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FORMATION OF THE ACTIVATED SLUDGE BIOCENOSIS DURING LANDFILL LEACHATE PRE-TREATMENT IN SBR

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Abstract: It has been proved that at the established SBR technological regime, 74–71% effectiveness of the removal of impurities expressed as COD (in the influent 955–1059 mg $O_2 \cdot dm^{-3}$, respectively) can be obtained at *Bx* 0.40–0.45 mg $COD \cdot mg^{-1} \cdot d^{-1}$. Ammonium nitrogen, in the concentration of up to 292 mg NH_4-$N \cdot dm^{-3}$ was removed in 93% at *Bx* 0.40–0.96 mg $COD \cdot mg^{-1} \cdot d^{-1}$. The simultaneous denitrification described by 93% effectiveness of total nitrogen removal occurred, too. Activated sludge had the attributes of an inengaged sludge in leachates pretreatment at *Bx* between 0.40–0.45 mg $COD \cdot mg^{-1} \cdot d^{-1}$. Its biocenosis consisted of zoogleal and filamentous bacteria, protozoa *Mastigota* nd., *Difflugia* nd., *Aspidisca* sp., *Lionotus* sp., *Oxytricha* sp., *Opercularia* sp., *Tokophrya* sp. and rotifera. At the critical values of *Bx* (0.96–1.64 mg $COD \cdot mg^{-1} \cdot d^{-1}$), when leachates pretreatment effectiveness sharply dropped, biocenosis of activated sludge consisted only of zoogleal and filamentous bacteria, hyphae fungi, *Mastigota* nd. and *Opercularia* sp.

INTRODUCTION

Landfilling is an important waste management technology aimed at protecting the biosphere from harmful influences. A total of 566 municipal landfill sites with organized leachate management are in operation in Poland [21]. Both a high content of toxic substances and the presence of numerous chemical and microbiological impurities make them a considerable threat to the environment as well as health and epidemiological hazard. Studies conducted at the Institute of Environmental Engineering Systems, Warsaw University of Technology, since 2003 showed that psychrophilic heterotrophic bacteria dominate quantitatively in leachates while spore forming bacteria and potentially pathogenic species of microscopic fungi occur in smaller numbers. The total number of mesophilic bacteria and thermotolerant coli form bacteria, the bacteria *Clostridium perfringens* and *Listeria monocytogenes*, are indicators of sanitary contamination of leachates [5]. As demonstrated by the U.S. Environmental Protection Agency, faecal coliform bacteria and *Streptococcus faecalis* occur in leachates from municipal landfills [21]. Thus, reliable methods of preventing the air and the ground-water environment in the vicinity of landfills from chemical and microbiological impurities must be developed to protect the environment. To achieve this, the degree of chemical and microbiological leachate contamination must be assessed and the efficiency of contaminant removal at individual unit treatment process must be evaluated. Treatment methods of the landfill leachates may be divided into physicochemical and biological or a combination of both. Main physicochemical methods used in landfill leachate treatment include adsorption on active carbon, coagulation/flocculation, membrane processes and advanced oxidation (O_3, O_3/H_2O_2, Fenton's reaction, O_3/UV, O_3/H_2O_2/UV, UV/TiO_2). The molecular weight of complex organic compounds decreases in these processes, which enhances their susceptibility to biodegradation; however, full mineralization of organic substances occurring in leachates is not obtained. A greater efficiency is achieved with combined biological and physicochemical methods such as nitrification/denitrification/O_3/UV, nitrification/denitrification/precipitation/O_3, precipitation/membrane reactor/reversed osmosis, biological pre-treatment/TiO_2/UV/biological oxidation, activated sludge/electron-beam radiation [3, 18, 20, 21, 24].

The efficiency of biological methods is dependent mostly on the chemical composition of leachates, including the content of organic compounds and their susceptibility to degradation defined by the COD/BOD_5 ratio, the content of nitrogen, phosphorus and heavy metals. This is in turn influenced by the landfill age. Therefore, biological methods are mostly suitable for treatment of leachates from young landfills in which organic compounds occur as easily biodegradable compounds. As conventional technologies of the activated sludge and biological filters do not guarantee a full elimination of chemical and biological contaminants even in the case of leachates from young landfills, much attention has lately been paid to the use of the activated sludge method in SBRs. This technology is based on a long-term supply of leachate to reactor under alternating aerobic and anoxic conditions in the fill phase. It provides a high efficiency of the removal of carbon, nitrogen and phosphorus compounds while sludge bulking is eliminated. The advantages of this method are the small size of SBRs which can be then located near landfills and the possibility of the installation of a cover protecting the soil and the air from contamination by pathogenic microorganisms occurring in leachates. Studies on the practical application of this technology to treat leachates were conducted by, for instance, Dimadopoulus *et al.* [4], Surmacz-Górska *et al.* [19], Loukidou and Zouboulis [13], Klimiuk and Koc-Jurczyk [7], Klimiuk and Kuligowska [8, 9], Neczaj *et al.* [15], Kuligowska *et al.* [11, 12], and Zhou *et al.* [27].

The aim of this study was to determine the influence of an SBR load of chemical contaminants defined by COD on the formation of the activated sludge biocenosis. The results will be used to assess the SBR technology for the sustainable development of the activated sludge biocenosis that guarantees a high efficiency of the removal of leachate contaminants.

