[Japanese Journal of Water Treatment Biology Vol.43 No.2 73-82 2007]

Excess Sludge Reduction and Biomass Characteristics in Swim-Bed Wastewater Treatment Process

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Abstract

The management of excess sludge in wastewater treatment accounts for a major portion of the cost for wastewater treatment and entails significant technical challenges. Swim-bed technology containing an innovation attached-growth material named biofringe (BF) allows the retention of large amounts of biomass, which could contribute to the reduction of excess sludge production. The treatment performances of a lab-scale reactor packed with BF material under various operational conditions and volumetric loading rates were investigated. The process demonstrated effective treatment of high-strength wastewater. COD removal efficiencies greater than 80% were consistently achieved even with volumetric loading rates as high as 7 kg-COD/m^3/d. Observed sludge yields (Y_{obs}) from 0.13 kg-MLSS/kg-COD$_{removed}$ to 0.29 kg-MLSS/kg-COD$_{removed}$ were obtained in swim-bed wastewater treatment. The occurrence of an abundance of protozoa and mentozoa in the activated sludge floc could account for this significantly low production of excess sludge. The long sludge retention time (SRT) was also regarded to be another reason for the low excess sludge production. The attachment of large amounts of biomass on the BF material might be due to the extremely high ratios of extracellular polymers (EPS) to biomass, which varied from 64% to 84%.

Key words: attached-growth, sludge reduction, excess sludge reduction, protozoa, mentozoa, extracellular polymers

INTRODUCTION

The activated sludge process is widely used for wastewater treatment. It has been developed for about one-hundred years, but many problems still exist. Especially the generation of a large amount of excess sludge that has to be wasted has been a big problem. The cost for excess sludge treatment has been estimated to be 50-60% of the total expense of wastewater treatment plants. Accordingly, there is much more interest in biological treatment methods with less excess sludge production.

Several strategies have been suggested for the purpose of sludge reduction. One is sludge disintegration. Cell lysis will release cell contents into the medium, thus providing an autochthonous substrate that contributes to the organic loading. Components of this substrate are reused in microbial metabolism while a portion of the carbon is liberated as products of respiration, which results in an overall reduction in biomass production. Since the biomass growth that subsequently occurs on this autochthonous substrate cannot be distinguished from growth on the original organic substrate, this strategy is also termed as cryptic growth[1]. Ozonation is another well known method of sludge disintegration. Yusui and Shibata[2] developed a process consisting of sludge ozonation and a biodegradation stages for treating municipal and industrial wastewaters. No excess sludge was withdrawn and no significant accumulation of inorganic solids occurred in the aeration tank when operated under the optimal rates. Kamiya and Hirotsuji[3] subsequently developed a new system combining both biological treatment and intermittent ozonation to reduce excess sludge production using less ozone, which also favorably controlled the sludge bulking. Results of their lab-scale experiments treating synthetic wastewater indicated that intermittent ozonation was preferred over continuous ozonation because it reduced sludge production by 50% with only 30% of the ozone dose required for continuous treatment and it resulted in improving the sludge settling characteristics[4-7]. Thermal treatment is another sludge disintegration method. This process has also been developed in combination with thermal and chemical treatment for sludge reduction. In addition, it was found that alkaline treatment by NaOH addition combined with thermal treatment (pH10, 60℃ for 20 min) was the most efficient sludge elimination process[8,9]. However, most disintegration technologies have an unfavorable cost: benefit ratio and are not wildly regarded as economically viable[10].

The second technology for sludge reduction is uncoupling metabolism. Bacterial anabolism is coupled to catabolism of substrate through rate limiting respiration[11]. However, uncoupled metabolism would occur if respiratory control did not exist and instead the biosynthetic processes was rate limiting. Therefore, excess free energy would be directed away from anabolism so that the production of biomass can be reduced. Uncoupled metabolism was observed under some conditions, such as in the presence of inhibitory compounds, heavy metals, excess energy uncoupling, abnormal temperatures, and limitation of nutrients[12,13]. Recently, many researchers have focused on sludge reduction induced by chemical uncoupling. Sludge production was reduced significantly, raging from 40% to 86.9%, by adding different chemical uncouplers to biological treatment[14-19]. However, the application of chemical uncouplers for sludge reduction may cause a reduction in COD removal, an increase in oxygen consumption and deterioration in activated sludge properties such as settling and dewatering. Most of chemical uncouplers tested are xenobiotic and potentially harmful to the environment. Thus, their application should be observed carefully and further research is needed on the environmental impacts of long-term use of chemical uncouplers[20].

