

Urszula WYDRO^{1*}, Elżbieta WOŁEJKO¹,
Andrzej BUTAREWICZ¹ and Tadeusz ŁOBODA¹

EFFECT OF SEWAGE SLUDGE ON BIOMASS PRODUCTION AND CONTENT OF MACRONUTRIENTS AND CHLOROPHYLL IN GRASS MIXTURES

WPLYW OSADU ŚCIEKOWEGO NA PRODUKCJĘ BIOMASY I ZAWARTOŚĆ MAKROSKŁADNIKÓW ORAZ CHLOROFILU W MIESZANKACH TRAW

Abstract: The aim of this study was to determine the effect of various doses of municipal sewage sludge on the growth of biomass of aboveground parts of lawn grass mixtures and their macronutrients and chlorophyll content. Four experiments on the lawns along the main roads of Białystok (at Popieluszki, Hetmanska, Piastowska and Raginisa Strs.) were founded. Three doses of sewage sludge (0.0 – control; 7.5 and 15.0 kg · m⁻²) and two grass mixtures: Eko and Roadside were factors in experiment. Samples of aboveground part of grasses were collected 3 times: in June, August and October 2011, after that dry matter of the aboveground part of grasses from 1 m², the total nitrogen content, total phosphorus content, total potassium content and chlorophyll *a* and *b* content were determined. The dose of sewage sludge significantly influenced N and K accumulation in the grasses. The maximum average nitrogen content (2.82 % of d.m.) was observed in grass from plots with the highest dose of sewage sludge, while the potassium from plots with sewage sludge dose 7.5 kg · m⁻². The accumulation of biomass of grasses in the study period was mainly differentiated by the sewage sludge dose and the sampling time. The average summary yield for the three months study (June, August and October) was from 229.83 to 430.70 g · m⁻². The highest results were obtained at a dose of sewage sludge 15.0 kg · m⁻². The sampling time and dose of sewage sludge significantly influenced chlorophyll *a* and *b* content. The average chlorophyll *a* content in June, August and October was: 0.60; 0.64 and 0.54 mg · g⁻¹ of f.m. respectively, while chlorophyll *b*: 1.00; 0.94; 0.39 mg · g⁻¹ of f.m. It was found a low average ratio of chlorophyll *a* to *b* in mixtures of grasses, which ranged from 0.54 in June to 1.82 in October to mixture Eko and 0.57 in June to 1.68 in October to mixture Roadside. Plants of the control plots showed higher average ratio of chlorophyll *a* to *b* in comparison with plants from plots with sewage sludge amendment.

Keywords: biomass, lawn grasses, macronutrients, chlorophyll, sewage sludge

¹ Division of Sanitary Biology and Biotechnology, Białystok University of Technology, ul. Wiejska 45E, 15–351 Białystok, Poland.

* Corresponding author email: uwydro@gmail.com

Introduction

Disposal of sewage sludge is becoming an increasing problem due to the increasing amount of biosolids. The main reason of this situation is the fact that amount of treated wastewater is increasing, which is associated with the dynamic development of sewerage system. Measurable result is an increasing percentage of the population supported by sewage treatment and a continuous increase in amount of municipal sewage sludge [1]. Therefore, it is necessary search for methods and ways of rational use of this biosolid.

One way of utilization of municipal sewage sludge is to use it for recultivation of degraded areas, biological fixation of surface from wind or water erosion or to improve water retention and possible fertilization of plants not intended for human consumption [2, 3]. It is justified by the fertilizer properties of the sewage sludge such as high content of nitrogen, phosphorus, carbon, macro- and microelements, which are necessary to improve soil conditions and plant growth [4–7]. Moreover, due to the high content of organic matter in biosolid, it is possible to improve soil physical properties such as soil aeration and its water holding capacity, helping to improve soil microbial activity, eg respiration and enzymatic activity [7–9]. Therefore, very promising future area of application of municipal sewage sludge is land adjacent to roads. Urban soils are usually formed from building waste, which means that their structure is more dense, have lower content of humus, lower water capacity and water permeability, and lower biological activity [10]. Furthermore, developing industry and transport communications are threats for urban soils because of their progressive chemical degradation that leads to a sustainable and progressive deterioration of their properties. It is a reason that urban soils require recultivation treatment [11].

Green city areas, including lawns, have various kinds of functions. On the one hand, well cared lawns enhance the aesthetic value of the city, on the other hand, play phytoremediation role, both for the air and soil generated by transport [12].

For the effective phytoremediation plants should be characterized by high resistance to difficult environmental conditions, have the ability to accumulate xenobiotics from the soil, as well as rapid growth and high dry mass production. Grasses are characterized by high phytoremediation potential [13]. Grasses in urban areas can fulfill the above requirements when they have good conditions for rapid growth and high dry mass production.

The aim of this paper was to assess the effect of different doses of municipal sewage sludge for the production of dry mass of lawn grass mixtures and the contents of nitrogen, phosphorus, potassium, chlorophyll *a* and *b* and the ratio of chlorophyll *a* to *b* in urban lawn grasses.

Materials and methods

Four experiments on the lawns along the main roads of Białystok: Popiełuszki Str., Hetmańska Str., Piastowska Str. and Raginisa Str. were founded. Each experimental area was 90 m² and each of them was divided into 18 plots with 5 m² area. The factors of the experiment were: 3 doses of sewage sludge and two mixtures of lawn grasses.

