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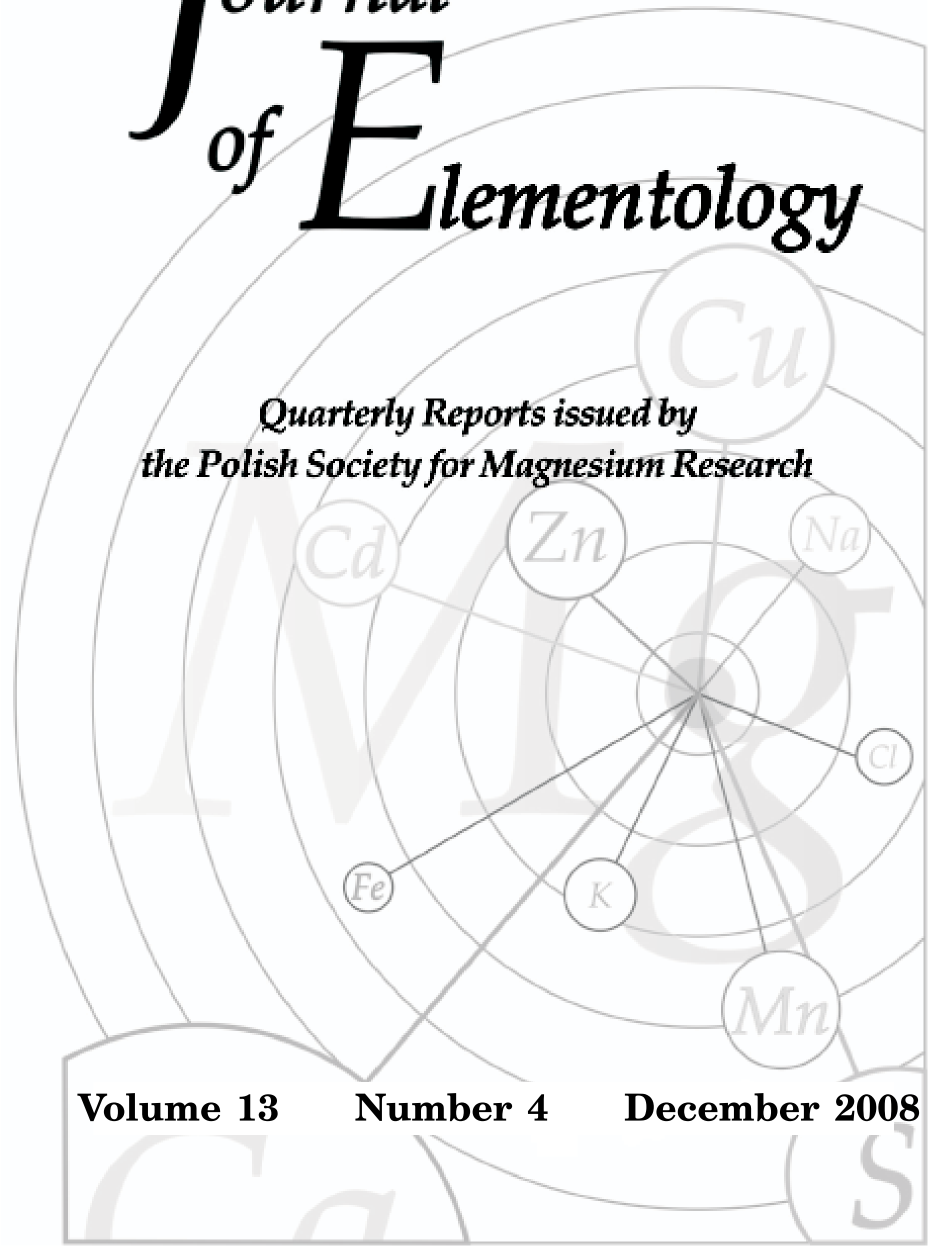

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Original papers

MUTUAL Cu, Fe AND Mn SOLUBILITY CONTROL UNDER DIFFERENTIATED SOIL MOISTURE STATUS

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Abstract

The effect of three different moisture statuses, i.e., 150, 200 and 300% FWC (Field Water Capacity) on copper (Cu), iron (Fe) and manganese (Mn) solubility and solution activity was investigated on soil samples characterised by different levels of copper contamination. Soils (200 g) were weighted into polyethylene containers (500 cm^3) and amounts of bidistilled water were properly added to reach the targeted moisture status of 150, 200 and 300% of the FWC. The incubation was held under laboratory conditions at the temperature of 19-20°C for a period of 30 days. Supernatants (10 cm^3) were collected at given intervals of time, filtered and analysed for pH (potentiometrically) as well as for Cu, Fe, and Mn (spectrophotometrically).

It was found that the activity of Cu^{2+} ions decreased gradually with increasing pH of the solution, irrespective of the moisture status and that this process was more pronounced for 150 and 200% than for 300% FWC. The activity of Fe varied in a narrow range i.e. –5.0 and –6.0 $mol_c dm^{-3}$ and was related to pH changes (in the range 4.0-7.5). The impact of increased moisture status on the solubility process was less pronounced. Further studies should be undertaken in order to elucidate such Fe behaviour. Manganese solution activity varied mostly between – 4.0 and –7.0 $mol_c dm^{-3}$, and was found to be less sensitive to pH changes. But it must be pointed out that the effect of high pH on the increase of Mn activity was limited, which implied that Mn^{2+} activity was moisture-dependent, basically. Care should be taken to avoid any submersion of soils subjected to contamination or pollution by trace metals, since any excess of stagnant water (anoxic conditions) leads to increased solubility and simultaneous activity of trace metals in the solution. This process is greatly strengthened by significant amounts of soil-born Fe and organic matter.

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Key words: copper, iron, manganese, solubility, solution pH, metal activity, soil moisture.

WZAJEMNA KONTROLA ROZPUSZCZALNOŚCI Cu, Fe I Mn W WARUNKACH ZRÓŻNICOWANEGO UWILGOTNIENIA GLEBY

Abstrakt

W pracy przedstawiono wpływ trzech różnych stanów uwilgotnienia gleby, tzn. 150, 200 i 300% PPW (polowej pojemności wodnej), na dynamikę rozpuszczalności oraz aktywność miedzi (Cu), żelaza (Fe) i manganu (Mn) w glebach o zróżnicowanym stopniu zanieczyszczenia miedzią. Próbki gleb o masie 200 g umieszczono w polietylenowych pojemnikach (500 cm^3), do których dodano odpowiednią ilość wody podwójnie destylowanej w celu zapewnienia właściwej wilgotności, tj. 150, 200 i 300% PPW. Zawartość pojemników inkubowano w warunkach laboratoryjnych w temp. 19-20°C przez okres 30 dni. W wyznaczonych odstępach czasu pobrano roztwory znad gleby (10 cm^3), przesączono i oznaczono w nich pH (potencjometrycznie) oraz Cu, Fe i Mn (spektrofotometrycznie).

Wykazano, że aktywność jonów Cu^{2+} malała stopniowo wraz ze wzrostem pH roztworów, niezależnie od stanu uwilgotnienia, z tym że ten proces zaznaczył się bardziej w przypadku 150 i 200% PPW w porównaniu z 300% PPW. Aktywność Fe wahała się w wąskim przedziale, tzn. –5.0 i –6.0 $mol_c dm^{-3}$, i zależała od zmian pH między 4,0 a 7,5. Ponadto, należy zaznaczyć, że wzrost uwilgotnienia na jego rozpuszczalność był słabo zaznaczony. Dalsze badania powinny być przeprowadzone w celu wyjaśnienia takiego zachowania żelaza. Aktywność manganu w roztworze wahała się między – 4,0 a –7,0 $mol_c dm^{-3}$ i była mniej wrażliwa na zmiany pH. Należy podkreślić, że wpływ wysokiego pH na wzrost aktywności Mn był ograniczony, co oznacza tym samym zależność aktywności jonów Mn^{2+} od uwilgotnienia gleby. Należy ostrożnie postępować z glebami zanieczyszczonymi lub skażonymi pierwiastkami śladowymi w celu uniknięcia ich zatapiania, gdyż nadmiar stojącej wody (warunki beztlenowe) prowadzi do wzrostu rozpuszczalności i jednocześnie aktywności tych metali w roztworze. Ten proces jest zdecydowanie silniejszy w warunkach występowania znacznych ilości Fe i materii organicznej.

Słowa kluczowe: miedź, żelazo, mangan, rozpuszczalność, aktywność metali, uwilgotnienie gleby.

INTRODUCTION

As a result of increased input from industry, traffic and agriculture, the average trace metal content in soils worldwide is increasing considerably. This has become a major point of concern during the last few decades since metal contents have reached potentially toxic levels in urban as well as agricultural soils in the vicinity of mining and metallurgical industries, thus creating areas frequently depicted as *hot spots* (HELIOS-RYBICKA 1996). Metal solubility, toxicity and transfer away from the contamination point depend on many soil chemical and physical properties. However, risk assessment is still entirely based on the total soil metal content. Ecotoxicological studies revealed, however, that metal speciation in the solution phase is one of the key factors that regulate metal toxicity in soil subjected to contamination

(Renner 1997, Meißner et al. 2008). Hence, metal toxicity (bioavailability) appears to be related closely to free metal activity in solution instead of the total soil metal content in the solid phase (Hare, Tessier 1996). Consequently, changes in soil chemical conditions that control the concentration and free metal activity (pH, solubility, desorption) induce changes in the availability without apparent changes in the total metal content (Spurgeon and Hopkin 1996, Mcbride et al. 1997). Soil parameters that affect metal solubility and speciation in the soil solution include moisture status, total metal content, pH, organic matter and clay content.

Soils may contain appreciable amounts of iron and manganese, which under conditions of water excess, activate redox processes of soils. Gotoh and Patrick (1972, 1974) studied the distribution of different forms of Fe and Mn in water logged soil over a wide range of closely controlled potential and pH conditions. These researchers reported that increases in water-soluble and exchangeable iron were favoured by a decrease in both redox potential and pH. Moreover, they found that at pH 5 almost all of the soil manganese was converted from reducible into water-soluble plus exchangeable. Schwab and Lindsay (1983) plotted the Fe^{2+} activity as a function of pH and reported that the Fe^{3+} activity is controlled by $FeCO_3$ (siderite) at pH below 8 and by $Fe_3(OH)_8$ (ferrosic hydroxide) at pH above 8. According to Hassan (1990), the solubility of manganese was controlled by MnO_2 at the beginning of the flooding period, whereas $MnCO_3$ (rhodochrosite) controlled this solubility up to 15 weeks.

Some reports have pointed out that contaminated soils are becoming frequently subjected to flooding (Helios-Rybicka and Adamiec 2001, Saha and Mandal 2000, Kelly et al. 2003).

This is of great concern since specific site contaminants are potentially dispersed into neighboring areas, thus creating some environmental threat. The monitoring of solubilized metals and their concentrations under flooding conditions is still a matter of great challenge due to several constraints (instability of bulk waters, continuous inputs of suspended organic and mineral particles, uncontrolled dilutions rates, etc.). Comprehensive investigations dealing with soil moisture status and how water excess induces the solubility and controls metal ion activity in the soil solution are still absent from scientific literature. Little is known about the effect of anoxic conditions on metal solubility and the subject has been less intensively studied because of experimental difficulties. With an increase in soil moisture, the solution concentration of most cationic metals and calcium tentatively increases due to strengthened solubility and desorption. But the commonly observed rise in solution pH leads to a decrease in metal concentration and ion activity in the solution due to adsorption processes (Kabata-Pendias 1993, Diatta et al. 2004).

The purpose of the current study was to evaluate the impact of three soil moisture statuses, i.e., 150, 200 and 300% FWC (Field Water Capacity) on mutual Cu, Fe, and Mn solubility and solution activity control of four soils under different levels of metal contamination, mainly that of copper. Another aim was to elucidate the effect of equilibrium pH on the activity of solubilized metals.

MATERIAL AND METHODS

Physical and chemical soil properties

Soil samples used in this study were collected (0-20 cm depth) from lands subjected to the Głogów Copper Smelter activity (N 51°41'03" and E 15°57'12", Poland). Prior to basic analyses, they were air-dried and crushed to pass through a 1 mm mesh sieve. Particle size distribution was determined according to the method of Bouyoucos-Casagrande modified by PRÓSZYŃSKI (rok). Organic carbon was determined by the dichromate wet oxidative method of Tiurin, in which soil organic carbon is oxidized with potassium dichromate in the presence of sulphuric acid. The unreacted potassium dichromate was back titrated with ferrous sulphate. Soil pH (in 0.01 mole $CaCl_2$) was potentiometrically measured in the soil solution (ratio 1:5), according to the Polish Standard (1994). Total dissolved solids (TDS) were determined conductively in supernatants collected during the incubation time and additionally in water extracts (soil/water ratio 1:5) at the end of the study. The specific surface area (SSA) was estimated by the ethylene glycol monoethyl ether (EGME) method as reported by CARTER et al. (1986). The cation exchange capacity (CEC) of the soils at their initial pH was determined by the barium chloride (0.10 mole $BaCl_2$) method as described by HENDERSHOT and DUQUETTE (1986), whereas the summation of exchangeable alkaline cations and acidity was done according to THOMAS (1982).

Experimental design and soils incubation

Prior to establishing the experiment, the water retention capacity of the soils was determined by the method of the capillary rise of soil moisture (LITYŃSKI et al. 1962). The method consists of filling in with a weighted soil a weighted metallic cylinder (diameter = 4 cm; length = 17 cm) supplied with a sieve located at 1 cm from the bottom of the cylinder, which is immersed in a crystallizer containing distilled water as shown below (Figure 1).

Once moisture appears on the surface of the soil, the cylinder containing the moist soil is taken out of the crystallizer and weighted after 1/2 to 1 hour. The difference between the moist and dry soil gives the amount of water the soil is able to retain.

The field water capacity (FWC) of soils used for the current experiment was as follows: 22.5% (No. 1 – slightly loamy sand); 31.0% (No. 2 – light

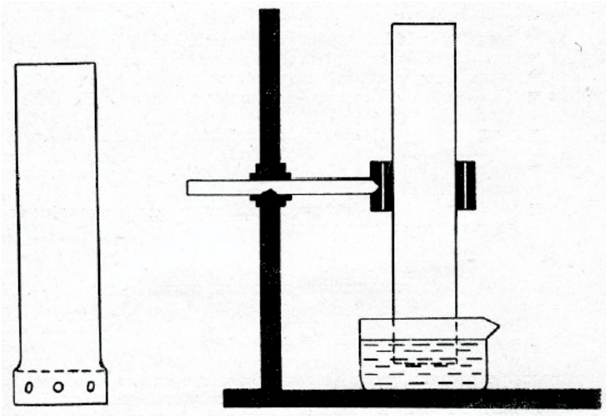

Fig. 1. Metallic cylinder and equipment used for the determination of the Field Water Capacity (FWC) of soils

loamy sand); 36.9% (No. 3 – light loamy sand) and 47.5% (No. 4 – silty medium loam). Soils (200 g) were weighted into polyethylene containers (500 cm^3) and amounts of bidistilled water were properly added to reach the targeted moisture status of 150, 200 and 300% of the FWC. The incubation was held under laboratory conditions at the temperature of 19-20°C for a period of 30 days. All objects were duplicated.

Supernatant collection and analysis

Supernatants (10 cm^3) were collected at given intervals of time, filtered and analysed for pH (potentiometrically) as well as for Cu, Fe, and Mn by the FAAS method (Flame Atomic Absorption Spectrophotometry, Varian 250 plus). The same amounts of bidistilled water (i.e., 10 cm^3) were added back to polyethylene containers in order to keep the moisture status constant. Computation of the results and processing of the graphs were performed by the use of Excel® sheet facilities.

RESULTS AND DISCUSSION

Some of the properties of the four soils used in the current study are listed in Table 1. The soil reaction was mostly acidic to *ca* neutral and varied within the range 4.3-6.5, in contrast to the organic carbon content, which ranged widely from 2.1 to 31.5 $g \cdot kg^{-1}$. The cation exchange (CEC) of soils varied accordingly from 11.3 to 29.7 cmol(+) kg^{-1}, which implied that the buffer properties were sufficiently developed and could probably affect

Table 1

Selected physical and chemical properties of investigated soils

Soil No.	Particle size (mm)			pH (0.01M $CaCl_2$)	C-org. ($g \cdot kg^{-1}$)	SSA[a] ($m^2 g^{-1}$)	CEC[b] cmol (+) kg^{-1}	Metals extracted by 6M HCl ($mg \cdot kg^{-1}$)		
	sand (1.0-0.05)	silt (0.05-0.002)	clay (<0.002)					Cu	Fe	Mn
	($g \cdot kg^{-1}$)									
1[c]	680	250	70	4.3	2.1	7.8	11.3	45.5	1085.3	1118.3
2	660	220	120	5.6	11.7	9.8	14.1	2041.3	2503.7	223.7
3	700	180	120	6.5	14.7	19.9	23.9	114.3	3484.3	247.5
4	390	300	310	5.4	31.5	68.3	29.7	10710.0	18180.0	3555.0

a: specific surface area; *b*: cation exchange capacity; *c*: 1 – slightly loamy sand; 2 – light loamy sand; 3 – light loamy sand; silty medium loam the solubility process. This could be related to a rather high level of both silt and clay contents, whose amounts ranged between 300 and 610 $g \cdot kg^{-1}$. The content of metals as listed in Table 1 varied significantly, e.g. from 45.5 up to 10 710 mg kg^{-1} in the case of copper (Cu). According to the guidelines elaborated by KABATA-PENDIAS et al. (1993), copper contents can be classified as follows: Soil No. 1 – natural content (degree 0); No. 3 – slight contamination (degree II) and for soils No. 2 and No. 3 – strong contamination (degree V) and pollution (degree V), respectively. Under conditions of water excess (submerged soils), some complex interacting geochemical reactions could be expected which would reduce or enhancne the solubility, desorption, diffusion of Cu, and interestingly iron (Fe) and manganese (Mn), basically (ROMKENS et al. 1996).

Copper solubility (activity) and solution pH at 150, 200 and 300% FWC

While most studies in environmental soil chemistry have focused on the adsorption or sorption of ions and molecules on soils, the solubility/desorption process is less studied but equally important. This is particularly true for soils that are subjected to (i) contamination or already contaminated and (ii) under conditions of temporary or permanent water excess. The results illustrated in Figures 2a, 3a, 4a for the respective moisture status 150, 200, 300 % FWC (Field Water Capacity) report the course of copper (Cu^{2+}) solubility expressed in terms of Cu^{2+} activity *versus* the equilibrium solution pH for the representative soils (No. 1, No. 2, No. 3 and No. 4). According to BARROW et al. (1981) and DIMIRKOU et al. (2002), the first Cu hydrolysis constant (pK_1) is about 8.0, (i.e., 7.2-8.1), which means that copper ions in the soil solution do not easily undergo hydrolysis process, which practically implies that the activity of Cu^{2+} ions should be significantly high throughout the pH values up to 8. This is particularly important since under a high contamination level and elevated moisture status, copper solubility, mobility and possible toxicity may gradually rise. Copper solubility shown in Figures 2a, 3a, 4a suggests that several ions and compounds may control its dynamics and activity in the soil solution. Most studies over the pH-dependence of copper solubility indicated that solubility and activity decreased with increasing pH (MA, DONG 2004). This can be illustrated by the copper solubility diagram reported by KABATA-PENDIAS and PENDIAS (1999), from which it could be deduced that up to pH *ca* 9.0, Cu^{2+}, $CuOH^{+}$, CuO, $Cu_2(OH)_2{}^{2+}$ and even $CuCO_3$ are the potential Cu forms and ions inducing copper solubility and solution activity. Some years ago HARTER (1983) assessed that at pH < 6.0, copper in solution was virtually present (≈98%) as Cu^{2+} and at pH 4.0 about 0.01% of the copper ions should be present as $CuOH^{+}$. This percentage may increase 10-fold as the pH is raised to 5.0. Thus, the effect of pH in depressing the activity of free ions in the soil solutions could also be attributed to an increase in the pH-dependent charges of organic matter and also of aluminium, iron and manganese oxides (ABD-ELFATTAH, WADA 1981, JARVIS 1981,

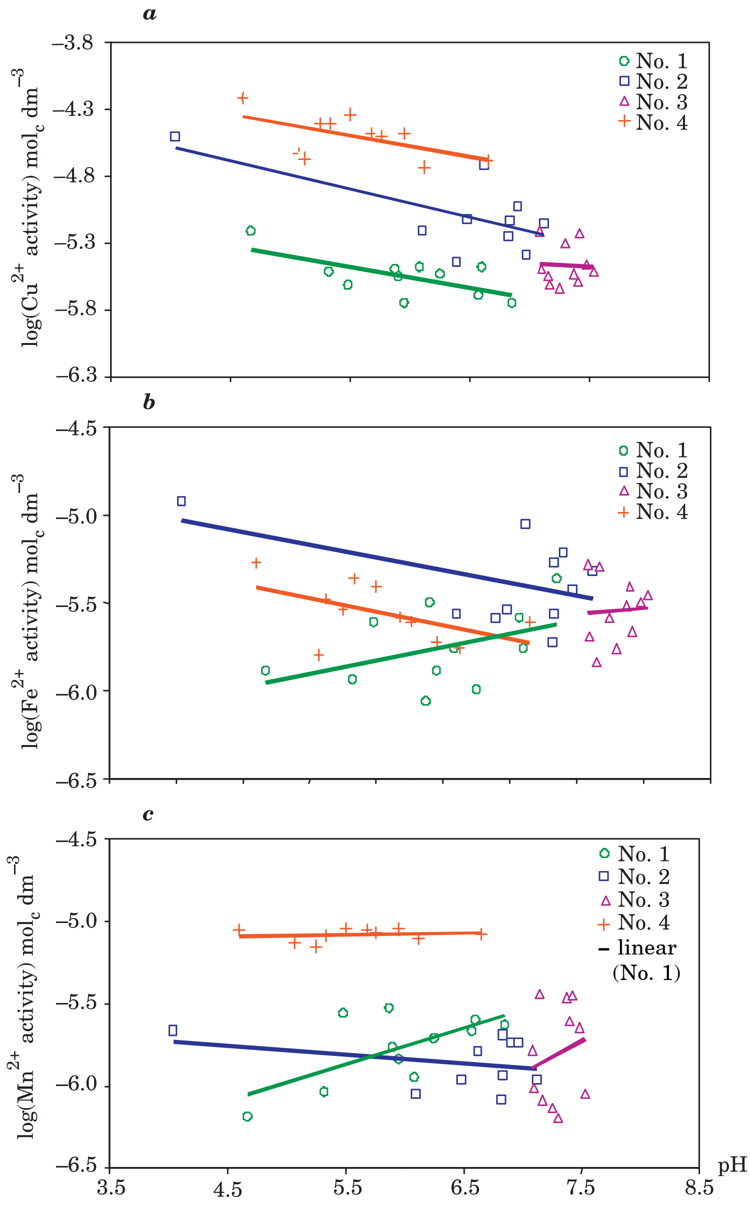

Fig. 2: Copper (*a*), iron (*b*) and manganese (*c*) activities in supernatants pH at 150% FWC (Field Water Capacity) for investigated soils (No. 1, No. 2, No. 3 and No. 4)

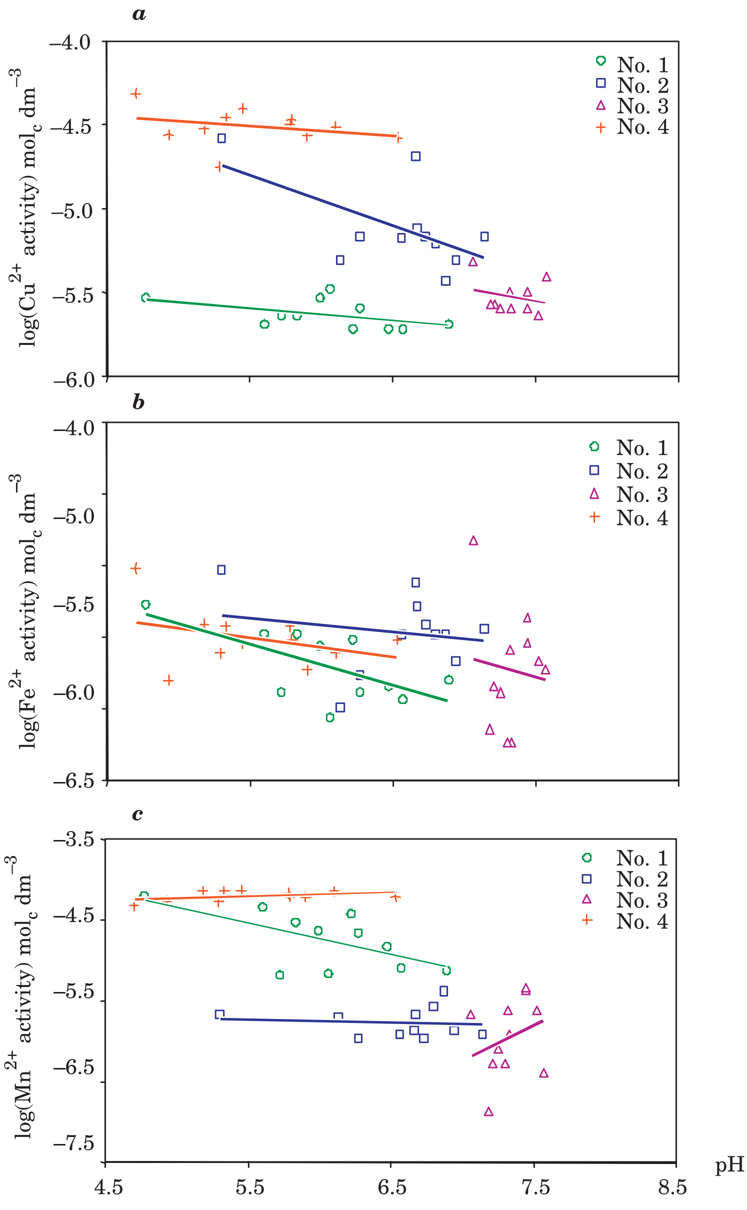

Fig. 3: Copper (*a*), iron (*b*) and manganese (*c*) activities in supernatants pH at 200% FWC (Field Water Capacity) for investigated soils (No. 1, No. 2, No. 3 and No. 4)

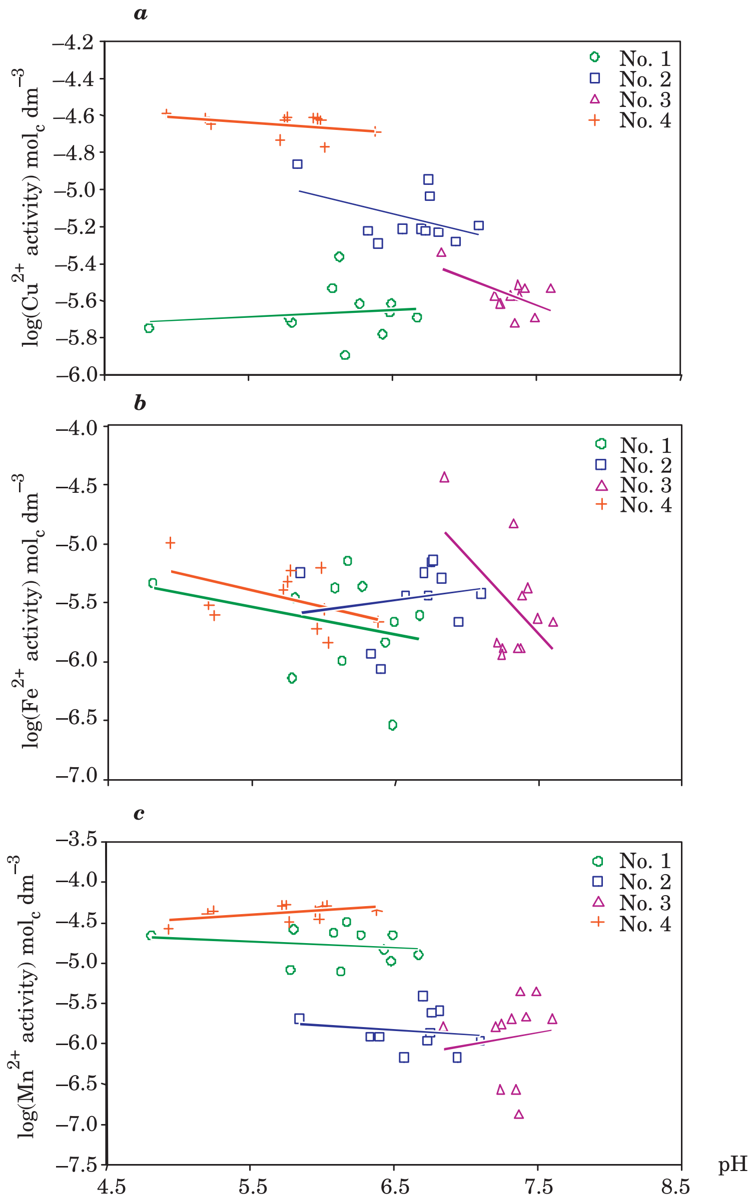

Fig. 4: Copper (*a*), iron (*b*) and manganese (*c*) activities in supernatants pH at 300% FWC (Field Water Capacity) for investigated soils (No. 1, No. 2, No. 3 and No. 4)

RITCHIE, JARVIS 1986, KING 1988). Therefore, if we assume that an increase of pH > 5.7 may lead to over 1.0% of copper ions hydrolyzing in a solution (SMITH, MARTELL 1976), then the mechanism that could be involved in the transformation can be presented as follows:

$$Cu^{2+}_{(aq)} + nH_2O \longrightarrow Cu(OH)_n^{2-n} + nH^+$$

that is at $n = 1$, the hydrolysis may concern:

$$Cu^{2+}_{(aq)} + HOH \longrightarrow CuOH^+ + H^+$$

The activity of Cu^{2+} ions decreased gradually with an increasing pH of the solution, irrespective of the moisture status. This trend was more pronounced for the 150 and 200% than for the 300% FWC, for which the activity of Cu^{2+} in soil No. 1 increased slightly, mainly in the pH range from 4.5 to 6.5. Such copper behaviour was in close agreement with the fact that in extremely diluted media the activity of ions increases and this reaction is activated by a relatively low solution pH. The impact of pH on copper activity in the case of the soils No. 3 deserves particular attention. As listed in Table 1, this soil was characterised by the highest pH value (6.5) and acid soluble copper amount of 114.3 $mg \cdot kg^{-1}$. Theoretically, copper activity in this medium could be expected to be similar to that observed for soil No. 1, since the acid soluble Cu amount was only 3-fold higher. Unexpectedly and interestingly, the values of the $\log(Cu^{2+})$ fluctuated within a similar range (from –4.3 to –5.8 $mol_c dm^{-3}$) except that more than 95% of all Cu activity values were comprised within the pH interval 7.0-7.5, irrespective of the soil moisture status. At such high pH, the dissolution of organic compounds may take place and Cu complexation directly reduces Cu activity levels according to the following reaction:

$$Cu^{2+} + H^xHUM \longrightarrow CuH^{(x-n)}HUM + nH^+$$

where HUM expresses the organic compound.

Copper forms stable complexes with dissolved organic compounds in solution and therefore higher levels will reduce the free ionic Cu^{2+} activity. According to ROMKENS and DOLFING (1998) at pH levels higher than 5 usually more than 99% of the total dissolved copper concentration is bound to dissolved organic compounds, which by developing their net negative surface charge acted as efficient carriers participating in copper solubility and mobility in soils, particularly under high moisture status. Copper activity of highly contaminated and polluted soils, i.e. No. 2 and No. 4, increased gradually along with an increasing moisture status. This could be attributed to the dilution effect, which practically strengthens Cu dissolution process and. Additionally, to the rise of pH along with an increase in the moisture status, as shown in Table 2. Similar observations were pointed out by CHUAN et al. (1996), SAHA and MANDAL (2004), KELLY et al. (2003) and WANG and STAUNTON (2005), who stated that soil submersion (flooding) rapidly induced strongly reducing conditions, with a consequent increase in pH, which acts in a dual way: on the one hand, it tends to lower Cu activity in the solution and, on the other hand, simultaneously increases Cu solubility throughout its complexation by pH-dependent dissolution of organic compounds.

Iron (Fe) and Mn solubility (activity) versus solution pH at 150, 200 and 300% FWC

The solubility and further activity of Fe and Mn in soil solutions proceed throughout redox processes, which are practically controlled by the soil moisture status, the length of anoxic conditions (occurrence of ferric – Fe^{3+} and ferrous – Fe^{2+} compounds) and simultaneously mediated by pH, whose changes from acidic to alkaline reaction induce the following reactions of Fe with hydroxide ions:

$$Fe^{3+}\ (Fe^{2+}) \xrightarrow{OH^-} Fe(OH)^{2+} \xrightarrow{OH^-} Fe(OH)_2^{+} \xrightarrow{OH^-} Fe(OH)_3$$

simple cation (*soluble*) — hydroxy metal cations (*relatively* soluble) — hydroxide (*insoluble*)

As shown in Figures 2b, 3b and 4b respectively for the moisture status 150, 200 and 300% FWC (Field Water Capacity), Fe activity varied in a narrow range i.e. -5.0 and -6.0 $mol_c dm^{-3}$, which implied that Fe transformations under current anoxic conditions could be related to more factors than to moisture status, basically. Solution reaction (pH) was one of the fundamental factors which decidedly influenced Fe solubility and activity. According to ATTA et al. (1996) the Fe reduction rate increased along with decreasing pH and was more pronounced at pH significantly below 7.0. This finding agrees well with the results illustrated in Figures 2b, 3b and 4b, which report the gradual decrease of Fe activity with increasing pH mainly for soils No. 1, No. 2 and No. 4 for pH values varying roughly between 5.55 and 6.63 (Table 2). The data included in Table 2 unequivocally show that anoxic conditions induced a slight rise in pH, which in turn could act as a limiting factor for Fe activity. This confirms that the more acidic the soil becomes, the more ferrous ions build up. As a consequence, larger ferrous iron concentrations after submergence are to be expected in more acidic soils since the hydrogen ion activity increases. This results in more iron being released from the crystalline or amorphous forms, which are present in the soil as free iron oxides, as earlier pointed out by PONNAMPERUMA (1986). Interestingly, pH changes recorded in the case of soil No. 3 (Table 2) deserve special attention, since they varied in a specifically close range i.e. 7.30-7.33, respectively for the soil moisture status 150-300% FWC. Importantly, Fe activity occurred at solution pH *ca* 7.5 (Figures 2b, 3b and 4b),

Table 2

Mean concentrations of soluble Cu and pH changes of soils incubated for 30 days at 150, 200 and 300% FWC (Field Water Capacity)

Soil No.	Moisture status (% of FWC – Field Water Capacity)					
	150		200		300	
	Cu concentrations ($mg \cdot dm^{-3}$) and solution pH					
	Cu	pH	Cu	pH	Cu	pH
No. 1*	0.234 ± 0.094	5.96	0.186 ± 0.039	6.04	0.178 ± 0.064	6.10
No. 2	0.773 ± 0.641	6.46	0.724 ± 0.564	6.55	0.578 ± 0.222	6.63
No. 3	0.283 ± 0.108	7.30	0.231 ± 0.065	7.33	0.216 ± 0.055	7.42
No. 4	2.603 ± 0.985	5.55	2.489 ± 0.624	5.54	1.772 ± 0.216	5.72

*1 – slightly loamy sand; 2 – light loamy sand; 3 – light loamy sand; 4 – silty medium loam

which implied that under alkaline conditions Fe solubility may occur but with a slightly lower Fe activity. This could be attributed to the formation of less soluble compounds such as hydroxides, and – with higher pH – carbonates. This observation is in line with the results reported by SCHWAB and LINDSAY (1983), who plotted the Fe^{2+} activity as a function of pH and pointed out that Fe^{3+} activity was controlled by $FeCO_3$ (siderite) at pH below 8.0 and by $Fe_3(OH)_8$ (ferrosic hydroxide) at pH above 8. Based on these statements, it can be deduced that Fe activity was less dependent on the moisture status but more on pH changes, which controlled the extent of Fe solubility. Siderite was most probably the main compound regulating Fe concentrations in the water solution of soils, since the pH did vary up to *ca* 7.5. The fact that Fe activity did not vary much (practically between –5.0 and –6.0 $mol_c dm^{-3}$) for solution pH between 4.0 and even 7.5 was strikingly unexpected. Further studies should be undertaken in order to elucidate this Fe behaviour.

The geochemical transformation of manganese (Mn) is by essence more complex than in the case of iron, due to the fact that the spectrum of Mn redox potential involves several steps i.e. from Mn^{7+} up to Mn^{2+} (KABATA-PENDIAS, PENDIAS 1999). According to BRADY (1984) in well-oxidized soils the manganic (Mn^{4+}) form is dominating whereas under waterloggind conditions the reduced manganous form (Mn^{2+}) prevails. The results reported in Figures 2c, 3c and 4c for the moisture status 150, 200 and 300% FWC (Field Water Capacity), respectively, are basically quantitative, therefore the Mn^{2+} was considered to represent the overall reduced forms of Mn under anoxic conditions of the current trial. Manganese solubility expressed in terms of its solution activity ($\log Mn^{2+}$) varied mostly between –4.0 and –7.0 $mol_c dm^{-3}$, irrespective of the effect of the moisture status and was found to be significantly sensitive to pH changes. On the other hand manganese solubility depended additionally on the soil-born manganese content (extracted in

6 moles HCl), since soils characterized by highest content (i.e. No. 1 and No. 4, Table 1) exhibited higher Mn activity, which gradually decreased along with the pH increase as observed for the soil No. 1. Interestingly, manganese solubility and activity course developed in the case of the soil No. 4, seemed to be more complex and may not be related to its soil-born content solely. A slight increase in Mn activity with a raise of pH was even unexpected, since the equilibrium solution pH of this soil did not exceed the pH value of 6.5. Two assumptions may be formulated: i) the extremely high Fe content detected in the soil No. 4 could be a substantial source of electrons, which acted as efficient Mn reducing agents in one hand and ii) the significantly high organic matter content (31.5 $g \cdot kg^{-1}$) as a potential precursor of electrons, which could additionally strengthen Mn reduction, on the other hand. This finding is in line with the data reported by Atta et al. (1996) who pointed out that the enrichment of submerged soils with organic matter induced an increase in the water-soluble Mn fractions by simultaneously decreasing the content of the residual Mn pool. According to Hassan (1990), the solubility of manganese was controlled by MnO_2 at the onset of flooding, whereas the $MnCO_3$ (rhodochrosite) controlled this solubility in prolonged periods of water logging, i.e., up to 15 weeks. In the current study, the suggested Mn solubility and activity could have been mediated by the following reactions:

$$\underset{\text{solid (insoluble)}}{MnO_2} + 4H^+ + 2\bar{e} \longrightarrow \underset{\text{simple cation (soluble)}}{Mn^{2+}} + 2H_2O$$

$$\underset{\text{hydroxycation (relatively soluble)}}{MnOOH^{3+}} + 3H^+ + 4\bar{e} \longrightarrow \underset{\text{simple cation (soluble)}}{Mn^{2+}} + 2H_2O$$

Manganese geochemical transformations observed for soil No. 3, as illustrated in Figures 2c, 3c and 4c, operated basically within pH ranges 7.0 and 7.6, similarly to those reported earlier for Fe. Interestingly, Mn^{2+} activity in a soil solution tended to increase along with pH increase, in contrast to the trends reported in many studies. Besides, it acted opposite to Fe. At such slightly alkaline pH of the solution, the hydroxy manganese forms could prevail, hence controlling the solubility as well as activity processes. The high-pH inducing manganese solubility and activity observed in the current study, which was characteristic for the slightly alkaline soil No. 3, agreed with the data reported by Atta et al. (1996) and Wang and Staunton (2005). But it should be pointed out that the effect of high pH on the increase of Mn activity was limited, which implied that this activity was basically moisture-dependent. The occurrence of $MnCO_3$ (Hassan 1990) as a manganese solubility/activity controlling factor was less probable under the current experimental conditions.

CONCLUSIONS AND STATEMENTS

1. The activity of Cu^{2+} ions decreased gradually with increasing pH of the solution, irrespective of the moisture status. This trend was more pronounced for the 150 and 200% than for 300% FWC (Field Water Capacity). This was attributed to the dilution effect, which practically strengthened the Cu dissolution process and, additionally, to the rise of pH along with the increased moisture status.

2. The activity of Fe varied in a narrow range i.e., –5.0 and –6.0 $mol_c dm^{-3}$, which implied that its transformations under current anoxic conditions was related more strongly to the solution pH than to the moisture status. The fact that the Fe activity did not vary greatly for a solution pH between 4.0 and 7.5 was strikingly unexpected. Further studies should be undertaken in order to elucidate such Fe behaviour.

3. Manganese solubility and solution activity varied mostly between –4.0 and –7.0 $mol_c dm^{-3}$, and was found to be less sensitive to pH changes. But it should be decidedly pointed out that the effect of high pH on the increase of Mn activity was limited, which implied that Mn^{2+} activity was moisture-dependent, basically.

4. Care should be taken to avoid any submersion of soils subjected to contamination and polluted by trace metals, since any excess of stagnant water (anoxic conditons) leads to increased solubility and simultaneously higher activity of trace metals in the solution. This process is evidently strengthened by significant amounts of soil-born Fe and organic matter.

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EVALUATION OF CHERRY TOMATO YIELDING AND FRUIT MINERAL COMPOSITION AFTER USING OF BIO-ALGEEN S-90 PREPARATION

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Abstract

The aim of the experiments carried out in a high, polyethylene in the years 2004-2006 was to examine the influence of the algae preparation Bio-algeen S-90 on yield, mineral composition of fruits, content of assimilatory pigments, intensity of assimilation and transpiration and photosynthetic water use efficiency index of the cherry tomato cv. Conchita F_1. The results on tomato yields proved significant differences in total and marketable yield under the influence of Bio-algeen. The biggest total and marketable yields were obtained when plants were treated three times with Bio-algeen. Bio-algeen also caused increase in content of mineral components in cherry tomato fruits. Three- and fourfold sprays increased the content of total nitrogen, phosphorus, potassium, calcium, zinc, iron and nitrates. Treating tomato plants with the preparation increased the content of assimilatory pigments of tomato. Increase in a number of Bio-algeen sprays led to higher contents of total, a and b chlorophyll and carotenoids in leaves. Intensity of assimilation and transpiration of the plants and water use efficiency index depended on number of plants spraying with Bio-algeen. Plants sprayed two and three times with Bio-algeen were characterized by greater transpiration and assimilation intensity, but also by smaller efficiency of water use index.

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Key words: vegetables, seaweeds, content of bioelements, water use efficiency index.

OCENA PLONOWANIA I SKŁADU MINERALNEGO OWOCÓW POMIDORA DROBNOOWOCOWEGO PO ZASTOSOWANIU PREPARATU BIO-ALGEEN S-90

Abstrakt

Celem badań prowadzonych w wysokim tunelu foliowym, w latach 2004-2006, było określenie wpływu stosowania preparatu z alg morskich Bio-algeen S-90 na plon, skład mineralny owoców, zawartość barwników asymilacyjnych, intensywność asymilacji i transpiracji oraz wskaźnik efektywności wykorzystania wody w procesie fotosyntezy roślin pomidora drobnoowocowego odmiany Conchita F_1. Wykazano istotne różnice w plonie ogółem i handlowym pod wpływem traktowania roślin preparatem Bio-algeen S-90. Największy plon ogółem i handlowy owoców uzyskano w przypadku 3-krotnego oprysku preparatem. Preparat Bio-algeen wpłynął również na zwiększenie zawartości składników mineralnych w owocach pomidora drobnoowocowego. Opryskiwanie 3- i 4-krotne roślin zwiększyło poziom azotu ogólnego, fosforu, potasu, wapnia, cynku, żelaza oraz azotanów. Traktowanie roślin preparatem zwiększyło zawartość barwników asymilacyjnych u pomidora. Zwiększanie liczby oprysków preparatem Bio-algeen wpłynęło na wyższą zawartość chlorofilu a, b i całkowitego oraz karotenoidów w liściach. Intensywność asymilacji i transpiracji roślin oraz wskaźnik efektywności wykorzystania wody zależał od liczby oprysków preparatem Bio-algeen. U roślin, które opryskiwano 2- i 3-krotnie, stwierdzono większą intensywność transpiracji i asymilacji, ale mniejszy wskaźnik efektywności wykorzystania wody.

Słowa kluczowe: warzywa, algi morskie, zawartość biopierwiastków, wskaźnik wykorzystania wody.

INTRODUCTION

Bio-algeen is a natural extract from sea thallophytic algae containing a brown pigment. It contains over seventy active components: macro and microelements (CZECZKO and MIKOS-BIELAK 2000). It is a source of the following chemical elements: nitrogen (0.02%), potassium (0.096%), phosphorus (0.006%), magnesium (0.21%), calcium (0.31%), iron (6.3 $mg \cdot kg^{-1}$), boron (16 $mg \cdot kg^{-1}$), molybdenum, cobalt and selenium (PATIER et al. 1993).

The aim of using extracts from sea algae in tomato cultivation is to form a strong, well developed root system (PERTUIT 1995). Om addition, biostimulators enhance plants' tolerance to uncomfortable growth conditions (water deficiency, temperature changes, diseases attack). Another important role of biostimulators is to increase assimilation rate and availability of nutrients. They also improve plants' growth, flowering and fruit setting as well as increase yield and quality of fruits (BECKET and VAN STANDEN 1989, WYSOCKA-OWCZAREK 2001).

Using extracts from sea algae in plants cultivation enhances the chlorophyll content in leaves. Chlorophyll level depends on betanine level in seaweed extract (BLUNDEN et al. 1996).

MATERIAL AND METHODS

In 2004-2006 experiments were carried out at the Department of Vegetable Crops of the University of Agriculture in Szczecin to examine the influence of the sea algae preparation Bio-algeen S-90 on yield, chemical composition of fruits, content of assimilatory pigments and intensity of plant assimilation and transpiration in the cherry tomato cv. Conchita F_1. Tomatoes were cultivated in a high, unheated polyethylene tunnel.

Tomato seeds were sown in a greenhouse on 20th March and seedlings were planted on 20th May 2004 and on 17th May 2005 and 2006 in an unheated foil tunnel in rows, using row spacing 1.4 x 0.25 m. The surface area of every plot was 3.5 m^2 (10 plants on the plot of ground).

Bio-algeen S-90 preparation was used in concentration 0.3% in the form of spraying conducted one, two, three and four times. A dose of 1 ml of Bio-algeen was applied to each plot every time. The first spraying was carried out at the stage of 2-3 proper leaves, second – before planting, third – at the beginning of flowering, fourth – at the initial stage of plants yielding.

Plants were headed in the first decade of July behind the sixth cluster. During the plant vegetation period the biometrical measurements were carried out (height of plant, diameter of stem, number of leaves, number of flowers and fruits).

Harvest of fruits was conducted from the third decade of July to the first decade of September. After the harvest total and marketable yield were evaluated.

Content of carotenoids and chlorophyll a and b were estimated in leaves. After the harvest, chemical analyses of macro- and microelements content were carried out. Total nitrogen was estimated according to the method recommended by Kjeldahl, phosphorus – according to the colorimetric method, potassium, magnesium, calcium; iron and zinc were estimated in a plasma spectrometer. Nitrate nitrogen ($N\text{-}NO_3$) content was estimated according to the potentiometric method. Chlorophyll content was estimated according to the method recommended by Arnon et al. (1956) in Lichtenthaler and Wellburn modification (1983). The measurements of gas exchange were carried out by the means of a gas analyser IRGA LCA-4 working in an open system.

The experiment was conducted in a randomized blocks design, in four replications. Results were statistically verified by Tukey's test at the significance level $\alpha = 0.05$.

RESULTS AND DISCUSSION

Results regarding tomato yielding proved significant differences in total and marketable yield under the influence of Bio-algeen. The greatest total and marketable tomato fruit yield was obtained in the years 2004-2006 when plants were sprayed three times with the preparation (Figure 1). Synthesis of the experimental results proved that the total yield in this case amounted to 6.64 kg·m^{-2} and was on average 1.2 kg·m^{-2} greater than the yield of a control combination and of the combination where Bio-algeen was used once. The marketable yield, obtained from the control plants and from those plants which were treated only once with Bio-algeen was significantly smaller, by an average 1.27 kg·m^{-2}.

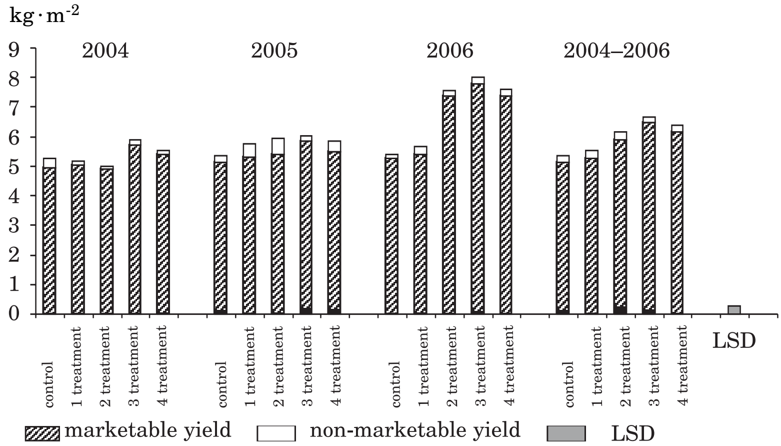

Fig. 1. The influece of Bio-algeen treatment on the yield of small-sized tomato in the years 2004-2006

Tomato is an important source of mineral salts, especially of potassium. Nurzyński and Michałojć (1998) determined 0.29-0.35% P, 3.84-3.95% K, 0.10--0.11% Ca and 0.17-0.18% Mg in tomato fruit dry matter. Kobryń (2002) noticed the mineral composition of tomato fruits is highly variable between cultivars. In the years 2002-2003 Dobromilska and Kujath (2004) estimated the levels of macroelements (g·kg^{-2}) as N 22.6, P 5.43, K 31.2, Ca 1.63, Mg 1.4 and microelements (mg·kg^{-1} d.m.) as Fe 80 and Zn 24.13 in fruit of the cherry tomato cv. Conchita F_1 cultivated in a high, polyethylene tunnel.

The chemical analyses of tomato fruits proved that in 2004 fruits contained more total-N, P, Ca, Mg and Zn and less K and Fe in comparison with 2005 (Table 1). During fruit ripening in 2004 the weather conditions were very convenient (greater insolation). Koźmiński and Michalska (2004) observed that insolation in Poland is a highly varied factor. The results

Table 1

The effect of Bio-algeen S-90 spraying on minerals and nitrates in cherry tomato fruits cv. Conchita F_1

Number of treatments	Year	Macroelements ($g \cdot kg^{-1}$ d.m)					Microelements ($mg \cdot kg^{-1}$ d.m)		Nitrates ($mg\ NO_3^- \cdot kg^{-1}$ d.m)
		N	P	K	Ca	Mg	Zn	Fe	
Control	2004	20.50	5.10	24.90	1.70	1.40	20.0	60.0	205.0
	2005	16.10	4.86	31.85	1.55	1.03	11.0	170.0	200.0
	x	18.30	4.98	28.38	1.63	1.22	15.5	115.0	202.5
1 treatment	2004	20.80	5.90	26.70	1.70	1.40	20.0	80.0	220.0
	2005	15.89	5.54	31.85	1.55	1.05	14.0	170.0	200.0
	x	18.35	5.72	29.28	1.63	1.23	17.0	125.0	210.0
2 treatments	2004	20.60	5.90	26.30	1.70	1.40	20.0	80.0	245.0
	2005	16.52	5.92	31.85	1.94	1.03	14.0	180.0	210.0
	x	18.56	5.91	29.08	1.82	1.22	17.0	130.0	227.5
3 treatments	2004	21.65	6.40	28.20	2.10	1.30	28.0	100.0	235.0
	2005	16.52	5.82	32.37	1.74	1.03	14.0	180.0	210.0
	x	19.09	6.11	30.29	1.92	1.17	21.0	140.0	222.5
4 treatments	2004	23.50	6.40	28.60	1.90	1.30	26.0	80.0	245.0
	2005	17.64	5.28	32.79	1.55	1.00	14.0	180.0	230.0
	x	20.57	5.84	30.70	1.73	1.15	20.0	130.0	237.5
Mean		18.97	5.71	29.55	1.75	1.20	18.1	128.0	220.0
$LSD_{\alpha=0.05}$		0.85	0.41	1.22	0.09	n.s.	1.20	13.8	15.5

of our chemical analyses on samples obtained in 2004-2005 proved that total-N content increased with increase of the preparation doses. Fruits obtained from plants sprayed four times with Bio-algeen accumulated most total-N. Content of total-N increased in comparison with the control object by 2.22 $g \cdot kg^{-1}$ d.m. Phosphorus content in fruits also increased from 4.98 $g \cdot kg^{-1}$ d.m. in the control object to 6.11 $g \cdot kg^{-1}$ d.m. when plants were treated three times with Bio-algeen. All the sprayings increased significantly P content in comparison with control.

Bio-algeen sprays significantly differentiated potassium content in tomato fruits, which in turn affected their colour. The preparation used three and four times increased K level in fruits to 30.49 $g \cdot kg^{-1}$ d.m. on average, whereas fruits of control plants contained by 2.11 $g \cdot kg^{-1}$ d.m. less potassium.

Table 2

The influence of Bio-algeen S-90 spraying on the assimilatory pigments content in small-sized tomato leaves (mg·g^{-1} f.m.)

Number of treatments	Years	Content of assimilatory pigments (mg·g^{-1} f.m.)			
		chlorophyll a	chlorophyll b	total chlorophyll	carotenoids
Control	2004	0.94	0.42	1.36	0.45
	2005	1.39	0.56	1.94	0.56
	x	1.17	0.49	1.65	0.51
1 treatment	2004	0.99	0.47	1.46	0.43
	2005	1.42	0.56	1.98	0.57
	x	1.21	0.52	1.72	0.50
2 treatments	2004	0.99	0.47	1.46	0.48
	2005	1.52	0.62	2.15	0.62
	x	1.26	0.55	1.81	0.55
3 treatments	2004	1.02	0.49	1.51	0.55
	2005	1.51	0.62	2.13	0.60
	x	1.27	0.56	1.82	0.58
4 treatments	2004	1.09	0.51	1.60	0.54
	2005	1.57	0.64	2.22	0.62
	x	1.33	0.58	1.91	0.58
Mean		1.25	0.60	1.78	0.54
LSD $_{\alpha=0.05}$		0.11	0.07	0.13	n.s

Calcium content in fruits increased as a result of double, triple and fourfold plant spraying with Bio-algeen. Fruits treated three times with the preparation contained most Ca (1.93 g·kg^{-1} d.m.).

Magnesium content did not depend on treatments involving the tested preparation, reaching on average 1.19 g·kg^{-1} d.m. Zn level in tomato fruits depended significantly on the number of sprays with Bio-algeen. As the number of sprays increased, the contnet of this element in tomato fruit likewise increased, especially when triple and fourfold sprays were used. As a mineral component of foodstuffs, zinc is very important to human health because it is included in an enzyme which reduces free radicals (MANGEL and KIRGBY 1983). Fe deficiency causes poor tomato fruit setting (WYSOCKA-OWCZAREK 2001). The Fe level in tomato fruit of cv. Conchita F_1 reached to 128 mg·kg^{-1} d.m. on average and was dependent on a number of sprays with the preparation. A significantly smaller (115 mg·kg^{-1} d.m.) content of this chemical component was estimated in control than in fruits of the plants sprayed two, three and four times (on an average 133 mg·kg^{-1} d.m.).

Bio-algeen caused a significant increase in the content of nitrates in fruits, especially in 2004. Fruits of the control plants contained on average, for both years of the experiments, 26.7 mg NO_3^-·kg^{-1} d.m. less than the plants sprayed two, three and four times.

Our analysis of the determination of assimilatory pigments in tomato leaves proved that chlorophyll and carotenoids were less abundant in 2004 than in the following year (Table 2). High temperature and higher insolation in August 2004 accelerated fruit ripening, yellowing of leaves and earlier termination of the vegetative season. Leaves of plants treated four times with Bio-algeen contained more total, a and b chlorophyll than leaves of the control plants (0.19, 0.09 and 0.26 mg·g^{-1} f.m., respectively). Bio-algeen did not differentiate the level of carotenoids in tomato leaves.

