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The use of algae in the removal of Cd and Cu in the process of wastewater recovery

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ABSTRACT

The research work concerned initiating metal biosorption by living algae by means of administering cadmium and copper. The mixed population of green algae used in the study was taken from a natural reservoir. The process was evaluated based on changes in metal concentrations in biomass when accurate doses of metals were spiked into the culture and for different reaction times. Changes of Cd(II) and Cu(II) concentrations in the culture medium were also analyzed. The study showed that mixed algal population was good biosorbent of Cu and Cd ions. It was observed that spiking of different doses of cadmium (II) and copper (II) considerably affected the effectiveness and efficiency of sorption. The use of similar concentrations of Cu(II) and Cd (II) ions during the experiments allowed one to compare the general effectiveness of sorption of both metals. The results at high contents of heavy metals (4.1 mg Cd/dm^3 and 4.3 mg Cu/dm^3) were better for copper. The effectiveness of the process was at the level of 70–85%, whereas for cadmium it was 40–70%.

Keywords: Biosorption; Cadmium; Copper; Algae

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1. Introduction

Heavy metals are more and more often found in natural waters in amounts which sometimes exceed acceptable concentrations. In water environment, heavy metals occur in a molecular form (e.g. colloids) or dissolved one (such as free ions, complex ions and chelated metals with organic and inorganic ligands) [1]. Dissolved forms of metals can be removed from water by means of physicochemical processes such as repelling free ammonia and precipitation with lime, coagulation, filtration, recarbonization, sorption and ion exchange [2]. However, heavy metals removal is not always effective enough. The process of recovery of waters polluted with compounds of many metals may also be difficult since the optimum conditions for the reduction of contents of particular metals may be different. The degree of heavy metal removal, e.g. in the process of precipitation, mostly depends on the pH of the water environment. The optimum value of this parameter differs from metal to metal. That is why after the recovery process, residual quantities of

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heavy metal ions may remain in the waters, which make it impossible to reuse the waters for industrial purposes. It is important that channel waters after the recovery process to the natural environment should comply with the requirements included in the Regulation of the Minister of Environment. The highest acceptable values of pollution indices for treated industrial wastes are as follows: max. 0.4 mg/dm^3 (mean daily value) and 0.2 mg/dm^3 (mean monthly value) for cadmium and 0.5 mg/dm^3 for copper [3]. Such low concentrations of heavy metal ions can be achieved, among others, using biological methods.

Biosorption of heavy metals is a process of their removal cooperation. Ions of heavy metals are either bound by functional groups of the cell wall or they penetrate into the cell [4]. The accumulation of metal ions in the cytoplasm is predominantly conditioned by the occurrence of living microorganisms. Metabolic energy, the activity of enzymatic systems and an efficient system of transport through the cytoplasmic membrane are necessary for the effective accumulation of metals by cells [5–7].

The process of biosorption involves several mechanisms responsible for the uptake of metal ions. These include particularly microprecipitation, chemisorption, complex reaction, chelation, and physical adsorption. These mechanisms may occur simultaneously with different efficiency. It depends on the kind of functional groups present in the algal cell wall and the physicochemical properties of water environment [7].

The biosorption process is highly effective. Its reaction time equals only several minutes, and it can concentrate metals in small quantities. Efficiency of the biosorption process depends on many factors. The most important of them are as follows: pH of the water environment, the concentration of heavy metals and the biosorbent, ionic strength, temperature, and the speed of mixing.

Microorganisms which are capable of carrying out a highly effective process of heavy metals biosorption are among other algae, available in practically unlimited quantities [8,9]. Certain species of these microorganisms have a high tolerance to the presence of heavy metal ions; algae also have a high surface area-to-volume ratio. Moreover, it has been proved that some differences in resistance to the toxicity of a given metal can occur within one species [10]. The use of algae to remove heavy metal compounds also allows for their recovery [9–11].