Municipal sewage sludge from the Municipal Wastewater Treatment Plant in Sokolka was used. Sewage sludge was stabilized and had a smear texture. For fertilization 3 doses of sewage sludge: 0.0, 7.5 and 15.0 kg · m⁻² were used. Doses of sewage sludge were established according to Kiryluk [14] who found in several years study that the most effective doses for turfing of municipal waste disposal areas were those above 40 Mg/ha. Sewage sludge in autumn 2010 was used.

Before establishment of experiment both sewage sludge and soil from each combination were analyzed according to Directive of Environmental Minister from July 13th, 2010 concerning municipal sewage sludges [2]. Analyzes were done by Regional Chemical and Agricultural Station in Bialystok (Tables 1 and 2).

Table 1

Selected physical and chemical properties of soils

Properties	Popieluszki	Hetmanska	Piastowska	Raginisa
pH	7.6	7.9	7.7	7.4
Sand [%]	75.69	75.89	71.87	84.35
Silt [%]	22.30	22.02	25.41	14.67
Clay [%]	2.01	2.09	2.72	0.98
Textural class	loamy sand	loamy sand	sandy loam	sand
P ₂ O ₅ [mg/100 g]	22.0	7.3	18.4	10.0

Table 2

Selected properties of municipal sewage sludge

Properties	Municipal sewage sludge
pH	6.7
Dry weight [%]	19.30
Organic matter [% d.m.]	58.40
Total P [% d.m.]	2.73
Total N [% d.m.]	3.99
Ammonium N [% d.m.]	0.14
Ca [% d.m.]	5.51
Mg [% d.m.]	0.66
Pb [mg/kg d.m.]	23.5
Cd [mg/kg d.m.]	< 0.50
Cr [mg/kg d.m.]	58.00
Cu [mg/kg d.m.]	194.00
Ni [mg/kg d.m.]	22.00
Zn [mg/kg d.m.]	1459.00
Hg [mg/kg d.m.]	1.04

In spring 2011, on the prepared plots two mixtures of lawn grasses were seeded: Eko (M1) from Nieznanice Plant Breeding Station which included 30 % of *Lolium perenne*

cv. Niga, 15 % of *Poa pratensis* cv. Amason, 22.6 % of *Festuca rubra* cv. Adio and 32.4 % of *Festuca rubra* cv. Nimba. and grass mixture Roadside (M2) from Barenbrug which included 32 % of *Lolium perenne* cv. Barmedia, 5 % of *Poa pratensis* cv. Baron, 52 % of *Festuca rubra rubra* cv. Barustic, 5 % of *Festuca rubra commutata* cv. Bardiva (BE) and 6 % of *Festuca rubra commutata* cv. Bardiva (NL).

There were obtained 72 test plots (4 locations 2 grass mixtures 3 doses of sewage sludge 3 replicates), each with 5 m² area.

Dry matter of the aboveground part of grasses from 1 m², total nitrogen content, total phosphorus content, total potassium content and chlorophyll *a* and *b* content were determined in grass samples collected in June (27.06.2011), August (16.08.2011) and October (14.10.2011). Aboveground parts of grass mixture were also harvested 21.07.2011 and 18.09.2011 and their dry matter was determined, but macronutrient and chlorophyll contents were not determined.

Aboveground parts of plants were cut from a random 33.3 × 33.3 cm area of each plot in order to determine dry matter. Grass samples were dried at 105 °C for 24 h, then at 75 °C, for complete water evaporation.

Total nitrogen in aboveground parts of grass was determined by the Kjeldahl method after sample mineralization in concentrated sulfuric acid [15].

Total phosphorus in aboveground parts of grasses was determined by molybdo-vanadate method after sample mineralization in concentrated sulfuric acid with hydrogen peroxide [15].

Total potassium concentration in plant material were determined using *Atomic Absorption Spectrometry* AAS (Varian SpectrAA-100A). Samples were mineralized in temperature about 450 °C and ashes were dissolved in concentrated nitric(V) acid.

For chlorophyll determination fresh plant material was homogenized in a mortar with addition of CaCO₃ and quartz sand. Chlorophyll was extracted with 80 % acetone. Chlorophyll *a* and *b* content was determined using HACH DR5000 spectrophotometer by measuring absorbance at λ = 663 and 645 nm. The content of chlorophyll *a* and *b* were calculated according to the formulas:

$$\text{Chlorophyll } a = (12.7 \cdot D_{663} - 2.7 \cdot D_{645}) \cdot V \cdot (1000 w)^{-1}$$

$$\text{Chlorophyll } b = (22.9 \cdot D_{645} - 4.7 \cdot D_{663}) \cdot V \cdot (1000 w)^{-1}$$

where: D₆₄₅ and D₆₆₃ – optical density at λ = 645 and 663 nm, respectively,

V – volume of the solution in [cm³],

w – fresh weight of the leaves sample in [g].

The results were statistically analyzed using analysis of variance with Tukey test at significance level at α = 0.05. The correlation between characteristics were calculated using Statistica 9.0.

Results and discussion

One of the consequences of the use of sewage sludge for plant fertilization may be change of plant chemical composition. The nutrients contained in biosolid are beneficial

for plant growth and development. On the other hand, the introduction of the sludge to the soil also entails risk of the circulation of heavy metals and other contaminants, which can cause adverse effects on physiological processes in plants [16]. Municipal sewage sludge used in the experiment satisfied the requirements of Regulation [3] on the application of sewage sludge to non-agricultural land recultivation. Furthermore, sludge are characterized by a high abundance of nitrogen and phosphorus, and the organic matter (Table 2). Municipal sewage sludge fertilization influenced concentration of nitrogen, potassium, chlorophyll *a* and *b* in plants and the accumulation of aboveground dry matter of grasses.

