Application of acrylic fiber biomass carrier in integrated fixed-film activated sludge (IFAS) process for wastewater treatment

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Abstract

Some problems remain unsolved for the conventional activated sludge (CAS) process and biological nutrient removal (BNR) process, such as excessive sludge production, low pollutant removal rate and a large space requirement. The integrated fixed-film activated sludge (IFAS) process could enhance the CAS process by providing a greater biomass concentration in the aeration tank thus provide the potential to reduce the tank size requirements. Applied as an aerobic IFAS process, the swim-bed technology involving biofringe® (BF) acrylic fiber biomass carriers, has been presented for effective treatment of organic and nitrogen pollutants.

In this study, the organic and nitrogen removal performances of the swim-bed technology in different systems treating domestic and high-strength synthetic wastewaters were evaluated. Meanwhile, excess sludge reductions in different reactors were also investigated.

In the first part of this study, the characteristics of two kinds of acrylic fiber biomass carries, a net type carrier named biofix® (BX) and BF carrier, were evaluated in three parallel reactors and the treatment performances among them were experimentally compared. Under volumetric loading rates (VLRs) from 1 to 30 kg BOD m^{-3} d^{-1}, organic removal, nitrification performance and reactor sludge characteristics among three reactors were compared. The results showed that the BF biomass carrier packed in a cycle aeration reactor demonstrated the most stable and predominant performance in sludge attachment and retention, nitrification performance, and organic pollutant removal.

In the second part of this study, the BF biomass carrier was partially packed in a plug-flow activated sludge reactor with a packing ratio of 15%, to investigate its feasibility in improving the sludge settleability and enhancing the pollutant degradation of the activated sludge process. High s-BOD_5, s-COD, and ammonium removal efficiencies of 99.1%, 96.5% and 83.6%, respectively were obtained under VLR of 4.5 kg s-COD m^{-3} d^{-1}. SVI values below 50 mL g^{-1} and high settling velocities demonstrated satisfactory settling characteristics of the sludge, which were attributed to the increase in the size of the biomass flocs as a result of use of the BF carrier material. Relatively low viscosity of mixed liquor also facilitated the sludge

settling performance as no sludge bulking problems were encountered throughout the experimental period. Existence of a large amount of protozoa and metazoa observed through microscopic examination was considered to contribute to the lower sludge yield compared with that for the CAS process. DNA results demonstrated microbial community shift between the seed sludge and the sludge sample after 378 days of operation with *Proteobacteria* to be predominant. The results demonstrated that the use of swim-bed technology enhanced treatment performance and provided process stability for the CAS process.

In the third part of this study, the BF biomass carrier was packed into the suspended type Modified Ludzack-Ettinger (MLE) system to evaluate the enhanced nitrogen removal and excess sludge reduction. Synthetic domestic wastewater with low C/N ratio was used as influent. This system was operated continuously under various VLRs from 0.5 to 1.75 kg COD m^{-3} d^{-1}. During 110 days of operation, 70% of total nitrogen (TN) removal, 80% of COD removal and 90% of BOD removal efficiencies were obtained stably, even at a very short hydraulic retention time (HRT) of 2.8 h. Meanwhile, observed sludge yield of 0.13 kg MLSS kg^{-1} $COD_{removed}$ was obtained during the whole experimental period and the lowest observed yield of 0.05 kg MLSS kg^{-1} $COD_{removed}$ was obtained under VLR of 1.75 kg BOD m^{-3} d^{-1}. Furthermore, the mixed liquor suspended solids (MLSS) concentrations reached more than 8,000 mg L^{-1}, which was closely related to the stable TN removal performance and extremely low excess sludge reduction of this combined system.

In the fourth part of this study, simultaneous nitrification and denitrification (SND) with excess sludge reduction and were evaluated in an attached growth treatment system consisting of a down-flow anaerobic fermentation (AF) reactor and an aerobic swim-bed (SB) reactor operated as an integrated fixed-film activated sludge (IFAS) process. The two reactors were packed with biomass carriers consisting of different configurations of the same acrylic-fiber material: a net-type carrier biofill® (BL) in the AF reactor and BF in the SB reactor. The system was operated continuously under various organic loading rates (OLRs) from 1.5 to 4.5 kg COD m^{-3} d^{-1} and nitrogen loading rates (NLRs) from 0.1 to 0.3 kg N m^{-3} d^{-1} using moderately high-strength synthetic wastewater. The AF reactor, located upstream of the SB reactor, provided hydrolysis, fermentation and anaerobic ammonification. During 184 days of operation, stable TN removal efficiencies ranging from 85% to 97% were obtained primarily due to SND in the SB reactor when the OLR was higher than 2.7 kg COD m^{-3} d^{-1}. In addition,

observed sludge yields for the whole system ranged from 0.13 to 0.17 kg MLSS kg^{-1} $COD_{removed}$; furthermore, MLSS were maintained at about 10,000 mg L^{-1} in the SB reactor. Exceptionally high COD and TN removal rates of 5.9 kg COD m^{-3} d^{-1} and 0.43 kg N m^{-3} d^{-1}, respectively, were observed in the SB reactor. These results demonstrated that excess sludge reduction and SND could be achieved concurrently by this combined AF-SB system.

In general, applied as an aerobic IFAS reactor, the swim-bed technology involving the BF biomass carrier demonstrated remarkable pollutant removal performance for both high-strength and low-strength organic wastewaters, especially in nitrogen removal and excess sludge reduction.

List of abbreviations and nomenclature

AF	Anaerobic fermentation
BF	Biofringe®
BL	Biofill®
BX	Biofix®
CAS	Conventional activated sludge
COD	Chemical oxygen demand (mg L^{-1})
CSL	Corn steep liquor
DO	Dissolved oxygen (mg L^{-1})
DN reactor	Denitrification reactor
F/M	Food to microorganism ratio (kg BOD kg^{-1} MLSS d^{-1})
HRT	Hydraulic retention time (h)
IFAS	Integrated fixed-film activated sludge
MLE	Modified Ludzak-Ettinger
MLSS	Mixed liquor suspended solids (mg L^{-1})
MLVSS	Mixed liquor volatile suspended solids (mg L^{-1})
NE	Nitrification efficiency (%)
NT reactor	Nitrification reactor
OLR	Organic loading rate (kg COD m^{-3} d^{-1})
NLR	Nitrogen loading rate (kg N m^{-3} d^{-1})
SB	Swim-bed
s-BOD	Soluble BOD
s-COD	Soluble COD
SND	Simultaneous nitrification and denitrification
SRT	Solids retention time (d)
SS	Suspended solids (mg L^{-1})
SVI	Sludge volumetric index (mL g^{-1})
TN	Total nitrogen (mg L^{-1})
VFA	Volatile fatty acid (mg L^{-1})
VLR	Volumetric loading rate (kg COD/BOD m^{-3} d^{-1})
WWTP	Wastewater treatment plant
Y_{obs}	Observed sludge yield (kg MLSS kg^{-1} $COD_{removed}$)

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Chapter 1 Introduction

1.1 Conventional activated sludge process

In a plug-flow conventional activated sludge (CAS) plant (Fig 1-1), the primary settled wastewater and acclimated microorganisms (activated sludge) are aerated in a basin or tank. After a sufficient aeration period, the flocculent activated sludge solids are separated from the wastewater in a sludge settling tank. The clarified wastewater flows forward for further advanced treatment or discharge. A portion of the clarifier underflow sludge is returned to the aeration basin for mixing with the primary treated influent to the basin and the remaining sludge is wasted to the sludge handling portion of the treatment plant. The portion circulated is determined on the basis of the ratio of mixed liquor volatile suspended solids (MLVSS) to influent wastewater biochemical oxygen demand which will produce the maximum removal of organic material from the wastewater. Circulation flow varies from 25 to 50 percent of the raw wastewater flow, depending on treatment conditions and wastewater characteristics.

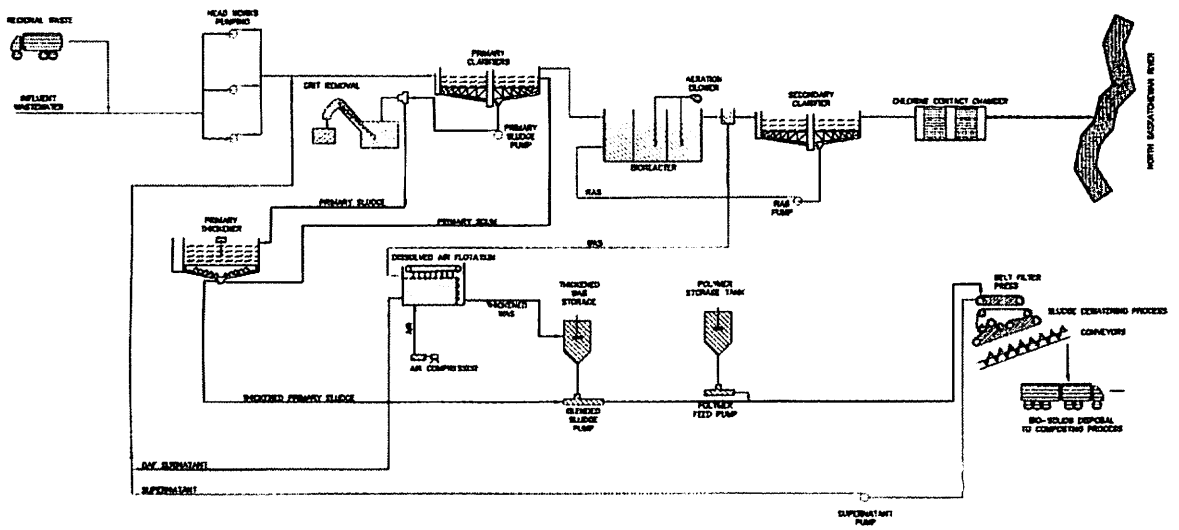

Fig. 1-1 Schematic of the conventional activated sludge process.

The antecedents of the CAS processes date back to the early 1880s to the work of Dr. Angus Smith, who invested the aeration of wastewater in tanks and enhancing of the oxidation of the organic matter. The process was named as activated sludge by Arden and Lockett because it involved the production of an activated mass of microorganisms capable of

aerobic stabilization of organic material in wastewater (Metcalf & Eddy, 1930).

The CAS process has been used widely around the world for biological treatment of domestic and industrial wastewaters. However, there are still some problems which attract much attention nowadays, such as high excess sludge production with the sludge yield coefficient ranging from 0.3 to 0.5 kg MLVSS kg^{-1} $COD_{removed}$ (Xing et al., 2003). Cost for disposal of excess sludge ranges from 25% to 60% of total operational costs of wastewater treatment plants (WWTPs) (Pérez-Elvira et al., 2006). Furthermore, costs for disposal of sludge continue to increase due to restrictions stemming from environmental legislative constraints (Lapinski et al, 2003). Compared with commonly used methods for disposal of excess sludge such as such as land spreading, landfill, incineration, and sea disposal (Bebin, 1997), an ideal way to solve this problem is to reduce sludge production at the source, i.e., at the wastewater treatment, before it becomes a problem (Wei et al., 2003).

Furthermore, the efficiency of nitrogen removal in the CAS processes is another critical issue due to eutrophication in receiving water bodies. Conventional nitrogen removal systems are based on the combination of nitrification and denitrification processes with spatial or temporal separation due to different environmental conditions required for the autotrophic nitrifiers and heterotrophic denitrifiers. Nitrification occurs under aerobic conditions while denitrification prevails in the absence of oxygen (Mu□nch, 1996). The obvious drawbacks of these conventional nitrogen removal processes -- preanoxic or postanoxic denitrification -- are the relatively large space requirement and relatively complicated operational controls.

1.2 Sludge reduction

Substrate is diverted from assimilation for biosynthesis to non-growth activities, so that wastewater treatment processes must be optimized to reduce the excess sludge production. Different strategies are recently developed for sludge reduction based on these mechanisms: lysis-cryptic growth, uncoupling metabolism, maintenance metabolism, and predation on bacteria (Low et al., 1999; Liu et al., 2001). The two latter strategies are relative easier and practical for application presently.

1.2.1 Maintenance metabolism in sludge reduction

Microorganisms satisfy their maintenance energy requirements prior to producing additional biomass, and this acknowledgement has exhibited possible approaches for sludge reduction during biological wastewater treatment. It is well known that increasing solids retention time (SRT) and decreasing sludge loading rate can reduce sludge production in aerobic wastewater treatment processes (Van Loosdrecht et al., 1999). The energy available to microorganisms is decided by substrate supply. On the other hand, endogenous respiration is the autodigestion of biomass. The major advantage of the endogenous metabolism is that the incoming substrate could be finally respired to carbon dioxide and water, while results in a lower biomass production (Martinage et al., 2000). By increasing biomass concentration in aeration tank, it would be possible to reach a situation in which the amount of energy provided equals with the maintenance demand. A relationship was presented to describe substrate utilization for maintenance and biomass production in substrate limited continuous microbial cultures (Low et al., 1999). Results showed that the biomass reduction occurred, i.e. biomass reduction by 44%, when the biomass concentration was increased from 1,700 to 10,300 mg L^{-1}. On the opposite, it is impossible to increase the sludge concentration significantly in CAS processes by means of sedimentation. However, membrane bioreactor (MBR) can be operated at long SRT even complete sludge retention which can be controlled independently from hydraulic retention time (HRT) by membrane filtration. By applying MBRs to the treatment of real sewage, Laera et al. (2005) reported that sludge yield values could be close to zero under the organic loading rate (OLR) around 0.07 kg COD kg VSS^{-1} d^{-1} during 180 days of continuous operation. The results showed that biomass evolved towards a minimized maintenance OLR, resulting in proportionality between the OLR and the equilibrium biomass concentration. At present, the sludge concentrations in MBR typically vary from 15,000 to 20,000 mg L^{-1} (Rosenberger et al., 2000). However, problems encountered at long SRT operation for MBR are poor oxygenation leading to increased aeration cost, and membrane fouling which requires frequent membrane cleaning and replacement. It is therefore not feasible to operate MBR with complete sludge retention in practice, and there should exist a minimal rate at which excess sludge is wasted in order to keep an optimal range of sludge concentration in MBR.

1.2.2 Predation on bacteria

A biological wastewater treatment process can be regarded as an artificial ecosystem, and activated sludge is an ideal habitat for several organisms (protozoa and metazoa) other than bacteria. One way to reduce excess sludge production is to increase the number of high trophic level such as protozoa and metazoa in activated sludge processes. These organisms predate on bacteria whilst decomposition of substrate remains unaffected. During transfer from a low trophic level to a high level, energy is lost due to inefficient biomass conversion (Ratsak et al., 1996). The protozoa can be divided into four groups: ciliates (free swimming, crawling and sessile), flagellates, amoeba, and heliozoan (Eikelboom, 2000). The metazoa consist normally of rotifera and nematoda. Other metazoa, such as Aeolosomatidae and Naididae, occur at a low number or occasionally as a bloom. Among these mini-animals, bdelloid rotifers (including *Philodina sp.)* and *Aeolosoma hemprichi sp.* which is one kind of worm have been reported to make great contributions to sludge reduction (Lapinski et al., 2003; Song et al., 2009). Moreover, a major worm bloom resulted in a low sludge volume index (SVI), lower energy consumption for oxygen supply and less sludge disposal (25-50% sludge reduction) (Wei et al., 2003). For enhancing the predation effects, a two-stage system was developed for sludge reduction (Ghyoot et al., 2000). The first stage (the bacterial stage) was operated as a chemostat at a short SRT to induce dispersed bacterial growth. The second stage was designed as a predator stage with a long SRT for growth of protozoa and metazoa.

1.3 Biological nitrogen removal

Biological nitrogen removal consists of two separate reactions to convert ammonia nitrogen (NH_4-N) to gaseous molecular nitrogen (N_2). The first reaction (nitrification) is the oxidation of the NH_4-N to nitrite-nitrogen (NO_2-N) and then to nitrate-nitrogen (NO_3-N) by ammonia-oxidizing bacteria (AOB) and nitrite-oxidizing bacteria (NOB), respectively. Typically, the NO_2-N concentration found in the bulk solution is less than 0.2 mg L^{-1}, because the rate of the whole process is limited by AOB. The second reaction (denitrification) involves the reduction of NO_3-N to molecular nitrogen. BOD is required for denitrification as electron donor and organic carbon source. Denitrification also produces alkalinity and can restore up to 50% of that was destroyed during the nitrification process. Many microorganisms are capable of denitrifying NO_3-N in the anoxic zone, but the most

commonly observed bacteria are *Pseudomonas* species. Münch et al. (1996) observed that the denitrification rate depends on the dissolved oxygen (DO) levels, they also found that the denitrification rate decreased gradually with time during aeration periods in sequencing batch reactors (SBRs). It has been reported that some species may be able to nitrify and denitrify under aerobic conditions. Gupta et al. (1999) reported that *Thiosphaera pantotropha sp.* is able to nitrify NH_4-N and denitrify NO_3-N using organics in synthetic sewage as a carbon source under aerobic conditions.

The following conditions are required for the occurrence of denitrification.

- Presence of NO_3-N or NO_2-N. These compounds are normally produced by nitrification and serve as the terminal electron acceptor in denitrification.
- Absence of DO. The presence of oxygen prevents the formation of the enzyme necessary for the substitution of nitrate for oxygen as the terminal electron acceptor.
- A facultative bacterial mass. Mass of organisms must be present with the necessary enzyme system to use nitrate instead of oxygen as the terminal electron acceptor.
- Presence of a suitable electron donor, or energy source. The carbonaceous energy sources for denitrification are internal (organic material naturally present in the wastewater), external (e.g. methanol added to the denitrification stage of the process), or self generated (nutrients released though the death of organisms in the process) substrates.

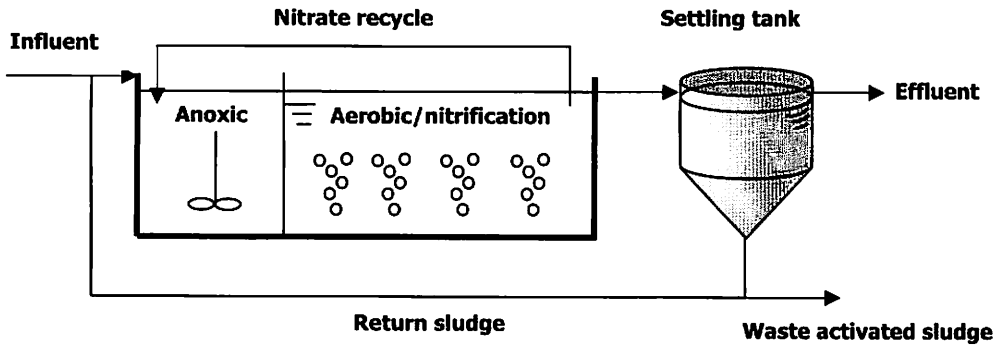

Fig. 1-2 Schematic of the MLE process.

The Modified Ludzak-Ettinger (MLE) process is one of the widely used conventional nitrogen removal processes in municipal wastewater treatment (Gayle et al., 1989). The

process consists of an anoxic tank followed by the aeration tank where nitrification occurs. NO_3-N produced in the aeration tank is recycled back to the anoxic tank (Fig. 1-2). Because the influent organic substrate serves as the electron donor for oxidation-reduction reactions using NO_3-N, the process is termed substrate denitrification. Since the anoxic process precedes the aeration tank, the process is also known as preanoxic denitrification.

Although this process has been widely used and proved effective, the problems of huge space requirement and relatively complicatedly operational control of this process still remain.

1.4 Integrated fixed-film activated sludge process

For enhancing the pollutants removal performance or upgrading the CAS process, several kinds of packing materials have been developed. These packing materials may be suspended in the activated sludge mixed liquor or fixed in the aeration tank. A term used to specify these processes is integrated fixed-film activated sludge (IFAS) process (Sen et al., 1994; Randall et al, 1996; Sriwiriyarat et al., 2005). These processes are intended to enhance the activated sludge process by providing a greater biomass concentration in the aeration tank and thus provide the potential to reduce the tank size requirements. Advantages claimed for these process enhancements are as follows:

- Increased treatment capacity
- Greater process stability
- Reduced sludge production
- Enhanced sludge settleability
- Reduced solids loadings on the sludge settling tank
- No increase in operation and maintenance costs

The IFAS process has received widespread acceptances in the engineering community as the most economical new way to expand wastewater plant capacity without building new aeration basins or clarifiers. It also can be used to increase treatment levels at existing plants in order to meet new permits for ammonia or total nitrogen. The IFAS process can provide a high surface area for attached growth by packing media into the aeration basins of an existing

activated sludge. Shock load and toxic load problems can be minimized in this process due to the existence of large amount of biomass on the integrated media. This increases the SRT without increasing the mixed liquor concentration in the system and overloading the secondary clarifier. These are just the potential problems with CAS systems. However, suspended type treatment systems are more flexible than fixed film systems for meeting higher effluent quality standards. The IFAS process is a hybrid system which provides both stability and flexibility.

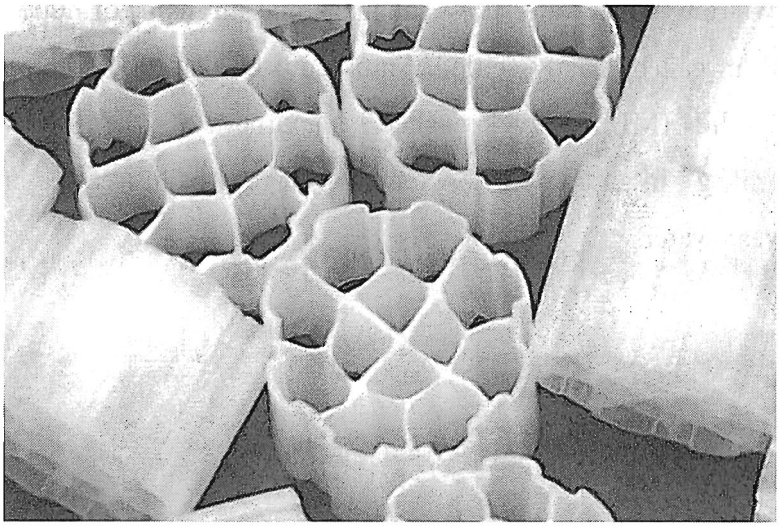

Fig. 1-3 Close-up of bioportz® suspending IFAS media.

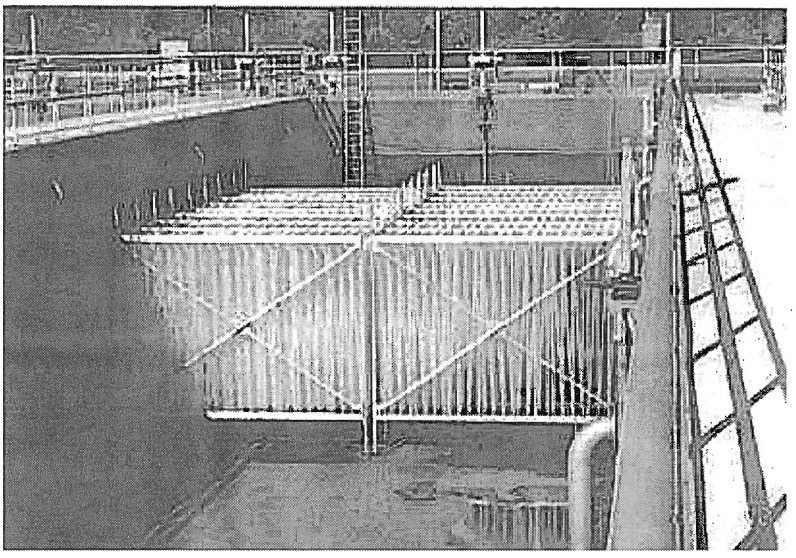

Fig. 1-4 Ringlace® installation in field.

There are now more than dozen different variations of IFAS processes. For suspended

packing IFAS processes, typicall examples include the captor®, linpor®, kaldnes® and bioportz® (Fig. 1-3) processes. For fixed packing IFAS processes, typical examples are the ringlace® (Fig. 1-4), biomatrix®, bio-2-sludge®, and submerged rotateing biological contactors (RBC) processes.

The swim-bed technology (Fig. 1-5) using biofringe® (BF, NET, Japan) carriers (Rouse et al., 2004) is exceptionally effective for wastewater treatment, especially for application as fixed packing aerobic IFAS processes. The "swimming motion" of the biofilm attached on BF carrier created by strong wastewater flow enhances mass transfer of nutrients to biofilm and the detached biofilm can be easily settled. Yamamoto et al. (2006) reported that the mixed liquid suspended solids (MLSS) level reached a maximum of 16,800 mg L^{-1} with low SVI values below 50 mL g^{-1} in an aerobic IFAS swim-bed reactor in treating anaerobic digestion liquor of swine wastewater.

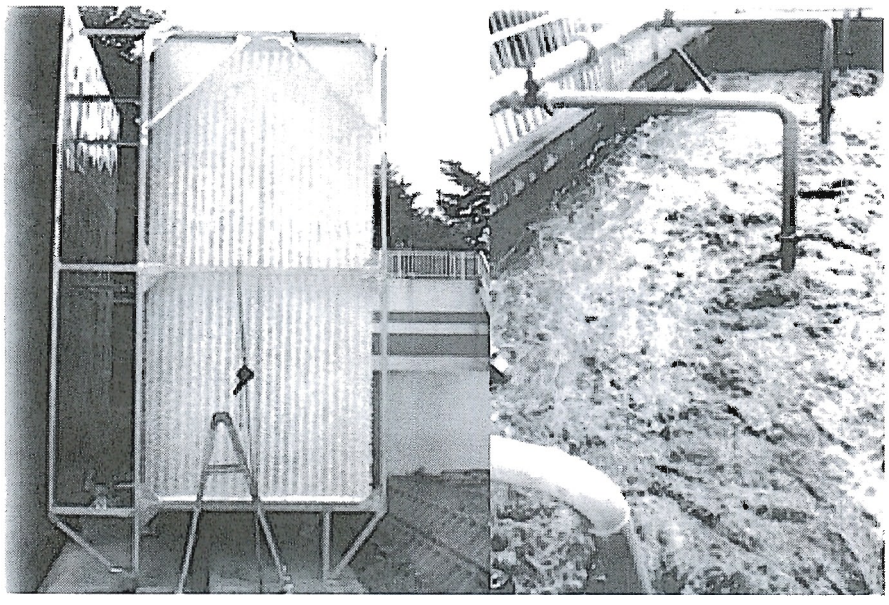

Fig. 1-5 Biofringe® installation in swim-bed aerobic tank.

1.5 Simultaneous nitrification and denitrification

Recent studies have revealed that nitrification and denitrification processes can occur concurrently in the same reactor (Halling-Sørensen et al., 1996). This process has been termed as simultaneous nitrification and denitrification (SND). Such processes exclude the need for

two separate reactors or intermittent aeration, thus result in a smaller footprint for the treatment plant and simplify the treatment system (Li et al., 2008). Compared with nitrogen removal through conventional nitrification and denitrification (such as the MLE process), SND provides several advantages: (1) it eliminates the need for either two separate tanks operated in serial or intermittent aeration operation in a single tank, thus continuous effluent output can be achieved with a smaller footprint (Guo et al., 2005); (2) it utilizes less carbon source and reduces sludge yield (Turka et al, 1987); (3) it requires less alkalinity demand which is consumed during nitrification but produced during simultaneous denitrification (Chang et al, 1999); (4) it consumes less energy due to the reduction in aeration requirement (Collivignarelli et al, 1999). Therefore, nitrogen removal via SND is of great economic interest for WWTPs.