The third approach for sludge reduction is to maximize the sludge retention time and increase the numbers of predators within the biological treatment system. Microorganisms satisfy their maintenance energy requirement in preference to producing additional biomass, which has possible applications for sludge reduction during biological wastewater treatment. By increasing the biomass concentration, it would theoretically be possible to reach a situation in which amount of potential energy provided equals the maintenance demand. The complete sludge retention had little impact on wastewater treatment performance except a slight increase in the sludge inorganic fraction[21].

The protozoa and metazoa can not only prey on the bacteria but also degrade the remaining substrate. During energy transfer from low to high trophic levels, energy is lost due to inefficiencies during biomass conversion. In the past, protozoa and metazoa were usually used as important indicators of process performances and efficiencies in biological wastewater treatment processes. Recently, many researchers have focused on sludge reduction induced by grazing on bacteria[20]. The minimization of sludge production by protozoa and metazoa predation on bacteria was investigated in two two-stage systems treating synthetic wastewaters, in which the second stage was a suspended-carrier biofilm reactor[22,23]. The sludge production in the predator stage was significantly decreased by 60-80% compared with that in the bacterial stage. The total sludge yield was 0.05 g-TSS/g-$COD_{removed}$ in the two-stage system fed acetic acid, whereas it was 0.17 g-TSS/g-$COD_{removed}$ in the other two-stage system fed methanol. Further study was carried out to investigate sludge reduction with this two-stage system treating different pulp and paper industry wastewaters (the second stage designed as activated sludge and biofilm reactors, respectively)[24]. Results of this study showed that the sludge yield (0.01-0.23 g-TSS/g-$COD_{removed}$) of the two-stage system was obviously lower than that (0.2-0.4 g-TSS/$COD_{removed}$) of the conventional activated sludge processes treating the same wastewater.

Swim-bed technology involving the novel acryl fiber material, biofringe (BF), is a new concept for the treatment of organic wastewater. The BF material allows for attachment of large amounts of biomass on a flexible matrix in a fixed position. By this approach, flexing of the matrix induced by wastewater flow creates a "swimming" motion that enhances mass transfer of nutrients to the attached growth (*i.e.*, biofilm). Thus, all the potential benefits of fluidized-bed reactors are retained. In addition, it was our hypotheses that sludge production in a BF process would be reduced due to the formation of a longer food chain. The abundance of protozoa and metazoa in a BF process should enhance the sludge reduction. The objective of this study was to evaluate the performance of a single BF reactor for excess sludge reduction. The characteristics of biomass were investigated as well.

MATERIALS AND METHODS

Reactor and operational conditions The reactor containing BF material used in this study was constructed by acryl resin, having downdraft and updraft sections in a parallel upright arrangement as shown in Fig. 1. It also had clear zones of approximately 70 mm at the bottom and 30 mm at the top (below and above the biofringe reaction zone in the downdraft section). The working volume was 21.6 *l*. Influent was introduced deeply within the updraft section using a peristaltic pump at a fixed flow rate of 2 *l*/h. Air was also introduced near the base of the updraft section, serving to mix and oxygenate the wastewater while circulating it through the reactor. The air flow rate was fixed at 10 *l*/min and was related to the highest water flow velocity, by a tracer study using polyvinyl alcohol beads (dia., 4 mm; s.g., 1.025), which were used to establish a correlation between air-flow rate and water flow velocity in the narrow updraft section.