Uptake and use of minerals by plants from sludge is dependent on many factors *ie* holding water capacity, redox potential, soil temperature, microbial activity in the rhizosphere. Each of this factors (either alone or in combination with the others) can stimulate or inhibit mineral uptake by the plant and its impact on their chemical composition [17].

Nitrogen is an element that plants take in the largest quantities. This is mainly due to the fact that it is a part not only of amino acids and proteins but also the nucleotides (such as adenosine-5'-triphosphate – ATP) and nucleic acids, some of the plant hormones, some of secondary metabolites, and other biologically important compounds [18]. Several studies have shown that nitrogen is one of the most important elements of yielding, and nitrogen fertilization is especially effective when the soil is rich in other nutrients [19].

The nitrogen concentration in the used grass mixtures was differentiated, but statistically insignificant. Factors that influenced the nitrogen concentration of the studied grasses were biosolid dose, sampling time and location (Table 3).

The lowest average concentrations of total nitrogen were observed in control plants (1.90 % of d.m.). Addition of the sludge to the soil resulted in increase of nitrogen concentration in grasses (on average 38.0 %) compared with control plots. Similar results were obtained by Gondek and Filipek-Mazur [20], who found higher nitrogen concentration in white mustard after the application of sewage sludge than without fertilization. The highest nitrogen concentration in the plant samples (2.80 %) collected from the plots with the highest dose sludge was observed. Sewage sludge used in the study of Jama and Nowak [21] caused increase of nitrogen concentration in the leaves of willow at a single dose (75.0 Mg (t)/ha), while double dose caused a decline of this component.

The average concentration of nitrogen in grass mixtures was lowest in samples collected in October (2.06 % of d.m.) and the highest in samples of August (2.54 % of d.m.). Nitrogen in the sludge mainly occurs in organic form [22]. Sewage sludge amendment causes intensive soil microbial activity and intensive development of rhizosphere [23, 24]. As a result of progressive mineralization of organic nitrogen, its mineral forms (NO_3^- and NH_4^+) are released and nitrogen becomes available to plants in optimum amounts [9, 25]. Availability of nutrients from sewage sludge is a function of climatic conditions during vegetation period, dose of sewage sludge and sludge C to N ratio [19]. Decrease in temperature reduces activity of soil microorganisms which are responsible for decomposition of organic matter [24]. October is the month when plant

vegetation is ceasing and plant demand for this component decreases. Furthermore, the biochemical processes occurring in the soil are inhibited and intensity of release of nitrogen is lower.

Table 3

Total nitrogen, phosphorus and potassium concentration [% of d.m.] in aboveground parts of grass mixtures (ECO and ROADSIDE) grown in Białystok at Popieluszki, Hetmanska, Piastowska and Raginisa streets. Plants were harvested in June, August and October 2011

Factor	N [% of d.m.]	P [% of d.m.]	K [% of d.m.]	Dry matter [g · m ⁻²]
A – grass mixtures				
Eko	2.33	0.25	1.60	111.14
Roadside	2.42	0.24	1.49	113.76
LSD _{0.05}	ns.*	ns.	0.80	ns.
B – dose of sewage sludge				
0.0 kg · m ⁻²	1.90	0.24	1.45	76.61
7.5 kg · m ⁻²	2.41	0.23	1.63	117.17
15.0 kg · m ⁻²	2.82	0.27	1.57	143.57
LSD _{0.05}	0.15	ns.	0.11	14.19
C – time of sampling				
June	2.52	0.24	1.73	158.37
August	2.54	0.22	1.78	126.70
October	2.06	0.27	1.13	52.28
LSD _{0.05}	0.15	ns.	0.11	14.19
D – localization				
Popieluszki St.	2.26	0.27	1.36	83.22
Hetmanska St.	2.17	0.22	1.46	105.55
Piastowska St.	2.22	0.24	1.82	154.73
Raginisa St.	2.84	0.25	1.55	106.30
LSD _{0.05}	0.20	ns.	0.15	18.25

* not significant differences.

Average accumulation of nitrogen by mixtures of lawn grasses was the highest at Raginisa Str. (2.84 % of d.m.), while the lowest at Hetmanska Str. (2.17 % of d.m.). It can be assumed that intensity of accumulation of this component by plants largely depended on soil condition at the given location and the velocity of mineralization of organic matter (C : N ratio in the soil mainly) as Gondek [19] was reported.

Phosphorus, like nitrogen, is an essential macronutrient for plant growth. Phosphorus concentration in the grasses in each time of the study was similar independently of all studied factors (Table 3). This indicates that the sewage sludge amendment did not influence the amount of this component in the aboveground parts of grass mixtures. Similar results obtained Jakubus [15]. The author argues that phosphorus from sewage sludge as compared with other biogenic elements was the least used by plants. This is

probably due to occurrence of this element in the forms of sparingly soluble and, therefore sparingly available to plants. Phosphorus in the soil undergoes strong chemical sorption. The rate of release of this component depends largely on the soil pH [20]. It should be noted that soil amendment with sewage sludge showed high pH in the range 7.4–7.9 (Table 1), which is characteristic for urban soils, which are usually created with debris-limestone additives and high pH they may be also caused by precipitation of alkaline dust [26]. Alkaline soil pH (above 7) may cause immobilization of phosphorus by forming its compounds with calcium and that entails, limiting its availability. Furthermore, no difference in phosphorus concentration in plants after application of sewage sludge may be due to low susceptibility to degradation of sewage sludge [27]. Various sludge mineralization rate determines the various degrees of nutrients release, and thus, their availability for plants.