Table 3

The effect of Bio-algeen S-90 spraying on assimilation and transpiration in cherry tomato

Number of treatments	Years	Assimilation (μmol CO_2·m^{-2}·s^{-1})	Transpiration (mmol H_2O·m^{-2}·s^{-1})	Water use efficiency index (mmol·mol^{-1})
Control	2004	7.01	0.82	8.55
	2005	4.73	0.05	94.60
	x	5.87	0.44	13.34
1 treatment	2004	7.22	0.98	7.37
	2005	4.61	0.15	30.73
	x	5.92	0.57	10.39
2 treatments	2004	8.39	1.87	4.49
	2005	9.78	1.06	9.23
	x	9.09	1.47	6.18
3 treatments	2004	11.46	1.99	5.76
	2005	9.23	1.20	7.70
	x	10.35	1.60	6.47
4 treatments	2004	7.74	0.94	8.23
	2005	3.28	0.46	7.13
	x	5.51	0.70	7.87
Mean		7.35	0.96	7.66
LSD $_{\alpha=0.05}$		1.21	0.12	3.97

Photosynthetic water use efficiency index, defined as a relation of assimilation (A) to transpiration (E), largely conditions plant productivity, especially under stress conditions of water deficit and deficient supply of nutrients (Tuner 1997). No dependence of high water use efficiency and high transpiration intensity was found, which confirms the assumption that tomato plants do not tolerate low supply of nutrients in soil (Table 3). Plants treated two and three times with Bio-algeen were characterized by more intense transpiration and assimilation, but also by the smaller photosynthetic water use efficiency index.

Under suitable plant growth conditions, intensity of CO_2 assimilation and transpiration are correlated with each other, as intense synthesis of organic compounds increases demand for water and nutrients. Plants which transpire more intensely are capable of nourishing all their organs better (Wróbel 2006).

CONCLUSIONS

1. Triple spraying tomato plants with Bio-algeen S-90 in the concentration of 0.3% significantly increased total and marketable yield.

2. Bio-algeen increased mineral components in fruits of the cherry tomato cv. Conchita F_1 (N, P, K, Ca, Zn and Fe), especially when was used several times.

3. Bio-algeen used in tomato cultivation increased a, b and total chlorophyll content in leaves.

4. Spraying of tomato plants with Bio-algeen two and three times depressed water use efficiency index in the process of photosynthesis.

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POTASSIUM CONTENT IN MAIZE AND SOIL FERTILIZED WITH ORGANIC MATERIALS

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Abstract

Since potassium is essential for plant nutrition, should this element be deficient in sewage sludge used for plant fertilization supplementary mineral potassium fertilization is necessary. The aim of the present study has been to evaluate the effect of fertilization with organic materials on maize yield, its potassium concentrations and the content of bioavailable forms of potassium in soil of different grain size distribution. The impact of fertilization on potassium concentrations in maize was examined in a 3-year, two-factor pot experiment (the factors were soil and fertilization). The grain size distribution of the test soil material was weakly loamy sand (psg), sandy silt loam (gpp) and medium silt loam (gśp). Sewage sludge originated from two different municipal mechanical and biological sewage treatment plants. Mixtures of sewage sludge with peat were prepared at a gravimetric ratio 1:1 converted to material dry matter. For chemical analyses the plant material was mineralized in a muffle furnace (at 450°C for 5 hrs) and the remains were dissolved in diluted nitric acid. Bioavailable potassium was determined with Egner-Riehm method in the soil material, which was dried and sifted through a 1mm mesh sieve. Potassium was determined by flame photometry in solutions of the plant material and soil extracts. Fertilization with sewage sludge and mixtures with peat had a more positive effect on maize yield than fertilization with mineral salts. In comparison with organic materials supplied to the soil, mineral salt treatment significantly increased potassium content in maize biomass. Mineral supplementation of potassium introduced with organic materials and its balancing did not increase soil abundance in bioavailable potassium in comparison with the initial abundance, although a diversified soil response to the applied fertilization was observed.

Key words: potassium, sewage sludge, maize.

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ZAWARTOŚĆ POTASU W KUKURYDZY I GLEBACH NAWOŻONYCH MATERIAŁAMI ORGANICZNYMI

Abstrakt

Biorąc pod uwagę niezbędność potasu w żywieniu roślin, deficytowa zawartość tego składnika w osadach ściekowych wykorzystywanych do nawożenia roślin wymaga jego uzupełnienia w formie mineralnej. Celem badań była ocena wpływu nawożenia materiałami organicznymi na plon części nadziemnych i korzeni kukurydzy, zawartości w nim potasu oraz zawartości form przyswajalnych tego składnika w glebach o różnym składzie granulometrycznym. Ocenę wpływu nawożenia na zawartość potasu w kukurydzy przeprowadzono w 3-letnim dwuczynnikowym doświadczeniu wazonowym (czynniki: gleba, nawożenie). Do badań użyto materiału glebowego o składzie granulometrycznym piasku słabo gliniastego (psg), gliny piaszczystej pylastej (gpp) i gliny średniej pylastej (gśp). Osady ściekowe pochodziły z dwóch różnych komunalnych oczyszczalni mechaniczno-biologicznych. Mieszaniny osadów ściekowych z torfem sporządzono w stosunku wagowym 1:1, w przeliczeniu na suchą masę materiałów. W celu oznaczenia potasu materiał roślinny mineralizowano w piecu muflowym (temp. 450°C, 5 h), pozostałość roztworzono w rozcieńczonym kwasie azotowym. W materiale glebowym wysuszonym i przesianym przez sito o średnicy oczek 1 mm potas przyswajalny oznaczono metodą Egnera-Riehma. W uzyskanych roztworach materiału roślinnego oraz ekstraktach glebowych potas oznaczono metodą fotometrii płomieniowej. Nawożenie osadami ściekowymi i mieszaninami osadów z torfem działało korzystniej na plony kukurydzy niż nawożenie solami mineralnymi. W porównaniu z zastosowanymi doglebowo materiałami organicznymi, nawożenie solami mineralnymi istotnie zwiększyło zawartość potasu w biomasie kukurydzy. Zrównoważenie solami mineralnymi ilości potasu wprowadzanego z materiałami organicznymi i jego zbilansowanie nie zwiększyło zasobności gleby w potas przyswajalny, w porównaniu z zasobnością wyjściową, chociaż zaobserwowano różne zmiany w glebach po zastosowanym nawożeniu.

Słowa kluczowe: potas, osady ściekowe, kukurydza.

INTRODUCTION

Good fertilization value of sewage sludge has been confirmed in numerous investigations (MAZUR 1995, WOŁOSZYK and KRZYWY 1999, KRZYWY et al. 2004). Sewage sludge can be used for soil and plant fertilization mainly because of the significant concentrations of organic matter and nutrients it contains (BARAN et al. 2002). If all rules for safe application of sewage sludge in agriculture are observed, this material may supplement or replace farmyard manure.

Sewage sludge may have a highly different chemical composition, sometimes comprising high amounts of nitrogen or phosphorus content but having little potassium (MAZUR 1996, KALEMBASA et al. 2001). Such low concentrations of this element in sewage sludge are due to the fact that potassium occurs in the form of easily soluble salts (KALEMBASA et al. 1999) which dissociate in water solutions, and this favours retention of potassium in sewage waters during the technological process of sludge separation.

Considering the crucial role of potassium in animal nutrition, deficit of this element in sewage sludge used for plant fertilization requires its supplementation in the mineral form.

The present research intended to asses the effect of fertilization with organic materials on the yield of maize shoot and root dry mass, their potassium concentrations and the content of bioavailable forms of this element in soils different in the grain size composition.

MATERIAL AND METHODS

The effect of fertilization on potassium content in maize was assessed in a two-factor (factors: soil and fertilization) pot experiment conducted in 2003-2005. The research was carried out on three soils and the experimental design, identical for each soil, comprised 7 treatments in three replications: without fertilization – (0); fertilization with chemically pure salts – (NPK); farmyard manure – (OB); sewage sludge A (OŚA); mixture of sewage sludge A with peat – (MOŚA); sewage sludge B (OŚB) and mixture of sewage sludge B with peat (MOŚB). The following soil material was used for the experiment: weakly loamy sand (psg), sandy silt loam (gpp) and medium silt loam (gśp), all collected from the arable layer (0-20 cm) of plough lands near Krakow. Sewage sludge originating from two different mechanical and biological municipal treatment plants and mixtures with peat were used for the experiment. Sewage sludge was mixed with peat at a weight ratio 1:1 per dry mass of organic materials. Peat containing 408 $g \cdot kg^{-1}$ d.m. comprised 88 $g \cdot kg^{-1}$ ash, 34.4 g $N \cdot kg^{-1}$d.m., 0.91g $P \cdot kg^{-1}$d.m. and 1.14 g $K \cdot kg^{-1}$d.m. The chemical composition of the organic materials and soil material (values per dry mass assessed at 105°C) is given in Tables 1 and 2.

Plastic pots used for the experiment contained 5.50 kg of air-dried soil material. Before the experiment, the soils were gradually moistened to 30% of maximum water capacity. Afterwards, sandy loam and medium silt loam were limed to obtain the reaction recommended in the Ministry decree (soil pH in agricultural areas no less than 5.6) (*Rozporządzenie*… 2002). Liming was applied separately in each pot. Chemically pure CaO was used in a dose calculated on the basis of soil hydrolytic acidity. Next, all the soils were left for 4 weeks and water loss was occasionally supplemented. Finally, organic fertilization was carried out in doses corresponding to 1.20 g $N \cdot pot^{-1}$. Phosphorus and potassium were supplemented with solutions of chemically pure salts [P – $Ca(H_2PO_4)_2 \cdot H_2O$ and K – KCl] to equalize quantities of these elements supplied with the organic materials. In the mineral (NPK) treatment the doses of nitrogen, phosphorus and potassium were identical to the ones in the organic material treatments. Doses of N, P and K were, respectively, 1.20 g $N \cdot pot^{-1}$ as NH_4NO_3, 1.26 g $P \cdot pot^{-1}$ as $Ca(H_2PO_4)_2 \cdot H_2O$ and

Table 1

Chemical composition of materials used in the experiment

Specification		FYM (OB)	Sewage sludge (OŚA)	Sewage sludge + peat (MOŚA)	Sewage sludge (OŚB)	Sewage sludge + peat (MOŚB)
Dry matter ($g \cdot kg^{-1}$)		189	310	343	418	372
pH (H_2O)		6.22	6.12	5.57	5.73	5.20
Organic matter ($g \cdot kg^{-1}$ d.m.)		679	353	652	552	771
Total forms						
N	$g \cdot kg^{-1}$ d.m.	21.6	17.0	24.7	37.4	35.1
S		7.24	8.81	6.23	14.62	7.85
P		22.60	5.48	3.00	19.32	7.64
K		26.69	2.71	1.88	2.81	1.64
Ca		4.83	15.66	13.31	9.22	11.95
Mg		6.26	4.86	2.82	2.55	1.59
Na		4.60	0.54	0.40	0.70	0.44
Cr	$mg \cdot kg^{-1}$ d.m.	6.07	19.74	10.25	37.88	17.47
Zn		531	899	488	1684	821
Pb		3.99	65.9	38.2	29.4	17.5
Cu		338.0	78.3	40.6	119.4	51.8
Cd		1.28	2.71	1.45	2.25	1.03
Ni		11.74	13.32	7.14	25.36	12.07
Hg		*	3.58	1.80	2.29	1.07

*trace

1.48 g $K \cdot pot^{-1}$ as KCl. In the second and third year of the experiment, because of the subsequent fertilizer effect and the abundance of the soils in bioavailable phosphorus and potassium, the applied doses were as follows: 0.80 g N; 0.2 g P and 1.40 g $K \cdot pot^{-1} \cdot year^{-1}$ as chemically pure salts.

Maize cv. San (FAO 240) was cultivated as a test plant, with five plants per pot left to grow. Maize (grown for green forage) was harvested at the 7-9 leaves stage. The growing season lasted 47 days in the first year, 66 days in the second year and 54 days in the third year. Throughout the experiment the plants were watered with distilled water to 50% of maximum water capacity. Once harvested, the plants were dried (at 70°C) to constant weight and the yield of dry mass of shoots and roots was determined. After crushing in a laboratory mill, the plant material was mineralized for 5 hours in a muffle furnace at 450°C. The remains were dissolved in diluted nitric acid 1:2 (v/v) (Ostrowska et al. 1991). The soil material collected each year

Table 2

Some properties of soils before the establishment of the experiment

Specification			Soil		
			(psg)	(gpp)	(gśp)
Granulometric composition Ø	1.0-0.1 mm	%	78	42	28
	0.1-0.02 mm		13	33	29
	< 0.02 mm		9	25	43
pH KCl			6.21	5.69	5.30
Hydrolitic acidity		$mmol(+) \cdot kg^{-1}$ d.m	11.2	23.4	33.2
Sum of alkaline cations			39.9	86.8	128.4
Total N		$g \cdot kg^{-1}$ d.m	0.96	1.25	1.72
Organic C			9.37	13.36	17.68
Total S			0.16	0.28	0.32
Available forms					
P		$mg \cdot kg^{-1}$ d.m.	79	217	29
K			166	359	138
Mg			134	154	126
$S\text{-}SO_4$			13.4	11.9	11.4

after the growing season was analyzed to determine changes in the physicochemical properties caused by the fertilization. In the dried material sifted through a 1mm mesh sieve, bioavailable potassium was determined using Egner-Riehm method (Ostrowska et al. 1991). In the plant material solutions and soil extracts, potassium was determined with the flame photometry method (FES) on a Philips PU 9100X apparatus. Plant reference material NCS DC73348 (China National Analysis Center for Iron & Steel) and soil reference material AG-2 (*Agro*MAT) were attached to each analytical series. The results were verified statistically using a fixed model, in which soil or fertilization was the factor. The statistical computations involved one-way ANOVA and the significance of differences was estimated using NIR Fisher test at significance level $p < 0.05$ (Stanisz 1998).

RESULTS AND DISCUSSION

The organic materials used for the experiment differed in the chemical composition, including potassium content, which was very small in sewage sludge and its mixtures with peat (Table 1). Peat added to sludge generally diminished the contents of individual elements in comparison with their concentrations in sludge alone.

The soil material used for the experiment belonged to various texture groups. In addition, it significantly differed in the chemical properties, including the content of available potassium forms (Table 2). A considerably high potassium content was assessed in the medium soil.

Yields of maize biomass (roots and shoots), as averages for all the treatments in the three years, were significantly smaller on the light soil (psg) (over 20% irrespective of the plant part) than the yields on the other two (heavier) soils (Figure 1). The difference in the yield harvested from sandy loam (gpp) and medium loam (gśp) was not significant for the shoots, where-

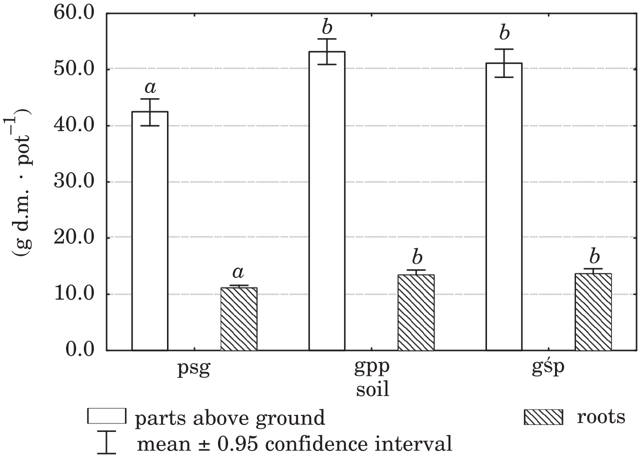

Fig. 1. Yields of aerial parts and roots of maize from fertilization objects (3-year period mean). Means followed by the same letters did not differ significantly at $p < 0.01$ according to Fisher test

as the average root biomass yield did not differ at all between these soils. Analysis of variance confirmed a favourable effect of organic fertilization on maize biomass yield (Table 3). Treatment with sewage sludge or its mixtures with peat as well as fertilization with farmyard manure produced much higher yields than exclusive mineral fertilization. Larger yields from the mineral salt treatments occurred only in the first year of the experiment (Figure 2). In the subsequent years, the response to fertilization with organic materials was positive despite the differences noticed on the lighter soils, which obscured the overall effect. Larger yields were produced when sewage sludge and peat mixture was applied compared to sludge alone. Fertilization efficiency of organic materials is determined mainly by their nitrogen content, particularly nitrogen mineral forms (SZULC et al. 2004). Nitrogen largely determines biomass yield. However, limited availability of other fer-

Table 3

Yields of dry mass of parts above ground and roots plants, total potassium content in maize and potassium available in soils (3-year period mean)

Object	Yield of biomass		K in the plant		K available in the soil
	(g d.m.·pot^{-1})		(g K·kg^{-1} d.m.)		(mg K·kg^{-1} d.m.)
	(Cz.n.)	(K)	(Cz.n.)	(K)	
Control (0)	22.1 ^{a}	7.4 ^{a}	17.5 ^{a}	9.9 ^{a}	80 ^{a}
NPK	42.5 ^{b}	10.1 ^{b}	37.5 ^{d}	18.9 ^{c}	127 ^{bc}
FYM (OB)	48.0 ^{c}	13.1 ^{cd}	30.5 ^{c}	16.8 ^{bc}	143 ^{c}
Sewage sludge (OŚA)	47.3 ^{c}	11.8 ^{c}	29.4 ^{bc}	16.8 ^{bc}	109 ^{abc}
Sewage sludge+peat (MOŚA)	49.3 ^{cd}	11.8 ^{c}	29.1 ^{bc}	15.9 ^{b}	116 ^{abc}
Sewage sludge (OŚB)	50.4 ^{cd}	14.1 ^{de}	28.7 ^{bc}	15.4 ^{b}	99 ^{ab}
Sewage sludge+peat (MOŚB)	55.6 ^{d}	15.8 ^{e}	25.6 ^{b}	15.6 ^{b}	108 ^{abc}

(Cz.n.) – aerial parts; (K) – roots
Means followed by the same letters in columns did not differ significantly at $p < 0.01$ according to Fisher test.

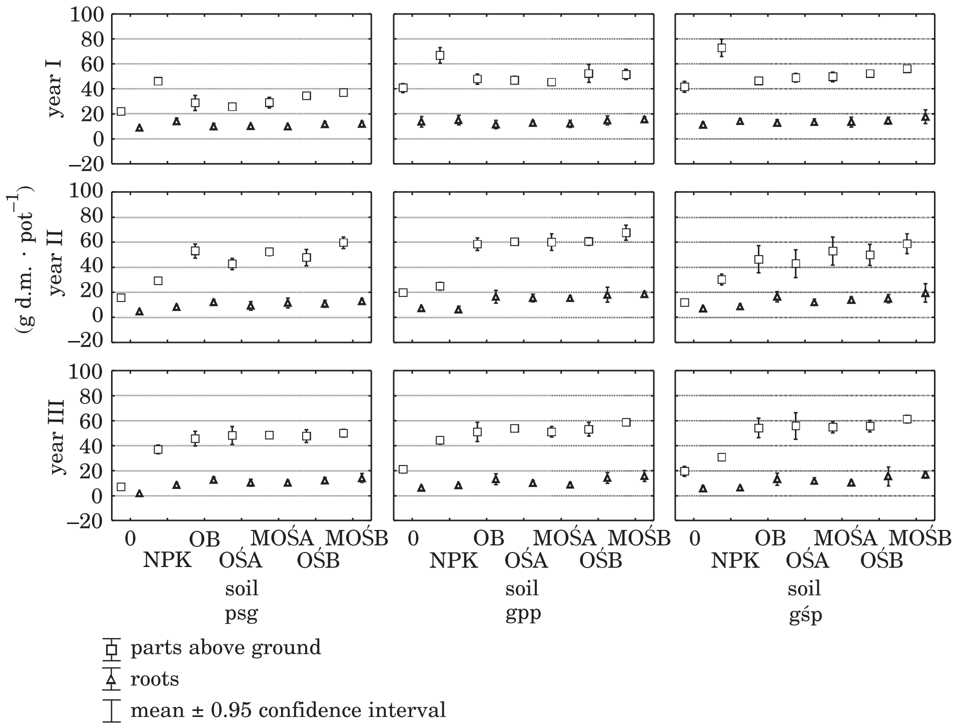

Fig. 2. Yield of aerial parts and roots of maize in each year of the experiment

tilizer components, such as potassium, directly influences mineral economy in crops. Over the three-year period of our investigations, fertilization with organic materials produced better results expressed by the biomass yields than treatments with mineral salts. This effect is not fully attributable to the activity of the applied sewage sludge or its mixtures with peat. It is also a subsequent effect of the activity of organic materials and supplementary fertilization with mineral salts in the second and third year of the experiment. Plant yields might have been affected by other components, such as sulphur, magnesium or microelements supplied to soil with organic materials, whose amounts were not balanced. According to WOŁOSZYK (2003) application of natural or organic fertilizer, due to the so-called subsequent effect, does not always raise crop yields. DRAB and DERENGOWSKA (2003) demonstrated a positive effect of sewage sludge on crop yielding while proving that yield, irrespective of the soil type, was conditioned by a dose of sewage sludge. According to WIATER et al. (2004), immediate fertilizer effect of sewage sludge on maize yield may be worse than that produced by mineral fertilization, although the subsequent effect of sewage sludge fertilizer activity could be more beneficial.

Irrespective of the type of soil and year of the experiment, the highest amounts of potassium were found in shoots and roots of maize fertilized with mineral salts (Table 3, Figures 3 and 4). The mean potassium content for the three years in this treatment was 37.5 g $K \cdot kg^{-1}$d.m. in shoots, thus being over 7 $g \cdot kg^{-1}$ d.m. larger than the content determined in shoots of plants fertilized with organic materials. Smaller differences occurred in the

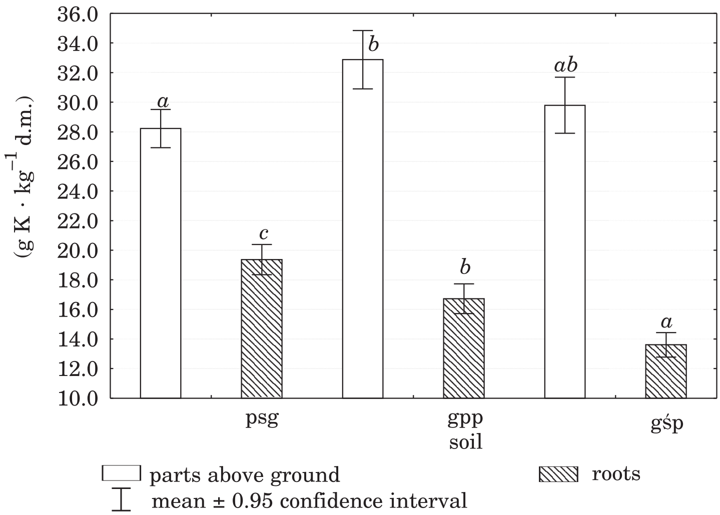

Fig. 3. Content of potassium in aerial parts and roots of maize from fertilization objects (3-year period mean). Means followed by the same letters did not differ significantly at $p < 0.01$ according to Fisher test

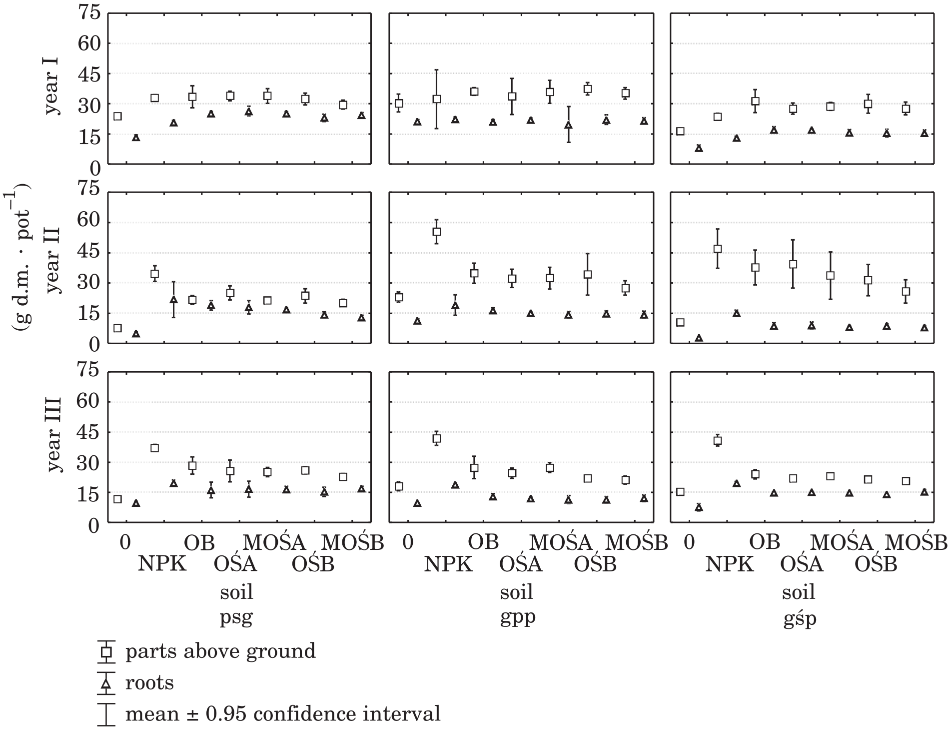

Fig. 4. Content of potassium in aerial parts and roots of maize in each year of the experiment

potassium content in roots. Significant variation was also noticed in the potassium content in maize biomass depending on the kind of soil (Figure 3). The content was the highest in sandy silt loam (gpp), but the average potassium content for all the treatments and years did not differ significantly in biomass of maize cultivated on weakly loamy sand (psg) and medium silt loam (gśp). This effect was certainly produced by the content of bioavailable potassium in soil. Application of macroelements as mineral fertilizers usually affects their concentrations in plants to a greater degree than fertilization with these elements in the form of natural and organic fertilizers or waste materials. It is so mainly because of the form in which the elements occur in these fertilizers as well as due to their occasionally low content (Kalembasa et al. 2001). According to Kalembasa and Symanowicz (1999), sewage sludge has a beneficial effect on the content of macroelements in plants. Wołoszyk (2003) claims that even when sewage sludge contains little potassium, it is not always necessary to supplement this component in the mineral form, and this depends on soil abundance in bioavailable potassium forms and the kind of crop. Drab and Derengowska (2003) revealed that even if nitrogen and phosphorus content in plants increased proportionately to the applied dose of sewage sludge, potassium content in the plant biomass did not change much, also when an additional dose of this element in the mineral form was used. On the other hand, WIATER et al. (2004) demonstrated a subsequent effect of fertilization with sewage sludge granulate on potassium content in maize.

The mean content of bioavailable potassium in the soil from all the treatments differed significantly (Figure 5). The highest amount of bioavailable forms of this element was determined in the soil amended with farmyard manure, whereas the lowest level of bioavailable potassium, apart from

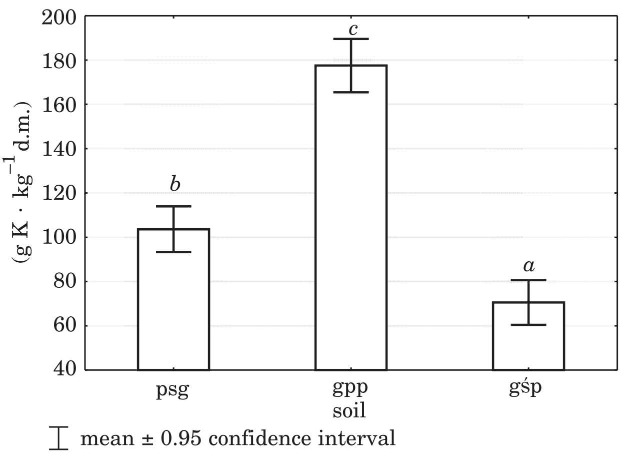

Fig. 5. Content of available potassium forms in soils from fertilization objects (3-year period mean). Means followed by the same letters did not differ significantly at $p < 0.01$ according to Fisher test

the control treatments, occurred after application of sewage sludge. Considerable variation was also observed in the content of bioavailable potassium forms in the soil in individual years (Figure 6). The research reveals that on average the content of bioavailable potassium in soil after the application of organic materials, particularly sewage sludge (OŚA and OŚB), was smaller in the soil amended with mineral salts (NPK) (Table 3). However, the analysis of variance did not confirm the significance of differences. The highest mean content of bioavailable potassium in soil, in comparison with the unfertilized treatment, was found after farmyard manure application (OB). Likewise, WOŁOSZYK (2003), who tested composts and sewage sludge, noticed that the content of bioavailable potassium forms diminished in comparison with the bioavailable forms of this element in soil prior to the experiment.

According to MCLEAN and BRYDON (1971), small doses of potassium do not always ensure proper plant supply with this element and the causes of this phenomenon are connected with unchangeable sorption of potassium ions.

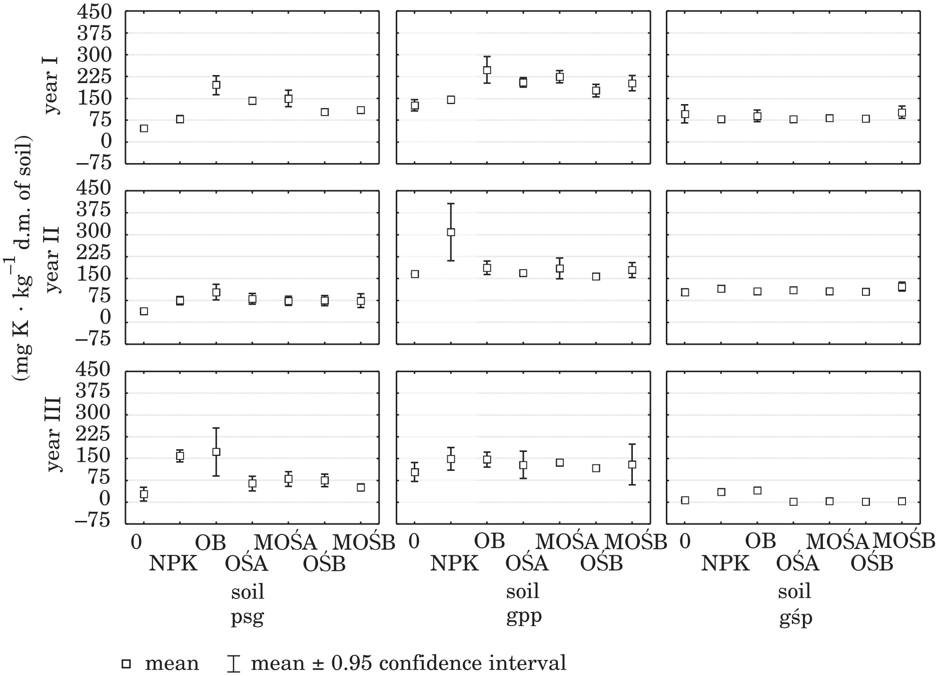

Fig. 6. Content of available potassium forms in soils in each year of the experiment

CONCLUSIONS

1. Fertilization with sewage sludge and mixtures of sludge with peat affected maize yields more favourably than mineral salt treatment.

2. Sludge mixtures with peat (in comparison with sludge used separately) influenced slightly more advantageously maize biomass yield and comparative potassium content in the plant biomass.

3. In comparison with the organic materials applied to the soil, mineral salt fertilization significantly increased potassium content in maize biomass.

4. Equalizing potassium quantities supplied with the organic materials by mineral salts and its balancing did not increase the soil abundance in bioavailable potassium in comparison with the initial abundance, although various changes were observed in the soils after the applied fertilization.

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EFFECT OF SELENIUM ON SELECTED MACRONUTRIENTS IN MAIZE PLANTS

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Abstract

Selenium is an essential element for humans, animals and some species of microorganisms. In higher plants, however, the role of selenium is still unclear. Because selenium enrichment may influence the nutrient balance of plants, a study was done to test the effects of selenite-Se ($Na_2SeO_3 \cdot 5H_2O$) on selected macronutrients content in maize (*Zea mays* L. var. *saccharata* Kcke. cv. Złota Karłowa) seedlings. Plants were grown in Hoagland I nutrient solution (pH 6,2) amended with selenite at 0 (control), 5, 25, 50 and 100 $\mu mol \cdot dm^{-3}$ for 14 days. The dry weight of the shoots was then analyzed for phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg) content. Phosphorus and calcium content increased, while potassium content decreased with increasing selenium treatments. No significant differences were found for magnesium level. Plant growth was affected by excessive selenium concentration. At low concentration (5 $\mu mol \cdot dm^{-3}$), selenium tended to stimulate the plant growth and the root elongation but at higher concentrations (50 and 100 $\mu mol \cdot dm^{-3}$) the dry mass accumulation and root tolerance index severely decreased.

The study revealed that disturbances of growth and reduction of plant's biomass at the presence of high selenium concentrations in the nutrient solution may have resulted from the disturbance of mineral balance of plants, namely accumulation of large amounts of calcium and phosphorus in shoot tissues.

Key words: selenium, maize, chemical composition.

WPŁYW SELENU NA ZAWARTOŚĆ WYBRANYCH MAKROELEMENTÓW W ROŚLINACH KUKURYDZY

Abstrakt

Selen jest pierwiastkiem niezbędnym dla ludzi, zwierząt oraz niektórych mikroorganizmów, jednak jego rola w roślinach wyższych nie jest w pełni poznana. Ponieważ wzbogacenie podłoża w selen może wpływać na równowagę mineralną roślin, przeprowadzono ba-

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dania dotyczące wpływu selenu w formie seleninu ($Na_2SeO_3 \cdot 5H_2O$) na zawartość wybranych makroelementów w siewkach kukurydzy (*Zea mays* L. var. *saccharata* Kcke. cv. Złota Karłowa). Rośliny rosły przez 14 dni w pożywce Hoaglanda I (pH 6,2) wzbogaconej w selen, który stosowano w następujących stężeniach: 0 (kontrola), 5, 25, 50 i 100 µmol·dm^{-3}. Po tym czasie w suchej masie organów nadziemnych analizowano zawartość fosforu (P), magnezu (Mg), potasu (K) i wapnia (Ca). Stwierdzono, że wraz ze wzrostem stężenia selenu w pożywce, w organach nadziemnych roślin wzrastała zawartość fosforu i wapnia, malała zaś zawartość potasu. Nie wykazano istotnych różnic w zawartości magnezu. Wprowadzenie do podłoża selenu wpływało również na wzrost roślin. Stwierdzono, że selen w niskim stężeniu (5 µmol·dm^{-3}) stymulował wzrost siewek oraz elongację systemu korzeniowego, ale w jego wyższych stężeniach tego pierwiastka (50 i 100 µmol·dm^{-3}) akumulacja biomasy oraz indeks tolerancji korzenia drastycznie malały. Z przeprowadzonych badań wynika, że zaburzenia wzrostu oraz redukcja biomasy roślin w obecności wysokich stężeń selenu w podłożu może być związana z zakłóceniem równowagi mineralnej roślin, a zwłaszcza z gromadzeniem w tkankach pędów znacznej ilości wapnia i fosforu.

Słowa kluczowe: selen, kukurydza, skład chemiczny.

INTRODUCTION

Selenium – an element chemically similar to sulfur – is a micronutrient essential for people, animals and some microorganisms. Traces of selenium play a principal role in cellular metabolism, namely in anti-oxidation reactions. However, selenium has quite a narrow spectrum of physiological action because its deficiency as well as excess exert negative effects on functions of an organism (Sembratowicz, Grela 1997).

Although all plants are able to take up and metabolize selenium, the assumption about its necessity for higher plants has not been fully confirmed yet. Plants differ in their ability to accumulate selenium in tissues, therefore they were divided into species capable of active and specific selenium accumulation (selenium accumulators) and those which cannot accumulate selenium (Adriano 1986).

Accumulation of high selenium amounts in tissues of plants that have no capability of its accumulation, inducing toxicity of the element, is strictly associated with non-specific incorporation of selenocytein and selenomethionin into proteins instead of cystein and methionin, respectively. It was found that the bond formed between selenium atoms in selenocystein is longer, weaker and more labile than disulfide binding, which may have influence on slight changes in the third-order structure of proteins and evoke disturbances of catalytic functions of enzymes (Mazzafera 1998, Terry et al. 2000). Replacing cystein by selenocystein in proteins may also disturb formation of disulfide bridges. Furthermore, selenium has a negative effect on protein synthesis because selenomethionin appearing instead of methionin probably impairs formation of peptide binding (Terry et al. 2000).

It is suggested that interaction with general minerals is one of more important factors due to which trace elements affect the plant's metabolism. Presence of heavy metals may induce deficiency of macronutrients and micronutrients necessary for a proper course of principal life processes (Baranowska-Morek 2003). Interaction between selenium and a given element depends on quantitative proportions and it may also cause synergistic effects (Pyrzyńska 2000). Selenium – like heavy metals – can modify uptake and accumulation of minerals which are important for metabolism (Kopsell et al. 2000, Pazurkiewicz-Kocot et al. 2003). Moreover, positive influence of selenium on changes in the activity and permeability of the cellular membrane has been found, and this may be one of the earliest symptoms of the influence of selenium on plants (Kinraide 2003).

The present experiments were aimed at evaluating the influence of selenium on plant growth and content of selected macronutrients (P, K, Mg, Ca) in maize seedlings growing at the presence of varied sodium selenate concentrations.

MATERIALS AND METHODS

The experiments were carried out in water cultures and their purpose was to investigate the influence of selenium on contents of selected macronutrients in maize plants (*Zea mays* L. var. *saccharata* Kcke. cv. Złota Karłowa). Seeds obtained from the Seed Center in Lublin were placed on wet filter paper 1 cm from the upper edge, covered with a wet filter band and rolled up. The tubes thus prepared were placed vertically in beakers filled with distilled water and left for 7 days at 25°C. Afterwards, healthy and well-developed seedlings were placed in glass jars of 1 dm^3 capacity (two in each jar) filled with 1.5-fold concentrated Hoagland I nutrient solution (pH 6.2). After 3 days, selenium in the form of sodium selenate ($Na_2SeO_3 \cdot 5H_2O$) was introduced at following concentrations: 0 (control), 5, 25, 50, or 100 $\mu mol \cdot dm^{-3}$. The maize plants grew in an air-conditioned plant growth room under controlled photon density stream within the photosynthetically active range of 270 $\mu mol \cdot m^{-2}\ s^{-1}$ at a photoperiod of 14/10 h and temperature 25/20°C (day/night).

After 14 days, the index of tolerance (IT), which defines the increase of root length of experimental plants expressed as per cent relative to control, was determined (Mc Neilly 1994). Also dry matter of roots and shoots was determined. Dry plant material was subjected to chemical analyses for determination of the content of the following macronutrients in aerial plant organs: phosphorus by vanadium-molybdate colorimetry; magnesium by colorimetry using titanium yellow; potassium and calcium by the AAS technique (Nowosielski 1974). The experiment was performed in three time-independent repetitions. In all the experiments, the least small difference (LSD) between the means was calculated using Tukey's test. Differences at $P = 0.05$ were considered as statistically significant.

RESULTS AND DISCUSSION

Due to chemical similarity of selenium to sulfur, most of the references deal with interaction between these two elements. It was found that plants take up, transport and metabolize sulfur and selenium using similar mechanisms. It is a common opinion that both anions are transported by sulfur carriers in cellular membranes, therefore the presence of sulfur may affect selenium accumulation and vice versa (TERRY et al. 2000, ELLIS and SALT 2003). However, selenium influence on other than sulfur macronutrients is a subject of only small number of studies and their results are not always univocal.

The current experiment revealed that introduction of selenium into the medium significantly interacted with the chemical composition of aerial parts of maize plants (Table 1). Phosphorus and calcium in dry matter manifested an increasing tendency along with an increase in the selenium concentration in the nutrient solution. Amounts of phosphorus slightly increased at the presence of 5 and 25 μmol Se·dm^{-3}; however, the differences were not statistically significant. Selenium introduced into the medium at the levels of 50 and 100 μmol·dm^{-3} caused about a 4-5-fold increase of phosphorus content in dry matter in relation to the control. Similar dependence was found for calcium. Its content in shoots was much elevated at the presence of 50 and 100 μmol Se·dm^{-3}, exceeding by about 5-6-fold the control value. WU and HUANG (1992), in their experiments on selenium affecting accumula-

Table 1

Influence of selenium on some macronutrients content in maize shoots
(mean of tree replications)

Selenium concentration in nutrient solution (μmol·dm^{-3})	(% d. w.)			
	Ca	K	Mg	P
0 (control)	0.280^{a}	3.705^{b}	0.275	0.375^{a}
5	0.340^{a}	4.210b^{c}	0.290	0.430^{a}
25	0.305^{a}	4.550^{c}	0.295	0.505^{a}
50	1.615^{b}	3.400^{ab}	0.340	1.835^{b}
100	1.340^{b}	2.830^{a}	0.340	1.450^{b}
LSD $_{0.05}$	0.413	0.831	n.s.	0.546

Means in each column followed by the same letter are not significantly different.

tion of macro- and micronutrients in fescue and clover, recorded similar dependencies regarding calcium concentrations, while contrary results were achieved for phosphorus. In experiments conducted by Kopsell et al. (2000), selenium did not have any effect on calcium accumulation; instead it caused a decline in phosphorus in cabbage plants. Singh and Malhotra (1976) as well as Mikkelsen et al. (1989) observed increased phosphorus content in plants cultivated at the presence of selenium.

Our analysis of potassium content revealed that at the presence of 25 μmol Se·dm^{-3}, the level of this macronutrient increased by 23%, while after adding 100 μmol Se·dm^{-3} into the nutrient solution, its content decreased by 24% as compared to the control. Other selenium rates did not exert significant influence on potassium contents in aerial parts of maize plants. Positive effects of selenium in the form of selenite on potassium accumulation were also observed by Pazurkiewicz-Kocot et al. (2003), who found that the content of potassium in maize leaves significantly increased when introducing 10 μmol Se·dm^{-3} into the medium, but a contrary dependence was recorded in roots. Kopsell et al. (2000) revealed that the potassium level in cabbage leaves increased linearly along as the selenium concentration in the medium rose.

Our analysis of the influence of various selenium concentrations on magnesium accumulation indicated that the content of the latter did not differ considerably as compared to the control plants. Arvy et al. (1995) achieved similar results when analyzing effects of selenium in forms of selenate and selenite on ionic balance of suspended cultures of *Catharanthus roseus* L.

Influence of selenium on plants largely depends on its chemical form and its concentration in nutrient solution. Our own studies revealed a stimulating effect of selenium at the concentration of 5 μmol·dm^{-3} on plant growth (Table 2). Dry matter of the maize roots and aerial organs exceeded the control by 79% and 62%, respectively, at the presence of 5 μmol·dm^{-3} selenium. Also slight stimulation of root system elongation was observed under these conditions (IT = 110%). Selenium concentrations above 25 μmol·dm^{-3} resulted in a dramatic biomass decrease, which resulted mainly from less developed aerial organs (Table 2). After introducing selenium into the medium at the rate of 100 μmol·dm^{-3}, root dry matter was lower by 56%, and that of aerial parts by 78% in relation to the control. The two highest selenium concentrations caused reduction of root growth potential. The IT of roots reached only 68% and 57% of the control values at the presence of 50 and 100 μmol Se·dm^{-3}, respectively.

The results achieved by Hartikainen et al. (2000) also confirm the fact that selenium interaction with plants depends on its concentration. At lower rates, selenium stimulated growth of ryegrass seedlings, while at high doses it acted as pro-oxidant reducing yields and inducing metabolic disturbances. Later experiments by Xue et al. (2001) on lettuce seedlings confirmed positive influence of low selenium concentrations on yielding of the species;

Table 2

Influence of selenium on dry weight and IT of maize plants
(mean of tree replications)

Selenium concentration in nutrient solution ($\mu mol \cdot dm^{-3}$)	Dry weight (g per jar)		Roots IT
	roots	shoots	
0 (control)	0.831^{b}	2.899^{c}	100.0^{a}
5	1.485^{c}	4.686^{d}	110.2^{a}
25	0.792^{b}	2.389^{c}	91.45^{a}
50	0.469^{a}	1.184^{b}	67.67^{b}
100	0.367^{a}	0.649^{a}	56.73^{b}
$LSD_{0.05}$	0.160	0.471	18.64

Means in each column followed by the same letter are not significantly different.

however, the effect depended on plant's development stage. Phytotoxicity of high selenium levels is associated mainly with non-specific replacement of sulfur amino acids by their selenium analogues (TERRY et al. 2000). Moreover, this effect may result from disturbances in mineral balance, which is indicated by analysis of chemical composition of maize shoots performed in the present experiment. It was found that the largest differences in the content of the macronutrients we determined, as compared to control plants, were closely connected with a considerable decrease in the dry matter of seedlings and reduction of root growth potential (Tables 1, 2).

CONCLUSIONS

1. Contents of macronutrients in aerial parts of maize depended on a selenium concentration in the nutrient solution. Selenium at concentrations of 50 and 100 $\mu mol \cdot dm^{-3}$ caused a significant increase in phosphorus and calcium. Potassium significantly increased at the presence of 25 µmol $Se \cdot dm^{-3}$, while decreasing under the influence of 100 µmol $Se \cdot dm^{-3}$. Presence of selenium in the medium did not have significant influence on magnesium.

2. Selenium at 5 $\mu mol \cdot dm^{-3}$ concentration stimulated maize seedling growth, while rates 50 and 100 $\mu mol \cdot dm^{-3}$ dramatically reduced dry matter of plants and inhibited the root system elongation.

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EFFECT OF SULFUR ON THE QUALITY OF WINTER RAPE SEEDS

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Abstract

The paper presents the results of three-year (2000-2003) experiments to determine the effect of the date of sulfur application (fall + spring or spring only) and the fertilizer rate (0, 30, 60, 90 kg·ha^{-1}) on the usability of winter rape seeds.

The pre-sowing and the spring rates of sulfur as well as the rate applied entirely in the spring did not lead to significant variations in crude fat concentrations, but a clear tendency towards an increase in the total protein content was observed in rape seeds. A one-way increase was reported to the level of 60 kg S·ha^{-1}.

An increase in the sulfur rate (regardless of the date of application) to 90 kg·ha^{-1} raised the concentrations of alkene glucosinolates, mainly gluconapin and progoitrin, and – to a lesser degree – of indole glucosinolates, mainly 4-hydroxyglucobrassicin, in rape seed cv. Lisek.

The splitting of the total sulfur rate into two rates (fall + spring) caused a higher increase in the concentrations of gluconapin, progoitrin and 4-hydroxyglucobrassicin than a single rate application in the spring.

Key words: winter rape, sulfur fertilization, crude fat, total protein, gluosinolates.

SIARKA A JAKOŚĆ NASION RZEPAKU OZIMEGO

Abstrakt

W pracy przedstawiono 3-letnie (2000-2003) wyniki badań nad wpływem terminu aplikacji siarki (jesień + wiosna lub tylko wiosna) i poziomu dawki (0, 30, 60, 90 kg·ha^{-1}) na wartość użytkową nasion rzepaku ozimego.

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Siarka aplikowana przedsiewnie i wiosną oraz wyłącznie wiosną nie różnicowała istotnie zawartości tłuszczu surowego, powodując jednak wyraźną tendencję do wzrostu zawartości białka ogółem w nasionach. Przyrost ten był jednokierunkowy do dawki 60 kg S·ha^{-1}.

Zastosowanie siarki (niezależnie od terminu zabiegu) aż do dawki 90 kg na 1 ha powodowało w nasionach odmiany Lisek przyrost zawartości głównych glukozynolanów alkenowych – glukonapiny i progoitryny, a w mniejszym stopniu indolowych, tj. 4-hydroksyglukobrassicyny.

Warto podkreślić, iż podział dawki siarki na cześć jesienną i wiosenną w większym stopniu wpływał na zwiększenie zawartości progoitryny, glukonapiny oraz 4-hydroksyglukobrassicyny niż jednokrotna wiosenna aplikacja tego składnika.

Słowa kluczowe: rzepak ozimy, nawożenie siarką, tłuszcz surowy, białko ogółem, glukozynolany.

INTRODUCTION

Sulfur fertilization increases the total fat content of rape seeds (Booth et al. 1991ab, Rotkiewicz et al. 1996, Jankowski 2007). Sulfur increases the concentration of essential unsaturated fatty acids in oil and enhances the usability of rapeseed oil (Krauze, Bowszys 2000).

Sulfur deficiency usually results in a lower quantity and quality of protein by reducing the levels of exogenous amino acids, which are an important diet component (Schnug, Haneklaus 1995, Zhao et al. 1997, Wielebski, Wójtowicz 2000, Jankowski 2007). Contrary results were reported in a study conducted by Rotkiewicz et al. (1996), where the application of 80 kg S·ha^{-1} lowered (by 0.2-0.8%) the total protein content in winter rape seeds. Sulfur fertilization may increase the level of glucosinolates in defatted seed residues (Withers et al. 1995, Bilsborrow et al. 1995, Budzyński, Ojczyk 1995, Rotkiewicz 1996, Wielebski, Muśnicki 1998ab, Fimes et al. 2000, Jankowski 2007), but it is less likely to induce such an increase in rapeseed oil (Niewiadomski 1984). Due to sulfur fertilization, the total concentrations of glucosinolates in low-erucic acid double-zero („00") rape varieties may increase by 20-30% (Rotkiewicz et al. 1996, Wielebski, Wójtowicz 2003, Jankowski 2007) and up to 80% (Wielebski, Wójtowicz 2003). Sulfur fertilization stimulates the biosynthesis of alkene glucosinolates (Zhao et al. 1995, Rotkiewicz et al. 1996, Wielebski 1997), mainly progoitrin and gluconapin (Wielebski 1997) as well as glucobrassiconapin (Rotkiewicz et al. 1996). This is because sulfur stimulates mainly the synthesis of methionine, the precursor of alkene glucosinolates (Zhao et al. 1994). At low sulfur fertilization rates, the concentration levels of those metabolites may decrease due to a heightened activity of myrosinase or a drop in the levels of available methionine (Bones et al. 1994).

The objective of this study was to determine the effect of a fertilizer rate (0, 30, 60 and 90 kg·ha^{-1}) and the date of sulfur application on the content of total protein and crude fat, the concentration levels and the structure of glucosinolates in winter rape seeds.

METHODS

The study presents the results of three-year strict field and laboratory experiments conducted at the Research and Experimental Station of the University of Warmia and Mazury, located in Bałcyny (N = 53°35′49″ E = 19°51′20.3″). Field experiments were performed in a randomized split-plot design, with four replications:

factor I – sulfur rate ($kg \cdot ha^{-1}$): (Ia) control without S fertilization; (Ib) 30; (Ic) 60; (Id) 90;

factor II – date of sulfur application:
(IIa) fall (BBCH – 00) and spring (BBCH – 30),
(IIb) spring only (BBCH – 30).

IIa	BBCH - 00	0	15	30	30
	BBCH - 30	0	15	30	60
IIb	BBCH - 30	0	30	60	90

The experiments were established on typical lessive silty soil developed from light loam, of quality class IIIa representing a very good rye complex (2000 and 2001) or a good wheat complex (2002) – Table 1. The content of available phosphorus was evaluated as average to very high, potassium – as average, magnesium and sulfate sulfur – as low. Topsoil was acidic

Table 1

Description of soil conditions

Specification	Growing season		
	2000/2001	2001/2002	2002/2003
Soil type	typical lessive soil		
Soil species	light loam		
Soil pH (1 M KCl)	5.3	5.9	5.9
Soil quality class	IIIa		
Soil suitability complex	very good rye complex		good wheat complex
Content of available nutrients ($mg \cdot kg^{-1}$ soil)			
– P	62.0	114.8	89.9
– K	107.9	103.8	207.5
– Mg	41	83	57
– $S\text{-}SO_4$	18.1	15.2	6.7

or slightly acidic (pH 5.3-5.9 in 1 M KCl). The rape forecrop were grain crops – spring barley (2000 and 2002) and winter wheat (2001). The experimental plot covered an area of 18 m^2.

A pre-sowing rate of 35 kg $P \cdot ha^{-1}$ in the form of triple superphosphate and 100 kg $K \cdot ha^{-1}$ in the form of highly concentrated potash salt were applied. The autumn pre-sowing N rate was 20 $kg \cdot ha^{-1}$. The spring nitrogen rate (160 $kg \cdot ha^{-1}$) was applied in two parts – 100 $kg \cdot ha^{-1}$ at stage BBCH 30 and 60 $kg \cdot ha^{-1}$ at stage BBCH 51. Nitrogen was applied in the form of ammonium nitrate (-S) or ammonium sulfate and ammonium nitrate (+S) as fertilizer balancing the nitrogen and sulfur rate.

Dressed winter rape seeds cv. Lisek were sown between the 10^{th} and 20^{th} of August (2001, 2002) and in the last week of August (2000) in the quantity of 120 germinating seeds per m^2 of plot area, at spacing of 20 cm.

Dicotyledonous weeds were controlled with a post-sowing application of 999 $g \cdot ha^{-1}$ metazachlorine and 249 $g \cdot ha^{-1}$ of quinmerac, and monocotyledonous weeds – with 52 $g \cdot ha^{-1}$ of haloxyphop-R. Insecticide was applied after the level of insect infestation had exceeded the standards given by the Institute of Plant Protection in Poznań. During the spring growing season in the first and second year of the experiment, a single treatment for the control of the rape blossom beetle was applied (300 $g \cdot ha^{-1}$ chloropiryphos + + 30 $g \cdot ha^{-1}$ cypermetrine). During the third experimental cycle in the spring growing season, three treatments for the control of rape pests were applied (1 x cabbage curculio, 2 x rape blossom beetle) with the use of 7.5 $g \cdot ha^{-1}$ deltametrine, 7.5 $g \cdot ha^{-1}$ cypermetrine and 10 $g \cdot ha^{-1}$ alpha-cypermetrine. Fungicides were not applied due to low disease incidence. Rape was harvested using a two-stage method at the beginning or end of the second week of July.

Growth stages were determined with the use of the BBCH scale – Biologische Bundesanstalt, Bundessortenamt und Chemical Industry (MUŚNICKI, MRÓWCZYŃSKI 2006).

Seed samples (treatment average) were subjected to a chemical analysis to determine the levels of crude fat (by electromagnetic resonance) and nitrogen (by Kjeldahl method, expressed as crude protein, according to the Polish Standard PN-75/A-04018/Az3:2002). The concentrations of glucosinolates were estimated by high-performance liquid chromatography (HPLC), as described by HEANEY et al. (1986).

Chemical analyses of soil and seeds were performed at the laboratories of the Chemical and Agricultural Station in Olsztyn, Institute of Animal Nutrition and Fodder Science, Department of Plant Raw Materials Processing and Chemistry, University of Warmia and Mazury in Olsztyn and the Center for Excellence of the Institute of Animal Reproduction and Food Research at the Polish Academy of Sciences in Olsztyn. The results of chemical analyses were verified by an analysis of variance in accordance with the established experimental method. The mean values of the investigated parameters in all treatments were compared with the use of Duncan's test. LSD values were stated for 5% error. Analyses of variance were performed with the use of STATISTICA® applications, and the remaining calculations were carried out in EXCEL® spreadsheet.

RESULTS

Agrometeorological conditions

Total atmospheric precipitation from August to October 2000, 2001 and 2002 exceeded the multi-year precipitation average by 2 to 54%. The above is emphasized due to possible changes in sulfur translocation in soil. In the fall of 2000, intense precipitation was reported only in the pre-sowing period and at the beginning of germination (in August). In 2001, precipitation in excess of the average monthly levels was noted in August and September, and in 2002 – in all months of the fall growing season (Table 2).

Table 2

Moisture conditions during the autumn growing season (August – October)

Growing season	Precipitation (mm)				Percentage of long-term precipitation total			
	VIII	IX	X	Σ	VIII	IX	X	Σ
2000/2001	141	47	5	47	181	82	9	102
2001/2002	82	99	35	99	105	174	65	114
2002/2003	87	61	144	61	112	107	267	154

The winter dormancy period lasted from 153 to 172 days in subsequent years of field experiments (2000-2003). Although according to multi-year statistics, January is the coldest month in Ostróda Lakeland, the coldest month during the study was December (2001/2002, 2002/2003) and January in only one of the experimental years (2000/2001). The drop in mean daily air temperature (below 5°C) in the winter was always accompanied by snow cover (with a depth of 5-10 cm), which minimized the effect of low temperature on plant wintering.

The spring and summer growing seasons in 2001-2003 were marked by moderate precipitation, which covered the water demand of winter oilseed rape. The highest precipitation was reported in the spring and summer season of 2000/2001 and 2002/2003. The monthly distribution of precipitation was highly diversified in the studied years. Insufficient precipitation to cover the water requirements of plants was reported annually only in April (Table 3).