The influent stock solution consisted of a bonito fish meat (Kyokuto medicine Co. Ltd., Japan) and peptone (Mikuni Chemistry Co. Ltd., Japan), and was diluted with tap water to obtain desired influent concentrations. The 5-day biochemical oxygen demand (BOD) was 74% of the chemical oxygen demand (COD) for influent solutions. $KHCO_3$ buffer solution was also added to the influent as a buffer solution. The synthetic influent compositons were showed in Table 1. In

Table 1 Influent Compounds

Compounds	Concentration(mg/*l*)
COD	450-3150
TN	45-315
Na^+	19
K^+	6
Ca^{2+}	20
Mg^{2+}	7
SO_4^{2-}	24
Alkalinity	195

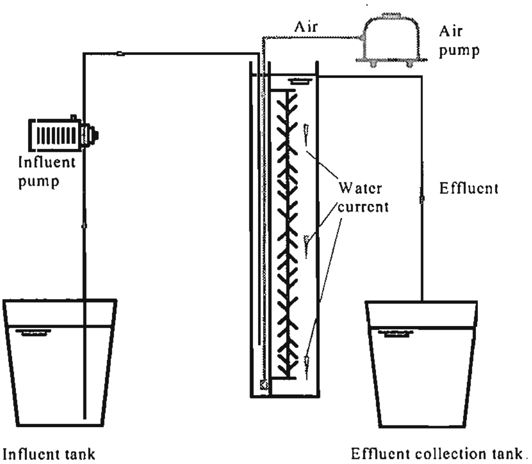

Fig. 1 Schematic diagram of experimental apparatus

Table 2 Operational conditions

Run	I	II	III	IV		V	
HRT (h)	10.8	10.8	10.8	10.8	10.8	10.8	
COD loading rate (kg-COD/m^3/d)	1	2	3	4	5	6	7
Influent COD (mg/l)	450	900	1350	1800	2250	2700	3150
Duration (days)	30	20	16	6	16	8	21

order to keep the operational temperature of 25℃, a heater was used in the wintertime and air-conditioning was applied in the summer time under relatively dark conditions.

The experiments were divided into five runs corresponding to the applied COD volumetric loading rates (VLRs). Table 2 gives an overview of the main operational characteristics for the whole study. At the end of each run, the reactor was stopped and the BF material was removed from the reactor. All the biomass attached on BF material was then detached by hand and collected along with biomass gleaned from cleaning the interior of the reactor and the total amount was quantified. The experiment was then immediately re-started using the biomass detached from the BF as seed sludge. To quantify the amount of solids lost in the effluent, the entire effluent was collected and representative samples were for SS determinations.

Biomass seed and retention matrix The reactor was initially seeded using activated sludge from a lab-scale fill-and-draw batch reactor. The synthetic medium used for the development and maintenance of the seed sludge was the same as that used in this study.

The biomass retention matrix was the BF material composed of fringe yarns (NET Co. Ltd., Japan, diameter, *ca.* 3 mm) attached to a support filament as shown in Fig. 2. The staple fiber of the fringe yarns was a hydrophilic acrylic composite. The material had a rough texture formed by bonding three threads with different heat resistances in a sinoidal pattern using a partial-melting process.

Analytical methods COD was measured by the closed reflux colorimetric method according to *Standard Methods*[25] with prior filtration using filter paper (No.5B) to remove undissolved components. BOD was measured using a respirometer (BODTrack; Hach Co., Ltd., Loveland, CO). The pH level was measured using a pH meter (IM-22P; TOA Electronics, Ltd., Tokyo, Japan). Dissolved oxygen (DO) was measured using a DO meter (OM-51; Horiba, Ltd., Kyoto, Japan). MLSS, SS and SVI were all determined according to *Standard Methods*. Activated sludge flocs were examined by light microscopy (Nikon, ECLIPSE E600) and pictures were taken using a digital camera (Nikon CooLpix 4500).

Extra cellular polymers (EPS) of suspended sludge and biomass attached to biofringe were extracted by the alkaline-washing method[26]. In the extracted solutions, proteins were measured using the method of Lowry *et al.*[27] and carbohydrates by the method of Dubois *et al.*[28]. Nucleic acids (combined RNA and DNA) were estimated by the UV absorption method[29].