Another component which grasses collect in large quantities is potassium. In our research all the experimental factors had a significant impact on the content of potassium in the aboveground part of grasses (Table 3). In average 7.40 % more potassium accumulated grass mixture Eko than Roadside. Taking into account dose of sludge, in average the most potassium accumulated grasses from plots, where 7.5 kg/m² of sewage sludge was used (1.63 % of d.m.) (Table 3). Grass fertilized with 15 kg/m² of sewage sludge and without fertilization accumulated 4.00 % and 12.40 % less potassium compared with plots with a single sewage sludge dose. The results are not reflected in the research of Harnisz and Ciecko [28], where they did not observed effect of sludge fertilizer on the potassium concentration in green mass of corn. A higher potassium concentration in the grasses collected in August and June was founded, but less in October (1.78, 1.73 and 1.13 % d.m., respectively). On average the highest accumulation of potassium in the dry mass was in grass from Piastowska Str. (1.82 % d.m.) and the lowest from Popieluski Str. (1.36 % of d.m.). According to the literature, potassium concentration in dry matter of plants is high and ranges from 2 to 5 %. Our studies indicate a low level of this element concentration in the test grass mixtures. According to many authors [7, 4, 29] potassium concentration in sewage sludge is low, because good solubility of potassium compounds what is a reason for its discharge with wastewater. Siuta [4] and Tujaka [29] paid attention to the fact that the use of sewage sludge to fertilization and recultivation should be supplemented with potassium. This is especially important because of the low levels of nutrients in urban soils. Ciecko and Harnisz [28] stated higher potassium content in plants after fertilization with sewage sludge compost with manure and straw, which was an additional source of potassium. Our results indicate that sewage sludge application without addition components rich with potassium does not meet needs of the plants for this component.

Dry matter production is an indicator of soil fertility and plant production potential [30]. The accumulation of dry matter by aboveground lawn grass mixtures in the study period was differentiated by the sewage sludge dose, the sampling time and location (Table 3). The average summary dry matter for the study period (June, August and October) was from 229.83 to 430.70 g · m⁻². Summary production of biomass from the plots with sewage sludge increased on average by 70 % compared with the plots without biosolids. The best results were obtained by using a double dose of sewage

sludge. Taking into account the sampling time, the maximum average dry matter was in June ($158.37 \text{ g} \cdot \text{m}^{-2}$). The minimum yield of dry matter was observed in October ($52.28 \text{ g} \cdot \text{m}^{-2}$). On average the largest dry matter during one harvest was obtained from plots on Piastowska Str. ($154.73 \text{ g} \cdot \text{m}^{-2}$), while the smallest on Popieluski Str. ($83.22 \text{ g} \cdot \text{m}^{-2}$). The studied dry matter of grass mixtures varied, but not statistically significant (Table 3).

Many authors supports yielding role of sewage sludge [16, 19, 30]. According to Gondek [19], production of dry matter and its chemical composition is highly modified not only as a result of fertilizer application but also as a result organic compounds transformation in soil. This transformation depends on frequently changing environmental factors.

Statistical analysis showed that the concentration of potassium and nitrogen in plant is correlated with the amount of dry matter and the correlation coefficients are $r = 0.60$ and 0.39 , respectively. This confirms the fact that nitrogen and potassium are one of a major elements which determine an increase of dry matter as also provides Kutik et al [31]. According to Starck [32], production of dry matter is mainly a result of photosynthetic activity of plants and nitrogen assimilation, which activate growth processes, *eg* in increase of assimilation area of leaves and better plant development [33].

Chlorophyll concentration in leaves is a factor, which can determine plant dry matter, as it allows a plant to trap solar energy and to produce assimilates [34].

Our study shows that municipal soil amendment sewage sludge influenced chlorophyll *a* and *b* concentration in lawn grass mixtures and their ratio. The concentration of chlorophyll *a* in aboveground parts of grasses was dependent on the used grass mixture, harvest time, location and to a lesser extent on the dose of sewage sludge (Table 4). The highest average concentration of chlorophyll *a* was found in August ($0.64 \text{ mg} \cdot \text{g}^{-1}$ of f.m.) and the smallest in samples collected in October ($0.54 \text{ mg} \cdot \text{g}^{-1}$ of f.m.). An average chlorophyll *a* concentration was higher by 5.20 % in Roadside mixture than in Eko mixture ($0.58 \text{ mg} \cdot \text{g}^{-1}$ of f.m.). Average chlorophyll *a* content was the highest in the grass collected from Piastowska Str. ($0.63 \text{ mg} \cdot \text{g}^{-1}$ of f.m.) and the lowest from Popieluski Str. ($0.56 \text{ mg} \cdot \text{g}^{-1}$ of f.m.). A sewage sludge fertilization increased the chlorophyll *a* concentration in average by 3.4 % compared with control plots (Table 4).

The concentration of chlorophyll *b* was differentiated by dose of sludge and sampling time (Table 4). The highest average chlorophyll *b* concentration in the grass samples collected in June was found ($1.00 \text{ mg} \cdot \text{g}^{-1}$ of f.m.) and the lowest in a samples in October ($0.39 \text{ mg} \cdot \text{g}^{-1}$ of f.m.). The increase concentration of chlorophyll *b* was proportional to the dose of sewage sludge. Sewage sludge resulted in its increase of average concentration of 37.10 % compared with control plots ($0.62 \text{ mg} \cdot \text{g}^{-1}$ of f.m.).