Table 3

Moisture conditions during the spring growing season (April – August)

Growing season	Precipitation (mm)					Percentage of long-term precipitation total				
	IV	V	VI	VII (first 20. days)	Σ	IV	V	VI	VII	Σ
Water demand (acc. to Klatt)										
–	45	70	75	30	220	129	124	110	37	91
Actual atmospheric precipitation (mm)										
2000/2001	44	31	49	110	234	126	54	72	167	107
2001/2002	10	90	73	37	210	29	158	107	53	98
2002/2003	24	79	61	83	247	69	139	90	146	117

CONTENT AND BIOLOGICAL YIELD OF CRUDE FAT

Sulfur fertilization had a non-significant effect on the crude fat content of rape seeds. In the first and third experimental cycle, sulfur fertilization had an adverse effect on crude fat concentration. Those years were characterized by relatively high precipitation in the period of rape ripening. The date of sulfur application (fall + spring, spring only) did not lead to significant variations in crude fat concentration levels in rape (Table 4).

Table 4

Crude fat content (% seed d.m.)

Growing season	Time of S application	S rate ($kg \cdot ha^{-1}$)				Mean
		0	30	60	90	
2000/2001	BBCH 00 and BBCH 30	43.1	43.0	41.5	43.0	42.7
	BBCH 30	43.1	41.9	41.7	42.6	42.3
2001/2002	BBCH 00 and BBCH 30	47.1	45.5	48.3	47.2	47.0
	BBCH 30	47.1	47.3	48.2	47.3	47.5
2002/2003	BBCH 00 and BBCH 30	46.0	46.0	44.9	45.9	45.7
	BBCH 30	46.0	45.5	46.6	44.8	45.7
2000/2001	–	43.1	42.5	41.6	42.8	42.5
2001/2002	–	47.1	46.4	48.3	47.3	47.3
2002/2003	–	46.0	45.8	45.9	45.4	45.8
–	BBCH 00 and BBCH 30	45.4	44.8	44.9	45.4	45.1
–	BBCH 30	45.4	44.9	45.7	44.9	45.2
Mean		45.4	44.9	45.3	45.2	–
LSD n.s.						

The biological yield of crude fat was determined by seed weight and the fat content of seeds. As a result of sulfur fertilization, biological yield increased by nearly 11% in the first year and by 4% in the two subsequent years of the experiment. The date of sulfur application had no influence on crude fat yield per hectare (Table 5).

Table 5

Biological yield of crude fat ($Mg \cdot ha^{-1}$)

Growing season	Time of S application	S rate ($kg \cdot ha^{-1}$)				Mean
		0	30	60	90	
2000/2001	BBCH 00 and BBCH 30	2.13	2.30	2.35	2.48	2.32
	BBCH 30	2.13	2.37	2.34	2.28	2.28
2001/2002	BBCH 00 and BBCH 30	2.02	2.08	2.12	2.09	2.08
	BBCH 30	2.02	2.09	2.13	2.08	2.08
2002/2003	BBCH 00 and BBCH 30	2.06	2.14	2.08	2.21	2.12
	BBCH 30	2.06	2.13	2.25	2.09	2.13
2000/2001	–	2.13	2.34	2.35	2.38	2.30
2001/2002	–	2.02	2.09	2.13	2.09	2.08
2002/2003	–	2.06	2.14	2.17	2.15	2.13
–	BBCH 00 and BBCH 30	2.07	2.17	2.18	2.26	2.17
–	BBCH 30	2.07	2.20	2.24	2.15	2.17
Mean		2.07	2.19	2.21	2.21	–
LSD: S rate – 0.070						

The data in Figure 1 indicate that crude fat yield per hectare increased up to the fertilization rate of 60 kg $S \cdot ha^{-1}$ on average in the studied years (Figure 1). Nevertheless, a statistically significant increment, in comparison with the control treatment, was reported only to the level of 30 kg $S \cdot ha^{-1}$ (Table 4).

CONTENT AND BIOLOGICAL YIELD OF PROTEIN

In comparison with the control, sulfur fertilization increased the total protein content of rape seeds in the first and third experimental cycle (Table 6). This increase varied over years and was observed only to the level of 60 kg $S \cdot ha^{-1}$. The date of sulfur application had only a minor effect (within statistical error limits) on total protein content (Table 6).

Regardless of the rate and date of application, sulfur fertilization increased the biological yield of protein by 190 $kg \cdot ha^{-1}$ in the first year and only by 10-60 $kg \cdot ha^{-1}$ in the remaining two years of the experiment (Table 7).

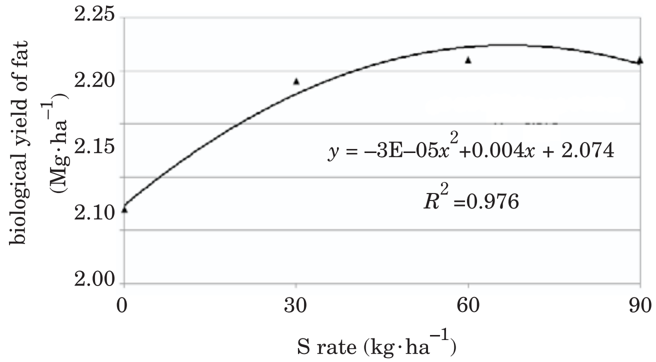

Fig. 1. Correlation between sulfur rate and the biological yield of crude fat, 2000-2003

Table 6

Total protein content (% seed d.m.)

Growing season	Time of S application	S rate (kg·ha^{-1})				Mean
		0	30	60	90	
2000/2001	BBCH 00 and BBCH 30	21.0	21.4	22.4	22.3	21.8
	BBCH 30	21.0	22.0	22.7	21.9	21.9
2001/2002	BBCH 00 and BBCH 30	21.1	20.4	21.1	21.2	21.0
	BBCH 30	21.1	21.0	21.1	21.1	21.1
2002/2003	BBCH 00 and BBCH 30	19.4	19.7	20.0	19.8	19.7
	BBCH 30	19.4	19.8	19.6	19.7	19.6
2000/2001	–	21.0	21.7	22.6	22.1	21.9
2001/2002	–	21.1	20.7	21.1	21.2	21.0
2002/2003	–	19.4	19.8	19.8	19.8	19.7
–	BBCH 00 and BBCH 30	20.5	20.5	21.2	21.1	20.8
–	BBCH 30	20.5	20.9	21.1	20.9	20.9
Mean		20.5	20.7	21.2	21.0	–
LSD n.s.						

Sulfur fertilization at the rate of 30 kg·ha^{-1} led to a significant increase in protein yield per hectare (by around 9% in comparison with control) in the experimental period. A successive increase in the sulfur rate (to 60 kg·ha^{-1}) resulted in an additional 3% growth in protein yield which remained within statistical error limits (Table 7). The data in Figure 2 show that the continued increase in sulfur rate (to 90 kg·ha^{-1}) did not affect the levels of this qualitative trait.

Table 7

Biological yield of total protein (Mg·ha)

Growing season	Time of S application	S rate (kg·ha^{-1})				Mean
		0	30	60	90	
2000/2001	BBCH 00 and BBCH 30	1.04	1.14	1.27	1.29	1.19
	BBCH 30	1.04	1.24	1.27	1.17	1.18
2001/2002	BBCH 00 and BBCH 30	0.90	0.93	0.92	0.94	0.92
	BBCH 30	0.90	0.93	0.93	0.93	0.92
2002/2003	BBCH 00 and BBCH 30	0.87	0.92	0.93	0.95	0.92
	BBCH 30	0.87	0.93	0.94	0.92	0.92
2000/2001	–	1.04	1.19	1.27	1.23	1.18
2001/2002	–	0.90	0.93	0.93	0.94	0.93
2002/2003	–	0.87	0.93	0.94	0.94	0.92
–	BBCH 00 and BBCH 30	0.94	1.00	1.04	1.06	1.01
–	BBCH 30	0.94	1.03	1.05	1.01	1.01
Mean		0.94	1.02	1.05	1.04	–
LSD: S rate – 0.065						

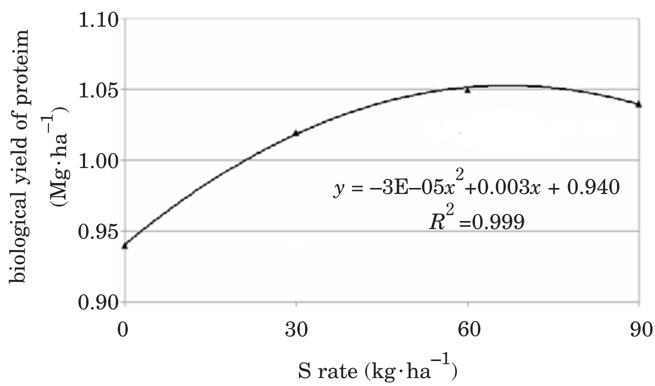

Fig. 2. Correlation between sulphur rate and the biological yield of total protein, 2000-2003

CONTENT AND COMPOSITION OF GLUCOSINOLATES

The total content of glucosinolates in rape seeds cv. Lisek was low. Sulfur fertilization at the rate of 30 – 60 – 90 kg·ha^{-1} gradually increased the accumulation of glucosinolates in defatted residues of winter rape seeds (Figure 3).

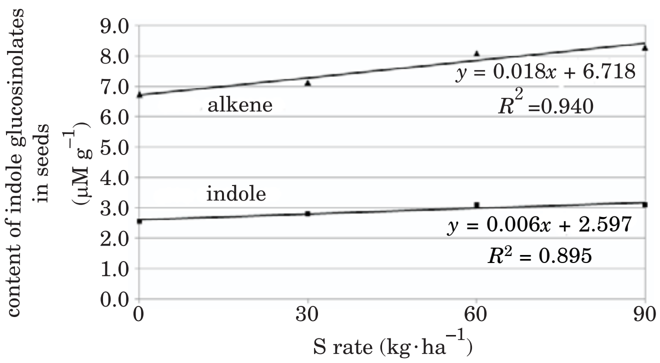

Fig. 3. Effect of sulfur rate on glucosinolates structure in winter rape seeds, 2000-2003

Sulfur stimulated the biosynthesis of alkene and indole glucosinolates to a similar extent (producing a similar ratio). The splitting of the total sulfur rate into two rates (fall + spring) had a more stimulatory effect on the accumulation of alkene and indole glucosinolates in defatted seed residues than a single rate application in the spring (Table 8).

Table 8

Effect of the rate and application time of sulfur on the total glucosinolate content of winter rape seeds (μM g^{-1} seeds), 2000-2003

Glucosinolates	S rate (kg·ha^{-1})						
	control	30		60		90	
		a+s	s	a+s	s	a+s	s
Alkene	6.74	7.43	6.82	8.30	7.90	8.45	8.15
Indole	2.55	2.88	2.73	3.12	3.07	3.10	3.09
Alkene / Indole	2.65	2.58	2.50	2.66	2.57	2.73	2.64

a+s – autumn + spring (BBCH 00 and BBCH 30)
s – spring (BBCH 30)

An increase in the content of alkene glucosinolates in winter rape seeds fertilized with sulfur was reported mostly in respect of gluconapin and progoitrin. In the group of compounds where tryptophan is the precursor, a particularly high increase was observed in 4-hydroxuglucobrassicin. The concentration of glucobrassicin increased to a lower degree, whereas the level of neoglucobrassicin remained unchanged (Table 9).

The date of sulfur application at particular rates did not affect the content of alkene and indole glucosinolates. A detailed analysis of the results

Table 9

Effect of the rate and application time of sulfur on the content of particular glucosinolates in winter rape seeds (μM g^{-1} seeds), 2000-2003

Glucosinolates		control	S rate (kg·ha^{-1})					
			30		60		90	
			a+s	s	a+s	s	a+s	s
Alkene	progoitrin	3.97	4.29	3.88	4.76	4.56	4.82	4.69
	sinigrin	0.06	0.06	0.06	0.07	0.07	0.07	0.07
	gluconapoliferin	0.13	0.15	0.15	0.19	0.18	0.19	0.18
	glucoalisin	0.20	0.21	0.24	0.27	0.22	0.29	0.25
	gluconapin	1.58	1.78	1.63	1.96	1.91	2.02	1.97
	glucobrassicanapin	0.80	0.94	0.86	1.05	0.96	1.06	0.99
Indole	4-hydroxyglucobrassicin	2.41	2.72	2.58	2.96	2.91	2.97	2.94
	glucobrassicin	0.09	0.11	0.10	0.11	0.11	0.08	0.10
	neoglucobrassicin	0.05	0.05	0.05	0.05	0.05	0.05	0.05

a+s – fall + spring (BBCH 00 and BBCH 30)
s – spring (BBCH 30)

obtained indicates that sulfur application in the spring resulted in lower concentrations of progoitrin, gluconapin and 4-hydorxyglucobrassicin in comparison with split dressing (fall + spring) (Table 9).

DISCUSSION

Deficiency of plant-available sulfur reduces the rate of protein biosynthesis (ZHAO et al. 1997). A positive effect of sulfur on protein concentrations in winter rape seeds of open-pollinated (traditional and double-zero varieties) and hybrid varieties was reported by WIELEBSKI and MUŚNICKI (1998a,b) and JANKOWSKI (2007). The results of the present experiments also indicate that sulfur fertilization at a rate of up to 60 kg ha^{-1} had a beneficial effect (which varied between the studied years) on the total protein content of winter rape seeds.

The effect of sulfur on the oil content of winter rape seeds is ambiguous. KRAUZE and BOWSZYS (2000) reported that sulfur fertilization, regardless of the application method, decreased the crude fat content of rape seeds. In the experiments performed by BUDZYŃSKI and OJCZYK (1995), ROTKIEWICZ et al. (1996), ZUKALOVÁ et al. (2001), WIELEBSKI and WÓJTOWICZ (2004), sulfur fertilization had no significant effect on the accumulation of crude fat in winter rape seeds. In a study conducted by JANKOWSKI (2007), the increase in sulfur rate (up to 90 kg·ha^{-1}) decreased the fat content of seeds by 0.6% d.m. In the current study, sulfur fertilization was not shown to have a significant effect on the fat content of rape seeds. It should be noted, however, that in years marked by high precipitation, sulfur fertilization decreased the crude fat content in seeds in the period of rape ripening (first and third experimental cycle).

Sulfur fertilization primarily deteriorates the quality of rape seeds due to elevated levels of glucosinolates (WITHERS et al. 1995, BILSBORROW et al. 1995, BUDZYŃSKI, OJCZYK 1995, ROTKIEWICZ et al. 1996, WIELEBSKI, MUŚNICKI 1998ab, FISMES et al. 2000, ZUKALOVÁ et al. 2001, WIELEBSKI, WÓJTOWICZ 2003, 2004, JANKOWSKI 2007). Sulfur fertilization may increase the total concentrations of glucosinolates in double-zero („00") rape varieties by as much as 30% (ROTKIEWICZ et al. 1996). The adverse effects of sulfur fertilization on the usability of rape seeds were also noted in this study. Regardless of the application time, an increase in the sulfur rate to the level of 90 kg·ha^{-1} raised the concentrations of alkene and indole glucosinolates in rape seeds.

Sulfur fertilization stimulates primarily the biosynthesis of alkene glucosinolates (ZHAO et al. 1995, BOOTH et al. 1995, ROTKIEWICZ et al. 1996, WIELEBSKI 1997, WIELEBSKI, WÓJTOWICZ 2003, 2004), mainly progoitrin, gluconapin (WIELEBSKI 1997) and glucobrassicanapin (ROTKIEWICZ et al. 1996). In the present study, sulfur fertilization had a stimulatory effect mainly on gluconapin and progoitrin (alkene glucosinolates), 4-hydroxyglucobrassicin and glucobrassicin (indole glucosinolates). Sulfur application in the spring resulted in lower concentrations of progoitrin, gluconapin and 4-hydroxyglucobrassicin in comparison with the application in the fall and spring.

CONCLUSIONS

1. The pre-sowing and the spring rates of sulfur as well as the rate applied entirely in the spring did not lead to significant variations in crude fat concentrations, but a clear tendency towards an increase in the total protein content was observed in rape seeds. This increase was reported to the level of 60 kg S·ha^{-1}.

2. An increase in the sulfur rate (regardless of the date of application) to 90 kg·ha^{-1} raised the concentrations of alkene glucosinolates, mainly gluconapin and progoitrin, and – to a lesser degree – of indole glucosinolates, mainly 4-hydroxyglucobrassicin, in rape seed cv. Lisek.

3. The splitting of the total sulfur rate into two rates (fall + spring) caused a higher increase in the concentrations of gluconapin, progoitrin and 4-hydroxyglucobrassicin than a single rate application in the spring.

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INFLUENCE OF FERTILIZATION WITH DAIRY SEWAGE SLUDGE SANITISED WITH COAL FLY ASH ON MICROBIOLOGICAL ACTIVITY AND CONCENTRATION OF HEAVY METALS IN GREY-BROWN PODZOLIC SOIL

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Abstract

The aim of the study was to determine how dairy sewage sludge (DSS) sanitised with coal fly ash affected microbiological properties of soil and level of heavy metals in soil. The experiment was performed under laboratory conditions (pot experiment). The experiment was carried out on grey-brown podzolic soil, formed from heavy loamy sand, of acidic reaction. Pots were filled with 4 kg of soil. The investigations were performed in three replications. Two rates of dairy sewage sludge fertilization: 1 and 2.5% DSS$\cdot$kg^{-1} of soil were applied. The soil in pots was watered to 60% of the total water capacity and incubated for 4 months. Analyses included assays of the total number of bacteria and fungi, number of cellulolytic bacteria, respiration and dehydrogenase activity and concentration of heavy metals.

It was confirmed that dairy sewage sludge had a significant effect on properties of soil. It positively affected microbiological activity of soil. The test doses (1; 2.5%$\cdot$kg^{-1}) of dairy sewage sludge sanitised with lignite ash caused stimulation of the growth of bacteria and fungi in soil. Dairy sewage sludge had an inhibiting effect on the dehydrogenase activity and stimulated the respiratory activity in the soil under study. The incorporation of dairy sewage sludge caused a non-significant increase of heavy metal content in soil, which was much lower than the norms.

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Key words: dairy sewage sludge, microbiological activity, microorganisms, soil.

WPŁYW NAWOŻENIA OSADEM ŚCIEKÓW MLECZARSKICH HIGIENIZOWANYM POPIOŁEM WĘGLA BRUNATNEGO NA AKTYWNOŚĆ MIKROBIOLOGICZNĄ I ZAWARTOŚĆ METALI CIĘŻKICH W GLEBIE PŁOWEJ

Abstrakt

Celem badań było określenie wpływu osadu ścieków mleczarskich higienizowanego popiołem węgla brunatnego na właściwości mikrobiologiczne i zawartość metali ciężkich w glebie. Badania przeprowadzono w warunkach laboratoryjnych, w doświadczeniu wazonowym. Doświadczenie założono na glebie płowej, wytworzonej z piasku gliniastego mocnego, o odczynie kwaśnym. W wazonach umieszczono po 4 kg gleby. Badania prowadzono w trzech powtórzeniach. W badaniach zastosowano dwie dawki osadu ścieków mleczarskich: 1 i 2,5% osadu $\cdot$ kg^{-1} gleby. Glebę w wazonach nawodniono do poziomu 60% całkowitej pojemności wodnej i inkubowano przez 4 miesiące. Analizy obejmowały oznaczenie: ogólnej liczebności bakterii i grzybów, liczebności bakterii celulolitycznych oraz aktywności respiracyjnej i dehydrogenaz, a także poziomu metali ciężkich.

Stwierdzono, że osad ścieków mleczarskich istotnie wpływał na właściwości gleby. Korzystnie oddziaływał na mikrobiologiczne właściwości gleby. Zastosowane dawki (1; 2,5% $\cdot$ kg^{-1}) osadu higienizowanego popiołem węglowym stymulowały rozwój bakterii i grzybów w glebie. Osad ścieków mleczarskich spowodował zahamowanie aktywności dehydrogenaz i stymulację aktywności respiracyjnej gleby. Wprowadzenie osadu spowodowało nieistotny wzrost zawartości metali ciężkich w badanej glebie, który był znacznie niższy niż przewidują dopuszczalne normy.

Słowa kluczowe: osad ścieków mleczarskich, aktywność mikrobiologiczna, mikroorganizmy, gleba.

INTRODUCTION

For productivity of agroecosystems and protection of the environment it is necessary to develop and implement management strategies that maintain the quality of soil, and these include conserving the amount of organic matter (Saviozzi et al. 1999). Sewage sludge is a useful source of major plant nutrients (nitrogen, phosphorus, sulphur and magnesium) and organic matter. However, sewage sludge can contain larger concentrations of heavy metals than most soils. There is a concern that once metals have been added to agricultural land and accumulate in the topsoil, they could have negative effects on soil fertility and microbial activity (Gibbs et al. 2006). For this reason it is very important to monitor the ecological state of soil after the application of such wastes (Ros et al. 2003, Kizilkaya, Bayrakli 2005).

In many EU countries a notable increase can be currently observed in agricultural utilisation of sewage sludge from the food industry (Butarewicz 2003, Davis, Hall 1997, Przewrocki et al. 2004). Sewage sludge, rich in organic matter and biogens, has an effect on soil environment, soil microorganisms included. Both counts of microorganisms and their biochemical activity undergo changes in soils subjected to the effect of such waste (SULLIVAN et al. 2005, JEZIERSKA-TYS, FRĄC 2005). Tests based on determination of microbial populations and biochemical activity are used for estimation of the fertility and productivity of soils, and provide us with comprehensive knowledge on changes taking place in soil environment (GOSTKOWSKA et al. 1998, KOBUS 1995, MYŚKÓW 1981). The concentration heavy metals in soil after incorporation of dairy sewage sludge helps to evaluate the risk of soil contamination by DSS (MORENO et al. 1999).

The objective of the present study was estimation of the effect of dairy sewage sludge sanitised with lignite ash on the total number of bacteria, fungi, cellulolytic bacteria, respiration activity and dehydrogenase activity and level of heavy metals in grey-brown podzolic soil.

MATERIALS AND METHODS

The study was conducted in a pot experiment in three replications. The object of the study was soil characterised by the grain size composition of sand fraction 65%, silt fraction 19%, fine silt and clay fraction 16%. The soil contained 0.45% of organic carbon and 0.036% of total nitrogen. The soil was amended with dairy sewage sludge sanitised with lignite ash. The sludge doses applied were 1 and 2.5% kg^{-1}. The soil in pots was brought to a moisture level of 60% total water capacity and incubated for 4 months in aerobic conditions. On the dates of analyses soil samples were taken from the pots and microbiological analyses were performed. Analyses were completed after 7, 14, 30, 60, 90 and 120 days of soil incubation. The scope of the microbiological analyses, performed with methods commonly used, covered total counts of bacteria, fungi and cellulolytic bacteria (RODINA 1968), the respiration activity (RÜHLING and TYLER 1973) and dehydrogenase activity (THALMANN 1968). In addition, the content of heavy metals in the soil samples was analysed using the AAS method after mineralization in concentrated nitric acid (V).

The results were processed statistically with ANOVA. The LSDs were calculated with Tukey's test at significance level of $\alpha = 0.05$. All statistical calculations were made with Statistica 7.1 Software.

RESULTS AND DISCUSSION

Figure 1 presents the results of the study on the effect of dairy sewage sludge doses applied in the experiment on counts of bacteria on particular dates of analysis, and their mean values for all experimental treatments.

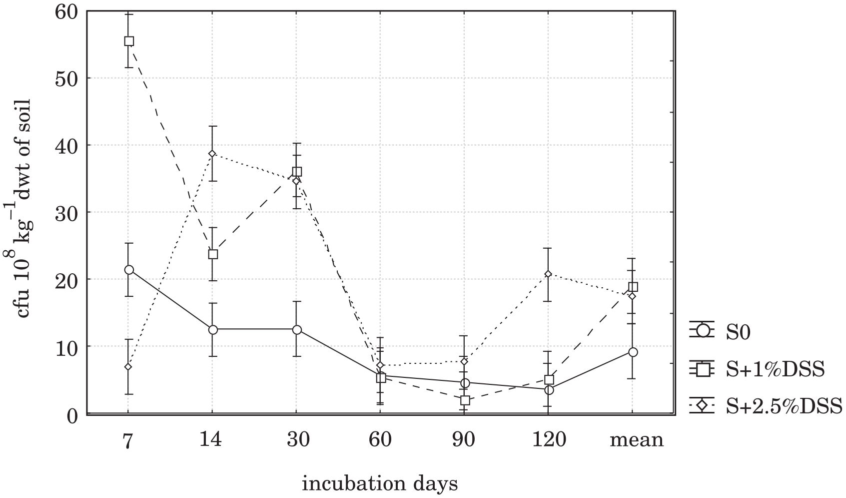

Fig. 1. Effect of dairy sewage sludge on total number of bacteria in soil

The data indicate that in the initial period of the experiment the sludge dose of 2.5% caused a decrease in the bacterial populations to levels considerably lower than the value obtained in the control treatment. The lower sludge dose (1%), on the other hand, caused a significant increase in the growth of the bacterial groups under study. In the subsequent stages of the study, i.e. on analysis dates II and III, a significant increase was observed in the bacterial populations under the effect of both sludge doses applied. On analysis dates IV and V, the populations of the microbial groups in treatments with sludge were similar or insignificantly higher than the values obtained in the control treatment. At the final stage of the experiment, in the treatment with the higher dose of sludge, a significant increase in the number of bacteria was recorded. The mean values of bacterial populations in the particular experimental treatments showed notable stimulation of the microbial groups by both sludge doses applied.

The data concerning the size of fungal populations (Figure 2) indicate that both sludge doses stimulated, throughout the whole study, the growth of this microbial group. A significant seasonal increase in the population of fungi affected the mean value for particular experimental treatments, as illustrated in Figure 2.

The effect of the doses of dairy sewage sludge on the number of microorganisms participating in mineralisation of cellulose is illustrated in Figure 3. The data show that on all the dates of analyses, with the exception of date V, stimulation was observed in the growth of cellulolytic bacteria, especially in the treatment with the higher dose of sludge. This is confirmed by the mean values for the particular treatments concerning the numbers of cellulolytic bacteria, as illustrated in Figure 3.

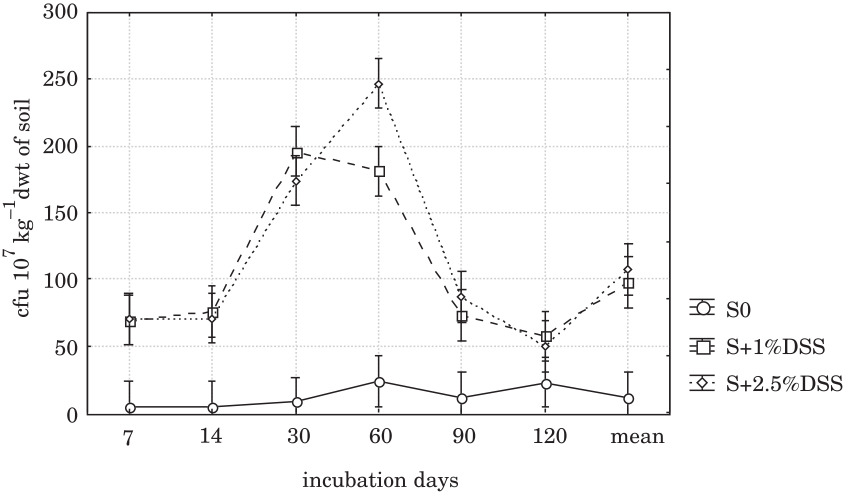

Fig. 2. Effect of dairy sewage sludge on total number of fungi in soil

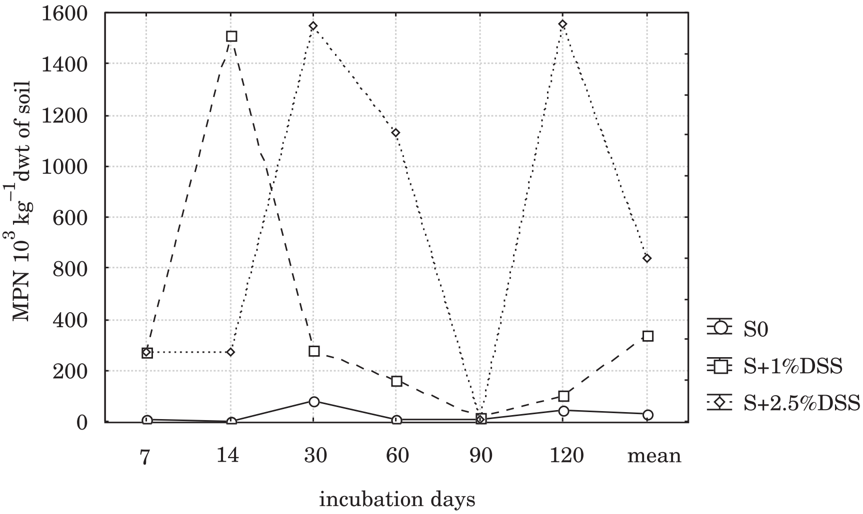

Fig. 3. Effect of dairy sewage sludge on the number of cellulolytic bacteria in soil

The respiration activity in the experimental treatments for the particular dates of analysis is illustrated in Figure 4. The highest respiration activity was characteristic of the treatment with the sludge dose of $2.5\% \cdot kg^{-1}$, but only until date III of analysis. A lower level of respiration activity was observed in the treatment with the sludge dose of 1%, but nevertheless it was higher than in the control treatment. On analysis dates IV and V the respiration activity in the treatments with sludge was significantly higher

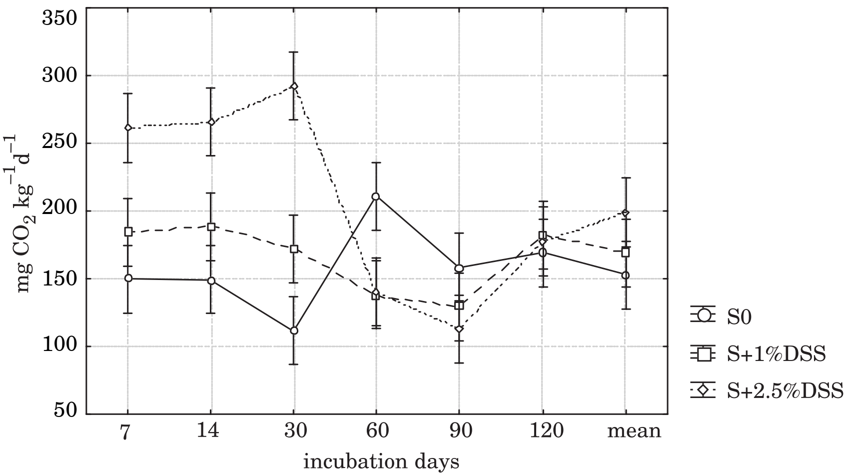

Fig. 4. Effect of dairy sewage sludge on respiration activity of soil

compared to the control treatment. In the final stage of the study, the respiratory activity of grey-brown podzolic soil with dairy sewage sludge was on a level slightly above the value obtained in the control. Mean values of respiration activity for the particular experimental treatments are given in Figure 4a. Notable stimulation of the respiration activity was caused by the sludge dose of 2.5%.

Figure 5 illustrates the data concerning the effect of sludge on dehydrogenase activity in the soil under study. Both the seasonal results and mean values for the experimental treatments indicate a decrease in this activity under the effect of both sludge doses (1 and $2.5\% \cdot kg^{-1}$) compared to the values obtained in the control treatment.

The dairy sewage sludge had influence on the concentration of heavy metals in soil (Table 1). The highest values of heavy metals were observed in the soil amended with the higher dose of DSS, but in all the treatments the concentration of heavy metal was lower than permissible (Rozp. Min. Środ. 2002).

The total number of bacteria and fungi is an index of the biological activity of soil and is used for the determination of the biological status of soil environment (Kucharski et al. 1992, Loc, Greinert 2000, Loc, Obertyńska 2003). The present study shows that the doses of dairy sewage sludge sanitised with lignite ash had a stimulating effect on the total number of bacteria and fungi. The stimulation in the growth of the analysed microbial groups should be attributed to enrichment of grey-brown podzolic soil with organic matter and mineral components brought in with sludge. Increase in soil microbial populations caused by dairy sewage sludge was also observed by other authors (Furczak, Joniec 2002, Jezierska-Tys, Frąc 2005, Lima et al.

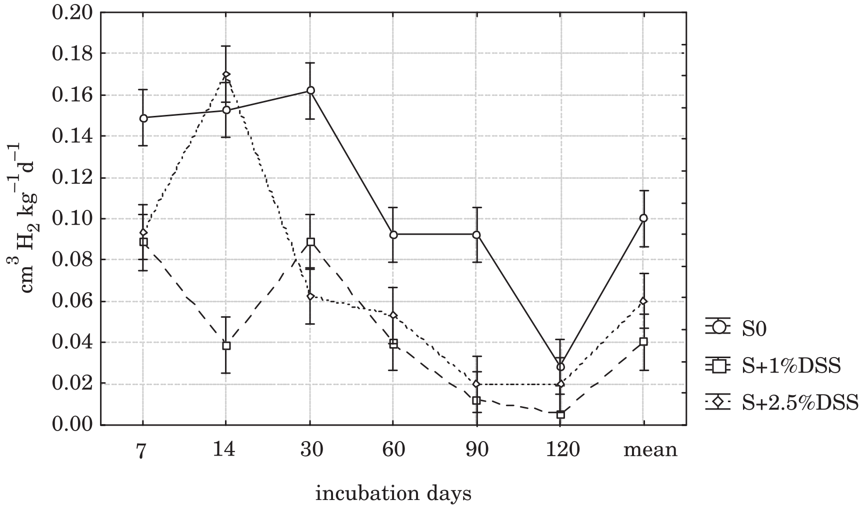

Fig. 5. Effect of dairy sewage sludge on dehydrogenase activity in soil

Table 1

Effect of dairy sewage sludge on concentration of heavy metals in soil (mg $\cdot$ kg^{-1} d.m. of soil)

Metal	Treatments		
Maximum concentration of metal in grey-brown podzolic soil (Rozp. Min. Środ. 2002)	SO control soil	S+1%DSS	S+2.5%DSS
Cd (1)	0.05	0.06	0.10
Cr (50)	2.45	2.44	12.8
Cu (25)	1.21	1.31	5.26
Ni (20)	1.00	1.10	3.62
Pb (40)	2.00	2.98	6.32
Zn (80)	2.23	5.26	17,1
Hg (0.8)	0.01	0.02	0.02

S – soil,
DSS – dairy sewage sludge

1996). The results obtained in our study indicate an effect of dairy sewage sludge on bacteria participating in mineralisation of cellulose, as evidenced by a significant increase in populations of cellulolytic bacteria following the application of sludge at both doses. Also FURCZAK and JONIEC (2005) observed increased populations of cellulolytic bacteria as a result of application of municipal sewage sludge.

Many authors (MYŚKÓW 1981, NANNIPIERI 1994, NANNIPIERI et al. 1999) claim that dehydrogenase activity may be used as an index of changes in biological activity in soil environment resulting from the application of mineral fertilisation, including amendment with sewage sludge. The results obtained in this study indicate a decrease in the dehydrogenase activity of the test soil under the effect of both doses of dairy sewage sludge sanitised with lignite ash. Also LAI et al. (1999) found that coal ash added to sewage sludge reduced dehydrogenase activity in soil. Examination of respiratory activity of soil is one of the methods for determination of activity of soil microorganisms and changes taking place under the effect of both natural and anthropogenic factors. The results of our study showed that dairy sewage sludge doses of 30 and 75 t ha^{-1} caused stimulation of respiratory activity of grey-brown podzolic soil. This was probably caused by increased levels of organic matter in the test soil owing to the introduction of organic matter with sludge. Our earlier study (JEZIERSKA-TYS and FRĄC 2005) also revealed a stimulating effect of dairy sewage sludge on the respiration activity of soil.

CONCLUSIONS

1. The test doses (1 and 2.5%) of dairy sewage sludge sanitised with lignite ash caused stimulation of the growth of bacteria and fungi in grey-brown podzolic soil.

2. Dairy sewage sludge had an inhibiting effect on the dehydrogenase activity and stimulated the respiratory activity in the soil under study.

3. The incorporation of dairy sewage sludge caused a non-significant increase in the content of heavy metals, which remained on a permissible level.

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COUNTS AND ACTIVITY OF MICROORGANISMS PARTICIPATING IN NITROGEN TRANSFORMATIONS IN SOIL, FOUR YEARS AFTER APPLICATION OF SEWAGE SLUDGE

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Abstract

The objective of the study was to determine the direction, intensity and duration of changes in abundance and activity of certain microbial groups active in nitrogen transformations in soil subjected to a 4-year fertilization treatment with municipal and industrial sewage sludge. The study was conducted on podzolic soil, whose accumulation horizon had been fertilized in 1998 with fermented sewage sludge at doses of 30 $Mg \cdot ha^{-1}$(1%), 75 $Mg \cdot ha^{-1}$(2.5%), 150 $Mg \cdot ha^{-1}$(5%), 300 $Mg \cdot ha^{-1}$(10%) and 600 $Mg \cdot ha^{-1}$(20%) and planted with willow (*Salix viminalis* L.). Four years after the application of sludge, microbiological and biochemical analyses were made in two soil horizons (0-20 cm and 20-40 cm). It was found that in the soil from the 0-20 cm depth significant stimulation of the growth of proteolytic fungi and bacteria continued, but only under the effect of the highest dose of sludge. Moreover, there was a notable stimulation of protease activity and nitrification process alongside a slight inhibition of ammonification. In the soil from the 20-40 cm layer stimulation of the growth of protein-decomposing fungi was observed as well as that of proteolytic and nitrification activity of soil, while ammonification was inhibited. However, the effect of sludge was generally less pronounced in the deeper soil layer than in the surface soil horizon.

Key words: municipal-industrial sewage sludge, podzolic soil, microbiological transformations of nitrogen, content $N\text{-}NH_4$ and $N\text{-}NO_3$.

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LICZEBNOŚĆ I AKTYWNOŚĆ MIKROORGANIZMÓW CZYNNYCH W PRZEMIANACH AZOTU W GLEBIE, W CZWARTYM ROKU OD WZBOGACENIA JEJ OSADEM ŚCIEKOWYM

Abstrakt

Celem pracy było określenie kierunku, natężenia i czasu utrzymywania się zmian w liczebności i aktywności niektórych grup mikroorganizmów czynnych w przemianach azotu, w glebie poddanej 4-letniemu oddziaływaniu osadu ścieków komunalno-przemysłowych. Badaniami objęto glebę bielicową, której poziom akumulacyjny nawieziono w 1998 r. przefermentowanym osadem ściekowym w ilości: 30 $Mg \cdot ha^{-1}$(1%), 75 $Mg \cdot ha^{-1}$(2,5%), 150 $Mg \cdot ha^{-1}$ (5%), 300 $Mg \cdot ha^{-1}$(10%) i 600 $Mg \cdot ha^{-1}$(20%). Następnie glebę obsadzono wierzbą (*Salix viminalis* L.) i w 4-tym roku od wprowadzenia osadu wykonano w dwu jej warstwach (0-20 cm i 20-40 cm) analizy mikrobiologiczne i biochemiczne. Stwierdzono, że w glebie z głębokości 0-20 cm utrzymywało się nadal istotne pobudzenie rozwoju bakterii i grzybów proteolitycznych, ale tylko pod wpływem najwyższej dawki osadu. Ponadto występowała wyraźna stymulacja aktywności proteazy i procesu nitryfikacji oraz niewielkie hamowanie amonifikacji. W glebie z warstwy 20-40 cm odnotowano również stymulację rozwoju grzybów rozkładających białko, aktywności proteolitycznej i nitryfikacyjnej gleby oraz hamowanie amonifikacji. Jednak oddziaływanie osadu było w tych warunkach na ogół słabsze niż w wierzchniej warstwie gleby.

Słowa kluczowe: osad ścieków komunalno-przemysłowych, gleba bielicowa, mikrobiologiczne przemiany azotu, zawartość $N\text{-}NH_4$ i $N\text{-}NO_3$.

INTRODUCTION

Numerous studies, for example HATTORI and MUKAI (1986), CZEKAŁA (2002) and BIELIŃSKA and ŻUKOWSKA (2002), indicate that sewage sludge is a rich source of both carbon and nitrogen organic matter, and of various mineral components. Introduced to soil, it is a valuable source of nutrients for various soilborne microbial groups, including the ones taking part in nitrogen transformations, thus affecting their activity and consequently the fertility of soils (SIUTA 1998, BARAN et al. 1999, BIELIŃSKA, ŻUKOWSKA 2002, ŻUKOWSKA et al. 2002, FURCZAK, JONIEC 2007b, JONIEC, FURCZAK 2007b). Therefore, one possible utilisation method of such waste is its application in agriculture for fertilisation of soils under alternative crop cultures (SIUTA 1998), for example willow (*Salix viminalis*). This species is characterized by high capacity for absorption of nitrogen, phosphorus and water and - most importantly – also of heavy metals, thus displaying phyto-remediation properties (SIUTA 1998, BARAN et al. 2000). The crop yield of willow is used in the furniture and wicker-products industry, in power generation as a fuel which is less noxious than coal, and in the chemical industry – as a source of cellulose and raw material for production of bio-alcohol (SZCZUKOWSKI et al. 2001). Studies on the effect of sewage sludge on abundance and activity of microorganisms involved in nitrogen transformations in soil are mainly fragmentary and most often conducted under laboratory conditions (HATTORI, MUKAI 1986, Kobus et al. 1990, Pascual et al. 2007), or else involve short-term field experiments with a variety of plants (Baran et al. 1999, Bielińska, Żukowska 2002, Garcia-Gil et al. 2004, Furczak, Joniec 2007b,c, Joniec, Furczak 2007a). The long-term effects of sewage sludge application on the a.m. microbiological and biochemical parameters in soil under a culture of willow have not been performed until present. Therefore, the objective of this study has been to estimate the direction, dynamics and duration of changes in the numbers of microbial groups and in the activity of biochemical processes related to nitrogen transformations in soil from a long-term field experiment, in the 4th year since soil amendment with sewage sludge.

Multi-year monitoring of microbiological effects of soil fertilisation with sewage sludge is not only of theoretical interest but has a practical application, such as estimating the possibility of utilisation of waste in a manner that is safe for the micro-biocenosis. Moreover, our study can help to assess to what extent the tests used here may be applicable to the estimation of microbiological effects of multi-year influence of sewage sludge on the soil environment.

MATERIAL AND METHODS

The study involved a field experiment set up in Końskie in 1998 by the Institute of Soil Science and Natural Environment Management, Lublin University of Agriculture. The experiment comprised plots of a surface area of 15 m^2 each. The accumulation horizon of podzolic soil developed from weakly loamy sand was fertilised with fermented sludge from municipal and industrial sewage produced at the Mechanical-Biological Sewage Treatment Plant in Końskie. The sludge, in accordance with the current standards in Poland (Rozp. MOŚ 2002), met the requirements set up for agricultural utilisation. It was applied at doses of 30 Mg$\cdot$ha^{-1} (1%), 75 Mg$\cdot$ha^{-1} (2.5%), 150 Mg$\cdot$ha^{-1} (5%), 300 Mg$\cdot$ha^{-1} (10%) and 600 Mg$\cdot$ha^{-1}(20%). Next, four weeks after the application, the soil was planted with willow (*Salix viminalis* L.). The control treatment in the experiment was soil under the same crop culture but without sludge amendment. The grain size composition and certain physicochemical and chemical properties of the soil and sludge used for the amendment are given by Baran et al. (2000).

In the fourth year of the experiment, i.e. 2001, samples of soil were taken from the depths of 0-20 cm and 20-40 cm in three sampling seasons (spring, summer and autumn) to perform the following determinations (with three replications): counts of protein-decomposing bacteria and fungi on the Frazier substrate (Rodina 1968), to which – in the case of fungi – antibiotics were added in accordance with the recommendations by Martin (1950); ammonification intensity, in 25-gram weighed portions of soil containing 0.1%

of asparagine, from which – after 3 days of incubation – ammonium ions were extracted and their content was determined according to Nessler's method (Nowosielski 1974); nitrification intensity, in 25-gram weighed portions of soil containing 0.1% of monobasic ammonium phosphate, from which – after 7 days of incubation – nitrate ions were extracted and their level was measured using the brucine method (Nowosielski 1974); protease activity, according to the method of Ladd and Butler (Ladd, Butler 1972); content of $N\text{-}NH_4$ and $N\text{-}NO_3$ ions in the soil after extraction of 25-gram weighed portions of the soil, following Nessler's (Nowosielski 1974) and the brucine (Nowosielski 1974) methods, respectively; soil reaction, potentiometrically in 1 $mol \cdot dm^{-3}$ KCl, and soil moisture with the gravimetric method. During the determinations of ammonification and nitrification, the moisture content of the incubated soil was maintained at the level of 50-60% of total hydraulic capacity. Mineral forms of nitrogen were extracted from the soil with 2% KCl at the 1:5 ratio.

The results were processed statistically with the method of analysis of variance. The significance of differences was determined with Tukey's test at p = 0.05.

Table 1

Moisture of soil

Dose of sludge ($Mg\ ha^{-1}$)	Depth (cm)	Moisture (% w.w.)			
		24. 05	26. 07	25. 10	Mean
0	0-20	16.10	13.07	13.96	14.38
30		19.25	12.97	15.73	15.98
75		17.81	14.61	17.04	16.49
150		22.40	15.38	22.49	20.09
300		22.39	18.93	17.97	19.76
600		28.02	22.68	20.86	23.85
0	20-40	12.97	11.37	14.28	12.87
30		15.72	10.90	14.34	13.65
75		14.76	14.29	15.96	15.00
150		14.70	12.80	18.62	15.37
300		16.92	11.17	18.32	15.47
600		24.53	19.17	21.97	21.89

RESULTS AND DISCUSSION

The study performed in the fourth year of the experiment showed that in the soil from 0-20 cm a significant stimulating effect of sewage sludge on the growth of protein-decomposing bacteria and fungi occurred only under the influence of the highest dose of sludge (Table 2). This indicates that the effect of sludge on the microbial groups examined was notably weaker than in the previous years of the experiment (FURCZAK, JONIEC 2007b, JONIEC, FURCZAK 2007a,c). Some changes in this respect were observable also in the soil from the depth of 20-40 cm (Table 2). Although by then no effect of sludge on the numbers of proteolytic bacteria was observed, the highest dose of sludge caused stimulation of the growth of "proteolytic" fungi, which was even stronger than in the soil from 0-20 cm (Table 2). While analysing periodic oscillations (Table 2) in the soil from the two horizons, it was noticed that generally both microbial groups occurred most abundantly in spring, which was probably caused by a temperature increase after the winter season.

The stimulation of the growth of protein-decomposing bacteria and fungi observed in the combination with the highest dose of sludge (Table 2) could have been caused by the persistently increased level of nutrients – under those conditions – for the microbial groups. This explanation is supported by ŻUKOWSKA et al. (2002), who showed a continuingly increasing content of organic carbon and total nitrogen in soil treated with sludge. Those compounds had certainly originated from the sludge added to soil and, additionally, were a product of microbiological transformation of organic matter introduced to soil with the sludge. It appears that also the growth of proteolytic bacteria and fungi could have been supported by improvement of other living conditions of these microbial groups, i.e. soil reaction and moisture (Tables 1, 3), and probably the water-air relations. Some authors report that in soils amended with sewage sludge (at greater doses in particular) the soil retention capacity increases, aggregate and semi-aggregate structure is formed, soil density decreases while the full water capacity of soil increases (BARAN et al. 1996a,b).

It has also been demonstrated that the proteolytic activity in the top layer of the soil (0-20 cm) was stimulated by the higher levels of the sludge, i.e. 150 $Mg \cdot ha^{-1}$, 300 $Mg \cdot ha^{-1}$ and 600 $Mg \cdot ha^{-1}$ (Table 4). Similar tendencies were observed by BARAN et al. (1999) in soil 0-20 cm deep originating from a 4-year field experiment with sewage sludge. It has been demonstrated (DICK 1994) that the primary producers of proteases in soil are protein-decomposing bacteria and fungi. Therefore, their stimulation in treatments with higher doses of sludge, and especially with the highest one (Table 2), was accompanied by an increase in the proteolytic activity (Table 4).

Table 2

Numbers of proteolytic bacteria and fungi in the soil

Dose of sludge ($Mg \cdot ha^{-1}$)	Depth (cm)	Proteolytic bacteria (cfu $10^9 kg^{-1}$ d.m. of soil)					"Proteolytic" fungi (cfu $10^6 kg^{-1}$ d.m. of soil)				
		24. 05	26. 07	25. 10	mean	(%)	24. 05	26. 07	25. 10	mean	(%)
0	0-20	3.2	2.3	1.7	2.4		85.2	41.5	45.6	57.4	
30		2.5	2.4	1.5	2.1	-12.5	63.6	50.1	38.6	50.7	-11.5
75		3.1	1.8	3.2	2.3	-4.2	66.8	65.7	31.2	54.6	-4.9
150		3.5	3.0	0.9	2.3	-4.2	57.1	53.8	36.7	49.2	-14.3
300		3.8	3.1	2.3	3.0	29.2	89.8	99.1	47.2	78.7	37.1
600		8.0	4.4	1.9	4.8	100.0	123.7	241.3	60.4	141.8	147.0
0	20-40	1.3	1.2	2.3	1.6		30.1	7.1	11.4	16.2	
30		2.2	1.3	2.1	1.9	18.8	16.2	9.6	15.2	13.7	-15.4
75		3.2	1.5	1.9	2.2	37.5	30.6	6.5	23.2	20.1	24.1
150		3.6	2.5	1.6	2.6	62.5	14.5	13.5	23.7	17.3	6.2
300		3.3	2.3	2.1	2.6	62.5	15.2	4.4	65.1	28.3	145.9
600		2.0	2.8	3.0	2.6	62.5	54.0	59.6	44.7	52.8	225.9
Mean		3.3	2.4	1.9	2.5		53.9	54.3	36.9	48.4	
Mean for depth		0-20 cm – 2.8; 20-40 cm – 2.2					0-20 cm – 72.1; 20-40 cm – 24.7				
LSD date		0.4					10.2				
LSD depth		0.3					6.9				
LSD depth x dose		1.1					28.8				
LSD treatments		2.1					54.4				

% – stimulation or inhibition by sludge

Table 3

Content of $N-NH_4$, $N-NO_3$ ions and reaction in the soil

Dose of sludge ($Mg \cdot ha^{-1}$)	Depth (cm)	Content of $N-NH_4$ (mg kg^{-1} d.m. of soil)					Content of $N-NO_3$ (mg kg^{-1} d.m. of soil)					Reaction (pH KCl)			
		24. 05	26. 07	25. 10	mean	(%)	24. 05	26. 07	25. 10	mean	(%)	24. 05	26. 07	25. 10	range
0	0-20	30.71	37.28	36.75	34.91		33.39	24.19	7.91	21.83		6.8	7.5	6.8	6.8 - 7.5
30		32.29	33.97	30.61	32.29	-7.5	38.85	22.72	13.77	25.11	15.03	7.4	7.5	7.7	7.4 - 7.7
75		34.58	24.09	29.34	29.34	-16.0	61.25	25.65	5.47	30.79	41.4	7.5	7.8	7.5	7.5-7.8
150		45.68	25.92	27.07	32.89	-5.8	60.51	30.07	22.56	37.71	72.6	7.6	7.9	7.7	7.6 - 7.9
300		46.12	35.77	22.65	34.75	-0.4	69.69	31.67	19.38	40.25	84.2	7.3	7.7	7.4	7.3 - 7.7
600		48.24	37.78	43.35	43.12	23.6	72.35	41.18	14.89	42.81	96.1	7.3	7.8	7.3	7.3 - 7.8
0	20-40	33.56	34.30	19.46	29.11		35.85	33.92	16.83	28.87		7.3	7.3	6.8	6.8 - 7.3
30		48.22	34.96	21.13	34.77	19.4	47.54	27.33	10.53	28.47	-1.4	7.1	7.1	7.0	7.0 - 7.1
75		55.72	18.63	28.57	34.31	17.9	45.68	27.20	10.68	27.85	-3.5	7.0	7.4	6.8	6.8 - 7.4
150		45.30	32.70	44.70	40.90	40.5	38.18	35.79	23.35	32.44	12.4	7.0	7.6	7.1	7.0 - 7.6
300		38.55	23.76	33.87	32.06	10.1	43.72	29.13	25.14	32.66	13.1	7.2	7.7	7.2	7.2 - 7.7
600		43.76	22.45	19.44	28.55	-1.9	46.57	46.93	25.57	39.69	37.5	7.1	7.3	7.2	7.1 - 7.3
Mean		41.81	30.13	29.75			49.47	31.32	16.34						
Mean for depth		0-20 cm – 34.56; 20-40 cm – 33.23					0-20 cm – 33.08; 20-40 cm – 31.66								
LSD date		1.61					2.91								
LSD depth		1.10					1.98								
LSD depth x dose		4.56					8.24								
LSD treatments		8.60					15.55					0.3			

% – stimulation or inhibition by sludge

Table 4

Activity of selected rocesses related to nitrogen transformations in the soil

Dose of sludge ($Mg \cdot ha^{-1}$)	Depth (cm)	Protease, mg of tyrosine (kg^{-1} d.m. of soil h^{-1})					Ammonification (mg $N\text{-}NH_4$ kg^{-1} d.m. of soil 3 d^{-1})					Nitrification (mg $N\text{-}NO_3$ kg^{-1} d.m. of soil 7 d^{-1})				
		24. 05	26. 07	25. 10	mean	(%)	24. 05	26. 07	25. 10	mean	(%)	24. 05	26. 07	25. 10	mean	(%)
0	0-20	9.94	10.63	15.05	11.88		246.40	185.44	295.64	242.49		22.86	41.06	46.01	36.64	
30		9.79	10.10	11.75	10.54	-11.1	198.28	112.35	310.35	206.99	-14.6	101.66	44.27	51.31	65.74	79.5
75		10.80	10.75	13.35	11.64	-2.0	232.69	130.95	284.02	215.88	-10.97	81.86	103.75	114.26	99.96	172.1
150		16.48	10.53	36.64	21.22	78.8	130.61	109.60	275.54	171.92	-29.1	187.22	183.69	298.26	223.06	524.2
300		22.87	15.35	19.43	19.22	61.9	115.74	116.85	318.45	183.68	-24.3	189.52	123.55	128.77	147.28	302.0
600		23.03	33.92	17.46	24.80	108.9	221.70	91.12	304.08	205.63	-15.2	288.40	196.45	208.74	231.20	531.0
0	20-40	6.62	6.94	3.87	5.80		207.03	227.25	358.10	264.13		7.20	21.03	37.72	21.98	
30		8.05	8.54	10.38	8.99	54.7	230.86	188.61	318.14	245.87	-6.9	2.02	73.47	44.22	39.90	81.5
75		4.02	5.04	9.71	6.26	7.8	222.00	265.64	316.98	268.21	1.5	1.95	19.75	21.03	14.24	-35.2
150		5.75	4.61	16.33	8.90	53.2	237.59	206.11	247.35	230.35	-12.8	6.17	87.75	37.30	43.74	99.0
300		3.00	5.01	12.18	6.73	15.8	218.31	135.33	294.81	216.15	-18.2	84.02	33.54	21.90	46.49	111.5
600		8.48	7.48	12.69	9.55	64.4	212.52	144.85	350.15	235.84	-10.7	95.87	74.26	29.72	66.62	203.1
Mean		10.74	10.74	14.90	12.13		206.15	159.51	306.13	223.92		89.06	83.55	86.60	86.41	
Mean for depth		0-20 cm – 16.55; 20-40 cm – 7.70					0-20 cm – 204.43; 20-40 cm – 243.43					0-20 cm – 133.98; 20-40 cm – 38.83				
LSD date		0.55					14.29					4.34				
LSD depth		0.37					9.71					2.95				
LSD depth x dose		1.55					40.42					12.28				
LSD treatments		2.92					76.27					23.17				

% – stimulation or inhibition by sludge

In some of the treatments, the activity of proteases was also observed to have increased in the soil from the deeper layer (20-40 cm), but the stimulation was weaker than in the 0-20 cm deep soil horizon (Table 4). Our comparison of the data given in Table 4 with the results obtained in the previous years (FURCZAK, JONIEC 2007b,c, JONIEC, FURCZAK 2007b) shows that the effect of sludge on the proteolytic activity in both soil layers weakened with time.