SCOPE OF THE STUDY

The technological parameters optimized in preliminary studies conducted within M. Szyłak-Szydłowski's PhD project [22] were used in leachate pre-treatment. The following were determined during the process control:

- sludge concentration (d.m.) in the reactor during the mixing and aeration phases,
- sludge volume index,
- in the leachates entering to and flowing out from the reactor: pH, COD_{Cr}, ammonia nitrogen (NH_4-N), nitrite nitrogen (NO_2-N), nitrate nitrogen (NO_3-N), total Kjeldahl nitrogen and total phosphate phosphorus,
- number of microorganism groups constituting the biocenosis of the activated sludge.

MATERIALS AND METHODS

Sampling

Leachates delivered to the collection well located near the landfill site designed for waste other than inert and hazardous waste, so-called municipal waste, in the south-eastern part of the town of Otwock via a drainage system were examined. The site has been in use since 1998, and its operation will be discontinued in 2012. Municipal wastes containing a high amount of biodegradable organic matter exceeding 20 $Mg \cdot d^{-1}$ are deposited in the site. The estimated site capacity is $12 \cdot 10^5$ Mg; only $2.7 \cdot 10^5$ Mg has been used so far. The landfill is lined with a 2 mm PEHD geomembrane. Wastes are stored in districts on plots, whose thickness is 1.5–2 m, and the lining thickness is 0.15 m. Leachate waters and landfill gas quantities in the landfill as well as surface and underground waters near the landfill are monitored. The leachates were characterized by COD 3080 mg $O_2 \cdot dm^{-3}$, total organic carbon 1.254 mg $C \cdot dm^{-3}$, BOD_5/COD – 0.181, SO_4^{2-} – 160 mg $SO_4^{2-} \cdot dm^{-3}$, $SO_4^{2-} \cdot Cl^-$ – 0.074, 0.163 mg $Cu \cdot dm^{-3}$, 2.065 mg $Zn \cdot dm^{-3}$, 0.207 mg $Pb \cdot dm^{-3}$, 0.017 mg $Cd \cdot dm^{-3}$, 0.216 mg $Cr \cdot dm^{-3}$, 0.003 mg $Hg \cdot dm^{-3}$, electrolytic conductivity 19.5 $mS \cdot cm^{-1}$ and pH 7.5 [5].

Leachate pre-treatment was conducted in an SBR with the working volume 6.9 dm^3, equipped with a mixer and the fine bubble aeration system that supplies oxygen concentration of 2 mg $O_2 \cdot dm^{-3}$. The activated sludge from the municipal waste treatment plant in Piaseczno near Warsaw was used as the seed of the reactor. The sludge concentration in the reactor remained in the range of 3–4 $g \cdot dm^{-3}$, and the sludge age (*0x*) at 12.5 d. The hydraulic retention time (*HRT*) was 16 hours, and the sludge load (*Bx*) ranged from 0.40 to 1.64 mg $COD \cdot mg^{-1} \cdot d^{-1}$, obtained by an increase in the percentage participation of leachates (5, 10, 15, 20 and 30%) in the mixture with synthetic wastewaters prepared according to the recipe given by Klimiuk and Wojnowska-Baryła [10].

The system worked in three 8-hour cycles per day. Each cycle consisted of a 45-minute fill phase, 30-minute mixing phase, 2.10 hour aeration phase, 45 minute mixing phase, 1.50 hour aeration phase, 1.30 hour settling phase and 30 minute decanting phase (including 25 minutes of decanting and 5 minute idling).

These parameters were "optimized" in so-called monitoring examinations (first stage of experiments), where the efficiency of impurities removal from the mixture of leachates and sewage at different duration time of each cycle phases was determined. These experiments are wider described in Szyłak-Szydłowski doctoral thesis.

Control examinations of the process in the ranges given above were conducted after two-weeks when *Bx* changed.

Chemical determinations

Chemical oxygen demand (COD) was determined with the dichromate method as given in the standard PN-74/C-04578.03. Mineral nitrogen forms were determined with spectrophotometric methods, ammonia nitrogen (NH_4-N) (direct nesslerisation) as given in PN-C-04576-4:1994, nitrite nitrogen (NO_2-N) as given in PN-EN 26777:1999, and nitrate nitrogen (NO_3-N) as given in PN-82/C-04576.08. Total Kjeldahl nitrogen was determined with the titration method as given in PN-73/C-04576.12. Phosphate phosphorus was determined with the spectrophotometric method (molybdenum method) as given in PN-91/C-04537/09. The sludge dry mass was determined with the weight method as given in PN-EN827.

Biological determinations

Microscopic observations of the activated sludge biocenosis were conducted with an Opton contrast-phase microscope. The determinations are given as the frequency of individual group of microorganism per cm^3 of the sludge.

RESULTS AND DISCUSSION

Examinations were conducted in two stages. The first stage, recognized as preliminary, so-called monitoring, was purposed of determination of duration time and arrangement of particular cycle phases in SBR, providing effectiveness of contaminations removal, with set sludge load value, expressed by COD (see materials and methods). The second stage, whose results are presented below, constituted so-called proper examinations, heading for defining influence of sludge load (*Bx*) on effectiveness of impurities removal, with arrangement and duration of particular stages of cycle in SBR, set in the first stage of examinations.