Results of our study suggest that one of the differentiating factors of chlorophyll concentration is a kind of grass mixtures. This confirms the observations of Gebczynski [35], who suggests that a level of chlorophyll *a+b* in plants may fluctuate and one of the major factor of variation of this parameter can be a strain. Additionally these results show that sewage sludge differentiated chlorophyll concentration. It is not confirmed by Woloszyk and Maciorowski [36] who worked with rape and winter wheat, and used sewage sludge compost. They had no effect on a chlorophyll content in a leaves.

Table 4

Average chlorophyll *a* and *b* concentration [mg/g f.m.] in aboveground parts of grass mixtures (ECO and Roadside) grown in Białystok at Popieluski, Hetmanska, Piastowska and Raginisa streets. Plants were harvested in June, August and October 2011

Factor	chl <i>a</i> [mg/g f.m.]	chl <i>b</i> [mg/g f.m.]
A – grass mixtures		
Eko	0.58	0.77
Roadside	0.61	0.78
LSD _{0.05}	0.02	ns.*
B – dose of sewage sludge		
0.0	0.58	0.62
7.5	0.59	0.81
15.0	0.61	0.89
LSD _{0.05}	0.03	0.07
C – time of sampling		
June	0.60	1.00
August	0.64	0.94
October	0.54	0.39
LSD _{0.05}	0.03	0.07
D – localization		
Popieluski St.	0.56	0.76
Hetmanska St.	0.58	0.74
Piastowska St.	0.63	0.81
Raginisa St.	0.60	0.79
LSD _{0.05}	0.04	ns.

* ns. – not significant differences.

Kachel-Jakubowska [37] highlights that chlorophyll concentration is correlated with nitrogen concentration, which is a component of chlorophyll [33]. The correlation coefficients obtained in our study ($r = 0.33$ and $r = 0.58$) confirm relation of chlorophyll *a* and *b* with nitrogen concentration in the grass and positive correlation of chlorophyll *a* and *b* concentration with biomass production ($r = 0.41$ and $r = 0.47$). Swedrzynska et al [38] in research on corn and oats found that, when nitrogen fertilization increased, level of chlorophyll and aboveground dry matter increased significantly. According to Cieccko et al [39], potassium is also responsible for increasing concentration of chlorophyll in assimilatory parts, which justifies the resulting correlation between the component of grasses and chlorophyll *a* and *b* concentration ($r = 0.34$ and 0.67).

The ratio of chlorophyll *a* to chlorophyll *b* was varied in studied grass in all experimental plots. Taking into account the location of grass mixtures, the highest and lowest average chlorophyll *a* to *b* ratios in plants growing on plots from Raginisa Str. was observed and it was from 0.64 to 1.15 for Eco mixture and from 0.63 to 1.02 for

Roadside mixture (Fig. 1a, 1d). Taking into account the sample time collection, the average ratio of chlorophyll *a* to *b* was from 0.54 in June to 1.82 in October for Eko mixture and from 0.57 in June to 1.68 in October for Roadside mixture (Fig. 1a, 1b).

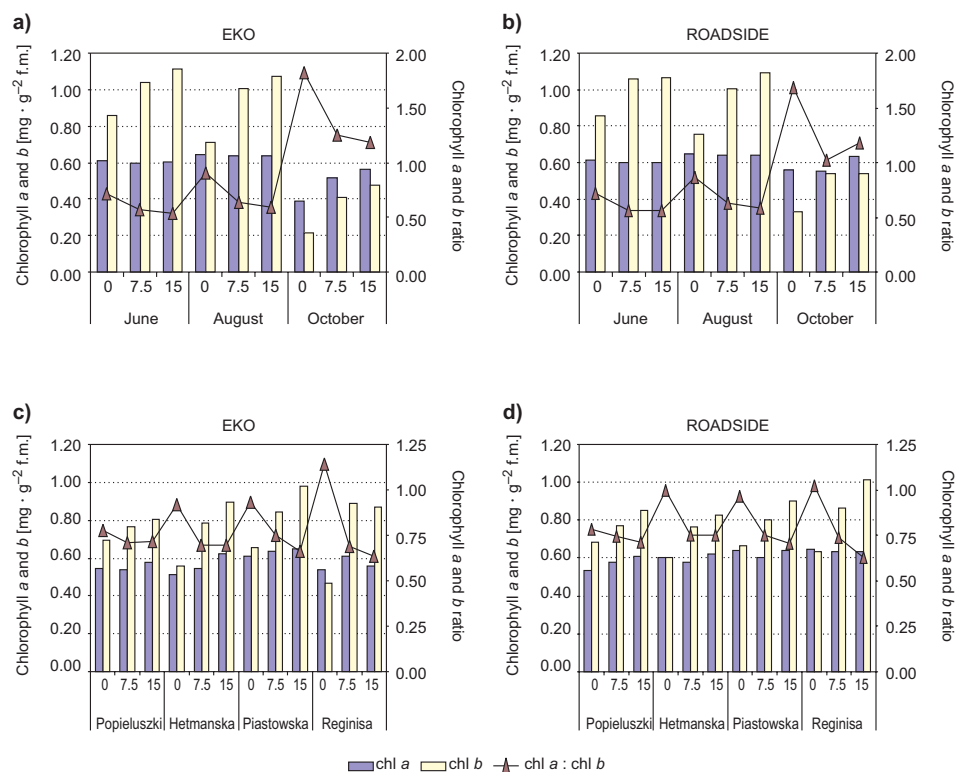


Fig. 1. Average chlorophyll *a* and *b* concentration [$\text{mg} \cdot \text{g}^{-1}$ of f.m.] and chlorophyll *a* to chlorophyll *b* ratio in aboveground parts of grass mixtures (a, c – EKO and b, d –ROADSIDE) grown in Białystok at Popieluszki, Hetmanska, Piastowska and Reginisa streets. Plants were harvested in June, August and October 2011