Using natural biosorbents to support water recovery processes is particularly worthwhile because of relatively low costs of their acquisition, high affinity to ions of many heavy metals and virtually non-limited number of binding sites [12,13].

2. Methodology of the research

2.1 Origin of the algae

The culture was a mixed population of algae (mainly *Chlorococcales* chlorophyta with the dominance of *Scenedesmus quadricauda*) obtained from the Poraj dam reservoir. Besides, the presence of bacteria and some amounts of tryptone were found.

The dam reservoir is located at 763.3 km of the Warta River (from the point where it joins the Oder). Apart from the Warta, streams such as Ordonówka, Złoty Stok, and Boży Stok flow into the reservoir. In the waters of the Poraj dam reservoir, massive algal bloom is observed in the summer every year. The blooms result in an increased photosynthesis process leading to alkalization of the environment, which promotes the precipitation of heavy metals from the depths to sediments [14]. Ions of heavy metals such as nickel(II) 15–59 mg/kg, cadmium(II) 1.5–2.3 mg/kg, and copper(II) 3.3–7.5 mg/kg were found in the sludge from the reservoir. In the mobile fractions of the sludge, cadmium(II) was predominant [15].

2.2 The culture medium

The culture medium was prepared in accordance with the applicable regulation (Commission Directive No. 92/69/EEC of 31/07/1992.). The composition of the basic solutions was as follows:

Solution I was NH_4Cl—1.5 g; $MgCl_2{\cdot}6H_2O$—1.2 g; $CaCl_2{\cdot}2H_2O$—1.8 g; $MgSO_4{\cdot}7H_2O$—1.5 g; KH_2PO_4—0.16 g.

Solution II was $FeCl_3{\cdot}6H_2O$—0.08 g; $Na_2EDTA{\cdot}2H_2O$—0.1 g.

Solution III was H_3BO_3—0.185 g; $MnCl_2{\cdot}4H_2O$—0.415 g; $ZnCl_2$—0.003 g; $CoCl_2{\cdot}6H_2O$—0.0015 g; $CuCl_2{\cdot}2H_2O$—0.00001 g; $Na_2MoO_4{\cdot}2H_2O$—0.007 g.

Solution IV was $NaHCO_3$—50 g.

The basic solutions were prepared as follows: The above-mentioned amounts of salts were weighed out to four volumetric flasks, complemented with distilled water up to 1 dm^3, then sterilized and stored at the temperature of 4°C in dark bottles.

The prepared solutions were used for the preparation of culture medium. 10 cm^3 of basic solution I was entered into a 1 dm^3 volumetric flask, and 1 cm^3 samples of basic solutions II, III, and IV were added; finally, it was complemented with deionized water up to the volume of 1 dm^3.

2.3. Determination metal ions in algal biomass

First, the algal biomass was mineralized (in accordance with PN—EN 14084:2004).

Weighed out dry mass samples of algae (0.168—0.265 g) were entered to 100 cm^3 test tubes. 10 cm^3 of chloroazotic acid was added to each tube prepared this way. Next the mineralization process was conducted in three successive temperature ranges: I—20 min at the temperature of 70˚C, II—30 min at the temperature of 100˚C, III—20 min at the temperature of 140˚C, using a VELP DK20 digestion unit. After cooling, the solutions were filtrated through qualitative filters, and then Cd(II) and Cu(II) ions were assayed with the use of atomic absorption spectrometry in flame in accordance with PN-81/C-04570/01 and with the use of NOVAA 400 spectrometer from Analityk Jena.

3. Procedure

3.1. Algal culture

The culture was carried out in a 120 dm^3 tank, at the temperature of 24˚C (±2˚C), lit with cool white fluorescent lights L36 W/840 in a continuous mode. After the introduction into the tank of the culture medium prepared accordingly, mixed algal culture acquired from the natural environment was entered into the culture. In order to keep the algae suspended, the cultures were slightly mixed. The culture pH was kept within the 7.5–7.8 range. The control of the process involved regular measurements of the number of algae (specimens) occurring in the culture medium with the use of a microscope and the Sedgewick–Rafter chamber. Before the experiment, the number of specimens in 1 cm^3 of the culture medium was 200,000.