It is commonly known that proteases play the main role in nitrogen transformations in soil and their activity is largely dependent on the content of C org and total N (CHAZIJEW 1982, BIELIŃSKA, ŻUKOWSKA 2002). The present study confirms this relationship, similarly to ŻUKOWSKA et al. (2002), who demonstrated a continuing increase of the level of a.m. fractions of organic matter in treatments with higher doses of sludge. This effect could have also been facilitated by increased numbers of various microbial groups (Table 2, FURCZAK, JONIEC 2007a). KOBUS (1995), SASTRE et al. (1996) and PAUL and CLARK (2000) report that dead micro-organisms are an important source of carbon and mineralised nitrogen, affecting directly the growth in the proteolytic activity of soil. Positive correlation of the proteolytic activity with the content of microbial biomass carbon in soil is also reported by WICK et al. (2002). While analysing periodic oscillations of the proteolytic activity, it was noticed that, contrary to the most abundant occurrence of proteolytic microorganisms in spring (Table 2), the proteolytic activity was the most intensive in autumn (Table 4). Taking into account the protective function of soil with respect to extracellular enzymes (KOBUS 1995), it is likely that this activity in autumn was a sum of the currently present proteolytic microorganisms and proteases released earlier.

The proteolytic activity of soil is also determined by such factors as soil reaction, moisture, temperature and oxygenation (CHAZIJEW 1982, CIEŚLA, KOPER 1990). This is supported by the present study, which showed continued improvement of living conditions of proteolytic microorganisms (especially soil moisture and, to a certain extent, reaction) – Tables 1, 3. Also the improvement of other soil properties (aggregate structure and water-air relations), as indicated by studies by BARAN et al. (1996a,b), could have provided an additional contribution to the stimulation of this activity.

Moreover, a slight although significant inhibition of the process of ammonification was observed in the soil from 0-20 cm sampled from the plots fertilized with the higher rates of sludge (150 $Mg \cdot ha^{-1}$ and 300 $Mg \cdot ha^{-1}$) – Table 4. This effect was also noticed in the soil from the depth of 20-40 cm, but it was less intensive than in the surface horizon and observable only in the treatment with the 300 $Mg \cdot ha^{-1}$ dose of the waste. It should be emphasized that the effect of sludge on the a.m. parameter weakened in comparison to the preceding years (FURCZAK, JONIEC 2007b,c, JONIEC, FURCZAK 2007b). As in the case of the protease activity, the intensity of ammonification in samples from both horizons of the soil was the highest in autumn (Table 4).

A decline in ammonification in incubated soil under the effect of increased doses of sludge was probably attributable mainly to the simultaneous intensification of nitrification under these conditions. As a result of the latter process, rapid oxidation of ammonium ions occurred, which caused a reduction of their content in the soil samples (Table 4). Slight inhibition of ammonification in the incubated soil was not accompanied by any reduction in the level of ammonium ions under natural conditions (Table 3). In some treatments a significantly higher content of these ions was even recorded. Kobus et al. (1990) and Herrero et al. (1998) reached similar conclusions studying the effect of sewage sludge on intensification of the process of ammonification.

Since $N\text{-}NH_4$ ions are generally more preferred by micro-organisms as a source of nitrogen than $N\text{-}NO_3$ ions (Paul, Clark 2000), it appears that the depressed ammonifying power determined on the basis of the content of ammonium ions could have also been, to some extent, an effect of its more intensive incorporation by microorganisms into their own cells. The above observation, as in the study by Herrero et al. (1998), is supported by the fact that the decrease in ammonification in the 4^{th} year of the experiment was accompanied by a simultaneous increase in the activity of dehydrogenases in soil (Furczak, Joniec 2007a), as these enzymes are responsible for transformations of ammonium ions in the processes of nitrification and their immobilization via incorporation into microbial biomass (Herrero et al. 1998, Paul, Clark 2000).

During the study, the most pronounced changes caused by sludge occurred in the process of nitrification (Table 4). This is indicated by a distinct, usually increasing with the dosage, stimulation of this process. This effect persisted in both layers of the soil, but it was stronger in the soil from 0-20 cm. Also the content of nitrate ions in the 0-20 cm layer was higher in almost all treatments with sludge compared to the control soil (Table 3), while in the soil from the deeper layer it was higher only in the treatment with 600 $Mg \cdot ha^{-1}$ content of the waste.

As in the case of the other biochemical tests under analysis, stimulation of nitrification was weaker than in the preceding three years of the experiment (Furczak, Joniec 2007b,c, Joniec, Furczak 2007b).

The results obtained for particular time periods of the study indicate that in both layers of the soil the intensity of the process of nitrification and the content of $N\text{-}NO_3$ ions were the highest in spring (Tables 3, 4).

In the soil with sludge, stimulation of oxidation of ammonium nitrogen was most likely caused by the higher level of nutrient substrate for nitrifiers, which is indicated by the content of $N\text{-}NH_4$ ions in the soil (Table 3) and by the persisting favourable living conditions for those microorganisms, i.e. increased soil moisture, somewhat higher reaction (Tables 1 and 4) and, probably, improved oxygenation of the soil. Studies by Baran et al. (1996a,b) indicate that sewage sludge introduced to soil notably improves its water-air relations.

In the fourth year after the application of sludge, under the effect of the in the presence higher sludge doses, a slight increase of pH in the soil from 0-20 cm appeared (Table 3). The effect was more pronounced in spring and autumn. The effect of sludge on the reaction of the deeper layer of soil was notably weaker and observable only in certain treatments of the experiment (Table 3).

CONCLUSIONS

1. In the fourth year after the introduction of municipal and industrial sewage sludge to soil, it was only the highest sludge dose that continued to stimulate the growth of proteolytic bacteria and fungi in the soil from 0-20 cm. In the 20-40 cm layer the effect was observable only for “proteolytic” fungi.

2. The higher doses of sludge (150 $Mg\cdot ha^{-1}$, 300 $Mg\cdot ha^{-1}$ and 600 $Mg\cdot ha^{-1}$) caused an increase in the proteolytic activity of the soil from 0-20 cm. In the deeper layer of the soil (20-40 cm), stimulation of this enzymatic parameter was recorded as well, but it was notably weaker.

3. In some of the treatments, a slight inhibition of the process of ammonification continued in the soil from the depth of 0-20 cm (150 $Mg\cdot ha^{-1}$, 300 $Mg\cdot ha^{-1}$). The effect was also noticed in the deeper layer of soil, where it was distinctly weaker. The content of ammonium ions, on the other hand, was higher in certain treatments in the 0-20 cm (dose of 600 $Mg\cdot ha^{-1}$) and in the 20-40 cm layers (doses of 30 $Mg\cdot ha^{-1}$, 75 $Mg\cdot ha^{-1}$ and 150 $Mg\cdot ha^{-1}$) than in the control.

4. The most intensive changes persisted in the course of the process of nitrification. In the soil from 0-20 cm a distinct stimulation of nitrification was observed for all the doses of sewage applied, although larger at higher fertilization rates. Also the content of nitrates was notably higher in almost all the treatments with sludge. The positive effect of sludge on the process of nitrification and on the concentration of $N\text{-}NO_3$ ions was observable also in the deeper layer of the soil, but it was less pronounced. This process turned out to be the most sensitive indicator of the analysed changes persisting in the soil amended with sludge.

5. In the fourth year of the experiment, there was a continuing albeit slight increase of soil pH in the 0-20 cm layer in all the treatments, while in the 20-40 cm layer the effect of sludge on the soil reaction was nearly absent.

6. The results indicate that the products of transformations of fermented sewage sludge introduced in the soil four years before continued to produce a generally stimulating effect on the analysed studied parameters of microbiological activity of the soil environment. The study may be useful

for comprehensive assessment of long-term effects of agricultural utilisation of such waste. Therefore, it is recommended that research on this matter be continued also in the future.

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CHANGES IN MAGNESIUM CONCENTRATIONS AND LOAD IN RUNOFF WATER FROM NITRATE VULNERABLE ZONES*

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Abstract

The objective of this study was to investigate magnesium concentrations and load, and to determine their seasonal changes in runoff water from catchments classified as nitrate vulnerable zones.

The results indicate that the average concentrations of 12.2 mg Mg·dm^{-3}, with fluctuations within the range of 3.3 to 26.2 mg·dm^{-3}, and average annual load of 14.3 kg Mg·ha^{-1}·year^{-1}, with fluctuations within the range of 4.8 to 41.6 kg Mg·ha^{-1}·year^{-1}, in runoff water from agricultural areas are determined by weather conditions (season), type of drainage system (ditches, drains) and fertilization intensity. In comparison with land drained by a network of drainage ditches, intensive farming in drained areas increases magnesium loss 2.5-fold from 10 kg per hectare of semi-intensively farmed area to 25 kg Mg·ha^{-1} in an intensively farmed area. The highest magnesium loss was reported in the non-growing season, and around 46% of total magnesium load was leached out in the winter. The magnesium loss was minimized during harsh winters and summer draughts (to around 1.2 kg Mg·ha^{-1}·year^{-1}) due to a seasonal absence of runoffs.

Key words: magnesium, load, nitrate vulnerable zone, agricultural sources.

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DYNAMIKA STĘŻEŃ I ŁADUNKU MAGNEZU W WODACH ODPŁYWAJĄCYCH Z OBSZARÓW SZCZEGÓLNIE ZAGROŻONYCH SPŁYWAMI AZOTU ZE ŹRÓDEŁ ROLNICZYCH

Abstrakt

Celem pracy była analiza stężeń i ładunków magnezu oraz określenie ich sezonowych zmian w wodach odpływających ze zlewni zakwalifikowanych do obszarów szczególnie narażonych na zanieczyszczenia wód azotanami ze źródeł rolniczych.

Uzyskane wyniki świadczą o tym, że średnie stężenie 12,2 mg Mg·dm^{-3}, z wahaniami od 3,3 do 26,1 mg·dm^{-3}, i średni roczny ładunek 14,3 kg Mg·ha^{-1}·rok^{-1}, z wahaniami od 4,8 do 41,6 kg Mg·ha^{-1}·rok^{-1}, w wodach odpływających urządzeniami melioracyjnymi z obszarów rolniczych zależą od warunków meteorologicznych (pora roku), rodzaju systemu melioracyjnego (rowy, dreny) oraz intensywności nawożenia. Intensywne użytkowanie zdrenowanych gruntów ornych, w porównaniu z glebami odwadniającymi siecią rowów, powoduje ponad 2,5-krotny wzrost wymycia magnezu z 10 kg z 1 ha obszaru o średnio intensywnym rolnictwie do 25 kg Mg·ha^{-1} z obszaru intensywnego rolnictwa. Największy odpływ występował w okresie pozawegetacyjnym, ok. 46% ogólnej masy ładunku odpłynęło zimą. W okresie mroźnych zim oraz suszy letnich następuje minimalizacja odpływu magnezu (do ok. 1,2 kg Mg·ha^{-1}·rok^{-1}) w wyniku okresowego zaniku odpływu wód.

Słowa kluczowe: magnez, stężenie, ładunek, obszar szczególnie zagrożony, źródła rolnicze.

INTRODUCTION

In addition to the nutrient load carried by precipitation, surface water also contains substances drained from the soil and transported with surface runoffs from agricultural areas. The volume and intensity of nutrient migration is largely affected by the quantity of water runoff which, in turn, is determined by the geological structure of the catchment, relief features, soil class, soil's buffering and sorption capacity, type of catchment use and climatic conditions determining water flow and its availability for plants (Solarski et al. 2000, Koc et al. 2002, Lipiński 2003). The quality of water varies seasonally due to changes taking place in the soil, biological sorption and changes in the soil's water balance in different seasons of the year (Koc et al. 1996, Miler et al. 2001). Mineral and organic fertilizer nutrients which are not plant available are partially leached out from the soil, transported to water-bearing horizons or directly to surface water. Intensive farming also leads to the evacuation of vital nutrients, including magnesium, a key element, essential for life processes (Borowiec et al. 1993, Koc 1999). Large quantities of magnesium are leached out from soils which are heavily fertilized with potassium and ammonium nitrogen as these elements have an antagonistic effect on exchange sorption. High level of mineral fertilization and regular application of high slurry rates containing K^+ and NH_4^+ could speed up magnesium loss.

The objective of this study was to investigate magnesium concentrations and load, and to determine their seasonal changes in runoff water from catchment areas classified as nitrate vulnerable zones.

MATERIALS AND METHODS

The study was carried out in 2005-2007 in the catchment of Lake Dobskie. The land in the lake's catchment is used by a farming estate, which covers a total area of 1,090 ha, including 841 ha of arable land. The farm specializes in hog production (around 2,600 head). The study covered four partial catchments of Lake Dobskie characterized by different fertilization intensity (intensive, semi-intensive and low intensity) and type of drainage system (ditches and drains – Figure 1):

Catchment no. 1 is an intensively farmed and drained catchment (521) of an area of 15.1 ha of arable land, soil quality class IIIb and IVa. In 2005 it was cropped with winter triticale. Mineral fertilization at a rate of N – 61 $kg \cdot ha^{-1}$ and organic fertilization (animal slurry) at a rate of 20 $m^3 \cdot ha^{-1} \cdot year^{-1}$ were applied. In 2006, spring barley was grown on these fields, fed with the

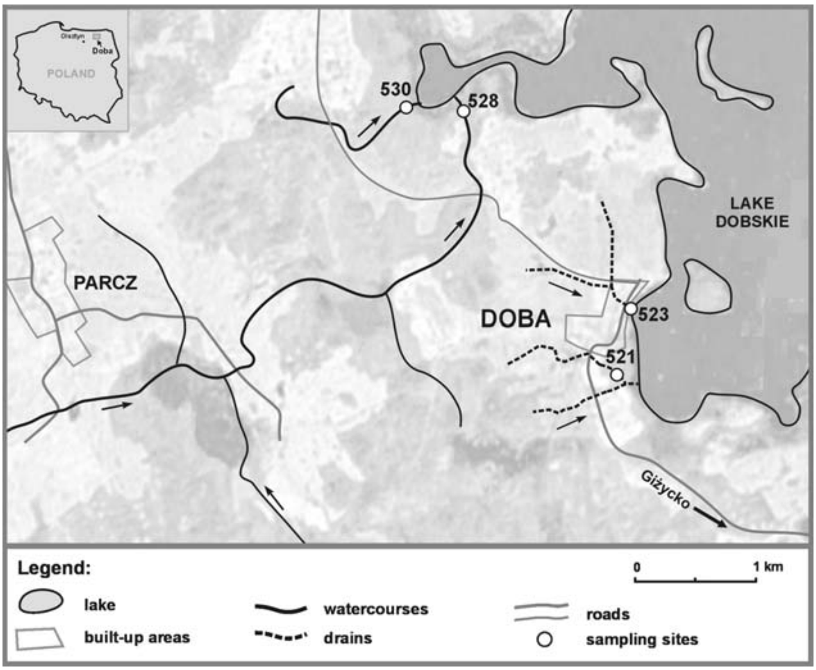

Fig. 1. Sampling sites in the catchment of Lake Dobskie

following fertilization rates: 98 kg $N \cdot ha^{-1}$, 46 kg $P_2O_5 \cdot ha^{-1}$, 60 kg $K_2O \cdot ha^{-1}$. Slurry was applied in June 2006. In 2007, winter rye was grown and fed 100 kg $N \cdot ha^{-1}$, 52 kg $P_2O_5 \cdot ha^{-1}$, 50 kg $K_2O \cdot ha^{-1}$.

Catchment no. 2 is a semi-intensively farmed and drained catchment (523) of an area of 41.7 ha. Situated in the vicinity of build-up areas in the village of Doba and a large farm, it consists of light soils of quality class V. The drainage system carries water away from fertilized farm fields (68 kg $N \cdot ha^{-1}$, 40 kg $P_2O_5 \cdot ha^{-1}$, 55 kg $K_2O \cdot ha^{-1}$ in 2005 for oats, 88 kg $N \cdot ha^{-1}$, 41 kg $P_2O_5 \cdot ha^{-1}$, 54 kg $K_2O \cdot ha^{-1}$ in 2006 for rye, and in 2007, the field was also sown with winter rye and the following fertilizer rates were applied: 140 kg $N \cdot ha^{-1}$, 52 kg $P_2O_5 \cdot ha^{-1}$, 50 kg $K_2O \cdot ha^{-1}$).

Catchment no. 3 is an intensively farmed catchment (528) covering the supply area of Pilwa Bay in the western part of Lake Dobskie. A surface watercourse drains an area of 1,616 ha which is 57%. afforested The watercourse was stagnant between July and October 2005 and between July and August 2006. Arable land in catchment no. 3 consists mostly of light loamy sands (soil quality class IV b). Triticale was grown in 2005, winter rape in 2006 and winter wheat in 2007. In the experimental period, the area was fertilized with 130 kg $N \cdot ha^{-1}$ and a spring – with slurry at the rate of 20 $m^3 \cdot ha^{-1}$ in 2005, and 206 kg $N \cdot ha^{-1}$ in 2006. In 2007, winter wheat was fertilized with 130 kg $N \cdot ha^{-1}$, 52 kg $P_2O_5 \cdot ha^{-1}$, 54 kg $K_2O \cdot ha^{-1}$.

Catchment no. 4 is a semi-intensively farmed catchment (530) draining the western part of Pilwa Bay catchment. It covers an area of 109 ha and is 60% afforested. The remaining parts of the catchment are covered by arable land on loamy sands (soil quality class V). In the analyzed period, the watercourse was stagnant in the summer and autumn. In 2005, oat was fertilized with 100 kg $N \cdot ha^{-1}$, 60 kg $P_2O_5 \cdot ha^{-1}$ and 50 kg $K_2O \cdot ha^{-1}$. In 2006 and 2007, rye was fertilized with 113 kg $N \cdot ha^{-1}$, 44 $K_2O \cdot ha^{-1}$ and 136 kg $N \cdot ha^{-1}$, 52 kg $P_2O_5 \cdot ha^{-1}$, 50 kg $K_2O \cdot ha^{-1}$, respectively.

The investigated areas are made up of soils with an average magnesium content of 3.4 to 7.0 Mg·100 g soil.

In line with the recommendations of the Nitrates Directive (DIRECTIVE 1991), the analyzed catchments were classified as a nitrate vulnerable zone where nitrogen pollution from agricultural sources has to be limited. This was determined in view of the nitrate content of underground water reaching 95 $mg \cdot dm^{-3}$ (Directive 2004a).

The following physical and chemical properties of water in the investigated area were determined: pH by potentiometry, temperature with the use of a thermometer integrated with an oxygen probe, oxygen saturation with the use of a WTW Multiline P3 meter, and electrolytic conductivity with the use of a Hanna conductivity meter. The flow of drained water and watercourses falling into the lake was measured according to a sample collection with the use of a VALEPORT 801 electromagnetic flow meter. Water samples were collected once a month in 2005-2007 and magnesium concentrations were determined by colorimetry with the use of Titanium yellow. Laboratory analyses were performed in accordance with the generally observed standards (HERMANOWICZ et al. 1999). The values of precipitation volume and ambient air temperatures were supplied by the Institute of Meteorology and Water Management based on the readouts of the meteorological station in Kętrzyn. The results were verified statically with the use of the Statistica 7 application.

Seasonal variations in water runoff, magnesium concentrations and load were computed by classifying samples into groups according to the season in which they were collected: winter (January – March), spring (April – – June), summer (July – September) and autumn (October – December). Magnesium loss was determined by multiplying magnesium levels and water flow volume. Magnesium loss per ha was determined by dividing the obtained loss values by the area of the drained catchment.

RESULTS AND DISCUSSION

Significant variations in weather conditions affecting plant development, magnesium uptake, water runoff and the loss of waterborne nutrients were observed in the investigated period. Based on the total volume and distribution of precipitation, the analyzed years were divided into two categories (according to KACZOROWSKA 1962): normal year (2005 – 545 mm) and two wet years (2006 – 640 mm; 2007 – 646 mm) – Figure 2. Variations in ambient air temperature were also noted with the average of 7.7°C, 8.1°C and 8.8°C in the successive years of the experiment. Periodic drying-up of the watercourses was observed in the summer and in early autumn throughout the entire experimental period. Below-zero air temperatures noted between December 2005 and March 2006 and in February 2007 led to the formation of snow and ice cover on the ground surface. The thawing of precipitation water in April 2006 and March 2007 resulted in the leaching of micronutrients from the ground into the watercourse.

Magnesium concentrations in water are determined by the rate of magnesium discharge from the soil and biosorption. The above processes are affected by physical and chemical properties such as oxygen saturation of water, temperature, pH and conductivity (Table 1). The highest oxygen saturation was reported in the spring and summer due to the intensive growth of vegetation, which significantly alters the properties of water.

Significant variations were also noted in water electrolytic conductivity (550-815 $\mu S \cdot cm^{-1}$). Conductivity values indicate that the content of water-soluble substances is higher in water evacuated via drains than via open watercourses and ditches, particularly in the autumn (KOC et al. 2002).

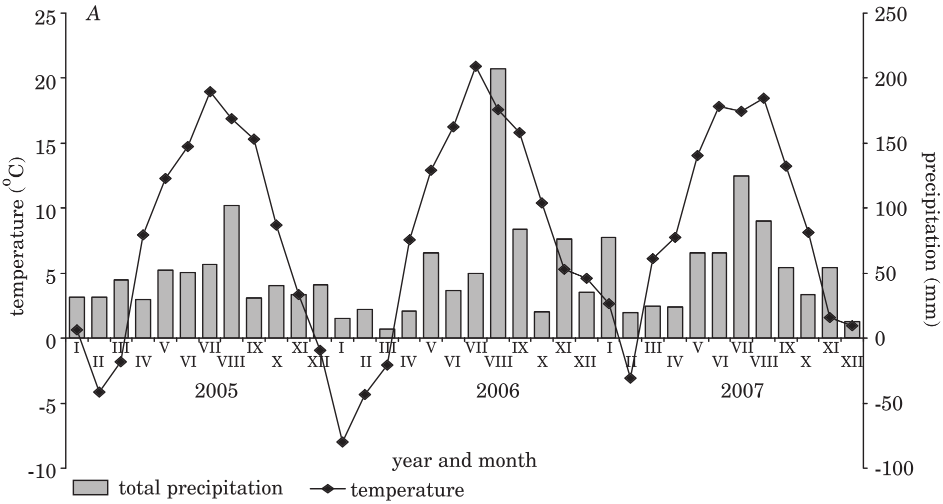

Fig. 2. Distribution of precipitation volume and ambient air temperature in 2005-2007

The fluctuations in Mg^{+2} levels in runoffs from agricultural catchments resulted from intensive farming as well as from the type of drainage system and weather conditions.

In the analyzed period, the above factors contributed to high variations in Mg^{+2} concentrations both in between and during the investigated years and seasons (Table 2).

An analysis of magnesium levels in water runoffs indicates that increased magnesium levels and the highest fluctuations in the range of 3.8 to 26.1 mg·dm^{-3} (SD=3.52, CV=49.45%) were noted in 2005, and fluctuations in the range of 3.3 to 15.7 mg·dm^{-3} (SD=3.52, CV=29.31%) were observed in 2007 in water drained from intensively farmed areas. Higher Mg^{+2} concentrations in runoffs from farming areas could be due to magnesium supplementation treatments as well as intensified Mg leaching from the soil in the non-growing season.

Lower fluctuations in magnesium levels (from 12.8 to 15.6 mg·dm^{-3} in 2006, and from 7.2 to 17.0 mg·dm^{-3} in 2007) were observed in watercourses with semi-intensively farmed catchments, and these findings are validated by the lowest standard deviation and coefficient of variation.

Throughout the entire experimental period, the highest magnesium levels, at 14.8 mg·dm^{-3} on average, were reported in the summer in water drained from intensively farmed areas (slurry application), although the analyzed nutrient was taken up by plants (Table 3). The above could be due to the fact that substances were dissolved in smaller quantities of water in seasons when more water was evaporated than precipitated (Koc et al. 2003). This hypothesis is supported by the electrolytic conductivity of water in the

Table 1

Seasonal variations in the physical and chemical properties of the inflows to Lake Dobskie in 2005-2007

Inflow (Type of use) \ Parameter	Season	Oxygen saturation (%)	Temperature (°C)	pH	Conductivity (μS·cm^{-1})
Drain 521 (intensive)	W*	58.3	4.5	7.2	762
	Sp	50.2	11.3	7.2	762
	Su	62.0	14.2	7.3	798
	A	62.0	7.5	7.3	815
	Y	58.1	9.1	7.3	782
Drain 523 (semi-intensive)	W	91.3	4.5	7.5	638
	Sp	84.4	9.6	7.6	672
	Su	92.4	13.6	7.6	681
	A	93.3	8.5	7.5	690
	Y	89.6	8.8	7.6	669
Watercourse 528 (intensive)	W	75.6	1.0	7.3	611
	Sp	73.0	17.0	7.8	612
	Su	86.0	15.4	7.7	550
	A	63.7	3.8	7.4	578
	Y	73.6	8.2	7.5	595
Watercourse 530 (semi-intensive)	W	89.2	1.3	7.7	580
	Sp	93.7	14.4	7.9	597
	Su	no flow			
	A	84.0	3.2	7.7	663
	Y	88.3	6.3	7.8	609

*W – winter; Sp – spring; Su-summer; A – autumn; Y – year

investigated period (Table 1). A slightly different trend was observed in water drained via watercourses. The highest Mg^{+2} concentrations of 14.1 mg·dm^{-3} were reported in the autumn in the watercourse draining semi- intensively farmed light soils, while the lowest levels at 11.3 mg·dm^{-3} were noted in runoffs from intensively farmed medium-heavy soils due to the biological sorption of magnesium by water vegetation. This process was not observed in water evacuated via drainage systems.

The highest seasonal fluctuations in magnesium levels (8.3-19.2 mg·dm^{-3}) were observed in water evacuated by the drainage system from the most intensively farmed area (Table 3). The above was validated by the results of prior research conducted in the Olsztyn Lakeland (Koc, Szymczyk 2003).

Based on an analysis of correlations between the investigated parameters, a negative correlation was determined between temperature, conductivity and magnesium concentrations only in the drain evacuating water from a semi-intensively farmed area (Table 4).

Table 2

Magnesium concentrations in the inflows to Lake Dobskie in 2005-2007

Type of inflow Type of use	2005			2006			2007			Mean for 2005-2007		
	mean X ----------- range of values	standard deviation (SD)	coefficient of variation (%) CV	mean X ----------- range of values	standard deviation (SD)	coefficient of variation (%) CV	mean X ----------- range of values	standard deviation (SD)	coefficient of variation (%) CV	mean X ----------- range of values	standard deviation (SD)	coefficient of variation (%) CV
Drain (521) intensive	13.8 ---------- 3.8-26.1	6.83	49.45	12.2 ---------- 7.7-17.8	3.07	25.24	12.0 ---------- 3.3-15.7	3.52	29.31	12.7 ---------- 3.3-26.1	4.75	37.47
Drain (523) semi-intensive	12.9 ---------- 7.1-19.6	3.44	26.68	10.7 ---------- 5.5-13.6	2.48	23.19	11.8 ---------- 7.8-24.5	4.77	40.34	11.8 ---------- 5.5-24.5	3.69	31.30
Watercourse (528) intensive	11.0 ---------- 7.0-13.6	2.23	20.28	12.6 ---------- 7.0-17.6	3.01	23.82	12.6 ---------- 8.2-24.5	4.38	34.76	12.2 ---------- 7.0-24.5	3.44	28.23
Watercourse (530) semi--intensive	10.9 ---------- 8.0-14.6	2.29	21.03	13.8 ---------- 12.8-15.6	1.09	7.89	12.1 ---------- 7.2-17.0	3.02	24.95	12.1 ---------- 7.2-17.0	2.54	20.97

Table 3

Average seasonal magnesium concentrations ($mg \cdot dm^{-3}$) in the inflows to Lake Dobskie in 2005-2007

Season	Type of inflow (type of use)	Year			Mean for 2005-2007
		2005	2006	2007	
Winter	drain 521 (intensive)	8.3	10.3	10.1	9.5
	drain 523 (semi-intensive)	9.3	10.8	15.5	11.9
	watercourse 528 (intensive)	10.0	12.0	15.6	12.5
	watercourse 530 (semi-intensive)	10.0	14.9	12.9	12.6
Mean		9.4	12.0	13.5	11.6
Spring	drain 521 (intensive)	17.2	13.7	11.8	14.3
	drain 523 (semi-intensive)	16.0	9.9	10.9	12.2
	watercourse 528 (intensive)	11.3	14.3	12.0	12.6
	watercourse 530 (semi-intensive)	10.5	12.8	8.8	10.7
Mean		13.8	12.7	10.9	12.4
Summer	drain 521 (intensive)	19.2	13.5	11.7	14.8
	drain 523 (semi-intensive)	13.9	11.0	9.0	11.3
	watercourse 528 (intensive)	dry	12.3	10.3	11.3
	watercourse 530 (semi-intensive)	dry	dry	dry	dry
Mean		16.5	12.3	10.3	12.5
Autumn	drain 521 (intensive)	10.6	10.6	14.5	11.9
	drain 523 (semi-intensive)	12.5	11.0	11.9	11.8
	watercourse 528 (intensive)	12.0	11.7	12.6	12.1
	watercourse 530 (semi-intensive)	14.6	13.4	14.3	14.1
Mean		12.4	11.6	13.3	12.5
Year	drain 521 (intensive)	13.8	12.2	12.0	12.7
	drain 523 (semi-intensive)	12.9	11.8	11.8	12.2
	watercourse 528 (intensive)	11.0	12.6	12.6	12.1
	watercourse 530 (semi-intensive)	10.9	13.8	12.1	12.3
Mean		12.1	12.6	12.1	12.3

The above supports the role of vegetation in removing Mg^{+2} from watercourses at favorable temperatures (KOC 1999).

A significant correlation (above 0.3) was observed between electrolytic conductivity and magnesium levels in water drained from an intensively farmed area. As confirmed by published sources, intensive fertilization supports the leaching of Mg^{+2} from the soil sorption complex (DOBRZAŃSKI, ZAWADZKI 1981).

The magnesium load evacuated by various means (ditches and drains) from farming areas was determined mostly by the level of farming intensity in the catchment (Table 5). Precipitation water collects and transports substances released from the soil into the water circulation system (PAŁKA-GUTOWSKA1996).

Table 4

Matrix of coefficients of correlation between magnesium concentrations and the selected physical and chemical properties of water in the inflows to Lake Dobskie

Parameter / Inflow	Oxygen saturation (%)	Temperature (°C)	pH	Conductivity (μS·cm^{-1})
Drain 521 (intensive)	0.02	0.24	0.13	0.39*
Drain 523 (semi-intensive)	0.26	-0.09	-0.12	0.30
Watercourse 528 (semi-intensive)	0.12	-0.05	-0.13	-0.38
Watercourse 530 (low intensity)	-0.16	-0.57*	-0.38	0.57*

*correlations were marked as significant at $p < 0.05$

Table 5

Seasonal and annual variation (kg·ha^{-1}·year^{-1}) in magnesium outflows per area unit in the investigated catchments

Season	Type of inflow (type of use)	Year			Mean for 2005-2007
		2005	2006	2007	
Winter	drain 521 (intensive)	14.9	1.9	5.2	7.3
	drain 523 (semi-intensive)	8.4	1.2	6.9	5.5
	watercourse 528 (intensive)	6.9	0.60	12.0	6.5
	watercourse 530 (semi-intensive)	9.7	0.8	10.8	7.1
Mean		10.0	1.1	8.7	6.6
Spring	drain 521 (intensive)	16.1	6.5	6.2	9.6
	drain 523 (semi-intensive)	3.2	0.7	1.4	1.8
	watercourse 528 (intensive)	3.7	0.5	1.9	2.1
	watercourse 530 (semi-intensive)	2.1	0.2	0.7	1.0
Mean		6.3	2.0	2.5	3.6
Summer	drain 521 (intensive)	8.2	1.8	1.0	3.7
	drain 523 (semi-intensive)	1.3	0.3	1.0	0.9
	watercourse 528 (intensive)	dry	0.3	0.7	0.3
	watercourse 530 (semi-intensive)	dry	dry	dry	dry
Mean		2.4	0.6	0.7	1.2
Autumn	drain 521 (intensive)	2.4	9.9	1.9	4.7
	drain 523 (semi-intensive)	1.9	2.6	1.7	2.1
	watercourse 528 (intensive)	1.2	4.4	2.5	2.7
	watercourse 530 (semi-intensive)	0.6	4.0	1.9	2.2
Mean		1.5	5.2	2.0	2.9
Year	drain 521 (intensive)	41.6	20.0	14.2	25.3
	drain 523 (semi-intensive)	14.8	4.8	11.0	10.2
	watercourse 528 (intensive)	11.8	5.9	17.2	11.6
	watercourse 530 (semi-intensive)	12.4	5.0	13.4	10.3
Mean		20.2	8.9	13.9	14.3

The highest annual Mg^{+2} load (per area unit) of 41.6 kg $Mg \cdot ha^{-1} \cdot year^{-1}$ was evacuated in 2005 from an intensively farmed area, mostly in the winter (38%) and in the spring (39%), as the highest water runoffs and magnesium concentrations were reported in those seasons.

In the course of the three experimental years, an average of 14.3 kg $Mg \cdot year$ was discharged per ha of agricultural areas. As regards both analyzed drainage systems, the highest magnesium outflows were reported in an intensively farmed area where the resulting loss was 2.5 times higher than in watercourses draining intensively and semi-intensively farmed areas as well the semi-intensively farmed area connected to a drainage network. A similar dependency was noted by Pulikowski et al. (2006). The highest Mg^{+2} loss was observed in the non-growing season, when around 46% of the relevant load (average for all catchments) was discharged in the winter due to magnesium leaching from the soil and the absence of magnesium uptake by plants (Banach et al. 2007).

Water and magnesium runoffs may be inhibited during a harsh winter. The absence of flow reported every summer in the two investigated watercourses limited magnesium outflows and minimized the resulting loss to 1.2 kg $Mg \cdot ha^{-1} \cdot year^{-1}$ over the investigated period.

CONCLUSIONS

1. The magnesium load evacuated with water runoffs from agricultural areas is determined by weather conditions, which modify runoff volume, the intensity of farming operations in the catchment and the type of drainage system. The greatest magnesium losses are observed during mild winters in drained and intensively fertilized areas.

2. Farming intensity in the catchment affected magnesium concentrations in drainage outflows. The highest average magnesium level of 12.7 $mg \cdot dm^{-3}$ with fluctuations in the range of 3.3 to 26.1 $mg \cdot dm^{-3}$ were reported in water drained from an intensively farmed area. Variations in the magnesium content of drainage water resulting from the level of farming intensity did not exceed 30%.

3. Around 50% of magnesium loss from drained areas was reported during mild and wet winters. Magnesium outflows were not observed during harsh winters. Summer loss accounted for around 26% of the average annual magnesium load and it was inhibited during draughts.

4. Every year, an average of 14.3 kg of magnesium is drained to surface water from 1 ha of agricultural catchments. The magnesium loss from intensively farmed areas is 2.5 times higher than in catchments characterized by semi-intensive farming.

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VERMICOMPOSTING OF DUCKWEED (*LEMNA MINOR* L.) BIOMASS BY *EISENIA FETIDA* (SAV.) EARTHWORM

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Abstract

This work presents the dynamics of *E. fetida* (Sav.) earthworm populations during vermicomposting of duckweed (*Lemna minor* L.) biomass in small containers, and provides properties of the vermicomposts produced. An experiment was conducted under laboratory conditions (in darkness, at an average temperature 25±5°C, with substrate moisture 70-75%). Test pots (3 replications for each duckweed treatment) were filled with one litre of garden soil, into which 100 individuals of *E.fetida,* of known biomass, were introduced per pot. Duckweed was fed to earthworms regularly, in two treatments: (1) duckweed + + cattle manure (1:1), and (2) duckweed only.

Earthworm number and biomass of tested populations were determined after 4 months of vermicomposting, and it was found that an average number of *E.fetida* in containers with duckweed and manure was 121±5 ind./container with a total biomass of 25.8±1.1 g. Populations in pure duckweed were significantly smaller ($p<0.05$), with 57±6 ind./container and a total biomass of 9.8±1 g. Cocoon production was also different across treatments. Populations in duckweed alone produced 55±13 cocoons /per container, significantly less ($p<0.05$) than the 231±37 cocoones when manure was added.

Duckweed vermicomposts were odourless and had good granular structure. Chemical characteristics of both vermicompost types (with or without a manure supplement) were desirable. Content of macroelements in duckweed vermicomposts was high, whereas microelements, cadmium and lead were within the permitted levels, making these vermicomposts extremely useful in environmental reclamation, including agriculture. The manure addition was important for characteristics and chemical content of duckweed vermicomposts. The vermicomposts produced from duckweed and manure contained more ash, N, P, K, Mg, Zn, Cu, Ni, Cr, Cd and Pb.

Key words: *Eisenia fetida* (Sav.), duckweed *Lemna minor* L., vermicompost, chemical composition.

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WERMIKOMPOSTOWANIE BIOMASY RZĘSY DROBNEJ (*LEMNA MINOR* L.) Z UDZIAŁEM DŻDŻOWNICY *EISENIA FETIDA* (SAV.)

Abstrakt

W publikacji zaprezentowano stan populacji dżdżownic kompostowych *E. fetida* (Sav.) podczas wermikompotowania biomasy rzęsy drobnej *Lemna minor* L. Opisano także cechy wyprodukowanych wermikompostów. Doświadczenie prowadzono w warunkach laboratoryjnych (w ciemności, średnia temp. 25±5°C, wilgotność podłoża 70-75%). Wazony testowe (w 3 powtórzeniach dla każdej kombinacji z utylizowanej rzęsy) wypełniono ziemią ogrodową o objętości 1 dm^3. Do tych podłoży wprowadzono po 100 osobników *E. fetida* o znanej biomasie. Do utylizacji podawano regularnie biomasę rzęsy, w dwóch kombinacjach: (1) rzęsa + obornik bydlęcy (1:1), (2) rzęsa.

Liczebność i sumę biomasy testowanych populacji dżdżownic wyznaczono po 4 miesiącach wermikompostowania. W wazonach z rzęsą i obornikiem stwierdzono istotnie wyższą średnią liczebność *E.fetida* (121±5 os./wazon) i sumę biomasy (25.8±1.1 g), w porównaniu ze zredukowaną populacją w czystej rzęsie (57±6 os./wazon i 9.8±1 g biomasy). Podobne zależności stwierdzono w przypadku produkcji kokonów. Dżdżownice złożyły istotnie mniej kokonów, utylizując czystą rzęsę, bez dodatku obornika bydlęcego (odpowiednio 55±13 i 231±37 kokonów/wazon).

Dodatek obornika bydlęcego różnicował także cechy wermikompostu z rzęsy drobnej. Wermikompost z rzęsy z dodatkiem obornika bydlęcego zawierał więcej popiołu, N, P, K, Mg, Zn, Cu, Ni, Cr, Cd oraz Pb.

Słowa kluczowe: *Eisenia fetida* (Sav.), rzęsa drobna *Lemna minor* L., wermikompost, skład chemiczny.

INTRODUCTION

Presently, natural methods of sewage treatment are preferred all over the world. One possible method of communal wastewater treatment is to employ a duckweed (*Lemna minor*. L.) system, which captures nutrients present in wastewater (Bonomo et al. 1997, Riggle 1998, Steen et al. 1999). *Lemna*-type systems are used mainly in natural, municipal sewage or agricultural organic waste treatment systems. Duckweed grows profusely in stagnant water, demanding hardly any agro-technical operations, but its growth is determined mainly by environmental resources (Hamid 1993). In clean water, duckweed grows slowly and contains much fiber, ash and carbohydrates, whereas in sewage-fed ponds it grows quickly and contains more protein (Miztar, Slinger 1976). In a biological sewage treatment plant, duckweed acts as a natural sewage treating factor by absorbing nutrients, mainly nitrogen and phosphorus. Depending on the climatic conditions, the system works at various efficiency levels, but during the growing season a large amount of biomass is created and becomes bio-waste. Duckweed requires environmental utilization after being removed from the water.

Since composting of duckweed biomass progresses slowly, an attempt was undertaken to vermicompost it with an aid of *Eisenia fetida* (Sav.) earthworms. As described in numerous publications (BARAN et al. 1996, MAZUR et al. 1996, GĄSIOR et al. 1998, KOSTECKA et al. 1998, ZABŁOCKI, KIEPAS-KOKOT 1998, MAZUR et al. 2000, BURY et al. 2001, KALEMBASA 2001, KANIUCZAK et al. 2001), the nutrient value of vermicompost is significantly influenced by the origin, type and proportions of the (mixed) organic wastes utilized.

MATERIAL AND METHODS

Earthworms and substrates

Earthworms of the species *Eisenia fetida* (Sav.) were obtained from a culture bank (maintained since 1996) at the Institute of the Natural Basis for Agriculture, Faculty of Biology and Agriculture – now Rzeszow University.

Duckweed (*L. minor*) was taken from the municipal wastewater treatment plant located in Łąka near Rzeszow. Cattle manure, also used in the experiment, was obtained from a local farmyard and stored for several months after a high temperature treatment. Garden soil, also utilized, was characterized by pH (H_2O) of 6.6; 12.0 g N; 0.44 g P; 5.0 g K; 35.0 g Ca; 0.94 g Mg; 0.87g Na kg^{-1} determined using standard laboratory procedures (OSTROWSKA at al. 1991).

Experimental Design

Vermicomposting of duckweed biomass was conducted under laboratory conditions (in darkness, at an average temperature of 25±5°C, and a substrate moisture content of 70-75%). Ten-litre test pots (with 3 replications for each duckweed combination) were filled with garden the soil described above.

Into each pot, 100 individuals [15 adult and 85 immature of *E. fetida* of known biomass (24.1±1.1 g)] were introduced. Two treatments were established: (1) 500 ml of duckweed + cattle manure (1:1 dry weight), (2) 500 ml of duckweed (dry weight). The duckweed biomass was supplied regularly (on demand) as a feed to the earthworms.

Every month, June – October, earthworms were utilizing the same type of wastes and the state of their populations was examined. Using manual substrate segregation (with all specimens counted and weighed plus cocoons counted), average numbers and total biomasses of earthworm populations were determined. The data were expressed as means with standard deviations. The hypothesis of equal variances in both studied populations was tested using an appropriate version of Student *t*-test, at a significance level 0.05.

The composition of duckweed vermicomposts was also determined, with 10 samples of each vermicompost taken for analysis. Standard methodologies for chemical-agricultural research were used to perform the following determinations: dry mass at a temperature of 105°C, raw ash at 550°C, total organic carbon (TOC) by the wet dichromate potassium oxidation method, pH and electrical conductivity measured at a 1:5 sample/water ratio.

Total nitrogen was determined using Kiejdahl method in an automatic Kiel-Foss device, while total phosphorus was measured by the vanado-molybdate method. The total amounts of Ca, K and Na were determined by the photometric method having digested the samples in HNO_3:$HClO_4$:H_2SO_4 at a ratio of 20:5:1 in a TECATOR device, whereas the elements Mg, Fe, Mn, Cu, Zn, Co, Ni, Cr, Cd and Pb were determined by atomic absorption spectrometry (AAS). The data were expressed as means with standard deviations.

RESULTS

At the end of the experimental period an average number of *E.fetida* in containers with duckweed and manure was 121±5 ind./container with a total biomass of 25.8±1.1 g. Populations in pure duckweed were significantly smaller ($p<0.05$) with 57±6 ind./container and a total biomass of 9.8±1 g (Figures 1, 2). Cocoon production was also different across treatments. Populations in duckweed alone produced 55±13 cocoons /per container, significantly less ($p<0.05$) than the 231±37 cocoons /per container when manure was added (Figure 3). The duckweed vermicomposts produced were rich in plant nutrients (Table 1).

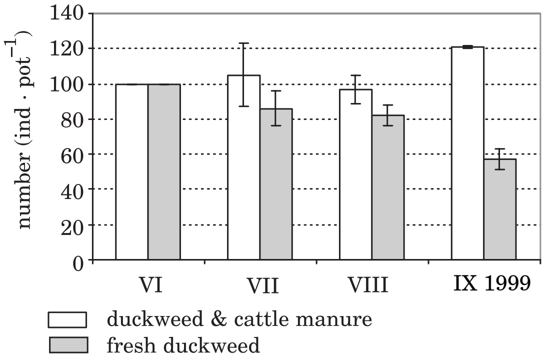

Fig. 1. Mean number (± SD) of *E. fetida* within given vermicomposting treatments

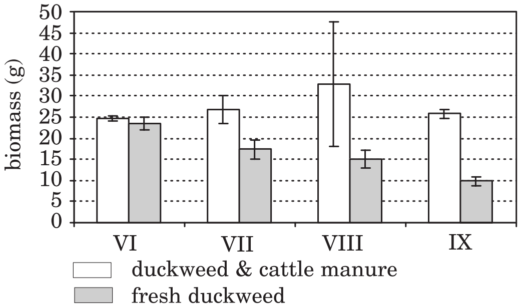

Fig. 2. Mean total biomass (± SD) of *E. fetida* within given vermicomposting treatments

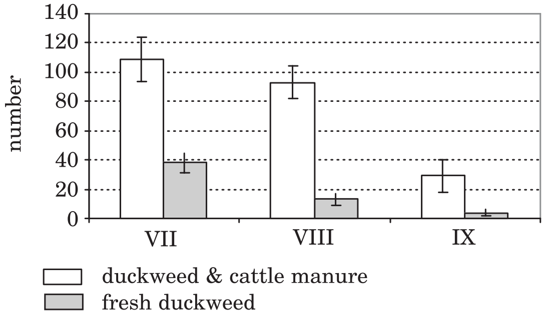

Fig. 3. Cocoon production (mean ± SD) of *E. fetida* within given vermicomposting treatments

DISCUSSION

Vermiculture biotechnology functions efficiently and quickly when density of earthworm is high (approximately 100 ind.$\cdot$dm^{-3}) (GADDIE, DOUGLAS 1977). It can be used to utilize different type of organic wastes (EDWARDS 1988, EDWARDS, BOHLEN 1996, ELVIRA et al 1997, 1999, NOGALES et al. 1999), and vermicompost thus produced can be useful and effective as a plant growth promoter (BUCKERFIELD et al. 1999, KOSTECKA, BŁAŻEJ 2000). As soils in many countries worldwide lose fertility and the amount of organic carbon and other minerals they contain is decreasing, we should use every opportunity to utilize organic waste to improve soil conditions.

Vermicomposting of duckweed caused a swift and odourless transformation of the substrate, which acquired characteristics, such as granular structure and content of nutrients, suitable for plants. Duckweed, nevertheless, was not a substrate that allowed populations of *E. fetida* to reproduce and grow to maximum levels. After four months of duckweed vermicomposting,

Table 1

Characteristics and chemical content of duckweed vermicomposts

Type of vermicompost		Produced from fresh duck-weed + cattle manure				Produced from fresh duck-weed			
Properties		average	SD	min	max	average	SD	min	max
%	ash	38.1	0.7	37.2	39.0	36.1	0.8	35	37
	TOC	36.5	0.3	36.0	36.8	37.6	0.5	37.1	38.3
pH in H_2O		-	-	5.5	6.0	-	-	4.3	4.6
$g \cdot kg^{-1}$ of dry mass	N	26.8	1.1	25	28	24.5	1.2	23	26
	P	5.7	0.2	5.4	6.0	4.4	0.4	4.0	4.8
	K	15.2	2.2	12.0	17.5	4.2	0.5	3.5	4.7
	Ca	2.3	0.2	2.0	2.5	2.9	0.2	2.6	3.1
	Mg	4.7	0.5	4.1	5.3	2.7	0.2	2.4	3.0
$mg \cdot kg^{-1}$ of dry mass	Na	844	214	600	1111	715	207	540	1055
	Fe	2463	423	2000	3000	2862	223	2600	3200
	Mn	638	34	600	680	665	35	635	726
	Zn	106	13	90	120	82	9.3	70	92
	Cu	52	8	40	60	45	17	22.5	65.0
	Ni	6.5	0.5	6.0	7.2	5.9	0.8	5.0	6.9
	Cr	5.3	1.3	4.0	7.0	3.84	1.11	2.55	5.02
	Co	0.87	0.14	0.7	1.05	1.6	0.5	1.20	2.34
	Cd	0.56	0.25	0.25	0.90	0.33	0.10	0.20	0.45
	Pb	8.9	0.9	7.8	10.0	7.97	0.85	7.02	9.27

the initial number and biomass had decreased by approximately 50%. This could have been a result of high earthworm density under experimental conditions. A reduction in the reproductive capacity of earthworm populations in vermicomposting beds and small containers has previously been recorded (MEYER, LOOTS 1999).

An addition of manure to the duckweed substrate supported maintenance of biomass and number of *E. fetida* populations (it also improved cocoon production). The presence of manure in the substrate also improved the characteristics of the vermicompost produced (larger amounts of N, P, K, Mg and Na than those produced with duckweed only). Manure also caused an increase in the pH of the vermicompost.

All vermicomposts produced with duckweed were characterized by large amounts of dry mass, including organic carbon and ash. However, these vermicomposts still included less ash than the ones produced with manure

(Gąsior et al. 1998) and household organic waste (Kostecka et al. 1999), sewage sludge, tannery waste plus straw (Mazur et al. 2000), duckweed plus sewage sludge 1:1 (Kaniuczak et al. 2001). The total amount of organic carbon (TOC) in these vermicomposts was higher than in the ones produced with manure (Gąsior et al. 1998), household organic wastes (Kostecka et al. 1999) and cattle manure (Nogales et al. 1999), sewage sludge (Baran et al. 1996), sewage sludge, tannery waste plus straw (Mazur et al. 2000), and lower than in vermicomposts produced with other wastes such as dry olive cake + cattle manure, olive cake + municipal biosolids, and dry olive cake + cattle manure + municipal biosolids (Nogales et al. 1999). Vermicomposting improves the quality of organic wastes, particularly through an increase in humic components which bind with heavy metals, making them less available to other organisms in the food chain. Stability of these organic and metallic complexes is highest at pH levels around 5, which are characteristic of many vermicomposts (Baran et al. 1996).

The pH level of all duckweed vermicomposts was lower than that of manure vermicomposts (Gąsior et al. 1998), household organic waste (Kostecka et al. 1999) and other wastes (Nogales et al. 1999).

Fresh duckweed is characterized by high amounts of nitrogen and phosphorus (Wo•niak, Zawora 1999) and vermicomposts made from it generally contain higher amounts of these elements than other vermicomposts (Gąsior et al. 1998, Kostecka et al. 1999, Nogales et al. 1999). It is important to stress that duckweed vermicomposts examined here contained more nitrogen, potassium and sometimes magnesium and sodium compared with vermicomposts produced by Mazur et al. (2000), or more nitrogen and potassium compared with vermicomposts produced by Bury et al. (2001) and Kalembasa (2001). These vermicomposts contained more nitrogen, phosphorus, potassium, magnesium and sodium compared with vermicomposts from duckweed and sewage sludge (Kaniuczak et al. 2001). The duckweed vermicomposts had a low ratio of C/N, so in order to avoid nitrogen loss to soils, carbohydrates should be provided (straw, cellulose, etc.). Vermicomposts produced from duckweed contained more microelements compared with household organic waste vermicomposts (Kostecka et al. 1999). The vermicomposts produced here, from duckweed and manure, contained more magnesium than vermicomposts from sewage sludge and tannery waste plus straw (Mazur et al. 2000), and also compared with vermicomposts produced from sewage sludge and sawdust (Zabłocki, Kiepas-Kokot 1998). Chemical analyses of our vermicomposts showed more magnesium and copper than found in vermicomposts produced from sewage sludge and additions: peat, sawdust, peat plus sawdust (Kalembasa 2001). Levels of iron, magnesium, zinc, copper and nickel were lower than in vermicomposts from sewage sludge plus additions: straw, hay, waste paper and leaves (Bury et al. 2001). Duckweed + + cattle manure vermicompost contained more zinc, copper, nickel, chromium, cadmium and lead and less iron and manganese than vermicomposts produced from pure duckweed. Vermicomposts produced from duckweed and duckweed plus manure contained less lead, cadmium and chromium than vermicomposts described by other authors (ZABLOCKI, KIEPAS-KOKOT 1998, MAZUR et al. 2000, BURY et al. 2001, KALEMBASA 2001). They also contained less cobalt, nickel, cadmium, chromium and lead compared with vermicomposts from duckweed plus sewage sludge (KANIUCZAK et al. 2001) and vermicomposts produced from sewage sludge and sawdust (ZABLOCKI, KIEPAS-KOKOT 1998). The levels of zinc, copper, nickel, chromium, lead and cadmium in duckweed vermicomposts was within the legal requirements for soil fertility improvement products (Rozp. Min. Roln. i Rozw. Wsi, 2001).

CONCLUSIONS

1. The earthworm *E. fetida* utilized duckweed biomass from a *Lemna*-type biological sewage treatment plant but had better population characteristics when cattle manure was added.

2. Duckweed vermicomposts were odourless and had good granular structure. Chemical characteristics of both vermicompost types (with or without a manure supplement) were desirable.

3. Content of macroelements in duckweed vermicomposts was high, whereas microelements, cadmium and lead were within permitted levels, thus making these vermicomposts extremely useful in environmental reclamation, including agriculture.

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RESPONSE OF BUTTER LETTUCE (*LACTUCA SATIVA* L.) TO DIFFERENT FORMS OF NITROGEN FERTILIZERS WITH CHLORINE AND SULPHATES

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Abstract

The objective of the experiment was to determine the effect of nutrients (different doses of chlorine, sulphates and calcium as well as different forms of nitrogen fertilizers: $Ca(NO_3)_2$ + NH_4NO_3, NH_4NO_3, NH_4NO_3 + $CaCl_2$, $NaNO_3$) on yield and content of N-NO_3, $N_{og.}$, P, K, Ca, Mg and Na in butter lettuce (*Lactuca sativa* L.) cv. Vilmorin. A pot experiment with four replications was repeated three times in 2007 (from 30th January and from 30th March) and in 2008 (from 31st March). It was established in the 'Plant House' of the Chair of Agricultural Chemistry and Environmental Protection UWM in Olsztyn. Lettuce seedlings were planted into pots containing 2 dm^3 substratum. Nitrogen was applied three times, every 10 days, each dose consisting of 100 mg $N \cdot dm^{-3}$ of substratum applied to soil; identical doses of phosphorus, potassium and magnesium were introduced to the substratum before planting lettuce. Irradiation period was 12 hours every day. Lettuce was harvested after 6 weeks. Determinations of N_{og} (Kjedahl's method), P (vanadium-molybdenum method), K, Ca, Na (flame photometry – ESA), Mg (atomic absorption spectrophotometry – ASA) were performed having wet mineralised plant samples in H_2SO_4. Concentration of nitrates (V) was determined in fresh matter (potentiometrically, using a ion-selective electrode) in a 0.03 $mol \cdot dm^{-3}$ CH_3COOH according to Nowosielski (1988). The smallest number of lettuce heads (statistically significant) was obtained after $NaNO_3$ fertilization. The concentration of N-NO_3 declined advantageously following an $NaNO_3$ treatment. In contrast, the highest accumulation of nitrates(V) occurred after application of lime saltpetre in conjunction with ammonium nitrate. The composition of minerals in butter lettuce cv. Vilmorin, such as phosphorus, potassium, magnesium and sodium, was more beneficially affected by fertilization with sulphate compounds (K_2SO_4 + $MgSO_4$) rather than chlorine compounds (KCl + $MgCl_2$). Soda nitre significantly depressed the content of magnesium and calcium in plants, had no effect on the concentration of phosphorus and increased accumulation of sodium and potassium.

Key words: *Lactuca sativa* L., macroelements, N-NO_3, nitrogen fertilizers.

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REAKCJA SAŁATY MASŁOWEJ (*LACTUCA SATIVA* L.) NA RÓŻNE FORMY NAWOZÓW AZOTOWYCH Z UDZIAŁEM CHLORU I SIARCZANÓW

Abstrakt

Celem pracy było określenie wpływu składników pokarmowych (zróżnicowanych dawek chloru oraz siarczanów i wapnia, a także różnych form nawozów azotowych: $Ca(NO_3)_2$ + NH_4NO_3, NH_4NO_3, NH_4NO_3 + $CaCl_2$, $NaNO_3$) na plon oraz zawartość $N\text{-}NO_3$, Nog., P, K, Ca, Mg i Na w sałacie masłowej (*Lactuca sativa* L.) odmiany 'Vilmorin'. Doświadczenie wazonowe w 4 powtórzeniach prowadzono 3-krotnie w 2007 r. (od 30.01. i od 30.03) i 2008 r. (od 31.03) w „Rośliniarni" Katedry Chemii Rolnej i Ochrony Środowiska UWM w Olsztynie. Rozsadę sałaty wysadzono do wazonów o pojemności 2 dm^3 podłoża. Azot aplikowano 3-krotnie, co 10 dni, w ilości 100 mg $N \cdot dm^{-3}$ podłoża; fosfor, potas i magnez w jednakowych dawkach stosowano przed wysadzeniem sałaty. Długość naświetlania roślin wynosiła 12 h dziennie. Zbiór przeprowadzono po 6 tygodniach. Oznaczenia zawartości Nog. (metodą Kjeldahla), P (metodą wanadowo-molibdenową), K, Ca, Na, (metodą fotometrii płomieniowej – ESA), Mg (metodą absorpcyjnej spektrometrii atomowej – ASA) dokonano po mineralizacji „na mokro" w H_2SO_4. Koncentrację azotanów(V) oznaczono w świeżej masie (potencjometrycznie z użyciem elektrody jonoselektywnej), w wyciągu 0,03 $mol \cdot dm^{-3}$ CH_3COOH wg NOWOSIELSKIEGO (1988). Najmniejszą masę główek sałaty (istotną statystycznie) otrzymano po nawożeniu $NaNO_3$. Korzystne zmniejszenie ilości $N\text{-}NO_3$ nastąpiło po zastosowaniu $NaNO_3$, natomiast największe nagromadzenie azotanów(V) stwierdzono po aplikacji saletry wapniowej łącznie z saletrą amonową. Na zawartość składników mineralnych w sałacie masłowej odmiany 'Vilmorin', takich jak fosfor, potas, magnez i sód, korzystniej wpływało nawożenie związkami siarczanowymi (K_2SO_4 + $MgSO_4$) niż chlorkowymi (KCl + $MgCl_2$). Saletra sodowa znacząco wpłynęła na zmniejszenie zawartości magnezu i wapnia w roślinach, nie miała wpływu na koncentrację fosforu, jednocześnie zwiększała akumulację sodu i potasu.