The studies showed that COD equal 955 mg $O_2 \cdot dm^{-3}$ on average in the influent was removed in 74%, and ammonia nitrogen characterized by the initial concentration 61 mg NH_4-$N \cdot dm^{-3}$ in 99% at *Bx* 0.40 mg $COD \cdot mg^{-1} \cdot d^{-1}$ obtained at the 5% leachate content in the mixture containing synthetic wastewaters. Rapid nitrification with the production of nitrates also occurred as the concentration of these compounds in the effluent wastewaters increased to 26.7 mg NO_3-$N \cdot dm^{-3}$. A significant reduction in the total nitrogen concentration from 106 to 33 mg $N_{tot} \cdot dm^{-3}$ indicated the occurrence of simultaneous denitrification. A low efficiency of phosphorus removal (only 26%, at the initial P-PO_4 concentration of 14.3 mg P-$PO_4 \cdot dm^{-3}$) observed at this sludge load should be noticed (Tab. 1).

A *Bx* increase to 0.45 mg $COD \cdot mg^{-1} \cdot d^{-1}$ obtained by a 10% addition of leachates to synthetic wastewaters resulted in a COD increase in the influent to 1059 mg $O_2 \cdot dm^{-3}$, ammonia nitrogen to 104 mg NH_4-$N \cdot dm^{-3}$ and nitrite nitrogen to 0.590 mg NO_2-$N \cdot dm^{-3}$. Consequently, total nitrogen in the influent reached 145 mg $N_{tot} \cdot dm^{-3}$. Concentrations of the other contaminants in wastewaters did not change significantly. Under these conditions, the removal efficiency of contamination defined by COD and ammonia nitrogen was still high (71 and 97%, respectively), and nitrification (NO_3-N was 15.8 mg NO_3-$N \cdot dm^{-3}$ in the effluent) and simultaneous denitrification (a decrease in total nitrogen concentration from 145 to 23 mg $N_{tot} \cdot dm^{-3}$) also occurred. The efficiency of phosphorus removal fell down was 27%.

A significant decrease in the removal efficiency of contamination defined by COD to only 45% was observed when *Bx* increased to 0.69 mg $COD \cdot mg^{-1} \cdot d^{-1}$ at the 15% addition of leachates to the mixture containing synthetic wastewaters. Influent COD was 1613 mg $O_2 \cdot dm^{-3}$, and the concentrations of individual nitrogen forms were: 202 mg NH_4-$N \cdot dm^{-3}$, 0.270 mg NO_2-$N \cdot dm^{-3}$ and 0.20 mg NO_3-$N \cdot dm^{-3}$. The efficiency of ammonia nitrogen removal was still high (95%) due to nitrification. Simultaneous denitrification of nitrates also occurred as indicated by the 82% removal of total nitrogen; a drop in the phosphorus content was slight (16%) (Tab. 1).

An increase in *Bx* to 0.96 mg $COD \cdot mg^{-1} \cdot d^{-1}$ following an increase in the percentage participation of leachates (20%) in the mixture containing synthetic wastewaters resulted in an increase in the influent COD to 2069 mg $O_2 \cdot dm^{-3}$ and nitrogen forms to 292 mg NH_4-$N \cdot dm^{-3}$, 1.07 mg NO_3-$N \cdot dm^{-3}$. Nitrite nitrogen concentration was 0.100 mg NO_2-$N \cdot dm^{-3}$.

Table 1. Effectiveness of the landfill leachates pretreatment in SBR depending on impurities sludge loading (mean values)

Participation of leachates in the mixture with synthetic wastewaters [%]	Sludge loading, [mg COD·mg^{-1}·d^{-1}]	Sample Parameter	COD [mg O_2·dm^{-3}]	$N\text{-}NH_4$ [mg·dm^{-3}]	$N\text{-}NO_2$ [mg·dm^{-3}]	$N\text{-}NO_3$ [mg·dm^{-3}]	N_{org} [mg·dm^{-3}]	$N_{Kjeldahl}$ [mg·dm^{-3}]	N_{tot} [mg·dm^{-3}]	$P\text{-}PO_4$ [mg·dm^{-3}]
5	0.40	influent	955	61	0.030	0.69	44	106	106	14.3
		effluent	249	0.69	0.030	26.7	8	9.2	33	10.7
		% removal	74	99	–	–	81	91	69	25
10	0.45	influent	1059	104	0.590	0.57	41	145	145	13.3
		effluent	305	3.50	0.290	15.8	4	8	23	9.7
		% removal	71	97	–	–	90	94	84	27
15	0.69	influent	1613	202	0.270	0.20	36	224	223	11.3
		effluent	871	10.7	1.400	15.9	11	21	39	9.4
		% removal	46	95	–	–	69	91	82	16
20	0.96	influent	2069	292	0.100	1.07	43	335	336	17.8
		effluent	1246	9.60	0.050	7.80	5	15	23	14.5
		% removal	40	97	–	–	88	96	93	18
30	1.64	influent	3406	351	1.040	4.67	38	389	395	20.3
		effluent	2879	52.1	0.070	1.90	15	68	70	14.8
		% removal	15	85	–	–	60	82	82	26

The removal efficiency of contamination expressed in COD was low (40%) at the high efficiency of ammonia nitrogen removal (97%). A significantly smaller increase in the amount of nitrates (to 7.80 mg NO_3-N·dm^{-3}) indicated an occurrence of nitrification and denitrification processes, proceeded with high intensity. A high decrease of total nitrogen (93%) indicated an intensive process of simultaneous denitrification. Phosphorus removal was still not very efficient (17%).