1.6 Problem statement

The swim-bed technology used BF biomass carriers has previously demonstrated effectiveness in treatment of high-strength synthetic organic wastewater and partial nitritation for anaerobic digestion liquor (Rouse et al., 2004; Yamamoto et al., 2006; Qiao et al., 2008). However, no comparison among different acrylic fiber biomass carriers and different aeration modes in reactors have been evaluated. In addition, the involvements of this technology in the CAS process, conventional nitrogen removal enhancement, and SND performance have not been reported yet.

1.7 Research objectives

The objectives of this study were to investigate the applicability of the IFAS swim-bed technology in COD removal, nitrogen removal, sludge settleability improvement and SND performance in different treating systems.

Part one is to evaluate the characteristics of two kinds of acrylic fiber biomass carries, a net type carrier named biofix® (BX, NET, Japan) and BF carrier, in three parallel reactors and compare the treatment performance among them. The objective in this part is to ascertain the favorable acrylic fiber biomass carrier and the most suitable reactor configuration.

Part two is to investigate the feasibility of partial packing of BF carrier in improving the sludge settleability and enhancing the pollutant degradation performance of the activated sludge process.

Part three is to evaluate the enhanced nitrogen removal and excess sludge reduction by packing BF biomass carrier into the MLE system for treating synthetic domestic wastewater with low C/N ratio.

Part four is to investigate the nitrogen removal and excess sludge reduction in a novel system, which combined a down-flow anaerobic fermentation (AF) reactor and an aerobic IFAS swim-bed (SB) reactor in treating moderately high strength organic wastewater.

This dissertation was divided into six chapters, including this introduction part, four parts reporting the results of these experiments, and last part for the conclusions and recommendations.

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Chapter 2 Basic comparison of treating performance among different acrylic fiber biomass carriers under different aeration modes

2.1 Introduction

Aerobic submerged fixed-film processes (Mohamed et al, 1998) consist of three phases: a packing carriers, attached biofilm and wastewater. Biochemical oxygen demand (BOD) and NH_4-N removed from the liquid flowing past the biofilm are oxidized. Oxygen is provided by diffused aeration into the packing or by being predissolved into the influent. Aerobic fixed-film processes include down-flow packed-bed reactors, up-flow pack-bed reactors, and up-flow fluidized-bed reactors. The type and ratio of packing are primary factors that affects the performance and operating characteristics of submerged attached growth processes. Designs differ by their packing configuration (Dumonta et al, 2008) and inlet and outlet flow distribution and collection.

The BF biomass carrier consists of support filament and fringe yarns (diameter, ca. 3 mm), which are made of polyester and hydrophilic acrylic fibers, respectively. Combined with the hydrophilic characteristic, the BF biomass carrier has special configuration that the inner part is in high density and the outer part is in rarefaction, so that sludge could attach to the BF carrier easily, quickly and permanently. In the unique swim-bed reactor, flexing of the matrix induced by wastewater flow creates a "swimming" motion that enhances mass transfer of nutrients to the attached biofilm. Therefore, this technology is labeled as swim-bed technology.

The BF carrier with flexible matrix structure can allow for a large amount of biomass attached on it, which will result in a long food chain bio-system. With the regular, dense and granule-like structure under suitable conditions, the biofilm detached from the BF carrier in swimming-bed reactor is similar to aerobic granule sludge, resulting in a good settleability, sequentially contribute to a high MLSS concentration in reactors (Yamamoto et al., 2006). The great biomass attributing to thick biofilm and high MLSS concentrations could induce SRT, high volumetric organic loads (VLRs) and high stability against shock loads. Therefore, combining with the high activity of the biofilm due to special "swimming" motion, the

swim-bed process could demonstrate a high efficiency treatment of domestic wastewater (Rouse et al., 2004).

The biofix® (BX) biomass carrier is one kind of carriers that has net-type structure with the same materials as the BF carrier. The unique structure of BX carrier brings about a perfect biomass attachment.

The objective of this chapter is to evaluate the characteristics of BF and BX biomass carriers under two kinds of aeration modes and compare the treatment performance among them. Organic pollutants removal, nitrification performance and bacterial morphology were evaluated under different VLRs using moderately high-strength synthetic wastewater.

2.2 Materials and methods

2.2.1 Synthetic wastewater

The synthetic wastewater used as influent was prepared by the dilution of concentrated bonito extract (40 g L^{-1}) and peptone (60 g L^{-1}) mixture liquor only with tap water and without any buffer or nutrient additions. The main compositions and their concentrations of the synthetic influent wastewater are shown in Table 2-1. BOD concentration was about 67% of the COD concentration and the ratio among the BOD, nitrogen and phosphorus for influent was 100: 32.5:1.5.

Table 2-1 Basic characteristics of the influent

Item	Concentration (mg L^{-1})
COD	1500
BOD	1000
TN	150

2.2.2 Experimental setup

This comparative experimental system consisted of a full aeration reactor packed with the

BF carrier (R1), a cycle aeration reactor packed with the BF carrier (R2) and a cycle aeration reactor packed with the BX carrier (R3), which were all constructed of acryl resin, with effective volumes of 10 L (depicted in Fig. 2-1a). Separated by a vertical baffle, reactors had downdraft and updraft sections of 115×115 and 115×30 mm, respectively.

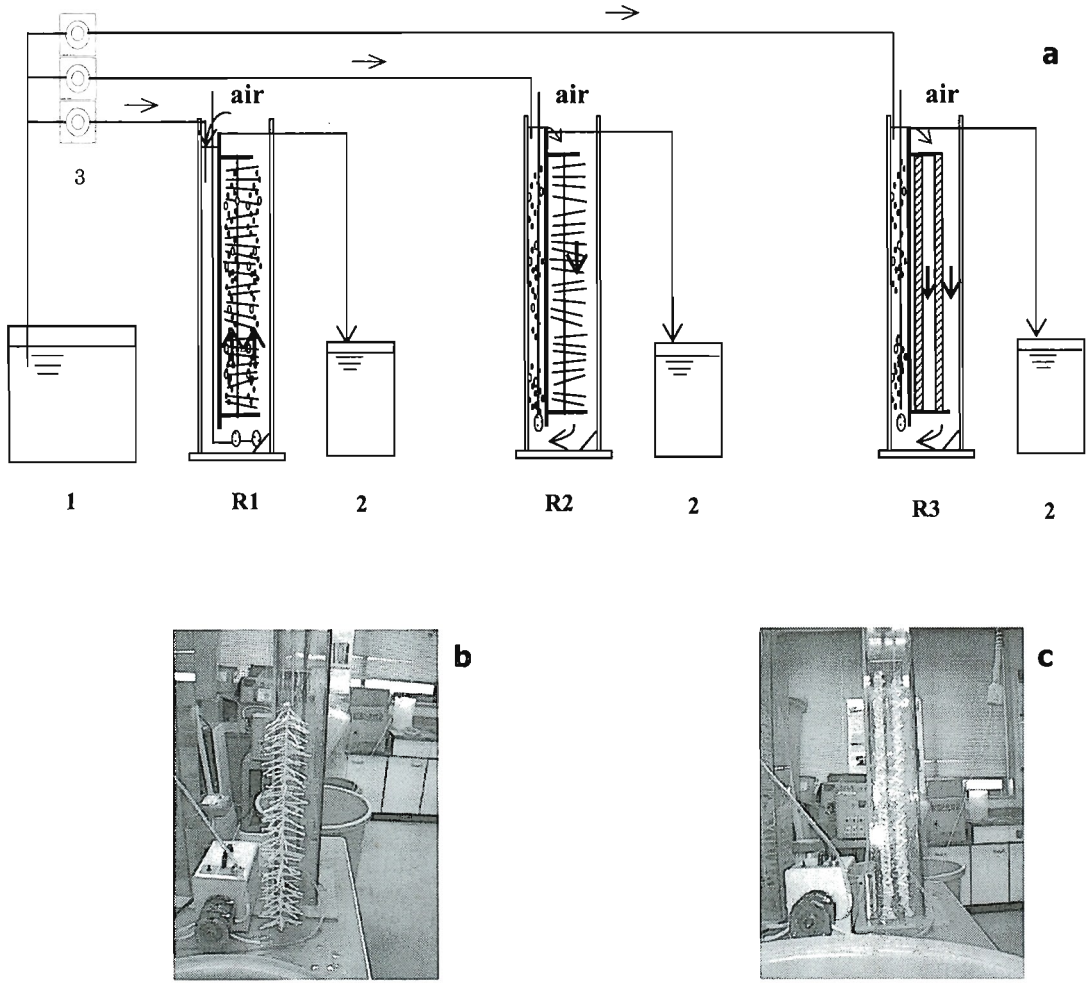


Fig. 2-1 (a) Schematic of the comparing system.
(1) influent tank; (2) effluent tank; (3) influent pump; (b) packing mode of BF in R1 and R2; (c) packing mode of BX in R3.

Support filament of the BF carrier was 500 mm in length. The specific surface area of BF carrier approximated 0.2 m^2 m^{-1}. Fringe yarns of the BF carrier were attached symmetrically, extending equal distances beyond each side of the support filament and twisting to give an even 3-dimensional distribution of the attachment matrix (about 30 circles per meter) (Fig. 2-1b). Two pieces of the BX (Fig. 2-1c) carrier were packed into the middle part of R3 with a

packing ratio of 50%. Each piece of the BX carrier was 500 mm in length and 20 mm in width with specific surface area of approximately 0.1 m^2 m^{-1}.

In R1, two sets of aerators were installed in the center of the bottom which were just under the BF packing. Thus, air could wash the BF carrier directly. In R2 and R3, air was introduced near the bottom of the updraft section, with a fixed flow rate of 10 L min^{-1} to aerate the wastewater and induce the down-flow hydraulic velocity of 20 cm s^{-1} in the downdraft section by providing density difference of the liquor, which led to a strong internal recirculation. There was a slanted edge (45°) baffle at the bottom of the reactor in order to direct the flow evenly from the downdraft section to updraft section.

2.2.3 System start-up and biofilm attachment

The reactors were initially inoculated with activated sludge from a lab scale fill-and-draw batch reactor under continuous aeration. The initial MLSS concentration was 730 mg L^{-1} for each reactor. During the sludge attachment period, the airflow rate for each reactor was regulated at 5 L min^{-1} to provide circulation velocity through the downdraft zone of 10 cm s^{-1} for quick biofilm attachment under relative weaker velocity conditions. After 24 hours of biofilm attachment, MLSS concentrations in R2 and R3 were extremely low (near zero) which indicated biofilm attachment was completed. Afterwards, air flow rate in each reactor was adjusted to 10 L min^{-1}, then the system was operated continuously in 3 months using the synthetic wastewater under increasing VLRs from 1 to 30 kg COD m^{-3} d^{-1} adjusted by increasing the HRT.

2.2.4 Experimental procedure and operational conditions

During 90 days of operation, the experiment was divided into seven runs according to various VLRs which were increased by changing HRT. The main operational conditions of this experiment were listed in Table 2-2.

During the period from Runs 1 to 3, VLRs were increased after both COD and BOD removal efficiencies were reached about 80% in all reactors. As for latter runs, the VLRs were increased weekly. Reactors were operated at constant temperature of 22 ± 2°C.

Table 2-2 Operational conditions during the whole experimental period

Period	Time (day)	VLR (kg BOD m^{-3} d^{-1})	Flow rate (L d^{-1})	HRT (h)
Run 1	1-20	1	10	24
Run 2	21-32	2	20	12
Run 3	33-55	3	30	8
Run 4	56-64	5	50	4.8
Run 5	65-71	10	100	2.4
Run 6	72-78	20	200	1.2
Run 7	79-86	30	300	0.8

2.2.5 Analytical methods

Concentrations of chemical oxygen demand (COD), BOD, NO_2-N, NO_3-N, suspended solids (SS), MLSS, MLVSS, and alkalinity were measured according to standard methods (APHA, 1995). COD concentration was measured by the closed reflux colorimetric method. TN concentration was determined by persulfate method. NO_2-N and NO_3-N concentrations were measured colorimetrically and spectrophotometrically, respectively. Alkalinity level was measured by titration. For soluble COD (s-COD) and soluble BOD (s-BOD), the final effluent samples were filtered (1 μm) before analysis. NH_4-N concentration was measured by the *o*-phenylphenol method (Kanda, 1995).

A digital pH meter (IM-22P, TOA Electronics, Japan) was used to measure pH. DO level in reactors was measured using a portable digital DO meter (OM-51, Horiba, Japan). Microorganisms in sludge of the reactors were observed by an electron microscope (Nikon Eclipse E600, Japan) with a digital camera (Nikon 4500, Japan).

2.3 Results and discussion

2.3.1 Comparison of sludge attachment performance among R1, R2 and R3

Biofilm attachment was accomplished within 24 hours after the start-up of this experiment, which was indicated by the very low MLSS concentrations in R2 and R3 (near zero).

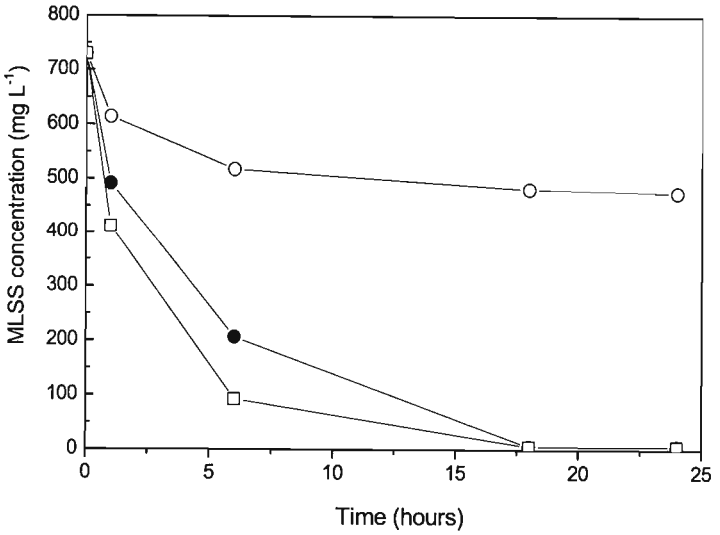


Fig. 2-2 Profiles of MLSS concentrations during startup of the experiment.

(○) R1; (●) R2; (□) R3.

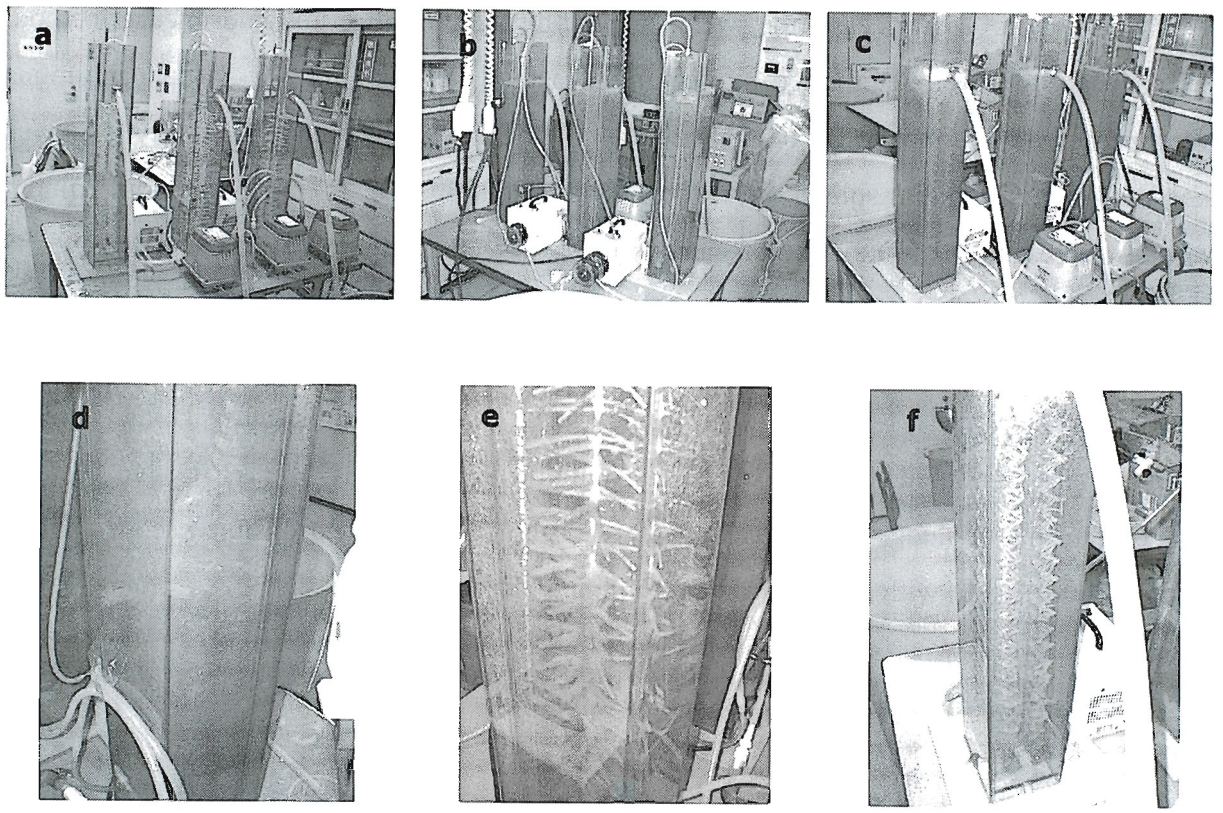


Fig. 2-3 Biofilm attachment performance in three reactors.

(a) reactors before biofilm attachment; (b) at the beginning of biofilm attachment; (c) 6 hour after biofilm attachment; (d) R1 at the end of biofilm attachment; (f) R2 at the end of biofilm attachment; (g) R3 at the end of biofilm attachment.

Fig. 2-2 shows the biofilm attachment performance for different carriers under two kinds of aeration modes. Comparing to the full aeration mode, the recycle aeration mode allowed for more and faster biomass attachment. At the end of attachment period, there was only 30% of seed sludge attached on BF carrier in R1. However, 80% of the seed sludge was attached on both BX and BF carriers within 6 h in cycle aeration reactors (R2 and R3). This results indicated that (1) the cycle aeration mode took obvious advantage over the full aeration mode in biofilm attachment; (2) BF and BX carriers attached sludge rapidly under suitable conditions due to their favorable configurations and hydrophilic characteristics. Moreover, the biomass attached on the BX carrier more rapidly than the BF carrier. At the end of biofilm attachment, almost no suspend activated sludges remained in R2 and R3. Furthermore, biofilm appeared much thicker along the carrier depth due to the down-flow current. Fig. 2-3 shows the biofilm attachment performance among the three reactors.

Following the biofilm attachment period, the influent feeding was started and the airflow rate was adjusted to 10 L min^{-1} during the whole experimental period.

2.3.2 Organic pollutant removal performance

Owing to the high effluent SS concentrations, effluent COD concentrations were much higher than s-COD concentrations. Treatment performances of all reactors were stable under low VLR of 1, 2 and 3 kg BOD m^{-3} d^{-1} (Runs 1 to 3). COD and s-COD removal efficiencies of all reactors were around 70% and 90%, respectively. There were not obvious differences among all reactors and this indicated that organic pollutants could be degraded easily under low VLRs irrelative of packing carriers and aeration modes. However, under high VLRs from run 4 to run 7, organic pollutant treating performance of R2 were better than those in other reactors (Fig. 2-4a, b). S-COD removal efficiencies of R2 maintained above 40% even under VLR of 30 kg BOD m^{-3} d^{-1} which was equal to a s-COD removal rate of 12 kg s-COD m^{-3} d^{-1}.

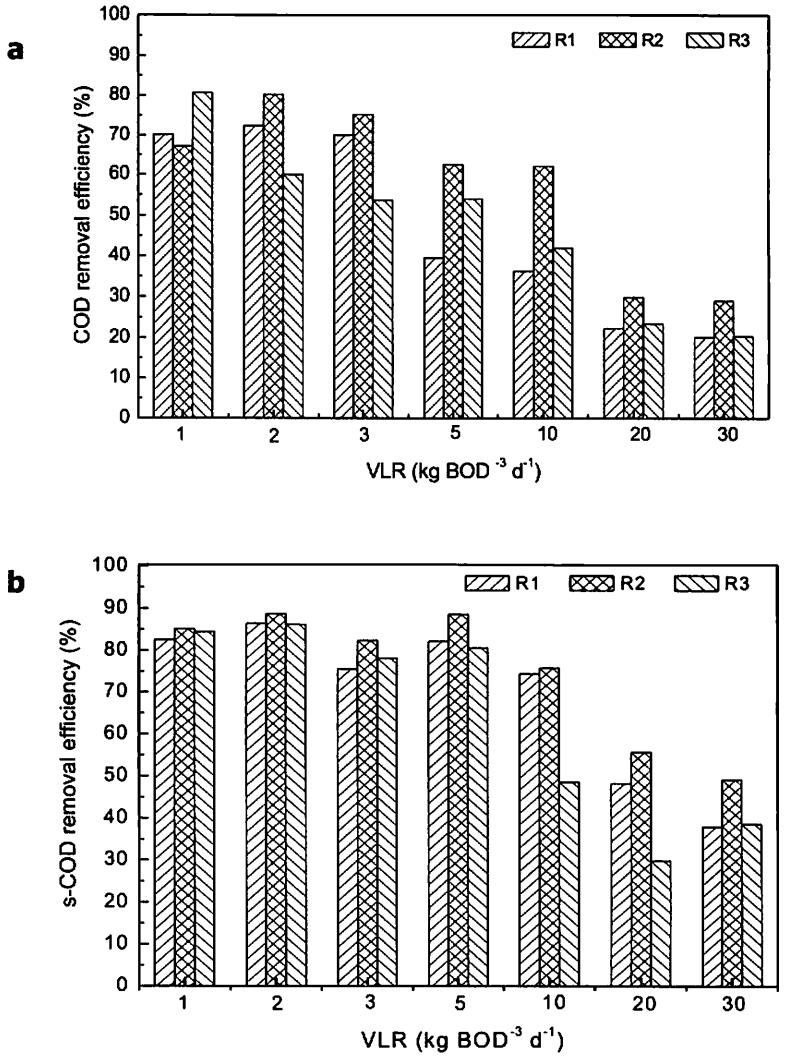


Fig. 2-4 Comparison of COD and s-COD removal performances.

(a) COD removal performance; (b) s-COD removal performance.

Fig 2-5 shows comparison of BOD and s-BOD removal performances for three reactors. Similar to COD removal, BOD removal had no obvious differences among three reactors. Effluent s-BOD concentrations of three reactors were approximate especially under low VLRs (Runs 1 to 3), with 90% s-BOD removal efficiencies. However, under high VLRs, attached biofilm in R1 was limited due to the aeration mode which contributed to high effluent SS and BOD concentrations compared with those in other reactors. On the opposite, BOD removal efficiencies of R2 were superior to other reactors due to its greater biomass attachment.

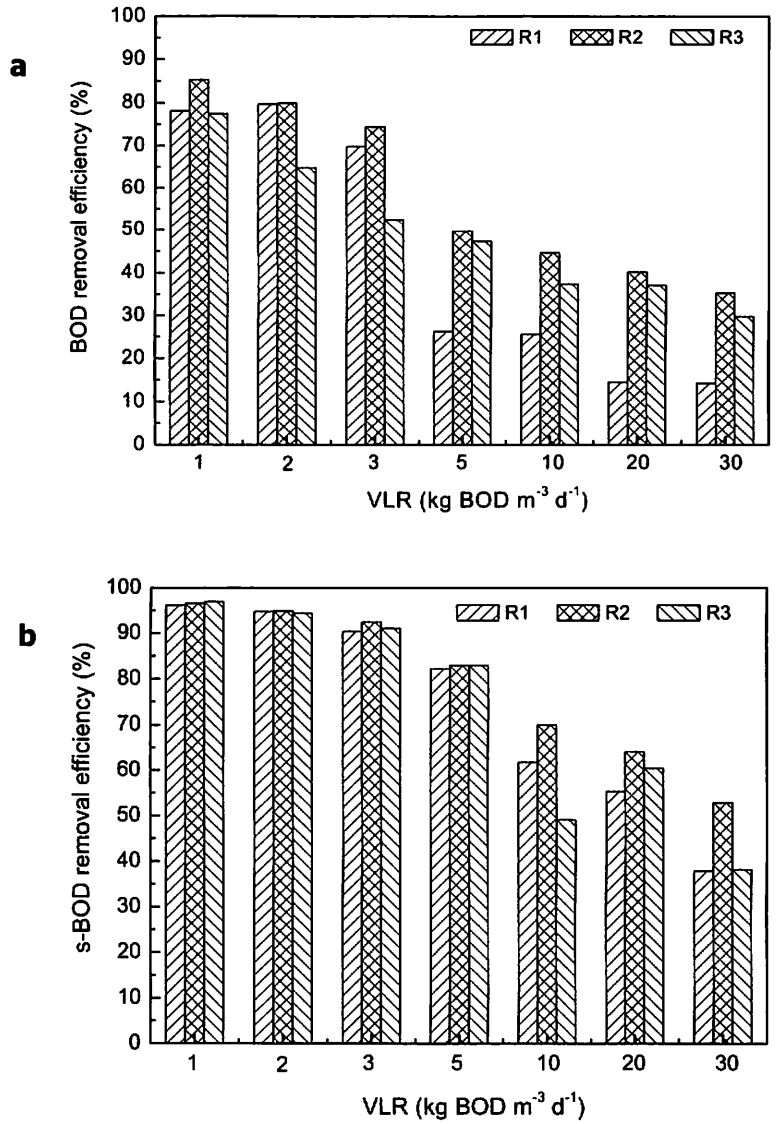


Fig. 2-5 Comparison of BOD and s-BOD removal performances among three reactors.

(a) BOD removal performance; (b) s-BOD removal performance.

Based on the COD and BOD removals, R2 took a priority for the removal of organic pollutant over the others, especially under high VLRs. As substrate is consumed, four distinct biomass growth phases develop sequentially: lag phase, exponential growth phase, stationary phase and death phase (Barker et al., 1997; Pan et al., 2008). Under VLRs of 1, 2 and 3 kg BOD m^{-3} d^{-1}, the biomass growth remained relatively constant over time, i.e. bacterial growth stayed on stationary phase. However, it was shifted to exponential growth phase with increasing VLRs. Because there was no limitation in substrate or nutrient supply during exponential growth phase, biomass were multiplying at their maximum rate. This led to high effluent SS concentrations. During the growth period under unlimited substrates and nutrients,

the only factor that affects the rate of exponential growth is temperature (Cleland et al., 2007; Baumann et al., 2004).