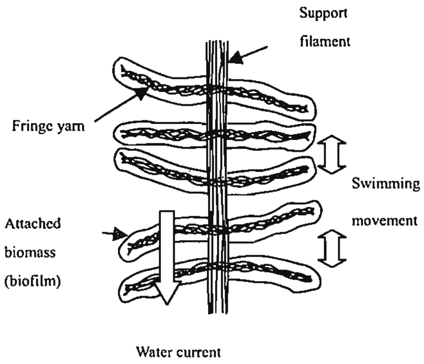

Fig. 2 Configuration of biomass carrier

Calculation methods The observed sludge yields (Y_{obs}) were calculated according to the following equations for each run.

$$Y_{obs} = \frac{g\text{-}X_{end} - g\text{-}X_{start}}{g\text{-}COD_{removed}} \tag{1}$$

$$g - X_{end} = X_{BF} + X_R + \sum_{i=1}^{n} X_i \tag{2}$$

The apparent effluent sludge yield (Y_e) basing on the effluent SS concentrations were calculated according to the following equation:

$$Y_e = \frac{\sum X_i}{g - COD_{removed}} \tag{3}$$

Where the term g-X_{start} is the total amount of seed biomass presented at the beginning of each run. g-X_{end} represents the total amount of the biomass at the end of the experiment, which including the biomass attached on the biofringe (X_{BF}), dispersed bacteria contained in the cleaning water for the reactor(X_R) and the sum of the biomass (X_i) in the daily collected effluent. The term g-$COD_{removed}$ is the total amount of removed COD in each run.

Basing on the assumption that the attached biomass on the biofringe was a constant amount after the biofilm became mature, the values of sludge retention time (SRT) and food to microorganisms ratio (F/M) respectively were estimated as:

$$SRT = \frac{V \cdot X_a}{Q_W \cdot X_e} \tag{4}$$

$$F/M = \frac{Q_s \cdot C_s}{X_a \cdot V} \tag{5}$$

where V (l) is the reactor volume, X_a (mg/l) is the average MLSS concentration in reactor, Q (l/d) represents flow rate, X_e (mg/l) is the average effluent SS concentration, and C_s (mg/l) is the average influent COD concentration.

RESULTS AND DISCUSSION

General treatment The degradation of organic constituents was evaluated by COD removals as shown in Fig. 3. The COD removals were maintained at a nearly constant level throughout the entire period of testing and over 80% of COD removal efficiency was achieved even at extremely high VLRs of 7 kg-COD/m^3/d.

Figure 4 and Fig. 5 show the daily changes in reactor MLSS and effluent SS concentrations, respectively. The MLSS

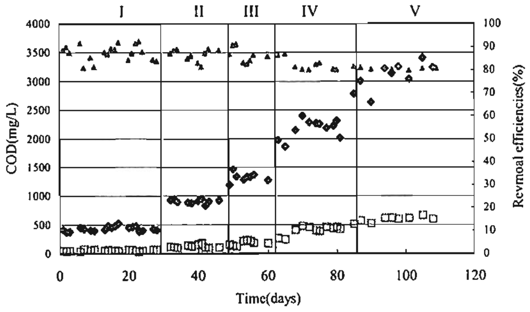

Fig. 3 Time courses of COD concentrations
◆ Influent, □ Effluent, ▲ Removal efficiency