A sudden drop in the efficiency of pre-treatment of leachates constituting 30% in the mixture with synthetic wastewaters (influent COD 3406 mg O_2·dm^{-3}, 351 mg NH_4-N·dm^{-3}, 1.040 mg NO_2-N·dm^{-3} and 4.67 mg NO_3-N·dm^{-3}) occurred at *Bx* of 1.64 mg COD·mg^{-1}·d^{-1}. Under these conditions, the efficiency of COD removal was 15% and the process of nitrification was inhibited, as indicated by high concentration of NH_4-N in the effluent. However, an 85% removal of ammonia nitrogen took place and the process of simultaneous denitrification occurred (total nitrogen elimination in 82%). A significant phosphorus removal from the wastewaters delivered to the reactor no longer occurred (26%).

In conclusion, the technological examinations showed that leachate pre-treatment may be conducted with a highly efficient removal of carbon compounds expressed as COD only at *Bx* of 0.40 to 0.45 mg COD·mg^{-1}·d^{-1} obtained by adding 5–10% leachates to synthetic wastewaters imitating municipal wastewaters. For this range of *Bx*, the influent COD was between 955 and 1059 mg O_2·dm^{-3}, and the elimination of the contamination described by it was 74–71%, respectively. Thus it significantly exceeded the efficiency obtained by Kuligowska *et al.* [11] equal 87.7; 79.3 and 70.9% for the influent COD equal 570, 580 and 612 mg O_2·dm^{-3}, respectively. A similar removal efficiency of contamination expressed as COD (85.5–79.3%, at the concentration in the influent equal 1090 mg O_2·dm^{-3}) was also obtained by Diamadopoulos *et al.* [4] in the process conducted for the aeration phase of 20–14 hours, respectively, and also by Kuligowska *et al.* [12] (83–77% – concentration in the influent 1380 mg O_2·dm^{-3}) at the sludge age 33–11 d, HRT 12–2 hours and an 18 hour aeration phase, respectively. According to Zhou *et al.* [27], the high sludge age (25 d) and the long aeration phase in the cycle (18 hours) encourage a high efficiency of the treatment process. Under these conditions, these authors obtained a 94% elimination of COD with the concentration of 5077 mg O_2·dm^{-3} and a 99% elimination of ammonia nitrogen with the concentration of 728 mg NH_4-N·dm^{-3}.

In the present examinations, a significant degree of ammonia nitrogen removal from the wastewaters was observed up to 20% leachate content in the mixture with synthetic wastewaters (mean concentration in the influent 292 mg NH_4-N·dm^{-3}, mean removal efficiency 97%). Therefore, a greater removal efficiency of this form of mineral nitrogen was obtained than in the wastewater treatment process conducted by Diamadopoulos *et al.* [4] and Kuligowska *et al.* [11]. The former obtained a 49–35% efficiency of NH_4-N removal (concentration in the influent 107 mg NH_4-N·dm^{-3}) for the aeration stage 20–14 hours, respectively, and the latter a 99.8–99.9% elimination of NH_4-N whose concentration equaled 60–130 mg NH_4-N·dm^{-3}.

The present studies show that the process of simultaneous denitrification indicated by a simultaneous considerable drop in the concentration of total nitrogen in the effluent (mean 93%) also occurred in the SBR up to 292 mg N-NH_4·dm^{-3} in the delivered wastewaters. The nitrification process of phase I and II occurred with significant intensity in the aeration phase up to Bx = 0.69 mg COD·mg^{-1}·d^{-1}, at which the influent COD was 1613 mg O_2·dm^{-3}. This is consistent with previous studies by Surmacz-Górska *et al.* [20], who observed that nitrification occurred without significant disturbances for COD values in the influent ranging between 1000 and 2000 mg $O_2 \cdot dm^{-3}$ in the SBR. When COD concentration increases, its hardly biodegradable part inhibits nitrification, and the authors recommend a longer wastewater aeration time to encourage its degradation. A significant removal of phosphorus was not observed during the pre-treatment process. Its amount in effluent wastewaters was 9.7–14.8 mg $P\text{-}PO_4 \cdot dm^{-3}$ for the load range examined in the study. Therefore, further treatment of effluent wastewaters with chemical methods must be conducted to reduce the phosphorus concentration to the level that does not cause eutrophication of water reservoirs receiving treated wastewaters.

Significant changes in the abundance and generic composition of microorganisms constituting the sludge biocenosis also occurred during leachate pre-treatment conducted at various values of *Bx* (Fig. 1). The activated sludge used to inoculation of SBR showed the properties of well worked out sludge in treating the municipal wastewaters as it was characterized by well formed brownish flocs in the number $208 \cdot 10^3 \cdot cm^{-3}$, with visible numerous aggregations of zoogloeal bacteria ($2.4 \cdot 10^3 \cdot cm^{-3}$) and typical colonies of *Zoogloea ramigera* ($12.6 \cdot 10^3 \cdot cm^{-3}$) or filamentous bacteria ($11.4 \cdot 10^3 \cdot cm^{-3}$). The microfauna consisted of 6 taxa belonging to the flagellate group of protozoa ($8.4 \cdot 10^3 \cdot cm^{-3}$), free-swimming protozoa of the genus *Aspidisca* ($4.2 \cdot 10^3 \cdot cm^{-3}$), *Lionotus* sp. ($4.8 \cdot 10^3 \cdot cm^{-3}$) and *Oxytricha* ($0.6 \cdot 10^3 \cdot cm^{-3}$), attached protozoa of the genus *Vorticella* and rotifers ($7.2 \cdot 10^3 \cdot cm^{-3}$).