Słowa kluczowe: *Lactuca sativa* L., makroelementy, $N\text{-}NO_3$, nawozy azotowe.

INTRODUCTION

Abundance and availability of mineral components in substratum condition the volume and quality of produced plant biomass. Wrongly balanced fertilization, particularly nitrogen treatments, often leads to unfavourable modifications of the chemical composition of crops. Excessive accumulation of nitrates(V) in green matter of plants is especially dangerous. Relevant references comprise many reports in the effect of substratum, agronomic practice, climatic conditions as well as genetic traits of crops on accumulation of nitrates and their effect on plant produce (DURMAN, CUSTIC 1990, KARIMAEI et al. 2004, JAROSZ, DZIDA 2006, KOWALSKA et al. 2006, KOZIK 2006, WOJCIECHOWSKA et al. 2006).

The purpose of this study has been to determine the effect of nutrients (different doses of chlorine, sulphates and lime as well as different forms of nitrogen: $Ca(NO_3)_2$+NH_4NO_3, NH_4NO_3, NH_4NO_3+$CaCl_2$, $NaNO_3$, on yield and content of $N\text{-}NO_3$, $N_{og.}$, P, K, Ca, Mg and Na in butter lettuce (*Lactuca sativa* L.) cv. Vilmorin.

MATERIAL AND METHODS

A two-factor fertilization experiment was established three times, in 2007 (on 30th January and 30th March) and in 2008 (on 31st March) in the Plant House of the Chair of Agricultural Chemistry and Environmental Protection, University of Warmia and Mazury in Olsztyn. Butter lettuce (*Lactuca sativa* L.) cv. Vilmorin served as the test plant. Lettuce seedlings were planted to pots containing 2 dm^3 substratum. Nitrogen was applied three times, at ten-day intervals, in doses consisting 100 mg N·dm^{-3} substratum: identical doses of phosphorus, potassium and magnesium were introduced before planting the lettuce (Table 1).

Table 1

Doses of nutrients in mg per dm^3 of substratum

No	Objects		N	P	K	Mg	Ca	Cl	S-SO_4	Na
0			0	0	0	0	0	0	0	0
1	sulphates or chlorides	$Ca(NO_3)_2$+NH_4NO_3	300	100	150	50	429	0 (94)	75 (0)	0
2		NH_4NO_3	300	100	150	50	0	0 (94)	75 (0)	0
3		NH_4NO_3+$CaCl_2$	300	100	150	50	429	381 (475)	75 (0)	0
4		$NaNO_3$	300	100	150	50	0	0 (94)	75 (0)	495

The irradiation period was 12 hours daily. Lettuce plants were harvested after 6 weeks. The determinations of N_{og} (Kjeldahl's method), P (vanadium molybdenum method), K, Ca, Na (flame photometry ESA), Mg (atomic absorption spectrophotometry – ASA) were performed after wet mineralization in H_2SO_4. Concentration of nitrates (V) was determined in fresh matter (potentiometrically using an ion selective electrode) in a 0.03 mol·dm^{-3} CH_3COOH extract according to Nowosielski (1988). The first experimental factor consisted of two forms of potassium and magnesium fertilizer: sulphate and chloride (KH_2PO_4+K_2SO_4+$MgSO_4$; KH_2PO_4+KCl+$MgCl_2$). The second factor encompassed 4 variants of nitrogen fertilizers, which involved two forms of nitrogen fertilizers: NO_3^- i NH_4^+; ($Ca(NO_3)_2$+NH_4NO_3, NH_4NO_3, NH_4NO_3+$CaCl_2$, $NaNO_3$). During the harvest, fresh matter of lettuce heads was weighed. The substratum prepared from mineral soil, before fertilization treatments, possessed the following physical and chemical properties: pH_{H2O} – 6.3, EC – 0.51; in 0.03 mol·dm^{-3} CH_3COOH extract: Ca – 1000.2 mg, Mg – 46.3 mg, K – 219.5 mg, Na – 185.6 mg, Cl – 66.1 mg, P – 23.2 mg, S-SO_4 – 218.6 mg, N-NH_4 – 10, N-NO_3 – 58.5 mg dm^{-3} of the substratum.

The results were processed statistically using analysis of variance for a two-factor experiment established in a completely randomized design, with an aid of Statistica 7.0 software.

RESULTS AND DISCUSSION

The fertilization treatments applied in this experiment significantly modified both yields and concentration of minerals in butter lettuce dry matter (Figures 1 and 2). The highest yield of butter lettuce was obtained when lime saltpetre was introduced to the substratum along with ammonium nitrate. The yield obtained when soda nitre had been applied was 18.7% lower compared to the highest yield, but it was still 47.5 g higher than the con-

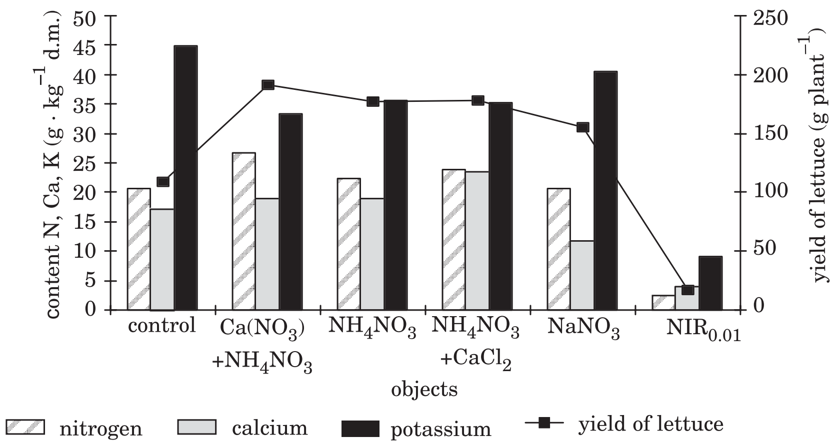

Fig. 1. Yield and content of N, Ca and K in dry matter of butter lettuce leaves depending on the form of nitrogen fertilizers

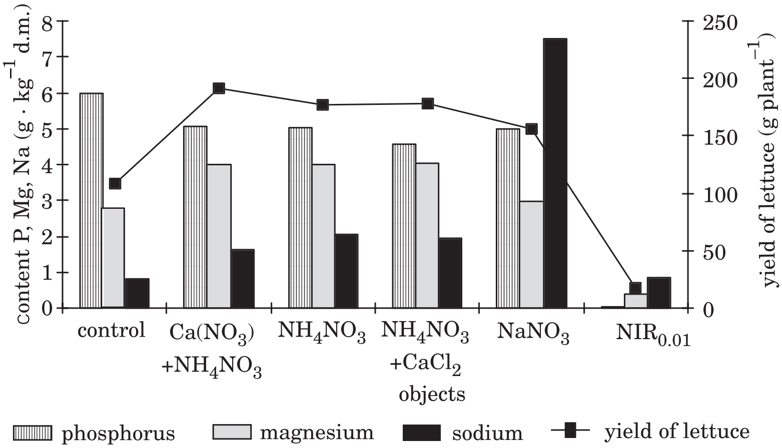

Fig. 2. Yield and content of P, Mg and Na in dry matter of butter lettuce leaves depending on fertilization

trol. Although lettuce is not tolerant to chlorine, in this experiment no significant changes in lettuce yield occurred as a result of introduction of ammonium nitrate or ammonium nitrate with calcium chloride and consequent increase in the concentration of chlorine in the substratum. These results are further confirmed by tests conducted by JAROSZ and DZIDA (2006) involving different forms of potassium fertilizer (KCl, KNO_3, K_2SO_4).

The level of total nitrogen in lettuce leaves was slightly higher (by 2.75 $g \cdot kg^{-1}$ d.m. on average) versus the control (Figure 1). The lowest concentration of this element occurred in the objects fertilized with soda nitre. When this compound was added together with calcium chloride, no further decrease in the total nitrogen concentration in lettuce leaves was recorded. The significantly highest amount of total nitrogen in dry matter was found following the fertilization treatment with $Ca(NO_3)_2$+NH_4NO_3, especially in the objects to which sulphates were introduced.

Lime applied as calcium saltpetre did not cause increased levels of calcium in lettuce leaves (Figure 1). Significant increase, however, in the concentration of this element was found after introduction of calcium in the form of $CaCl_2$. Significant decrease in the content of calcium occurred after an application of soda nitre (a 31.7% decline versus the control).

The forms of nitrogen fertilizers tested in the current experiment caused a significant drop in the content of potassium in lettuce leaves as compared to the control (Figure 1). Similar results are reported by JAROSZ and DZIDA (2006), who discovered that higher nitrogen rates were associated with decreasing concentrations of potassium in lettuce. The content of potassium in lettuce dry matter was least affected by nitrogen added to the substratum in the form of soda nitre. In the other experimental variants, the concentration of potassium was depressed by 9.4 to 11.7 K kg^{-1} d.m. relative to the zero object.

The content of magnesium in lettuce heads rose significantly (by an average 1.25 $g \cdot kg^{-1}$ d.m. versus the unfertilized lettuce) following the introduction of lime saltpetre, ammonium nitrate as well as its combination with calcium chloride (Figure 2). Soda nitre caused a much smaller increase (0.2 $g \cdot kg^{-1}$ d.m.). Similarly to the results reported by PERUCKA et al. (2007), no effect of additionally introduced calcium ions on the concentration of magnesium in lettuce leaves was determined.

The concentration of sodium in butter lettuce leaves depended primarily on the form of nitrogen fertilizers applied (Figure 2). In the objects fertilized with soda nitre, the content of sodium in lettuce leaves rose nearly 9.3-fold relative to the control object. In the other objects, the content of Na was from two- to 2.5-fold higher than in the unfertilized object.

No statistically proven differences were found in the concentration of phosphorus in lettuce, as dependent on the different forms of nitrogen fertilizers tested in this experiment (Figure 2). Significant differences in the magnesium content occurred in the concentration of this element appeared depending whether the lettuce was fertilized with chlorides or with sulphates (Figure. 3). Sulphates stimulated accumulation of magnesium. The differences in the levels of the other macroelements (N, P, K, Ca, Mg and Na) in lettuce leaves depending on sulphate or chloride fertilization were not significant statistically.

The chloride and sulphate form did not have statistically verified effect on the concentration of nitrates in lettuce. However, it was possible to notice the influence of these two forms on the concentration of nitrates depending on the age of the lettuce organs (Figure 4). The oldest leaves contained more nitrates (V) following an application of chloride rather than sulphate forms, whereas inner leaves accumulated more N-NO_3 under the

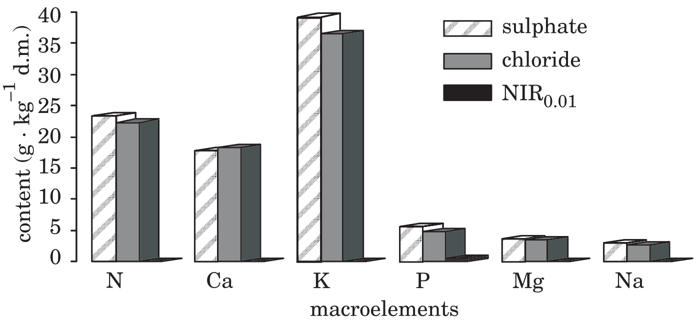

Fig. 3. Yield and content of N, Ca, K, P, Mg and Na in dry matter of butter lettuce leaves depending on the sulphate and chloride fertilization

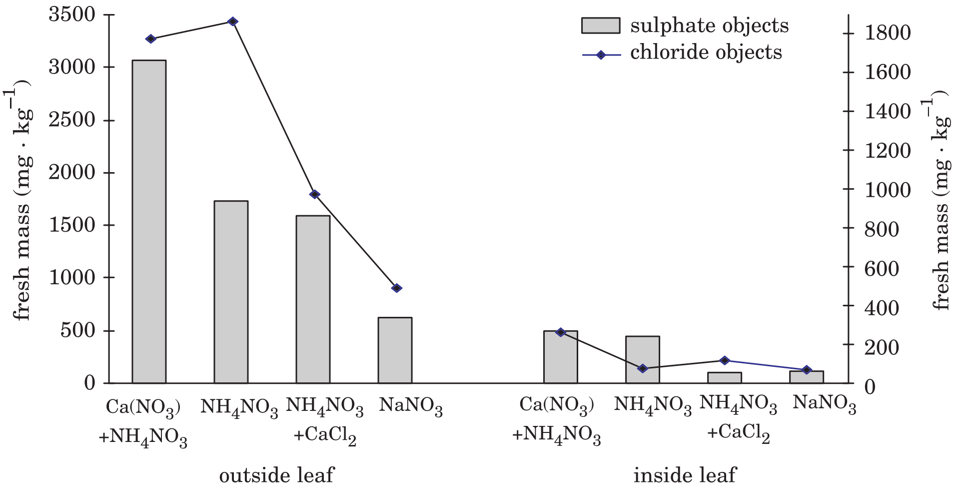

Fig. 4. Concentration of nitrates(V) in outer and inner leaves of butter lettuce in mg·kg^{-1} fresh matter depending on the experimental factors

influence of sulphate rather than chloride fertilizers. When $CaCl_2$ was included in a fertilization dose, the amount of N-NO_3 in lettuce leaves was lower than in the ammonium nitrate fertilized objects. There are many reports on the beneficial effect produced by chlorine used as KCl consisting of depressed levels of nitrates in crops (Michałojć 2000, Nurzyński et al. 2001, Jarosz, Dzida 2006, Kozik 2006). The highest levels of nitrates were found in the lettuce fertilized with ammonium nitrate combined with lime saltpetre. The lowest concentration of nitrates, four-fold less than caused by the $Ca(NO_3)_2$+NH_4NO_3 fertilization treatment, was found in lettuce plants treated with $NaNO_3$.

CONCLUSIONS

1. The lowest lettuce yield was produced when the crop had been fertilized with $NaNO_3$.

2. Depressed amounts of N-NO_3 occurred as a result of using $NaNO_3$. In contrast, the highest accumulation of nitrates(V) was caused by an application of lime saltpetre combined with ammonium nitrate.

3. Fertilization with sulphate compounds (KH_2PO_4+K_2SO_4+$MgSO_4$) rather than with chloride ones (KH_2PO_4+KCl+$MgCl_2$) had a more beneficial effect on the concentration of mineral components in butter lettuce cv. Vilmorin, such as nitrogen, phosphorus, potassium, magnesium and sodium.

4. Soda nitre significantly depressed the concentration of magnesium and calcium in lettuce, increased accumulation of sodium and potassium, but had no effect on the concentration of phosphorus.

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EFFECT OF MANGANESE DEFICIENCY ON GAS EXCHANGE PARAMETERS, LEAF GREENNESS (SPAD) AND YIELD OF PERENNIAL RYEGRASS (*LOLIUM PERENNE* L.) AND ORCHARD GRASS (*DACTYLIS GLOMERATA* L.)

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Abstract

The objective of this study was to determine the effect of manganese deficiency in soil on the rate of photosynthesis and transpiration, water use efficiency, leaf greenness and the yield of selected cultivars of perennial ryegrass (*Lolium perenne* L.) and orchard grass (*Dactylis glomerata* L.). During the growing season, the rate of photosynthesis and transpiration was measured using a LI-COR 6400 gas analyzer (Portable Photosynthesis System), and leaf greenness was estimated with a Minolta SPAD-502 chlorophyll meter. Water use efficiency (WUE) was calculated based on instantaneous values of photosynthesis and transpiration. Dry matter yield was determined by green matter drying to constant weight at 105°C. The results of the study indicate that the response of grasses to manganese deficiency in soil was dependent on plant species and cultivar. In the present experiment perennial ryegrass cultivars showed a stronger response to manganese deficit than orchard grass cultivars. Their response involved a decrease in the rate of photosynthesis and transpiration, and in the chlorophyll content of leaves. Among the tested cultivars, perennial ryegrass cv. Maja was found to be most sensitive to manganese deficiency in soil, as confirmed by the highest decrease in the values of all examined parameters.

Key words: manganese deficiency, perennial ryegrass, orchard grass, photosynthesis, transpiration, water use efficiency (WUE), leaf greenness (SPAD), yield.

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WPŁYW NIEDOBORU MANGANU NA WSKA•NIKI WYMIANY GAZOWEJ, INDEKS ZIELONOŚCI LIŚCI (SPAD) ORAZ PLONOWANIE ŻYCICY TRWAŁEJ (*LOLIUM PERENNE L*). I KUPKÓWKI POSPOLITEJ (*DACTYLIS GLOMERATA* L.)

Abstrakt

Celem pracy była ocena wpływu niedoboru manganu w glebie na intensywność fotosyntezy i transpiracji, współczynnik wykorzystania wody, indeks zieloności liści oraz plonowanie wybranych odmian życicy trwałej (*Lolium perenne* L.) i kupkówki pospolitej (*Dactylis glomerata* L.). W okresie wegetacji mierzono intensywność fotosyntezy i transpiracji za pomocą przenośnego analizatora gazowego LI-COR 6400 oraz indeks zieloności liści za pomocą optycznego chlorofilometru Minolta SPAD-502. Na podstawie chwilowych wartości fotosyntezy i transpiracji wyliczono współczynnik wykorzystania wody (WUE). Plon suchej masy określono przez wysuszenie zielonej masy w temperaturze 105°C, do stałej wagi. Wykazano, że reakcja traw na niedobór manganu w glebie zależy od gatunku rośliny i odmiany. Większą reakcję na deficyt manganu, polegającą na ograniczeniu procesu fotosyntezy i transpiracji oraz zmniejszeniu zawartości chlorofilu w liściach, wykazywały odmiany życicy trwałej niż kupkówki pospolitej. Najbardziej wrażliwa na niedobór manganu w glebie była odmiana życicy trwałej Maja, u której stwierdzono w największym stopniu ograniczenie wszystkich badanych parametrów.

Słowa kluczowe: niedobór manganu, życica trwała, kupkówka pospolita, fotosynteza, transpiracja, WUE, indeks SPAD, plonowanie.

INTRODUCTION

Manganese is essential for the growth and development of plants (Jankowski et al. 2000, Cioroi, Florea 2003). This element is a growth stimulator and an activator of numerous enzymatic processes. It also participates in nitrogen assimilation, the synthesis of proteins and vitamin C, respiration, as well as in the reactions of splitting water to liberate oxygen during photosynthesis (Grzyś 2004). Moreover, manganese affects chlorophyll persistence, and its deficiency causes chlorophyll breakdown under the influence of strong light. Manganese deficit disturbs normal growth and development of plants. The symptoms of manganese deficiency in grasses include leaf chlorosis, a slower growth rate and failure to response to nitrogen and iron (Falkowski et al. 1990). Manganese deficiency occurs most often in crops grown on neutral carbonate soils and on soils with a high humus content (Kabata-Pendias, Pendias 1999, Jodełka et al. 2000, Halasova et al. 2001, Kopittke, Menzies 2004).

The objective of this study was to determine the effect of manganese deficiency in soil on the rate of photosynthesis and transpiration, water use efficiency, leaf greenness and the yield of selected cultivars of perennial ryegrass and orchard grass.

MATERIALS AND METHODS

The experiment was conducted in a greenhouse of the University of Warmia and Mazury in Olsztyn. Kick-Brauckmann pots were filled with 10 kg of soil developed from loose sand, characterized by a low content of phosphorus (31 $mg \cdot kg^{-1}$), potassium (42 $mg \cdot kg^{-1}$), magnesium (13 $mg \cdot kg^{-1}$) and manganese (8.7 $mg \cdot kg^{-1}$). Soil reaction was slightly acidic – pH 5.8 dm^{-3} in 1 mol KCl. The experiment was performed in four replications. In each pot, 2 to 3 seeds of grass were sown at 10 points, and the plants were thinned immediately after emergence, leaving 7 plants per pot. The experiment involved two cultivars of perennial ryegrass (*Lolium perenne* L.): tetraploid Maja and diploid Argona, and two cultivars of orchard grass (*Dactylis glomerata* L.): tetraploid Dala and diploid Areda. Control pots were fertilized with a nutrient solution containing 1.00 g N as $CO(NH_2)_2$, 0.25 g P as KH_2PO_4, 1.00 g K as K_2SO_4 and 0.50 g Mg as $MgSO_4 \cdot 7H_2O$. A micronutrient solution (30 $ml \cdot pot^{-1}$), composed of 2.65 g Fe in EDTA, 0.09 g $MnCl_2 \cdot 4H_2O$, 0.1 g $ZnCl_2$, 0.03 g $CuCl_2 \cdot 2H_2O$, 0.12 g H_3BO_3 and 0.01 g $(NH_4)_6Mo_7O_{24} \cdot 4H_2O$ per kg of soil, was also applied. The remaining pots were not fertilized with manganese. During the growing season leaf greenness was estimated with a Minolta SPAD-502 chlorophyll meter, while the rate of photosynthesis and transpiration was measured using a LI-COR 6400 gas analyzer (Portable Photosynthesis System), at air temperature of around 25°C, a constant CO_2 concentration of 400 ppm and illumination of 1000 μmol m^{-2} s^{-1}. Four measurements were performed for each regrowth. Mean values for regrowths are presented in the paper. Water use efficiency (WUE) was calculated based on instantaneous values of photosynthesis and transpiration. The aboveground parts of plants were cut three times over the growing season. Dry matter yield was determined by green matter drying to constant weight at 105°C. The results were processed statistically with the use of Statistica software. The significance of differences was verified by Tukey´s test at $p = 0.99$.

RESULTS AND DISCUSSION

The tested cultivars showed low intensity of photosynthesis, ranging from 5.58 to 8.88 μmol CO_2 $m^{-2}s^{-1}$ on average (Table 1). Cv. Argona was marked by the highest rate of photosynthesis. Manganese deficiency in soil significantly limited photosynthesis intensity in all tested cultivars. The present results are consistent with the findings of other authors (Ohki 1985, Sienkiewicz-Cholewa 2002) who also observed a lower rate of photosynthesis in manganese-deficient treatments. In the present experiment the rate of photosynthesis decreased by 23% under manganese deficit conditions, com-

Table 1

Intensity of photosynthesis (μmol CO_2 m^{-2} s^{-1})

Cultivars	Fertilization	1st cut	2nd cut	3rd cut	Mean
Areda	control object manganese deficiency	5.98 ^{bc} 6.49 ^{bc}	9.17 ^{d} 7.43 ^{a}	7.38 ^{e} 4.62 ^{a}	7.51 ^{d} 6.18 ^{b}
Dala	control object manganese deficiency	7.16 ^{c} 5.55 ^{b}	8.39 ^{bc} 8.69 ^{cd}	5.92 ^{c} 6.59 ^{d}	7.16 ^{cd} 6.95 ^{c}
Argona	control object manganese deficiency	9.46 ^{d} 5.81 ^{b}	10.25 ^{e} 8.00 ^{b}	6.93 ^{d} 4.51 ^{a}	8.88 ^{e} 6.11 ^{b}
Maja	control object manganese deficiency	6.30 ^{bc} 3.07 ^{a}	10.43 ^{e} 8.13 b	9.00 ^{f} 5.53 ^{b}	8.58 ^{e} 5.58 ^{a}
Mean for cultivars					
Areda Dala Argona Maja		6.23 ^{b} 6.36 ^{b} 7.63 ^{c} 4.68 ^{a}	8.30 ^{a} 8.54 ^{a} 9.13 ^{b} 9.28 ^{b}	6.00 ^{b} 6.26 ^{c} 5.72 ^{a} 7.28 ^{d}	6.84 ^{a} 7.05 ^{a} 7.49 ^{b} 7.08 ^{a}
Mean for fertilization					
Control object Manganese deficiency		7.22 ^{b} 5.23 ^{a}	9.56 ^{b} 8.06 ^{a}	7.31 ^{b} 5.31 ^{a}	8.03 ^{b} 6.20 ^{a}
Mean for cuts		6.23 ^{a}	8.81 ^{b}	6.31 ^{a}	7.12 ^{a}

*homogeneous statistical groups (values marked with same letter did not differ statistically)

pared to control treatments. The response of perennial ryegrass cultivars to manganese deficiency was stronger than that of orchard grass cultivars, as confirmed by a higher decrease in photosynthesis intensity. This could be due to the ability of orchard grass to accumulate and store large quantities of manganese (DOBOSZYŃSKI, WASILEWSKI, 1983, MŁYNARCZYK, OLKOWSKI 1986, JARGIEŁŁO et al. 1991). Cv. Maja was found to be least resistant to manganese deficiency. The rate of photosynthesis decreased by 35% in this cultivar. In both species and in all cultivars the highest rate of CO_2 assimilation was recorded in the second regrowth.

Manganese deficiency limited water transpiration per unit leaf area. The value of this indicator was on average 24% lower in manganese-deficient treatments, in comparison with control treatments (Table 2). Orchard grass responded only slightly to manganese deficiency – the rate of transpiration in the tested cultivars of this species did not differ significantly subject to the level of manganese fertilization. On the other hand, the value of this parameter decreased by 34% and 42% in perennial ryegrass cultivars not fertilized with manganese. The highest rate of transpiration and the highest reduction in water evaporation under manganese deficit conditions were noted in cv. Maja. These results contradict the findings of OHKI (1985) who postulated that manganese deficiency had no effect on transpiration in common wheat. It should be also stressed that particular regrowths differed with respect to the rate of transpiration per unit leaf area. The highest rate

Table 2

Intensity of transpiration (m mol H_2O m^{-2} s^{-1})

Cultivars	Fertilization	1^{st} cut	2^{nd} cut	3^{rd} cut	Mean
Areda	control object manganese deficiency	0.90^{a} 1.32^{d}	0.92^{d} 0.68^{a}	0.52^{c} 0.28^{a}	0.78^{c} 0.76^{c}
Dala	control object manganese deficiency	1.04^{b} 1.05^{b}	0.77^{b} 0.80^{c}	0.53^{c} 0.51^{c}	0.78^{c} 0.79^{c}
Argona	control object manganese deficiency	1.21^{c} 0.81^{a}	1.31^{f} 0.81^{c}	0.38^{b} 0.28^{a}	0.96^{d} 0.64^{a}
Maja	control object manganese deficiency	2.03^{e} 1.05^{b}	1.19^{e} 0.76^{b}	0.36^{b} 0.26^{a}	1.19^{e} 0.69^{b}
Mean for cultivars					
Areda		1.11^{b}	0.80^{a}	0.40^{c}	0.77^{a}
Dala		1.05^{a}	0.78^{a}	0.52^{d}	0.78^{a}
Argona		1.01^{a}	1.06^{c}	0.33^{b}	0.80^{b}
Maja		1.54^{c}	0.97^{b}	0.31^{a}	0.94^{c}
Mean for fertilization					
Control object		1.29^{b}	1.04^{b}	0.45^{b}	0.93^{b}
Manganese deficiency		1.06^{a}	0.76^{a}	0.33^{a}	0.71^{a}
Mean for cuts		1.18^{c}	0.90^{b}	0.39^{a}	0.82^{b}

Explanations, see Table 1

of transpiration was recorded in the first regrowth, and then it gradually decreased.

Manganese deficiency in soil had no considerable influence on water relations in the investigated species and cultivars of grass. In the cultivars Argona and Maja water use efficiency decreased only slightly, by 3% and 5% respectively, while in the cultivars of orchard grass WUE increased to a slight degree (Table 3). A comparison of water use efficiency in the tested cultivars revealed that it reached the highest level in cv. Maja. In all cultivars the highest water use efficiency was observed in the third regrowth, which resulted from a low rate of transpiration.

Leaf greenness measurements showed that orchard grass cultivars contained less chlorophyll in leaves than perennial ryegrass cultivars (Table 4). Among the cultivars of perennial ryegrass, cv. Maja had a significantly higher chlorophyll content (44.83 SPAD units on average). Manganese deficit resulted in a decrease in chlorophyll concentrations, by 5.5% on average. Cv. Maja showed the strongest response to manganese deficiency, although the chlorophyll content of leaves decreased in this cultivar by only 8% on average. Literature on the subject provides no information on the impact of manganese fertilization on the chlorophyll content of grasses, which makes a detailed discussion of the results impossible. However, in a study conduct-

Table 3

Water use efficiency (µmol CO_2 m^{-2} s^{-1} · m mol H_2O m^{-2} s^{-1})

Cultivars	Fertilization	1st cut	2nd cut	3rd cut	Mean
Areda	control object	6.70 ^{c}	10.02 ^{b}	14.26 ^{bc}	10.33 ^{ab}
	manganese deficiency	4.93 ^{b}	10.96 ^{c}	16.64 ^{de}	10.84 ^{bc}
Dala	control object	6.89 ^{c}	10.93 ^{c}	11.28 ^{a}	9.70 ^{a}
	manganese deficiency	5.29 ^{b}	10.90 ^{c}	12.93 ^{ab}	9.71 ^{a}
Argona	control object	7.85 ^{c}	7.84 ^{a}	18.38 ^{e}	11.36 ^{c}
	manganese deficiency	7.16 ^{c}	9.88 ^{b}	15.99 ^{cd}	11.01 ^{bc}
Maja	control object	3.10 ^{a}	8.78 ^{b}	24.86 ^{g}	12.25 ^{d}
	manganese deficiency	2.93 ^{a}	10.78 ^{c}	21.07 ^{f}	11.56 ^{cd}
Mean for cultivars					
Areda		5.82 ^{b}	10.49 ^{c}	15.45 ^{b}	10.58 ^{b}
Dala		6.09 ^{b}	10.91 ^{d}	12.11 ^{a}	9.70 ^{a}
Argona		7.50 ^{c}	8.86 ^{a}	17.18 ^{c}	11.18 ^{c}
Maja		3.01 ^{a}	9.78 ^{b}	22.96 ^{d}	11.92 ^{d}
Mean for fertilization					
Control object		6.13 ^{b}	9.39 ^{a}	17.19 ^{a}	10.91 ^{a}
Manganese deficiency		5.08 ^{a}	10.63 ^{b}	16.66 ^{a}	10.79 ^{a}
Mean for cuts		5.60 ^{a}	10.01 ^{b}	16.93 ^{c}	10.85 ^{b}

Explanations, see Table 1

Table 4

Leaf greenness index (SPAD)

Cultivars	Fertilization	1st cut	2nd cut	3rd cut	Mean
Areda	control object	36.13 ^{bc}	42.03 ^{c}	40.85 ^{c}	39.67 ^{b}
	manganese deficiency	34.45 ^{a}	40.05 ^{b}	38.38 ^{b}	37.63 ^{a}
Dala	control object	35.43 ^{ab}	40.05 ^{b}	41.03 ^{c}	38.84 ^{b}
	manganese deficiency	36.43 ^{bc}	38.55 ^{a}	36.48 ^{a}	37.15 ^{a}
Argona	control object	39.95 ^{d}	43.18 ^{c}	41.58 ^{c}	41.57 ^{c}
	manganese deficiency	37.80 ^{c}	40.43 ^{b}	41.30 ^{bc}	39.84 ^{b}
Maja	control object	44.98 ^{e}	48.10 ^{e}	46.90 ^{e}	46.66 ^{e}
	manganese deficiency	40.45 ^{de}	45.85 ^{d}	42.60 ^{d}	43.00 ^{d}
Mean for cultivars					
Areda		35.29 ^{a}	41.04 ^{b}	39.61 ^{a}	38.64 ^{a}
Dala		35.93 ^{a}	39.30 ^{a}	38.75 ^{a}	37.99 ^{a}
Argona		38.88 ^{b}	41.80 ^{b}	41.44 ^{b}	40.70 ^{b}
Maja		42.72 ^{c}	46.98 ^{c}	44.75 ^{c}	44.83 ^{c}
Mean for fertilization					
Control object		39.12 ^{b}	43.34 ^{b}	42.59 ^{b}	41.69 ^{b}
Manganese deficiency		37.28 ^{a}	41.22 ^{a}	39.69 ^{a}	39.40 ^{a}
Mean for cuts		37.83 ^{a}	42.05 ^{b}	41.62 ^{b}	40.50 ^{b}

Explanations, see Table 1

Table 5

Dry matter yield (g $\cdot$ pot^{-1})

Cultivars	Fertilization	1st cut	2nd cut	3rd cut	Mean
Areda	control object manganese deficiency	7.43^{bc} 6.80^{a}	7.63^{a} 6.78^{a}	5.45^{ab} 4.53^{a}	20.50^{b} 18.10^{a}
Dala	control object manganese deficiency	8.15^{bc} 8.18^{bc}	7.43^{a} 7.08^{a}	5.68^{bc} 5.78^{bc}	21.25^{bc} 21.03^{bc}
Argona	control object manganese deficiency	7.50^{bc} 8.05^{bc}	7.13^{a} 6.50^{a}	6.45^{bc} 6.10^{bc}	21.08^{bc} 20.65^{bc}
Maja	control object manganese deficiency	8.73^{c} 6.48^{ab}	7.05^{a} 6.63^{a}	6.70^{c} 6.25^{bc}	22.48^{c} 19.36^{b}
Mean for cultivars					
Areda		7.11^{a}	7.20^{a}	4.99^{a}	19.30^{a}
Dala		8.16^{b}	7.25^{a}	5.73^{b}	21.14^{b}
Argona		7.78^{ab}	6.81^{a}	6.28^{bc}	20.86^{b}
Maja		7.61^{ab}	6.84^{a}	6.48^{c}	20.93^{b}
Mean for fertilization					
Control object		7.95^{b}	7.31^{b}	6.07^{b}	21.33^{b}
Manganese deficiency		7.38^{a}	6.75^{a}	5.66^{a}	19.79^{a}
Mean for cuts		7.71^{b}	7.06^{b}	5.87^{a}	20.64^{c}

Explanations, see Table 1

ed by OHKI (1985) manganese deficit in soil reduced chlorophyll concentrations in the leaves of common wheat, similarly as in the tested grass species. In the current experiment the lowest chlorophyll content of leaves was recorded in the first regrowth, regardless of a cultivar.

The yield of orchard grass cv. Areda was found to be the lowest, while the yields of the other investigated cultivars remained at a comparable level (Table 5). A lower yield of this cultivar was also noted in our earlier studies (OLSZEWSKA 2004, 2005, 2006). Manganese deficiency decreased the yield of grasses, by around 7% on average. The greatest effect of manganese deficit on yielding was observed in perennial ryegrass cv. Maja, whose yield declined by approximately 14% in comparison with control treatments. It should be stressed that this cultivar showed the strongest response to deficit in soil Mn, reflected by the highest drop in the values of all examined parameters.

CONCLUSIONS

1. The response of grasses to manganese deficiency in soil was dependent on plant species and cultivar. Perennial ryegrass cultivars showed a stronger response to manganese deficit than orchard grass cultivars. Their

response involved a decrease in the rate of photosynthesis and transpiration, and in the chlorophyll content of leaves.

2. Among the tested cultivars, perennial ryegrass cv. Maja was found to be most sensitive to manganese deficiency in soil, as confirmed by the highest decrease in the values of all examined parameters.

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EFFECT OF COPPER DEFICIENCY ON GAS EXCHANGE PARAMETERS, LEAF GREENNESS (SPAD) AND YIELD OF PERENNIAL RYEGRASS (*LOLIUM PERENNE* L.) AND ORCHARD GRASS (*DACTYLIS GLOMERATA* L.)

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Abstract

Copper is essential for the growth and development of plants. This micronutrient affects chlorophyll content, photosynthesis process and water relations in the plant. The objective of this study was to determine the effect of copper deficiency in soil on the rate of photosynthesis and transpiration, water use efficiency, leaf greenness and the yield of selected cultivars of perennial ryegrass (*Lolium perenne* L.) and orchard grass (*Dactylis glomerata* L.). During the growing season, the rate of photosynthesis and transpiration was measured using a LI-COR 6400 gas analyzer (Portable Photosynthesis System), and leaf greenness was estimated with a Minolta SPAD-502 chlorophyll meter. Water use efficiency (WUE) was calculated based on instantaneous values of photosynthesis and transpiration. Dry matter yield was determined by green matter drying to constant weight at 105°C. The results of the study indicate that copper deficiency significantly decreased the rate of photosynthesis and transpiration, chlorophyll concentration in leaves and the yield of all investigated cultivars. Perennial ryegrass cv. Maja was found to be most resistant to copper deficiency – it was characterized by a high rate of photosynthesis and transpiration, and by the highest chlorophyll content. The yield of cv. Maja attained under copper deficit conditions was comparable to that of other cultivars grown under control conditions.

Key words: copper deficiency, perennial ryegrass, orchard grass, photosynthesis, transpiration, water use efficiency (WUE), leaf greenness (SPAD), yield.

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WPŁYW NIEDOBORU MIEDZI NA WSKAŹNIKI WYMIANY GAZOWEJ, INDEKS ZIELONOŚCI LIŚCI (SPAD) ORAZ PLONOWANIE ŻYCICY TRWAŁEJ (LOLIUM PERENNE L). I KUPKÓWKI POSPOLITEJ (*DACTYLIS GLOMERATA* L.)

Abstrakt

Miedź jest mikroelementem niezbędnym do prawidłowego wzrostu i rozwoju roślin. Wpływa na zawartość chlorofilu, proces fotosyntezy oraz gospodarkę wodną roślin. Celem pracy była ocena wpływu niedoboru miedzi w glebie na intensywność fotosyntezy i transpiracji, współczynnik wykorzystania wody, indeks zieloności liści oraz plonowanie wybranych odmian życicy trwałej (*Lolium perenne* L.) i kupkówki pospolitej (*Dactylis glomerata* L.). W okresie wegetacji mierzono intensywność fotosyntezy i transpiracji za pomocą przenośnego analizatora gazowego LI-COR 6400 i indeks zieloności liści za pomocą optycznego chlorofilometru Minolta SPAD-502. Na podstawie chwilowych wartości fotosyntezy i transpiracji wyliczono współczynnik wykorzystania wody (WUE). Plon suchej masy określono przez wysuszenie zielonej masy w temperaturze 105°C do stałej wagi. Wykazano, że niedobór miedzi istotnie ograniczył intensywność fotosyntezy, transpiracji, poziom chlorofilu w liściach oraz plonowanie wszystkich badanych odmian. Odmianą najbardziej odporną na niedobór miedzi była Maja, która odznaczała się wysoką intensywnością fotosyntezy i transpiracji oraz największą zawartością chlorofilu. W warunkach niedoboru miedzi plonowała ona na poziomie pozostałych odmian uprawianych w warunkach kontrolnych.

Słowa kluczowe: niedobór miedzi, życica trwała, kupkówka pospolita, fotosynteza, transpiracja, WUE, indeks SPAD, plonowanie.

INTRODUCTION

Copper is essential for the growth and development of plants. This micronutrient influences chlorophyll content, participates in the processes of protein and carbohydrate synthesis, and positively affects the activity of nitrate reductase (Falkowski et al. 1990, Kuduk 1996, Terelak et al. 1998, Kabata-Pendias, Pendias 1999, Barczak et al. 2006). Moreover, copper has an impact on the photosynthesis process and water relations in the plant. Copper deficiency decreases the rate of photosynthesis and increases the rate of respiration (Ruszkowska, Wojcieska-Wyskupajtys 1996), and it induces changes in the structure of leaf epidermis and stem tissue. Under copper deficit conditions, stomata, parenchymal and sclerenchymal cells of the stem become smaller, while xylem cells undergo deformations. These undesirable changes limit water flow in the soil-plant system, leading to plant wilting (Dyki, Borkowski 2000). The copper content of plants varies widely depending on the part of plant, growth stage, species and variety, as well as on copper concentrations in soil and climate conditions (Kabata-Pendias 1996, Ruszkowska et al. 1996a).

The objective of this study was to determine the effect of copper deficiency in soil on the rate of photosynthesis and transpiration, water use efficiency, leaf greenness and the yield of selected cultivars of perennial ryegrass and orchard grass.

MATERIALS AND METHODS

The experiment was conducted in a greenhouse of the University of Warmia and Mazury in Olsztyn. Kick-Brauckmann pots were filled with 10 kg of soil developed from loose sand, characterized by a low content of phosphorus (31 mg·kg^{-1}), potassium (42 mg·kg^{-1}), magnesium (13 mg·kg^{-1}) and copper (4.5 mg·kg^{-1}). Soil reaction was slightly acidic - pH 5.8 dm^{-3} in 1 mol KCl. The experiment was performed in four replications. In each pot, 2 to 3 seeds of grass were sown at 10 points, and the plants were thinned immediately after emergence, leaving 7 plants per pot. The experiment involved two cultivars of perennial ryegrass (*Lolium perenne* L.): tetraploid Maja and diploid Argona, and two cultivars of orchard grass (*Dactylis glomerata* L.): tetraploid Dala and diploid Areda. Control pots were fertilized with a nutrient solution containing 1.00 g N as $CO(NH_2)_2$, 0.25 g P as KH_2PO_4, 1.00 g K as K_2SO_4 and 0.50 g Mg as $MgSO_4 \cdot 7H_2O$. A micronutrient solution (30 mg·pot^{-1}), composed of 2.65 mg Fe in EDTA, 0.09 mg $MnCl_2 \cdot 4H_2O$, 0.1 mg $ZnCl_2$, 0.03 mg $CuCl_2 \cdot 2H_2O$, 0.12 mg H_3BO_3 and 0.01 mg $(NH_4)_6Mo_7O_{24} \cdot 4H_2O$ per kg of soil, was also applied. The remaining pots were not fertilized with copper. P, K and Mg were applied pre-sowing at a single rate, while nitrogen and microelements were applied at three rates: pre-sowing, after the first and second cutting. During the growing season leaf greenness was estimated with a Minolta SPAD-502 chlorophyll meter, while the rate of photosynthesis and transpiration was measured using a LI-COR 6400 gas analyzer (Portable Photosynthesis System), at air temperature of around 25° C, a constant CO_2 concentration of 400 ppm, illumination of 1000 μmol m^{-2} s^{-1} and soil moisture content of around 70% of field water capacity. Measurements were taken on the youngest, fully developed leaf of shoots selected randomly in each treatment. Four measurements were performed for each regrowth, and readouts were taken at one-week intervals. Mean values for regrowths are presented in the paper. Water use efficiency (WUE) was calculated based on instantaneous values of photosynthesis and transpiration. The aboveground parts of plants were cut three times over the growing season: cut I – 9 July, cut II – 30 August, cut III – 25 October. Dry matter yield was determined by green matter drying to constant weight at 105°C. The results were processed statistically with the use of Statistica software. The significance of differences was verified by Tukey`s test at $p = 0.99$.

RESULTS AND DISCUSSION

Copper deficiency disturbs the conversion of light energy into chemical energy, thus reducing the rate of photosynthesis and photosynthetic activity in plants. The average rate of photosynthesis was low in the leaves of all investigated grass cultivars, and it ranged from 5.59 to 8.88 μmol CO_2 $m^{-2}s^{-1}$ (Table 1). Cv. Maja was marked by the highest rate of photosynthesis. The course of this process was widely differentiated in particular regrowths. The lowest rate of photosynthesis was observed in the first regrowth, while the highest – in the second regrowth. Copper deficiency in soil significantly limited photosynthesis intensity in the leaves of all tested cultivars. Compared to control treatments, the rate of photosynthesis decreased by 25%. Tetraploid cultivars were more resistant to copper deficiency than diploid cultivars. Among the tested cultivars, cv. Dala was found to be most resistant to copper deficiency. The rate of photosynthesis decreased to the slightest degree in this cultivar, i.e. by 15% on average, compared to 22-36% in the other cultivars. Orchard grass is known for its ability to accumulate copper. FALKOWSKI et al. (1990) reported that the concentration of this micronutrient was substantially higher in orchard grass than in perennial ryegrass under copper deficit conditions. This probably explains the weaker response of orchard grass to copper deficiency, in comparison with perennial ryegrass.

In the present study copper deficiency decreased transpiration intensity, on average by 31% (Table 2). The investigated cultivars differed in their responses to copper deficit. The weakest response was recorded in cv. Maja, while the strongest in cv. Argona – the rate of transpiration decreased in these cultivars by approximately 16% and 42% respectively. DYKA and BORKOWSKI (2000) demonstrated that in tomato plants copper deficiency caused a reduction in the thickness of the xylem layer and deformation of xylem cells as well as the closing of stomata, thus disturbing water transport and limiting transpiration. Perennial ryegrass cultivars evaporated more water than orchard grass cultivars. Particular regrowths differed with respect to the rate of transpiration, which was the fastest in the first regrowth and the slowest in the third regrowth.

Water relations in the plant can be determined based on water use efficiency (WUE). The highest water use efficiency was observed in the third regrowth, which resulted from a low rate of transpiration (Table 3). In orchard grass cultivars, higher values of WUE were recorded in treatments not fertilized with copper, whereas in perennial grass cultivars – in control treatments. Among the tested cultivars, the lowest WUE (mean value for regrowths) was reported for cv. Argona, while differences between the remaining cultivars were statistically non-significant.

Copper participates in chlorophyll biosynthesis (GRZYŚ 2004) – around 70% of copper can be found in chloroplasts. Copper deficiency in soil had a signifi-

Table 1

Intensity of photosynthesis (μmol CO_2 m^{-2} s^{-1})

Cultivars	Fertilization	1st cut	2nd cut	3rd cut	Mean
Areda	control object copper deficiency	5.98*^{cd} 3.36 ^{a}	9.17 ^{c} 8.29 ^{b}	7.38 ^{d} 5.12 ^{a}	7.51 ^{e} 5.59 ^{a}
Dala	control object copper deficiency	7.16 ^{e} 6.22 ^{d}	8.39 ^{b} 6.98 ^{a}	5.92 ^{b} 5.02 ^{a}	7.16 ^{d} 6.07 ^{b}
Argona	control object copper deficiency	9.46 ^{f} 4.36 ^{b}	10.25 ^{d} 6.82 ^{a}	6.93 ^{c} 5.87 ^{b}	8.88 ^{f} 5.69 ^{a}
Maja	control object copper deficiency	6.30 ^{d} 5.13 ^{bc}	10.43 ^{d} 8.96 ^{c}	9.01 ^{e} 5.91 ^{b}	8.58 ^{f} 6.66 ^{c}
Mean for cultivars					
Areda Dala Argona Maja		4.67 ^{a} 6.69 ^{c} 6.91 ^{c} 5.71 ^{b}	8.73 ^{b} 7.68 ^{a} 8.54 ^{b} 9.69 ^{c}	6.25 ^{b} 5.47 ^{a} 6.40 ^{b} 7.46 ^{c}	6.55 ^{a} 6.61 ^{a} 7.28 ^{b} 7.62 ^{c}
Mean for fertilization					
Control object Copper deficiency		7.22 ^{b} 4.77 ^{a}	9.56 ^{b} 7.76 ^{a}	7.31 ^{b} 5.48 ^{a}	8.03 ^{b} 6.00 ^{a}
Mean for cuts		5.60 ^{a}	8.66 ^{b}	6.39 ^{a}	7.02 ^{a}

*homogeneous statistical groups (values marked with same letter did not differ statistically)

Table 2

Intensity of transpiration (m mol H_2O m^{-2} s^{-1})

Cultivars	Fertilization	1st cut	2nd cut	3rd cut	Mean
Areda	control object copper deficiency	0.90 ^{b} 0.64 ^{a}	0.92 ^{e} 0.52 ^{b}	0.52 ^{c} 0.35 ^{a}	0.78 ^{d} 0.50 ^{a}
Dala	control object copper deficiency	1.04 ^{c} 0.92 ^{b}	0.77 ^{c} 0.44 ^{a}	0.53 ^{c} 0.36 ^{a}	0.78 ^{d} 0.57 ^{b}
Argona	control object copper deficiency	2.03 ^{e} 0.87 ^{b}	1.19 ^{f} 0.84 ^{d}	0.36 ^{a} 0.37 ^{a}	1.19 ^{f} 0.69 ^{c}
Maja	control object copper deficiency	1.21 ^{d} 1.10 ^{c}	1.31 ^{g} 0.84 ^{d}	0.38 ^{a} 0.48 ^{b}	0.96 ^{e} 0.81 ^{d}
Mean for cultivars					
Areda Dala Argona Maja		0.77 ^{a} 0.98 ^{b} 1.45 ^{d} 1.15 ^{c}	0.72 ^{b} 0.60 ^{a} 1.01 ^{c} 1.08 ^{d}	0.43 ^{b} 0.44 ^{b} 0.36 ^{a} 0.43 ^{b}	0.64 ^{a} 0.68 ^{b} 0.94 ^{d} 0.88 ^{c}
Mean for fertilization					
Control object Copper deficiency		1.29 ^{b} 0.88 ^{a}	1.04 ^{b} 0.66 ^{a}	0.45 ^{b} 0.39 ^{a}	0.93 ^{b} 0.64 ^{a}
Mean for cuts		1.09 ^{c}	0.85 ^{b}	0.42^{a}	0.79 ^{b}

Explanations, see Table 1

Table 3

Water use efficiency (μmol CO_2 m^{-2} s^{-1} · m mol H_2O m^{-2} s^{-1})

Cultivars	Fertilization	1st cut	2nd cut	3rd cut	Mean
Areda	control object copper deficiency	6.70 ^{b} 5.28 ^{a}	10.02 ^{b} 16.03 ^{d}	14.26 ^{c} 14.84 ^{c}	10.33 ^{b} 12.05 ^{d}
Dala	control object copper deficiency	6.89 ^{b} 6.74 ^{b}	10.93 ^{c} 15.95 ^{d}	11.28 ^{a} 13.96 ^{c}	9.70 ^{ab} 12.23 ^{d}
Argona	control object copper deficiency	4.66 ^{a} 5.02 ^{a}	8.63 ^{a} 8.17 ^{a}	19.11 ^{e} 16.09 ^{d}	10.80 ^{c} 9.80 ^{ab}
Maja	control object copper deficiency	5.23 ^{a} 4.67 ^{a}	7.97 ^{a} 10.63 ^{bc}	23.89 ^{f} 12.38 ^{ab}	12.36 ^{d} 9.23 ^{a}
Mean for cultivars					
Areda		5.99 ^{b}	13.02 ^{c}	14.55 ^{b}	11.19 ^{b}
Dala		6.81 ^{c}	13.44 ^{c}	12.62 ^{a}	10.96 ^{b}
Argona		4.83 ^{a}	8.40 ^{a}	17.60 ^{c}	10.28 ^{a}
Maja		4.95 ^{a}	9.30 ^{b}	18.14 ^{c}	10.80 ^{b}
Mean for fertilization					
Control object		5.87 ^{b}	9.39 ^{a}	17.14 ^{b}	10.80 ^{a}
Copper deficiency		5.43 ^{a}	13.70 ^{b}	14.32 ^{a}	10.81 ^{a}
Mean for cuts		5.65 ^{a}	11.04 ^{b}	15.73 ^{c}	10.81 ^{b}

Explanations, see Table 1

cant effect on chlorophyll concentrations in leaves. Leaf greenness (SPAD) values decreased by approximately 7-8% in all regrowths under copper deficit conditions (Table 4). Cv. Maja showed the strongest response to copper deficiency, which was manifested by an 11% decrease in the chlorophyll content of leaves. At the same time, this cultivar had the highest chlorophyll concentration (on average 42.73 SPAD). The lowest chlorophyll content of leaves was recorded in the first regrowth, regardless of a cultivar. This is consistent with our earlier findings (Olszewska 2006, Olszewska et al. 2008).

Perennial ryegrass cv. Maja was characterized by the highest yield of all tested cultivars, both under optimum fertilization conditions and under copper deficit conditions (Table 5). In this cultivar a high yield corresponded with a high rate of photosynthesis and transpiration, and with the highest chlorophyll content of leaves. Deficit in soil Cu caused a yield decline in all the cultivars, but statistically significant differences were observed only with regard to cv. Areda. Dry matter yield dropped on average by around 8%, and the highest decline was noted in cv. Dala (approx. 11%). In all the analyzed cultivars the strongest response to copper deficiency was reported in the second regrowth, which was reflected by the highest yield decrease.

Table 4

Leaf greenness index (SPAD)

Cultivars	Fertilization	1st cut	2nd cut	3rd cut	Mean
Areda	control object copper deficiency	36.13 [cd] 33.48 [a]	40.85 [b] 38.90 [a]	42.03 [d] 40.45 [bc]	39.67 [b] 37.61 [a]
Dala	control object copper deficiency	35.43 [bc] 34.40 [ab]	41.05 [bc] 37.25 [a]	42.03 [d] 38.85 [a]	39.50 [b] 36.83 [a]
Argona	control object copper deficiency	39.95 [e] 36.88 [cd]	43.18 [d] 41.43 [bc]	41.58 [cd] 39.65 [b]	41.57 [c] 39.32 [b]
Maja	control object copper deficiency	41.98 [f] 37.85 [d]	48.10 [e] 42.2 [c]	45.90 [e] 40.38 [bc]	45.33 [d] 40.14 [b]
Mean for cultivars					
Areda Dala Argona Maja		34.80 [a] 34.91 [a] 34.41 [b] 39.91 [c]	39.88 [a] 39.95 [a] 42.30 [b] 45.15 [c]	41.23 [b] 39.64 [a] 40.61 [b] 43.14 [c]	38.64 [a] 38.17 [a] 40.44 [b] 42.73 [c]
Mean for fertilization					
Control object Copper deficiency		38.37 [b] 35.63 [a]	43.29 [b] 39.94 [a]	42.88 [b] 39.83 [a]	41.51 [b] 38.48 [a]
Mean for cuts		37.01 [a]	41.82 [b]	41.16 [b]	39.99 [b]

Explanations, see Table 1

Table 5

Dry matter yield ($g \cdot pot^{-1}$)

Cultivars	Fertilization	1st cut	2nd cut	3rd cut	Mean
Areda	control object copper deficiency	7.38 [a] 7.23 [a]	7.63 [c] 6.50 [ab]	5.45 [ab] 5.03 [a]	20.45 [bc] 18.75 [a]
Dala	control object copper deficiency	8.15 [a] 6.98 [a]	7.43 [bc] 6.40 [ab]	5.67 [ab] 5.62 [ab]	21.25 [bc] 19.00 [ab]
Argona	control object copper deficiency	7.50 [a] 7.68 [a]	7.13 [bc] 6.35 [a]	6.45 [cd] 5.98 [cd]	21.07 [bc] 20.00 [ab]
Maja	control object copper deficiency	8.73 [a] 7.93 [a]	7.05 [bc] 6.65 [bc]	6.70 [d] 6.35 [cd]	22.48 [c] 20.93 [bc]
Mean for cultivars					
Areda Dala Argona Maja		7.30 [a] 7.56 [a] 7.59 [a] 8.33 a	7.06 [a] 6.91 [a] 6.74 [a] 6.85 [a]	5.23 [a] 5.65 [a] 6.21 [b] 6.53 [b]	19.60 [a] 20.13 [a] 20.54 [ab] 21.70 [b]
Mean for fertilization					
Control object Copper deficiency		7.93 [a] 7.45 [a]	7.31 [b] 6.48 [a]	6.07 [b] 5.74 [a]	21.31 [b] 19.67 [a]
Mean for cuts		7.69 [c]	6.89 [b]	5.91 [a]	20.49 [d]

Explanations, see Table 1

CONCLUSIONS

1. Copper deficiency significantly decreased the rate of photosynthesis and transpiration, chlorophyll concentration in leaves and the yield of all the investigated cultivars.

2. Perennial ryegrass cv. Maja was found to be most resistant to copper deficiency – it was characterized by a high rate of photosynthesis and transpiration, and by the highest chlorophyll content. The yield of cv. Maja attained under copper deficit conditions was comparable to that of other cultivars grown under control conditions.