2.3.3 Nitrogen transformations

Profiles of NH_4-N concentrations in three reactors are shown in Fig. 2-6. Effluent NH_4-N concentrations had no obvious difference among three reactors. Under lower VLRs below 5 kg BOD m^{-3} d^{-1}, aerobic ammonification was performed well in all reactors. The decreases in NH_4-N concentrations in Run 1 were mainly due to the nitrification performance. The free ammonia (FA) concentrations in the reactors which were calculated by the equations presented by Anthonisen et al. (1996) were much higher than 1 mg L^{-1} (data not shown), which had been reported to be the inhibition level to nitrifiers (Anthonisen et al., 1996; Vadivelu et al., 2006). The high FA concentrations caused the high effluent NH_4-N concentrations without NO_X-N (NO_2-N and NO_3-N) production under VLRs of 5 kg BOD m^{-3} d^{-1} (Fig. 2-8). When VLRs were higher than 5 kg BOD m^{-3} d^{-1}, ammonification was not exhibited completely and effluent NH_4-N concentrations decreased gradually due to the short HRT (less than 4.8 h).

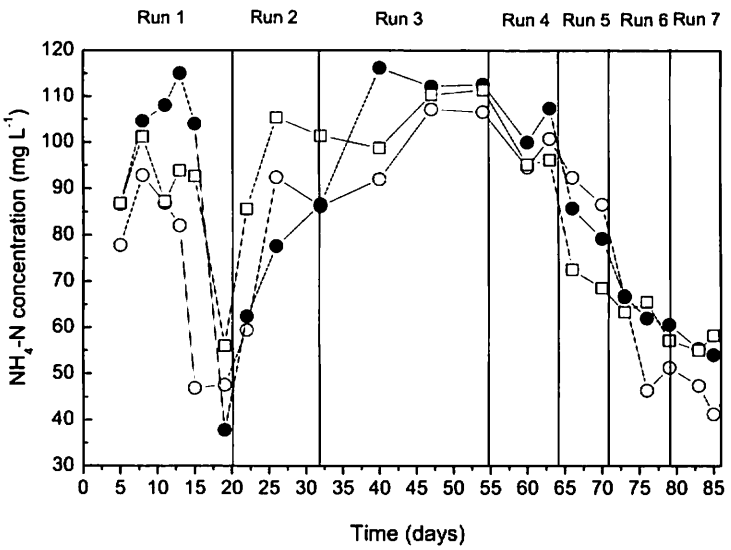


Fig. 2-6 Profiles of NH_4-N concentrations.

(○) R1; (●) R2; (□) R3.

The changing patterns in effluent pH values were similar to those observed in NH_4-N concentrations (Fig. 2-7). Under low VLRs, pH was changed mainly by the nitrification

performance. On the other hand, pH was decreased due to the inhibition of ammonification under higher VLRs.

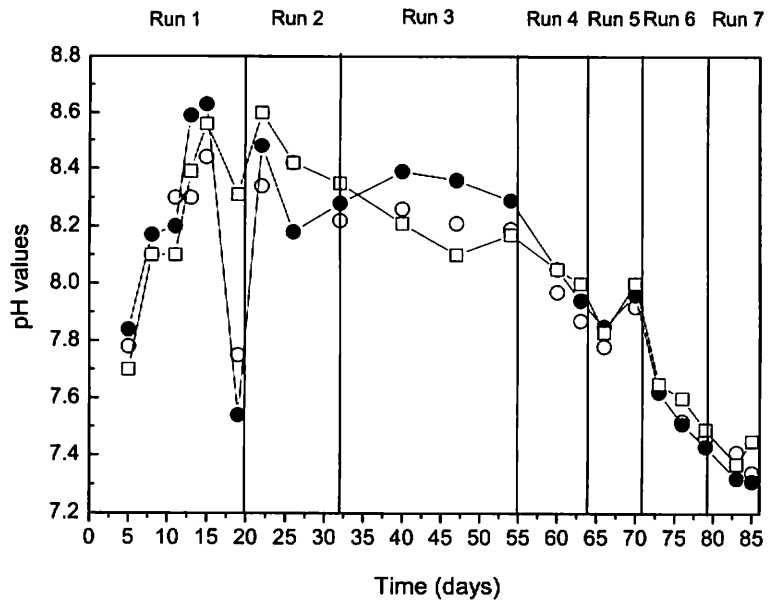


Fig. 2-7 Profiles of pH during the whole experimental period.

(O) R1; (●) R2; (□) R3.

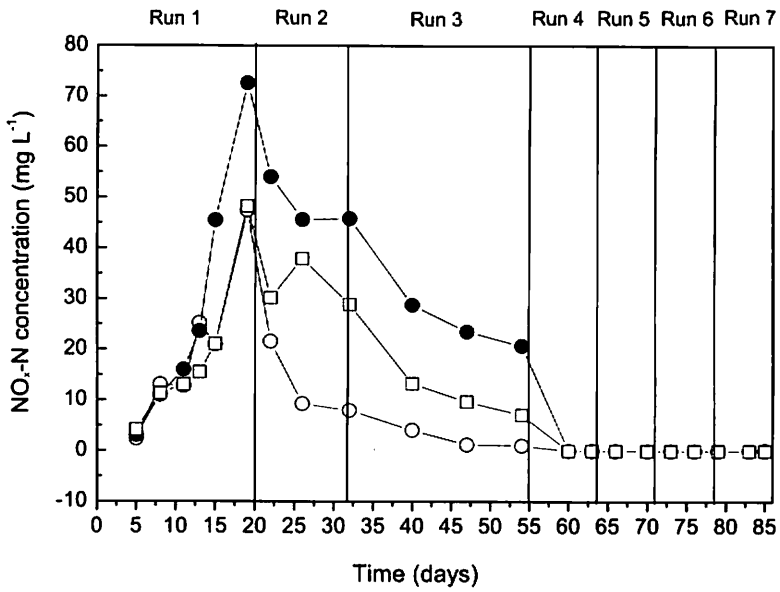


Fig. 2-8 Profiles of NO_X-N (NO_2-N and NO_3-N) concentrations during the whole experimental period.

(O) R1; (●) R2; (□) R3.

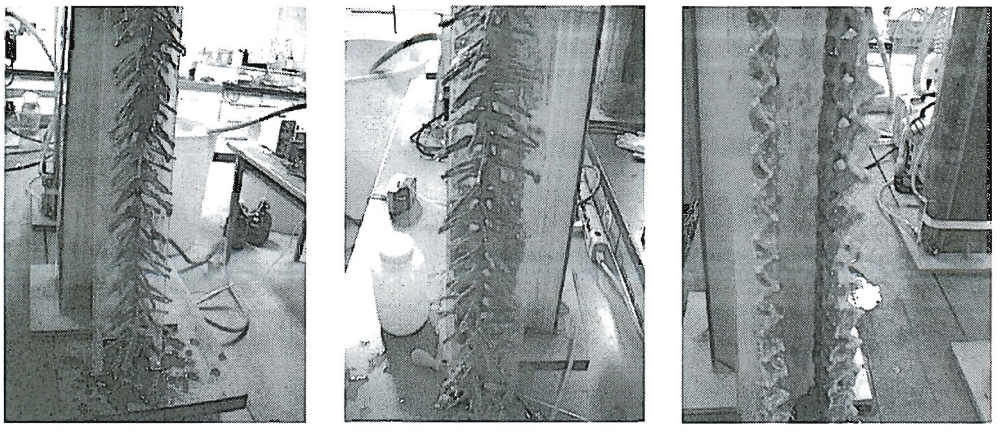
In this study, nitrification performance of each reactor was evaluated by the sum of NO_2-N and NO_3-N which reflected the activity of nitrifying bacteria. Nitrification process is a two-step process involving two groups of bacteria (Li et al., 2006; Gheewala et al., 2004). In

the first stage, NH_4-N is oxidized to NO_2-N by ammonia-oxidizing bacteria (AOB). Then NO_2-N is oxidized to nitrate by nitrite-oxidizing bacteria (NOB) in the second stage. Both of them are autotrophic bacteria with longer generation time than facultative and heterotrophic bacteria. Therefore, nitrification occurred under low VLRs (< 5 kg BOD m^{-3} d^{-1}) with long HRT (> 8 h) and NO_2-N was the predominant in oxidized nitrogens (data not shown). At the end of Run 1, the total amount of NO_2-N and NO_3-N in R2 reached to 70 mg L^{-1} (Fig. 2-8) which was 35% higher than that in R1 and R3, indicating a better nitrification efficiencies were obtained in R2. Under low VLRs (Runs 1 to 3), the changing tendencies of NO_2-N and NO_3-N concentrations among all reactors were similar. Nitrification efficiencies in R2 were better than that in other reactors. Following the increase in VLRs and decrease in HRT, effluent NO_2-N and NO_3-N concentrations decreased gradually. There was nearly no production of NO_2-N and NO_3-N when SRTs were decreased below 4.8 h (Run 4) in all reactors. DO concentrations in all reactors were maintained more than 1 mg L^{-1}. So that, the deterioration of nitrification could be related to the enhanced growth of heterotrophs rather than oxygen limitation. Overall, nitrification efficiencies of R2 were better than that of R1 and R3 under low VLRs below 5 kg BOD m^{-3} d^{-1}.

Commonly, nitrogen is biologically removed through microbial growth and nitrification-denitrification process. Because only aeration tanks were used in this study, TN was removed through microbial growth rather than denitrification pathway. TN removal efficiencies in all reactors were similar and only about 10-15% (data not shown).

2.3.4 Sludge characteristics

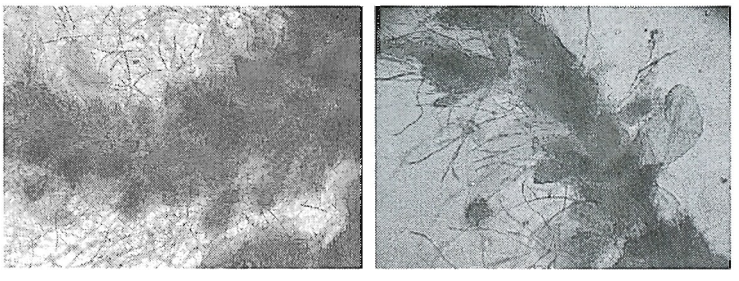
At the end of Run 7, attached biofilms on carriers were detached completely in three reactors and the total weights of activated sludge were determined. The weight differences between these values and the initial MLSS concentrations were considered to be the estimated weights of attached biofilms. Biomass amounts on carrier of three reactors were determined to be 6.1, 25.8 and 14.1 g, respectively. Biofilm biomass attached on BF carrier in R2 was nearly 2 times higher than that on BX carrier in R3 under the same operational condition. This could interpret the reason for better treatment performance for organic pollutants and nitrification in R2. High amount of biomass attached on BF carrier due to its excellent attachment capability was the main reason for good treatment performance in R2.



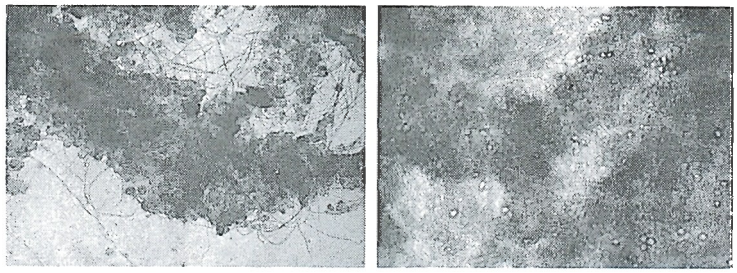
R1 R2 R3

Fig. 2-9 Photographs of biomass attached on biomass carriers at the end of the experiment.

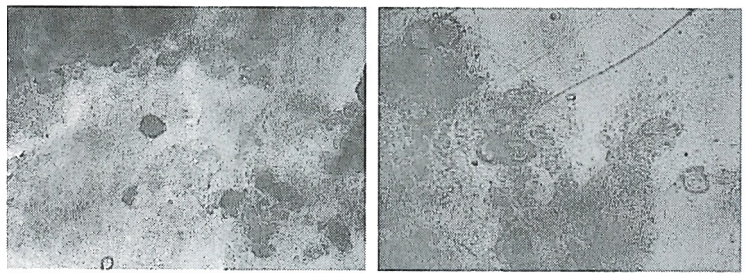
Under VLR of 1 kg BOD m^{-3} d^{-1}, filamentous microorganisms were appeared in all reactors. The amount of filamentous bacteria was small in R2 and sludge was more compact comparing with that sludge in other reactors. On the other hand, large number of filamentous bacteria were observed in R1 and R3. When VLR increased to 2 and 3 kg BOD m^{-3} d^{-1}, filamentous bacteria in R2 reduced gradually and floc forming bacteria (*Zoogloea sp.*) became abundant. Floc forming Zoogloea enhanced the sedimentation of activated sludge and contributed to a better effluent quality. In addition, *Epistylis sp.* appeared in R1 which was regarded to be the indicator of the better pollutant removal performance. Under low VLRs, the amount of biomass in R3 was less than that in other reactors. Following the increase in VLRs to 5 and 10 kg BOD m^{-3} d^{-1}, the number of *Epistylis sp.* in R2 were increasing. However, with VLR increased to 20 and 30 kg BOD m^{-3} d^{-1}, *Epistylis sp.* disappeared due to high organic pollutant concentrations and low DO levels. Treatment performances of all reactors became worse under such high VLRs, and a large number of tiny protozoa became abundant in all reactors.



R1

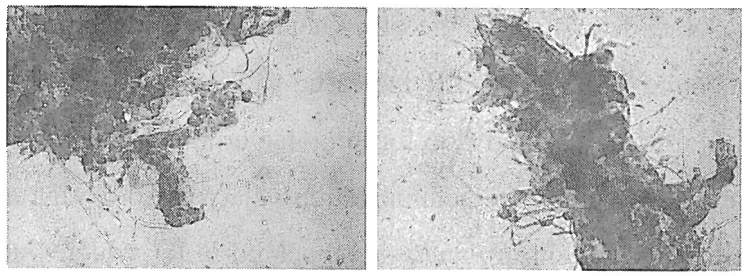


R2

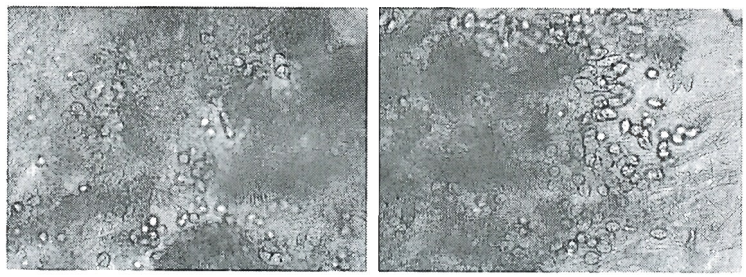


R3

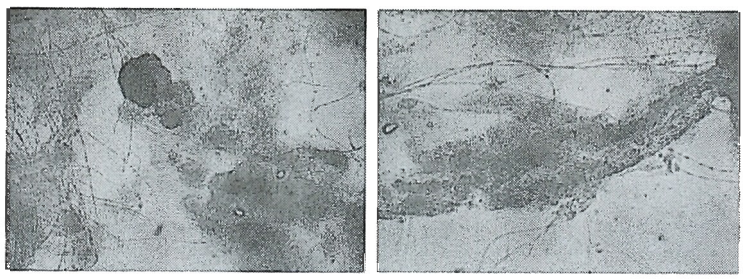
(a) 3 kg BOD m^{-3} d^{-1}



R1

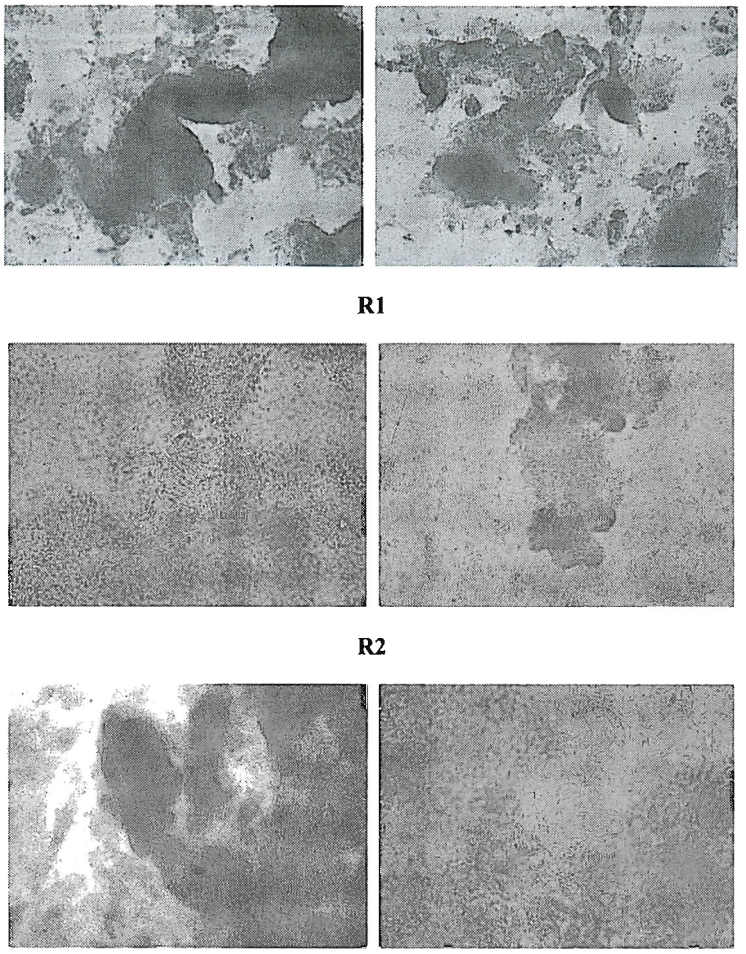


R2



R3

(b) 5 kg BOD m^{-3} d^{-1}



(c) 20 kg BOD m^{-3} d^{-1}

Fig. 2-10 Comparison of microscopic observations among three reactors under different VLRs.

(a) 3 kg BOD m^{-3} d^{-1}; (b) 5 kg BOD m^{-3} d^{-1}; (c) 20 kg BOD m^{-3} d^{-1}.

2.4 Conclusions

In this chapter, the characteristics of BF and BX biomass carriers and their pollutant treating performances were compared among three parallel reactors. COD removal efficiencies, nitrification efficiencies and microorganism morphologies were evaluated under different VLRs.

(1) Though the BX biomass carriers attached biomass a little more rapidly than the BF carrier, the BF carrier could attach more biomass than the BX carrier in continuous operation. At the end of this experiment, attached sludge biomass in R2 was about two times of that in R3 under the same operational condition. Meanwhile, comparing with full aeration mode in

R1, recycle aeration mode in R2 possessed the advantage of more and rapid biomass attachment.

(2) COD removal performances of three reactors were good and stable under low VLRs. While under high VLRs, treatment performance of R2 was better than those for other reactors owing to high amount of attached biomass on BF carrier.

(3) Nitrification efficiencies of R2 were better than those in R1 and R3 under low VLRs below 5 kg BOD m^{-3} d^{-1}. Under high VLRs, the transformations of influent organic nitrogen in three reactors were mainly characterized by ammonification without NO_2-N and NO_3-N production. Ammonification also deteriorated with the increase in VLRs.

(4) Filamentous bacteria appeared in all reactors under the low VLRs. Amount of filamentous bacteria was relatively low and dense activated sludge was obtained in R2. Filamentous bacteria disappeared gradually and floc forming bacteria (*Zoogloea sp.*) was abundant in all reactors with the increase in VLRs.

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Chapter 3 Application of swim-bed technology to enhance treatment performance of activated sludge process

3.1 Introduction

There still exist some unsolved problems in CAS process, such as difficulties in solid-liquid separation caused by sludge bulking, large space requirement for construction of a settling tank, high excess sludge production and relatively low pollutant removal capacity (Cornelissen et al., 2002). Fixed bed and fluidized-bed attached-growth processes have demonstrated high sludge retention capability, reduced sensitivity to toxicity, co-existence of aerobic and anoxic metabolic activities, and reactor compactness (Pastorelli et al., 1997). However, maintenance associated with solids accumulation and possible packing plugging can affect the performance of fixed bed process. Careful inlet and outlet designs for good flow distribution and higher power to keep fluidized condition are also required in fluidized-bed process (Wang et al., 1986; Fox et al., 1988).

Using the swim-bed technology, high reactor MLSS concentrations from 10 to 15 g L^{-1} (excluding the sludge biomass of biofilm), with good sludge settling characteristics, have been achieved without sludge bulking under high VLRs from 3 to 5 kg BOD m^{-3} d^{-1} with excellent organic pollutant removal performance (Cheng et al., 2005). Furthermore, long SRT can be easily maintained with sludge recycling from settling tank to swim-bed reactor, which reduces sludge production due to long food chain system and predation on bacteria. This biological wastewater treatment method can save operational costs and avoid potential harmful environmental impacts in comparison with physical or chemical wastewater treatment methods.

In this chapter, the swim-bed technology was applied in an activated sludge system. The main purpose was to investigate the possibility of improving sludge characteristic by partially packing BF carrier in a plug-flow activated sludge system and to evaluate the treatment capability in terms of organic matter removal under high loading rate conditions.

3.2 Materials and methods

3.1.1 BF biomass carrier

The BF carrier consisted of support filament and fringe yarns (diameter, *ca.* 3 mm), which were made of polyester and hydrophilic acrylic fibers, respectively. The BF had special structure such that the core point had high density while the outside was loosely knit so that sludge could attach to BF easily and quickly. The fringe yarns waved due to the effect of high velocity water current as shown in Fig. 3-1. Since the movement of BF imitated swimming motion, we named this treatment process as swim-bed process.

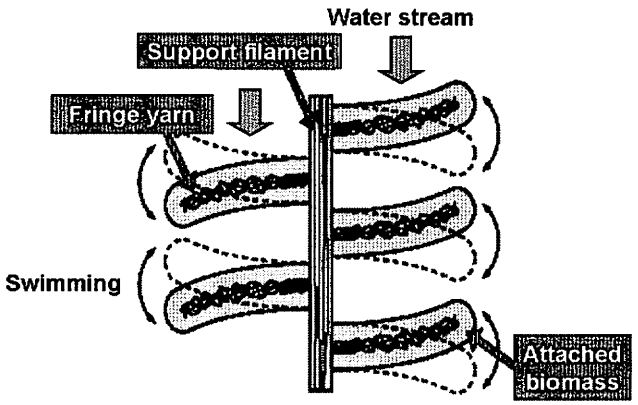


Fig. 3-1 Schematic of BF and swimming motion.

3.1.2 Experimental setup and operational conditions

A four-celled reactor made of acryl resin and having a total volume of 34.8 L was used in this study as shown in Fig. 3-2. The first reactor cell with a working volume of 10.8 L was packed with 0.5 m length of the BF carrier. The estimated packing volume of the BF carrier was approximately 5 L resulting in a packing ratio of about 15% of the total reactor volume. The working volumes of the other 3 cells were 8 L, each (named as AS1, AS2, AS3). The cells were initially seeded with activated sludge from lab-scale fill-and-draw batch reactor and the initial MLSS concentration was set at about 4,000 mg L^{-1}. The air flow rate was fixed at 10 L min^{-1} and the reactor temperature was maintained at 25°C. A settling tank, made of acrylic resin, with a effective volume of 2 L was also used. Settled sludge in the settling tank was stirred at 4 rpm and returned to the BF reactor at a 100% recycle rate.

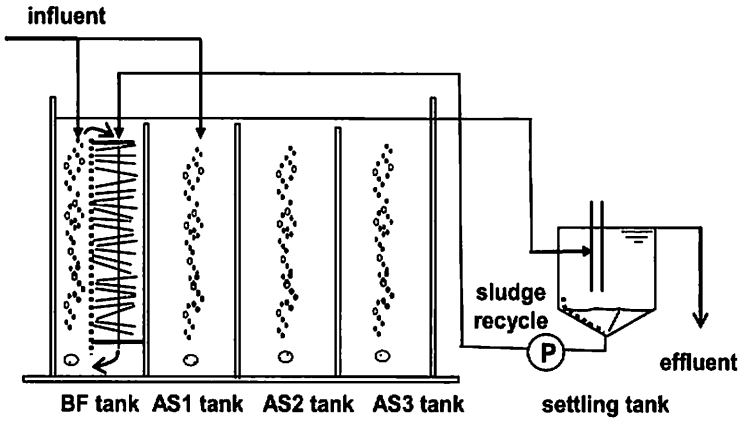


Fig. 3-2 Schematic of the experimental setup

The synthetic influent wastewater consisting of meat extract and peptone mixture was prepared as a stock solution and diluted with tap water to obtain the desired concentration. The total nitrogen (TN) was 10% of the soluble COD (s-COD), while the soluble 5-day BOD ($s\text{-}BOD_5$) was 67 % of the s-COD. $NaHCO_3$ (1M) was used as the buffer solution. The VLR was increased by adjusting the influent concentration and the HRT. In order to mitigate the direct influence of VLR on the BF reactor cell, the influent was split and was introduced to AS1 reactor cell on day 23. On day 200, pH controller was installed in the AS2 reactor cell to ensure that the effluent pH was more than 5 by adding 1.25 N NaOH solution.

3.1.3 Analytical techniques

Viscosity was measured by B-viscosity meter (BL; Toki, Tokyo). Floc size distribution analysis was conducted using a laser diffraction particle size distribution analyzer (Horiba LA-920, Kyoto, Japan). TN concentration was measured according to standard methods (APHA, 1995).

PCR-DGGE was performed by enzyme bacteriolysis methods using 16S rDNA for bacterial community analysis. Sample taken from mixed liquor in the BF reactor cell was centrifuged at 9,000×g for 10 minutes and these harvested cells were used for the extraction of DNA. DNA extraction and purification was performed 7 times with Phenol/Chloroform/Isoamyl alcohol. PCR reaction mixture was composed of 30 ng of the extracted DNA, 0.5 unit of KOD-plus-DNA polymerase (Toyobo, Osaka, Japan), and 5 pmol of each forward and reverse

primer. The forward primer was 357F with GC-clamp while the reverse primer was 534R (Muyzer et al., 1993). PCR conditions were as follows: 94°C for 2 min with thermal cycling consisting of 9 cycles each of 15 sec. at 94°C, 30 sec. at 60°C, and 30 sec. at 68°C. Electrophoresis was performed using Bio-Rad Ccode® system at 100 V and 60 °C for 16 h according to manufacturer's protocol. After electrophoresis, the gel was stained with SYBR Gold Nucleic Acid Gel Stain for 30 min. and DNA bands were verified using EM-20E UV-transilluminator (UVP, Upland, CA, USA). Besides the DGGE analysis, the amplified DNA with 357F and 534R primers were also cloned as follows: the amplified products were inserted into *Hinc* II site of pBluescript II KS^+ (Strantagene) and transformed *E.coli.* DH10B. Plasmid DNA was extracted and used to sequence the inserted DNA by dideoxy sequencing method with M13 and M4 primers. The obtained DNA sequences were analyzed by homology search with NCBI BLAST program.

Other analytical methods are the same as those described in Chapter 2.