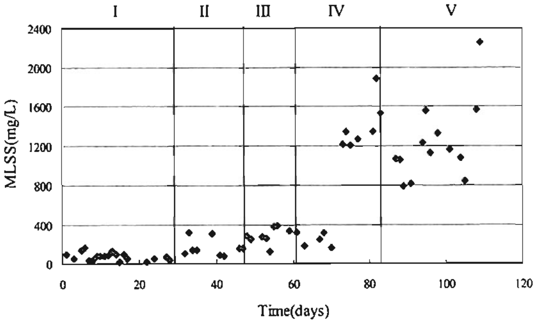

Fig. 4 Time courses of MLSS concentrations
◆ MLSS

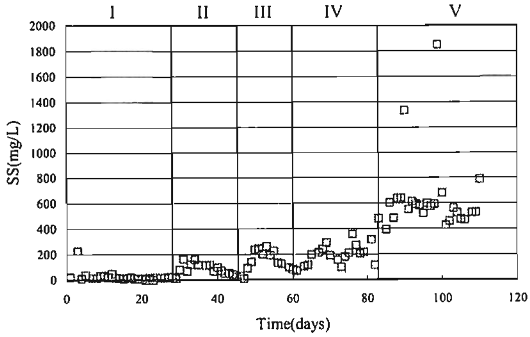

Fig. 5 Time courses of SS concentrations
□ Effluent SS

concentrations varied greatly, these results show that the MLSS and effluent SS levels increased with an increase in VLRs. They were not stable even under the same operational conditions. In fact, the reactor MLSS and effluent SS concentrations should be same since there was no setting tank in this process. However, the effluent was collected every day by a tank in order to calculate the sludge production. The values of SS that sampled from the collected effluent presented one day's average SS levels, while MLSS concentrations were the instant values. This implied that the reactor MLSS levels fluctuated greatly even within one day's operation. This would be closely related to attachment/detachment profile of biofilm. Suspended solids in a BF reactor can be regarded as the fragments of biofilm[26]. The average effluent SS varied from 10.3 mg/*l* to 640.9 mg/*l* with the increase in VLRs from 1 kg-COD/m^3/d to 7 kg-COD/m^3/d.

Sludge production The observed sludge yield (Y_{obs}) and apparent sludge yield for effluent SS (Y_e) were determined according to equations (1) and (3), respectively. Figure 6 shows the values of Y_{obs} and Y_e in each run. The levels of observed sludge yields (Y_{obs}) ranged from 0.13 kg-MLSS/kg-COD$_{removed}$ to 0.29 kg-MLSS/kg-COD$_{removed}$ and the levels of apparent sludge yields (Y_e) varied from 0.06 kg-MLSS/kg-COD$_{removed}$ to 0.21 kg-MLSS/kg-COD$_{removed}$ when the VLRs increased from 1 kg-COD/m^3/d to 7 kg-COD/m^3/d. The sludge yields were reduced considerably in the first four runs. Although sludge yield increased a lot in Run V corresponding to a COD VLR of 7 kg-COD/m^3/d, they were still lower (30-50%) than that reported values for typical activated sludge process[30]

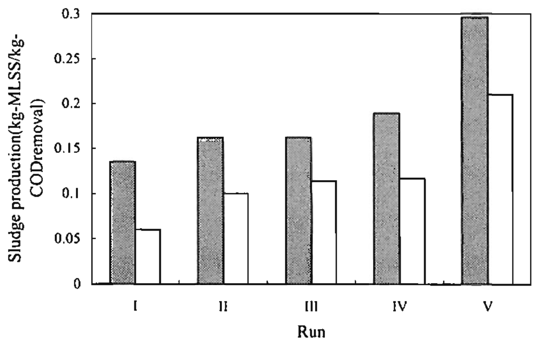

Fig. 6 Sludge production in each run
▨ Y_{obs} ☐ Y_e

Table 3 Summarization of the SRT values and the F/M ratios for Run

Run	I	II	III	IV	V
SRT (d)	49	17	11	15	15
F/M (kg-COD/m^3/d)	0.33	0.69	0.97	0.73	0.38

Sludge characteristics

Sludge retention time and F/M ratio

SRTs and F/M ratios for all runs were calculated by Equations (4) and (5), respectively (shown in Table 3). Although the lab-scale study was conducted with short run periods, the SRTs were longer than those of conventional activated sludge processes ranging from 3-6 days. The longer SRT benefited sludge reduction. The results of a pilot-scale side-stream MBR treating synthetic wastewater at SRTs ranging from 30 days to 2 days showed that both sludge yield and biomass viability generally increased with decreasing SRT[31]. Little excess sludge production was obtained in a pilot cross-flow MBR plant with complete sludge retention. High reactor MLSS concentrations of 40-50 g/*l* were obtained and only 6 % of influent carbon was assimilated.

The F/M ratios from Run II to Run IV were higher than optimal F/M ratios applied in conventional activated sludge processes. However, better removal efficiencies were still achieved in these three runs, which demonstrate that the swim-bed process is well suited for the wastewater treatment under high F/M ratios.