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MAGNESIUM, CALCIUM, POTASSIUM AND SODIUM CONTENT IN GROUNDWATER AND SURFACE WATER IN ARABLE LANDS IN THE COMMUNE (*GMINA*) OF KĄTY WROCŁAWSKIE

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Abstract

This paper discusses some aspects of the research conducted in the hydrological years 2000/2001-2002/2003 on arable areas around several small water bodies located on the outskirts of villages in the commune of Kąty Wrocławskie. The aim of the paper was to assess the content of selected chemical elements in the groundwater and small water bodies.

The water bodies included in the research appeared a few decades ago as a result of human activity; in Zybiszów and Bliż they are small post-mine water bodies, whereas in Smolec and Rybnica they are ponds filling former clay excavation sites. Their surface ranges widely between 0.05 and 2.2 ha, while the average depth reaches 1.2 to 3.5 m. Since no flows come to these water bodies, they are fed only by ground and rain water. In the research period the water level of the bodies fluctuated between 3 and 40 cm, while the maximum changes in the groundwater level were above 1 m.

The examined waters contained elevated levels of elements, the fact which is directly related to the kind of soils in the region. Another factor affecting the content of elements is whether or not soils are used agriculturally. It has been observed that arable areas are distinguished by positive correlation of magnesium, potassium and sodium concentrations in ground and surface waters. Increased content of these elements in Rybnica suggests that the waters receive pollutants from nearby houses.

In the groundwater examined the ratio of calcium and magnesium concentrations ranged from 2.7 to 6.9, whereas in the surface water it varied from 1.2 to 5.1. Values below

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3 were obtained for both types of water only in Rybnica, which proves the influx of sewage from households.

Key words: water, magnesium, calcium, potassium, sodium.

ZAWARTOŚĆ MAGNEZU, WAPNIA, POTASU I SODU W WODACH GRUNTOWYCH ORAZ POWIERZCHNIOWYCH NA UŻYTKACH ROLNYCH W GMINIE KĄTY WROCŁAWSKIE

Abstrakt

W pracy przedstawiono wybrane elementy badań prowadzonych w latach hydrologicznych 2000/2001-2002/2003 na użytkach rolnych wokół kilku niewielkich zbiorników wodnych zlokalizowanych na obrzeżach wsi w gminie Kąty Wrocławskie. Celem pracy była ocena zawartości wybranych pierwiastków w wodach gruntowych i małych zbiornikach wodnych.

Zbiorniki objęte badaniami powstały przed kilkudziesięciu laty w efekcie działalności człowieka – w Zybiszowie i Bliżu są to niewielkie zbiorniki kopane, natomiast w Smolcu i Rybnicy powstały w miejscach dawnych wyrobisk gliny. Ich powierzchnia jest zróżnicowana i wynosi od 0,05 do 2,2 ha, a średnia głębokość od 1,2 do 3,5 m. Nie dopływają do nich żadne cieki, a zasilanie następuje jedynie przez dopływ wód gruntowych oraz opadowych. W okresie badawczym położenie zwierciadła wody w tych zbiornikach zmieniało się w zakresie 3-40 cm, natomiast maksymalne zmiany położenia zwierciadła wody gruntowej przekraczały 1 m.

Badane wody zawiarają duże ilości pierwiastków, co ma bezpośredni związek z rodzajem gleb i użytkowaniem ich jako pola orne. Na terenach użytkowanych rolniczo obserwuje się dodatnie skorelowanie stężenia magnezu, potasu i sodu w wodach gruntowych i powierzchniowych. Podwyższone zawartości tych pierwiastków na obiekcie w Rybnicy wskazują na zanieczyszczenie wód spływem z terenów zabudowanych.

W analizowanych wodach gruntowych stosunek stężenia wapnia do magnezu wynosił od 2,7 do 6,9, natomiast w powierzchniowych od 1,2 do 5,1. Wartości mniejsze od 3, dla obu rodzajów wód, stwierdzono tylko na obiekcie w Rybnicy, co potwierdza hipotezę, że dopływają tam zanieczyszczenia z terenów zabudowanych.

Słowa kluczowe: woda, magnez, wapń, potas, sód.

INTRODUCTION

The composition and concentration of substances in ground and surface water is a resultant of two factors: the geological structure of the earth's crust, including the intensity with which it is leached, and anthropogenic activity associated with agriculture, industry and public utilities. As water travels through the soil's profile, various water-soluble substances are released (PULIKOWSKI et al. 2006). Most of them are macroelements and alkali elements. Water acts as a solvent, enabling plants to absorb substances essential for their growth. Any excess of water content in soil causes an adverse outflow of nutrients. The actual mass of the components carried away depends mostly on the amount of water leaving particular surface (Pulikowski 2004). The amount of rainfall (Szymczyk, Cymes 2005), the type of soil and the manner in which it is used (Cymes, Szymczyk 2005) are indirectly responsible for the load of components carried away from the soil. Leaching calcium and magnesium is additionally made more intensive by draining devices (Kobus, Glińska-Lewczuk 2005). In the case of heavily salted soils, their profile is washed with clean water in order to reduce the concentration of, for instance, sodium (Kelleners et al. 2000).

Calcium and magnesium are important macroelements, taken up by plants in considerable amounts. Magnesium is a constituent of chlorophyll and activates a number of enzymatic reactions. Calcium incrusts cell membranes, influences the accumulation of oxalates, regulates water supply and metabolic processes. It also favours the forming of cloddy structure, as it triggers off coagulation of soil colloids (Dobrzański, Zawadzki 1981).

Alkali elements – sodium and potassium – also play a significant role in vital processes of plants. Potassium plays a major role in synthesis and respiration processes and, even more importantly, regulates the hydration of tissues. Sodium, on the other hand, influences the physicochemical properties of plasma and water supply. The uptake of these elements is strictly dependent on the relationship between their contents in soil (Dobrzański, Zawadzki 1981).

The purpose of this paper was to assess the content of selected elements in groundwater and small water bodies in the commune (*gmina*) of Kąty Wrocławskie.

CHARACTERISTICS OF SUBJECTS AND METHODOLOGY OF RESEARCH

This paper discusses selected elements of the research conducted in the hydrological years 2000/2001-2002/2003 in the commune (*gmina*) of Kąty Wrocławskie on arable areas around several small water bodies located on the outskirts of the following villages: Bliż, Rybnica, Smolec and Zybiszów. The area included in the research is located in the region of Wrocław Plain (Równina Wrocławska) (Kondracki 1994), west of Wrocław. The surface of the plain is distinguished by minor denivelations. The arable lands there are mainly fertile black earth created mostly from silts and silty loams, sporadically lined with more permeable deposits.

The average long-term sum of rainfall in this area (on the basis of the data obtained from the meteorological point of the Experimental Station for Cultivar Testing in Zybiszów) was 568 mm, out of which 371 mm (*ca* 65%) fell during the vegetation period (April-September). Annual sums of rainfall in the following years of the research period 2000/2001-2002/2003 were 644 mm, 577 mm and 488 mm, while the sums of rainfall recorded during the vegetation periods came to, respectively, 466 mm, 362 mm and 295 mm. The subsequent hydrological years of the research period were characterized as a wet year (2001), a normal year (2002) and a dry year (2003). Vegetation periods (April-September) of these years were described, respectively, as: medium wet, normal and medium dry. With regard to thermal conditions, the analyzed period can be considered relatively warm, with average air temperatures in summer semi-years (May-October) and winter semi-years (November-April) usually somewhat above average. Only in 2002/2003 the winter semi-year was cooler.

The water bodies included in the research has appeared a few decades ago due to human activity; in Zybiszów and Bliż they are small post-mine water bodies, whereas in Smolec and Rybnica they are ponds filling in former clay excavation sites. They differ in the surface area, with the largest pond in Smolec (Sb) – *ca* 2.2 ha. Much smaller are the water bodies in Rybnica (Rb) – *ca* 0.25 ha, in Zybiszów (Zb) – *ca* 0.06 ha and three bodies in Bliż, namely (Bb1), whose surface reaches 0.09 ha, Bb2 – 0.05 ha and Bb3 – 0.12 ha. The average depths of these bodies are: Sb – 3.5 m, Rb –1.4 m, Zb – 1.8 m, Bb1 – 1.8 m, Bb2 – 1.5 m and Bb3 – 1.2 m. Since no flows come to these water bodies, they are fed only by ground and rain water. The ponds in Bliż are connected and water flows from Bb1 through Bb2 to Bb3, with the excess of water from the latter pond carried away through a ditch to a small stream called the Kasina. Throughout the research period, the level of groundwater was systematically monitored by means of installed indicating devices. In piezometers situated on adjacent lands depths of the groundwater table were recorded. For the purpose of chemical analyses water samples were taken from all the bodies (Bz1, Bb2, Bb3, Rb, Sb, Zb) and piezometers: Bp (Bliż); Zp1, Zp2 (Zybiszów); Rp1, Rp2 (Rybnica) and Sp (Smolec).

Annual fluctuations in the water level of the particular bodies fell into the range from several to 40 cm at maximum. The water bodies were usually filled up to the maximum in spring or, briefly, after heavy summer rains. In the following years of the research, the water table levels were 24 cm, 14 cm and 30 cm in Rybnica; 31 cm, 33 cm and 40 cm in Smolec; and 34 cm, 32 cm and 36 cm in Zybiszów. The water table of the bodies in Bliż was generally very stable; small changes, oscillating around 3-15 cm, were due to the fact that these bodies form a mutually connected system, in which excess water can run freely to a nearby water flow.

The course of changes in the groundwater table in the piezometers located near the water bodies coincided broadly with the changes in the atmospheric conditions. The groundwater table was usually nearest the ground surface in spring after thaw. Later, the water level gradually decreased to reach its maximum depth in the second half of the hydrological year. Occasionally, the groundwater table rose distinctly also after heavy summer rains, as it happened in the end of July and the beginning of August 2001.

The groundwater table in the piezometers at particular test subjects fluctuated within different ranges. In piezometer Rp1 the depth of the water table altered within 78 and 175 cm, while in Rp2 the minimum and maximum depths were 109 and 184 cm. The groundwater table in piezometer Sp in Smolec fluctuated during the research period from 97 to 210 cm. In piezometer Zp1 in Zybiszów the depths changed from 62 from 110 cm, whereas in Zp2 – from 185 to 234 cm. In Bliż in piezometer Bp the groundwater table changed in individual years from the minimum value of 95 cm beneath the ground level to the maximum level of 180 cm.

In order to conduct physicochemical analyses, water samples were taken once a month during the vegetation period (April-September). Calcium and magnesium contents were marked by the versene method while sodium and potassium were determined by flame photometry (HERMANOWICZ et al. 1999). The determinations were performed in the Water and Sewage Laboratory at the Institute of Environmental Development and Protection, University of Environmental and Life Sciences, Wrocław. The differences between average composition of the groundwater coming from various measuring points were estimated by means of unidirectional variance analysis (test F) at significance level p=0.05. Statistical calculations were performed with an aid of Statistica 7.1 software.

RESULTS AND DISCUSSION

The groundwaters were characterized by pH ranging from 6.5 do 8.4; a somewhat smaller range, from 7.2 to 8.3 pH, was observed for wells located near households. The surface waters had a slightly higher pH, which ranged from 7.3 to 8.8. On the whole, they are neutral or slightly alkaline waters.

The content of magnesium in the groundwaters was significantly different and it ranged from 25.0 mg $Mg \cdot dm^{-3}$ in the well in Bliż to 74.0 mg $Mg \cdot dm^{-3}$ in Rp1 in Rybnica (Figure 1). The values obtained in most of the subjects were approximate to those provided in the literature (PULIKOWSKI 2004, FIEDLER, OTHERS 2005). It is noteworthy that the values obtained at the piezometers in Rybnica were up to 2-3-fold higher than the remaining results. This is probably related to the local geological structure. The concentrations of magnesium in the surface waters were slightly depressed, indicating a direct relationship with the concentration of this element in the groundwater (Figure 2). The values obtained in our study are considerably high in comparison to those reported by KOC et al. (2001) for water bodies of similar dimensions located in Olsztyn Lake District (Pojezierze Olsztyńskie)

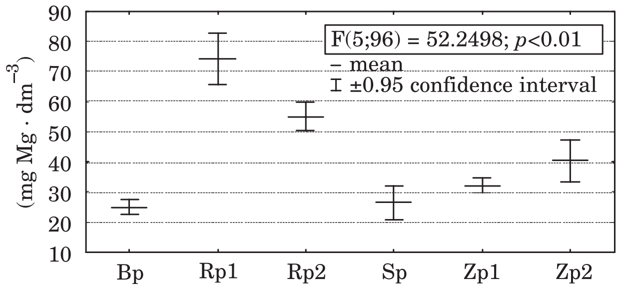

Fig. 1. Concentration of magnesium in groundwater

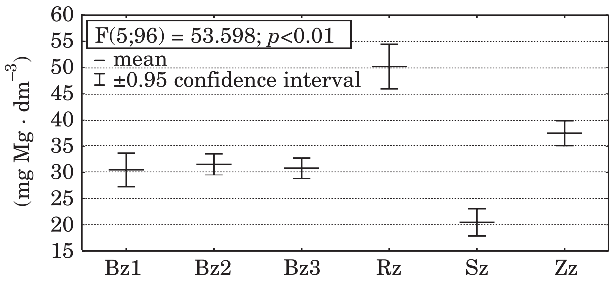

Fig. 2. Concentration of magnesium in surface waters

or Miler et al. (2001) in Wielkopolska Region. The area examined in the present research comprised black earth used as arable fields, thus the conditions are favourable for the leaching of magnesium and calcium (Cymes, Szymczyk 2005).

The concentration of calcium in the groundwater was characterized by significantly weaker changeability, with the exception of well Zp2, where an average concentration reached 280 mg Ca·dm^{-3}, being evidently higher than in the other wells (Figure 3). In the case of calcium no direct relationship between the concentrations in ground and surface waters was discerned (Figure 4). It is the acidic pH that is crucial for the calcium content and this is why processes increasing the pH, such as acid rains or nitrification, increase the concentration of this element in water. (van Lonn, Duffy 2007). As for the bodies situated in Bliż, it may be presumed that the increased concentration of this element is related to the waters that flow there from the farmsteads and households (Tandyrak et al. 2005).

In natural waters the content of calcium is 3-4-fold higher than that of magnesium, whereas waters which are more heavily salted, e.g. seawater, contain 3-4-fold more magnesium (Gomółka, Szaynok 1997). In the examined

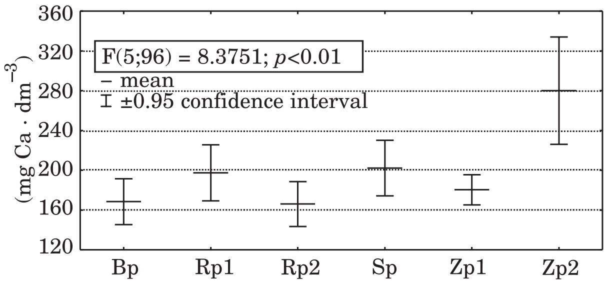

Fig. 3. Concentration of calcium in groundwater

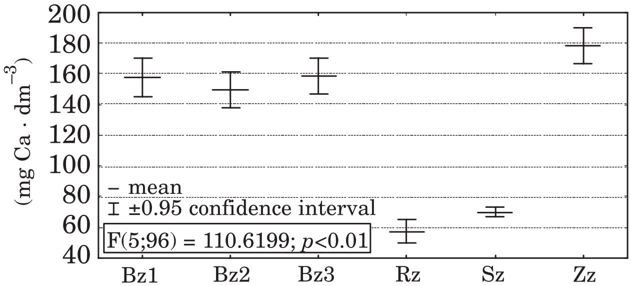

Fig. 4. Concentration of calcium in surface waters

groundwaters, calcium and magnesium concentrations were in the ratio from 2.7 to 6.9; in the surface waters the ratio was from 1.2 to 5.1. Values smaller than 3, for both types of water, were obtained only in Rybnica, which may indicate that these waters are polluted by an inflow coming from the local households.

Potassium content in the groundwaters varied substantially and ranged from 6.0 mg K·dm^{-3} in Bliż to 26.9 mg K·dm^{-3} in well Rp1 in Rybnica (Figure 5). Similar values were obtained in other studies (MILER et al. 2001, DURKOWSKI 2005). An equally high potassium concentration occurred in surface waters coming from the subject in Rybnica, where it reached 65 mg K·dm^{-3}. The research by DURKOWSKI (2005) demonstrates that such high potassium concentrations occur in urban areas. An analogous situation was observed with respect to the surface waters (Figure 6); potassium concentration in body Rb was definitely higher than elsewhere and also diverged from the values noted in agricultural lands (DURKOWSKI, WORONIECKI 2001, KOC et al. 2001).

Also in the case of sodium the highest concentrations were obtained in the subject located in Rybnica, with the values of 109.7 mg Na·dm^{-3} for

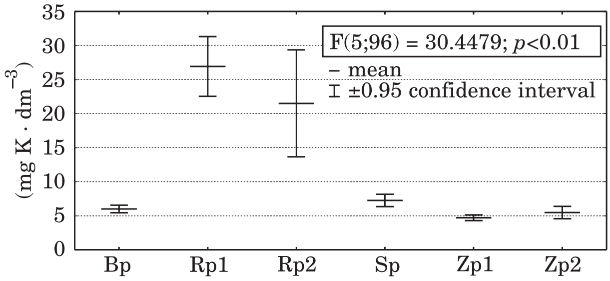

Fig. 5. Concentration of potassium in groundwater

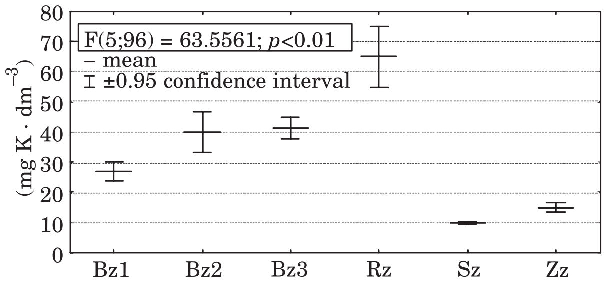

Fig. 6. Concentration of potassium surface waters

groundwaters and 59.6 mg Na·dm^{-3} for surface waters (Figures 7 and 8). Such high concentrations of this element do not occur on arable lands (Miler et al. 2001, Pulikowski 2004), the fact which confirms the assumptions that the composition of water in this subject may be influenced by wastewater flowing to there from local households (Orzepowski et al. 2008).

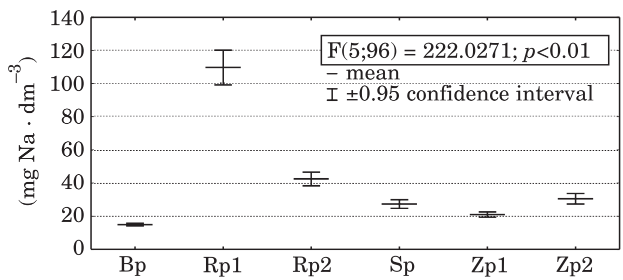

Fig. 7. Concentration of sodium in groundwater

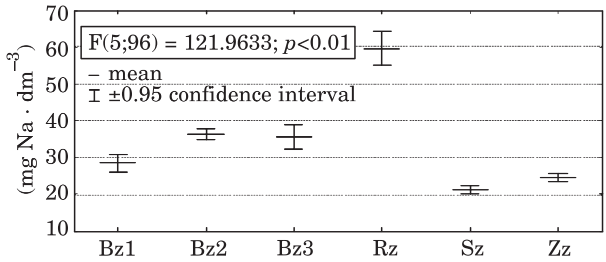

Fig. 8. Concentration of sodium in surface waters

CONCLUSIONS

1. The tested waters are characterized by a significant content of the examined elements, the finding which is directly related to the type of soils and their use as arable fields.

2. On arable areas a positive correlation of magnesium, potassium and sodium contents is observable, both in ground and surface waters.

3. Increased values of magnesium, potassium and sodium, as well as a low ratio of calcium to magnesium concentrations in the subject in Rybnica may be indicative of eutrophication of the waters in this area owing to an influx of pollutants from farmsteads and households nearby.

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EFFECT OF FOLIAR AND SOIL APPLICATION OF COPPER ON THE LEVEL AND QUALITY OF WINTER RAPESEED YIELDS

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Abstract

In Poland nearly 40% of land under agricultural use is characterised by a progressing deficit of copper available to plants. Inventory studies revealed that 25% of rapeseed plantations are undernourished with this component.

The objective of the study was to estimate the yield-forming effects of foliar and soil fertilisation of rapeseed with copper, to identify the optimum dose of Cu for the crop species, and to compare the effects of the two methods of application on the quality of the seeds. The study comprised two three-year strict field experiments, in which pre-sowing fertilisation of rapeseed with copper was applied at rates of 4, 8 and 12 kg Cu$\cdot$ha^{-1}, as well as foliar spraying at the optimum dose of 250 Cu g$\cdot$ha^{-1} in the phase of closed bud. The experiments were set up on light soils of acid and light acid reaction and with a low or medium content of available copper.

Significant increases in rapeseed yields, compared to the control treatment (without Cu), were obtained in treatments fertilised to the soil with doses of 8 and 12 kg Cu$\cdot$ha^{-1} and in the treatment with foliar spraying. In plants from treatments without copper fertilisation, insufficient levels of the content of this component were found. Sufficient copper nutrition of plants caused a significant increase in yields of rape seeds. In some of the treatments, there was a tendency for the copper content in seeds to increase favourably, especially in the case of foliar application and of the higher copper doses applied to the soil. Also, some cases of increased oil content in seeds were recorded, even by several percent compared to seeds from treatments without Cu fertilisation.

With the existing copper deficit in soils in Poland, fertilisation of rapeseed with this component appears to be crucial.

Key words: rape, copper, fertilization, yield of seeds, oil content.

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WPŁYW DOLISTNEGO I DOGLEBOWEGO NAWOŻENIA MIEDZIĄ NA WIELKOŚĆ I JAKOŚĆ PLONÓW RZEPAKU OZIMEGO

Abstrakt

W kraju blisko 40% użytków rolnych cechuje pogłębiający się niedobór miedzi przyswajalnej dla roślin. Badania inwentaryzacyjne wykazały 25% plantacji rzepaku niedożywionych w ten składnik.

Celem badań była ocena efektów plonotwórczych dolistnego i doglebowego nawożenia rzepaku ozimego miedzią, wskazanie optymalnej dawki Cu dla tego gatunku oraz porównanie wpływu obu sposobów aplikacji na cechy jakościowe nasion. Przeprowadzono 2 trzyletnie ścisłe doświadczenia polowe, w których zastosowano przedsiewnie nawożenie rzepaku miedzią dawką 4, 8 i 12 kg Cu$\cdot$ha^{-1} oraz oprysk dolistny o optymalnej dawce 250 Cu g$\cdot$ha^{-1} w fazie zwartego pąka Doświadczenia prowadzono na glebach lekkich o odczynie kwaśnym i lekko kwaśnym oraz niskiej bądź średniej zawartości przyswajalnej miedzi.

Istotne zwyżki plonów nasion rzepaku w stosunku do obiektu kontrolnego (bez Cu) uzyskano na obiektach nawożonych doglebowo dawką 8 i 12 kg Cu$\cdot$ha^{-1} oraz pod wpływem dolistnej aplikacji. W roślinach z obiektów nie nawożonych miedzią stwierdzono niedostateczne dla rzepaku zawartości tego składnika. Optymalne odżywienie roślin miedzią spowodowało istotny wzrost plonów nasion rzepaku. W poszczególnych doświadczeniach pojawiła się tendencja do korzystnego zwiększania się zawartości miedzi w nasionach szczególnie po zastosowaniu dolistnego nawożenia oraz wyższych dawek doglebowych.

Odnotowano również przypadki zwiększenia zawartości tłuszczu w nasionach nawet o kilka procent w stosunku do obiektów bez nawożenia Cu.

Wobec niedoborów miedzi w glebach kraju, nawożenie rzepaku tym składnikiem staje się niezbędne.

Słowa kluczowe: rzepak, miedź, nawożenie, plon nasion, zawartość tłuszczu.

INTRODUCTION

In the nearest years, rapeseed may gain strategic importance for the Polish agriculture and economy in general. Until now, the cultivation of the crop species has been limited primarily by the demand of the national oil-producing industry. Recently, a new major customer for rapeseed, i.e. the industry producing ecological diesel fuel, bio-fuel, has appeared. In accordance with the EU requirements, in the future Poland will be required to systematically increase rapeseed production (KUŚ 2002). Rapeseed oil production may be increased through reliable and high yields of rapeseed with the required level of oil content.

Production of high crop yields, stimulated by basic fertilisation, causes gradual depletion of nutrients from soil, mainly microelements which are not added to soil by fertilisation. Microelemental deficiency grows, resulting in limited plant growth and development. Copper is among the most deficient components of soils in Poland (CZUBA 2000, GEMBARZEWSKI 2000). Studies on the fertility of Polish soils of Poland in 1994-1999 showed that low copper content occurs in 36% of arable soils (Dębowski, Kucharzewski 2000). Insufficient copper content for rapeseed plants was observed in 28% of fields (Sienkiewicz-Cholewa 2001).

Rapeseed belongs to plants which are sensitive to excessive levels of copper, yet – like cereals – it takes up approx. 40 g of Cu per 1 ha (10 g per 1 ton of seed yield). This indicates a high requirement of rapeseed for this element. As copper performs important physiological functions in the plant, rapeseed fertilisation with this component appears to be crucial in view of the existing deficit of copper in soils (Katyal, Randhawa 1983, Shorrocks 1990).

MATERIAL AND METHODS

In 2003-2006, two three-year strict experiments were carried out at the Experimental Stations Baborówko and Osiny, which included varied copper fertilisation of rapeseed. Against the background of the optimum basic NPK fertilisation as established in the IUNG Fertilisation Recommendations, the experiments included soil fertilisation with increasing doses of copper and foliar fertilisation at a dose accepted as optimum for plants (Szukalski 1987). The experiments were set up in a random block system, with 4 replications, and included the following experimental treatments:

1) control (without Cu) – K;
2) Cu to soil, at a dose of 4 $kg \cdot ha^{-1}$ – Cu1;
3) Cu to soil, at a dose of 8 $kg \cdot ha^{-1}$ – Cu2;
4) Cu to soil, at a dose of 12 $kg \cdot ha^{-1}$ – Cu3;
5) Cu in foliar application, at a dose of 250 $g \cdot ha^{-1}$ – Cu4 (BBCH 50-59).

In the experiments two population cultivars of rapeseed were tested – cv. Lisek at ES Baborówko and cv. Californium at ES Osiny. The area of the experimental plots was 24 m^2.

The plots were fertilised to soil with copper in the form of technical salt – copper sulphate $CuSO_4 \cdot 5H_2O$. Foliar fertilisation in the form of spraying was performed at the budding phase of rapeseed (BBCH 50-59), using 0.2% water solution of copper sulphate.

Content of macro- and micronutrients in the experimental soils and in rape seeds was determined using the methods commonly applied at agrochemical stations (Metody Badań Laboratoryjnych w Stacjach Chemiczno-Rolniczych 1980). The content of copper was determined with the AAS method. Crude fat in seeds was determined with the Soxlet method after extraction with ethyl ether.

The yields obtained have been worked out statistically using variance analysis. The significance of cross-object differences in variance analysis were evaluated with the Tukey's test (α = 0.05).

The experiments were conducted on light soils, slightly acidic in reaction and with low or medium content of available copper (Table 1). Determinations made for the soils revealed high and medium levels of content of available forms of P, K and Mg and high content of zinc and manganese, fully covering the nutritional requirements of rapeseed. Estimation of the content of the nutrients was performed on the basis of the valid limit values in accordance with the IUNG Fertilisation Recommendations (1985).

Table 1

Chemical characteristics of soils under trials

Experimental station (soil)		pH in KCl	P	K	Mg	Cu	Mn	Zn
			(mg·kg^{-1})					
Baborówko (light loamy sand)	I*	5.6	68 ^{h}	124 ^{m}	90 ^{h}	2.3 ^{m}	132 ^{m}	9.8 ^{h}
	II	5.8	76 ^{h}	100 $_{m}$	100 ^{h}	1.5 ^{l}	96 ^{m}	10.4 ^{h}
	III	6.4	65 ^{m}	104 ^{m}	98 ^{h}	4.5 ^{m}	112 ^{m}	13.0 ^{h}
Osiny (heavy loamy sand)	I	6.0	65 ^{m}	108 ^{m}	105 ^{h}	1.6 ^{l}	168 ^{m}	6.3 ^{m}
	II	6.0	64 ^{m}	124 ^{m}	93 ^{h}	2.5 ^{m}	201 ^{m}	12.0 ^{h}
	III	6.0	76 ^{h}	83 ^{m}	110 ^{h}	1.8 ^{m}	156 ^{m}	14.3 ^{h}

*I,II,III – consecutive years of the experiment;
Concentration in soil: *l* – low, *m* – medium, *h* – high

RESULTS

One of the approved indicators of rapeseed requirement for copper was the effect of fertilisation with this component on the level of seed yields. Rapeseed yields obtained in the particular experiments were varied, from 3.0 to 7.3 t·ha^{-1}, and depended largely on the weather conditions. The lowest yield at the level of 3.0-7.3 t·ha^{-1} was obtained at Baborówko II. Plant emergence and growth was limited by drought conditions. Plant density in autumn was low (50 per m^{2}). Some of the population was destroyed by frost. Relatively high yields within the range of 4.0-4.5 t·ha^{-1} were achieved at Baborówko I, III and Osiny II, III. The highest yield of seed – 7.3 t·ha^{-1} was noted in a trial at Osiny Station after favourable winter survival. Weather conditions during the vegetation period were conducive to plant development.

In the first two years of the experiments, a statistically proven increase in rapeseed yields was obtained in treatments with soil fertilisation at the highest dose of copper – 12 kg Cu·ha^{-1} (Cu3) and under the effect of foliar fertilisation with copper at the dose of 0.250 kg Cu·ha^{-1} (Cu4). The increase in seed yields was in the range of 2-6% (Table 2). Pre-sowing fertilisation with the lowest doses, 4 and 8 kg Cu·ha^{-1} (Cu1), did not result in any increase in rapeseed yields. In the 3rd year of the experiments no yield response was recorded at either of the two experimental locations.

Table 2

Rape seed yields in separate trials in t·ha^{-1}

Cultivar		Fertilization					
		0	Cu1	Cu2	Cu3	Cu4	$LSD_{0.05}$
Lisek	I	4.09 ^{a}	4.13 ^{ab}	4.17 ^{ab}	4.22 ^{b}	4.22 ^{b}	0.111
	II	2.96 ^{a}	2.93 ^{a}	3.00 ^{ab}	3.00 ^{ab}	3.17 ^{b}	0.205
	III	4.00	4.08	4.07	4.10	4.18	n.s.
Californium	I	7.31 ^{a}	7.57 ^{ab}	7.71 ^{ab}	7.72 ^{b}	7.93 ^{b}	0.392
	II	4.52 ^{a}	4.56 ^{ab}	4.60 ^{ab}	4.83 ^{ab}	4.94 ^{b}	0.425
	III	5.30	5.34	5.44	5.39	5.44	n.s.

Yields marked with the same letter are not significantly different acc. to Tukey's test;
n.s. – differences not significant

In the control treatments (without Cu) copper levels were insufficient for plants – on average they reached 4.7 mg·kg^{-1} whereas the optimum copper content for rapeseed given by Bergmann is 5-12 mg·kg^{-1} (BERGMANN 1986). Under the effect of the fertilisation applied in our experiments, the concentration of copper in the plants increased reaching 4.9, 4.9, 5.4, 9.3 mg·kg^{-1} in treatments Cu1, Cu2 , Cu3 and Cu4, respectively. Optimum nutrition of rapeseed with copper was ensured by the soil fertilisation at the dose of 12 kg Cu·ha^{-1} and the foliar fertilisation.

Oil content in rapeseed without copper fertilisation was notably lower than the mean values given by COBOR (Centre for Studies on Crop Plant Cultivars): 44.6 and 45.2 % d.m in cv. Lisek and Californium, respectively. Copper application, both to the soil and in the form of foliar fertilisation, resulted in an increase in the oil content of seeds of both rapeseed cultivars. Pre-sowing application of the higher doses of copper – 8 and 12 kg (Cu2 and Cu3) and foliar application at the dose of 0.250 kg Cu·ha^{-1} caused a significant increase in oil concentration with relation to the control treatments. Fertilisation to the soil with the lowest dose of 4 kg Cu·ha^{-1} (Cu1) did not result in any improvement in the oil content of seeds of the two rapeseed cultivars. The highest increase in seed oil content, by 2.5-2.7%, was recorded after the foliar application of copper on soils with low levels of copper (Baborówko II, Osiny I).

Table 3

Oil content in rapeseed (%)

Cultivar		Fertilization					
		0	Cu1	Cu2	Cu3	Cu4	$LSD_{0.05}$
Lisek	I	43.8	42.8	44.5	44.4	45.7	0.56
	II	43.0	43.2	43.4	44.5	45.7	0.84
	III	42.3	41.7	42.7	42.7	43.5	0.82
Californium	I	44.0	44.8	45.7	45.2	45.9	0.99
	II	43.3	43.3	43.7	43.0	43.5	0.95
	III	43.0	43.0	43.8	44.0	44.6	0.93

Copper concentration in seeds of the two rapeseed cultivars was considerably lower than the average of 3.1 $mg \cdot kg^{-1}$ d.m., recorded in Poland in the 1970's (KAMIŃSKA i in. 1976). Seeds harvested from the treatments without copper fertilisation (K) contained on average 2.2 mg $Cu \cdot kg^{-1}$ for both cultivars. Under the effect of the dose of 12 $kg \cdot ha^{-1}$ applied to the soil, and of the foliar fertilisation, the level of copper concentration increased to 2.8--3.0 $mg \cdot kg^{-1}$ d.m.

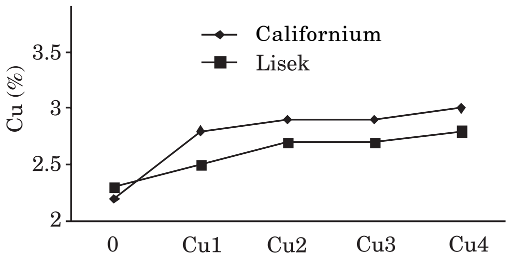

Fig. 1. Mean copper content in the seeds of the rapeseed cultivars

DISCUSSIONS

The need of fertilising rapeseed with copper, at a high level of basic fertilisation, on light soils with a low content of copper was indicated by the results of a pot experiment conducted by RUSZKOWSKA, ŁYSZCZ (1975) with traditional rapeseed cultivars "0". With a high level of NPK fertilisation, the increase in rapeseed yields was as much as 40%. The yield-forming effect of copper in the cultivation of "00" rapeseed cultivars is also indicated by the results of a strict field experiment conducted by BOBRZECKA, SALAMONIK (1997).

These authors found a significant increase in rapeseed yields on grey-brown podzolic soil and alluvial soil with low and medium content of available copper after the application of copper at 10 kg Cu·ha^{-1}. In a study conducted by SIENKIEWICZ-CHOLEWA (2001), on soils with a medium content of Cu, synthesis of yields obtained in the experiments indicated a 6% increase in rapeseed yields.

In this study, the best results were achieved with the foliar fertilisation at the dose of 250 g Cu·ha^{-1}. Increase in the dose of copper applied to the soil resulted, in most of the experiments, in a slight increase of the seed yield. This shows that on soils with pH in the range of 5.4-6.4 adsorption of copper is high, which limits its availability to plants. Copper could have also partly accumulated in plant roots, which happens when high doses of the component are introduced in soil. Many authors argue that analysis of the index parts of plants – leaves – is not an indicator that would reliably reflect the actual status of copper supply for plants (KORZENIOWSKA, STANISŁAWSKA-GLUBIAK 2003, STANISŁAWSKA-GLUBIAK, KORZENIOWSKA, IGRAS 2007).

Oil is the most important parameter of rapeseed quality. Apart from the genetic factor, the content of oil in rape seeds is largely determined by mineral fertilisation of the plants. Decrease in rapeseed oil content may result from deficit of nutrients, among others of such microelements as zinc and copper that control the metabolic transformations in the plants.

The pre-sowing fertilisation with copper applied in this study at the rates of 8 and 12 kg Cu·ha^{-1} and the foliar fertilisation at the dose of 0.250 kg Cu·ha^{-1} resulted, in most of the experiments, in a significant increase of oil concentration in the final yield of rapeseed as compared to the control treatments. A favourable increase in rapeseed oil content was also obtained by BOBRZECKA, SALAMONIK (1997) on a light soil with low copper content. Under the effect of foliar fertilisation with 0.200 kg Cu·ha^{-1}, the level of oil content increased by 3.5% d.m.

Natural supplementation of the required amount of copper in fodders and feeds is expected to maintain a suitable level of this component in plant products. Analysis of rape seeds from the experiments showed that seeds from treatments without copper fertilisation had a low copper content compared to the national mean value used for comparison. This indicates a notable deterioration of rapeseed quality over the years, which is due to an increasing deficit of copper in soils. The chemical composition of seeds is determined genetically but can be modified, to a certain extent, by the environmental factors. Under the effect of the copper fertilisation applied in our trials, a tendency towards increased copper concentration in rape seeds was observed in the two test rapeseed cultivars. This is favourable in terms of the consumption value of oil as well as of the fodder value of rapeseed oil cake and meal. Studies indicate that the content of copper in plant products and its consumption have decreased notably, resulting in increased incidence of several human and animal diseases (ŻECHAŁKO-CZAJKOWSKA 1992, KUBIŃSKI 1996).

The favourable effect of copper fertilization on the level of yields, content of oil and concentration of copper in seeds provides evidence supporting the necessity of including copper in rapeseed fertilisation.

CONCLUSIONS

1. The application of rapeseed fertilisation with copper under conditions of low and medium level of this component in soil resulted in 2-6% increase in rapeseed yields. A statistically significant increase in rapeseed yields was obtained under the effect of foliar fertilisation with copper at 250 $g \cdot ha^{-1}$ and of the soil fertilisation with copper at 12 $kg \cdot ha^{-1}$.

2. The foliar fertilisation and pre-sowing fertilisation with copper at 8 and 12 $kg \cdot ha^{-1}$ led to a significant increase in the concentration of oil in seeds of the test rapeseed cultivars. Under the effect of copper fertilisation, a favourable tendency towards increased copper concentration in seeds of the test rapeseed cultivars was observed.

3. Foliar copper application produced a stronger effect on the concentration of this element and fat level in rape seed was more pronounced than soil Cu fertilization

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QUALITY OF WELL WATERS IN CONTEXT OF THE CONTENT OF NITROGEN AND PHOSPHORUS COMPOUNDS IN THE UPPER NAREW RIVER VALLEY*

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Abstract

The paper dealt with the evaluation of concentration and spatial distribution of nitrogen and phosphorus forms in dug wells in selected farms in the upper Narew River valley. The study was also aimed at assessing the influence of selected elements of a farm on quality of well water. Examinations were carried out in 8 villages in Podlasie region. Three farm dug wells were selected for examination in each village. All wells take water from the first water-carrying level. The study included well waters within the section of the Narew River from the villages Sobótka to Złotoria. The checkpoints were set in: Sobótka, Narew, Ciełuszki, Kaniuki, Zawyki, Uhowo, Topilec, and Złotoria localized along the Narew and its adjacent lands. Water samples were collected in spring, summer and autumn 2006. Ammonia, nitrates (III), nitrates (V), and phosphates were determined in water samples filtered through micropore filters (d = 0.45 μm) by means of colorimetry applying a spectrometer HACH (DR-2000). Parameters of the analytical procedures were adjusted in accordance to the Decree of Minister for Environment (2004). In total, 288 determinations in three series were made. For each of the three wells from every village, the arithmetic means of the analyzed parameters were calculated on the basis of samples collected on three dates. The assessment of underground water quality was made according to the Decree of Minister for Environment of 11 February 2004 on on surface and underground water status, screening performance, interpretation and presentation of the status of such waters. In addition, the Decree of the Minister for Health of 29 March 2007 (Rozporządzenie Ministra Zdrowia, 2007) was referred to. The results were compared to concentrations of forms of particular components ($N-NH_4$, $N-NO_2$, $N-NO_3$) included in the Decree of the Minister for Environment (2004) and the Decree of the Minister for Health (2007) in order to designate water quality classes and principal chemical requirements to the well wa-

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ters examined. The study revealed that the analyzed waters did not meet standards set for potable water due to exceeding permissible values for ammonia and nitrates (V) concentrations. The distance of a well from the nearest farm buildings affected ammonia concentration in well water.

Keywords: well, nitrates, phosphates.

JAKOŚĆ WÓD STUDZIENNYCH W DOLINIE GÓRNEJ NARWI POD WZGLĘDEM STĘŻENIA ZWIĄZKÓW AZOTU I FOSFORANÓW

Abstrakt

Celem pracy było określenie stężenia oraz rozkładu przestrzennego form azotu i fosforanów w studniach gospodarskich (kopanych) z wybranych gospodarstw wiejskich doliny górnej Narwi. Dodatkowym celem pracy było określenie wpływu wybranych elementów zagrody wiejskiej na jakość wód studziennych. Badania prowadzono w 8 wsiach położonych w województwie podlaskim. W każdej wsi do badań wybrano po 3 studnie. Wszystkie studnie czerpią wodę z pierwszego poziomu wodonośnego. Badaniami objęto wody studzienne na odcinku rzeki Narew od wsi Sobótka do wsi Złotoria. Miejsca poboru próbek wody wyznaczono w miejscowościach: Sobótka, Narew, Ciełuszki, Kaniuki, Zawyki, Uhowo, Topilec, Złotoria zlokalizowanych wzdłuż rzeki Narew w jej bezpośrednim sąsiedztwie. Próbki wody pobierano 3-krotnie: wiosną, latem, jesienią 2006 r. W próbkach wody przefiltrowanych przez filtry mikroporowate o średnicy porów 0,45 μm oznaczono metodą kolorymetryczną, na spektrofotometrze HACH typu DR-2000, azot amonowy, azotynowy, azotanowy i fosforany. Parametry metod analitycznych dostosowano do zaleceń Rozporządzenia Ministra Środowiska (2004). Ogółem wykonano 288 oznaczeń w trzech seriach badawczych. Obliczono średnią arytmetyczną zawartość badanych parametrów w próbkach pobranych z 3 studni każdej wsi w trzech terminach. Ocenę jakości wód podziemnych oparto na Rozporządzeniu Ministra Środowiska z dnia 11 lutego 2004 r. w sprawie klasyfikacji dla prezentowania stanu wód powierzchniowych i podziemnych, sposobu prowadzenia monitoringu oraz sposobu interpretacji i prezentacji stanu tych wód, z uwzględnieniem Rozporządzenia Ministra Zdrowia z dnia 29 marca 2007 r. Wyniki porównywano ze stężenirm form poszczególnych składników ($N\text{-}NH_4$, $N\text{-}NO_2$, $N\text{-}NO_3$) ujętych w Rozporządzeniach MŚ (2004) i MZ (2007) w celu przypisania badanym wodom klas jakości oraz podstawowych wymagań chemicznych. Wykazano, że: wody nie odpowiadały standardom wody przeznaczonej do spożycia ze względu na przekroczenia wartości dopuszczalnych stężeń azotu amonowego i azotanowego, wody o najniższej jakości stwierdzono w studniach ze wsi Złotoria i Zawyki. W pracy określono również wpływ odległości studni od zabudowań inwentarskich na stężenie azotu amonowego w ich wodach.

Słowa kluczowe: studnia, azotany, fosforany.

INTRODUCTION

Resources of underground waters in Podlasie region occur in Quaternary, Tertiary, Cretaceous, and Jurassic formations. Underground waters in Quaternary formations are practically present all over the region (http://www.mos.gov.pl/).

The depth of water surface in river valleys and depressions of the upper Narew River valley ranges within 0.1-1.0 m, while the height can be up to 5-8 m; it is the main water source in dug wells. Waters at this level vary greatly, depending on the intensity of rainfall and spring thawing (*Studium*... 2004).

Quality of ground water originating from farms is an indicator of this point source of agricultural contamination, mainly associated with animal production, whose influence on area pollution attracts more attention than plant production (Sapek 2006).

Farms and adjacent land are closely associated with animal production. Animal waste, such as solid and liquid manure as well as poultry droppings, is potential source of ground water contamination with nutrients, mainly nitrogen and phosphorus compounds (Durkowski et al. 1997).

The paper contains an evaluation of the concentration and spatial variability of nitrogen forms and phosphates in farm dug wells localized in the upper Narew River valley. In addition, the purpose of this study has been to assess the influence of selected elements of a farm on well water quality.

MATERIAL AND METHODS

The study was carried out in 8 villages situated in Podlasie region (Figure 1). Three farm dug wells were selected for examination in each village. All wells take water from the first water-carrying level. The study included well waters within the section of the Narew from the villages Sobótka to Złotoria. The checkpoints were set in: Sobótka, Narew, Ciełuszki, Kaniuki, Zawyki, Uhowo, Topilec, and Złotoria, all lying along the Narew and at its adjacent lands. Water samples were collected in spring, summer and autumn 2006.

Samples were collected in such a way to preserve their native character. When the wells were permanently covered, water was taken from pumps or taps after letting some water out to ensure the same parameters as those of underground water. The distance between the wells and the farm buildings, houses and the Narew River was also measured. All the data are presented in Table 1.

Ammonium nitrogen, nitrite nitrogen, nitrate nitrogen and phosphates were determined in water samples filtered through micropore filters ($d = 0.45$ µm) by means of colorimetry applying a spectrometer HACH (DR-2000). Parameters of the analytical procedures were adjusted in accordance to the Decree of the Minister for Environment (2004).

In total, 288 determinations in three series were made. For each of the three wells from every village, the arithmetic means of the parameters were calculated on a basis of samples collected on three dates.

Fig. 1. Localization of checkpoints for well water sampling

The underground water quality was assessed in compliance with the Decree of the Minister for Environment of 11 February 2004 (*Rozporządzenie...* 2004) on surface and underground water status, screening performance, interpretation and presentation of the status of such waters. In addition, the Decree of the Minister for Health of 29 March 2007 (*Rozporządzenie...* 2007) was referred to. The results were compared to concentrations of forms of particular components ($N\text{-}NH_4$, $N\text{-}NO_2$, $N\text{-}NO_3$) specified in the Decree of the Minister for Environment (2004) and the Decree of the Minister for Health (2007) in order to designate water quality classes and principal chemical requirements to the well waters examined.

Pearson's correlation coefficient was used to evaluate the dependence of the parameters examined on a distance between a given well and the selected characteristics of a farm. The dependence is illustrated by Figure 2.

The data clustering method, based on a notion of a distance between objects or variables in multi-dimensional space, was also used to analyze the results. The technique makes it possible to present grouped objects or their traits in the form of a bundle diagram. Calculation of Euclidean distance is a direct way to estimate the distance between objects. This measure determines the real geometric distance between objects in space, and is calculated using raw data. The method consists of presenting similarity between objects or their features (variables) as a function of distance. Objects

Table 1

Localization of the wells

Village	Well	Distance from farm buildings	Distance from houses	Distance from the Narew
		(m)		
Złotoria	1	13	8	100
	2	8	15	50
	3	10	30	100
Topilec	1	6	5	100
	2	50	5	40
	3	6	5	150
Uhowo	1	0	3	200
	2	10	5	30
	3	20	3	50
Zawyki	1	0	20	150
	2	20	5	70
	3	15	30	30
Kaniuki	1	30	20	50
	2	3	20	30
	3	25	1	50
Ciełuszki	1	30	3	10
	2	30	15	50
	3	9	10	50
Narew	1	30	7	200
	2	17	5	200
	3	16	7	50
Sobótka	1	0	15	50
	2	35	8	50
	3	5	18	70

are grouped in sets called clusters that combine particular variables. The variables are more similar to one another when the distance between them is smaller.

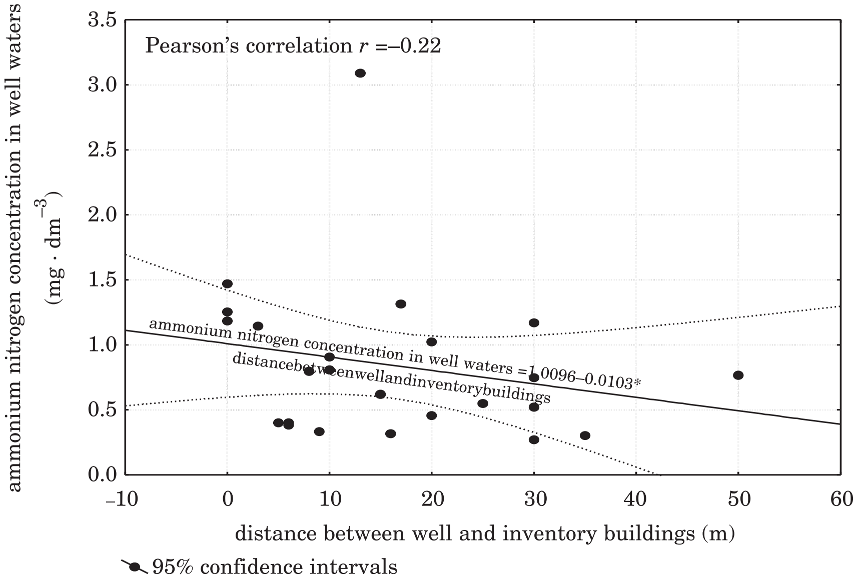


Fig. 2. Dependence of ammonia concentration in well waters on the distance between well and farm buildings

RESULTS AND DISCUSSION

The quality of the well waters we studied differed in particular villages. Most of the water samples were characterized by excessive content of ammonium nitrogen (according to the Decree of the Minister for Environment 2004), which indicates direct contact of water intake points with organic contaminants. They may originate not only from animal waste, but also from household sewage wrongly managed on a farm. The highest concentrations of ammonium nitrogen were found in the wells in Złotoria (1.6 $mg \cdot dm^{-3}$ – IV quality class for underground waters), while the lowest ones (0.52 $mg \cdot dm^{-3}$ – IV quality class) occurred in the wells in Topilec (Figure 3). Both values, nevertheless, exceeded norms regulating the quality of potable waters (0.39 $N\text{-}NH_4$ $mg \cdot dm^{-3}$).

Ammonium nitrogen present in waters usually originates from biochemical decomposition of organic nitrogen compounds, which proves a direct contact of water intake point with organic contaminants (BŁASZCZYK 1993).

SAPEK and PIETRZAK (1996) as well as URBANIAK and SAPEK (2003) claim that concentration of ammonia in water from piezometric wells localized on ploughed grounds is similar to that found on grasslands.

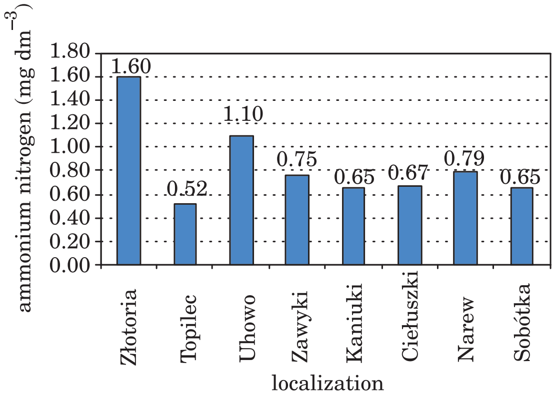


Fig. 3. Mean values of ammonium nitrogen

The highest level of nitrite nitrogen was recorded in the wells in Zawyki village (0.14 mg·dm^{-3} – out-of-class waters), whereas the lowest in Ciełuszki (0.02 mg·dm^{-3} – III class of underground waters quality) – Figure 4. These values did not exceed permissible level (0.39 mg·dm^{-3}) set by the Minister for Health (DECREE of 2007).

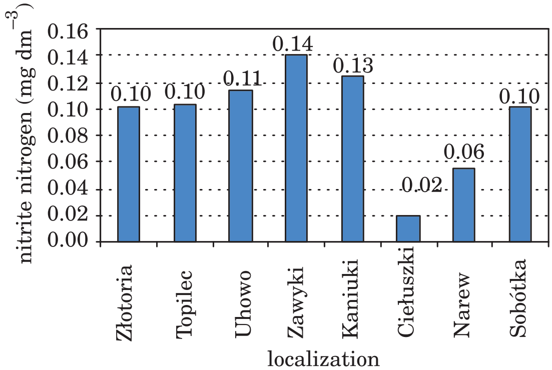


Fig. 4. Mean values of nitrite nitrogen

The highest content of nitrate nitrogen was recorded in waters from the wells localized in Zawyki (13.51 mg·dm^{-3} – IV quality class), while the lowest one in water samples collected in Uhowo (3.14 mg·dm^{-3} – II quality class for underground waters) – Figure 5. The concentration of nitrate nitrogen found in Zawyki village exceeded norms for potable waters (11.3 mg·dm^{-3}). The level of water saturation with nitrate nitrogen, which is the most common nitrogen compound in well waters, is one of the princi-

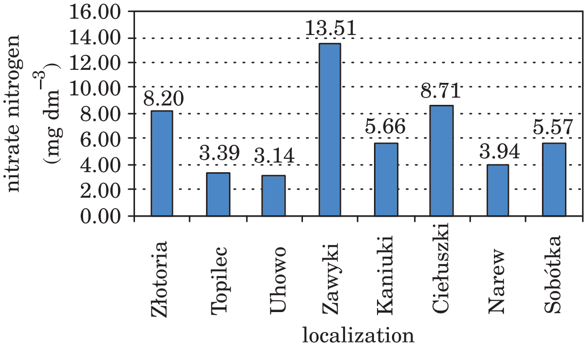


Fig. 5. Mean values of nitrate nitrogen

pal criteria for determination of well waters as potable waters (SĄDEJ, PRZEKWAS 2006). SKORBIŁOWICZ et al. (2001) observed that the well waters we analyzed were primarily affected by factors related to the agricultural and residential character of the villages, which largely contributed to the contamination of shallow waters supplied for consumption. Farmsteads and households are the main sources of contamination of well waters with nitrogen and phosphorus compounds.

ZAHN and GRIMM (1993) assumed that elevated concentration of chlorides in ground water is an indicator of the presence of nitrate nitrogen.

The highest phosphate concentration was recorded in well waters collected in Zawyki (3.07 $mg \cdot dm^{-3}$– III quality class), whereas the lowest one appeared in water from Ciełuszki village (0.84 $mg \cdot dm^{-3}$ – II class of underground water quality) – Figure 6. The content of phosphates in potable water is not regulated by any decree.

Contamination of drinking water with nitrogen compounds is usually accompanied by phosphate contamination. As phosphates can stimulate biological life, it is accepted that its presence at levels higher than 0.5 $mg \cdot dm^{-3}$ is unwanted (DURKOWSKI et al. 1997). The elevated concentrations of phosphates in the well waters we studied, likewise increased ammonium nitrogen, may have been a result of wrong handling of household wastewater and sewage, which contained residues of detergents (WIATER, SKOWROŃSKA 2002).

SAPEK and SAPEK (2005) claim that toiletries used for personal hygiene on a farm habitants, detergents for washing clothes and dishes, other detergents used to clean houses and other farm buildings as well as veterinary preparations, including those used for milking and washing milk containers and tanks, are the main sources of farm and ground water contamination, mainly with sodium and phosphorus.

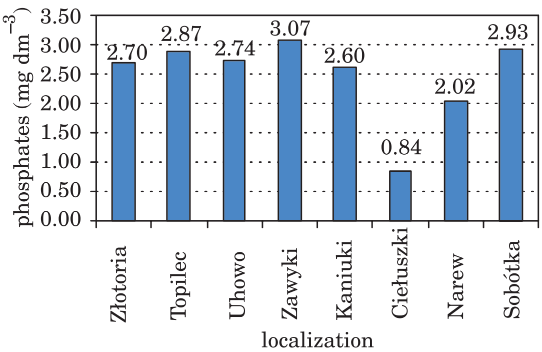
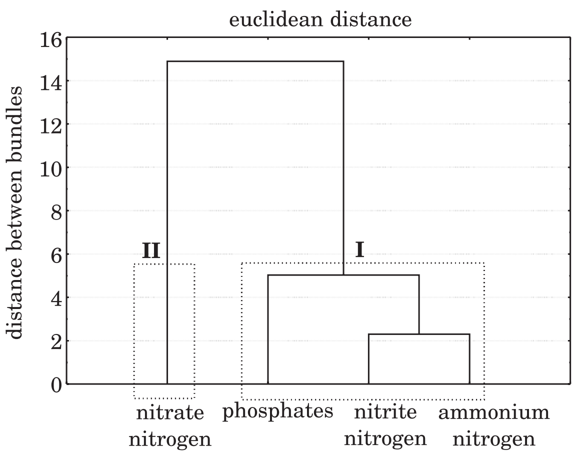


Fig. 6. Mean values of phosphates

A weak dependence of ammonia concentration in the well waters on the distance of a well from a nearby farmstead was discovered. It was manifested by the negative value of Pearson's coefficient r=-0.22 at p = 0.008. The mathematical model generated for this dependence, presented in Figure 2, makes it possible to predict the ammonium nitrogen concentration in well waters on a basis of a known distance of a well from a farm building.