3.2. Procedure of the experiments

50 cm^3 of the culture medium grafted with mixed algal culture including a known number of algae (10,000,000 specimens) was introduced into each of seven glass 100 cm^3 bioreactors. Subsequently, the solutions of cadmium and cuprum compounds in three different concentrations were introduced (Table 1). The lowest and mean concentrations approximated the highest acceptable values of pollution indices for treated industrial wastes [4].

The process was carried out for three reaction times: (1) immediate measurement, (2) after 1 h, and (3) after 24 h of the administration of heavy metal ions.

After the lapse of a specific incubation time, the whole content of bioreactors was centrifuged for 3 min at the speed of 3,000 rpm. The centrifuged algae were dried to solid mass in the temperature of 105˚C and then ground down in a mortar. Next, the obtained biomass samples were mineralized, and the contents of Cu(II) and Cd(II) were determined.

From each sample of the decanted culture medium obtained after the centrifugation, 20 cm^3 was taken through a qualitative filter and acidified with 1 cm^3 of concentrated HNO_3 to pH approx. 2. The samples were kept in the temperature of 4˚C until the moment of determination of Cu(II) and Cd(II) concentrations.

All the assays were carried out twice, achieving repeatable results. Only in the case of a handful of samples, the results were divergent, and the experiment had to be repeated.

4. Discussion of the results

In previous studies conducted by the author, it had been shown that a mixed population of chlorophyta (with the dominance of *Chlorococcales*, mainly *S. quadricauda*) from a natural water reservoir was a much more effective biosorbent of heavy metal ions than a pure culture of *Pseudokirchneriella subcapitata* algae bred in laboratory conditions. The research was conducted with the use of ions of cadmium (II) and copper (II), and it was found that the biodiversity of algae positively influenced the effectiveness of the process of removing ions of these metals [16].

The aim of the research was to determine the sorption capacity regarding Cu(II) and Cd(II) with participation of a mixed algal population, with the use of different amounts of metals and different reaction times.

The effectiveness of biosorption of cadmium (II) and copper (II) was controlled by determining

Table 1
Heavy metal concentrations used in the study

		Bioreactor						
Metal entered	Unit	1	2	3	4	5	6	7
$CdCl_2 \times H_2O$	mg Cd(II)/dm^3	0.07	0.82	4.10	–	–	–	Control sample
$CuCl_2$	mg Cu(II)/dm^3	–	–	–	0.08	0.86	4.30	

concentrations of heavy metal ions in the algal biomass and the culture medium after established incubation times. The process effectiveness was evaluated in relation to the control culture without any compounds of Cu(II) or Cd(II). Thanks to the same unit applied (mg/dm^3), the effectiveness of biosorption process was calculated on the basis of the concentration of the metal remaining in the culture medium, comparing it with the concentration introduced during the experiment.

Before entering ions of Cu(II) and Cd(II) into the culture medium, the content of these metals in the control culture was determined. It was found that the participation of Cd(II) in the algal biomass was 0.008 mg/g$_{d.m.}$, and in the medium, 0.006 mg/dm^3. As for the participation of Cu(II) in the algal biomass, it was 0.009 mg/g$_{d.m.}$, and in the medium, 0.007 mg/dm^3.

The presence of copper ions in algal biomass is justified since copper, which is a microelement, was a component of the culture medium. The content of ions of Cd(II) in the algal biomass and the medium indicates the pollution of the water environment from which the algae are taken with compounds of these metals. It should also be emphasized that concentrations of Cu(II) and Cd(II) ions occurring both in the biomass and in the culture medium are so low that their influence on the result of the experiment was regarded as negligible.