3.1.4 Calculation methods

In this chapter, nitrification efficiency (NE) was calculated according to Eq. (3-1).

$$NE\ (\%) = 100 \times (NO_3\text{-}N_{eff} + TN_{inf} - TN_{eff})/TN_{inf} \quad (3\text{-}1)$$

The SRT and food to microorganism (F/M) ratio were calculated from equations (3-2) and (3-3), respectively for experimental run with MLSS concentrations of more than 10,000 mg L^{-1}:

$$SRT = (VX_a)/(QX_e) \quad (3\text{-}2)$$

$$F/M = (Q_sC_s)/(VX_a) \quad (3\text{-}3)$$

where V = the reactor volume (L); X_a = the average MLSS concentration in reactor (mg L^{-1}); Q_s = the flow rate (L d^{-1}); X_e = the average effluent SS concentration (mg L^{-1}); C_s = the average influent s-COD concentration (mg L^{-1}).

The observed sludge yield (Y_{obs}) was calculated according to Eq. (3-4) for the operational period when the MLSS concentration was more than 10,000 mg L^{-1}.

$$Y_{obs} = (W_{end} - W_{start} + \sum QX_e + \sum W_{out})/\sum Q(S_o - S_e), \quad (3\text{-}4)$$

where W_{end} = total amount of biomass at the end of each VLR, including suspended and

biofilm biomasses in all reactor cells and that in the settling tank (mg); W_{start} = total amount of seed biomass at the beginning of each VLR, (mg); Q = influent flow rate (L d^{-1}); X_e = effluent SS concentration (mg L^{-1}); W_{out} = total amount of excess sludge withdrawal (mg); S_o = influent COD concentration (mg L^{-1}); S_e = effluent COD concentration (mg L^{-1}).

3.3 Results and discussion

3.3.1 General treatment performance

The experimental reactor was operated for a period of 382 days. S-COD VLR was increased stepwise by increasing the influent s-COD concentration and the influent flow rate as shown in Fig. 3-3a. On day 95, sludge was washed out from the reactor because of clogging of connection tubes, triggering a VLR decrease for about six days (days 95-100). However, after the clogging was cleared, the VLR was increased gradually reaching 4.5 kg s-COD m^{-3} d^{-1}. High s-COD removal rates were consistently achieved throughout the experimental period at VLRs ranging from 0.8 to 4.5 kg s-COD m^{-3} d^{-1}. The average s-COD removal rate was around 96.0%. An average s-COD removal rate of 96.5% was obtained even under the highest VLR of 4.5 kg s-COD m^{-3} d^{-1}. In addition, the average s-BOD_5 removal rate was 99.1%, which indicated that the influent biodegradable organic matter was almost removed. Fig. 3-3c shows the time courses of nitrogen concentrations and nitrification rates during the experimental period. All of the influent nitrogen was composed of organic nitrogen. From the onset of the reactor operation, high effluent NO_3-N concentrations were observed. On days 202 and 232, high effluent NH_4-N concentrations over 100 mg L^{-1} were observed due to the high pH levels (7.98-8.28) caused by failure of pH control. Soon after the pH controller was fixed, the system recovered producing high NO_3-N and low NH_4-N concentrations in the effluent. Accumulation of nitrite was not observed with average effluent NO_2-N concentration of about 0.12 mg L^{-1}.

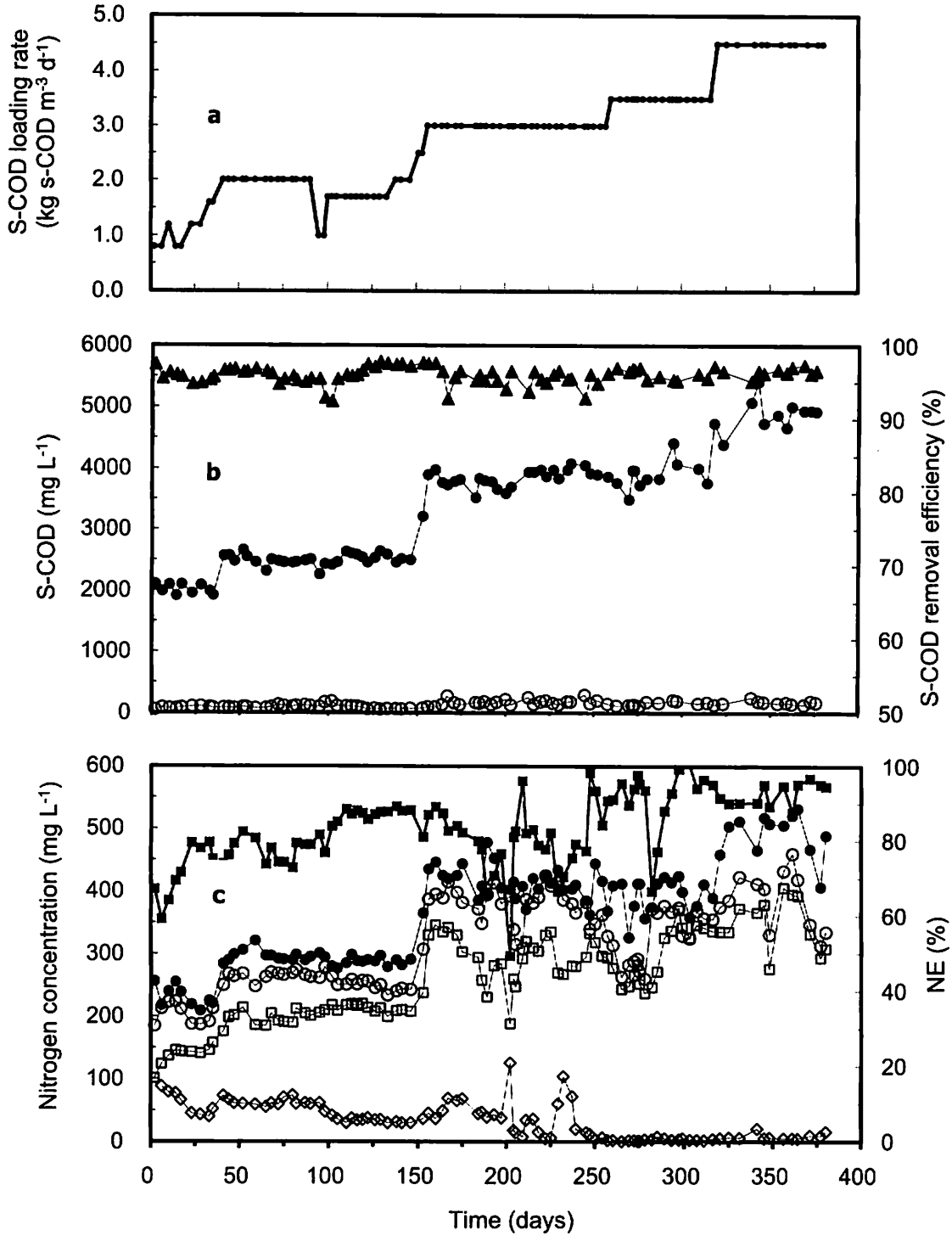


Fig. 3-3 Treatment performance results during the experimental period.

(a) profiles of s-COD VLR; (b) profiles of s-COD concentrations and removal rates: (●) influent s-COD; (○) effluent s-COD; (▲) s-COD removal rate; (c) Profiles of nitrogen concentrations and nitrification rates: (●) influent TN; (○) effluent TN; (□) effluent NO_3-N concentrations; (◇) effluent NH_4-N concentrations; (■) NE.

As shown in Fig. 3-3c, high NE was achieved during the experimental period with an average value of 83.6%. This high NE, combined with high effluent NO_3-N concentrations, demonstrated successful accomplishment of nitrification in the reactor. Compared to the results for one-through reactor reported by Rouse et al. (2004), the nitrification treatment results of this study are more promising. This could be attributed to the long SRT along with the availability of a settling tank, which was efficient for preventing the washout of nitrifying bacteria. High NEs were also achieved under low effluent pH levels (5.2 ± 0.6) before installing the pH controller. Nevertheless high NO_3-N concentrations were maintained. Considerable simultaneous denitrification was not observed. However, it will be possible to get high nitrogen removal by installing denitrification reactor in this treatment system. With the increase in VLR, it was found that the average TN removal efficiencies also increased, e.g., 8.76%, 16.7% and 21.7% at VLR of 3.0, 3.5 and 4.5 kg s-COD m^{-3} d^{-1}, respectively. This could be attributed to the formation of anaerobic zone on the BF media due to the extremely high reactor MLSS concentrations. Based on the results of this study, simultaneous high s-COD removal and high NE could be obtained in this type of system even at high VLRs.

3.3.2 Sludge characteristics

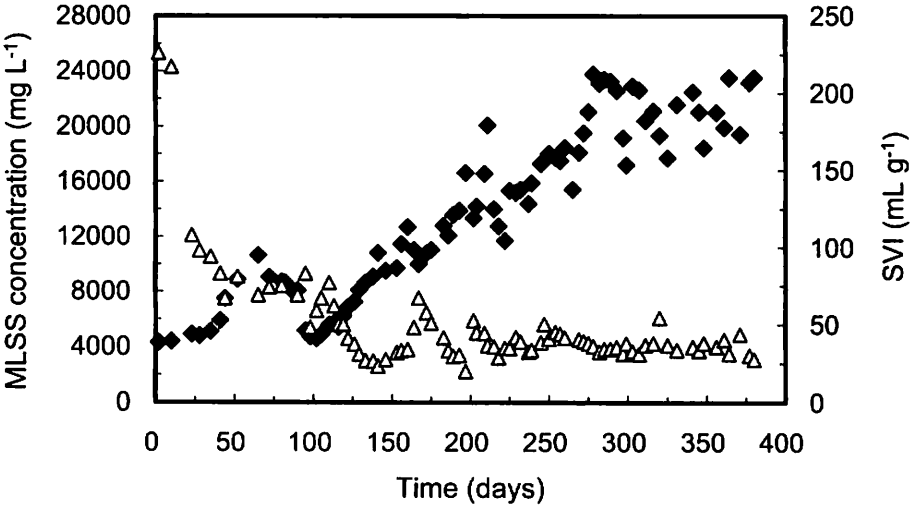


Fig. 3-4 Profiles of MLSS and SVI.

(◆) MLSS; (△) SVI.

MLSS concentrations and SVI values in the AS1 reactor cell were targeted and measured to elucidate the function of BF carrier. The results are shown in Fig. 3-4. MLSS concentrations increased with increase in s-COD VLR soon reaching a concentration of over 10,000 mg L^{-1}

on around day 70. On day 95, sludge was washed out due to clogging of a connection tube. However, together with increase in VLR, the MLSS concentration increased again after the clogging was cleared. The MLSS concentration reached more than 20,000 mg L^{-1} on day 275 at a VLR of 3.5 kg s-COD m^{-3} d^{-1} and the sludge had an SVI of less than 50 mL g^{-1}. The MLSS concentration remained at an average value of 20,000 mg L^{-1} from day 275 to day 382. Similar characteristics of sludge were also observed in AS2 and AS3 cells indicating that the sludge properties transferred from the BF cell to AS3 cell. Based on these results, it was concluded that the BF carrier was beneficial for the improvement of sludge characteristics. From the measurement of VSS content of about 93%, a large part of sludge was shown to consist of organic matter.

Fig. 3-5 illustrates the relationship between s-COD VLR, MLSS and SVI in the BF reactor cell. The MLSS concentration increased to more than about 12,000 mg L^{-1} at a VLR of 4.0 kg s-COD m^{-3} d^{-1}. Furthermore, the MLSS concentration exceeded 20,000 mg L^{-1} of MLSS at a VLR of 7.0 kg s-COD m^{-3} d^{-1}. On the other hand, the SVI values remained below 50 mL g^{-1} at a VLR of over 4.0 kg s-COD m^{-3} d^{-1}. Under this trend of high MLSS concentrations and low SVI levels was consistent up to a VLR of more than 4.0 kg s-COD m^{-3} d^{-1}. These results are consistent with the results of Cheng et al. (2005). Therefore, it was concluded that application of high VLR to BF reactor was an important parameter for getting high reactor MLSS concentrations with low SVI values. It was also suggested that partially packed BF carriers in the treatment process could improve the characteristic of the sludge in conventional activated sludge system.

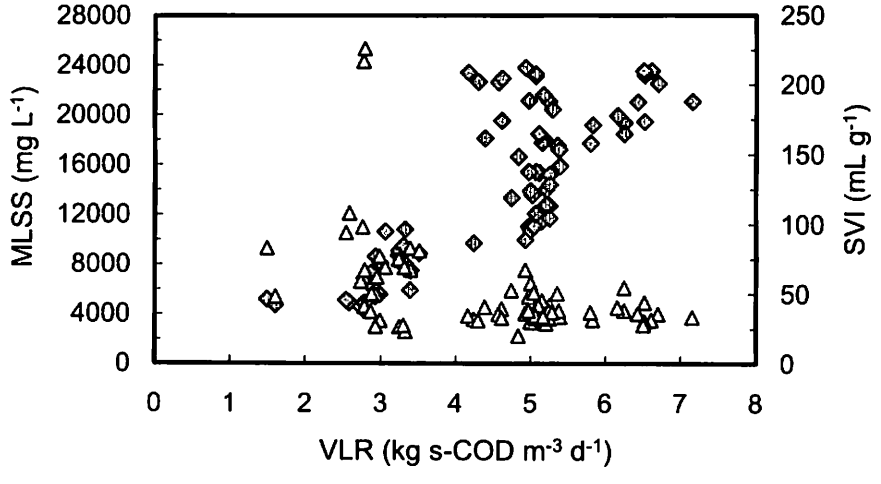


Fig. 3-5 Relationship between s-COD VLR, MLSS and SVI in the BF reactor cell.

(◇) MLSS; (△) SVI.

Table 3-1 Summarization of the SRT values and the F/M ratios under various VLRs

S-COD loading rate (kg s-COD m^{-3} d^{-1})	Duration (days)	SRT (days)	F/M (kg BOD kg^{-1} MLSS d^{-1})
3	63	66	0.14
3.5	63	59	0.11
4.5	65	41	0.14

Generally, SRTs are controlled between 5 to 10 days in CAS process to prevent the problems associated with solid-liquid separation (Hashimoto et al., 1981). Nevertheless, with long SRT ranging from 41 to 66 days in this research (Table 3-1), stable operation was still possible without problems in the solid-liquid separation. F/M ratios are normally maintained between 0.2 to 0.4 kg s-BOD kg^{-1} MLSS d^{-1} to form agglomeration for providing good settleability of activated sludge floc and to maintain good s-BOD removal (Shimizu et al., 2001). In our experiments, good treatment performance was observed at relatively lower F/M ratios of 0.11 to 0.14 kg s-BOD kg^{-1} MLSS d^{-1} (Table 3-1). The actual F/M ratios were considered to be lower than these calculated values if the amount of attached sludge on BF carrier was taken into consideration.

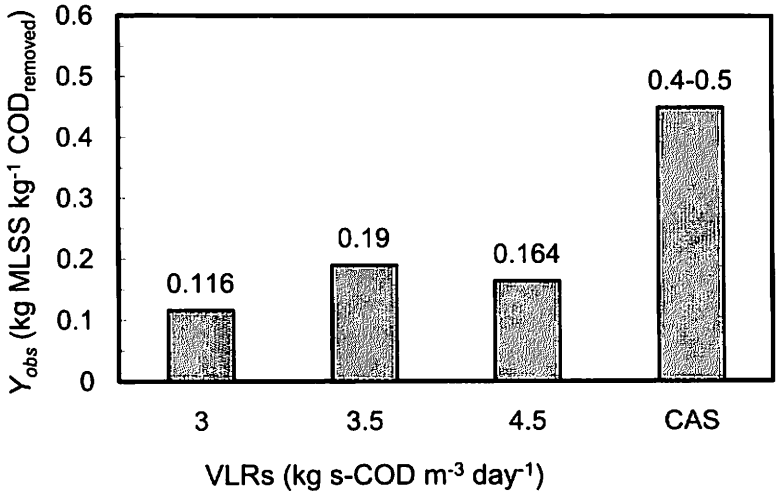


Fig. 3-6 Sludge yields for different VLRs.

Fig. 3-6 shows observed sludge yields (Y_{obs}) for VLRs of 3, 3.5 and 4.5 kg s-COD kg^{-1} MLSS d^{-1}. The calculated Y_{obs} values ranged from 0.116 to 0.190 kg MLSS kg^{-1} $s\text{-}BOD_{removed}$. These values were low compared with reported Y_{obs} values for CAS processes (Metcalf &

Eddy, 1991; Ghyoot et al., 1999). The reasons of these low Y_{obs} values are attributed to long SRTs.

Sludge settling capabilities were also examined through settling tests using diluted activated sludge taken from the AS1 reactor cell and the seed sludge tank in our laboratory. Sludge A was taken on day 305 from the AS1 reactor cell and diluted to 6,700 mg L^{-1}. Sludge B and C were taken from the seed sludge tank and diluted to 3,040 mg L^{-1} and 6,170 mg L^{-1}, respectively to imitate MLSS concentration in common activated sludge (sludge B) and activated sludge having same MLSS concentration as that of sludge A (sludge C). The results of the sludge settling tests are shown in Fig. 3-7. It is evident that BF carriers improved the sludge settling characteristics due to improved compressibility and settleability.

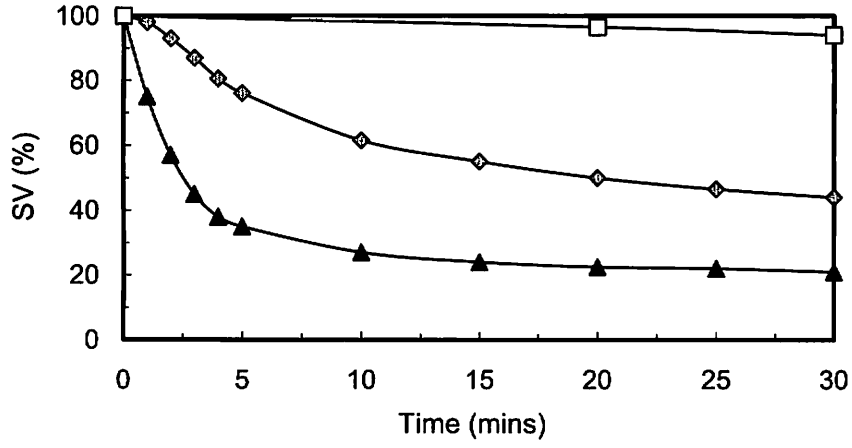


Fig. 3-7 Comparison of sludge settling curves for sludge in this study with activated sludge. (▲) sludge A; (◆) sludge B; (□) sludge C.

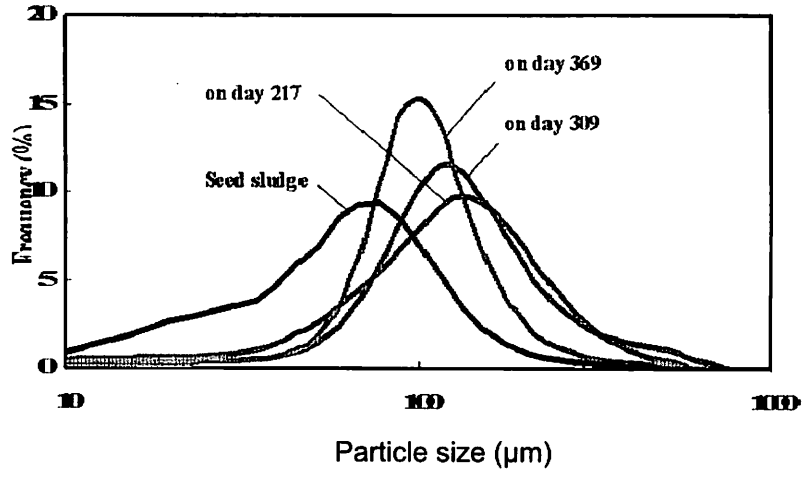


Fig. 3-8 Changes in particle size distribution during the experimental period.

Fig. 3-8 shows the particle size distribution of the seed sludge and the sludge samples taken from the AS1 reactor cell on days 217, 309, and 369 at VLRs of 3.0, 3.5, and 4.5 kg s-COD m^{-3} d^{-1}, respectively. The particle size distribution varied with an increase in VLR. Although the average floc diameter decreased with the increase in VLR, the frequency increased significantly. It was interesting to note that the test sludge samples taken from the AS1 reactor cell had same SVI values, but had the tendency to form smaller size floc with the increase in VLR.

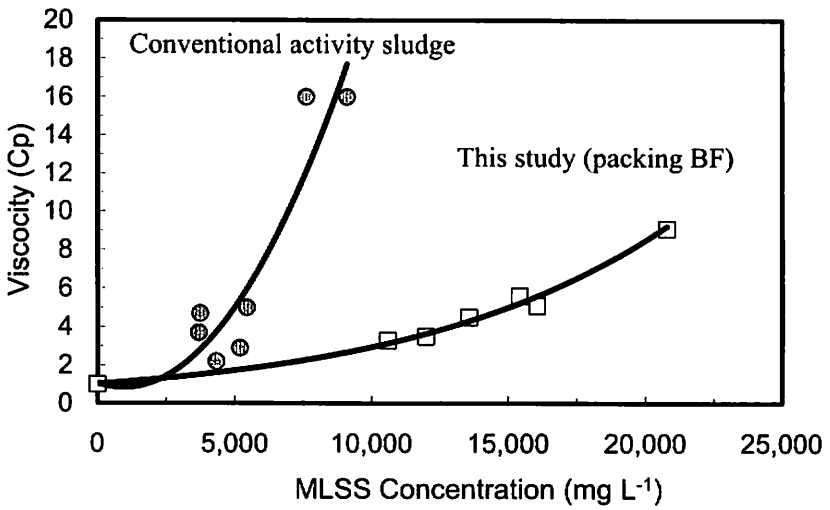
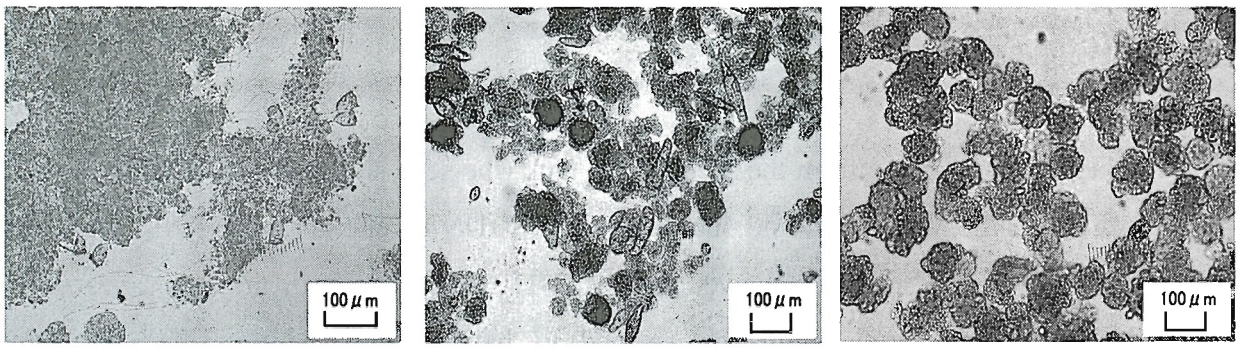


Fig. 3-9 Relationship between MLSS and viscosity.

Sludge viscosities were also measured using viscosity meter and defined distilled water as 1 Cp (MLSS = 0 mg L^{-1}). To compare with the sludge obtained in this study, activated sludge samples were taken from several wastewater treatment plants (WWTP) and their viscosities were measured. Further, Fig. 3-9 illustrates the relationship between MLSS concentration and viscosity for activated sludge taken from systems operating with and without the BF carrier. The exponential relationship observed between MLSS concentration and viscosity of sludge is consistent with the results reported by other researchers (Khongnakorn et al., 2007; Reid et al., 2008). Significantly lower viscosities at high MLSS concentrations were observed in this study. For example, the viscosity was 5 Cp at MLSS concentration of 5,000 mg L^{-1} in conventional activated sludge, while 5 Cp at MLSS concentration of 15,000 mg L^{-1} for activated sludge partially packing with the BF carrier (this study). This result shows that high MLSS concentration sludge with low viscosity was only realized using swim-bed technology. Generally, excessively high MLSS concentration, accumulation of untreated organic

compounds or presence of excess polymeric flocculant molecules contribute to increase in sludge viscosity (Nakagaki et al., 1991). Extremely high viscosity of mixed liquor will cause deterioration of sludge settleability and oxygen transfer efficiency. In this research, low sludge viscosities were observed even at high MLSS concentrations demonstrating potential advantage of the swim-bed technology.



a) on day 18 **b) on day 189** **c) on day 364**

Fig. 3-10 Microscopic photographs of activated sludge in AS1 reactor cell:
(a): on day 18, MLSS, 4,386 mg L^{-1}; SVI, 217 mL g^{-1}; (b): on day 189, MLSS,13,560 mg L^{-1}; SVI, 30 mL g^{-1}; (c): on day 364, MLSS, 23,510 mg L^{-1}; SVI, 31 mL g^{-1}.

Fig. 3-10 illustrates the results of microscopic observation of the reactor sludge on days 18, 189 and 364. At the beginning of the operation, the sludge was observed to have low density and composed of filamentous microorganisms (Fig. 3-10a). With the increase in VLR, granular-like flocs were confirmed (Fig. 3-10b). This granular-like sludge exhibited good settleability even at high MLSS concentration and contributed to the stable treatment performance of the system. Fig. 10c shows a photograph of the sludge sample taken on day 364 when the MLSS concentration was 23,510 mg L^{-1}. The photograph indicates a dense sludge. Moreover, high population of protozoa and metazoa was generally observed in the sludge sample. The presence of protozoa, e.g., *Opercularia.sp*, *Aspidisca.sp* is well known as indicator microorganism observed under high efficiency and high VLR conditions. The presence of large number of metazoa, e.g., *Philodina.sp*, *Rotaria.sp* and *Nais.sp*, however, is generally observed at low VLRs and occurrence of high nitrification (Kojima et al., 1996). These microorganisms formed a long food chain, maintained stable ecosystem and lead to low Y_{obs} and high organic removal efficiency (Kojima et al., 1996; Lapinski et al., 2003; Wei et al., 2003). Especially, massive growth of *Philodina.sp* contributed to agglomeration of activated sludge and subsequently clear effluent (Kojima et al., 1996). Agglomeration of sludge,

however, may be impacted by shear stress and turbulence in the reactor due to aeration or reactor geometry. Liu et al. (2005), for example, reported that shear stress generated by mixing water influenced sludge particle diameter and SVI. In our research, it was suggested that the turbulent flow caused by aeration at the rate of 10 L min^{-1} and the BF carrier in the four-celled reactor led to the glanular-like floc formation.