The dendrogram (Figure 7), which includes the analyzed parameters, presents two arrangements: set I formed by phosphates, nitrite nitrogen, and ammonium nitrogen; set II consisting of a single indicator – nitrate nitrogen. The dendrogram illustrates a significant distance connecting set II with set I, which considerably differentiates both bundles. The bundle forming set II contains only one object called a separated point. The system presents the arrangement of nitrogen and phosphorus forms, which indi-



Fig, 7. Euclidean distances between parameters cates that the occurrence of these components in waters from the wells we investigated results from the vicinity of organic contamination sources (ammonium nitrogen, nitrite nitrogen, and phosphates – set I). Set II (nitrate nitrogen) indicates that the concentration of this nitrogen form is determined by nitrification processes and its elution by runoffs from fields and farms.

CONCLUSIONS

1. The well waters examined did not meet standards for potable waters established by the Decree of Minister for Health because they exceeded permissible values of ammonium nitrogen and nitrate nitrogen concentrations.

2. The lowest quality waters were found in the wells in villages Złotoria and Zawyki.

3. The distance from a well to the nearest farm building had an influence on ammonium nitrogen concentration in the well water.

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PHOSPHORUS RUNOFF FROM SMALL AGRICULTURAL CATCHMENTS UNDER DIFFERENT LAND USE INTENSITY*

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Abstract

The study was carried out in the hydrologic year 2006 and comprised two small agricultural catchments in the Masurian Lakeland. Both catchments possessed very large water retention potential (presence of buffer zones and surface waters in the catchments) but they differed in the intensity of agricultural production.

The study has demonstrated that the concentration of phosphorus in the surface waters flowing from the catchments ranged from 0.12 to 0.43 $mg \cdot dm^{-3}$. The catchments were characterised by low indices of water overland flow (0.32-1.67 $dm^3 \cdot s^{-1} \cdot km^{-2}$), which was a decisive factor shaping the runoff of phosphorus (from 0.013 to 0.060 $kg \cdot ha^{-1} \cdot year^{-1}$. A subcatchment dewatered by a pipe drain system, which was used for comparison, had a water flow of 3.54 $dm^3 \cdot s^{-1} \cdot km^{-2}$ and exported much more phosphorus with its surface waters (0.39 $kg \cdot ha^{-1} \cdot year^{-1}$). The results obtained during our study emphasise an important role played by small landscape water retention elements in both catchments and the efficiency of buffer zones at a contact of water bodies and fields in reducing the runoff of biogenic substances, especially when agricultural production carried out on a given catchment is intensive.

Key words: phosphorus, migration of biogenic substances, water flow, agricultural catchments, biogeochemical barriers.

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STRATY FOSFORU Z MAŁYCH ZLEWNI ROLNICZYCH O RÓŻNEJ INTENSYWNOŚCI UŻYTKOWANIA

Abstrakt

Badania prowadzono w roku hydrologicznym 2006, w dwóch małych zlewniach rolniczych położonych w obrębie Pojezierza Mazurskiego. Badane zlewnie charakteryzowały się dużymi zdolnościami retencyjnymi (obecność stref buforowych i wód powierzchniowych w zlewni) oraz zróżnicowanym poziomem intensywności rolniczego użytkowania.

Wykazano, że koncentracja fosforu w wodach odpływających z badanych zlewni rolniczych kształtowała się w zakresie 0,12÷0,43 mg·dm^{-3}. Zlewnie charakteryzowały się niskimi wskaźnikami odpływu wody (0,32÷1,67 dm^{3}·s^{-1}·km^{-2}), co w największym stopniu wpłynęło na odpływ fosforu, kształtujący się na poziomie od 0,013 do 0,060 kg·ha^{-1}·rok^{-1}. W porównawczej zlewni drenarskiej, o wskaźniku odpływu 3,54 dm^{3}·s^{-1}·km^{-2}, wykazano wielokrotnie wyższy eksport fosforu z wodami – 0,39 kg·ha^{-1}·rok^{-1}. Uzyskane wartości wskazują na rolę małej retencji krajobrazowej w badanych zlewniach oraz skuteczność stref buforowych na styku wód i pól uprawnych, ograniczających odpływ biogenów, zwłaszcza w warunkach intensywnej produkcji rolniczej na obszarze zlewni.

Słowa kluczowe: fosfor, migracja biogenów, odpływ wody, zlewnie rolnicze, bariery biogeochemiczne.

INTRODUCTION

The runoff of phosphorus from drainage basins to surface waters is a serious environmental threat associated with the increasing eutrophication of water bodies (Kajak 2001, Schippers et al. 2006). In larger hydrographic systems, owing to the gradual elimination of point pollution sources achieved by constructing and developing wastewater treatment plants in urbanised areas, the volume of phosphorus load reaching surface waters will typically depend now on the size of phosphorus runoff from agricultural catchments (Ekholm, Mitikka 2006). The risk of contaminating surface waters by non-point sources of pollution is most evident in farmlands under intensive agricultural practice; this effect is accompanied by a large decrease in the contribution of point sources of pollution (Carpenter et al. 1998). Non-point pollution sources are much more difficult to reduce, hence it is important to monitor the runoff of phosphorus from agricultural drainage basins. The actual amounts of phosphorus flowing from unit surface depend on several factors, such as landscape features, type of soils, weather conditions, type and intensity of agricultural production. Considering the diversity of such conditions for different locations and their time-related variability, any attempt at evaluating the volume of phosphorus that is not based on long-term and systematic studies will be burdened with a large error, the fact that has been supported by many reports published in our country (Giercuszkiewicz-Bajtlik 1990, Koc 1998, Bajkiewicz-Grabowska 2002).

The objective of this study has been to determine the volume of phosphorus exported from two catchments, both characterised by a relatively low index of unit surface water runoff. The underlying hypothesis was that the export of biogenic substances from areas characterised by the surface water runoff being much lower than the average should be equally low. Another aim of the research was to compare the phosphorus runoff index values according to the intensity of agricultural use of both basins.

MATERIAL AND METHODS

The study was carried out in the hydrologic year 2006 and comprised two catchments lying in geographical region of Masurian Lakeland (Pojezierze Mazurskie). Although both objects – Sętal and Doba – can be defined as small agricultural drainage basins, they are clearly different in the intensity of agricultural production. Sętal basin is under extensive agricultural use, whereas the area drained to Dobskie Lake is at a serious risk of being polluted by agricultural points of pollution, as it is farmed rather intensively.

Sętal (Figure 1) is a subcatchment of a larger catchment of the middle Łyna River, lying 20 km north of Olsztyn. It covers about 24 km^2 and is drained by a surface stream called the Sętalska Struga. The control plane taken for our study (point 439) on this stream lies about 500 m away from its flow into the Łyna River. The study also included the drainage basin of the northern side of the Sętalska Struga (point 440). The whole basin covers 1,500 ha (Table 1), whereas the other, northern stream (nameless) drains 980 ha in the northern part of the catchment. The land utilisation structure of these objects is dominated by arable land (60%) and grassland (25%), mostly under extensive farming. In the 1990s half of the land was laid fallow. The catchment has a rather small contribution of wooded areas (10%) and a large share of surface waters (5%). Our study also comprised the tributaries of Nowe Włóki Lake (points 431A and 432A), lying in the upper part of this basin, which drained 50 and 172 ha, respectively, and consisted mainly of arable lands. All the above subcatchments are dominated by sandy loam and loamy sand soils, which were classified as soil valuation classes IVa and IVb in the Polish soil classification system.

Doba object contained three subcatchments of Dobskie Lake (Figure 1). The subcatchment designated as number 522 covered 28 ha. It contains a surface stream which flows to the main basin of Dobskie Lake near the village Doba and in its upper part is fed by pipe drains, which drain arable lands. The farmland in this catchment lies on medium compact soils (loamy sand) classified as IIIb and IV soil valuation classes. In the year of our study, the arable lands received pre-sowing mineral fertilization corresponding to 110 kg $N \cdot ha^{-1}$. In the spring and autumn of that year the fields were

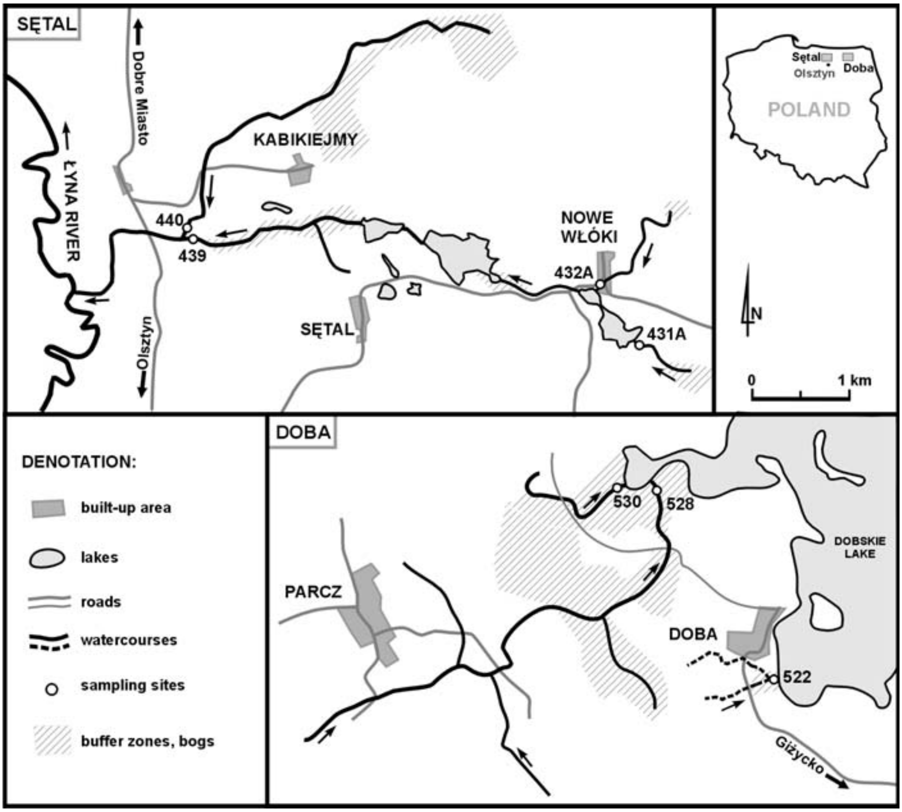

Fig. 1. Location and plans of Sętal and Doba catchments

Table 1

Basic hydrologic data of the control planes of set at the examined watercourses

Indicator	Sętal				Doba		
	431A	432A	439	440	522	528	530
Catchment area (ha)	50	172	1505	980	28	1616	109
Mean water flow ($dm^3 \cdot s^{-1}$)	0.25	0.53	23.9	3.12	0.99	12.2	0.41
Min-max flow ($dm^3 \cdot s^{-1}$)	0.0 1.9	0.0 2.9	1.6 107.3	0.8 9.7	0.3 2.9	0.0 47.8	0.0 1.7
Water overland flow ($dm^3 \cdot s^{-1} \cdot km^{-2}$)	0.50	0.31	1.67	0.32	3.54	0.75	0.38

also fertilized with FYM, in total 15 $m^3 \cdot ha^{-1}$. About 15% of the catchment area is overgrown with groups of trees, mainly near the mouth of the watercourse.

The subcatchment of point number 528 contains the recharge area of Pilwa Bay located at the westernmost end of Dobskie Lake. This surface stream drains a large area (1,616 ha) mostly (57%) covered by forest. The arable lands in this subcatchment are mainly light loamy sands (soil valuation class IVb). The arable lands received mineral fertilization (130 kg $N \cdot ha^{-1}$ and 60 kg $P \cdot ha^{-1}$) and liquid manure (15 $m^3 \cdot ha^{-1}$) with the latter divided into two rates: spring and autumn. The stream designated as number 530 dewaters the western part of the subcatchment of Pilwa Bay. The surface area of this subcatchment is 109 ha and 60% of this land is afforested. The remaining part consists of arable lands, which belong to the soil fraction of light loamy sand and receive mineral fertilization only (100 kg $N \cdot ha^{-1}$ and 60 kg $P \cdot ha^{-1}$).

Doba catchment contains an area distinguished by the Order of the Director of the Regional Water Management Board in Warsaw (*Rozporządzenie...* 2004) as being under a particularly severe ecological threat, where the phosphorus runoff from agricultural sources should be limited. This area comprises the whole basin dewatered by the pipe drains (no 522) and the eastern part of the watercourse catchment no 528.

Our study on the migration of phosphorus in the above catchments was carried out in the hydrologic year 2005/2006, which was characterised by the rainfall rated as average on a long-term scale (654 mm) and an average annual air temperature of 7.5°C. The year had low precipitation in early spring (March, April), which reduced the thawing water runoff. Such a distribution of precipitation alongside the dry previous year meant that the first half of the year was poor in rainfall, but the annual rainfall reached the long-term average owing to heavy rains in August and September.

The study consisted of measurements of the intensity of water flows, performed weekly using an electronic meter Valeport Model 550. In each month water samples were collected from all the watercourses to perform laboratory analyses. Total phosphorus and its mineral forms were determined by spectrophotometry using ammonium molybdate and tin (II) chloride.

RESULTS

The results showed that the concentration of phosphorus in the analysed water was only slightly variable, even though the research objects were highly different in their basic drainage characteristics. All the control planes of the watercourses were characterised by a low index of water outflow, ranging from 0.31 to 3.54 $dm^3 \cdot s^{-1} \cdot km^{-2}$ (Table 1). In the smaller subcatchments drained by periodic watercourses (431A, 432A, 530), the value of this index was sometimes less than 0.5 $dm^3 \cdot s^{-1} \cdot km^{-2}$ (Table 1). The runoff in watercourse 522, supplied in its upper section by a pipe drain system, was the largest but even there the value of a unit flow was lower than an average value for lake districts, which is assessed at 5.0-6.0 $dm^3 \cdot s^{-1} \cdot km^{-2}$ (Fal 1993).

The mean annual concentrations of total phosphorus at the particular control planes oscillated within 0.12-0.43 $mg \cdot dm^{-3}$ (Table 2). The maximum phosphorus concentration was found in the watercourse carrying the water drained from heavily fertilized fields (522), but even in this case the phosphorus volume never reached the extremely high values cited in the literature (Koc, Skwierawski 2004). The waters flowing from the catchment of Dobskie Lake through the surface streams, which are separated from the fields by marshes (points 528 and 530) contained much lower concentrations of phosphorus, comparable to those in the waters from the other object, the Sętalska Struga, where the catchment surface area is under a much less intensive agricultural production.

In the annual distribution, higher levels of phosphorus occurred in most

Table 2

Mean concentrations of total phosphorus (P_{tot}) and phosphate phosphorus ($P\text{-}PO_4$) in water flowing from Setal and Doba catchments ($m \cdot dm^{-3}$)

Site number	P_{total}			$P\text{-}PO_4$		
	winter half year	summer half year	year	winter half year	summer half year	year
Sętal						
431A	0.12	0.13	0.12	0.08	0.03	0.05
432A	0.12	0.37	0.24	0.10	0.31	0.20
439	0.11	0.27	0.19	0.04	0.07	0.0
440	0.22	0.29	0.26	0.04	0.04	0.04
Doba						
522	0.48	0.36	0.43	0.11	0.19	0.14
528	0.22	0.26	0.24	0.06	0.05	0.06
530	0.15	-	0.15	0.02	-	0.02

of the sites during the summer half-year. This dependence was more pronounced for total phosphorus amounts, whereas the levels of phosphorus mineral forms were more diverse (Table 2).

The concentration of phosphate phosphorus was clearly the highest in the watercourse flowing to the northern part of Nowe Włóki Lake (432A, Table 2). The elevated levels of phosphorus in this stream should be attributed to the presence of built-up areas in this subcatchment.

While analysing the results, no statistically significant differences were discovered (using Duncan's test at p = 0.05) in the concentrations of phosphorus between the objects located in Sętal (on average 0.203 mg $P_{tot} \cdot dm^{-3}$) and Doba catchments (0.195 mg $P_{tot} \cdot dm^{-3}$), although the agricultural use of land in the catchment drained by the watercourses feeding Dobskie Lake was much more intensive.

As the variability of the phosphorus concentration at the particular sites was rather low, the volume of phosphorus flowing from the analysed catchments depended mainly on the flow intensity, which in both catchments was rather low, suggesting high water retention capacity. And this, in turn, was a decisive factor affecting the volume of phosphorus runoff. The unit surface phosphorus runoff in both catchments was very small, even at the control planes of those watercourses where water continued to flow throughout the whole year (439, 440).

The runoffs of P_{tot} (total phosphorus) were varied at the particular sites (Table 3). Approximately similar and all very low runoffs were found in the smallest subcatchments, dewatered by periodically flowing streams, i.e. the subcatchments of the watercourses supplying Nowe Włóki Lake (431A and 432A) and the subcatchment of the stream flowing to Pilwa Bay in Dobskie Lake (530). The runoffs of phosphorus from these subcatchments never exceeded 0.02 $kg \cdot ha^{-1} \cdot year^{-1}$. Also the northern part of the Sętalska Struga catchment (440), dewatered by a continually flowing watercourse characterised by very small annual water flow intensity variation, was determined to carry away very small volumes of phosphorus: 0.023 $kg \cdot ha^{-1} \cdot year^{-1}$ (Table 3).

Table 3

Loads of total phosphorus (P_{tot}) and phosphate phosphorus ($P\text{-}PO_4$) in water flowing from Sętal and Doba catchments ($kg \cdot ha^{-1} \cdot year^{-1}$)

Site number	P_{total}			$P\text{-}PO_4$		
	winter half year	summer half year	year	winter half year	summer half year	year
Sętal						
431A	0.012	0.008	0.020	0.008	0.003	0.011
432A	0.005	0.015	0.020	0.004	0.012	0.016
439	0.014	0.046	0.060	0.004	0.012	0.016
440	0.013	0.010	0.023	0.002	0.001	0.003
Doba						
522	0.284	0.108	0.391	0.074	0.060	0.134
528	0.044	0.015	0.058	0.010	0.007	0.017
530	0.013	-	0.013	0.002	-	0.002

The watercourse fed by the pipe drains (522), taken as a comparative object for the other sites, drained an area similar in the farming intensity of arable lands but less afforested, where the water runoff was larger but the volume of phosphorus removed with the surface water was lower by several orders than in the other objects (0.39 $kg \cdot ha^{-1} \cdot year^{-1}$). Such a value of phosphorus runoff can be regarded as typical for catchments dominated by arable lands (KOC 1998, KOC, SZYMCZYK 2003). The variability in the amounts of mineral phosphorus forms was shaped likewise (Table 3).

The results obtained during our study, being lower (except for site 522) than the values reported in the literature (GIERCUSZKIEWICZ-BAJTLIK 1990, KOC, SZYMCZYK 2003), did not show any relationship between the phosphorus runoff and intensity of agricultural use of land. According to GRANIŃSKA et al. (2005), the largest export of phosphorus from a catchment can occur in areas which contain a large share of farmland used intensively or big urban settlements. In contrast, the lowest loads of phosphorus in watercourses were found in drainage basins covered with extensive grasslands, marshes or lakes. The results obtained for Dobskie Lake catchment, considered to be particularly vulnerable to an influx of pollutants from agricultural sources to its watercourses, suggest that migration of phosphorus can be reduced in drainage basins which possess natural barriers to biogenic substances on their way away from a catchment. Biogeochemical barriers can affect the runoff of phosphorus mainly by halting the surface flow of waters. This is important as phosphorus carried by surface streams travels much faster than in underground waters (ARCHEIMER, LIDEN 2000).

The present study has shown that the average indices of the phosphorus runoff from a catchment to its surface streams do not take into consideration the presence of buffer zones along the watercourses, which can act as biogeochemical barriers, thus limiting the influx of pollutants to waters. In one of the examined catchments, that is the drainage basin of Dobskie Lake, the watercourses which dewater the area lie in close proximity to intensively farmed lands at the total length equal 30% of their whole aggregate length. In addition, a large share of water-logged lands near the watercourses means that the concentrations and loads of phosphorus exported from the catchment were relatively small (Tables 2, 3). Under such conditions, the indices used for determination of the volumes of phosphorus loads reaching a lake from its catchment should be corrected by including the presence of areas highly capable of retaining water and accumulating matter, such as phosphorus.

CONCLUSIONS

1. The concentration of phosphorus in waters flowing from the examined agricultural catchments ranged from 0.12 to 0.43 $mg \cdot dm^{-3}$. Although the farming production in Doba catchment was intensive, no significant differences were found regarding the concentrations of phosphorus in both analysed catchments, which differed in the intensity of land use. The results indicate that land use intensity as a factor affecting the environment does not produce an unambiguous effect on the levels of phosphorus in areas under agricultural production.

2. The low levels of phosphorus runoff determined in both catchments were caused mainly by the atmospheric factors in the time period preceding our study, and by the landscape relief, which determined small values of water flow from the catchments (0.32-1.67 $dm^3 \cdot s^{-1} \cdot km^{-2}$). These conditions affected most strongly the runoff of phosphorus, which remained on a low level (0.013 to 0.060 $kg \cdot ha^{-1} \cdot year^{-1}$). The comparative catchment, partly dewatered through pipe drains, where the water flow was much higher (3.54 $dm^3 \cdot s^{-1} \cdot km^{-2}$), was found to experience a much larger phosphorus runoff via surface watercourses (0.39 $kg \cdot ha^{-1} \cdot year^{-1}$).

3. The values of the phosphorus runoff indices for the examined catchments prove that buffer zones at the contact between watercourses and fields under intensive agricultural production are very important, which means that they should be maintained and created, particularly in areas under intensive agricultural production.

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THE INFLUENCE OF SOIL REACTION ON THE EFFECTS OF MOLYBDENUM FOLIAR FERTILIZATION OF OILSEED RAPE

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Abstrakt

Thirty-three field trials have been carried out on soils of different acidity (pH=4.1-7.1) to investigate the effect of foliar fertilization with molybdenum on oilseed rape. Three rates of the element: 30, 60 and 120 g $Mo \cdot ha^{-1}$, were applied on two dates: in the spring, a few days after the growing season started, and during the early stem formation stage. In the group of trials on very acidic and acidic soils, seed yields were on average 0.08 $t \cdot ha^{-1}$ higher. The regression equation demonstrated that a relative seed yield increase obtained as a result of molybdenum fertilization depended on the concentration of manganese and percentage of the silt fraction in the soil. Oilseed rape grown on slightly acidic and neutral soils responded to molybdenum fertilization with an average seed yield increase of 0.02 $t \cdot ha^{-1}$. The rate of molybdenum had a significant effect on rape yields only in the group of trials set up on slightly acidic and neutral soils. No significant difference was found between 60 and 120 g $Mo \cdot ha^{-1}$. However, both of these rates were more effective than the lowest dose of 30 g $Mo \cdot ha^{-1}$. No evidence was found to support any relationship between the date of molybdenum application and seed yields or any interaction between Mo rates and application dates in either group of the trials.

Key words: Mo doses, time of application, yield increase, soil pH.

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WPŁYW ODCZYNU GLEBY NA EFEKTY DOLISTNEGO NAWOŻENIA RZEPAKU MOLIBDENEM

Abstrakt

Na glebach o różnej kwasowości (pH=4,1-7,1) przeprowadzono 33 doświadczenia polowe, w których badano efekt dolistnego nawożenia rzepaku molibdenem. Stosowano dawki 30, 60 i 120 g Mo·ha^{-1} w 2 terminach: wiosną, kilka dni po ruszeniu wegetacji, oraz na początku formowania łodygi. W grupie doświadczeń z glebami bardzo kwaśnymi i kwaśnymi uzyskano zwyżki plonów nasion, średnio 0,08 t·ha^{-1}. Według równania regresji, względna zwyżka plonów wskutek nawożenia Mo zależała od zawartości manganu oraz procentowego udziału frakcji pyłu w glebie. W grupie doświadczeń z glebami lekko kwaśnymi i obojętnymi rzepak reagował zwyżką plonów – średnio 0,2 t·ha^{-1}. Wielkość dawki molibdenu miała istotny wpływ na plonowanie rzepaku tylko w grupie gleb lekko kwaśnych i obojętnych. Nie stwierdzono istotnej różnicy między dawkami 60 g i 120 g Mo·ha^{-1}. Obie były jednak skuteczniejsze niż dawka 30 g Mo·ha^{-1}. Nie udowodniono wpływu terminu oprysku molibdenem na plony nasion ani współdziałania wielkości dawki Mo z terminem jej stosowania w obu grupach doświadczeń.

Słowa kluczowe: dawki Mo, terminy aplikacji, zwyżka plonu, pH gleby.

INTRODUCTION

Oilseed rape as a crop is gaining in economic importance all over the world. Its seeds are a valuable material for production of edible oil as well as biofuels. Oilseed rape is known to be a demanding crop in terms of macroelemental nutrition, but the information on microelements it requires remains scarce. The available literature focuses on *Cruciferae* family (Katyal, Randhawa 1983) and oilseed rape (Shorrocks 1990) being very susceptible to B and Mo deficit.

In Poland, about 60% of arable land consists of acidic soils, in which molybdenum availability to plants is limited. Countries in which soils are sandy and acidic (Gupta, MacLeod 2006), for example south-western Australia (Brennan 2006), struggle with a similar problem of limited molybdenum availability. There, many crops, particularly large wheat plantations, receive molybdenum fertilizers, as liming is not economically viable (Brennan, Bolland 2007). According to Finck (1998), oilseed rape should be fertilized with Mo under the low pH conditions and high nitrogen application. Sulphur application can also decrease the Mo uptake by oilseed rape (Balik et al. 2007).

Oilseed rape, which is not always cultivated on soils providing the crop with optimum reaction, is likely to suffer from insufficient molybdenum supply. The objective of our study has been to test the response of winter oilseed rape to foliar fertilization with molybdenum under the conditions of different soil pH.

MATERIALS AND METODS

The trails were performed on commercially grown winter oilseed rape plantations in 8 Polish provinces. In total, 33 testing sites had been selected. The soils were highly varied in soil reaction (4.1 to 7.1 pH in 1 mol $KCl \cdot dm^{-3}$). Most of the tests were performed on acidic and slightly acidic soils, characterised by various abundance in macroelements (Table 1).

Table 1

Percent distribution of trials according to soil acidity and fertility

Soil fertility	P	K	Mg	Mo	Soil reaction	
Very low	7	7	18	-	very acid	12
Low	22	18	36	14	acid	36
Medium	43	47	36	75	slightly acid	36
High	21	14	10	11	neutral	16
Very high	7	14	-	-	alkaline	-

Strict experiments with 4 replications were set up on fields under winter oilseed rape. The experiments were established as two-factor split-plots with a single control object. The design of the experiments was as follows:

The control – no Mo fertilization

– factor I – date of fertilization with Mo 1) in spring, a few days after the onset of the growing season, 2) the early stem formation stage;

– factor II – molybdenum rates: 1) 30 g $Mo \cdot ha^{-1}$, 2) 60 g $Mo \cdot ha^{-1}$ and 3) 120 g $Mo \cdot ha^{-1}$.

Molybdenum was applied as an aqueous solution of ammonium molybdate which contained 54% Mo.

In spring, immediately before Mo fertilization, soil samples were taken. Oilseed rape seeds, in turn, were sampled during the harvest. Laboratory examination of the soil and plant material was carried out using the methods commonly applied at chemical and agricultural laboratory stations. Granulometric composition of the soils was examined according to the procedure described by Casagrande-Prószyński; soil pH was tested in 1 mol $KCl \cdot dm^{-3}$ and the available forms of phosphorus and potassium were determined by Egnar-Riehm method, while these of magnesium were assayed using Schachtschabel method. The results were processed statistically. Differences in seed yields between the fertilization objects were tested using analysis of variance for multiple experiments. The tables which compile yields and mean values for the first and second factor levels are set alongside the values of the so-called ‘contrast’, i.e. the difference in yields between the control and an average yield for molybdenum fertilized objects. Correlation and regression calculations, obtained with an aid of Statographics software, were used to draw conclusions.

RESULTS AND DISCUSSION

Certain pot experiments conducted in the 1980s and earlier in Poland demonstrated that oilseed rape grown on acid soils was lower in Mo with soil liming being capable of raising Mo concentration in plant tissues. It could be hypothesised that the response of oilseed rape to foliar fertilization with Mo would be more pronounced under low soil pH than on soils of the reaction closer to neutral one. The results of our studies, however, do not confirm this hypothesis. The analysis of variance for the yields showed that on very acidic and acidic soils (pH=5.5), an average seed yield rise attributed to molybdenum foliar sprays was 0.08 $t \cdot ha^{-1}$, thus being within the significance limit (Table 2).

Table 2

Average yields of oilseed rape ($t \cdot ha^{-1}$) from experiments on very acidic and acidic soils

Time of application (factor I)	Mo rate ($g \cdot ha^{-1}$) (factor II)			Average of factor I
	30	60	120	
1	2.91	2.92	2.96	2.93^{A}
2	2.90	2.92	3.00	2.94^{A}
Average of factor II	2.91^{a}	2.92^{a}	2.98^{a}	2.94
Average of control plots				2.86
Contrast				0.08*

*, **, *** – significance of differences at $\alpha < 0.05$; $\alpha < 0.01$; $\alpha < 0.001$
Different letters indicate significant differences at a 5% level according to Tukey's test, factor I – capital letter, factor II – small letter.

In the other group of trials, set up on slightly acidic and neutral soils, the effect of molybdenum fertilization was stronger than on soils of the reaction ≤=5.5. Statistically significant increase in seed yield in this group equalled 0.2 $t \cdot ha^{-1}$ (Table 3).

The rate of molybdenum had a significant effect on oilseed rape yields only in the group of trials on slightly acidic and neutral soils, where no significant differences were observed between the effects produced on seed yields by the fertilization rates 60 and 120 g $Mo \cdot ha^{-1}$, although they were both more effective than the rate of 30 g $Mo \cdot ha^{-1}$. No effect of the date

Table 3

Average yields of oilseed rape ($t \cdot ha^{-1}$) from experiments on slightly acid and neutral soils

Time of application (factor I)	Mo rate ($g \cdot ha^{-1}$) (factor II)			Average of factor I
	30	60	120	
1	2.93	3.05	3.09	3.02^A
2	3.01	3.05	3.05	3.03^A
Average of factor II	2.97^a	3.05^b	3.07^b	3.03
Average of control plots				2.82
Contrast				0.21 ***

*, **, *** – significance of differences at $\alpha < 0.05$; $\alpha < 0.01$; $\alpha < 0.001$
Different letters indicate significant differences at a 5% level according to Tukey's test, factor I – capital letter, factor II – small letter.

of molybdenum application on seed yields was found. Neither did we observe any interaction between the rate of the element and the date of the fertilization treatment in any of the two groups of the experiments.

Some research carried out in Australia showed that on very acidic soils it was possible to attain higher wheat yields when Mo fertilization had been applied. The actual yield increment depended on the concentration of molybdenum in seed material. The lower the Mo content in the seeds, the better the yield stimulating effect of Mo fertilization (Brennan, Bolland 2007). Rates of Mo between 35 and 140 $g \cdot ha^{-1}$ had a significant effect. Common bean has also been examined towards increasing the Mo concentration in seeds by using very high rates of Mo, between 90 and 720 $g \cdot ha^{-1}$, applied to leaves (Vieira et al. 2005). By enriching seed material with Mo, it is possible to do without later Mo fertilization treatments.

In Germany, a study based on 70 field experiments showed that there was no relationship between soil reaction and the response of oilseed rape to Mo foliar fertilization (Finck, Sauermann 1998). Very few cases of seed yield increase occurred when the Mo concentration in rape leaves was below the optimum level, which is 0.4 mg $Mo \cdot kg^{-1}$ d.m. (Bergmann 1992). However, a fertilization rate of 40 Mo $g \cdot ha^{-1}$, which was applied in the reported study, seems to be too low for oilseed rape. According to the Australian data, application of comparable rates of molybdenum to another crop such as grapevine made the yields increase by 70-750% (Williams et al. 2005).

In our own studies, small increase in oilseed rape yields as a result of Mo fertilization which occurred only on very acid and neutral soils could be explained by the fact that on such soils there are a number of yield limiting factors (Brennan et al. 2004). These include low availability of Mo as much as toxicity of Al and Mn, limited availability of P and inhibited uptake of Ca and Mg. In a situation where several yield-determining factors occur on a minimum level, rising one of them up to the optimum position may not suffice to guarantee higher yields (WALLACE 1984). On soils characterised by a higher pH, the factors which inhibit the growth and development of crops are eliminated, which explains a more pronounced effect of foliar application of molybdenum.

Simple correlation calculations, computed on the basis of the data from the whole set of the experiments, did not show any relationship between the value of soil pH and yield increase obtained as a result of Mo fertilization (Figure 1). Within the whole range of soil pH values, there were both yield increases and decreases caused by Mo fertilization, but on soils of the pH ≤=5.5 yield the increments were smaller while declining yields occurred more often and were sometimes bigger than on soils of higher reaction.

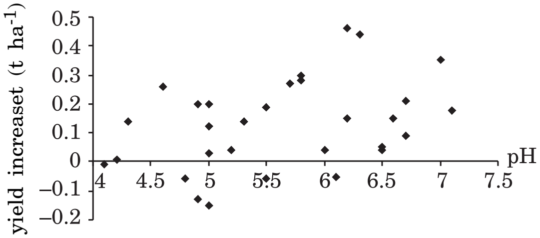

Fig. 1. Relationship between seed yield increase after Mo application and soil pH

In both groups of trials, regression correlation was searched between a relative yield increase obtained on molybdenum fertilized objects versus the control and some physicochemical properties of soil, such as abundance of available forms of macro- and microelements in soil or the content of floatable particles and silt fractions. For the soils characterised by pH ranging within 5.6-7.1 we were unable to create a regression model with the above variables as the population we investigated was uniform in this respect. The relationship for the trials established on very acidic and acidic soils was defined with the following regression function:

$$y = 1.106015 - 0.004865 \text{ silt} + 0.000801 \text{ Mn} \quad R^2 = 0.731^{***}, n = 14$$

where:

y = relative yield increase for 120 g $Mo \cdot ha^{-1}$ versus the control (%),

silt – content of silt fraction in soil (%),

Mn – concentration of available Mn in soil ($mg \cdot kg^{-1}$).

With this function it was demonstrated that on the soils of pH up to 5.5. the effect of molybdenum fertilization on yield increase was negatively correlated with the silt fraction but positively correlated with the percentage of available manganese in soil. A review of the relevant references indicated that higher yields could be obtained owing to some interactions between nutrients (FAGERIA 2001). Manganese can impede the uptake of molybdenum (KABATA-PENDIAS 2001). In the above equation, increased Mn concentration in soil was accompanied by a stronger response of oilseed rape to molybdenum fertilization, which reflects the Mn – Mo antagonism. Excess of manganese in soil, which impedes the uptake of molybdenum by plants, was responsible for their stronger response in yields to Mo foliar fertilization.

The negative effect of the silt fraction in soil on oilseed rape yield increase stimulated by Mo fertilization is difficult to explain. Most probably, there is some indirect relationship. The silt fraction exerts quite strong effect on water and air ratios in soil, which are associated with the oxidation / / reduction potential as well as molybdenum availability to plants.

CONCLUSIONS

1. Despite the commonly held opinion that molybdenum fertilization is a recommendable measure only on acidic soils, the highest winter oilseed rape yield increases stimulated by molybdenum foliar application were obtained on soils of pH > 5.5. On acid and very acidic soils, the effects of Mo fertilization were much lower and sometimes even negative, which meant that the seed yield was depressed.

2. On soils of pH > 5.5, the best results were obtained when 60-120 g $Mo \cdot ha^{-1}$ was applied, irrespective of the fertilization date (a few days after the vegetative season had begun or during the early stem formation stage). This seems to suggest that the exact date for Mo fertilization can be adjusted to combine it with a specific plant protection treatment.

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CONTENT OF CARBON, HYDROGEN AND SULPHUR IN BIOMASS OF SOME SHRUB WILLOW SPECIES

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Abstract

Carbon, hydrogen and sulphur were determined in biomass (shoots and roots) of five species of shrub willow: *Salix viminalis*, *Salix dasyclados*, *Salix triandra*, *Salix purpurea* and *Salix alba*. Samples of the biomass collected from a strict experiment were dried to constant weight at 105°C. The content of carbon, hydrogen and sulphur was determined in an automatic determinator ELTRA CHS 500. In addition to the chemical determinations, willow plants were assessed, in the first year of cultivation, in terms of their morphological traits and yields. The results underwent statistical analysis using Excel spreadsheets and Statistica PL software programme.

An average content of carbon in the biomass of the five analysed willow species was 496.33 $g \cdot kg^{-1}$ d.m. Among the five species, the highest carbon content was discovered in the biomass of *Salix purpurea* (501.32 $g \cdot kg^{-1}$ d.m.). An average content of hydrogen in aerial parts of shrub willow was significantly higher than in their roots (507.95 vs 465.88 $kg^{-1} \cdot$ d.m.). The content of hydrogen in all the species of shrub willow was on average 71.11 $g \cdot kg^{-1}$ d.m., with the highest hydrogen amount in biomass attributed to *Salix triandra* (74.98 $g \cdot kg^{-1}$ d.m.). This species, on the other hand, had the lowest concentration of sulphur in its biomass (on average 0.43 $g \cdot kg^{-1}$ d.m.). The average content of sulphur in aerial parts was significantly lower than in roots of willow plants (0.52 vs 0.95 $g \cdot kg^{-1}$ d.m.). This relationship held true for all the five species of willow plants.

An average yield of dry biomass obtained from the five shrub willow species in the year when the experiment was established was 5.81 $Mg \cdot ha^{-1}$. The highest dry biomass yield was produced by *Salix viminalis* (7.22 $Mg \cdot ha^{-1}$).

Key words: shrub willow, productivity, biomass, carbon, hydrogen, sulphur.

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ZAWARTOŚĆ WĘGLA, WODORU I SIARKI W BIOMASIE WYBRANYCH GATUNKÓW WIERZB KRZEWIASTYCH

Abstrakt

W biomasie pięciu gatunków wierzby krzewiastej: *Salix viminalis, Salix dasyclados, Salix triandra, Salix purpurea* oraz *Salix alba*, w pędach i korzeniach oznaczono zawartość węgla, wodoru i siarki. Pobrane ze ścisłego doświadczenia polowego próby biomasy wierzby wysuszono w temp. 105°C do uzyskania stałej masy. Zawartość węgla, wodoru i siarki w biomasie oznaczono w automatycznym analizatorze ELTRA CHS 500. Ponadto określono cechy morfologiczne oraz produktywność roślin *Salix* spp. w pierwszym roku uprawy. Wyniki badań opracowano statystycznie z użyciem arkusza kalkulacyjnego Excel oraz programu komputerowego Statistica PL.

Zawartość węgla w biomasie badanych gatunków wierzby krzewiastej wynosiła średnio 496,33 $g \cdot kg^{-1}$ s.m. Spośród badanych gatunków najwyższą zawartość węgla oznaczono w biomasie *Salix purpurea*, średnio 501,32 $g \cdot kg^{-1}$ s.m. Zawartość węgla w pędach nadziemnych była średnio istotnie wyższa (507,95 $g \cdot kg^{-1}$ s.m.) niż w korzeniach (465,88 $g \cdot kg^{-1}$ s.m.). Zawartość wodoru u badanych gatunków wynosiła średnio 71,11 $g \cdot kg^{-1}$ s.m. Istotnie najwyższą średnią zawartość wodoru w biomasie stwierdzono u gatunku *Salix triandra* (74,98 $g \cdot kg^{-1}$ s.m.). Gatunek *Salix triandra* zawierał istotnie najmniej siarki w biomasie, średnio 0,43 $g \cdot kg^{-1}$ s.m. Średnia zawartość siarki w pędach nadziemnych była istotnie niższa niż w korzeniach, odpowiednio 0,52 $g \cdot kg^{-1}$ s.m. i 0,95 $g \cdot kg^{-1}$ s.m. Zależność tę stwierdzono u wszystkich badanych gatunków wierzb.

Plon suchej biomasy badanych gatunków wierzb uzyskany w roku założenia doświadczenia wyniósł średnio 5,81 $Mg \cdot ha^{-1}$. Spośród badanych gatunków istotnie najwyższy plon suchej biomasy uzyskano u *Salix viminalis* – 7,22 $Mg \cdot ha^{-1}$.

Słowa kluczowe: wierzba krzewiasta, produktywność, biomasa, węgiel, wodór, siarka.

INTRODUCTION

Poland needs to increase the share of biomass in the total energy balance so as to improve its energy supply safety and enhance conservation of the natural environment. Using plant biomass for production of energy does not add to the warming effect. While growing, plants absorb the same amounts of carbon dioxide during photosynthesis as they release when their biomass is burnt (NALBORCZYK 2002, KISIEL et al. 2006). Production of energy from hardcoal creates a serious burden to the environment, such as elevated emission of carbon dioxide, sulphur dioxide, nitrogen oxides and flying ashes. In contrast to hardcoal combustion, biomass incineration makes it possible to achieve considerable reduction in SO_2 and organic pollutants, including polyaromatic hydrocarbons and flying organic compounds (KUBICA 2001)

Thus, comparison of fossil fuels and energy crops as energy sources should take into account the environmental burden caused by production of such fuels and emission of pollutants during energy generation. The available references lack detailed information concerning the content of carbon and hydrogen atoms in the cellulose and lignin biomass of shrub willows, which largely determine the calorific value of biomass-based solid fuels. The content of sulphur in energy crops should also be investigated, as it conditions the emission of this element to atmosphere while burning biofuels.

MATERIAL AND METHODS

The study was based on a field experiment conducted at the Research Station in Bałdy, the University of Warmia and Mazury in Olsztyn, Poland. The experiment was established in April 2006, on soil classified according to the Polish soil valuation system as defective wheat complex (class III b).

The first factor in the experiment comprised five species of shrub willow: *Salix viminalis*, *Salix dasycladosm*, *Salix triandra*, *Salix purpurea* and *Salix alba*. The second factor consisted of the type of willow biomass: aerial parts (leafless shoots) and underground biomass (roots).

The plants were not fertilised in the first year and the soil reaction was neutral. The grafts were planted in belts: 2 rows in a belt with 0.75 m space between the rows, followed by 0.90 m space between two belts and again 2 rows at 0.75 m spacing. On each plot, grafts were planted in 4 rows. The plants in a row were spaced at 0.25 m, which gave 48 thousand plants per hectare. The area of each plot was 23.1 m^2. The experiment was set up with three replications. At the end of the growing season, in November 2006, the willow plants underwent biometric measurements. The diameter of shoots was measured 50 cm above the ground surface on 10 randomly selected plants from the 2^{nd} and 3^{rd} row on each plot. Height of shoots (in m) was also measured on 10 plants per plot. Samples of biomass was taken for laboratory analyses. For this purpose, one whole plant was harvested from each plot (leafless aerial shoots and roots).

In the laboratory, proper samples of shoots and roots were obtained from the biomass of the five willow species. Biomass samples were dried up to constant weight at 105°C. The per cent contribution of shoots and roots in the dry matter of the *Salix* spp. plants was established. The yield of dry biomass ($Mg \cdot ha^{-1}$) was computed from the yield of fresh biomass of shoots and their moisture content. Dried samples of biomass were ground in an IKA analytic mill, using a 0.25 mm mesh sieve. The ground samples were again dried to constant weight, after which they were subjected to chemical analyses. The content of carbon, hydrogen and sulphur in the biomass of the analysed species of *Salix* spp. was determined in an automatic determinator ELTRA CHS 500.

The results of the study were processed statistically using Excel spreadsheets and Statistical software programme. Arithmetic means were calculated for all the analysed traits. In order to determine the mean concentrations of carbon, hydrogen and sulphur in the biomass (shoots and roots) of each willow species, corresponding weighted averages were computed. Using Duncan's significance test, LSD values at p=0.05 were derived. Finally, simple correlation coefficients for all the tested traits were computed.

RESULTS AND DISCUSSION

In the first growing season, aerial shoots accounted for 72.39% of dry biomass of willow plants, while the roots made up the remaining 27.61% (Figure 1). The lowest contribution of roots to the whole biomass was found for *S. triandra* (20.93%); the highest one – for *S. purpurea* (30.23%).

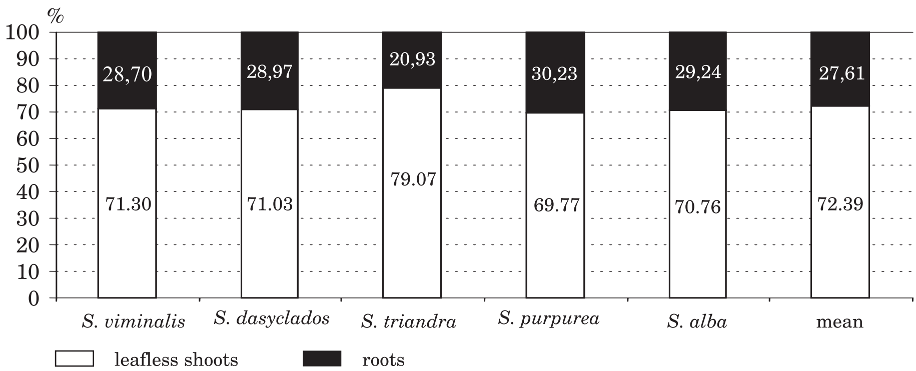

Fig. 1. Contribution of leafless shoots and roots in dry biomass obtained from *Salix* spp. plants

An average height of the shoots grown by the five willow species was 2.19 m (Table 1). Significantly higher shoots were developed by *S. viminalis* (2.70 m). *Salix dasyclados* grew comparably tall shoots, whereas the other willow species produced much lower shoots.

The thickest shoots were grown by *S. dasyclados* (14.73 mm). Similar diameters of shoots were obtained for *S. viminalis* and *S. triandra*. On the other hand, the diameter of shoots grown by *S. purpurea* and *S. alba* was significantly smaller compared to *S. dasyclados*.

The yield of dry aerial biomass obtained in the first growing season was on average 5.81 Mg·ha^{-1}. Among the five tested species of shrub willow, *Salix viminalis* produced the highest dry biomass yield (7.22 Mg·ha^{-1}), with *S. dasyclados* and *S. triandra* yielding on a comparable level. *Salix purpurea*, on the other hand, produced the lowest yields of dry matter (3.95 Mg·ha^{-1}). Yields of aerial biomass obtained in the first growing season being lower than in the consecutive years can be explained by the fact that

Table 1

Height and shoot diameter, yield of dry matter of the five shrub willow species

Species	Height of shoots (m)	Diameter of shoots (mm)	Dry biomass yield ($Mg \cdot ha^{-1}$)
Salix viminalis	2.70	13.27	7.22
Salix dasyclados	2.61	14.73	6.89
Salix triandra	2.02	11.70	6.78
Salix purpurea	1.68	7.70	3.95
Salix alba	1.95	11.50	4.19
Average	2.19	11.78	5.81
$LSD_{0.05}$	0.14	3.13	1.57

the plants needed to develop their root system, to the detriment of the growth and development of aerial parts. Kalembasa et al. (2006) report that yields of dry matter obtained from leafless shoots produced by *Salix viminalis* in the first growing season range from 3.21 to 6.51 $Mg \cdot ha^{-1}$, depending on fertilization. The analogous yields produced by a hybrid of *S. viminalis* and *S. purpurea* were much lower, from 0.45 to 1.04 $Mg \cdot ha^{-1}$. In the following years (2nd and 3rd growing season), yields of dry matter produced by *Salix viminalis* increased to 30 $Mg \cdot ha^{-1}$. In our own tests, the yielding potential of *Salix* spp. plants in the first year was much lower than in the following seasons (Stolarski 2002, Szczukowski et al. 2004).

The content of carbon in the biomass of the five willow species was on average 496.33 $g \cdot kg^{-1}$ (Table 2). The highest carbon content was determined in the biomass obtained from *Salix purpurea* (on average 501.32 $g \cdot kg^{-1}$ d.m.).

Table 2

Content of carbon in biomass of the five shrub willow species ($g \cdot g^{-1}$ d.m.)

Species (a)	Type of biomass (b)		Average
	shoots	roots	
Salix viminalis	506.89	475.09	497.77
Salix dasyclados	503.41	468.48	493.29
Salix triandra	505.86	456.29	495.48
Salix purpurea	514.53	470.84	501.32
Salix alba	509.07	458.68	494.34
Average	507.95	465.88	496.33
$LSD_{0.05}$	a – 5.8	b – 3.6	a × b – 8.2

Similar carbon concentrations were determined in the biomass of *Salix viminalis*. The average content of carbon in aerial parts was significantly higher than in roots (507.95 vs 465.88 g·kg^{-1} d.m.). Significantly more carbon was determined in the biomass obtained from shoots of *Salix purpurea* (514.53 g·kg^{-1} d.m.), whereas the lowest carbon level was found in roots of *Salix triandra* (456.29 g·kg^{-1} d.m.). The content of carbon in the biomass of *Salix* spp. was significantly positively correlated with the content of hydrogen (r=0.43) and significantly negatively correlated with the content of sulphur (r=-0.79), at n=30 (for 30 pairs of replications). Some other own studies showed that one-year shoots of *Salix viminalis* contained 484.1 g carbon per 1 kg^{-1} d.m. (STOLARSKI et al. 2005). Other chemical analyses carried out on various types of biomass in China by CUIPING et al. (2004) proved that an average content of carbon in willow timber was 467.9 g·kg^{-1} d.m., as compared to 483.2 g·kg^{-1} d.m. in birch wood, 421.1 g·kg^{-1} d.m. wheat straw and 637.8 g·kg^{-1} d.m. bituminous coal. The average content of carbon in the biomass of shrub willow cv. Wodtur (*Salix dasyclados* species) was 518.3 g·kg^{-1} d.m., and of cv. Sprint (*Salix viminalis* species) – 513.2 g·kg^{-1} d.m. (STOLARSKI et al. 2008). Regarding the above studies, the lowest content of carbon has been determined in biomass of Jerusalem artichoke (430.8 g·kg^{-1} d.m.).

The content of hydrogen in biomass of the analysed species of willow was on average 71.11 g·kg^{-1} d.m. (Table 3). Statistically, the highest significant content of hydrogen in biomass was determined for the species *Salix triandra* (74.98 g·kg^{-1} d.m.). The average content of hydrogen in shoots was significantly higher than that found in roots (71.78 vs 69.33 g·kg^{-1} d.m.). The highest concentration of hydrogen in biomass was found in shoots produced by *Salix triandra* (76.34 g kg^{-1} d.m.), while the lowest one occurred in roots of *Salix dasyclados* (66.40 g·kg^{-1} d.m.). The level of hydrogen in biomass of *Salix* spp. was significantly negatively correlated with the content of sulphur (r=-0.52, n=30). While analysing the relationships between

Table 3

Content of hydrogen in biomass of the five shrub willow species (g·kg^{-1} d.m.)

Species (a)	Type of biomass (b)		Average
	shoots	roots	
Salix viminalis	71.56	70.17	71.16
Salix dasyclados	67.10	66.40	66.90
Salix triandra	76.34	69.84	74.98
Salix purpurea	72.89	69.99	72.01
Salix alba	71.02	70.24	70.79
Average	71.78	69.33	71.11
$LSD_{0.05}$	a – 1.80	b – 1.10	a × b – 2.60

the analysed biometric features and the levels of the particular elements, it was only in one case that a strong negative correlation was established, namely between the diameter of shoots and content of hydrogen, where r=-0.56 was calculated for the mathematical model of y = -0.0433 x + 7.5584 (n=15). The content of hydrogen in one-year shoots of *Salix viminalis* in another experiment carried out by the same authors (STOLARSKI et al. 2005) was 68.7 $g \cdot kg^{-1}$ d.m. Some other tests showed that the concentration of hydrogen in biomass of *Salix* spp. was only slightly varied and ranged between 66.3 $g \cdot kg^{-1}$ d.m.for *Salix viminalis* to 66.8 $g \cdot kg^{-1}$ d.m. for *Salix dasyclados* (STOLARSKI et al. 2008). CUIPING et al. (2004) report that an average content of hydrogen in willow wood was 71.0 $g \cdot kg^{-1}$ d.m., compared to 83.6 $g \cdot kg^{-1}$ d.m. in birch wood, 65.3 $g \cdot kg^{-1}$ d.m.in wheat straw and 39.7 $g \cdot kg^{-1}$ d.m. in bituminous coal, where it was the lowest.

The content of sulphur in biomass of the five analysed species of shrub willow was on average 0.64 $g \cdot kg^{-1}$ d.m. (Table 4). The species called *Salix triandra* had the smallest, statistically confirmed, levels of sulphur in biomass (on average 0.43 $g \cdot kg^{-1}$ d.m.). The average content of sulphur in aerial shoots was significantly lower than in roots (0.52 versus 0.95 $g \cdot kg^{-1}$ d.m.).

Table 4

Content of sulphur in biomass of the five shrub willow species ($g \cdot kg^{-1}$ d.m.)

Species (a)	Type of biomass (b)		Average
	shoots	roots	
Salix viminalis	0.56	0.92	0.66
Salix dasyclados	0.56	1.06	0.70
Salix triandra	0.35	0.72	0.43
Salix purpurea	0.57	0.85	0.66
Salix alba	0.57	1.20	0.75
Average	0.52	0.95	0.64
$LSD_{0.05}$	a – 0.03	b – 0.02	a × b – 0.05

This tendency held true for all the analysed willow species. The lowest amounts of sulphur were determined in shoots of *Salix triandra* (0.35 $g \cdot kg^{-1}$ d.m.), whereas the highest ones appeared in roots of *Salix alba* (1.20 $g \cdot kg^{-1}$ d.m.). Some other experiments showed that the content of sulphur in one-year shoots of *Salix viminalis* was 0.43 $g \cdot kg^{-1}$ d.m. (STOLARSKI et al. 2005). KALEMBASA et al. (2005) observed that the total amount of sulphur in biomass of maiden grass (*Miscanthus* spp.) decreased as the grass continued to grow, with 1.67 g sulphur 1 kg^{-1} d.m. in 1st decade of June declining down to 0.50 g S kg^{-1} d.m. in 1st decade of September. The average content of sulphur for all the clones analysed in the above trial, irrespective of the date of sampling, was 0.91 g·kg^{-1} d.m. The amount of sulphur in the biomass of shrub willow *Salix dasyclados* was 0.44 g·kg^{-1} d.m., and in that of *Salix viminalis* – 0.40 g·kg^{-1} d.m. (STOLARSKI et al. 2008). By comparison, the content of sulphur in biomass of *Miscanthus* grasses was 0.48 g·kg^{-1} d.m. in *Miscanthus giganteus* and 0.58 g·kg^{-1} d.m. in *Miscanthus sacchariflorus.* The highest amount of sulphur determined in the above experiments was found in the biomass of prairie cordgrass (1.34 g·kg^{-1} d.m.). The content of sulphur in hardcoal is in general higher than in biomass of energy crops. According to ZAWISTOWSKI (2003), the content of sulphur in hardcoal is varied, ranging from 2.5 to 13.5 g·kg^{-1} d.m. When comparing the emission of sulphur during combustion of fuels, KUBICA (2001) suggested that that it was 20-fold higher for hardcoal than for timber.

CONCLUSIONS

1. The species *Salix purpurea* was characterised by the highest content of carbon in biomass (on average 501.32 g·kg^{-1} d.m.). The content of carbon in shoots produced by this species was significantly the highest among the analysed types of biomass. On the other hand, the lowest content of carbon was determined in roots of *Salix triandra*.

2. The content of hydrogen in biomass of shrub willow ranged within 66.90 (*S. dasyclados*) to 74.98 g·kg^{-1} d.m. (*S. triandra*).

3. Among the five tested species of shrub willow, the highest average content of sulphur was found in the biomass of *S. triandra*; the lowest – in biomass produced by *S. alba*.

4. Aerial shoots of all the tested species were characterised by higher concentrations of carbon, hydrogen and lower amounts of sulphur than the roots.

5. Relatively high amounts of carbon and hydrogen along with the low content of sulphur in the biomass of shrub willow mean that the plant can be treated as an energy source alternative to hardcoal.

6. The yield of dry biomass of the five tested species of shrub willow obtained in the first growing season was 3.95 (*Salix purpurea*) to 7.22 g·kg^{-1} d.m. (*Salix viminalis*).