4.1. Changes of cadmium (II) concentration in algal biomass and culture medium

The results of study on the dynamics of removal of Cd (II) ions are presented in Table 2. During the experiment, clear tendencies concerning the increase or decrease in cadmium (II) ions concentration levels in time were observed, both in the biomass and in the culture medium. It was found that when introducing low concentrations of Cd(II), the biosorption process occurs differently in comparison when higher doses of this element are introduced. When applying the lowest dose of Cd(II), 0.07 mg/dm^3, gradual desorption of metal ions from the biomass to the medium was observed (Fig. 1). Applying higher doses of cadmium (II) ions, 0.82 and 4.1 mg/dm^3, ensured better and better effectiveness of the process with time (Figs. 2 and 3).

Introducing 0.07 mg/dm^3 of ions of cadmium (II) into the medium grafted with algae resulted in about 60% effectiveness (0.32 mg/g$_{d.m.}$) of the biosorption process in the initial phase of the study. The effectiveness of the process gradually decreased to approximately 50% (0.27 mg/g$_{d.m.}$) after 24 h of the study (Fig. 1). The highest effectiveness of the process of sorption of cadmium ions in biomass was observed at the application of 0.82 mg Cd(II)/dm^3. Taking into consideration the content of ions of this metal in culture medium at the immediate measurement (0.152 mg/dm^3), it was calculated that the algal biomass sorbed approx. 75% (2.09 mg/g$_{d.m.}$) of the entered Cd(II). After 24 h, the effectiveness of the process rose up to approx. 95% (3.01 mg/g$_{d.m.}$) (Fig. 2). At the application of 4.1 mg/dm^3 of ions of Cd(II), the effectiveness of the process was from 40% (6.80 mg/g$_{d.m.}$) in the initial phase of the experiment up to approx. 70% (10.4 mg/g$_{d.m.}$) after 24 h incubation (Fig. 3). It is supposed that higher concentrations of Cd(II) ions would cause a reduction of the process effectiveness.

The content of ions of cadmium which remained in the culture medium during the administration of 0.82 mg/dm^3 of Cd(II) is lower than the acceptable values of this element (daily average 0.4 mg/dm^3 and monthly average 0.2 mg/dm^3) for treated industrial waste. As for the result of removing higher cadmium concentrations (4.1 mg/dm^3) from the medium, it exceeds the acceptable values provided in the regulation [4], hence after the recovery process, the waters may not be discharged to the natural environment.

Table 2
The dynamics of changes in cadmium ions concentrations in algal biomass and in culture medium within 24 h exposure

Cadmium concentration entered (mg/dm^3)	Cadmium (II) concentrations in algae after the determined reaction times (mg/g$_{d.m.}$)			Cadmium (II) concentrations in culture medium after the determined reaction times (mg/dm^3)		
	Immediate measurement	1 h	24 h	Immediate measurement	1 h	24 h
0.07	0.32	0.31	0.27	–[a]	0.026	0.035
0.82	2.09	2.40	3.01	0.152	0.067	0.024
4.1	6.80	7.75	10.40	2.560	2.082	1.315

[a] Result below the limit of quantification.

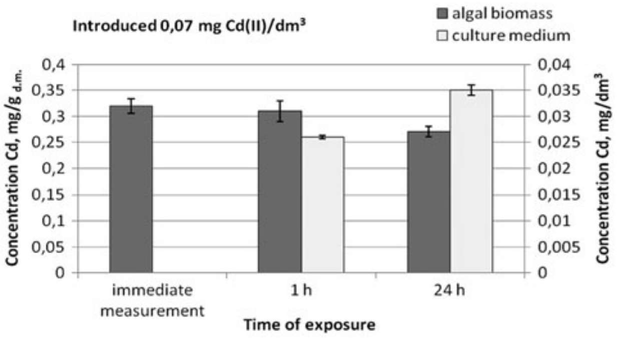

Fig. 1. Changes in the concentration of cadmium(II) in algal biomass (mg/g$_{\text{d.m.}}$) and in culture medium (mg/dm^3) depending on the time of exposure.