3.3.3 Microbial analysis

Two sludge samples, one from the seed sludge tank (the seed sludge) and one from the BF reactor on day 378, were taken and subjected to 16S rDNA gene analysis by PCR-DGGE method. The DGGE results are showed in Fig. 3-11. Since bands in the two lanes gave different patterns, bacterial population seemed to have changed during the long-term operation. The cloned DNAs were used to infer the bacterial members in the consortia, as shown in Table 3-2. A large number of bacteria belonging to phylum *Proteobacteria* was commonly observed in both samples. Changes in the day 378 sludge sample were observed as a wide variety of classes in phylum *Proteobacteria* were detected along with a variety of other phyla.

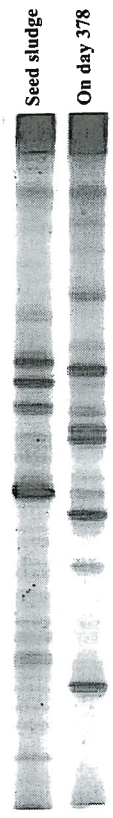


Fig. 3-11 DGGE results of different days from the BF reactor.

Table 3-2 Summary of analysis of the cloned bacterial 16S rDNA genes

Taxon(Phylum-class)	Seed Sludge		378th Sludge	
	No. of clone	percentage of total clones(%)	No. of clone	percentage of total clones(%)
Proteobacteria	13	65.0	22	57.9
α-proteobacteria	-	-	(5)	(22.7)
β-proteobacteria	(6)	(46.2)	(7)	(31.8)
γ-proteobacteria	(7)	(53.8)	(10)	(45.5)
Actinobacteria	-	-	2	5.3
Bacteroidetes	3	15.0	7	18.4
Gemmatimonadetes	-	-	2	5.3
Unclassified	4	20.0	5	13.2
Total	20	100	38	100

In both the samples, *Beta Proteobacteria* contained a large numbers of *Thermomonas.sp* and *Xanthomonas.sp* belonging to *Xanthomonadaceae* (family), and *Comamonas.sp* and *Acidovorax.sp* belonging to *Comamonadaceae* (family). *Xanthomonadaceae* and *Comamonadaceae* can consume DO and decompose organic substances such as peptone (Vauterin et al., 1995; Bergey et al., 2005a; Bergey et al., 2005b). These organisms might have contributed to the high s-COD removal rate achieved in this study. *Nitrosomonas oligotropha*, known as ammonia-oxidizing bacteria (AOB), was identified as a member of *Beta Proteobacteria* in the day 378 sample. This species is reported to be detected under low NH_4-N concentration conditions (Bollmann et al., 2002). Although high nitrification efficiencies were obtained in the reactor, none of the nitrite-oxidizing bacteria (NOB) such as *Nitrospira* or *Nitrobacter* were identified in *Beta Proteobacteria* members. It is reported that the AOB numbers are about ten times higher than that of NOB even that both of them share the same reaction rate (Fujita et al., 2007; Fujita, 2008). Therefore, non detection of NOB might be due to low population numbers as Ha et al. reported in their sample (Ha et al., 2005).

3.4 Conclusions

This study investigated the potential of enhancing treatment performance of conventional activated sludge process by installing BF carrier, known to create characteristics of swim-bed technology. Stable and high organic removal efficiencies were observed at VLRs of up to of 4.5 kg s-COD m^{-3} d^{-1} as the average s-COD and s-BOD_5 removal efficiencies were 96.5% and 99.1%, respectively. The reactor also exhibited high ammonium removal efficiency as an average removal efficiency of 83.6% was obtained. Partially packing of the BF carrier in the

reactor enhanced biomass retention and increased the particle size of flocs as compared to the seed sludge. The low SVI values below 50 mL g^{-1} and high settling velocities demonstrated satisfactory settling characteristics of the biomass, which could be attributed to the increase in the size of the biomass flocs. The relatively low viscosity of mixed liquor also facilitated the sludge settling performance as no sludge bulking problems occurred throughout the entire experimental period. Microscopic observation revealed abundance of protozoa and metazoa, which contributed to low sludge yields as compared with the conventional activated sludge. DNA analysis verified that *Proteobacteria sp.* was predominant in the sludge samples taken on day 378. Although high nitrification efficiencies were obtained, no NOB were detected in the sludge sample due to low population count of NOB. The results of this study demonstrate that use of swim-bed technology has the potential to enhance treatment performance and provide process stability to the conventional activated sludge process.

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Chapter 4 Nitrogen removal performance by combining Modified Ludzak-Ettinger (MLE) process with acrylic fiber packing in treating synthetic domestic wastewater with low C/N ratio

4.1 Introduction

The need for nitrification in wastewater treatment arises from the following reasons: (1) the effect of ammonia on receiving water with respect to DO concentrations and fish toxicity, (2) the need for nitrogen removal to control eutrophication, and (3) the need to supply nitrogen control for water reuse applications including groundwater recharge.

Biological denitrification is an integral part of biological nitrogen removal, which involves both nitrification and denitrification. Compared with other alternatives, such as ammonia stripping, breakpoint chlorination and ion exchange, biological nitrogen removal is generally more cost-effective and used more frequently.

The Modified Ludzak-Ettinger (MLE) process is one of the conventional nitrogen removal processes in municipal wastewater treatment (Gayle et al., 1989). The process consists of an anoxic tank followed by the aeration tank where nitrification occurs. Nitrate produced in the aeration tank is recycled back to the anoxic tank. Because the influent organic substrate serves as the electron donor for oxidation-reduction reactions using nitrate, the process is termed substrate denitrification. Since the anoxic process precedes the aeration tank, the process is also known as preanoxic denitrification.

For upgrading activated sludge processes, the IFAS (Sen et al., 1994) has been developed recently. The advantage for IFAS process over CAS process is the improvement of treatment capacity provided by the high biomass concentration in aeration tank, consequently enhancing volumetric nitrification rates, reducing sludge production and improving sludge (Metcalf & Eddy, 2003). The swim-bed technology using BF carriers (Rouse et al., 2004) is very effective for wastewater treatment, especially for application as fixed packing aerobic IFAS process. Thus, packing BF carriers seems to enhance the treating ability of MLE process.

In order to enhance the nitrogen removal efficiencies of the MLE process, the BF biomass carriers was applied for its powerful sludge attachment and retention capacities. In this chapter, the BF biomass carriers were packed into both aeration and anoxic tanks of the MLE process, which was then shifted to an IFAS-MLE process. This novel process combined MLE process with swimming-bed reactor was established to evaluate the nitrogen removal efficiencies for the treatment of synthetic wastewater with low C/N ratio.

4.2 Materials and methods

4.2.1 Synthetic wastewater

Table 4-1 Basic characteristics of the influent

Item	Concentration (mg L^{-1})
COD	300
BOD	200
TN	65

The synthetic wastewater used as influent was prepared by the dilution of concentrated bonito extract (40 g L^{-1}) and peptone (60 g L^{-1}) mixture liquor with tap water with ammonia chloride additions. The concentration of synthetic wastewater was shown in Table 4-1. BOD concentration was about 67 % of the COD concentration and the ratio among the BOD, nitrogen and phosphorus for the influent was 100: 32.5: 1.5.

4.2.2 Experimental setup

This system consisted of a denitrification (DN) reactor, a nitrification reactor (NT) and a sludge settling tank, which were all constructed of acryl resin, with effective volumes of 8, 10, and 5 L, respectively (shown in Fig. 4-1).

One mechanical mixer (MAZELA 2, EYELA, Japan) was installed in the DN reactor to provide the complete contact between substrates and bacteria at a stirring speed of 250 rmp.

Support filament of the BF carrier was 350 mm in length. The specific surface area of BF carrier approximated 0.2 m^2 m^{-1}. Fringe yarns of the BF carrier were attached symmetrically, extending equal distances beyond each side of the support filament and twisting to give an even 3-dimensional distribution of the attachment matrix (about 30 circles per meter). The DN reactor was operated at constant temperature of 22 ± 2 °C.

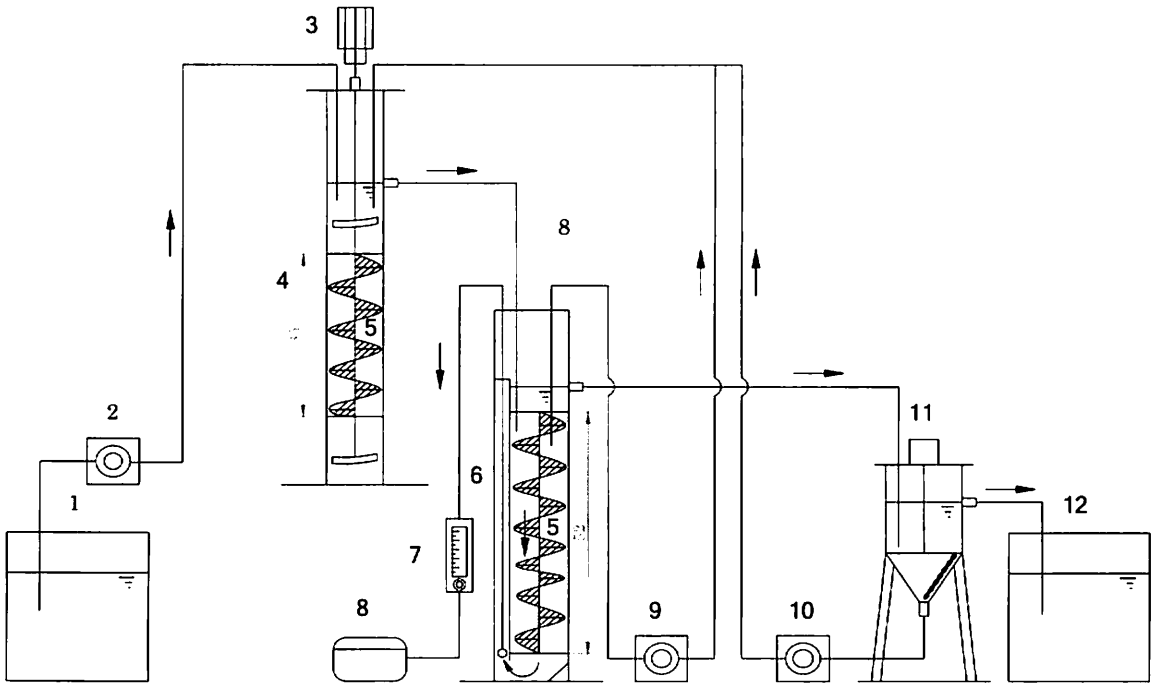


Fig. 4-1 Schematic of the experimental setup.

(1) influent tank; (2) influent pump; (3) stirrer; (4) denitrification (DN) reactor; (5) BF biomass carrier; (6) nitrification (NT) reactor; (7) air flowmeter; (8) air pump; (9) mixing liquid return pump; (10) sludge return pump; (11) sludge settling tank; (12) effluent tank.

The NT reactor had downdraft and updraft sections of 115×115 and 115×30 mm respectively, separated by a vertical baffle. Support filament of the BF carrier was 500 mm in length. The specific surface area of BF carrier approximated 0.2 m^2 m^{-1}. Fringe yarns of the BF carrier were attached and twisted as the same mode in the DN reactor. Air was introduced near the bottom of updraft section, with a fixed flow rate of 10 L min^{-1} to aerate the wastewater and induce a down-flow hydraulic velocity of 20 cm s^{-1} in downdraft section by providing density difference of the liquor, which led to a strong "swimming motion" for the carrier filaments. The NT reactor was also operated at constant temperature of 22 ± 2°C.

Surface area of the sludge settling tank was 0.03 m^2. Sludge in settling tank was gently

mixed by a chain and returned to the DN reactor at 100% recycle rate, meanwhile the mixing liquor in the NT reactor was also recycled to the DN reactor at 100% recycle rate to provide nitrate as substrate for denitrification.

4.2.3 System start-up and sludge acclimation

For startup of this experiment, 90 g (dry weight) of activated sludge was inoculated in the reactor with tap water for an initial MLSS concentration of 5,000 mg L^{-1} for both reactors, and airflow was set at 5 L min^{-1} to circulate the solution through the reaction zone at a velocity of 10 cm s^{-1} during biofilm attachment period. The attachment of 62 g of sludge during a 36 h period of biofilm attachment amounted to 73.8 g m^{-1} of BF support filament for the reactor.

Following the sludge attachment periods, the continuous operation was carried out using the synthetic influent. After the first week, the reactors were considered to be acclimated and airflow rates were increased to 10 L min^{-1}.

4.2.4 Experimental procedure and operational conditions

Table 4-2 Operational conditions during the whole experiment period

Period	Time (day)	VLR (kg BOD m^{-3} d^{-1})	Flow rate (L d^{-1})	HRT (h)
Run 1	1-18	0.5	46	9.5
Run 2	19-33	1.0	93	4.8
Run 3	34-88	1.5	140	3.2
Run 4	89-110	1.75	160	2.8

During 110 days of operation, the experiment was divided into four runs based on various VLRs, which were increased by changing HRT only. Table 4-2 shows the operational conditions during the whole experimental period.

4.2.5 Analytical methods

Except SS, SV_{30}, SVI and TN, analytical methods are the same as those described in Chapter 2. SS, SV_{30}, SVI and TN concentrations were analyzed according to standard methods (APHA, 1995.).

4.2.6 Calculation methods

In this chapter, observed sludge yields (Y_{obs}) were calculated according to Eq. (3-4).

The relationship between TN removal rate and recycle rate is shown in Eq.4-1 (Payne et al., 1981).

TN removal efficiency (%) = r/(r+1) x100 (4-1)

where r is recycle rate allowing for both mixing liquid and sludge recycles.

4.3 Results and discussion

4.3.1 COD and BOD removal performances

The effluent COD concentrations fluctuated at the beginning of the experiment. When the reactor was operating stably, COD removal became stable with removal efficiencies above 80% and effluent COD concentration less than 60 mg L^{-1}. Under low VLRs, COD removal efficiencies decreased slightly just after increasing the VLRs. Accompanying microbial sludge acclimation, the removal efficiencies were recovered in a short time. However, different from the lower removal efficiencies under low VLRs, the removal efficiencies were unchanged when the VLR was increased from 1.5 to 1.75 kg BOD m^{-3} d^{-1}. This demonstrated that the process had a certain capability of anti-shock loads under stable operational conditions (Fig. 4-2a). With the increase in VLRs, soluble COD (s-COD) removal efficiencies gradually decreased. However, COD removal efficiencies increased due to decrease in effluent SS concentrations by mature bio-system.

Comparing with COD removal, BOD removal was more stable. After the VLR was increased to 1.5 kg BOD m^{-3} d^{-1}, the effluent BOD concentrations were below 20 mg L^{-1} with

BOD and s-BOD removal efficiencies were around 90% and 95%, respectively (Fig. 4-3a). The BOD concentrations in DN reactor were very low, almost the same as effluent BOD concentrations. This indicated that influent organic matters were used for denitrification as carbon source in the DN reactor. Similar to the COD removal tendency, the BOD removal efficiencies decreased slightly when increasing loads under low VLRs. However, above VLR of 1.5 kg BOD m^{-3} d^{-1}, the process was stable under shock loads and maintained the stable operation after the MLSS concentrations reached a certain level.

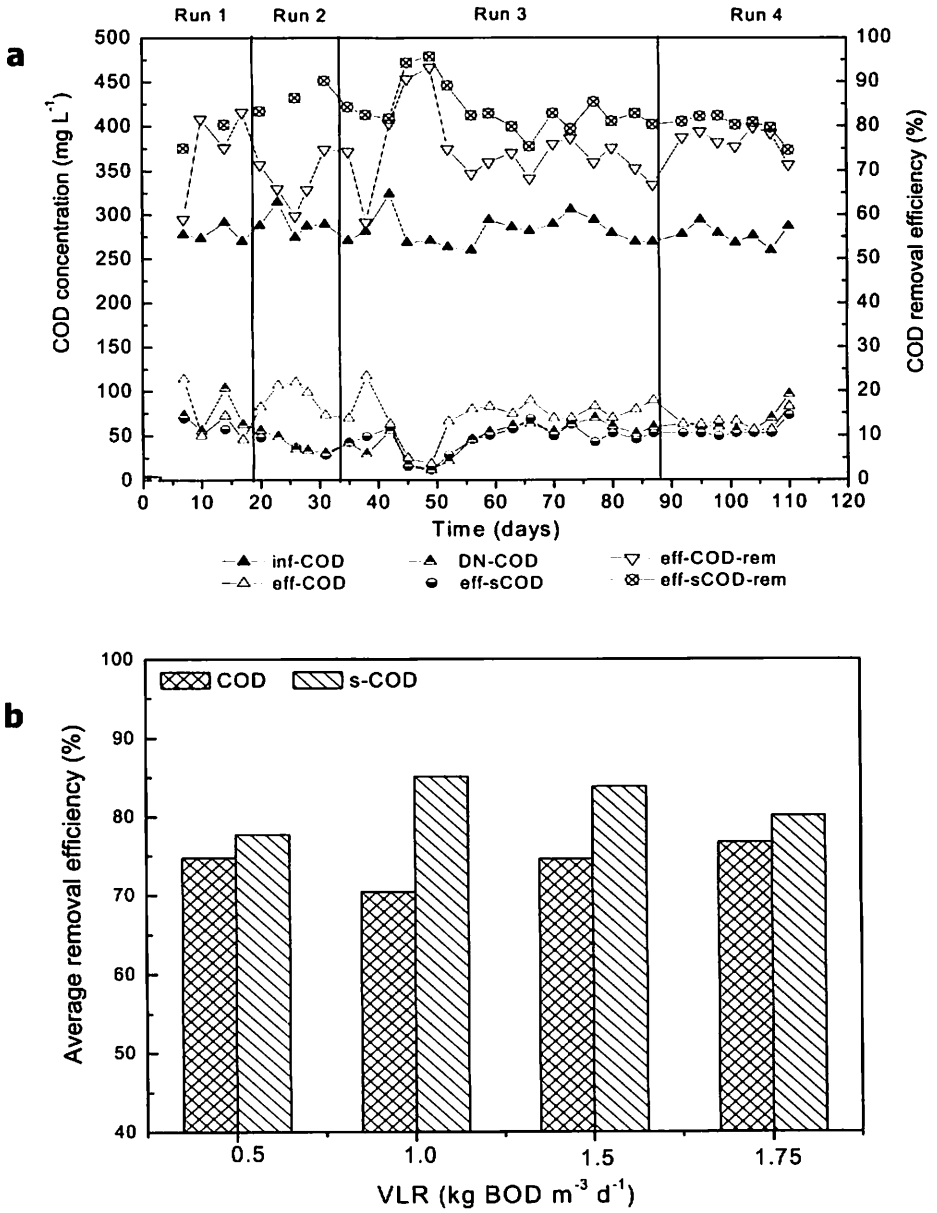


Fig. 4-2 COD and s-COD removal performances.

(a) Changes in COD concentrations and COD removal efficiencies; (b) average COD and s-COD removal efficiencies.

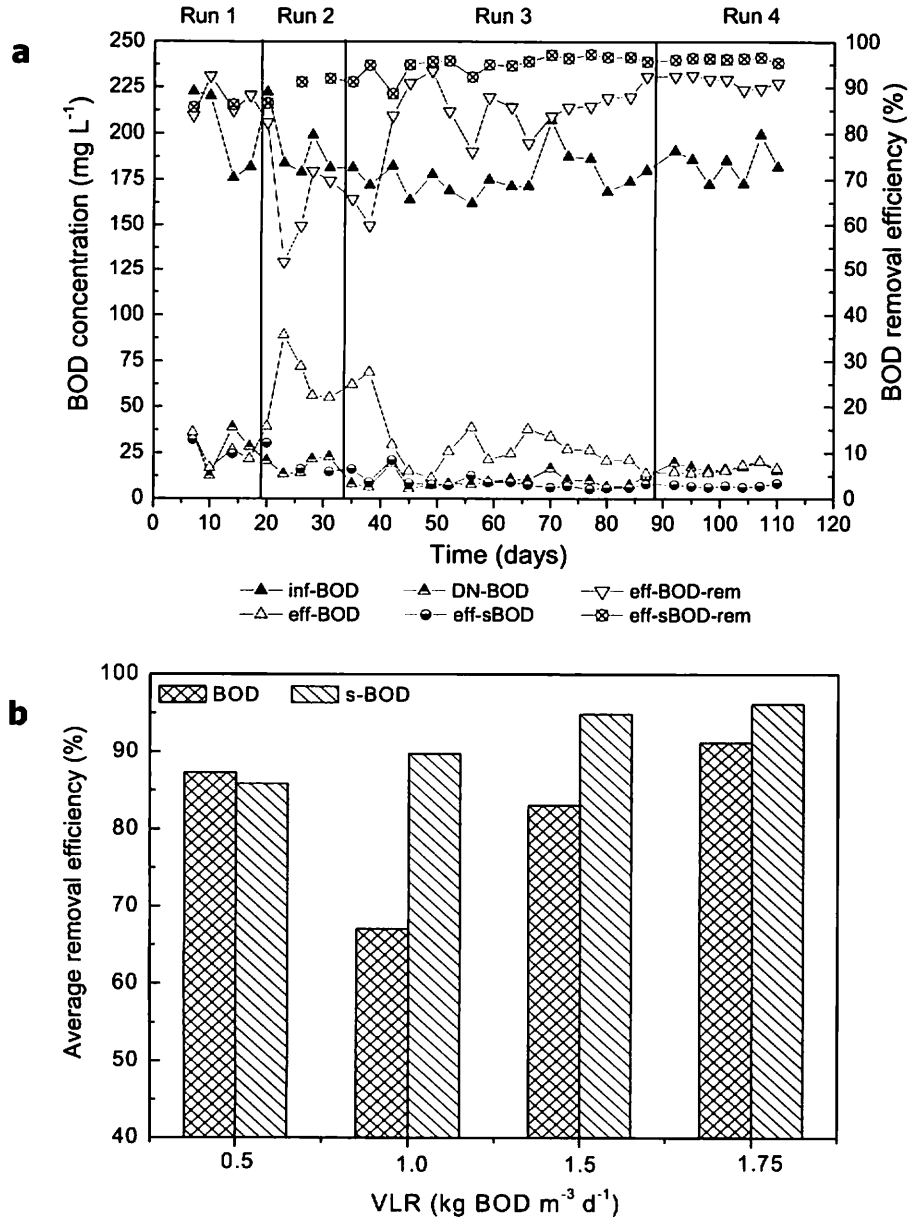


Fig. 4-3 BOD and s-BOD removal performances.

(a) Changes in BOD concentrations and BOD removal efficiencies; (b) Average BOD and s-BOD removal efficiencies.

For the traditional nitrogen removal process, there is a demand for COD as organic carbon source and C/N ration should be at least more than 5 for completely denitrification (Modin et al., 2007; Ma et al., 2009). In this experiment, influent COD and TN concentrations were 300mg L^{-1} and 65mg L^{-1} on average and the C/N ratio was about 4.6. This means that organic carbon source was not enough for complete denitrification in this study. So that, COD should be consumed entirely due to the shortage of organic carbon source. Therefore, high COD and

BOD removal efficiencies were obtained in all runs. BOD concentrations in the DN reactor were as low as that in the NT reactor. This illustrated that organic matter was consumed by denitrification as carbon source once entering the DN reactor.

4.3.2 Nitrogen removal performance

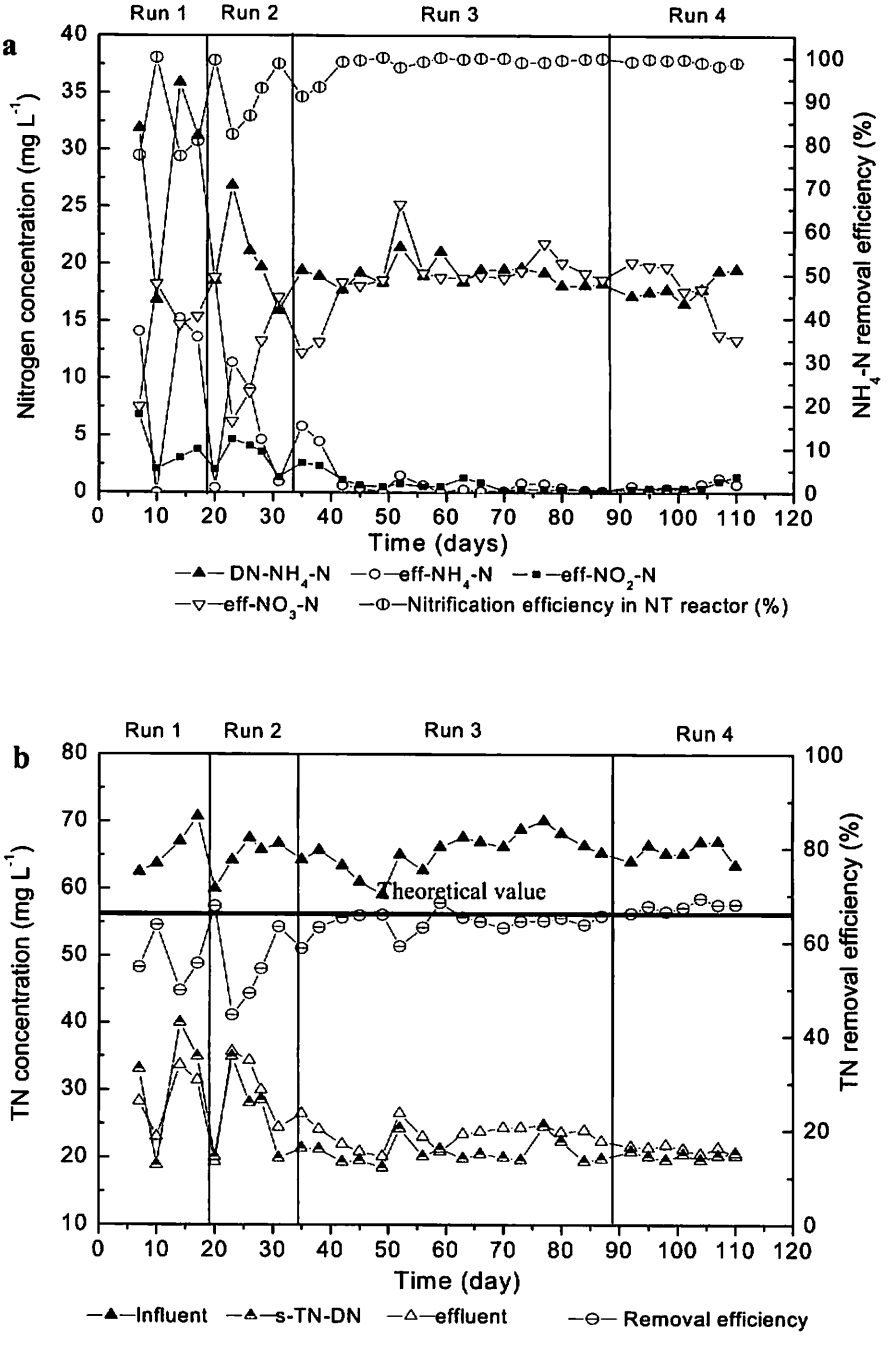


Fig. 4-4 Nitrogen removal performance.

(a) changes in nitrogen transformations; (b) TN removal efficiencies.