After the 7-day period of leachates dosage in the 5% quantitative ratio to municipal wastewaters (mean influent COD 955 mg $O_2 \cdot dm^{-3}$) in the amount ensuring *Bx* at 0.40 $COD \cdot mg^{-1} \cdot d^{-1}$, sludge flocs became light brown, big, merged, settling well (sludge index 102 $cm^3 \cdot g^{-1}$), with numerous aggregations of zoogloeal bacteria ($95.4 \cdot 10^3 \cdot cm^{-3}$) and typical colonies of *Zoogloea ramigera* ($4.2 \cdot 10^3 \cdot cm^{-3}$) and filamentous bacteria ($18 \cdot 10^3 \cdot cm^{-3}$). The microfauna consisted of 5 taxa of protozoa (*Mastigota* nd., *Aspidisca* sp., *Oxytricha* sp., *Opercularia* sp., shelled amoebae) and rotifers ($3.0 \cdot 10^3 \cdot cm^{-3}$). The genera *Aspidisca* ($4.8 \cdot 10^3 \cdot cm^{-3}$) and *Opercularia* ($13.8 \cdot 10^3 \cdot cm^{-3}$) dominated in the group of protozoa. The continuation of the pre-treatment process at the above reactor load caused a decrease in the number of zoogloeal aggregations of bacteria (to $12.2 \cdot 10^3 \cdot cm^{-3}$), protozoa of the flagellate group and belonging to the genus *Aspidisca* (up to $0.6 \cdot 10^3 \cdot cm^{-3}$) and a significant increase in the number of *Opercularia* sp. (up to $33 \cdot 10^3 \cdot cm^{-3}$) in the sludge. Free-swimming protozoa of the genus *Lionotus* ($0.6 \cdot 10^3 \cdot cm^{-3}$) and shelled amoebae ($1.2 \cdot 10^3 \cdot cm^{-3}$) also occurred (Tab. 2). The total number of protozoa in the activated sludge was thus similar to that recognized as the maximum number typical for well working activated sludge ranging from 10^3 to $5 \cdot 10^4 \cdot cm^{-3}$ [1, 14].

An increase in the sludge load to 0.45 $COD \cdot mg^{-1} \cdot d^{-1}$ by adding 10% leachates to synthetic wastewaters caused flocs dispersion and a drop in their size but it did not deteriorate sludge settling properties (sludge index was 96 $cm^3 \cdot g^{-1}$). The number of zoogloeal bacterial aggregations was similar to that recorded for the load of 0.40 mg $COD \cdot mg^{-1} \cdot d^{-1}$ ($23.4 \cdot 10^3$ – $72 \cdot 10^3 \cdot cm^{-3}$); *Zoogloea ramigera*, however, disappeared and the number of *Opercularia* sp. decreased to $2.8 \cdot 10^3$ – $4.2 \cdot 10^3 \cdot cm^{-3}$. The adverse influence of the sludge on the microfauna in the wastewaters dosaged to the reactor was indicated by an increase in the number of *Mastigota* nd. ($2.1 \cdot 10^3 \cdot cm^{-3}$), the disappearance of shelled amoebae, protozoa of the genera *Lionotus* and *Oxytricha*, at the end of the study period – as well as *Aspidisca* sp. and rotifers and the appearance of telotrochs deattached from the stalks of attached ciliates. Attached ciliates of the genus *Tokophrya* sp. ($0.6 \cdot 10^3 \cdot cm^{-3}$) occurred