Microorganisms Frequent microscopic observations of sludge were performed. Table 4 gives an overview of the observation results. Many *Mstigophora* were observed in Run I. *Ciliophora*, which preyed on the free bacteria, appeared in both Run I and Run II (shown in Fig. 7(a)). These two species of protozoa were abundant in the activated sludge in Run I and their dominance was closely related to clear effluent and good sludge settling ability. Others also observed that *Ciliophora* were

Table 4 Enumeration results for protozoa and metazoa (count/m*l*)

	filamentous microorganism	*Ciliate*	*Rotifer*	*Chaetonotus*	*Fragellate*	*Dyplogaster*
I	+	1.6×10^3–2.0×10^5	2.5×10^2–6.3×10^2	1.5×10–1.0×10^2	6.3×10^2–3.0×10^3	1.3×10–2.0×10^2
II	—	7.9×10^2–1.3×10^3	1.6×10^3–3.2×10^3	2.5×10^2–3.2×10^2	—	2.5×10–1.0×10^2
III	—	—	6.3×10^4–1.3×10^5	—	—	—
IV	—	—	2.5×10^2–2.0×10^3	—	—	—
V	—	—	—	—	—	—

generally the most abundant in protozoa communities in a wastewater treatment plant[32)]. They are responsible for decreasing the numbers of free bacteria and also play an important role in consuming dissolved organic matter[33)]. Many researchers have reported that *Ciliophora* contributes to the reduction of excess sludge production. The growth of *Ciliophoras* in a two-stage pure culture chemostat system was associated with 12–43% sludge reduction[34)] and *Ciliophoras* and *Mstigophoras* contributed to sludge reduction in an aerobic treatment process[22)]. *Chaetonotus* was also found during the first two runs (shown in Fig. 7 (a)) and an increase in number was observed in the second run. *Chaetonotus* usually use small protoza and filamentous algae as food and they are also known to be related for sludge reduction. *Rotatoria* occurred throughout the first four runs and their numbers increased with increases in VLRs from 1 kg-COD/m^3/d to 3 kg-COD/m^3/d (showed in Fig. 7 (b)). Many *Rotatoria* were still found in Run IV although their number decreased compared to Run III. It has been suggested that *Rotatoria* play an important role in the activated sludge process[35)]. The growth of *Rotatoria* is mainly dependent on the amount of food available and they consume several times of their body weight every day[36)]. This can be used to explain why the greatest abundance of *Rotatoria* was observed in Run III at a VLR of 3 kg-COD/m^3/d. The *Rotatoria* can graze on algae and particles in size ranging from 0.2–10μm according to the research work of Lapinski[37)]. The total biomass of the treatment system in which *Rotatoria* grazed on suspended particles was subsequently reduced by about 10% (TSS) within 48h at the *Rotatoria* density used. Consistent with that, a preliminary experiment suggested that total solids removal was also significantly reduced by *Rotatoria* grazing. In this study, considerable sludge reduction was obtained due to the *Rotatoria* occurrence[37)].

It was apparent that high organic removal efficiencies and sludge reductions were closely related to the occurrence of the protozoa and metazoa in large numbers. Their high diversity accounted for considerable sludge reduction in the first two runs. No increases in the observed sludge yields were observed with increases in VLRs from 2 kg-COD/m^3/d to 3 kg-COD/m^3/d. This result was due to the abundance of the *Rotatoria* at loadings of 3

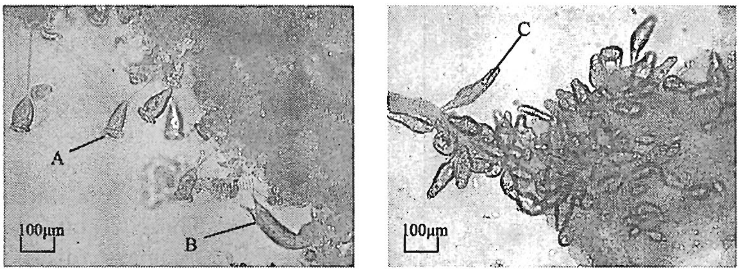

Fig. 7 Microscopic photographs of activated sludge on BF material showing protozoa and mentozoa
A: *Ciliapophora*, B: *Chaetonotus*, C: *Rotatoria*

kg-COD/m^3/d. Increases in both observed sludge and excess effluent sludge yields for Run V were associated with a decrease in protozoa and metazoa populations. Thus, it is not recommended to operate single swim-bed reactor at VLRs over 7 kg-COD/m^3/d.