The time course of nitrogen transformations is presented in Fig. 4-4a. The influent organic nitrogen could be easily degraded to ammonia nitrogen via ammonification in the DN reactor. In steady states, effluent NH_4-N and NO_2-N concentrations were almost zero, leaving only NO_3-N in the effluent. Nitrification efficiencies in the NT reactor were maintained above 95% by taking the NH_4-N of DN reactor as influent NH_4-N in NT reactor. Meanwhile, denitrification efficiencies in DN reactor also kept above 95% by taking the NO_3-N from recycled mixing liquid and recycled sludge as the influent NO_3-N in the DN reactor. The results illustrate that both nitrification capacity of the NT reactor and denitrification capacity of the DN reactor could be maintained at a very high level in this system.

The theoretical value of TN removal efficiency was 66.7% when r was 2. according to Eq. 4-2. As shown in Fig. 4-4b, the experimental TN removal efficiencies approached the theoretical value of 66.7% (presented by the horizontal line), which demonstrated that the obatined values in this study approximated to the theoretical value. Under VLRs of 1.5 and 1.75 kg BOD m^{-3} d^{-1}, average TN removal efficiencies were 64.2% and 70.4%, respectively, and effluent TN concentrations were below 20 mg L^{-1} which were lower than the theory value. Besides nitrogen assimilation of microorganism growth, simultaneous nitrification and denitrification were also attributed to the excess TN removal. With the growth of biomass, the thick biofilm could provide an anoxic micro-environment for denitrifying bacteria metabolism in aerobic reactor. Furthermore, denitrification could occur under aerobic conditions by heterotrophic nitrifying bacteria, thus simultaneous nitrification and denitrification occurred through the conversion of ammonia to gaseous nitrogen products (Robertson and Kuenen, 1990; Patureau et al., 1994). However, SND performance in this experiment was limited by excessive DO level and lack of organic carbon source (Zhu et al., 2007). Therefore, the primary contribution of nitrogen removal was due to preanoxic denitrification pathway.

High amount of nitrifying bacteria were enriched on the BF carrier owing to the high attachment and retention capacity of BF carrier. This enabled high TN removal efficiencies with low influent C/N ratio (Wen et al., 2003) and even under high VLRs. Microbial consortia for both reactors were maintained at a good growth condition for nitrifiers and denitrifiers. Thus, the TN removal efficiencies were stable even under high VLR of 1.75 kg BOD m^{-3} d^{-1}.

4.3.3 Sludge characteristics in the NT reactor

The changes in MLSS concentrations and SVI values are shown in Fig. 4-5. MLSS concentrations in both NT and DN reactors were almost the same due to the well mixing and recycling conditions of this system. When VLR was increased to 1.5 kg BOD m^{-3} d^{-1}, MLSS concentrations increased quickly to 5,000 mg L^{-1}, then raised up to 6,000mg L^{-1} gradually, finally maintained about 8,000 mg L^{-1} under VLR of 1.75 kg BOD m^{-3} d^{-1}.

When VLR was set to 1.5 kg BOD m^{-3} d^{-1}, SVI values quickly increased at first and then gradually decreased to 100. Afterwards, the SVI values kept stable around 150 under the VLR of 1.75 kg BOD m^{-3} d^{-1}.

MLSS concentrations could not increase to a high value in the conventional activated sludge (CAS) process due to poor sludge settlability. Solid/liquid separation in settling tank under high MLSS concentration was not easy. Generally, the levels of MLSS and SVI for CAS processes are reported to be 2,000 to 3,000 mg L^{-1} and below 150 ml g^{-1}, respectively (Giokas, 2003; Sezgin, 1982).

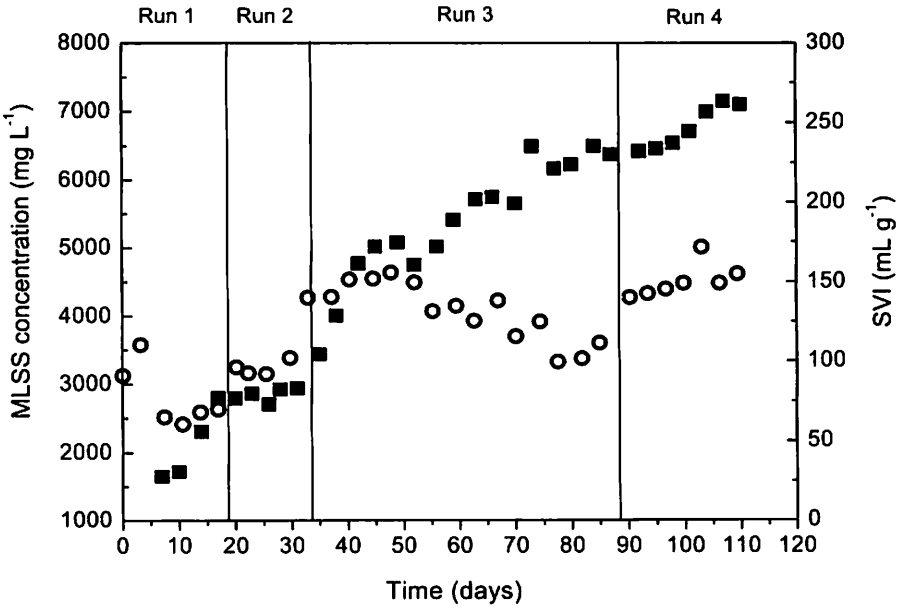
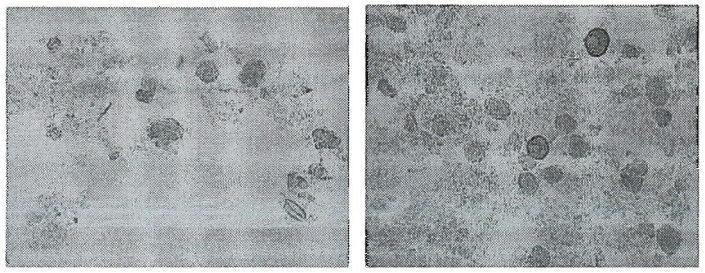
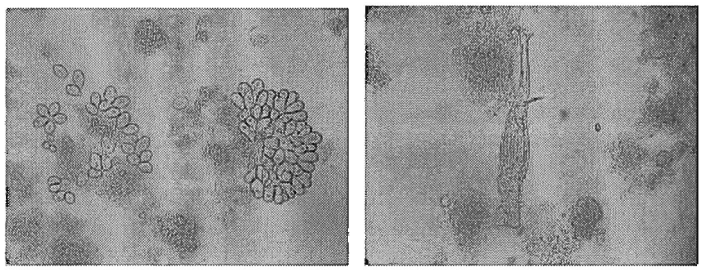


Fig. 4-5 Profiles of MLSS levels and SVI values.

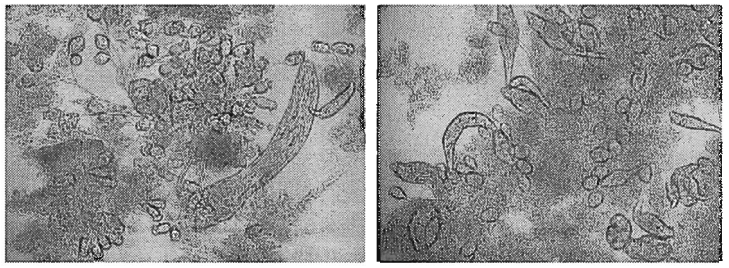
(■) MLSS; (O) SVI.



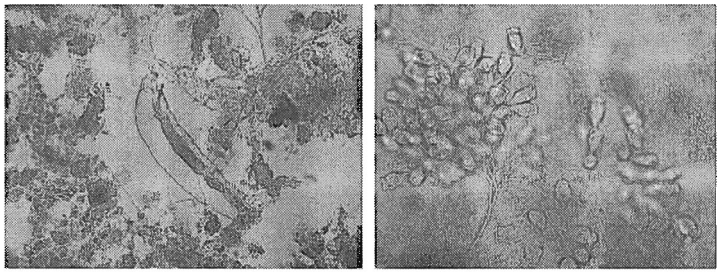
(a) 0.5 kg BOD m^{-3} d^{1}



(b) 1.0 kg BOD m^{-3} d^{-1}



(c) 1.5 kg BOD m^{-3} d^{-1}



(d) 1.75 kg BOD m^{-3} d^{-1}

Fig. 4-6 Microscopic photographs of microorganisms

This system combining MLE process with swimming-bed technology could maintain MLSS concentrations around 8,000 mg L^{-1}, which was about 3 times higher than that in CAS processes. The IFAS process has been widely accepted to claim for the advantage of enhanced sludge settleability and increased MLSS concentrations (Metcalf & Eddy, 2003). Gebara et al. (1999) reported that SVI was improved from 350 to 38 successfully by fitting plastic nets in a CAS reactor, and the BOD removal efficiencies were consequently increased from 72.5% to

97.5%. In another aspect, the so-called swimming motion by using BF carrier could assist the enhancement in sludge settleability which resulted in low value of SVI (Yamamoto et al., 2006). Despite of the low influent BOD concentrations and very short HRT, the highest SVI values could be maintained around 150, which was acceptable and resulted in a very high reactor MLSS concentrations in this study. High reactor MLSS concentration and high pollutant removal rates could be successfully confirmed in this experiment.

The key point of this system was the control of SVI under low influent substrate concentrations. It is hardly to recover the activated sludge system from sludge bulking (Mara and Horan, 2003; Thompson et al., 2003). Factors affecting SVI values are complicated and interactional. F/M ratio and SRT may play important roles in this study. The ways to control and harmonize each operational parameter should be investigated experimentally for maintaining a preferable SVI.

During the continuous operation, activated sludge appeared to aggregate gradually (Fig. 4-6). With the increase in VLRs, the protozoa and metazoa became abundant with high densities. Firstly many *Epistylis sp.* appeared (Fig. 4-6b), then *Rotatoria sp.* became predominant (Fig. 4-6c), finally *Microbiotus sp.* appeared under VLR of 1.75 kg BOD m^{-3} d^{-1} (Fig. 4-6d).

4.3.4 Sludge yield

Under VLR of 1.5 kg BOD m^{-3} d^{-1}, effluent SS concentrations were under 30 mg L^{-1} with an average of 15.2 mg L^{-1}. Then, effluent SS concentrations were under 10 mg L^{-1} with an average of 6.94 mg L^{-1} under VLR of 1.75 kg BOD m^{-3} d^{-1} (Fig. 4-7).

The sludge yields calculated under different VLRs are shown in Fig. 4-8. The total sludge yield during the whole experiment was 0.13 kg MLSS kg $COD_{removed}^{-1}$, and the lowest value was only 0.05 kg MLSS kg^{-1} $COD_{removed}$ under VLR of 1.75 kg BOD m^{-3} d^{-1}. This values were markedly lower than that for CAS process (0.3 to 0.5 kg MLSS kg^{-1} $COD_{removed}$), even lower than that in some membrane bioreactors (Xing et al., 2003), and will be of great interest for WWTPs in practical applications.

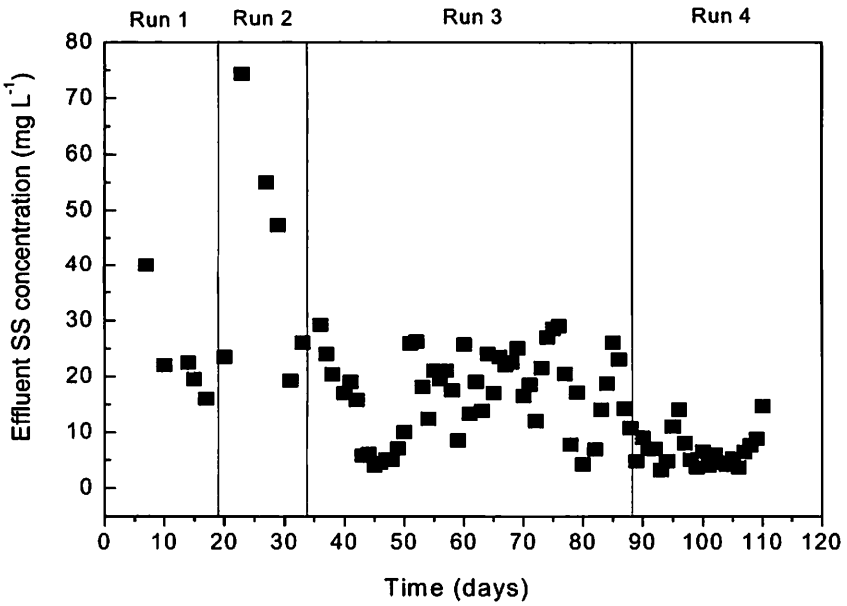


Fig. 4-7 Profile of effluent SS concentrations.

The primary reason of the low sludge yield is supposed to be the micro-fauna's predation on bacteria. During the shift from a low trophic level to a high level, energy is lost due to inefficient biomass conversion (Ratsak et al., 1996). It is well known that aerobic biofilm processes, depending on the biofilm thickness, generally have a more complex microbial ecology than that for activated sludge with films containing bacteria, fungi, protozoan, rotifers, and possibly annelid worms (WEF, 2000), thus it could obtain excess sludge reduction due to the longer food chain compared with that in activated sludge process.

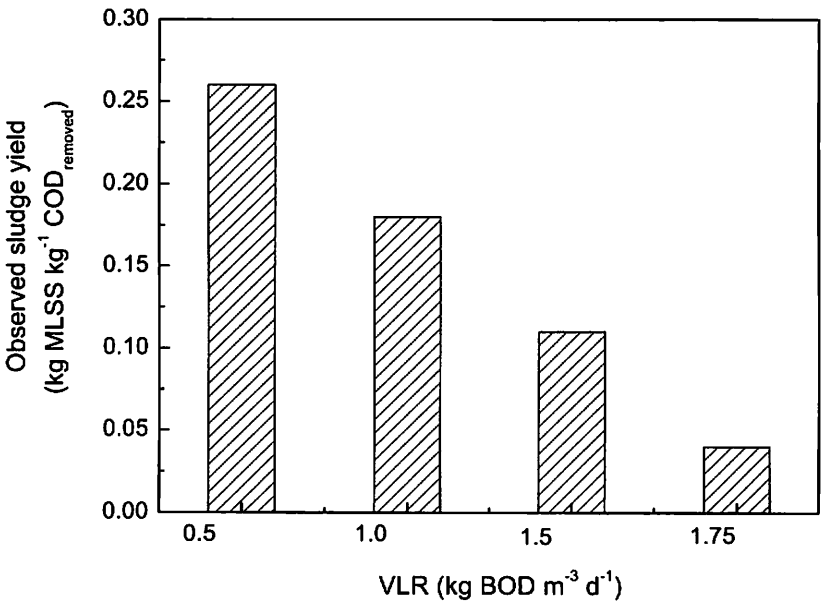


Fig. 4-8 Observed sludge yields under different VLRs.

With the increase in MLSS concentrations, the microbial community of activated sludge became very abundant and contributed to longer food chain bio-system (Cresson et al., 2008). The great emergencies and densities of protozoa and metazoan ensured the sludge yield reduction by predation on bacteria (Fig. 4-6). Therefore, sludge yield could be minimized in this system.

Furthermore, microscopic observation indicated that biodiversity of microbial community on the BF biomass carriers was much higher than that existed in the suspend sludge. The extensive biological communities resulted in the formation of long food chain, thus sludge yield could be minimized by predation.

4.4 Conclusions

In this chapter, a modified MLE process packing with the BF carriers both in anoxic and aerobic reactors was applied for COD and nitrogen removal for treating synthetic domestic wastewater with low C/N ratio. This study had focused on excess sludge reduction and nitrogen removal performance. Several advantages of using this system are listed below:

(1) Under VLRs from 0.5 to 1.75 kg COD m^{-3} d^{-1}, organic pollutants could be effectively removed with COD and BOD removal efficiencies of 80% and 90%, respectively. Moreover, BOD removal efficiencies reached up to 88.5% only within 14 days, which suggested that the quick biofilm attachment and quick start-up could be performed in this combined system. The final effluent was clear with an average effluent SS concentration of 10 mg L^{-1}.

(2) Excellent sludge attachment and retention capacity of the BF biomass carrier could effectively increase the amount of reactor sludge, thus removal efficiencies could be enhanced. In this study, both favorable nitrification in the NT reactor and denitrification in the DN reactor were carried out and the preferable TN removal was obtained under a very short HRT (only 2.8 h in Run 4). TN removal efficiencies of about 70% was obtained at the recycle ratio of 2.

(3) The MLSS concentration in both reactors could reach up to 8,000 mg L^{-1} and no filamentous activated sludge bulking occurred. This result suggested that the system

performed well even under extremely high reactor MLSS concentrations.

(4) The observed sludge yield during the whole experiment was 0.13 kg MLSS kg^{-1} $COD_{removed}$, and the lowest value was 0.05 kg MLSS kg^{-1} $COD_{removed}$ under VLR of 1.75 kg BOD m^{-3} d^{-1}. Those low observed sludge yields would be of great interest for practical applications of our proposed system.

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Chapter 5 Simultaneous nitrification and denitrification with excess sludge reduction in an attached growth system combining anaerobic fermentation and aerobic swim-bed processes

5.1 Introduction

The CAS process has a long history of being the most widely used biological wastewater treatment method. However, there are still some problems which attract much attention nowadays, such as large excess sludge reduction and low nitrogen removal efficiency.

During the past decade, many researchers have investigated the benefits of IFAS for individual enhancements of COD removal, partial nitritation, nitrification, denitrification and even enhanced biological phosphorus removal (EBPR) in CAS processes, by packing suspended or fixed biomass carriers into separate anoxic and aerobic reactors (Wang et al., 2000; Al-Sharekh et al., 2001; Sriwiriyarat et al., 2005; Chung et al., 2007). Using ringlace® media, Randall et al. (1996) documented that between 30% and 88% of the produced nitrates were denitrified under aerobic conditions. Similarly, Sen et al. (1994) observed 35% and 39% of denitrification occurred in aerobic tanks of single-sludge anaerobic-anoxic-aerobic processes packed with ringlace® and sponges, respectively. Recently, various biofilm processes, both in suspended and attached phases, have demonstrated the SND ability in a single aerobic reactor by having anoxic zones within the biofilm depth (Chen et al., 1998; Wang et al., 2006; Wen et al., 2008). Therefore, it is proposed that an aerobic IFAS process could accomplish steady total nitrogen removal via SND under suitable operational conditions.

In addition, anaerobic hydrolysis and fermentation (also referred to as acidogenesis) are commonly used as pretreatment for treating high-strength organic wastewater (Shin et al., 2005; Ma et al., 2009). Furthermore, anaerobic hydrolysis can be an effective method for improving denitrification efficiency by providing favorable electron donors such as sulfide and acetate (Rustrian et al, 1997; Garrido et al., 2001).

In this chapter, a combined system, which included an anaerobic fermentation (AF) reactor

and an aerobic IFAS swim-bed (SB) reactor, was constructed for treating moderately high-strength synthetic organic wastewater. The purpose of this study is to evaluate such a combined system in terms of COD removal, TN removal and excess sludge reduction. The possible mechanisms involved in SND were also discussed.

5.2 Materials and methods

5.2.1 Synthetic wastewater

The synthetic wastewater used as influent during the main experimental period of this study was prepared by diluting concentrated corn steep liquor (CSL, San-ei Sucrochemical, Japan) with tap water free of any buffer or nutrient additions. The characteristics of the influent were as follows: COD from 1000 to 2000 mg L^{-1}, TN from 65 to 130 mg N L^{-1}, and pH from 3.6 to 4.9 (Table 5-1). The C:N:P ratio of the influent was about 30:2:1. The CSL stock was produced by lactic acid fermentation and was thus acidic and high in lactate and volatile fatty acids (VFAs) (Table 5-2). During the acclimation period, another synthetic wastewater composed of bonito extract and peptone mixture was used.

5.2.2 Experimental setup

This system consisted of an AF reactor, a SB reactor and a sludge settling tank, which were all constructed of acrylic resin, with effective volumes of 6, 10, and 2.5 L, respectively (depicted in Fig. 5-1a). The AF and SB reactors had cross sectional sizes of 110×110 and 145 ×115 mm, and heights to effluent port of 500 and 635 mm, respectively.

Sixty pieces of the BL (biofill, NET, Japan) (Fig. 5-1b) carriers were packed into the middle section of the AF reactor with a packing ratio of 50%. A centrifugal pump with flow rate of 20 L min^{-1} was used to provide a down-flow internal circulation with a velocity of 2.5 cm s^{-1} to enhance the substrate-biomass contact in the AF reactor, and a tube distributor was set at the end of recycle pipe. The AF reactor was operated at constant temperature of 35 ± 1°C by a thermostat heater. A gas-liquid separating device was provided to collect the evolved gas for quantification and composition analysis.

The SB reactor had downdraft and updraft sections of 115×115 and 115×30 mm respectively, separated by a vertical baffle. Support filament of the BF carrier was 500 mm in length. The specific surface area of BF carrier approximates 0.2 m^2 m^{-1}. Fringe yarns of the BF carrier were attached symmetrically, extending equal distances beyond each side of the support filament and twisting to give an even 3-dimensional distribution of the attachment matrix (about 30 circles per meter) (Fig. 5-1c). Air was introduced near the bottom of the updraft section, with a fixed flow rate of 10 L min^{-1} to aerate the wastewater and induce the down-flow velocity of 20 cm s^{-1} in the downdraft section, which also induced a strong "swimming motion" for the carrier filaments. In addition, there was a slanted (45°) baffle in the bottom of the reactor to direct the flow evenly from the downdraft section to updraft section. The SB reactor was operated at constant temperature of 25 ± 1°C, except when higher room temperatures occurred in summer.

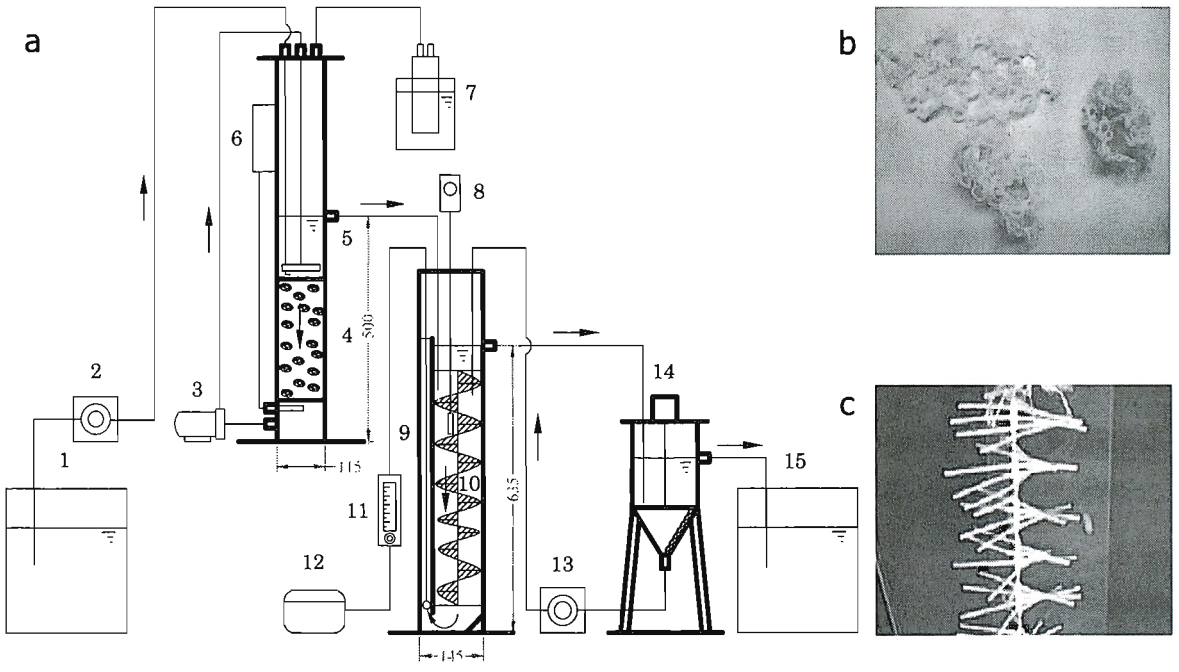


Fig. 5-1 (a) Schematic of the AF-SB system.

(1) influent tank; (2) influent pump; (3) AF recycle pump; (4) BL carriers; (5) AF reactor; (6) thermostat heating device in AF reactor; (7) gas-liquid separator; (8) thermostat heater in SB reactor; (9) SB reactor; (10) BF carrier; (11) air flow meter; (12) air pump; (13) sludge return pump; (14) settling tank; (15) effluent tank; (b) photograph of BL carriers; (c) photograph of BF carrier.

Surface area of the settling tank was 0.017 m^2. Sludge in settling tank was gently mixed by a chain and returned to the SB reactor at a 100% recycle rate.

5.2.3 System start-up and sludge acclimation

Both AF and SB reactors were initially inoculated with approximately 4,000 mg L^{-1} (as MLSS) of activated sludge from a fill-and-draw batch reactor. During the sludge attachment period without feeding influent, the recycle pump was started for internal recirculation of the AF reactor, and the airflow rate of SB reactor was maintained at 5 L min^{-1} to provide circulation velocity through the downdraft zone of 10 cm s^{-1} to enhance biofilm attachment under relative weaker velocity conditions. After 24 hours of biofilm attachment, the MLSS concentrations in the AF and SB reactors sharply, which demonstrated that both BL and BF carriers had excellent sludge attachment and retention capacities due to their hydrophilic characteristics and favorable configurations. Then, with the air flow rate in the SB reactor adjusted to 10 L min^{-1}, the system was operated continuously for about 1 month using synthetic wastewater containing fish meat extract and peptone mixture under low OLRs (0.5 to 1.5 kg COD m^{-3} d^{-1}) for acclimation of the sludge. During the sludge acclimation period, ammonification was observed in the AF reactor as well as some nitrification in the SB reactor (results not shown). The system was then shut down to collect all sludge for biomass calculation. Then the sludge obtained from the acclimation was inoculated again (anaerobic and aerobic sludges were used for the AF and SB reactors, respectively) according to the above mentioned biofilm attachment protocol at the start-up for the first stage (Run 1) during the main experimental period. Subsequently, the influent was shifted into CSL.

5.2.4 Experimental procedure and operational conditions

During 184 days of operation, the experiment was divided into four runs according to various OLRs, which were increased by changing hydraulic retention time (HRT) and influent COD concentration. The operational conditions during the whole experimental period are summarized in Table 5-1. Due to the objective of excess sludge reduction, no sludge was withdrawn intentionally from settling tank, though some organic solids were lost as effluent suspended solids (SS), sludge sampling events and occasional sludge washout. The initial OLR of each run after start-up was kept relatively low value for 3 to 5 days and then increased to the intended OLR. For accurate calculation of sludge yield, all sludge in the system was collected and sampled at the end of each run. Then, the sludge obtained from the previous run was inoculated again in the two reactors with a suitable volume as the seed

sludge for the new run. Interior surface growth in the feed lines and on the SB reactor walls was removed once a week.

