

THE YIELD AND CONTENT OF TRACE ELEMENTS IN BIOMASS OF *MISCANTHUS SACCHARIFLORUS* (MAXIM.) HACK. AND IN SOIL IN THE THIRD YEAR OF A POT EXPERIMENT

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Abstract

In the third year of a pot experiment (in a greenhouse), the carryover influence of fertilization with fresh sewage sludge and, for comparison, mineral fertilization on the content and uptake of Fe, Mn, Mo, B, Ba, Sr, As, Sn, Li, and Ti by the biomass of *Miscanthus sacchariflorus* grass. The yield of silver-grass biomass was evaluated on the basis of two harvests (June and December). Trace elements in the tested grass as well as in the soil (after harvest) were determined by means of the ICP-AES technique after sample combustion in a muffle furnace. Significant influence of sewage sludge fertilization on yield of silver-grass biomass was found. Grass harvested in autumn contained higher contents of some analyzed trace elements (Fe, Mn, Sr, Ba, and Ti) than that harvested in summer. The uptake of trace elements uptake by the silver-grass biomass was higher from the objects fertilized with sewage sludge than the ones receiving mineral fertilization. After three years of cultivation, the soil contained more Fe, Mn, Ba, Sr, Ti, B, and Li as compared to the content before the experiment.

Key words: *Miscanthus sacchariflorus*, biomass, fertilization, sewage sludge, trace elements.

**PLON I ZAWARTOŚĆ PIERWIĄTKÓW ŚLADOWYCH W BIOMASIE TRAWY
MISCANTHUS SACCHARIFLORUS (MAXIM.) HACK ORAZ W GLEBIE,
W TRZECIM ROKU DOŚWIADCZENIA WAZONOWEGO**

Abstrakt

W trzecim roku doświadczenia wazonowego (w warunkach szklarni) badano wpływ następczego nawożenia świeżym osadem ściekowym oraz – dla porównania – nawożenia mineralnego na zawartość i pobranie Fe, Mn, Mo, B, Ba, Sr, As, Sn, Li i Ti przez biomasę trawy *Miscanthus sacchariflorus*. Plon biomasy określono na podstawie dwóch zbiorów miskanta (w czerwcu i grudniu). Pierwiastki śladowe w testowanej trawie oraz glebie (po zbiorze) oznaczono metodą ICP-AES, po mineralizacji próbek na sucho, w piecu muflowym. Stwierdzono istotny wpływ nawożenia osadem ściekowym na plonowanie biomasy miskanta cukrowego.

W trawie zebranej jesienią stwierdzono większą zawartość niektórych analizowanych pierwiastków (Fe, Mn, Sr, Ba i Ti) niż w trawie zebranej latem. Pobranie pierwiastków śladowych z plonem biomasy miskanta było większe z obiektów nawożonych osadem ściekowym niż z obiektu nawożonego nawozami mineralnymi. Po trzech latach uprawy stwierdzono w glebie większą zawartość Fe, Mn, Ba, Sr, Ti, B i Li, w porównaniu z zawartością przed założeniem eksperymentu.

Słowa kluczowe: *Miscanthus sacchariflorus*, biomasa, nawożenie, osad ściekowy, pierwiastki śladowe.

INTRODUCTION

Introduction of new, energy plants into cultivation is stimulated by progressing depletion of sustainable resources of fossil fuels in Poland and worldwide (EL BASSAM 1995). Among energy crops, grasses of *Miscanthus* genus are the most important ones (KOCHANOWSKA, GAMRAT 2007). Silver-grass (*Miscanthus sacchariflorus*) deserves special attention owing to its due modest soil requirements, possibility to grow it on soil amended with sewage sludge, tolerance to water deficit and growth under unfavorable habitat conditions, e.g. mine waste dumps or along roads and highways (MAJTKOWSKI, MAJTKOWSKA 1998, 2000, MALINOWSKA et al. 2006, KOCHANOWSKA, GAMRAT 2007, KALEMBASA, MALINOWSKA 2007a).