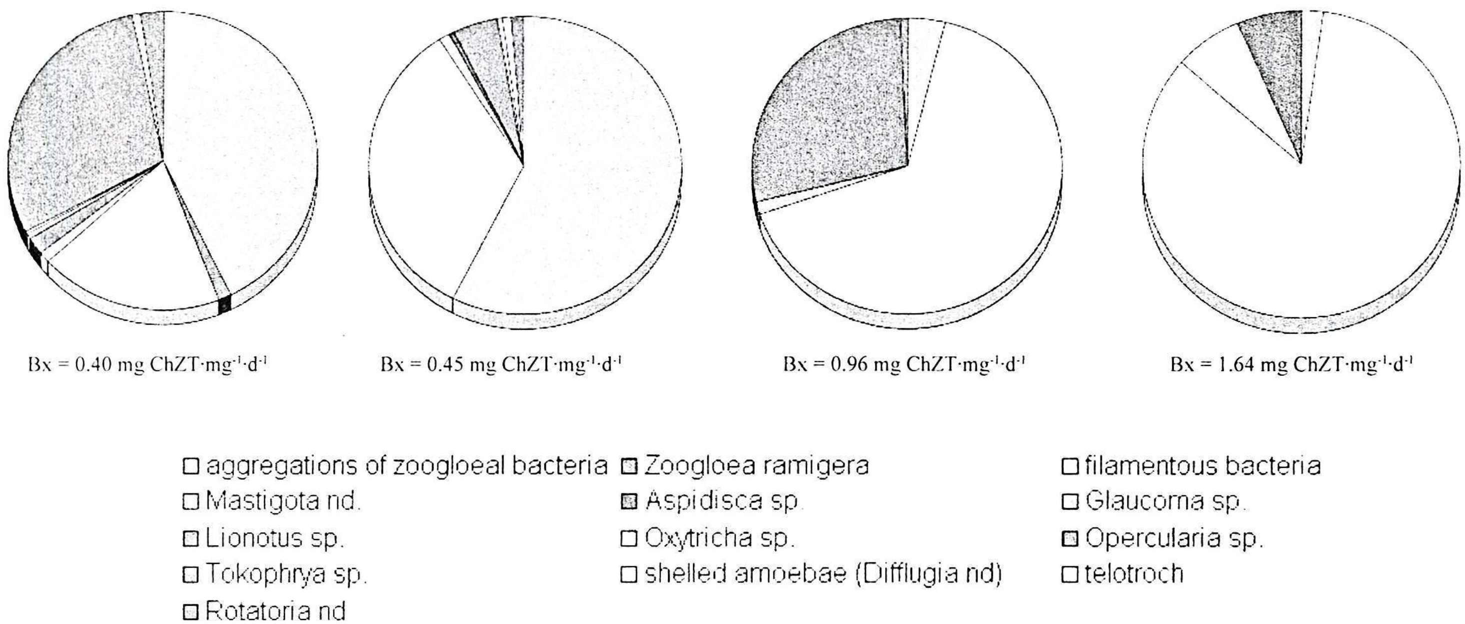

Fig. 1. Results of biological examinations of activated sludge in Sequencing Batch Reactor at sludge loading values of $Bx = 0.40–1.64$ mg $ChZT·mg^{-1}·d^{-1}$

Table 2. Results of biological examinations of activated sludge in Sequencing Batch Reactor

Participation of leachates in the mixture with synthetic wastewaters [%]		5		10		20	30
Sludge loading [mg COD·mg^{-1}·d^{-1}]		0.40		0.45		0.96	1.64
Sludge index [cm^{3}·g^{-1}]		102		96		80	72
Date of examination		12.05.05	24.05.05	20.06.05	12.07.05	29.09.05	25.11.05
General characteristic of the sludge		large, mergred flocs (not countable), light brown, well settling, lack of free-swimming bacteria	well formed, dark beige flocs, very large, which make their counting impossible, lack of free-swimming bacteria	brown, tiny, lightly detached flocs, well formed and settling, lack of free-swimming bacteria	lots of tiny, detached flocs, so counting is impossible, only clean colonies of zoogloeal bacteria were counted, lack of free-swimming bacteria	brown, fairly merged, beset flocs, lack of free-swimming bacteria	brown, rather merged, beset flocs, lack of free-swimming bacteria, hyphae fungi are present (3·10^{3}·cm^{-3})
Quantity of microorganisms in 1 cm^{3} of sludge (– not recorded)	sludge flocs	–	–	–	–	93·10^{3}	85.2·10^{3}
	aggregations of zoogloeal bacteria	95.4·10^{3}	12.2·10^{3}	23.4·10^{3}	72·10^{3}	3·10^{3}	1.2·10^{3}
	Zoogloea ramigera (Itzigsohn, 1868)	4.2·10^{3}	–	–	–	–	–
	filamentous bacteria	18·10^{3}	16.8·10^{3}	32.4·10^{3}	14·10^{3}	52.2·10^{3}	43.8·10^{3}
	Mastigota nd.	1.8·10^{3}	0.6·10^{3}	–	2.1·10^{3}	–	3.6·10^{3}
	Aspidisca sp.	4.8·10^{3}	0.6·10^{3}	0.6·10^{3}	–	–	–
	Glaucoma sp.	–	–	–	–	1.2·10^{3}	–
	Lionotus sp.	–	0.6·10^{3}	–	–	–	–
	Oxytricha sp.	0.6·10^{3}	–	–	–	–	–
	Opercularia sp.	13.8·10^{3}	33·10^{3}	4.2·10^{3}	2.8·10^{3}	22.2·10^{3}	3.5·10^{3}
	Tokophrya sp.	–	–	0.6·10^{3}	–	–	–
	shelled amoebae (*Difflugia nd.*)	–	1.2·10^{3}	–	–	–	–
	telotroch	–	–	–	1.4·10^{3}	–	–
	Rotatoria nd.	3.0·10^{3}	1.8·10^{3}	1.8·10^{3}	–	0.6·10^{3} (dead)	–

periodically in the sludge. Four taxa of microorganisms constituted the microfauna at the above sludge load.

A sudden deterioration of the quality of the activated sludge caused an increase in its load to 0.96 mg COD·mg^{-1}·d^{-1} by dosage 20% leachates in the mixture with synthetic wastewaters. Further flocs fragmentation and encystation, accompanied by a drastic decrease in the number of bacteria forming zoogloeal aggregations (down to 3·10^3·cm^{-3}) in them, a reduction in the number of the microfauna to two taxa (*Glaucoma* sp. – 1.2·10^3·cm^{-3} and *Opercularia* sp. – 22.2·10^3·cm^{-3}) and the death of rotifers, occurred. At the same time, the number of filamentous bacteria increased significantly in the sludge (to 52.2·10^3·cm^{-3}), which may indicate a high resistance of these bacteria to toxic compounds occurring in leachates.

The sludge load of 1.64 mg COD·mg^{-1}·d^{-1} at the 30% addition of leachates to synthetic wastewaters enhanced the phenomena described above. The number of aggregations of zoogloeal bacteria fell down to 1.2·10^3·cm^{-3}, while that of filamentous bacteria was still very high (43.8·10^3·cm^{-3}). Moulds (3000·cm^{-3}) occurred in the sludge. These organisms did not influence adversely sludge settling properties and the volume index was still low and equaled 72 cm^3·g^{-1}. Flagellates (3.6·10^3·cm^{-3}) and *Opercularia* sp. (3.5·10^3·cm^{-3}) were the only animal organisms in the sludge.