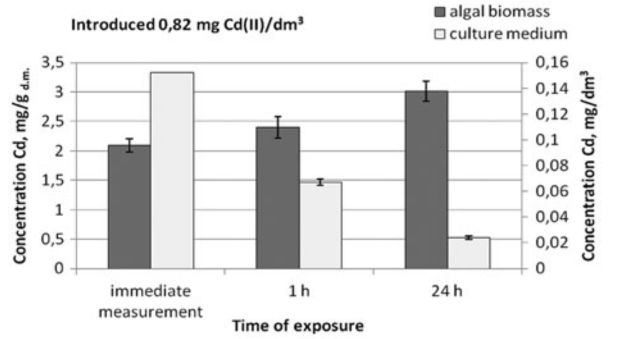

Fig. 2. Changes in the concentration of cadmium(II) in algal biomass (mg/g$_{\text{d.m.}}$) and in culture medium (mg/dm^3) depending on the time of exposure.

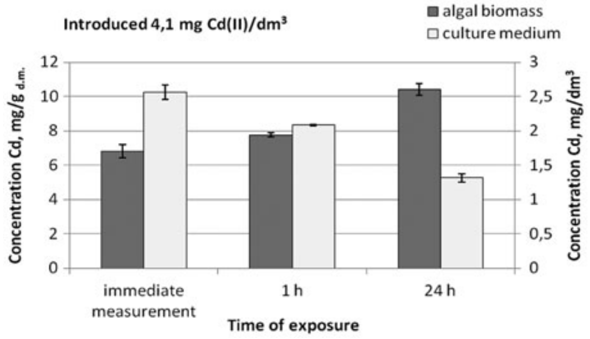

Fig. 3. Changes in the concentration of cadmium(II) in algal biomass (mg/g$_{\text{d.m.}}$) and in culture medium (mg/dm^3) depending on the time of exposure.

4.2. *Changes of copper (II) concentration in algal biomass and culture medium*

In the study focused on the dynamics of the copper (II) sorption process, a clear increase or decrease tendencies were not observed like in the case of cadmium (II) (Figs. 4 and 5). Only the highest introduced concentration of Cu(II) (4.3 mg/dm^3) resulted in an appropriate balance of the content of this element in the medium and the biomass (Fig. 6).

Administering low concentrations of copper (II) (0.08 mg/dm^3) did not cause any significant changes in the effectiveness of the biosorption process in time (Table 3). The immediate measurement of Cu(II) content in biomass was 0.29 mg/g$_{\text{d.m.}}$, over the first hour of the process, 0.32 mg/g$_{\text{d.m.}}$, and after 24 h exposure, 0.30 mg/g$_{\text{d.m.}}$ (Fig. 4).

When introducing 0.86 mg/dm^3 of copper (II), the contents of ions of this element in biomass observed at the immediate measurement (2.66 mg/g$_{\text{d.m.}}$) and over the 1 hour of the process (2.67 mg/g$_{\text{d.m.}}$) were comparable. Only after 24 h incubation, did the process efficiency rise by approximately 17% (3.23 mg/g$_{\text{d.m.}}$). However, taking into account the content of ions of copper (II) in the culture medium, it is estimated that the biomass absorbed approximately 80–95% of Cu(II) (Fig. 5). High effectiveness of the process, from approx. 70% (9.35 mg/g$_{\text{d.m.}}$) to 85% (11.49 mg/g$_{\text{d.m.}}$) was also observed when applying 4.3 mg/dm^3 Cu(II) (Fig. 6).