The research aimed at determining the biomass yield and total content of trace elements (Fe, Mn, Mo, B, Ba, Sr, As, Sn, Li, and Ti) in silver-grass (*Miscanthus sacchariflorus* (Maxim.) Hack) fertilized with three rates of fresh sewage sludge and mineral fertilizer (NPK) in the third years of a pot experiment.

MATERIAL AND METHODS

In spring 2005, a pot experiment was established in a greenhouse according to a completely randomized design with three replications. Pots of 20 l capacity each were filled with soil of heavy loamy sand granulometric composition (according to PTG), which was characterized by following properties: $\text{pH}_{\text{KCl}} = 6.60$, organic carbon content $30.5 \text{ g} \cdot \text{kg}^{-1}$, total content of some trace elements ($\text{mg} \cdot \text{kg}^{-1}$ soil): Fe – 5187; Mn – 145.8; Mo – 0.049; B – 0.673; Ba – 82.18; Sr – 29.06; As – 0.625; Sn – 0.663; Li – 1.70; Ti – 49.42. The content of organic carbon was determined by the oxidation-titrimetric method; total amounts of trace elements were analyzed applying the ICP-AES technique after soil sample combustion in a muffle furnace at 450°C . The influence of organic (fresh sewage sludge) and mineral fertilization on total content of trace elements in biomass of silver-grass (*Miscanthus sacchariflorus*) harvested on two dates (June and December 2007) in the third year of cultivation was studied.

The following objects were set up:

- control (with no fertilization);
- mineral NPK fertilization applied once before rhizomes set (mineral N was used as urea, whereas the N amount was calculated according to a dose of 20% fresh weight of sewage sludge in reference to the soil weight);
- 10% fresh weight of sewage sludge in relation to the soil weight, which corresponded to 20 g N pot^{-1} ;
- 20% fresh weight of sewage sludge in relation to the soil weight, i.e. 4 kg of fresh weight per pot, which corresponded to 40 g N pot^{-1} ;
- 30% fresh weight of sewage sludge in relation to the soil weight, i.e. 6 kg of fresh weight per pot, which corresponded to 60 g N pot^{-1} .

Fresh sewage sludge was used once (before sowing silver-grass) by mixing it with soil in pots. Phosphorus (triple superphosphate) and potassium fertilizers (potassium sulfate) were applied to all objects, but maintaining the N:P:K ratio at 1:0.8:1.2.

The plant material samples were combusted at 450°C in a muffle furnace for 15 hours. Then, 10 ml diluted HCl (1:1) was added to the crucibles and the content was evaporated to decompose carbonates and to separate the silicates. After adding another 5 ml 10% HCl, the content was passed through hard filter paper into a measuring flask (100 cm^3 capacity) and the volume was adjusted with distilled water. The solutions thus obtained were subjected to determinations of total content of Fe, Mn, Mo, B, Ba, Sr, As, Sn, Li, and Ti by the ICP-AES technique using an emission spectrophotometer combined with inductively-coupled plasma. After silver-grass harvest, soil samples were collected from particular fertilization objects and levels of the above trace elements were determined applying the ICP-AES technique.

Based on the harvested biomass yield and the content of trace elements, their uptake by silver-grass was calculated.

The results were statistically processed, and the differences between mean values for harvest dates, fertilization objects and their interactions were verified by means of variance analysis (FR Analvar 4.1 software). Significance of differences ($\text{NIR}_{0,05}$) was assessed using Tukey's test.

RESULTS AND DISCUSSION

In the third year of growing *Miscanthus sacchariflorus*, the biomass yield harvested in June ($266.9 \text{ g} \cdot \text{pot}^{-1}$) was twice as high as in December ($132.0 \text{ g} \cdot \text{pot}^{-1}$; Table 1), which resembled the results from the second year (KALEMBASA, MALINOWSKA 2007b). Significant differences in the silver-grass biomass yields were found in the third year after fresh sewage sludge application.