According to literature, ratio of chlorophyll *a* to chlorophyll *b* of higher plants is about 3. In our study, a much lower ratio of chlorophylls was obtained. Taking into account the date of harvest, the lowest ratio of chlorophyll *a* to *b* occurred in June and August, while in the grass collected in October higher ratio of chlorophylls was found (Fig. 1a–d). The reason for variations in chlorophyll *b* concentration was probably a dose of sewage sludge, which influence was more significant for chlorophyll *b* than for chlorophyll *a*. Probably, in the first months of research, plants were affected by stress caused by application of sewage sludge and month to month the plants have evolved adaptive mechanisms. According to Starck [40], changes in chlorophyll *a* and *b* in unfavorable conditions may be also a result of repair mechanisms, which could explain the low ratio of chlorophylls in August and higher in October. Many authors draw attention to heavy metals in sewage sludge, which in excessive amounts may adversely

affect the plants [36, 41]. Moreover Sikorski [42] points out that the chlorophylls are considered to be the most sensitive vegetables pigments and heavy metals speed up changes chlorophylls concentration [43].

In the case of grass grown under control conditions, the average ratio of chlorophyll *a* to *b* was low, but in all cases, it was higher in comparison with the plants from fertilized plots (Fig. 1a–d). During ontogeny plants are exposed to various biotic and abiotic stressors. A stress causes certain plant reaction which consist to changing metabolic processes and as a result of its growth and development [44]. Pollution caused by transport in areas along a routes could be an additional stress factor that contributed to the low chlorophyll *a* to *b* ratio in plants collected from control plots in the first months of study.

Conclusion

1. Sewage sludge soil amendment of urban lawns influenced plant nitrogen, potassium, chlorophyll *a* and chlorophyll *b* concentrations, the ratio of chlorophyll *a* to *b* the yield of dry matter.

2. Nitrogen concentration in the studied mixtures of lawn grasses increased proportionally with dose of sewage sludge. The highest concentration of this component in the grasses was when $15.0 \text{ kg} \cdot \text{m}^{-2}$ of sewage sludge was used. Accumulation of nitrogen in plants depended on the sampling time and location.

3. None of the experimental factors did influence significantly the concentration of phosphorus in lawn grass mixtures.

4. Potassium concentration in lawn grass mixtures was low in all variants of fertilization. Level of potassium in grasses depended on the dose of sewage sludge, mixtures of grasses and location.

5. Dry matter of investigated lawn grass mixtures was determined by the dose of sewage sludge, the time of sampling and location. The highest grass dry matter was from plots where the highest dose of sludge ($15.0 \text{ kg} \cdot \text{m}^{-2}$) was used.

6. Sludge fertilization influenced chlorophyll *a* and *b* concentration in the studied grasses; an increase of dose caused an increase in chlorophyll concentration.

7. The availability of main macronutrients for plants should be considered taking into account not only dose of applied sewage sludge but also other environmental factors including impact of transport.

Acknowledgement

This work was done with financial support of NCN, project N305 367438.

References

- [1] Uchwała Rady Ministrów Nr 233 z dnia 29 grudnia 2006 r w sprawie „Krajowego Planu Gospodarki Odpadami 2010”. Załącznik „Krajowy Plan Gospodarki Odpadami 2010”.
- [2] Instytut Inżynierii Środowiska. Określenie kryteriów stosowania osadów ściekowych poza rolnictwem. Częstochowa: Politechnika Częstochowska; 2004.