Table 3
The dynamics of changes in copper ions concentrations in algal biomass and in culture medium within 24 h exposure

Copper concentration entered (mg/dm^3)	Copper (II) concentrations in algae after the determined reaction times (mg/g$_{\text{d.m.}}$)			Copper (II) concentrations in culture medium after the determined reaction times (mg/dm^3)		
	Immediate measurement	1 h	24 h	Immediate measurement	1 h	24 h
0.08	0.29	0.32	0.30	–[a]	–[a]	–[a]
0.86	2.66	2.67	3.23	0.082	0.054	0.080
4.3	9.35	10.31	11.49	1.321	1.070	0.284

[a] Result below the limit of quantification.

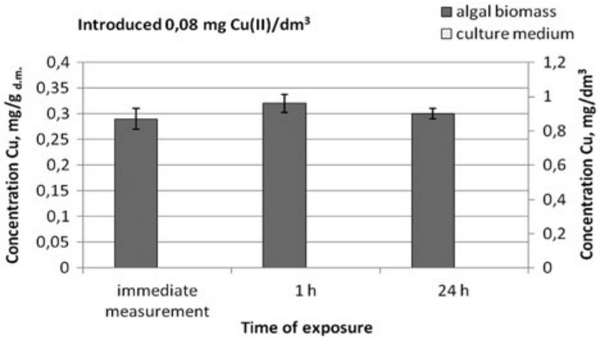

Fig. 4. Changes in the concentration of copper(II) in algal biomass (mg/$g_{d.m.}$) depending on the time of exposure.

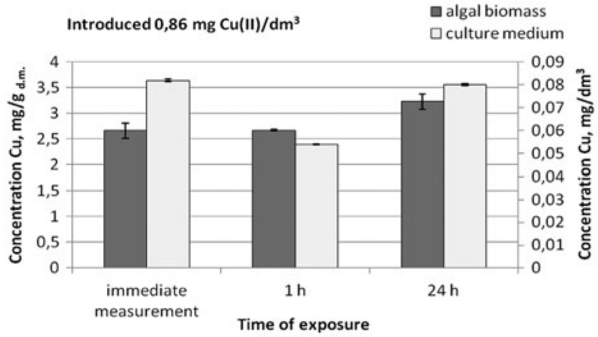

Fig. 5. Changes in the concentration of copper(II) in algal biomass (mg/$g_{d.m.}$) and in culture medium (mg/dm^3) depending on the time of exposure.

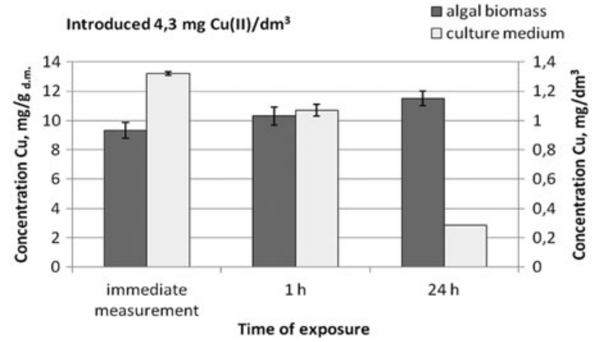

Fig. 6. Changes in the concentration of copper(II) in algal biomass (mg/$g_{d.m.}$) and in culture medium (mg/dm^3) depending on the time of exposure.

The effect of removing 0.86 mg/dm^3 of ions of copper previously introduced into the culture medium meets the highest acceptable value of copper concentration for treated industrial wastes (0.5 mg/dm^3). However, the administration of higher concentrations of copper (4.3 mg/dm^3) does not result in exceeding the acceptable values only in the case of 24 h incubation, when the effectiveness of Cu(II) ions from the medium is the highest.