Total amounts of the examined trace elements in silver-grass biomass varied and depended on the harvest date and fertilization rate (Table 1).

Statistical analysis revealed significant influence of the harvest date on total content of most of the studied trace elements (except molybdenum and arsenic) in silver-grass biomass. About twice as much Fe, Mn, and Sr, as well as more Ba and Ti were found in the autumn (December) than in the summer harvest. In the second year of silver-grass cultivation, KALEMBASA, MALINOWSKA (2007b) also recorded higher levels of heavy metals in the biomass harvested in autumn as compared to summer. Bio-accumulation of Mn, Mo, and Sr apparently increased on both dates due to fresh sewage sludge fertilization, while for the other elements such dependence was not observed. Much more lithium was determined in the silver-grass biomass grown on not-fertilized and mineral-fertilized soil than in that harvested from the soil amended with sewage sludge. The content of an element in a soil environment does not always condition its uptake by a plant (GRZYWNOWICZ, STRUTYŃSKI 1999). Sewage sludge fertilization under *Miscanthus sacchariflorus* (even at the highest applied rate – 30%) did not cause excessive accumulation of the tested trace elements, which may have been complexed with organic matter, being therefore converted into forms that are unavailable for plants.

A much higher uptake of the analyzed elements (Table 2) was recorded in biomass of silver-grass growing on soil fertilized with sewage sludge, which was associated with the size of yields achieved from those objects. The largest amounts of the trace elements were assimilated by yield of grass fertilized with 30% and 20% FW sewage sludge rates; more with the summer than with the autumn harvest (except Fe and Mn). KRZYWY et al. (2004) reported that uptake of microelements by *Miscanthus giganteus* straw increased along with the increasing rates of sewage sludge applied.

Table 1

The content of trace elements (mg kg⁻¹) in the biomass *Miscanthus sacchariflorus* harvested in the third year of cultivation in a pot experiment

Fertilization	The yield (g pot ⁻¹)	Fe	Mn	Mo	B	Ba	Sr	As	Sn	Li	Ti
Summer harvest (I)											
Control object	113.3	81.9	16.6	0.41	5.70	3.57	11.3	3.09	2.02	39.9	1.09
NPK	140.0	76.9	15.2	0.43	2.40	3.13	13.6	2.95	2.77	67.6	1.19
10%	333.3	67.0	21.3	0.52	4.66	3.28	11.1	2.65	1.52	11.6	0.59
20%	373.3	92.2	36.1	1.28	6.38	3.76	16.8	2.58	2.67	14.5	1.06
30%	375.0	88.3	35.2	0.95	4.21	4.09	14.1	2.19	2.32	9.76	1.37
Mean	266.9	81.3	24.9	0.72	4.67	3.57	13.4	2.69	2.26	28.7	1.06
Autumn harvest (II)											
Control object	53.3	159.2	22.8	0.59	0.80	4.86	23.0	2.51	0.21	10.3	0.97
NPK	60.0	229.0	19.2	0.44	0.98	4.90	17.3	1.77	0.22	13.1	2.47
10%	140.0	257.0	65.5	0.82	1.12	4.59	30.4	2.81	0.26	4.05	1.67
20%	206.7	153.6	69.2	0.82	1.39	5.29	23.8	2.86	0.24	3.99	1.69
30%	200.0	170.6	67.6	0.88	1.41	5.51	26.9	2.72	0.29	4.25	2.05
Mean	132.0	193.9	48.9	0.71	1.14	5.03	24.3	2.53	0.25	7.14	1.77
NIR _{0.05} :											
A (terms of harvest)	68.0	10.4	1.84	n.i.	0.49	0.39	1.01	n.s.	0.10	4.60	0.22
B(fertilization)	154.3	23.5	4.17	0.38	1.12	n.s.	2.30	n.s.	0.23	10.4	0.50
A/B interaction	152.1	23.2	4.11	0.37	1.11	n.s.	2.26	n.s.	0.23	10.3	0.49
B/A interaction	218.2	33.3	5.89	0.54	1.59	n.s.	3.25	n.s.	0.33	14.8	0.71

n.s. – non-significant

10% of waste activated sludge

20% of waste activated sludge

30% of waste activated sludge

The chemical analysis of the soil collected after three years of silvergrass cultivation (Table 3) revealed increase in the content of Fe, Mn, Ba, Sr, and Ti and a slight increase in the level of B and Li caused by the applied fertilization, as compared to the amounts before the experiment.