Table 5-1 Operational conditions over the main experimental period

Parameters		Run 1	Run 2	Run 3	Run 4
Duration	(day)	0-22	23-56	57-115	116-184
OLR_{wh}	(kg COD m^{-3} d^{-1})	1.5	2.7	3.5	4.5
OLR_{SB}	(kg COD m^{-3} d^{-1})	2.4	4.0	5.1	6.1
NLR_{Wh}	(kg N m^{-3} d^{-1})	0.1	0.18	0.23	0.30
NLR_{SB}	(kg N m^{-3} d^{-1})	0.16	0.28	0.36	0.47
HRT_{wh}	(h)	16	10.67	10.67	10.67
HRT_{AF}	(h)	6	4	4	4
HRT_{SB}	(h)	10	6.67	6.67	6.67
HRT_{ST}	(h)	2.5	1.67	1.67	1.67
Temperature in SB	(°C)	25 ± 1	26 ± 2	30 ± 2	20-32
Influent COD	(mg L^{-1})	1000	1200	1555	2000
Influent TN	(mg L^{-1})	65	80	105	130
Influent pH		4.7 ± 0.2	4.3 ± 0.2	4.1 ± 0.2	3.8 ± 0.2

Subscripts: (wh) whole system; (SB) SB reactor; (AF) AF reactor; (ST) settling tank.

At the ends of Runs 1, 2 and 4, the MLSS concentration in the SB reactor was determined by sampling the suspended sludge. Then, attached biofilm on the BF carrier was detached completely in the reactor. The MLSS concentration of this mixed sludge was also determined after thorough mixing. The calculated weight difference between the two sludges was considered to be the estimated weight of attached biofilm. However, the estimated biomass of attached biofilm was larger than the actual value, because the occupied volume of the attached biofilm was neglected in this estimating method. Therefore, at the end of Run 3, another protocol was adopted for separating the suspended and attached sludge in the SB reactor. After stopping the influent and sludge return from settling tank, tap water was feed at a flow rate of 30 L d^{-1} directly into the SB reactor with aeration. Almost all suspended sludge was washed out from the SB reactor and only attached biofilm remained in the reactor after 24 h.

5.2.5 Analytical methods

VFAs concentrations were determined by using a CTO-10AS liquid chromatography (Shimadzu, Japan). Gas produced from the AF reactor was analyzed by using a GC-14B gas chromatograph (Shimadzu, Japan).

Other analytical methods are the same as those described in Chapters 2 and 3.

5.2.6 Calculation methods

In this Chapter, observed sludge yields (Y_{obs}) were calculated according to Eq. (3-4), but W_{out} in this chapter presents the total amount of sampled sludge for analysis and occasional washout sludge (mg).

Nitrification efficiencies in the SB reactor were calculated according to Eq. (5-1) :

$$NE = 100 \times (TN_{AF} - NH_4\text{-}N_{fi})/TN_{AF}, \quad (5\text{-}1)$$

where NE = nitrification efficiency in the SB reator (%); TN_{AF} = AF effluent TN concentration (mg L^{-1}); $NH_4\text{-}N_{fi}$ = final effluent NH_4-N concentration (mg L^{-1}).

Final effluent COD concentrations were corrected in order to estimate the oxygen demand only due to the organic substance according to Eq. (5-2) :

$$COD_{fi} = COD_{total} - 1.14 \times NO_2\text{-}N_{fi}, \quad (5\text{-}2)$$

where COD_{fi} = corrected final effluent COD concentration (mg L^{-1}); COD_{total} = measured final effluent COD concentration (mg L^{-1}); $NO_2\text{-}N_{fi}$ = final effluent NO_2-N concentration (mg L^{-1}); 1.14 = the molecular ratio of oxygen to nitrogen.

5.3 Results and discussion

5.3.1 Hydrolysis and fermentation performance in the AF reactor

Table 5-2 shows the changes in VFAs and lactate concentrations during Runs 3 and 4. The average influent total VFAs (as COD) of about 250 and 340 mg L^{-1} increased to about 870 and 1,200 mg L^{-1} in the AF effluent in Runs 3 and 4, respectively. The increase of about 250%

in total VFAs suggested that a strong acid fermentation reaction occurred in the AF reactor. Meanwhile, most influent lactate was fermented to acetate during AF treatment. At the end of Run 4, the gas production rates ranged from 1.5 to 2 L d^{-1}, and the main components were methane (CH_4) and carbon dioxide (CO_2) at about 50% and 15%, respectively. However, the calculated CH_4 conversion efficiency was only about 0.015 L g^{-1} $COD_{removed}$, which is much lower than a standard value of 0.40 L g^{-1} COD at 35°C (Metcalf & Eddy, 2003). Zhang et al. (2008) obtained a gas production level (CH_4 was about 76%) of up to 0.38 L g^{-1} COD in an up-flow anaerobic sludge bed (UASB) reactor using PVA-gel beads as the biomass carriers for treating high-strength CSL. Compared with these results, the methanogenic activity of the AF reactor was negligible, due to different operational conditions, such as HRT, pH, OLR, substrate concentration and biomass carriers. In this study, the HRT of the AF reactor was usually only about 4 h that was not long enough for further VFAs fermentation, leading to accumulation of VFAs and reduction in pH, further inhibiting methanogenic activity.

Table 5-2 VFAs and lactate concentrations in Runs 3 and 4

Day	OLR_{wh}[a] kg COD m^{-3} d^{-1}	Sa.[b]	Acetate mg L^{-1}	Propionate mg L^{-1}	Isobutyrate mg L^{-1}	Butyrate mg L^{-1}	Isovalerate mg L^{-1}	Lactate mg L^{-1}	Total VFAs mg COD L^{-1}	VFAs & lac[c] mg COD L^{-1}
		Inf	23	140	0	0	0	374	236	636
80	3.5	AF	299	235	6	78	19	4	866	870
		SB	0	0	15	0	0	0	27	27
		Inf	30	145	0	0	0	383	252	662
102	3.5	AF	326	226	8	79	17	9	881	891
		SB	0	0	0	0	0	1	0	1
		Inf	25	210	0	0	0	488	343	865
132	4.5	AF	389	339	7	62	40	9	1136	1145
		SB	0	0	0	0	0	0	0	0
		Inf	36	203	0	0	0	499	344	878
158	4.5	AF	398	346	7	95	51	10	1238	1248
		SB	0	0	0	0	0	0	0	0

Superscript: (a) OLR for the whole system; (b) sample: Inf: influent; AF: AF effluent; SB: SB effluent; (c) sum of total VFAs and lactate.

Influent organic nitrogen, as in protein form, was hydrolyzed to amino acids and then further converted to the ammonia form in the AF reactor. The release of ammonium by anaerobic ammonification in the AF reactor would be preferable so as to allow for sequential

aerobic nitrification (Chen et al., 2009). This was considered beneficial to SND performance in the SB reactor. Moreover, the increase in alkalinity caused by the release of ammonia replenished the alkalinity consumed during acid fermentation as was evidenced by the elevated reactor pH, which ranged from 5.0 to 5.5 during most of the experimental period (Fig. 5-2).

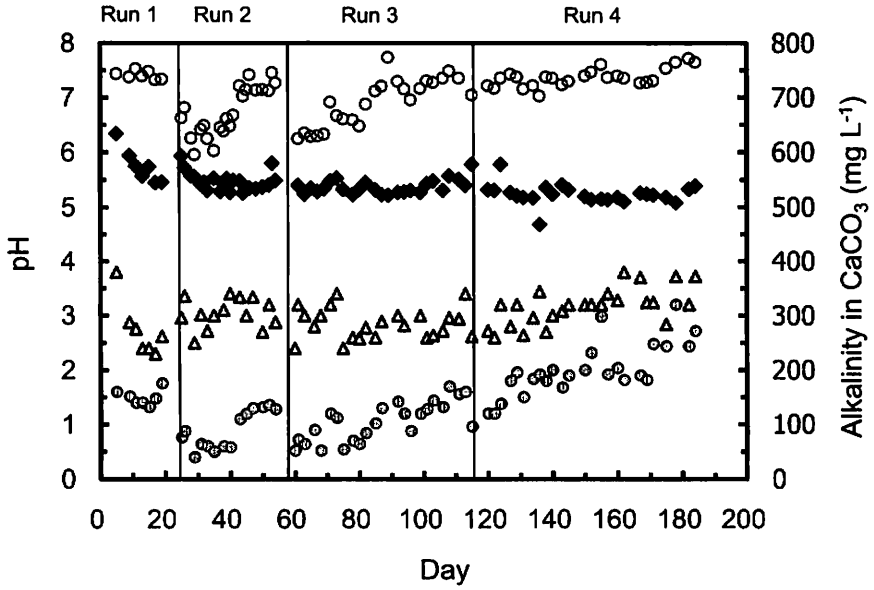


Fig. 5-2 Profiles of pH and alkalinity in AF and final effluents.

(◆) AF effluent pH; (○) final effluent pH; (△) AF effluent alkalinity; (●) final effluent alkalinity.

Effluent SS concentrations from the AF reactor fluctuated in the range of 30 to 150 mg L^{-1} and were usually under 100 mg L^{-1} due to the good sludge retaining capacity of the BL carriers. At the end of each run, the attached biofilm on the BL carriers was collected and quantified. Based on that data, the MLSS levels in AF reactor were estimated to be 3,950, 5,500, 8,800, and 11,000 mg L^{-1} in all runs sequentially, while the MLVSS/MLSS ratios of all samples were about 0.93. These high biomass concentrations could be responsible for the stable fermentation performance in the AF reactor.

5.3.2 COD removal performance

Fig. 5-3 shows daily COD removal performance for the whole system. Influent COD was removed efficiently over the whole system with average removal efficiencies of 94% to 99% during steady-state operations. Due to fermentation rather than methanogenesis being exhibited in the AF reactor, total COD removal efficiencies for the AF reactor were in the

range of only 5% to 20%, with an average of 13.5%. Thus, the actual OLRs for the SB reactor (OLR_{SBS}) were calculated to be 2.4, 4.0, 5.1 and 6.1 kg COD m^{-3} d^{-1} over Runs 1, 2, 3 and 4, respectively.

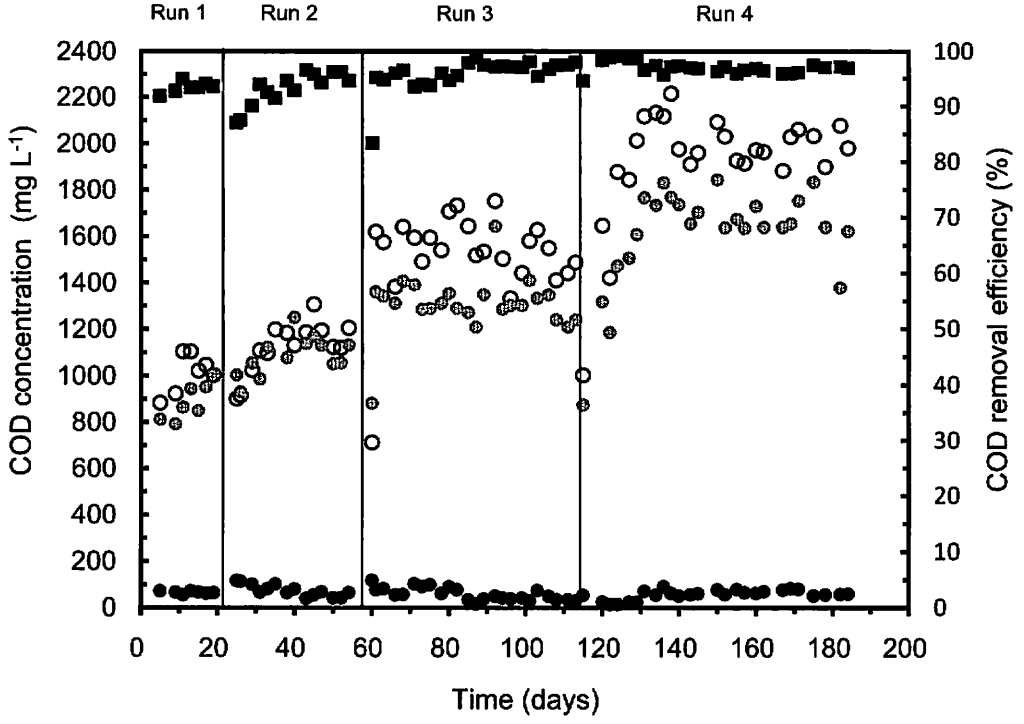


Fig. 5-3 Changes in COD removal performance.

(■) COD removal efficiency; (○) influent COD; (◍) AF effluent COD; (●) final effluent COD.

From Runs 1 to 3, slightly higher system effluent COD concentrations (100 to 120 mg L^{-1}) were observed during the start-up period of each run due to sludge acclimation in the SB reactor. However, after 7 to 10 days of operation, final effluent COD concentrations gradually decreased to 70, 50 and 40 mg L^{-1}, with COD removal efficiencies of 94%, 96% and 97%, respectively. However, just after the start-up period in Run 4, the final effluent COD concentration was only 30 mg L^{-1} even though the influent COD was higher than previous periods. This exceptionally high COD removal performance is attributed to the difference in seeding sludge in Run 4. Firstly, the initial amount of sludge for Run 4 was 147 g (dry weight), which was larger than those for other runs. Secondly, the biofilm attached on the BF carrier was separated from suspended sludge in the SB reactor at the end of Run 3, thus all this part of the sludge was inoculated in Run 4, accounting for a ratio of 33% of the total seed biomass. Therefore, the large amount of seed sludge and the high proportion of attached biofilm in the seed sludge enhanced the removal performance during the early period of Run 4.

Subsequently, the COD removal efficiencies were greater than 97% with the final effluent COD level below 80 mg L^{-1}.

The SB reactor thus demonstrated high COD removal efficiencies irrespective of OLR, with the highest COD removal rate of 5.9 kg COD m^{-3} d^{-1} occurring at an OLR_{SB} of 6.1 kg COD m^{-3} d^{-1} in Run 4. In a similar study[14] focused on COD removal in a suspended hybrid biological (suspended packing IFAS) reactor treating domestic wastewater, a COD removal efficiency of 83.7% was obtained at an ORL of 3.04 kg COD m^{-3} d^{-1}.

5.3.3 Nitrogen removal performance

Nitrogen removal performance is shown in Fig. 5-4. By calculation (Anthonisen et al., 1976), it can be inferred that no nitrogen loss due to volatilization of ammonia gas in the AF reactor occurred because of the low pH (5 to 5.5) throughout most of the main experimental period (Fig. 5-2). TN concentrations in the AF effluent were measured without filtration, so that these values were fluctuated and sometimes higher than the influent TN concentrations. The average TN removal efficiency of the AF reactor was about 3% attributable to bacteria growth requirements. Consequently, the actual NLRs for the SB reactor (NLR_{SB}s) were calculated to be 0.16, 0.28, 0.36 and 0.47 kg N m^{-3} d^{-1} for Runs 1 to 4, respectively.

In Run 1, effluent NH_4-N levels from the AF reactor increased from 50 to 62 mg L^{-1} and effluent NH_4-N levels from the SB reactor were in the range of 10 to 15 mg L^{-1} with about 82% of nitrification efficiency. However, NO_2-N accumulation was observed in the SB reactor with a concentration of 19 mg L^{-1} on day 5 with no NO_3-N detected in the final effluent. Nitrite-oxidizing bacteria (NOB) are more sensitive than ammonia-oxidizing bacteria (AOB) to free ammonia inhibition (Anthonisen et al., 1976), which could have caused nitrite accumulation during the early period of operation. Free ammonia concentration was estimated to be 0.2 mg L^{-1} on day 5, which would inhibit NOB more than AOB (Anthonisen et al., 1976). However, final effluent NO_2-N levels gradually decreased to 1.5 mg L^{-1} by day 17 while NO_3-N levels increased to 14 mg L^{-1}. This result indicated that NOB were gradually enriched. During that period, pH values decreased from 7.6 to 7.3 (Fig. 5-2), accompanying with a gradual improvement of nitrification. TN removal with an efficiency of 38.5% was observed by the end of Run 1.

Within 5 days after the start-up of Run 2, TN removal efficiencies reached 42.6%. Then, the efficiencies gradually increased to above 84% from day 43 to the end of this run with final effluent TN levels less than 12 mg L^{-1}. Concurrently, the final effluent NH_4-N levels fluctuated around 10 mg L^{-1} during the start-up period, but were always below 3 mg L^{-1} with nearly complete nitrification efficiencies reaching more than 98%. Furthermore, no NO_2-N accumulation in the SB reactor occurred during Run 2. Final effluent NO_3-N levels were about 40 mg L^{-1} during the first 10 days in this run and decreased sharply to below 10 mg L^{-1} afterwards.

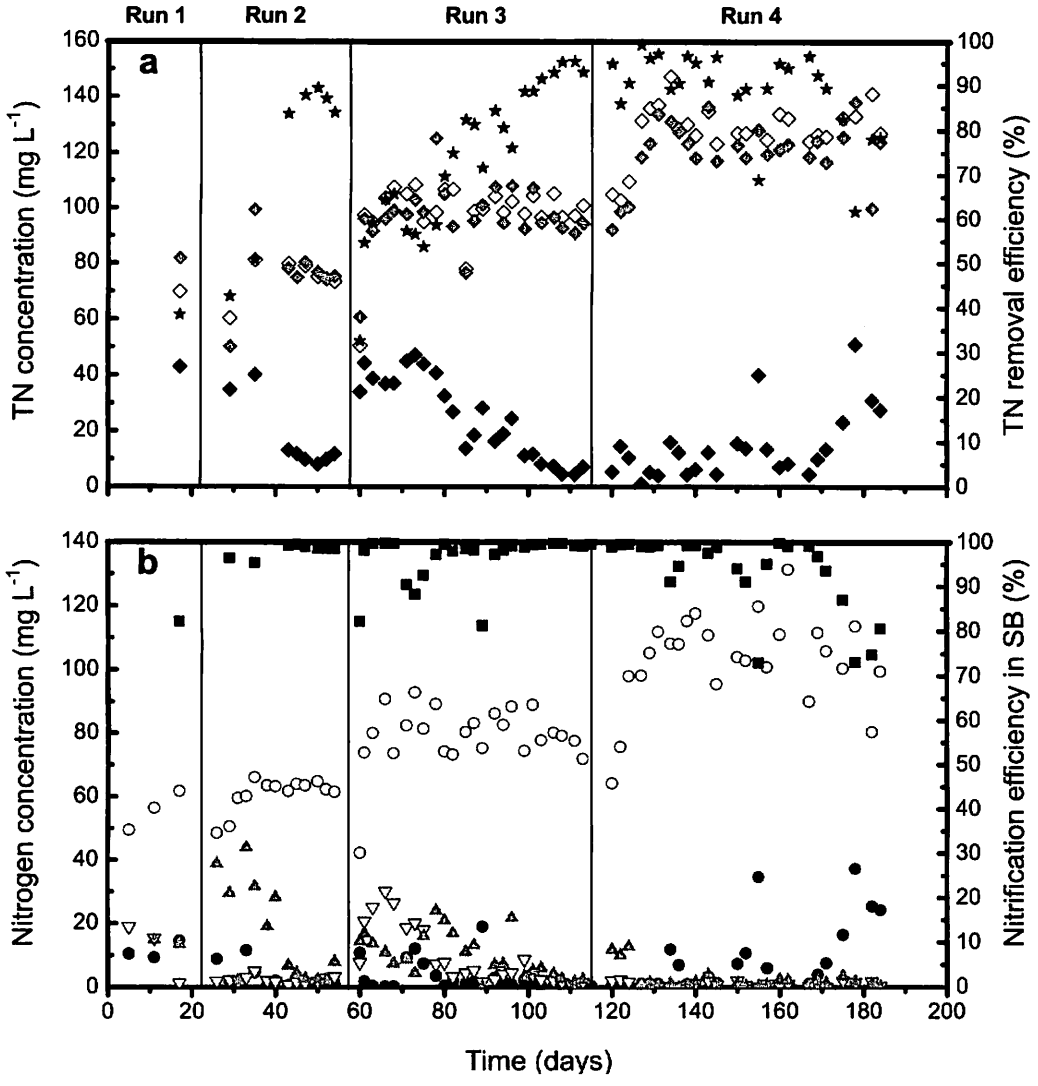


Fig. 5-4 (a) Changes in TN removal performance.

(★) TN removal efficiency; (◇) influent TN concentration; (✙) AF effluent TN concentration; (◆) final effluent TN concentration; (b) profiles of nitrogen concentrations and nitrification efficiency in SB: (■) nitrification efficiency in SB; (○) AF effluent NH_4-N concentration; (●) final effluent NH_4-N concentration; (▽) final effluent NO_2-N concentration; (▲) final effluent NO_3-N concentration.

In Run 3, influent TN levels were kept low during the start-up period, then increased to about 105 mg L^{-1} on day 60. On day 61, TN removal efficiencies reached 54.6%, while final NH_4-N, NO_3-N and NO_2-N concentrations were 2, 17 and 21 mg L^{-1}, respectively. In addition, NO_2-N accumulation was similar to that observed during the start-up of Run 1. Afterwards, TN removal efficiencies gradually increased to 82% on day 85. The final effluent NO_2-N levels decreased to 5 mg L^{-1}, while NH_4-N and NO_3-N concentrations oscillated in a range from 1 to 12 mg L^{-1} and 5 to 24 mg L^{-1}, respectively. On day 88, trouble with the air supply resulted in a lack of aeration for a whole night, leading to a sudden increase in NH_4-N to 18.94 mg L^{-1} with a decrease in TN removal efficiency to 71.6%. However, the system recovered quickly and TN removal efficiencies were restored to 84% by day 92, and then reached the highest value of 95% by the end of Run 3.

During the start-up period of Run 4, a TN removal efficiency of 94.9% was obtained in only 5 days of operation. The reasons for this quick response were thought to include the low DO concentration (1.7 to 2 mg L^{-1}) in the SB reactor and the large amount of inoculum applied. From day 125, the influent TN was increased to 130 mg L^{-1}, but TN removal efficiencies were still above 90% and reached 98% on day 127. On days 134 and 155, TN removal efficiencies decreased with final effluent NH_4-N levels increasing to a maximum of 35 mg L^{-1}, though NO_3-N and NO_2-N levels were unchanged (<3 mg L^{-1}). These deteriorations of TN removal were caused by the sludge washout which occurred twice due to deteriorated settling characteristics and pipe clogging. However, these deteriorated TN removal efficiencies were recovered within 2 or 3 days. Owing to temperature decrease (from 26 to 20°C) and sludge washout in the SB reactor, TN removal efficiencies decreased from 90% to 78% from day 175 to the end of this experiment with only leaving NH_4-N in the final effluent. Over the whole period in Run 4 when elevated NH_4-N levels appeared in the SB reactor, final effluent NO_3-N and NO_2-N levels were still maintained below 3 mg L^{-1} (Fig. 5-4b). These results show that the nitrification process is the rate-limiting step for TN removal in the SB reactor.

Over the course of this study, TN removals mainly occurred in the SB reactor. After the initial phase (Run 1), nitrifying organisms accumulated in the SB reactor and then high TN removal efficiencies of 85% to 97% were achieved during steady-state operations in all subsequent runs. During the brief period in Run 4 (days 120 to 171), the highest averaged TN

removal rate of 0.43 kg N m^{-3} d^{-1} occurred in the SB reactor.

5.3.4 Sludge characteristics in the SB reactor

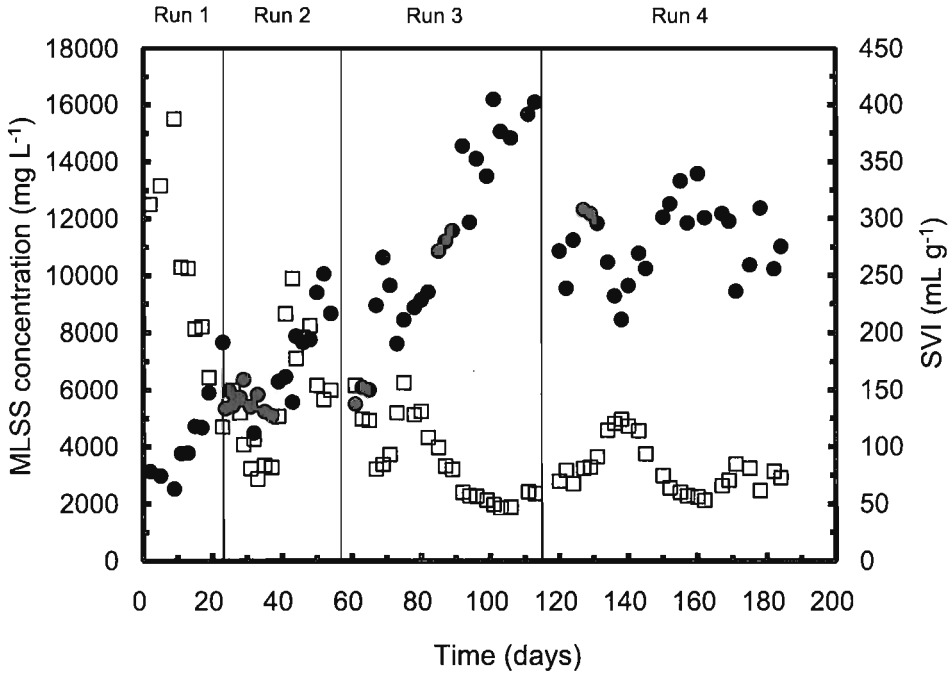
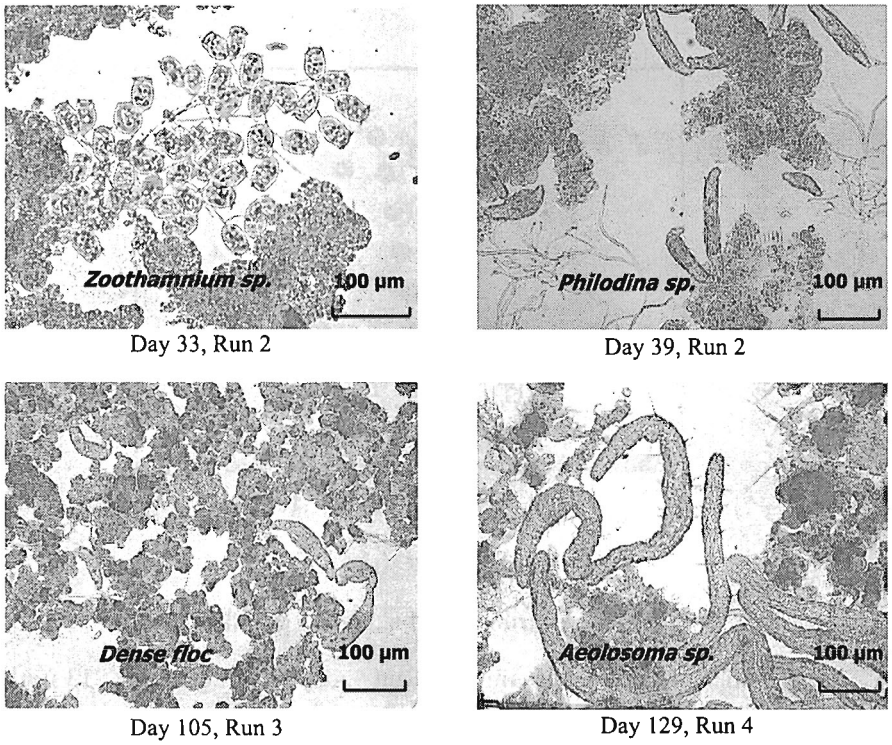


Fig. 5-5 Profiles of MLSS level and SVI value of sludge in the SB reactor.