- [3] Rozporządzenie Ministra Środowiska z dnia 2010 r w sprawie komunalnych osadów ściekowych (DzU 2010, Nr 137, poz 924).
- [4] Siuta J. Uwarunkowania i sposoby przyrodniczego użytkowania osadów ściekowych. Inż Ekol. 2003;9:7-42.
- [5] Wieczorek J, Gambuś J. Porównanie działania obornika i komunalnych osadów ściekowych na plonowanie i skład chemiczny słonecznika w doświadczeniu lizymetrycznym. Zesz Probl Post Nauk Roln. 2009;537:359-368.
- [6] Czekąła J, Mielnik L. Zmiany ilościowe i jakościowe związków próchnicznych zachodzące podczas kompostowania osadów ściekowych z udziałem kory sosnowej i trocin. Zesz Probl Post Nauk Roln. 2009;537:57-66.
- [7] Singh RP, Agrawal M. Potential benefits and risks of land application of sewage sludge. Waste Manage. 2008;28:347-358. DOI: 10.1016/j.wasman.2006.12.010.
- [8] Pathak A, Dastidar MG, Sreekrishnan TR. Bioleaching of heavy metals from sewage sludge: A review. J Environ Manage. 2009;90:2342-2353. DOI: 10.1016/j.jenvman.2008.11.005.
- [9] Wong JWC, Lai KM, Fang M, Ma KK. Effect of sewage sludge amendment on soil microbial activity and nutrient mineralization. Environ Int. 1998;24(8):935-943.
- [10] Greinert A. Gleby i grunty miejskie. In: Stan środowiska w Zielonej Górze w 1999 roku. Zielona Góra: Woj Inspekt Ochrony Środow. 2000;107-117. ISBN 83-7217-096-7.
- [11] Greinert A. Ochrona i rekultywacja terenów zurbanizowanych. Monografia nr 97. Zielona Góra: Wyd Politech Zielonogórskiej; 2000. ISBN 83-85911-12-X.
- [12] Gawroński SW. Fitoremediacja a tereny zieleni. Zieleń Miejska. 2009;10:28-29.
- [13] Macek T, Uhlík O, Jecna K, Novakova M, Lovecka P, Rezek J, Dudkova V, Stursa P, Vrchtova B, Pavlikova D, Demnerova K, Mackova M. Advances in Phytoremediation and Rhizoremediation. In: Singh A, Kuhad RC, Ward OP, editors. Advances in Applied Bioremediation, Soil Biology. 2009;17:257-277. DOI 10.1007/978-3-540-89621-0_14.
- [14] Kiryłuk A. Mieszanki traw i osad ściekowy w procesie rekultywacji wysypiska odpadów komunalnych. Lublin: AR Lublin PTG; 2002:85-86.
- [15] Ostrowska A, Gawliński S, Szczubiałka Z. Metody analizy i oceny właściwości gleb i roślin. Katalog. Warszawa: Wyd. IOŚ; 1991.
- [16] Jakubus M. Ocena przydatności osadów ściekowych w nawożeniu roślin. Woda – Środowisko – Obszary Wiejskie. 2006;6,2(18):87-97.
- [17] Kalembsa D, Malinowska E. Zmiany zawartości metali ciężkich w *Miscanthus sacchariflorus* (Maxim) Hack pod wpływem nawożenia osadem ściekowym. Łąkarstwo w Polsce. 2007;10:99-110.
- [18] Adamczyk B, Godlewski M. Różnorodność strategii pozyskiwania azotu przez rośliny. Kosmos. Problemy Nauk Biologicznych. 2010;59,1-2(286-287):211-222.
- [19] Gondek K. Wpływ nawożenia nawozami mineralnymi, obornikiem od trzody chlewnej i komunalnymi osadami ściekowymi na plon i niektóre wskaźniki jakości ziarna pszenicy jarej (*Triticum aestivum* L). Acta Agrophys. 2012;19(2):289-302.
- [20] Gondek K, Filipek-Mazur B. Ocena efektywności nawożenia osadami ściekowymi na podstawie plonowania roślin i wykorzystania składników pokarmowych. Acta Sci Pol Formatio Circumiecius. 2006;5(1):39-50.
- [21] Jama A, Nowak W. Pobieranie makroskładników z osadów ściekowych przez wierzbę krzewiastą (*Salix viminalis* L) i jej mieszańce. Nauka Przyr Technol. 2011;5(6):123-130.
- [22] Maćkowiak Cz. Wartość nawozowa osadów ściekowych. Inż Ekol. 2011;3:135-145.
- [23] Joniec J, Furczak J. Liczebność wybranych grup drobnoustrojów w glebie bielicowej pod uprawą wierzby użyźnionej osadem ściekowym w drugim roku jego działania. Annales UMCS Sec E. 2007;LXII(1):93-104.
- [24] Oleszczuk P. Zanieczyszczenia organiczne w glebach użyźnianych osadami ściekowymi. Część II. Losy zanieczyszczeń w glebie. Ecol Chem Eng. 2007;14(S2):186-198.
- [25] Skowron P, Filipek T, Fidecki M. Wpływ osadu ściekowego z oczyszczalni na plonowanie buraka cukrowego. Acta Agrophys. 2007;10(1):193-198.
- [26] Czarnowska K. Akumulacja metali ciężkich w glebach, roślinach i niektórych zwierzętach na terenie Warszawy. Rocz Glebozn. 1980;31(1):77-115.
- [27] Czekąła J. Wybrane właściwości osadów ściekowych z oczyszczalni regionu Wielkopolski. Cz 2. Zawartość węgla i azotu we frakcjach związków próchnicznych. Acta Agrophys. 2002;70: 83-90.