In conclusion, the studies showed that the activated sludge in the SBR had properties of a well working sludge when 5–10% of leachates were dosaged in the mixture with synthetic wastewaters at which the influent COD was 955–1059 mg O_2·dm^{-3}, 61–104 mg N-NH_4·dm^{-3}, 0.030–0.590 mg NO_2-N·dm^{-3}, 0.57–0,69 mg NO_3-N·dm^{-3} and 13.3–14.3 mg P-PO_4·dm^{-3}, and *Bx* 0.40–0.45 mg COD·mg^{-1}·d^{-1}. Besides zoogloeal and filamentous bacteria forming the flocs body, the microfauna of the sludge was composed of 5 and 4 taxa, respectively. Similar abundance of taxa in the activated sludge was recorded by Kuligowska *et al.* [11] during the treatment of wastewaters characterized by COD of 612 mg O_2·dm^{-3}. Flagellates and attached protozoa of the genera *Opercularia* and *Vorticella* were the most important representatives of the microfauna. Filamentous bacteria were not recorded. These authors showed an even greater diversity of the microfauna (15–22 taxa) in the activated sludge adapted for the treatment of wastewaters characterized by COD raging from 570 to 580 mg O_2·dm^{-3}. Shelled amoebae of the genus *Centropyxis* sp., attached ciliates *Acineta uncinata*, *Epistilis plicatilis*, *Opercularia coarctata* and free-swimming ciliates *Aspidisca cicada* dominated in the microfauna then.

An increase in the leachate participation in the wastewaters dosaged to the reactor to 20 and 30% and by this *Bx* values to 0.96 and 1.64 mg COD·mg^{-1}·d^{-1} caused a considerable decrease in the abundance of zoogloeal bacteria in the sludge, a sudden disappearance of the majority of protozoa and rotifers. At the same time, this encouraged the growth of filamentous bacteria. Apart from zoogloeal bacteria and attached protozoa of the genus *Opecularia*, this group of bacteria may be considered to be a characteristic component of the activated sludge biocenosis adapted for the pre-treatment of the study leachates (Fig. 2).

The disappearance of the microfauna in the activated sludge at *Bx* of 0.96 to 1.64 mg COD·mg^{-1}·d^{-1} should be attributed to the presence of carbon compounds that are difficult to biodegrade or toxic in the study leachates. This is indicated by the high COD value in the treated wastewaters of 1246 and 2879 mg O_2·dm^{-3}, respectively. The influence of the concentrations of mineral nitrogen forms was smaller as they did not exceed

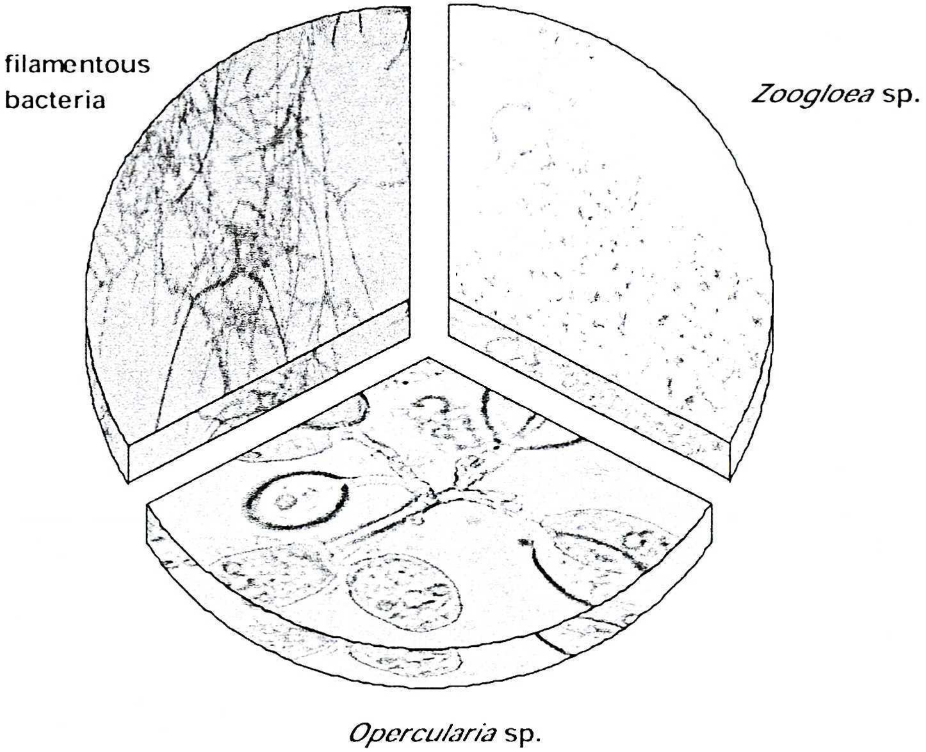

Fig. 2. The main groups of activated sludge biocenosis taking part in landfill leachates pretreatment in SBR

the values characteristic of toxic activity or inhibiting microfauna growth in the activated sludge (Tab. 3). The content of heavy metals in the pre-treated wastewaters did not inhibit the growth of protozoa and rotifers in the sludge, too. According to Abraham *et al.* [1], protozoa belonging to *Aspidisca cicada*, *Chilodonella uncinata*, *Vorticella convallaria* and *Vorticella microstoma* species tolerate concentrations of Fe > 2 $mg\cdot dm^{-3}$, Zn > 0.5 $mg\cdot dm^{-3}$, Cu > 0.06 $mg\cdot dm^{-3}$ and Cr = 0.10 $mg\cdot dm^{-3}$, that is significantly exceeding the concentrations of these metals in the pre-treated wastewaters.