(●) MLSS; (□) SVI.



Day 33, Run 2 Day 39, Run 2

Day 105, Run 3 Day 129, Run 4

Fig. 5-6 Microscopic observations of sludge in the SB reactor.

Fig. 5-5 shows the changes in MLSS and SVI in the SB reactor during the main experimental period. MLSS levels gradually increased and reached a maximum of 16,000 mg L^{-1} with a lowest SVI value of about 50 by the end of Run 3. This MLSS concentration was several times higher than that in CAS processes and higher than that in most IFAS processes, and comparable to that in some membrane bioreactors (MBRs) (Li et al., 2008). Subsequently, the MLSS oscillated around 11,000 mg L^{-1} during Run 4. Such high MLSS concentrations enabled the SB reactor to remove COD and NH_4-N at extremely high rates.

The low SVI and high MLSS in the SB reactor could be explained by following factors. Firstly, the IFAS process is widely known to enhance sludge settleability (Metcalf & Eddy, 2003) as has been verified by Gebara (1999). Secondly, the "swimming motion" of the BF carrier can be inferred to enhance the sludge settleability due to the strong hydraulic shearing force produced by the down-flow water, which causes (1) the biofilm to consolidate or become more densely packed (Laspidou et al., 2004b), and (2) the detached biofilm and suspended flocs to consistently obtain a uniformed shape (Wilén et al., 2003) as shown in Fig. 5-6 (day 105). Thirdly, the sludge settleability was improved by enhanced aggregation due to the proliferation of rotifiers (*Philodina sp.*) (Lapinski et al., 2003) (Fig. 5-6, days 39 and 105). Lastly, the increment of floc size (Fig. 5-7) was also in favor of the improvement of sludge settleability.

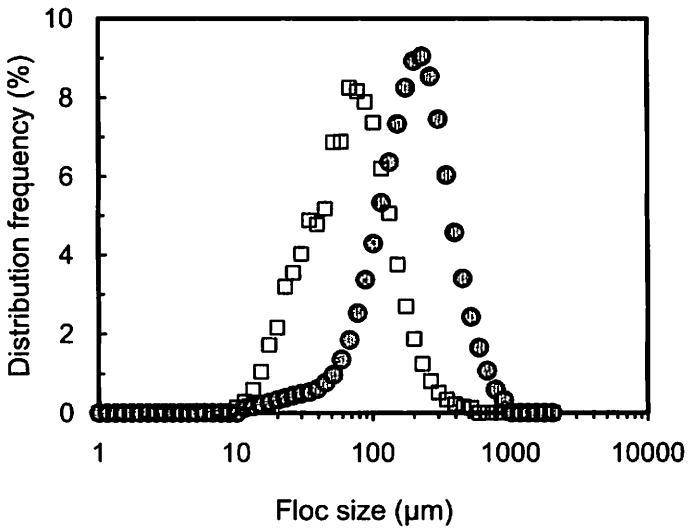


Fig. 5-7 Floc size distribution of sludge in the SB reactor.

(□) Seed sludge, mean size: 76 μm; (◎) Run 4, day 149, mean size: 213 μm.

In addition, the MLVSS/MLSS ratios of aerobic sludge were high (92-94%), which

indicates ideal conditions for biological treatment.

5.3.5 Excess sludge reduction

Y_{obs} values in this study were calculated to be 0.16, 0.13, 0.14 and 0.17 kg MLSS kg^{-1} $COD_{removed}$ in the four runs, sequentially. These values were markedly lower than typical values for CAS processes (0.3 to 0.5 kg MLVSS kg^{-1} COD), which is a point of great interest for WWTPs.

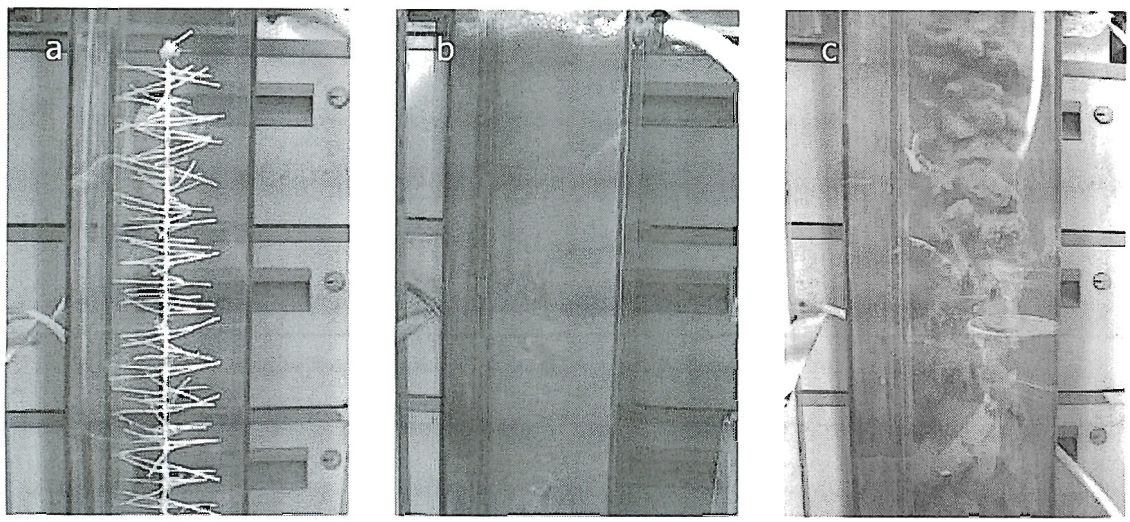


Fig. 5-8 Photograph of biofilm attached the BF carrier.

(a) vacant reactor; (b) running state in Run 3; (c) attached biofilm after suspended sludge removal at the end of Run 3.

One reason for these low observed sludge yields is regarded to be the microfaunas (protozoa and metazoa) predation on bacteria in the SB reactor. During transfer from a low trophic level to a high level, energy is lost due to inefficient biomass conversion (Ratsak et al., 1996). It is well known that aerobic biofilm processes generally have a more complex microbial ecology than that for activated sludge processes (WEF, 2000). Therefore, the low excess sludge production for the SB reactor was regarded to be related to the longer food chain compared with that in activated sludge. However, most predators such as protozoa and rotifers were reported to proliferate only in aerobic systems with high COD removal and nitrification efficiencies due to their sensitivity to DO limitation and inhibitory substances such as high concentration of ammonia (Metcalf & Eddy, 2003). Owing to the constantly high

COD removals and nitrification efficiencies in the SB reactor in all runs, various kinds of protozoa and metazoa appeared in the sludge of the SB reactor. Among these, *Philodina sp.* were predominant in Runs 2 and 3 (6,800 ind. mL^{-1}, on day 113). Especially, *Aeolosoma sp.*, which has more potential for sludge reduction than other predators due to its bigger size (Wei et al., 2003) was counted in 280 ind. mL^{-1} on day 130 in Run 4. Another reason for the effective reduction of excess sludge in this study was the unique microenvironment of the biofilm attached on the BF carrier. The configuration of the attached biofilm is shown in Fig. 5-8. The biofilm was very thick, containing black zones in poorly exposed areas due to formation of anaerobic conditions. Thus, the biofilm could provide an integrated aerobic, anoxic and anaerobic environment, allowing various trophic levels to exist, thereby reducing sludge yield[3] (Lin et al., 2009)

5.3.6 Mechanism of nitrogen removal via SND in the SB reactor

Table 5-3 Comparison of SND performance among different reactor setups

Reactor	Carriers	Influent	COD/N $g\ g^{-1}$	TN_{inf}[a] $mg\ L^{-1}$	COD removal $kg\ COD\ m^{-3}\ d^{-1}$	TN removal $kg\ N\ m^{-3}\ d^{-1}$	DO $mg\ L^{-1}$	pH	MLSS $g\ L^{-1}$	Reference
Moving Bed biofilm	PE balls[b]	Domestic wastewater	9.2	35	0.95	0.13	2	N/M	N/M	Wang et al., 2006
Fluidized bed	Rubber[c]	Sanitary wastewater	9	30-45	N/M	0.14	2	7-8	N/M	Wen et al., 2008
Immobilized-cell	PVA gel[d]	Beef + peptone	7.5	48	3.37	0.25	2.4-2.7	7.5	5-7[e]	Chen et al., 1998
MBR[f]	Fibrous[g]	Glucose + starch + NH_4Cl	N/M	160	N/M	63.6%[h]	0.3[i]	N/M	12.6-13.5	Li et al., 2008
AIC-MBR[j]	None	Sugar + NH_4Cl	9.59	42	0.86	0.073	0.8 (0.4)[k]	7.8	5 ± 0.3	Meng et al., 2008
Modified MBR[l]	None	Sucrose + NH_4Cl	9.3	215	1.28	0.13	N/M	7.6-8.5	N/M	Fu et al., 2009
Swim-bed (SB)	BF	Fermented CSL, VFAs	13-15	85-130	3.9-5.9	0.24-0.43	1.45-3.75	7-7.5	16	This study

Superscript: (a) influent TN concentration; (b) polyethylene carriers shaped like small balls with a diameter of 25 mm; (c) fragmentized rubber with a mean diameter of 3mm; (d) phosphorylated polyvinyl alcohol gel beads; (e) VSS (volatile suspended solid) concentration, immobilized on PVA gel beads; (f) internal-loop airlift MBR; (g) fibrous carrier composed of polythene and polyamide; (h) TN removal efficiency; (i) DO level in anoxic zone (47% of the total volume of reactor); (j) airlift internal circulation MBR; (k) 0.8 and 0.4 $mg\ L^{-1}$ in aerobic and anoxic zones, respectively; (l) airlift internal circulation MBR.

N/M: not mentioned.

The presented results in all literatures and this paper are all the optimal parameters in these studies.

Commonly, nitrogen is removed through cell assimilation (microbial growth) and the biological nitrification-denitrification process. To clarify the nitrogen removal mechanism in this study, the microbial growth was firstly evaluated. The sludge yield and the nitrogen content (about 7%) of sludge were measured, so that the nitrogen removal by cell assimilation was estimated to be about 14-18% of TN removal. Additionally, only 3% of TN was removed in the AF reactor. Therefore, TN removal in our treatment system appeared to be primarily due to simultaneous nitrification and denitrification (SND) in the SB reactor.

Many researchers have reported SND in various kinds of reactors, such as biofilm reactors (Chen et al., 1998; Wang et al., 2006; Wen et al., 2008) and modified airlift MBRs with continuous aeration rather than intermittent aeration (Li et al., 2008; Meng et al., 2008; Fu et al., 2009) as shown in Table 5-3. In addition, He et al. (2009) investigated the effect of biological factors such as DO, COD/N ratio and pH on SND performance in a membrane bioreactor. Those key factors are discussed below in light of the results observed.

DO level for SND in the SB reactor

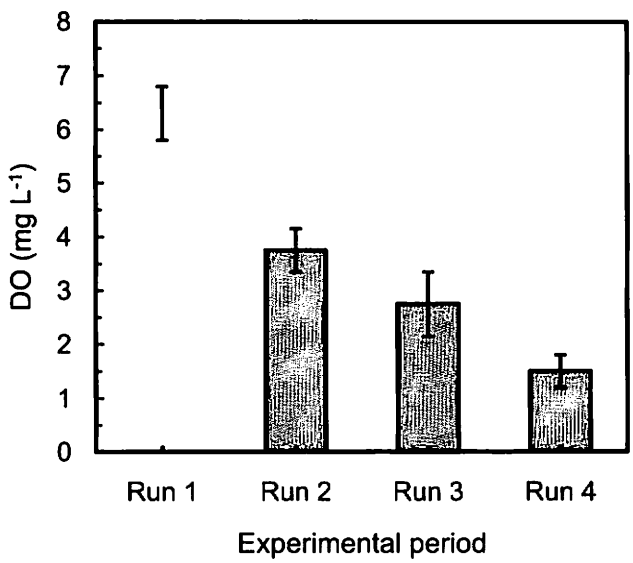


Fig. 5-9 DO levels in the SB reactor in different runs: bars indicate standard deviation.

DO level is regarded to be the most important factor for the occurrence of SND in most studies as shown in Table 3. He et al. (2009) also implied that a good SND performance was generally obtained at a DO level of 0.8 mg L^{-1} in a MBR. However, biofilm process could

achieve SND at a considerably higher DO level (1.5 to 3.75 mg L^{-1}, Table 5-3) due to gradients in the biofilm.

For a poorly soluble substrate (e.g. oxygen), the penetration depth into flocs or biofilm is relatively shallow (typically 100-150 mm for oxygen) (Denac et al., 1983). Furthermore, Laspidou et al. (2004a) found that the DO concentration at the bottom of biofilm with a 280-μm thickness was 0.49 mg L^{-1}, which is generally considered to be the limitation (0.5 mg L^{-1}) for denitrification (Wang et al., 2006). For the uniformly distributed aerobic condition in the SB reactor, the average DO concentrations in the bulk liquid were 6.3, 3.75, 2.75 and 1.5 mg L^{-1} during Runs 1, 2, 3 and 4, respectively (Fig. 5-9). It should be noted that these DO concentrations in the SB reactor were measured at 20 cm under the effluent port of the reactor. Generally, the measured DO levels were about 0.3 mg L^{-1} higher than those at the bottom of downdraft section in the reactor. This indicates that the lowest DO concentration (in Run 4) of the bulk liquid in the SB reactor was also higher than 1.0 mg L^{-1}.

The attached-growth in the SB reactor was approximately 27, 30, 49 and 55 g at the end of Run 1, 2, 3 and 4, respectively. It is not easy to measure the thickness of the biofilm. Thus, the average thickness could only be estimated by an approximate biofilm density in literatures. Taking the 49 g in Run 3 as an example, allowing for the specific surface area of BF being 0.2 m^2 m^{-1}, the average thickness could be estimated over than 3.1 mm even using the biggest biofilm composition density of 160 kg m^{-3} in the literature (Laspidou et al., 2004a). Actually, the biofilm thickness was far beyond this value owing to the 3-dimensional configuration of the BF carrier (Fig. 5-1c), and the biofilm appeared thicker along the BF yarns depth (Fig. 5-8). Even using the estimated value, the biofilm thickness was also far beyond the thickness (280 μm) of oxygen limitation. Thus, the thick biofilm attached on the BF carrier could create a favorable anoxic environment for denitrification.

The DO transfer limitation also might exist in some bigger suspended flocs in the SB reactor. Fig. 5-7 shows that the mean size of the suspended flocs in Run 4 was 213 μm. Although the average DO concentration in the bulk liquid was around 6.3 mg L^{-1} in Run 1, denitrification occurred with the final effluent NO_3-N below 15 mg L^{-1}. At the extremely high DO level in the bulk liquid, biofilm could limit DO penetration much more than suspended flocs, thus this denitrification was mainly performed in the biofilm rather than suspended

flocs. Furthermore, TN removal efficiencies deteriorated in Run 4 (days 134 and 155) because of the sludge washout and subsequent sharp decrease in MLSS concentration (Fig. 5-5). Accompanying the washout of suspended sludge, the TN removal deteriorated owning to the decrease in nitrification rather than denitrification. Thus, it can be deduced that the suspended sludge was mainly engaged in nitrification and denitrification occurred mainly in the biofilm.

pH for SND in the SB reactor

The relationship between the TN removal efficiency and the pH is shown in Fig. 10, which indicates clearly that the most suitable pH for SND in this study was 7 to 7.5. This value is a little lower than the reported values listed in Table 5-3, but very close to the result reported by He et al. (2009). In addition, the alkalinity of the final effluent increased due to denitrification (Fig. 5-2).

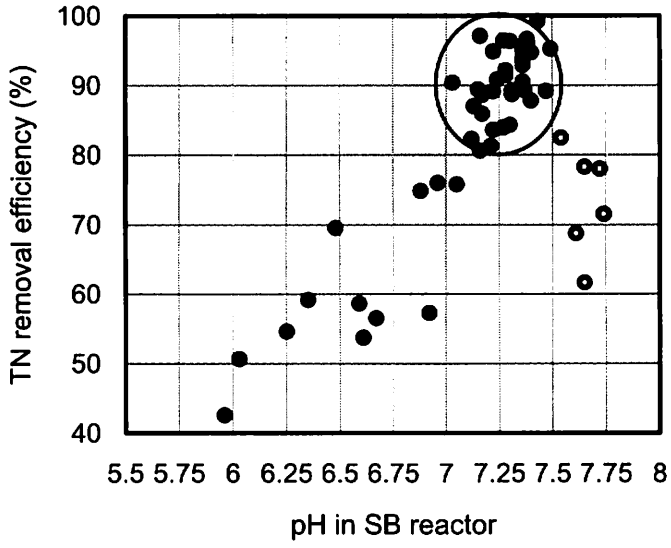


Fig. 5-10 Relationship between pH in the SB reactor and TN removal.
(●) results in normal operation; (○) results in mechanical troubles or sludge washout.

COD/N ratio and VFAs for SND in the SB reactor

COD/N ratio is also very important to SND, because denitrification process requires sufficient organic carbon as electron donor. The stoichiometric requirement for conventional denitrification is 2.86 g COD g^{-1} N (Metcalf & Eddy, 2003), allowing for the electron

transmitting balance between organic carbon and NO_3-N. However, previous studies indicated that the suitable COD/N ratio ranged from 7.5 to 9.6 g COD g^{-1} N for good SND performance (Table 5-3). A low COD/N will enhance nitrification capacity, but the TN removal will be impaired by insufficient electron donors for denitrification (Fu et al., 2009). Conversely, a high COD/N ratio will provide enough electron donors for denitrification, but the TN removal will be impaired because the nitrification reaction will be inhibited due to oxygen competition with heterotrophic bacteria (Meng et al., 2008). Thus, it would appear that the COD/N ratio (13-15 g COD g^{-1} N) in this study was too high for optimal SND performance; however, this was not the case, apparently due to the good nitrification attributed to the exceptionally high MLSS in the SB reactor.

Denitrifying organisms were reported to utilize acetate and propionate as electron donors preferentially due to their simple metabolic pathway in denitrification process (Elefsiniotis et al., 2004). In this study, the main components of the VFAs produced by the AF reactor were acetate and propionate, which were completely consumed in the SB reactor (Table 5-2). This result could explain high SND performance in the SB reactor.

In addition, Lakshmi et al. (2008) reported that the external mass transfer coefficient increased with increase in biofilm thickness, possibly due to the increases in bioparticle diameter and biofilm porosity concurrently. The very thick biofilm attached on the BF carrier could facilitate the mass transfer of substrates. Thus, VFAs produced in the AF reactor and NO_3-N produced in the SB reactor could diffuse into the biofilm and satisfy the growth requirement of denitrifying organisms within it.

Furthermore, the VFAs produced in the AF reactor could also be consumed by the aerobic heterotrophs in the SB reactor. The large amount of organic carbon produced in the AF reactor was probably consumed out of the anoxic part of the biofilm. Therefore, it is reasonable to assume that the endogenous carbon source was utilized for the denitrification process in this case. However, the substrate (VFAs) competition mechanism between aerobic heterotrophic bacteria (in suspended flocs and on the outer layer of biofilm) and anoxic heterotrophic denitrifying organisms in the biofilm remains unclear. With the aid of microelectrode techniques and microbial activity analysis (Zhou et al., 2008), further investigation might be desirable to study the VFAs, NO_3-N, redox potential (ORP) and DO profiles within the

biofilm for a better understanding of the SND mechanism in the SB reactor.

Another possibility of TN removal in the SB reactor is aerobic denitrification (Oguz et al, 2007). They implied that aerobic denitrification pathway could be activated by the addition of VFAs. However, the DO concentration always kept at 3 mg L^{-1}, and no any other limitations existed in their experiment. Due to the difference in experimental conditions, the aerobic denitrification pathway is negligible in our study.

In short, the SB reactor could produce the major favorable factors for SND, such as the thick biofilm which facilitated the denitrification, the high MLSS levels which were beneficial to good nitrification, the suitable pH and the suitable substrates for denitrification. These factors accounted for the higher TN removal rates (0.24 to 0.43 kg N m^{-3} d^{-1}) in the SB reactor than those in other SND reactors (Table 5-3).

5.4 Conclusions

In this study, an attached growth system was evaluated for carbon and nitrogen removals in treating moderately high-strength synthetic organic wastewater. The results revealed excess sludge reduction of the whole system and SND in the aerobic SB reactor.

(1) The combined system obtained 84% to 97% of TN removals mainly via SND at OLRs from 2.7 to 4.5 kg COD m^{-3} d^{-1} and excess sludge productions were from 0.13 to 0.17 kg MLSS kg^{-1} $COD_{removed}$.

(2) The AF reactor contributed hydrolysis and fermentation to the treatment train. VFAs and ammonia produced in the AF reactor were suitable substrates for the subsequent SND process.

(3) The SB reactor, applied as an IFAS process, demonstrated good treatment performance for COD and TN removals and excess sludge reduction. Maximum COD and TN removal rates of 5.9 kg COD m^{-3} d^{-1} and 0.43 kg N m^{-3} d^{-1}, respectively, were obtained. Moreover, the MLSS concentration in the SB reactor reached a maximum of 16,000 mg L^{-1}.

(4) Nitrification that occurred in the SB reactor is thought to be mainly due to the activity of

suspended bacteria, while denitrification occurred mainly in the biofilm. The thick biofilm attached on the BF carrier was considered to be essential for SND, allowing for good SND performance over a very wide range of bulk DO concentration (1.5 to 3.75 mg L^{-1}).

5.5 References

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Chapter 6 Conclusions and recommendations

6.1 Conclusions

Applied as an aerobic IFAS process, the swim-bed technology using the BF biomass carriers has been proved to be an effective approach to upgrade the CAS process and enhance the nitrogen removal performance.

1) Through comparison among two kinds of acrylic fiber biomass carries, a net type carrier in three parallel reactors with different aeration modes, results showed that the BF biomass carrier packed in a cycle aeration reactor demonstrated the most stable and predominant performance in sludge attachment and retention, organic pollutant removal, ammonification and nitrification.

2) The BF biomass carrier was partially packed in a plug-flow activated sludge reactor with a packing ratio of 15%. High s-BOD_5, s-COD, and ammonium removal efficiencies of 99.1%, 96.5% and 83.6%, respectively were obtained under VLR of 4.5 kg s-COD m^{-3} d^{-1}. SVI values below 50 mL g^{-1} demonstrated satisfactory settling characteristics of the sludge, which were attributed to the increase in the size of the biomass flocs as a result of the application of BF carrier. Relatively low viscosity of mixed liquor also facilitated sludge settling performance. Low sludge yield is attributed to the existence of a large amount of protozoa and metazoa. The results of demonstrated that partially packing of the BF carrier could enhance treatment performance and provided process stability to the CAS process.

3) Through fixed packing with BF, suspended type MLE process was shifted to an IFAS process. During a 110 days of operation, 70% of TN removal, 80% of COD removal and 90% of BOD removal efficiencies were obtained stably, even at a very short HRT of 2.8 h. Meanwhile, observed sludge yield of 0.13 kg MLSS kg^{-1} $COD_{removed}$ was obtained during the whole experimental period and the lowest observed yield of 0.05 kg MLSS kg^{-1} $COD_{removed}$ was obtained under VLR of 1.75 kg BOD m^{-3} d^{-1}. Furthermore, the MLSS concentrations reached more than 8,000 mg L^{-1}.

4) Excess sludge reduction and SND were evaluated in an attached-growth treatment system consisting of a down-flow anaerobic fermentation (AF) reactor and an aerobic swim-bed (SB) reactor operated as an IFAS process. The AF reactor, located upstream of the SB reactor, provided hydrolysis, fermentation and anaerobic ammonification. Stable TN removal efficiencies ranging from 85% to 97% were obtained via SND in the SB reactor. In addition, observed sludge yields for the whole system ranged from 0.13 to 0.17 kg MLSS kg^{-1} $COD_{removed}$; furthermore, the MLSS levels were maintained at about 10,000 mg L^{-1} in the SB reactor. Exceptionally high COD and TN removal rates of 5.9 kg COD m^{-3} d^{-1} and 0.43 kg N m^{-3} d^{-1}, respectively, were observed in the SB reactor.

In general, applied as an aerobic IFAS reactor, the swim-bed technology demonstrated remarkable pollutant removal performance for high-strength organic wastewater and low-strength, especially in nitrogen removal and excess sludge reduction.

6.2 Recommendations

In order to extend the application of BF biomass carrier to full-scale aerobic reactors, the following recommendations are put forward:

(1) Based on the results obtained, experiments using various real wastewaters are suggested.

(2) The phosphorus removal capability of swim-bed reactors was revealed low in this study, thus a new technology which enables phosphorus removal must be developed.

(3) The mechanism of the extremely high MLSS concentration obtained in the swim-bed reactor should be further investigated.

(4) The molecular biological analysis, such as denaturing gradient gel electrophoresis (DGGE) and fluorescence in situ hybridization (FISH) must be utilized to investigate excellent performance of swim-bed technology.

Appendix: Publications related to this dissertation

Journal papers

1. **Xiaochen Xu**, Hu Jin, Toichiro Koyama and Kenji Furukawa. Excess sludge reduction and simultaneously nitrification and denitrification (SND) performance by combining anaerobic fermentation with aerobic swim-bed reactors both using acrylic-fiber biomass carriers. Japanese Journal of Water Treatment Biology, 45 (3), 2009 (in press).
2. Yusuke Watanabe, Sen Qiao, **Xiaochen Xu**, Jiali Yang, Takashi Nishiyama, Takao Fuji, Toichiro Koyama, Zafar Bhatti and Kenji Furukawa. Studies on the Improvement of activated sludge treatment capability by using swim-bed technology. Japanese Journal of Water Treatment Biology, 45 (3), 2009 (accepted).

Conference proceedings:

1. **Xiaochen Xu**, Toichiro Koyama and Kenji Furukawa. Novel waste water treatment process combining an anaerobic filter and a swim-bed reactor using acrylic fiber biomass carriers. In: proceedings of an international conference symposium of environmental science and technology, Nov. 2007, Beijing, China. (CD-ROM), 695-706.
2. **Xiaochen** Xu, Hu Jin, Naoya Kawakami, Toichiro Koyama and Kenji Furukawa. Novel wastewater treatment process combining anaerobic and aerobic treatment using acrylic fiber biomass carriers. In: proceeding of the 43th annual conference of Japanese Society of Water Treatment Biology (JSWTB), Nov. 2007, Miyagi Japan,
3. Yusuke Watanabe, **Xiaochen Xu**, Kenji Furukawa and Toichiro Koyama. Study on high concentration activated sludge process using swim-bed technology. In: proceedings of the 42th annual conference of Japan Society on Water Environment (JSWE), March 2007, Nagoya, Japan.