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SODIUM AND POTASSIUM IN THE GROUNDWATER IN AREAS NEAR THE MAŚLICE MUNICIPAL REFUSE DUMP IN WROCŁAW*

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Abstract

Sodium and potassium are alkaline metals commonly occurring in natural environment. In clean groundwater their content usually does not exceed the value of 100 mg·dm^{-3} (Na) and 90 mg·dm^{-3} (K). Leakage waters of unsealed or improperly sealed refuse dumps constitute a potential grave source of water environment contamination. In the initial years of refuse dumping the leakage may contain up to 2,500 mg·dm^{-3} of sodium and up to 3,100 mg·dm^{-3} of potassium. The leakage from "old" dumps contains up to 3,700 mg·dm^{-3} sodium and up to 1,580 mg·dm^{-3} potassium.

The purpose of this paper was to demonstrate the character and the dynamics of the changes in sodium and potassium concentrations in the leakage waters of the Maślice Refuse Dump in Wrocław and in the groundwater in the adjacent areas. Since sand-gravel deposits lie in the base of the dump and the groundwater level is fairly high, contact occurs between the groundwater and the dumped waste as well as contamination transfer. Only part of the dump has a sealing and a drainage carrying the leakage to a tank. In the late 1990s the exploitation of the dump was terminated and rehabilitation started with a view to limiting the access of the water to the refuse dump.

The paper presented the results of the research covering the period from 1995 to 2007 on sodium and potassium contents in the groundwater coming to the dump, in the dump leakage and in the groundwater coming out of the dump.

The research has shown that both the concentrations and the proportions between sodium and potassium contents in the groundwater coming to the dump stayed within the natural ranges, exceeding them only in 2000-2001. The leakage, despite closing and rehabilitating the dump, was still characterized by fairly high and balanced concentrations of the

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analyzed elements. Also sodium and potassium contents in the groundwater coming out of the dump were high and balanced, which proves a continuous inflow of pollution through the unsealed base of the facility.

Key words: municipal refuse dump, groundwater, leakage waters, sodium, potassium.

SÓD I POTAS W WODACH PODZIEMNYCH NA TERENACH OTACZAJĄCYCH SKŁADOWISKO ODPADÓW KOMUNALNYCH „MAŚLICE" WE WROCŁAWIU

Abstrakt

Sód i potas są pospolicie występującymi w środowisku metalami alkalicznymi. W czystych wodach podziemnych ich zawartości najczęściej nie przekraczają 100 mg·dm^{-3} (Na) i 90 mg·dm^{-3} (K). Poważnym źródłem zanieczyszczenia środowiska wodnego mogą być odcieki z nieuszczelnionych lub nieprawidłowo uszczelnionych składowisk odpadów komunalnych. W początkowych latach składowania odpadów odcieki mogą zawierać do 2500 mg·dm^{-3} sodu i do 3100 mg·dm^{-3} potasu. Odcieki ze „starych" składowisk zawierają do 3700 mg·dm^{-3} sodu i do 1580 mg·dm^{-3} potasu.

Celem pracy było wykazanie charakteru i dynamiki zmian stężeń sodu i potasu w odciekach ze składowiska odpadów komunalnych „Maślice" we Wrocławiu oraz w wodach podziemnych na przyległych terenach. Ponieważ w podłożu składowiska zalegają utwory piaszczysto-żwirowe, a poziom lustra wód podziemnych jest dość wysoki, następuje kontakt wód podziemnych ze składowanymi odpadami oraz transport zanieczyszczeń. Tylko część składowiska ma uszczelnienie i drenaż odprowadzający odcieki do zbiornika. Na przełomie lat 1999-2000 zakończono eksploatację składowiska i rozpoczęto rekultywację, której głównym zadaniem było ograniczenie dostępu wody do złoża odpadów.

W pracy przedstawiono wyniki badań prowadzonych w latach 1995-2007 dotyczących zawartości sodu i potasu w wodach podziemnych dopływających do składowiska, odciekach składowiskowych oraz w wodach podziemnych wypływających za składowiskiem.

Wykazano, że zarówno stężenia, jak i proporcje między zawartościami sodu i potasu w wodach podziemnych dopływających do składowiska mieściły się w naturalnych zakresach, przekroczonych jedynie w latach 2000-2001. W odciekach, pomimo zamknięcia i rekultywacji składowiska, nadal stwierdzano dość wysokie i wyrównane stężenia analizowanych składników. Również zawartości sodu i potasu w wodach podziemnych odpływających za składowiskiem były wysokie i wyrównane, co świadczy o utrzymującym się dopływie zanieczyszczeń przez nieuszczelnione podłoże obiektu.

Słowa kluczowe: składowisko odpadów komunalnych, wody podziemne, odcieki składowiskowe, sód, potas.

INTRODUCTION

Sodium and potassium are alkaline metals commonly occurring in natural environment. In clean groundwater their content rarely exceeds the value of 100 mg·dm^{-3} (Na) and 90 mg·dm^{-3} (K). Na/K ratio in weakly mineralized water usually ranges from 0,2 to 0,9; the ratio increases along rising mineralization (usually up to 30-200) (Macioszczyk, Dobrzyński 2002).

Apart from natural sources, considerable amounts of sodium and potassium can reach water environment as a result of leakage from unsealed or improperly sealed refuse dumps. According to literature in the initial years of refuse dumping the leakage may contain up to 2,500 mg·dm^{-3} of sodium and up to 3,100 mg·dm^{-3} of potassium. The leakage from "old" dumps contains up to 3,700 mg·dm^{-3} of sodium and up to 1,580 mg·dm^{-3} of potassium (WILLIAMS 2002, ŻYGADŁO 2001, PAPADOPOULOU et al. 2007). During the examination of leakages occurring in the model deposits sodium concentrations ranged from 1,000 to 5,000 mg·dm^{-3}, while potassium concentrations were slightly lower (from 580 to 3,800 mg·dm^{-3}). Another factor which significantly affects the amount of sodium and potassium in dump leakages is the amount of water infiltrating the refuse dumps. With larger rainfall volume the concentrations diminish and the leakage volume grows, which gives rise to insignificant changeability of the charges of the removed impurities (KARTHIKEYAN et al. 2007, THORNTON et al. 2005).

The purpose of this paper was to demonstrate the character and the dynamics of the changes in sodium and potassium concentrations in the leakage waters of the Maślice Refuse Dump in Wrocław and in the groundwater in the adjacent areas.

MATERIAL AND METHODS

The research of the groundwater and the leakages was conducted in the area surrounding a closed municipal waste dump called Maślice in Wrocław. In the late 1960s neutralization process of the waste was begun, with an aid of excavation (covering a surface area of about 7ha) remaining after the exploitation of sand deposits. Sand-gravel deposits lie in the base of the dump and the groundwater level is fairly high (usually *ca.* 1-2 m above the foot of the dump). Under these conditions, both contact between the groundwater and the dumped waste as well as transfer of impurities are commonplace. The groundwater flows from south-west to north-east towards the Odra River (SZYMAŃSKA-PULIKOWSKA 2001). Only part of the dump (*ca.* 2 ha, built in 1994) has a sealing and a drainage carrying leakage to a tank, from which the research samples were taken. At the end of 1990s the exploitation of the dump was terminated and rehabilitation started. It involved reinforcing the slopes of the waste dump, sealing the dome with synthetic mineral material as well as, on the side of the groundwater inflow (in 2002), fixing a screen down to the impermeable layer to prevent the inflow. In 2004 the leakage tanks were filled in (SZYMAŃSKA-PULIKOWSKA 2005).

The paper presents the results of the research on sodium and potassium contents in the groundwater coming to the dump, in the dump leakage and in the groundwater coming out of the dump. The research into groundwater content was conducted from 1995 to 2007, whereas the examination of dump leakages was terminated the moment the tank had been filled in. Research material was taken 3 times a year. The analyzed dump leakages were from the derived from the tank and the groundwater originated from four piezometers located on the inflow (2) and the outflow (2) side of the dump. The water stagnant in the piezometer well was pumped out twice prior to taking samples of the groundwater. The contents of sodium and potassium in the taken samples were marked according to the methodology described in the literature (NAMIEŚNIK, JAMRÓGIEWICZ 1998, HERMANOWICZ et al. 1999). The statistical analysis of the research results was conducted by means of Statistica 7.1 programme.

RESULTS AND DISCUSSION

Table 1 depicts the characteristic values (mean, standard deviation, variance coefficient) and mean sodium-potassium concentration ratio in the consecutive years of the research into the groundwater flowing to the dump. Mean annual sodium contents ranged from 11.43 to 303.7 mg·dm^{-3} and these of potassium from 1.735 to 30.57 mg·dm^{-3} (ten times less). The highest concentrations were observed at the onset of the research period (1995--1996) and in the years 2000-2001. Only in this last period the contents of sodium and potassium exceeded the natural concentrations range (MACIOSZCZYK, DOBRZYŃSKI 2002). However, the results obtained for potassium content displayed a greater variability. The proportions between sodium and potassium concentrations fluctuated within the range between 2.524 and 14.04; reaching the highest values in 2001 (high sodium concentrations) and 2003 (distinctive decrease in potassium content).

Table 2 depicts the characteristic values (mean, standard deviation, variance coefficient) and mean sodium-potassium concentration ratio in the consecutive years of the research into dump leakages (1995-2003, excluding 1998). Mean annual sodium contents in the leakages ranged from 1,013 to 3,711 mg·dm^{-3}, reaching higher values after the dump was closed (since 2000). Mean annual sodium concentrations ranged from 905.8 to 2,350 mg·dm^{-3}. Na/K coefficient values fluctuated between 0.749 and 2.372. The results were characterized by a very significant variability, which diminished in the last years of the research (2002-2003). That is also when sodium and potassium concentrations became balanced; however, they still remained at a high level.

Research conducted by other authors also show high and balanced contents of the examined elements in leakages from refuse dumps, or their parts, exploited for a long time. The dumps in Pomorze Środkowe emitted leakages containing 273.7-1,708 mg Na·dm^{-3} and 268.3-1,143 mg K·dm^{-3}

Table 1

Characteristic values and mean sodium to potassium ratios
in groundwaters coming to the dump

Years		μ ($mg \cdot dm^{-3}$)	Na/K	σ	ν (%)
1995	Na	63.33	5.099	39.60	52.86
	K	12.42		6.563	62.53
1996	Na	43.85	2.702	22.99	47.14
	K	16.23		7.653	52.43
1997	Na	19.18	2.524	10.25	46.14
	K	7.600		3.507	53.41
1998	Na	29.04	3.596	7.877	19.35
	K	8.075		1.563	27.13
1999	Na	53.15	7.935	45.09	58.16
	K	6.698		3.895	84.84
2000	Na	303.7	9.935	297.2	44.18
	K	30.57		13.51	97.86
2001	Na	179.6	13.23	184.1	37.50
	K	13.58		5.093	102.5
2002	Na	46.68	7.802	34.46	45.87
	K	5.983		2.745	73.82
2003	Na	47.51	14.04	37.53	41.37
	K	3.383		1.400	79.01
2004	Na	17.22	4.314	11.14	68.72
	K	3.992		2.743	64.68
2005	Na	11.43	6.588	6.586	49.13
	K	1.735		0.852	57.64
2006	Na	23.83	3.733	5.856	38.31
	K	6.383		2.446	24.57
2007	Na	44.25	7.416	43.45	56.35
	K	5.967		3.362	98.20

Explanations: μ – mean, σ – standard deviation, ν – variance coefficient

(Sianów) as well as 619.3-2,460 mg $Na \cdot dm^{-3}$ and 710.8-2,048 mg $K \cdot dm^{-3}$ (Karlino) (JANOWSKA, SZYMAŃSKI 1999). Leakages from older sections of the refuse dump in Sierakowo (near Szczecin) contained 570-1,590 mg $Na \cdot dm^{-3}$ and 542-1,494 mg $K \cdot dm^{-3}$, whereas the newest section emitted leakages containing 103-638 mg $Na \cdot dm^{-3}$ and 85-516 mg $K \cdot dm^{-3}$ (MELLER et al. 2001).

Table 2

Characteristic values and mean sodium to potassium ratios in dump run-offs

Years		μ (mg·dm^{-3})	Na/K	σ	ν (%)
1995	Na	1013	1.118	273.4	146.7
	K	905.8		423.5	169.2
1996	Na	1615	0.749	156.7	121.2
	K	2157		216.5	118.6
1997	Na	1243	1.103	784.7	264.5
	K	1127		1008	408.9
1999	Na	1604	1.201	2518	2.593
	K	1335		986.3	29.64
2000	Na	3711	1.579	1484	65.52
	K	2350		672.7	152.2
2001	Na	2700	2.372	2174	315.2
	K	1138		636.9	81.21
2002	Na	1780	1.037	321.1	82.74
	K	1717		350.3	66.07
2003	Na	2461	1.162	439.5	73.75
	K	2118		172.0	85.44

Explanations, see Table 1

Thus the concentrations were approximate to the values demonstrated by the authors quoted previously (MOR et al. 2006, THORNTON et al. 2005, WILLIAMS 2002).

Table 3 depicts the characteristic values (mean, standard deviation, variance coefficient) and mean sodium-potassium concentration ratio in the consecutive years of the research into groundwater flowing out behind the dump. The examined waters contained on average from 481.0 to 4,084 mg Na·dm^{-3} and from 209.2 to 2,520 mg K·dm^{-3} per year. As in the case of the leakages, the highest sodium and potassium concentrations occurred after closing the dump (2000-2003). The proportions between mean annual sodium and potassium contents ranged between 1.163 and 2.567.

Such large amounts of the examined elements and the values of Na/K coefficient prove a considerable influence of the leakages on the composition of groundwater flowing out behind the dump. Since mean annual concentrations of sodium and potassium tended to be even higher in the discussed leakages, it is possible to suppose that the impurities getting into the ground from the old unsealed part of the dump contained even more of the examined elements. The research results of the waters flowing to the dump and the waters flowing out behind it were characterized by a similar variability.

Table 3

Characteristic values and mean sodium to potassium ratios
in waters flowing from the dump

Years		μ ($mg \cdot dm^{-3}$)	Na/K	σ	ν (%)
1995	Na	1049	1.163	729.1	69.52
	K	901.9		815.8	90.45
1996	Na	1786	1.634	532.5	29.82
	K	1093		686.8	62.84
1997	Na	1486	1.550	945.2	63.61
	K	958.9		894.0	93.23
1998	Na	2443	1.851	227.4	9.310
	K	1320		1008	76.38
1999	Na	2436	2.085	1198	49.17
	K	1168		905.3	77.48
2000	Na	3853	1.529	2262	58.71
	K	2520		2917	115.8
2001	Na	3983	2.567	1184	29.72
	K	1552		1218	78.46
2002	Na	3203	1.553	900.5	28.11
	K	2063		346.0	16.77
2003	Na	4084	2.015	1404	34.39
	K	2027		733.8	36.21
2004	Na	1400	2.188	1175	83.92
	K	639.8		550.9	86.10
2005	Na	481.0	2.299	355.7	73.95
	K	209.2		193.0	92.25
2006	Na	1790	1.687	1155	64.51
	K	1061		825.4	77.78
2007	Na	793.6	2.212	576.3	72.61
	K	358.7		296.3	82.61

Explanations, see Table 1

During research conducted on the dump in Radiowo (PACHUTA, KODA 2001) the groundwater polluted by leakages from the unsealed facility contained similarly high sodium concentrations (up to 4,730 $mg \cdot dm^{-3}$) but much lower potassium concentrations (up to 35 $mg \cdot dm^{-3}$). The groundwaters in the vicinity of other dumps (Mor et al. 2006, Srivastava, Ramanathan 2008) were also distinguished by an increased sodium content (up to 800 mg $Na \cdot dm^{-3}$) and a relatively low content of potassium (to 60 mg $K \cdot dm^{-3}$).

CONCLUSIONS

1. The concentrations of sodium and potassium in the groundwater coming to the Maślice Refuse Dump stayed within the natural ranges (except the years 2000-2001).

2. Despite closing and rehabilitating the dump, the leakage still contained fairly high and balanced amounts of sodium and potassium, implying that the dump will continue to contaminate the environment for a long time in the future.

3. High and balanced sodium and potassium contents in the groundwater leaving the dump prove a continuous inflow of impurities to the base of the dump.

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COMPARATIVE ANALYSIS OF FLUORIDE CONTENT IN SHEEP MANDIBLES FROM ARCHEOLOGICAL EXCAVATIONS IN SZCZECIN ACCORDING TO INDIVIDUAL AGE AND TIME OF BEING DEPOSITED IN SOIL

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Abstract

Bones can be a very good marker of environmental contamination by fluoride. Bones in a living organism have a different composition than in a dead one. As a result of adsorption from soil, bones from archeological excavations usually have more fluoride than those in a living body, and a significant portion of the fluorides they contain are acquired after death.

This paper presents the results of a studies on fluoride content of sheep mandibles from archeological excavation sites in Szczecin. An attempt was undertaken to define how the chronological age of the bones and the time they had been lying in soil affected the bones.

The material consisted of sheep mandibles from several excavations sites: Szczecin Mścięcino, Szczecin Rynek Warzywny, and Szczecin Zamek Książąt Pomorskich (Szczecin Castle of Pomeranian Dukes). Cultural layers in these excavations were mostly formed from humus and humus with sand and clay. The fluoride content was determined by an ion-selective electrode with the pH/mV Orion 920A.

Fluoride content was determined in 270 mandibles, which were classified into sheep age categories and according to the archeological age of the bones. The individual and archeological age of the sheep bones was determined by archeologists during the initial tests of the bones. A comparative evaluation of the significance of differences in the average

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fluoride content in the bones was performed by means of a single factor analysis of the orthogonal variance. The least significant differences were estimated by Tukey's test.

The results show that the fluoride content depends on the individual age of animals and the chronological age of bones. The fluoride content of the sheep mandibles increased along with the individual age of the animals. Moreover, chronologically younger bones contained significantly less fluoride than older ones. In the sheep mandibles which lay longer in soil, the fluoride content tended to increase with the chronological age, while in chronologically youngest bones the tendency was reverse.

Key words: fluoride, fossil bones, mandibles, sheep.

ANALIZA PORÓWNAWCZA ZAWARTOŚCI FLUORKÓW W ŻUCHWACH OWIEC Z WYKOPALISK ARCHEOLOGICZNYCH MIASTA SZCZECINA W ZALEŻNOŚCI OD WIEKU OSOBNICZEGO I CZASU ZALEGANIA W GLEBIE

Abstrakt

Kości mogą być bardzo dobrymi biomarkerami skażenia środowiska fluorkiem. Inny jest skład kości w organizmie żywym, a inny w martwym. W wyniku adsorpcji z gleby kości archeologiczne zazwyczaj zawierają więcej fluorków niż świeże, a znaczącą część fluorków stanowią te wbudowane po śmierci.

W pracy przedstawiono wyniki badań zawartości fluorków w żuchwach owiec z wykopalisk archeologicznych miasta Szczecina. Podjęto próbę określenia, w jaki sposób wiek osobniczy oraz czas zalegania kości w glebie wpływa na zmianę stężenia fluorków w żuchwach owiec.

Materiał do badań stanowiły żuchwy owiec z terenów, w których prowadzono wykopaliska archeologiczne: Szczecin Mścięcino, Szczecin Rynek Warzywny i Szczecin Zamek Książąt Pomorskich. Warstwy kulturowe w tych wykopaliskach tworzyły głównie próchnica oraz próchnica z domieszką piasku i gliny. Zawartość fluorków w kościach oznaczano potencjometrycznie z zastosowaniem elektrody jonoselektywnej z użyciem pH/jonometru firmy Orion 920A. Przebadano zawartość fluorków łącznie w 270 żuchwach, które posegregowano według utworzonych kategorii wieku osobniczego owiec i wieku archeologicznego kości. Wiek osobniczy oraz archeologiczny kości owiec został ustalony przez archeologów podczas wstępnych badań kości. Oceny porównawczej istotności różnic między średnimi zawartościami fluorków w kościach dokonano za pomocą jednoczynnikowej analizy wariancji ortogonalnej. Najmniejsze istotne różnice obliczono testem Tukeya.

Stwierdzono, że zawartość fluorków w żuchwach owiec była zależna zarówno od wieku osobniczego zwierząt, jak i wieku archeologicznego kości. Zawartość fluorków w żuchwach owiec zwiększała się wraz z wiekiem osobniczym zwierząt. Ponadto kości młodsze pod względem wieku archeologicznego zawierały istotnie mniej fluorków niż kości starsze. W żuchwach owiec zalegających dłużej w glebie stwierdzono także tenedencję wzrostu zawartości fluorków wraz z wiekiem osobniczym zwierząt, natomiast w kościach najmłodszych pod względem archeologicznym tendencja ta była odwrotna.

Słowa kluczowe: fluorki, kości archeologiczne, żuchwy, owce.

INTRODUCTION

Fluoride content in living organisms has a significant meaning for evaluation of the environmental contamination by this element. Bones, nails, antlers, hair, teeth, snails' shells or birds' egg shells as well as urine and blood are used as biomarkers of the fluoride environmental contamination (Mandat et al. 1990, Gutowska et al. 2002, Piotrowska et al. 2006). Friedrich (2002) claims that up to 90% of the retained fluorides in an organism is deposited in bones, teeth, nails and hair.

Fluorine is deposited in bones as hydroxyapatite. The hydroxyl ion is substituted by the F^- ion forming fluoroapatite, in which the distance between the calcium and fluoride ions is smaller. Fluoroapatite crystals are bigger, more stable, more resistant to dissolving and less subject to rebuilding (Grynpas 1990). According to Machoy (1987) the fluoride ion can substitute the hydroxyl ion in bones even up to 5.7%, and according to Dąbkowska et al. (1995) – in a range of several per cent.

The composition of bones in a living organism is different from that of bones in a dead animal. When a bone is buried in soil, the physiological equilibrium which was earlier established between this tissue and the surroundings is disturbed. In soil there follows a dynamic interaction between bones and the geochemical forces. The influence on the diagenetic bone tissue have: the pH of soil, microorganisms, structure of soil, mineralogy, and the organic content, so called external factors. The internal factors are: density, volume, microstructure and bone biochemistry (Szostek, Bałuszyńska 1997).

The reason of the fluoride accumulating in bones, mainly, after the death, is the influence of surrounding remains of the soils mainly (Noceń et al. 1994). As a result of the adsorption from soil, archaeological bones contain more fluoride than the living ones, and a significant part of fluorides constitute those which were sorption after the death (Chlubek et al. 1996). Posthumously, in the soil environment, bone enrichment in the fluorides is a very slow process. That is why a significant increase of the fluoride content in the archaeological bones remains is evident only in the bones which lay in soil tens of years or even several centuries (Noceń 1997).

This paper concerns an evaluation of the fluoride content in sheep mandibles from the archeological excavations of Szczecin, as well as an attempt of defining a dependence of the mandible fluoride content in the individual sheep age and the chronological age.

MATERIALS AND METHODS

The material for the study consisted of sheep mandibles from archaeological excavations in Szczecin. The bones for the analysis were rendered by the Chair of Animal Anatomy of University of Agriculture in Szczecin. The animal remains come from three archaeological excavation sites: Szczecin Mścięcino, Szczecin Rynek Warzywny, and Szczecin Zamek Książąt Pomorskich (Szczecin Castle of Pomeranian Dukes, latter called the Castle).

The material from Mścięcino site was excavated in 1954 while unearthing the remnants of an early Middle Age settlement and later, in 1968, during archaeological excavations of an early Middle Age castle. The sheep were probably kept for wool and hide rather than meat (KUBASIEWICZ 1955, STĘPIEŃ 1994).

The excavations from the Castle come from the sites set in different places. The material was excavated in the castle yard and basement in the 1970s. The sheep were a cross of the bigger copper sheep with the smaller peat sheep (KUBASIEWICZ 1960).

The last site – Szczecin Rynek Warzywny – was located close to the Castle, between Castle and the Odra River. The excavations were conducted between 1954 and 1965. The excavated bones were mainly mandibles of the ancient copper sheep, with very few bones of the peat sheep (KUBASIEWICZ 1957, KUBASIEWICZ, GAWLIKOWSKI 1967).

The individual and chronological age of the sheep bones was established by the archaeologists during initial tests of the bones.

Cultural layers in these excavations were mostly formed from humus and humus with sand and clay (GARCZYŃSKI 1955).

Fluoride content in 270 bones was determined (Table 1). The mandibles were classified into categories embracing individual sheep age and chronological age of bones. These categories and numbers of mandibles in each category are given in Tables 2 and 3. Adult individuals are those of above 24 months of age.

Table 1

Specification of the analyzed bones

Site of excavations	Number of mandibles	Chronological age (century)
Szczecin Mścięcino	26	9th - 11th
Szczecin Zamek	123	2nd half 7th - 20th
Szczecin Rynek Warzywny	121	2nd half 9th - 1st half 13th
Total	270	2nd half 7th - 20th

Table 2

Categories of individual sheep age and number
of mandibles in categories

Categories of individual sheep age		Number of bones
1.	<6 months	17
2.	6-12 months	80
3.	12-18 months	36
4.	18-24 months	27
5.	> 24 months	108

Table 3

Categories of chronological bones age and number
of mandibles in categories

Categories of chronological bones		Number of bones
I	2nd half 7th - early 9th	10
II	9th - 11th	159
III	late 11th - 2nd half 12th	41
IV	late 12th - 14th	42
V	early 16th - 18th	10
VI	19th - 20th	7

The fluoride content in the bones was determined by potentiometry using a pH/mV Orion 920A with an ion-selective fluoride electrode, according to Durda et al. (1986).

All analyses were performed in three replications. The comparative evaluation of significance differences in mean fluoride contents in bones was executed by ANOVA. The LSD values were estimated by Tukey's test. In order of determine how the individual age and the time of lying in soil influenced the fluoride content in mandibles, there was a 3D surface matched for the experiment data, by means of the method of the smallest squares with the distance weighing. It depicts the tendency of changes in fluoride content according both to individual age and chronological age of bones.

RESULTS AND DISCUSSION

The results of the experiment showed that the fluoride content in the sheep mandibles depended on both the individual sheep age and the chronological age of the bones.

According to the individual age, the mean fluoride content in the mandibles was the lowest in the bones of sheep age 6-12 months (category 2), where it was 123.4 mg $F^{-} \cdot kg^{-1}$ (Figure 1a). The bones of the youngest individuals contained 128.0 mg $F^{-} \cdot kg^{-1}$. As SZOSTEK, BAŁUSZYŃSKA (1997) claim that a higher fluoride content in mandibles of the youngest sheep, compared with sheep 6-12 months of age, can be caused by higher mineralization of bones in younger animals and easier fluoride sorption from soil by bones of young dead individuals. From the group of bones of the young individuals

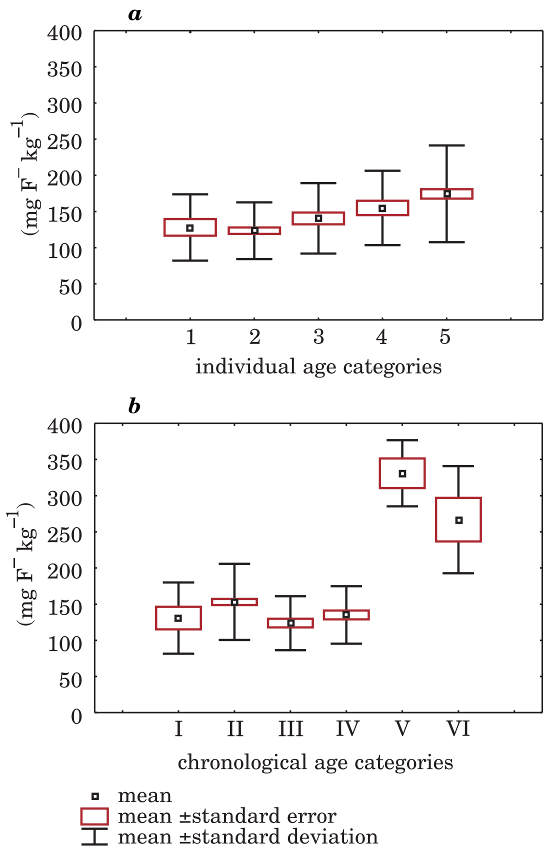

Fig. 1. The fluoride content in sheep mandibles from Szczecin excavations according to individual age of sheep (*a*) and chronological age of bones (*b*)

(6-12 months of age) on, the fluoride content tends to increase as the age of sheep goes up. A highly significant statistical difference in the fluoride content was observed between the categories 2 and 5 (p < 0.001 – Table 4). ALSO NOCEŃ (1997) claimed that along with the individual age increase, the accumulation of fluorides increased in archeological bones.

According to the chronological age of bones, an average fluoride content in archeological bones was found in mandibles from the late 11^{th}-2^{nd} half 12^{th} century (category III) and in mandibles lying in soil longest, from the 2^{nd} half of the 7^{th}- early 9^{th} century (category I), where it reached123.7 mg $F^{-}\cdot kg^{-1}$ and 130.8 mg $F^{-}\cdot kg^{-1}$, respectively (Figure 1b). The highest F^{-} content appeared in bones from the early 16^{th} to the 18^{th} century (category V) – 331.0 mg $F^{-}\cdot kg^{-1}$, and in bones from the 19^{th}- 20^{th} century (category VI) – 226.8 mg $F^{-}\cdot kg^{-1}$. The bones which lay in soil the shortest time showed the highest content of fluoride. This can be attributed to the surface layers of the excavation soil containing more fluoride than the deeper ones. The

Table 4

Statistical analysis of differences between the fluoride content in sheep mandibles from Szczecin excavations

According to the individual sheep age		
Individual age categories	($mg\ F^{-}\cdot kg^{-1}$)	
	mean	standard deviation
1.	128.0	45.92
2.	123.4	39.33
3.	140.4	48.71
4.	154.9	51.42
5.	174.3	66.89
Statistical analysis of difference	2-5***	
According to the chronological bones age		
Chronological age categories	($mg\ F^{-}\cdot kg^{-1}$)	
	mean	standard deviation
I	130.8	49.31
II	153.0	52.56
III	123.7	37.42
IV	135.0	39.78
V	331.0	45.78
VI	266.8	74.03
Statistical analysis of difference	I-V***, I-VI***, II-V***, II-VI***, III-V***, III-VI***, IV-V***, IV-VI***	

same conditions are observed in areas subjected to fluorine emissions (e.g. the Police Chemical Plant near Szczecin, which emits large amounts of fluorine). The excavation layers from which the bones used for our examinations were excavated contained large amounts of humus, which retained fluoride (MEINHARDT 1994, GAŁĄZKA 1996). The statistical analysis showed highly significant differences between the bones of the two youngest groups, and the other categories of the chronological age of bones (Table 4).

The surface diagram of the fluoride content according to the individual age of sheep and the chronological age of bones can suggest that in the bones from the period of up to 2nd half of the 12th century (I, II and III categories of chronological age) the fluoride content increased only slightly along with the individual sheep age, while in the bones which lay in soil a shorter time a reverse tendency was observed – the fluoride content in mandibles decreased along with the individual sheep age (Figure 2). It was also ascertained that in all the categories of individual age of sheep, the fluoride content in mandibles increased as the time they had been deposited in soil was shorter, a tendency which was most evident in the youngest bones (category 1 and 2).

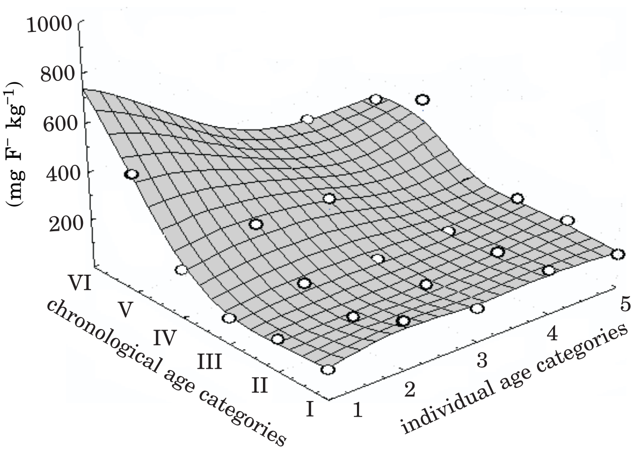

Fig. 2. Tendency of the fluoride content in sheep mandibles from Szczecin excavations according to individual sheep age and chronological bones age

CONCLUSIONS

1. The fluoride content in the sheep mandibles from the archeological excavations in Szczecin increased along with the individual age of animals.

2. As regards the chronological age, the younger bones contained significantly more fluorides than the older ones.

3. In the sheep mandibles which lay in soil longer, an increasing tendency in the fluoride content along with the individual age of animals occurred, while in the youngest chronological bones a contrary tendency was noticed.

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Review paper

THE IMPACT OF NICKEL ON HUMAN HEALTH

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Abstract

In 2008, nickel received the shameful name of the "Allergen of the Year." According to dermatologists the frequency of nickel allergies is still growing, and it cannot be explained only by fashionable piercing and nickel devices used in medicine (like coronary stents).

Occupational exposure of several million workers worldwide has been shown to give rise to elevated levels of nickel in blood, urine and body tissues. In these cases, workers are exposed to airborne fumes and dusts containing nickel and its compounds and therefore inhalation is the main route of uptake. Nonoccupational sources of nickel exposure for the general population include mainly drinking water and food. Recently, tests of kitchen kettles showed substantial leaching of nickel into drinking water when boiled in kettles with exposed nickel-plated elements.

Three types of adverse health impacts as a result of exposure to nickel are discussed in the text. Acute health effects generally result from short-term exposure to high concentrations of pollutants. Chronic noncancer health effects may result from long-term exposure to relatively low concentrations of pollutants. Inhalation of nickel also can cause cancer of the lungs, nose and sinuses. Cancers of the throat and stomach have also been attributed to inhalation of nickel. However, the exact mechanism by which nickel causes cancer is still questionable and needs further investigation. The most popular hypotheses to explain this phenomenon are presented in the text.

Key words: nickel, sources of nickel, exposure, toxicity, carcinogenesis, human.

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WPŁYW NIKLU NA ZDROWIE CZŁOWIEKA

Abstrakt

W 2008 nikiel zyskał niechlubne miano „alergenu roku". Według dermatologów, wciąż rosnąca częstość alergii na nikiel nie może być tłumaczona jedynie modą na przekłuwanie ciała lub zwiększone zastosowanie w medycynie urządzeń zawierających nikiel (np. stenty naczyń wieńcowych). Wykazano, że kilka milionów robotników na całym świecie narażonych zawodowo na nikiel i jego związki, ma podwyższony poziom tego pierwiastka we krwi, moczu i tkankach organizmu. W tych przypadkach pracownicy są wystawieni na opary i pyły zawierające nikiel, a zatem głównym sposobem pobierania tego pierwiastka jest inhalacja. Dla większości populacji źródła niklu stanowią jednak woda pitna i żywność. Niedawno w testach czajników kuchennych z odsłoniętymi elementami grzewczymi, wykonanymi ze stopów niklu wykazano uwalnianie znacznych ilości niklu do wody pitnej.

W pracy omówiono trzy główne typy szkodliwego dla zdrowia wpływu ekspozycji na nikiel. Ostre zatrucia, jako wynik krótkoterminowej ekspozycji na wysokie stężenia polutantów, chroniczne nienowotworowe efekty powstające na skutek długotrwałej ekspozycji na stosunkowo niskie dawki oraz nowotwory płuc, nosa i zatok spowodowane inhalacją powietrza zanieczyszczonego niklem. Również rak gardła i żołądka przypisuje się wdychaniu związków niklu. Dokładny mechanizm kancerogenezy wywołanej przez nikiel pozostaje niejasny i wymaga dalszych badań. W pracy omówiono najbardziej popularne hipotezy wyjaśniające to zjawisko.

Słowa kluczowe: nikiel, źródła niklu, ekspozycja, toksyczność, kancerogenność, człowiek.

INTRODUCTION

Nickel is the 28th element in the periodic table. It is a silver-white metal found in several oxidation states (ranging from -1 to +4), however, the +2 oxidation state [Ni(II)] is the most common in biological systems (DENKHAUS, SALNIKOW 2002). Nickel easily forms nickel-containing alloys, which have found an ever increasing use in modern technologies for over a hundred years now. Global input of nickel to the human environment is approximately 150 000 and 180 000 metric tonnes per year from natural and anthropogenic sources, respectively, including emissions from fossil fuel consumption, and the industrial production, use, and disposal of nickel compounds and alloys (KASPRZAK et al. 2003).

SOURCES OF NICKEL

Nickel is widely distributed in the environment, and can be found in air, water, and soil. Natural sources of atmospheric nickel include dusts from volcanic emissions and the weathering of rocks and soils. The level of nickel in ambient air is small (about 6-20 $ng \cdot m^{-3}$), but levels up to

150 ng $Ni \cdot m^{-3}$ could be present in air contaminated by anthropogenic sources. In water, nickel derives from biological cycles and solubilization of nickel compounds from soils, as well as from the sedimentation of nickel from the atmosphere. Uncontaminated water usually contain about 300 ng $Ni \cdot dm^{-3}$. Farm soils contain approximately 3-1000 mg $Ni \cdot kg^{-1}$ soil, but the Ni concentration can reach up to 24 000-53 000 $mg \cdot kg^{-1}$ Ni in soil near metal refineries and in dried sludge, respectively. At pH<6.5, nickel compounds in soil are relatively soluble, whereas at pH>6.7, most nickel exists as insoluble hydroxides. Nickel salts of strong acids (chloride, nitrate, and sulfate) and organic acids are soluble in water whereas nickel salts of weak inorganic acids, as well as metallic nickel, nickel sulfides, and nickel oxides are poorly water-soluble (BARCELOUX 1999, DENKHAUS, SALNIKOW 2002, SUTHERLAND, COSTA 2002).

The majority of nickel production is used for the creation of stainless steel, nickel alloys, and nickel cast iron that comprise objects, such as coins, electrical equipment, tools, machinery, armaments, jewelry, and household utensils. Nickel compounds are used also for electroplating, electroforming, nickel-cadmium alkaline batteries, dye mordant, catalysts, and electronic equipment. Nickel containing alloys include nickel plating of nonprecious metals, surgical steel (0.5-30% Ni), white gold (10-15% Ni), German silver (10-15% Ni), solders, hard-gold plating, and sterling silver. As the result of increasing consumption of nickel-containing products nickel compounds are released to the environment at all stages of production and utilization and may constitute a hazardous factor to human health (BARCELOUX 1999, DENKHAUS, SALNIKOW 2002).

NICKEL EXPOSURE

Exposures by inhalation, ingestion or skin contact occur in nickel and nickel alloy production plants as well as in welding, electroplating, grinding and cutting operations. Airborne nickel levels in excess of 1 $mg \cdot m^{-3}$ have been found in nickel refining, in the production of nickel alloys and nickel salts, and in grinding and cutting of stainless-steel. Although in these industries, modern control technologies have markedly reduced exposures in recent years, several million workers worldwide are exposed to airborne nickel and its compounds (KASPRZAK et al. 2003, SEILKOP, OLLER 2003).

Occupational exposure has been shown to give rise to elevated levels of nickel in blood, urine and body tissues, with inhalation as the main route of uptake. Nonoccupational sources of nickel exposure include food, air and water, but the levels found are usually several orders of magnitude lower than those typically found in occupational situations.

Drinking water and food are the main sources of exposure for the general population. The maximum concentration of total nickel allowed by law in Poland in drinking water is 20 $\mu g \cdot dm^{-3}$ (Dz.U.07.61.417). Average concentrations of total nickel in drinking water ranges from 3-7 $\mu g \cdot dm^{-3}$, but it increases in vessels that contain corroded nickel plating. The first drawn water from hot water taps plated with nickel may contain concentrations of 1-1.3 mg $Ni \cdot dm^{-3}$ (BARCELOUX 1999). Moreover, some domestic appliances contain nickel alloys. Tests of kitchen kettles showed substantial leaching of nickel into water when boiled in kettles with exposed nickel-plated elements. The concentration of nickel in boiled water increased from 4.55 and 0.8 $\mu g \cdot dm^{-3}$ (unboiled hard and soft water) up to 244 and 51 $\mu g \cdot dm^{-3}$, respectively. When water was boiled in concealed element kettle, the concentrations of nickel were 5.23 and 1.9 $\mu g \cdot dm^{-3}$, respectively (COT 2003). Most food contains below 0.5 mg $Ni \cdot kg^{-1}$ wet weight. Foodstuffs with high nickel contents are cocoa (up to 8.2-12 $mg \cdot kg^{-1}$ fresh wet weight), oatmeal, spinach, dry legumes, hazelnuts, dark chocolate, soya beans, and soya products (SUTHERLAND, COSTA 2002).

In vitro studies suggest that stainless steel endoprosthesis (14-36% nickel) may release nickel; however, the clinical significance of this amount of nickel remains to be determined. Also administration of nickel-contaminated medications (e.g., albumin, radiocontrast media, hemodialysis fluids) leads to significant exposures (MCGREGOR et al. 2000, KASPRZAK et al. 2003). The amount of nickel absorbed during medical procedures depends on the composition of the equipment in contact with the blood and body fluids. For example, the estimated uptake of nickel during a typical dialysis procedure in the 1980s was 100 mg Ni per treatment (SUNDERMAN 1983).

NICKEL EFFECT ON HEALTH

Human exposure to highly nickel-polluted environments causes a variety of pathologic effects. The toxic effects of nickel on the lung were recognized first by Agricola in the 16th century. Some fatal cases were noted following exposure to nickel carbonyl, and by the early 1930s, nickel was a recognized cause of contact dermatitis. Elevated incidences of lung and nasal cancer in workers exposed to nickel were also observed (SUNDERMAN et al. 1988, SEILKOP, OLLER 2003). In 2008, nickel received the shameful name of "Allergen of the Year" (GILLETTE 2008). According to the dermatologist the frequency of nickel allergy is still growing, and it can't be explained only by fashionable piercing and nickel devices used in medicine (like coronary stents and endoprostheses). All those observations caused that the interest in the nickel impact on human health increased (SIVULKA 2005).

Like many environmental agents, the toxic effect of nickel is related to the way it gets into an organism. Nickel can enter body *via* inhalation, ingestion and dermal absorption, but the route by which nickel enters cells is determined by its chemical form. For example, fat soluble nickel carbonyl can cross cell membranes by diffusion or through calcium channels (CANGUL et al. 2002), while insoluble nickel particles enter the vertebrate cells by phagocytosis (HECK, COSTA 1982).

The main transport protein of nickel in blood is albumin, but nickel can bind also to histidine and α2-macroglobulin (GLENNON, SARKAR 1982, KASPRZAK et al. 2003), and in this form is distributed throughout the tissues. A number of nickel-binding proteins including α1-antitrypsin, α1-lipoprotein and prealbumin were also described (NIELSEN et al. 1994). The highest nickel concentrations are found in the bone, lung, kidney, liver, brain and endocrine glands. Nickel is also found in breast milk, saliva, nails and hair. Transplacental transfer of nickel has been demonstrated in rodents. Nickel does not accumulate in the body; it is excreted in the urine, feces, bile and sweat (VALKO et al. 2005).

Contact with nickel compounds can cause a variety of adverse effects on human health, such as nickel allergy in the form of contact dermatitis, lung fibrosis, cardiovascular and kidney diseases and cancer of the respiratory tract (OLLER et al. 1997, MCGREGOR et al. 2000, SEILKOP, OLLER 2003). Chronic noncancer health effects may result from long-term exposure to relatively low concentrations of pollutants. Acute health effects generally result from short-term exposure to high concentrations of pollutants and they manifest as a variety of clinical symptoms (nausea, vomiting, abdominal discomfort, diarrhea, visual disturbance, headache, giddiness, and cough).

The most common type of reaction to nickel exposure is a skin rash at the site of contact. Skin contact with metallic or soluble nickel compounds can produce allergic dermatitis. This health problem caused by exposure to nickel affects people both at and away from work. Data indicate that women have greater risk for dermatitis, possibly due to a more frequent contact with nickel-containing items: jewelry, buttons, watches, zippers, coins, certain shampoos and detergents, pigments etc. (VAHTER et al. 2002, SZCZEPANIAK, PROKOP 2004). About 10% of women and 2% of men in the population are highly sensitive to nickel. Sensitization to the metal is generally caused by direct and prolonged skin contact with items that release nickel ions.

In large doses (>0.5 g), some forms of nickel may be acutely toxic to humans when taken orally (DALDRUP et al. 1983, SUNDERMAN et al. 1988). The acute lethality of nickel following oral exposure is dependent on the chemical form of nickel. A fatal case of nickel poisoning was reported for a $2^1/_2$--year-old girl who had ingested 15 g of $NiSO_4$ (3.3 g elemental Ni) and died of a cardiac arrest (DALDRUP et al. 1983). Death due to nickel-induced Adult Respiratory Distress Syndrome was reported for a worker spraying nickel using a thermal arc process (RENDALL et al. 1994). Death occurred after

13 days, and a total nickel intake was estimated at nearly 1 g. Nausea, vomiting, abdominal, headache, cough, shortness of breath, and giddiness were reported for 32 workers of an electroplating plant who drank water contaminated with nickel chloride and nickel sulfate (1.63 $g \cdot dm^{-3}$) (SUNDERMAN et al. 1988). Some studies have also provided information indicating the deterioration of nickel-induced dermatitis for women following exposure to dietary nickel (ATSDR 1988, SUTHERLAND, COSTA 2002).

Only a small portion of nickel ingested is absorbed by the body. From nickel balance experiments, HORAK and SUNDERMAN (1972) have estimated that about 10% of the nickel in a normal diet is absorbed. Alternatively, other studies have shown that an average nickel resorption from a normal diet is between 20 and 25% (MYRON et al. 1978, FLYVHOLM et al. 1984). Adverse health effects after oral exposure occurred only when nickel levels exceed many times levels of the metal normally occurred in food or drinking water and are decidedly rare cases.

The most hazardous route of exposure to nickel is by inhalation (SUTHERLAND, COSTA 2002). The chemical form of the metal and its solubility is a key determining factor in the toxicity mechanisms. Water-soluble nickel compounds can be absorbed by the lungs into the bloodstream and removed by the kidneys. Insoluble nickel compounds, however, can build up and remain in the lungs for a longer time. Inhalation of soluble nickel causes irritation of the nose and sinuses and can also lead to loss of the sense of smell or perforation of the nasal septum. Long-term exposure may lead to asthma, bronchitis or other respiratory diseases. The most acute nickel poisoning is caused by $Ni(CO)_4$. Exposure to nickel carbonyl can cause headaches, nausea, vomiting, chest pain and breathing problems, in the case of high exposure it may even lead to pneumonia and death. Inhalation of nickel can also cause cancer of the lungs, nose and sinuses (ZHICHENG 1994). Cancers of the throat and stomach have also been attributed to inhalation of nickel. Nickel carbonyl and insoluble nickel compounds (Ni_3S_2, NiO) are the forms of nickel responsible for cancer. Epidemiological studies have demonstrated increased mortality from cancers of the lung and nasal cavities in nickel refinery workers who were chronically exposed by inhalation of nickel-containing dusts and fumes (SEILKOP, OLLER 2003).

Nickel subsulphide (Ni_3S_2) is a well-known respiratory carcinogen. When it is inhaled, particles of Ni_3S_2 lodge themselves deep in the lungs, where they reside in contact as a solid with epithelial cells. These particles are cleared by macrophage cells, which remove them through the digestive tract. Under a condition of high exposure, the macrophage capacity for removal could be perturbed and Ni_3S_2 particles may be taken into epithelial cells by endocytosis. In this way nickel is delivered to the nucleus of lung epithelial cells and can cause a heritable change in chromosomes. It was also demonstrated that Ni_3S_2 induced lesions of both double- and single-stranded DNA in human cells; Ni_3S_2 treatment of cultured HeLa cells induced a 1.5-fold increase in 8-hydroxy-2'-deoxyguanosine compared with a control (KAWANISHI et al. 2001).

When mice were orally administered acute doses of $NiCl_2$ (from 3.4 to 108.8 mg·kg^{-1} body weight), a significant dose-dependent increase in DNA damage was observed in comparison with controls (DANADEVI et al. 2004). The results of research by CAVALLO et al. (2003) confirm involvement of nickel ($NiSO_4$) in production of reactive oxygen metabolites and in inhibition of DNA repair at doses comparable to environmental exposures such as concentrations found in biological fluids. It was demonstrated that some nickel complexes such as $[NiCR]^{2+}$ and $[Ni(CR\text{-}2H)]^{2+}$ bind to the minor groove of double-stranded DNA (MATKAR et al. 2006). Moreover, macrocyclic nickel complex $[Ni(CR\text{-}2H)]^{2+}$ can damage DNA *in vivo* and *in vitro* even in the absence of oxidizing agents. This activity leads to DNA strand breaks and *in vivo* cytotoxicity. Nuclear nickel may be also involved in production of oxygen radicals ($^{\bullet}OH$, H_2O_2) which could damage DNA. CHEN et al. (2003) showed that the level of hydroxyl radical in the Ni-treated group was much higher than in control. Moreover, nickel has been also shown to inhibit DNA repair in a way that may play a role in its toxicity. It has been proposed that nickel may bind to DNA-repair enzymes and generate oxygen-free radicals which cause *in situ* protein degradation. This irreversible damage to the proteins involved in DNA repair, replication, recombination, and transcription could be important for the toxic effects of nickel (LYNN et al. 1997). It happens especially when such DNA is associated with tumor suppression genes; under this condition, cancer cells could replicate at a high rate, thus reducing the time available for repair of the DNA damages (SUTHERLAND, COSTA 2002).

Often co-exposure to a second carcinogen caused a synergistic cancer increase. For example, intramuscular injection of nickel sulfide with 3,4-benzopyrene in rats produced more sarcomas in shorter time than with nickel sulfide alone (MAENZA et al. 1971). When the transforming potential of soluble nickel(II) was compared with such potential of other carcinogens, the efficiency of immortalization by nickel(II) was found to be higher than that by other carcinogens, including benzo[a]pyrene, diol epoxide, N-methyl-N-nitrosourea or, g or X-rays (TROTT et al. 1995).

It was also found that nickel(II) chloride with a classical carcinogen, such as UV radiation (UVR) had synergistic effect on skin cancer induction in Skh1 hairless mice (UDDIN et al. 2007). Mice drinking water containing Ni had significantly higher skin concentration of nickel compared with mice having no nickel in water. It was shown that co-carcinogenic effect of oral nickel with UVR as a matter of cancer yield and incidence was directly correlated with nickel concentration in the skin. Since humans are exposed to both UVR from sunlight and to nickel *via* environmental exposure, there is a potential co-carcinogenic hazard posed by environmental metals (arsenic, chromium, nickel) with UVR, which may be more serious than the hazard of the metals alone (UDDIN et al. 2007).

Some studies have also revealed that compounds of the essential metals: Mn(II), Mg(II) and Zn(II) given to rats with Ni_3S_2, significantly reduced local tumor incidence in a dose dependent manner. Mg(II) was the strongest and Zn(II) was the weakest inhibitor (KASPRZAK et al. 2003).

MOLECULAR MECHANISMS OF NICKEL CARCINOGENESIS

According to the IARC evaluation (IARC 1997) there are sufficient evidences in humans for the carcinogenicity of nickel sulfate and of the combinations of nickel sulfides and oxides encountered in the nickel refining industry. Hence, they can be classified in Group 1, i.e. cancerogenic to humans. As there is inadequate evidence in humans for the carcinogenicity of metallic nickel, it may be carcinogenic to humans (Group 2B).

The mechanism by which nickel causes cancer is still questionable and needs further investigation. Exposure of cells to the metal induces a variety of gene expression changes.

Nickel is known to be a calcium channel blocker (ZAMPONI et al. 1996), and several studies related toxic and carcinogenic effect of nickel with changes in calcium metabolism. It has been suggested also that high levels of nickel may impair absorption or utilization of iron when iron status is low.

It was revealed that acute treatment of rodent cells with nickel is very efficient at turning off the expression of thrombospondin I (TSP I) (SALNIKOW et al. 1994, SALNIKOW et al. 1997). The TSP protein is a regulator of tumor development; high level of TSP suppresses growth of blood vessels into the tumor body. It was shown, that ATF-1 transcription factor is hyperactivated in nickel-transformed cells and plays the role of a negative regulator of thrombospondin I. Therefore, the loss of TSP I expression in tumors promotes angiogenesis and stimulates tumor growth. Another transcription factor, which level was found to be increased after acute exposure to nickel, is hypoxia-inducible factor 1 (HIF-1) (SALNIKOW et al. 2002). During tumor development, HIF-1 facilitates angiogenesis that is essential for tumor growth. Like hypoxia, Ni(II) induces HIF-1 and therefore activates genes responsible for the up-regulation of glucose metabolism and glycolysis even in the presence of oxygen, the vascular endothelial growth factor, and the tumor marker *Cap43* (ZHOU et al. 1998, SALNIKOW et al. 2000).

Nickel is also known to cause inflammatory response for example through regulation of expression of transcription factors involved in inflammatory processes. It was shown that activation of NF-κB by nickel causes modulation of cellular and tissue responses, and can explain nickel-induced allergic effects and contact skin hypersensitivity (VIEMANN et al. 2007). NF-κB is a transcription factor important for apoptosis, inflammatory response, and expression of adhesion molecules. Intercellular adhesion molecule-1, vascular cell adhesion molecule-1, and endothelial leukocyte adhesion molecule-1 were found to be up-regulated by Ni(II) in cultured human endothelial cells (GOEBELER et al. 1993). A strong increase of NF-κB binding with DNA was found after stimulation of HUVEC with Ni(II) (GOEBELER et al. 1995, DENKHAUS, SALNIKOW 2002, KASPRZAK et al. 2003).

There is also a possibility of involvement of *p*53 gene mutations in nickel-induced transformation (DENKHAUS, SALNIKOW 2002). *P*53 is a tumor suppressor gene and transcription factor involved in the regulation of cell proliferation and apoptosis. Mutations in *p*53 are the most common genetic alterations found in human cancer. The *p*53 gene was reported to be mutated in human kidney epithelial cells chronically exposed to and eventually transformed by nickel (MAEHLE et al. 1992).

The *FHIT* (Fragile Histidine Triad) is a tumor suppressor gene located in a fragile chromosomal site sensitive to deletions. Its expression is frequently reduced or lost in tumors and pre-malignant lesions. The product of the gene, Fhit protein (phosphohydrolase), induces apoptosis through a complex interaction with its substrate, diadenosine triphosphate. Ni(II) was found to strongly inhibit the enzymatic activity of Fhit protein *in vitro* and also suppress Fhit expression in nickel-transformed BALB/c-3T3 cells (KOWARA et al. 2004).

In another study, nickel chloride was found to induce lipid peroxidation in the plasma of human blood *in vitro* in a concentration-dependent and time-dependent manner. The hydroxyl radical production increased in a concentration-dependent manner after Ni treatment for 1 h. Furthermore, a decreasing trend in a-tocopherol levels in plasma was observed after nickel exposure. Incubation with gluthathione, catechin, and mannitol decreased lipid peroxidation and reduced hydroxyl radical formation induced by Ni, but a greater decrease of a-tocopherol levels in plasma occurred with catechin (CHEN et al. 2002).

Further progress in understanding molecular mechanisms of nickel carcinogenicity has been achieved by the finding that nickel compounds increase the extent of DNA methylation and histone deacetylation, which leads to the inactivation of gene expression. Although the mechanisms by which nickel induces DNA hypermethylation are unknown, a possible model including the ability of nickel to substitute for magnesium, increase chromatin condensation and trigger *de novo* DNA methylation has been proposed (LEE et al. 1995). It is possible that inactivation of tumor suppressor gene by hypermethylation could assist in nickel-induced cell transformation. In addition to gene silencing by hypermethylation, a suppressive effect of nickel on histone H4 acetylation *in vitro* has been reported for both yeast and mammalian cells (BRODAY et al. 2000). Despite numerous reports of the DNA and chromatin damage observed in nickel-exposed cells and tissues, the mutagenic potential of this metal is generally considered to be low (FLETCHER et al. 1994).

CONCLUSIONS

Nickel is a ubiquitous metal, which finds increasingly more applications in modern technologies. Contact with nickel compounds (both soluble and insoluble) can cause a variety of adverse effects on human health. The most important and frequent are nickel allergy in the form of contact dermatitis, lung fibrosis, cardiovascular and kidney diseases, and lung and nasal cancers. There is evidence that some nickel compounds are carcinogens to humans. However, the exact mechanism of nickel-induced carcinogenesis is still unclear.

In 2008, nickel received the name of the "Allergen of the Year". According to dermatologists the frequency of nickel allergies is still growing and all the sources of human exposure to nickel should be recognized and examined.

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