The present studies are sufficient to suggest that the efficiency of leachate pre-treatment may be enhanced by increasing the number of microorganisms adapted to degrading the contaminations occurring in them. These are zoogloeal and filamentous bacteria as well as protozoa of the genus *Opercularia* in the case of the study leachates. The studies by Żubrowska-Sudoł [28] and Podedworna and Żubrowska-Sudoł [16] showed that moving-bed sequencing batch biofilm type reactors (MBSBBR) in which the growth of microorganisms occurs as activated sludge and biofilm on the surface of movable plastic carriers with a large specific surface create conditions conducive to an intensive multiplication of microorganisms. This technology allows a large load of contaminations to be taken in without changing the reactor volume, guarantees nitrification regardless of the wastewater retention time and activated sludge age, as well as makes the process of integrated denitrification and biological phosphorus removal possible under conditions of

Table 3. Comparison of the concentrations of different forms of mineral nitrogen in pretreated wastes at the critical sludge load values with toxic concentrations for activated sludge microfauna

Concentration of different forms of mineral nitrogen at the critical sludge load values (*Bx*) [$mg \cdot dm^{-3}$]				Inhibition or toxic treatment (activity) on activated sludge microfauna	Author, year
Bx	$N\text{-}NH_4^+$	$N\text{-}NO_2^-$	$N\text{-}NO_3^-$		
0.96 mg $COD \cdot mg^{-1} \cdot d^{-1}$	influent			Rotatoria – *Brachionus plicatilis*. Inhibition of glucosidase activity, at 2.4 ppm deionized ammonium	Araujo *et al.* (2000) [2]
	292	0.100	1.07		
	effluent			Protozoa – *Euplotes vannus* LC50 after 2 hours 7870 mg $N\text{-}NH_4 \cdot dm^{-3}$, threshold concentration inhibiting growth 100 mg $N\text{-}NH_4 \cdot dm^{-3}$	Xu Henglong *et al.* (2004) [25]
	9.60	0.050	7.80		
1.64 mg $COD \cdot mg^{-1} \cdot d^{-1}$	influent			Protozoa – *Paramecium bursaria* LC50 after 2 hours 95.94 mg $N\text{-}NH_4 \cdot dm^{-3}$ 27.3 mg $N\text{-}NO_2 \cdot dm^{-3}$	Xu Henglong *et al.* (2005) [26]
	351	1.040	4.67		
	effluent			Inhibition of protozoa and rotifers growth at ammonium nitrogen concentration above 30 $mg \cdot dm^{-3}$	Puigagut *et al.* (2005) [17]
	52.1	0.070	1.90		

a decreased load of organic compounds. The biomass development on carriers also limits the need for sludge recirculation and settling in secondary settling tanks, and thus it also eliminates sludge bulking. It is of great importance in the case of the study leachates as filamentous bacteria, dominated in the activated sludge biocenosis adapted for their pretreatment, as well as moulds at $Bx = 1.64$ mg $COD \cdot mg^{-1} \cdot d^{-1}$, also. Earlier on, conventional reactors with an immobilized biomass were used for wastewater treatment by Imai *et al.* [6] and Welander *et al.* [23], and SBR systems with microorganism carriers by Loukidou and Zouboulis [13].

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KSZTAŁTOWANIE SIĘ BIOCENOZY OSADU CZYNNEGO W PROCESIE PODCZYSZCZANIA ODCIEKÓW ZE SKŁADOWISK ODPADÓW W REAKTORZE SBR

W pracy wykazano, że przy przyjętym reżimie technologicznym pracy SBR uzyskać można 74–71% efektywności usuwania zanieczyszczeń wyrażonych w ChZT (stężenie w dopływie 955–1059 mg $O_2 \cdot dm^{-3}$) przy *Bx* w zakresie 0,40–0,45 mg ChZT·$mg^{-1} \cdot d^{-1}$. Azot amonowy w stężeniu do 292 mg NH_4-N·dm^{-3} usuwany był w 97% przy *Bx* w zakresie 0,40–0,96 mg ChZT·$mg^{-1} \cdot d^{-1}$. Jednocześnie zachodził proces symultanicznej denitryfikacji obrazowany 93% efektywnością usuwania $N_{całk}$. Osad czynny wykazywał cechy osadu wpracowanego do podczyszczania odcieków przy *Bx* w zakresie 0,40–0,45 mg ChZT·$mg^{-1} \cdot d^{-1}$. Jego biocenozę stanowiły bakterie zooglealne i nitkowate, pierwotniaki *Mastigota* nd., *Difflugia* nd., *Aspidisca* sp., *Lionotus* sp., *Oxytricha* sp., *Opercularia* sp., *Tokophrya* sp. i wrotki. Przy wartościach krytycznych *Bx* (0,96–1,64 mg ChZT·$mg^{-1} \cdot d^{-1}$), przy których gwałtownie spadła efektywność podczyszczania odcieków, biocenozę osadu stanowiły jedynie bakterie zoogleалne i nitkowate, grzyby strzępkowe oraz *Mastigota* nd. i *Opercularia* sp.