5. Conclusions

The study showed that a mixed algal population was a good biosorbent of ions of copper and cadmium. It was observed that introducing different concentrations of cadmium (II) and copper (II) considerably affected the effectiveness and efficiency of sorption of those ions. Furthermore, the achieved degree of removal of heavy metal ions is considered very high: 95% for cadmium at the presence of 0.82 mg/dm^3 of Cd(II) ions and 80–95% for copper at the presence of 0.86 mg/dm^3 of Cu(II) ions. In nearly all cases, longer processing time led to the improvement of the efficiency removal. The only exception was the case with a low concentration of cadmium (0.07 mg/dm^3), when the tendency was the opposite. The use of similar concentrations of Cu(II) and Cd (II) ions during the experiments allowed one to compare the general effectiveness of sorption of both metals. The results at the administration of high contents of heavy metals (4.1 mg Cd/dm^3 and 4.3 mg Cu/dm^3) were better for copper. The effectiveness of the process was at the level of 70–85%, whereas for cadmium, it was 40–70%.

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References

[1] Y.N. Mata, M.L. Blázquez, A. Ballester, F. González, J.A. Muñoz, Characterization of the biosorption of cadmium, lead and copper with the brown alga *Fucus vesiculosus*, J. Hazard. Mater. 158 (2008) 316–323.

[2] M. Świderska-Bróż, Removal of Heavy Metals Contained in the Wastewater from Glassmaking Industry, Communication of Wrocław University of Technology Institute of Environmental Protection Engineering, 1981.

[3] Regulation of the Minister of Environment of 24 July 2006 on the criteria that have to be met when disposing of sewage to water or ground and on the substances that are particularly detrimental to the water environment (in Polish).

[4] B. Volesky, Sorption and Biosorption, BV Sorbex, Montreal-St. Lambert, Canada, 2003.

[5] V.K. Gupta, A.K. Shrivastava, J. Neeraj, Biosorption of chromium (VI) from aqueous solutions by green algae *Spirogyra* species, Water Res. 35 (2001) 4079–4085.

[6] E. Fourest, J.C. Roux, Heavy metal biosorption by fungal mycelial by-products: Mechanism and influence of pH, Appl. Microbiol. Biotechnol. 37 (1992) 399–403.

[7] B. Volesky, Detoxification of metal-bearing effluents: Biosorption for the next century, Hydrometallurgy 59 (2001) 203–216.

[8] E. Romera, F. González, A. Ballester, M.L. Blázquez, J.A. Muñoz, Comparative study of biosorption of heavy metals using different types of alga, Bioresour. Technol. 98 (2007) 3344–3353.

[9] S. Klimmek, H.J. Stan, A. Wilke, Comparative analysis of the biosorption of cadmium, lead, nikel and zinc by alga, Environ. Sci. Technol. 35 (2001) 4283–4288.

[10] K. Chojnacka, Biosorption and Bioaccumulation in Practice, Nova Science Publishers, New York, NY, 2009.

[11] K. Chojnacka, Biosorption and bioaccumulation the prospects for practical applications, Environ. Int. 36 (2010) 299–307.

[12] M. Rajfur, A. Kłos, M. Wacławek, Sorption of copper (II) ions in the biomass of alga *Spirogyra* sp, Bioelectrochemistry 87 (2012) 65–70.

[13] A. Fargasova, Combined effects of Mn(II), Mo(VI), Ni (II), Cu(I), Cu(II) and V(V) on freshwater algae *Scenedesmus quadricauda* Breb. Strain greifswald 15 and benthic larvae of *Chironomus plumosus*, Chemia i Inżynieria Ekologiczna 7(10) (2000) 1011–1021.

[14] M. Bogdalski, W. Sułkowski, The impact of the dam reservoir “Poraj” on the cleanness of the Warta River, Treatment, recovery and protection of waters, Publishing House of Częstochowa University of Technology 22 (1998) 169–177 (in Polish).

[15] A. Rosińska, L. Dąbrowska, PCBs and heavy metals in bottom sediments of the dam reservoir Poraj, Eng. Environ. Prot. 11 (2008) 455–469 (in Polish).

[16] K. Kipigroch, M. Janosz-Rajczyk, R. Mosakowska, Sorption of copper (II) and cadmium (II) ions with the use of algae, Desalin. Water Treat. 52 (2014) 3987–3992.