Table 2

Uptake of other elements (mg pot⁻¹) by yield of *Miscanthus sacchariflorus* in the third year of cultivation in a pot experiment

Fertilization	Fe	Mn	Mo	B	Ba	Sr	As	Sn	Li	Ti
Summer harvest (I)										
Control object	9.28	1.88	0.05	0.65	0.40	1.28	0.35	0.23	4.52	0.12
NPK	10.76	2.13	0.06	0.34	0.44	1.90	0.41	0.39	9.46	0.17
10% of waste activated sludge	22.33	7.09	0.17	1.55	1.09	3.69	0.88	0.51	3.88	0.20
20% of waste activated sludge	34.42	13.48	0.48	2.38	1.40	6.28	0.96	0.99	5.42	0.40
30% of waste activated sludge	33.10	13.19	0.36	1.58	1.53	5.27	0.82	0.87	3.66	0.51
Mean (I)	21.95	7.55	0.22	1.30	0.97	3.68	0.69	0.60	5.39	0.28
Autumn harvest (II)										
Control object	8.44	1.21	0.03	0.04	0.26	1.23	0.13	0.01	0.55	0.52
NPK	13.74	1.15	0.03	0.06	0.29	1.04	0.11	0.01	0.79	0.15
10% of waste activated sludge	35.98	9.18	0.12	0.16	0.64	4.25	0.39	0.04	0.57	0.23
20% of waste activated sludge	31.75	14.31	0.17	0.29	1.09	4.92	0.59	0.05	0.83	0.35
30% of waste activated sludge	34.12	13.52	0.18	0.28	1.10	5.37	0.54	0.06	0.85	0.41
Mean (II)	24.81	7.87	0.10	0.17	0.68	3.36	0.35	0.03	0.72	0.24
Mean (I+II)	23.38	7.71	0.16	0.73	0.83	3.52	0.52	0.32	3.05	0.26

Table 3

The content of trace elements (mg kg⁻¹) in soil after the harvest of *Miscanthus sacchariflorus*, in the third year of cultivation in a pot experiment

Fertilization	Fe	Mn	Mo	B	Ba	Sr	As	Sn	Li	Ti
Control object	3867	111.4	0.06	0.67	44.25	18.99	0.19	0.46	1.13	28.89
NPK	3077	86.2	0.12	0.58	37.68	18.98	0.66	0.35	1.25	33.63
10% of waste activated sludge	4220	123.9	0.11	0.79	41.51	19.95	0.85	0.96	1.41	30.37
20% of waste activated sludge	4323	105.7	0.12	0.63	44.12	19.17	0.93	1.11	1.45	32.02
30% of waste activated sludge	4341	112.5	0.03	0.63	41.13	19.52	1.49	0.72	1.50	30.98
Mean	3966	107.9	0.09	0.66	41.74	19.32	0.82	0.72	1.35	31.18

CONCLUSIONS

1. Significantly higher biomass yields of silver-grass cultivated on sewage sludge fertilized soil were recorded in the third year of a pot experiment.

2. Fertilization with sewage sludge enhanced the accumulation of Mn, Mo, and Sr, while mineral fertilization raised the concentration of Li in silver-grass biomass, as compared to the control object.

3. Much higher content of some examined trace elements (Fe, Mn, Sr, Ba, and Ti) was found in the test plant biomass harvested in late autumn than in summer.

4. The uptake trace elements by the biomass yields of silver-grass was higher in the objects fertilized with sewage sludge at the highest (30%) and medium rates (20%) than by plants harvested from the control and mineral-fertilized objects.

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