- [28] Ciećko Z, Harnisz M. Wpływ kompostów z osadów ściekowych na zawartość potasu, wapnia i magnezu w wybranych roślinach uprawnych. Cz I. Nawożenie w kształtowaniu środowiska. Zesz Probl Post Nauk Roln. 2002;484:77-86.
- [29] Tujaka A. Ocena możliwości przyrodniczego wykorzystania osadów ściekowych z wybranych oczyszczalni ścieków. Zesz Probl Post Nauk Roln. 2009;525:445-452.
- [30] Kiryłuk A. Ocena przydatności mieszanek traw i osadu ściekowego do biologicznej rekultywacji wysypiska odpadów komunalnych. II Międzynar Konfer Nauk-Tech: Rekultywacja terenów zdegradowanych. Szczecin: Wyd AR Szczecin; 2003:161-168.
- [31] Kutik J, Cincerova A, Dvorak M. Chloroplast ultrastructural development during the ontogeny of the second leaf of wheat under nitrogen deficiency. Photosynthetica (Prague). 1993;28:447-453.
- [32] Starck Z. Różnorodne funkcje węgla i azotu w roślinach. Kosmos. Probl Nauk Biol. 2006;55,2-3(271-272):243-257.
- [33] Olszewska M, Grzegorzczak S, Bałuch-Małecka A. Wymiana gazowa i indeks zieloności liści *Trifolium repens* uprawianej w mieszkach z *Festolium braunii* i *Lolium perenne* w zależności od zróżnicowanego nawożenia azotem. Łąkarstwo w Polsce. 2008;11:147-156.
- [34] Ciećko Z, Grzegorzewski K, Żołnowski A, Najmowicz T. Oddziaływanie nawożenia mineralnego na plonowanie i zawartość cukru w korzeniach oraz zawartość chlorofilu w liściach buraka cukrowego. Biuletyn Inst Hodowli i Aklimatyzacji Roślin. 2004;234:137-143.
- [35] Gębczyński P. Zmiany ilościowe wybranych składników chemicznych w procesie mrożenia i zamrażalniczego składowania głównych i bocznych róż brokuła. Acta Sci Pol Tech Alimentaria. 2003;2(1):31-39.
- [36] Maciorowski R, Wołoszyk C. Bezpośredni i następczy wpływ kompostów sporządzonych na bazie komunalnego osadu ściekowego na fotosyntezę rzepaku jarego i pszenicy ozimej. Cz I. Nawożenie w kształtowaniu środowiska. Zesz Probl Post Nauk Roln. 2003;494:273-286.
- [37] Kachel-Jakubowska M. Zawartość chlorofilu w nasionach rzepaku poddanych procesowi suszenia. Inż Roln. 2009;8(117):39-45.
- [38] Swędryńska D, Niewiadomska A, Klama J. Koncentracja chlorofilu w blaszkach liściowych kukurydzy i owsa jako wskaźnik żywotności roślin inokulowanych bakteriami z rodzaju *Azospirillum*. Ekol Tech. 2008;4:165-170.
- [39] Ciećko Z, Żołnowski A, Wyszowski M. Plonowanie i zawartość skrobi w bulwach ziemniaka w zależności od nawożenia NPK. Annales UMCS, Sec E. 2004;LIX (1):399-406.
- [40] Starck Z. Mechanizmy integracji procesów fotosyntezy i dystrybucji biomasy w niekorzystnych warunkach środowiska. Zesz Probl Post Nauk Roln. 2002;481:113-123.
- [41] Bielińska EJ. Charakterystyka ekologiczna gleb ogrodów działkowych z terenów zurbanizowanych. J Research and Application in Agricultural Engineering. 2006;51(2):13-16.
- [42] Sikorski ZE, editor. Chemiczne i funkcjonalne właściwości składników żywności. Warszawa: WNT; 1994:399-424.
- [43] Stankiewicz M, Wawrzyniak-Kulczyk M. Poznaj – zbadaj – chroń środowisko, w którym żyjesz. Warszawa: Wyd WSiP; 1997:16-26.
- [44] Olszewska M. Wpływ niedoboru magnezu na wskaźniki wymiany gazowej, indeks zieloności liści (SPAD) i plonowanie *Lolium perenne* i *Dactylis glomerata*. Łąkarstwo w Polsce. 2005;8:141-148.

WPLYW OSADU ŚCIEKOWEGO NA PRODUKCJĘ BIOMASY I ZAWARTOŚĆ MAKROSKŁADNIKÓW ORAZ CHLOROFILU W MIESZANKACH TRAW

Zakład Biologii Sanitarnej i Biotechnologii
Politechnika Białostocka

Abstrakt: Celem podjętych badań było określenie wpływu różnych dawek komunalnego osadu ściekowego na przyrost biomasy części nadziemnych, a także na zawartość makroskładników oraz chlorofilu w mieszkach traw gazonowych. Założono cztery doświadczenia na trawnikach wzdłuż głównych ciągów komunikacyjnych Białegostoku: przy ul. Popiełuszki, Hetmańskiej, Raginisa i Piastowskiej. Czynniki w doświadczeniu były trzy dawki osadu ściekowego: 0 (kontrola), 7,5 i 15 kg · m⁻² oraz dwie mieszanki traw gazonowych: Eko i Roadside. Próbkę części nadziemnych traw pobrano w trzech terminach: w czerwcu, sierpniu i październiku 2011 r., po czym oznaczono w nich: ilość biomasy części nadziemnych traw z 1 m²,

ogólne formy azotu, fosforu i potasu oraz określono zawartość chlorofilu *a* i *b*. Osad ściekowy wpłynął istotnie na akumulację N i K w trawach. Maksymalną średnią zawartość azotu (2,82 % s.m.) zaobserwowano w trawach z poletek z największą dawką osadu, z kolei potasu – w trawach z poletek z dawką osadu wynoszącą 7,5 kg · m⁻². Gromadzenie biomasy traw w okresie badań było różnicowane głównie przez ilość dawek osadu ściekowego oraz termin pobierania próbek. Średni sumaryczny plon suchej masy roślin dla trzech miesięcy badań (czerwiec, sierpień i październik) wynosił od 229,83 do 430,70 g · m⁻². Najlepsze efekty uzyskano przy zastosowaniu dawki osadu ściekowego wynoszącego 15 kg · m⁻². Termin zbioru oraz dawka osadu wpłynęły istotnie na zawartość chlorofilu *a* i *b* w badanych trawach. Średnie zawartości chlorofilu *a* w czerwcu, sierpniu i październiku wynosiły odpowiednio: 0,60; 0,64 i 0,54 mg · g⁻¹ ś.m., z kolei chlorofilu *b*: 1,00; 0,94; 0,39 mg · g⁻¹ ś.m. Stwierdzono niski średni stosunek chlorofilu *a* do *b* w badanych mieszankach traw, który wynosił od 0,54 w czerwcu do 1,82 w październiku dla mieszanki Eko oraz od 0,57 w czerwcu do 1,68 w październiku dla mieszanki Roadside. Rośliny z poletek kontrolnych wykazywały wyższy średni stosunek chlorofilu *a* do *b* w porównaniu do roślin z poletek użyźnionych osadem.

Słowa kluczowe: biomasa, trawy gazonowe, makroskładniki, chlorofil, osady ściekowe