Characterization of extracellular polymers (EPS) At the end of each run, the EPS of biofilm were analyzed. The results were shown in Table 5. The total EPS in the attached growth of this study were significantly higher than reported values of various activated sludges[38]. Protein was the main component of EPS, which was consisted with the results of Rouse *et al.*[26]. The extremely high level of EPS-protein was the characteristic for biofilm growth (i.e., growth on an attachment medium) as compared to the flocculent or granular sludge.

In biofilm, EPS provides the framework, into which microbial cells are embedded. The attached biomass on BF material varied from 42.0 g-MVLSS/m-BF to 251 g-MLVSS/m-BF in this study (Fig. 8). The high levels EPS obtained in the present study contributed to the high amounts of the biomass attached on BF.

Table 5 Compositions of extracellular polymers in biofilm samples for each Run

	Run I	Run II	Run III	Run IV	Run V
Proteins	66	58	49	48	62
Carbohydrate	11	8	12	8	8.7
Nucleic acids	9	10	12	8	11
Total	86	76	73	64	82

All values are reported as percent (%) of dry biomass (i.e. MLVSS)

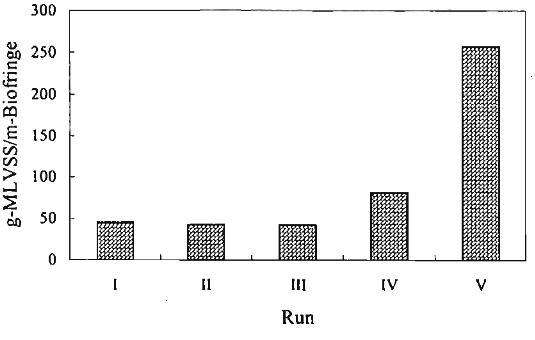

Fig. 8 Biomass changes with VLR changes
▨ Attached growth biomass

EPS came from the natural secretions of bacteria, cell lysis and hydrolysis production[39]. The ratios of EPS to the biomass first reduced in the first four runs by enhancing the VLRs and re-increased in Run V. The trend of EPS changes responding to the VLRs changes illustrated that the provided nutrient would affect the ratios of EPS to the biomass. Miqueleto *et al.* also proved that the EPS production depended on the VLRs applied in the reactor[40]. Zhang found the EPS of biofilm were degraded by their own producers and other microorganisms in the starvation phase[39]. Li got the similar result when investigating the influence of starvation phase on the properties and the development of aerobic granules[41]. On the contrary, the relative higher EPS concentrations were obtained in the Run I and Run V. The F/M ratios were lower than the other three runs. Further study was needed to make clear the affection factors to the EPS concentrations.

CONCLUSIONS:

1. Good treatment performances were achieved using the swim-bed technology. COD removal efficiencies greater than 80% were obtained even at VLRs up to 7 kg-COD/m^3/d.

2. The BF material allowed for the attachment of large amounts of biomass. The attached biomass ranged from 42.0 g-MLVSS/m-BF to 251 g-MLVSS/m-BF. The large amounts of attached-growth extended the SRTs varying from 11 to 49 days and contributed to the abundance of metazoa and protozoa, which contributed to the significant sludge reduction. The observed sludge yields and effluent sludge yields ranged from 0.13 kg-MLSS/kg-$\text{COD}_{\text{removed}}$ to 0.29 kg-MLSS/kg-$\text{COD}_{\text{removed}}$ and from 0.06 kg-MLSS/kg-$\text{COD}_{\text{removed}}$ to 0.21 kg-MLSS/kg-$\text{COD}_{\text{removed}}$, respectively.
3. High levels of the EPS/biomass ratios were observed, which could enhance the attachment of the biomass on the BF matrix. These results demonstrated that the EPS/biomass ratios were affected by the VLRs.

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(Submitted 2006. 12. 6)
(Accepted 2007. 4. 16)