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Multiple factors influencing anaerobic acidogenic pretreatment in an up-flow non-woven biofilm reactor

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HIGHLIGHTS

- A specific non-woven reactor is favorable for start-up of acidogenesis.
- Multi-factors have the comprehensive effects on VFAs production.
- The primary factors for VFAs composition are analyzed.

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ABSTRACT

Anaerobic acidogenesis in an up-flow non-woven biofilm reactor was investigated to convert chemical oxygen demand (COD) into volatile fatty acids (VFAs) as carbon source in the subsequent nitrogen removal process in synthetic domestic sewage pretreatment. A specifically designed non-woven padding inserted vertically into the reactor proved very favorable for acidogenic bacterial attachment. The characterization of the microorganisms was performed using scanning electron microscopy and fluorescent in situ hybridization. Multiple factors, including hydraulic retention time (HRT), pH, and influent COD and NH_4^+—N concentrations, played comprehensive roles in acidogenesis. Based on the Taguchi orthogonal array, the ordering sequence of the influential factors for VFAs production was influent NH_4^+—N ($R = 41.4$) > pH ($R = 37.3$) > influent COD ($R = 20.2$) > HRT ($R = 14.8$). The suitable conditions were HRT = 1.5 h, pH = 8.5, influent COD = 100 mg L^{-1}, and influent NH_4^+—N = 30 mg L^{-1}. Moreover, acetic acid, propionic acid, and *n*-butyric acid prevailed over other VFAs. Finally, the most influential factor for each VFA was demonstrated.

1. Introduction

Environmental issues on domestic sewage have become increasingly prominent with the upsurge of the discharge of nitrogen compounds into wastewater treatment plants [1]. A serious lack of readily biodegradable chemical oxygen demand (COD) as carbon source is a common problem that a low C/N ratio leads to the costly addition of external carbon sources for denitrification [2]. Generally, several-fold mg of COD (effective compounds are in fact VFAs substrates) is required for biological removal of 1 mg of NH_4^+—N. Thus, it would be of great value to maximize the VFAs production before denitrification in order to provide sufficient internal carbon source for denitrifying domestic sewage, or rather, this problem can be solved by anaerobic acidogenic pretreatment in the production of volatile fatty acids (VFAs). Which are more suitable carbon sources for denitrifying bacteria and have a higher rate as electron donors than methanol [3]. More recently, anaerobic acidogenesis has been also recognized as a further and promising solution for denitrifying domestic sewage without sufficient electron donors [4].

Factors such as the type of reactor, temperature, pH, hydraulic retention time (HRT), protein-to-carbohydrate ratio, as well as influent COD and NH_4^+—N [5–9], have been demonstrated in previous studies to affect anaerobic acidogenesis of high strength industrial wastewaters (e.g., slaughterhouse, pharmacy, COD above 20 g L^{-1}). With the results of these researches and considering that a single factor under ideal laboratory conditions cannot be practically achieved in full-scale plants, it could be concluded that the combination of the said each factors plays a crucial role in relation to control acidogenesis of the domestic sewage. However, information on the induction sequence of different influential factors on VFAs production is still limited, especially acidogenesis designed for domestic sewage. Assessing the effect of each factor on the entire operation can facilitate the rapid onset of acidogenesis. The

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combined influences of influent COD, NH_4^+—N, pH, HRT on production and composition of VFAs should be preferentially investigated, since the said four factors are the common controllable parameters for wastewater treatment plants in China.

In addition, although data on acidification of domestic wastewaters are in high demand, few studies have been carried out previously to assess the effects of the whole range of engineered reactor design parameters on acidification, especially for those municipal sewage treatment plants need to upgrade in order to be competent to perform acidogenesis [10,11]. Apparently, this type of reactor must meet the requirement of separating the acidogenic phase from the methanogenic phase through the different growth rates of acidogens and methanogens [12]. A prolonged period of cultivation should be avoided because of the competition for substrates between the said bacteria [13]. Some properties of acid-forming bacteria also need to be considered to establish rapidly a proper community of acidogenic bacteria in this type of reactor. For example, the settleability of acidogenic bacteria is much poorer than that of other anaerobic sludge, which means that a short HRT is favorable for acidogenic bacterial growth [14]. Based on these reasons, an attached growth reactor, which can prevent the washout of acid-forming bacteria from the reactor, may be an appropriate choice for maintaining a certain population of acidogenic bacteria.

In order to produce maximal VFAs from domestic sewage for subsequent denitrification, this paper focused on multiple effects of four controllable factors including pH, HRT, influent COD, and influent NH_4^+—N on both of production and composition of VFAs. Also, a novel up-flow non-woven biofilm reactor was specially designed for anaerobic acidogenic process. The advantages of the reactor and the suitable operating conditions of acidogenesis in pretreating domestic sewage were investigated.

2. Materials and methods

2.1. Up-flow non-woven biofilm reactor and synthetic wastewater

A sketch of the up-flow non-woven biofilm apparatus is shown in Fig. 1. A non-woven porous polyester (0.01 m^2 area) padding coated with a pyridinium-type polymer was inserted vertically into the reactor. A stirrer was used for mixing seed sludge to suspension consistency throughout the reactor at the start of the process. The subsequent sedimentation tank was equipped with a scraper to prevent accidental sludge loss. The experimental apparatus was hermetic and maintained at room temperature. Seed sludge was obtained from a local municipal wastewater plant (Dalian, China), and the initial mixed liquid suspended solids (MLSSs) used was 6000 mg L^{-1}. The synthetic wastewater contained sucrose $(NH_4)_2SO_4$, and KH_2PO_4 as nutrients. A trace element solution, which consisted of 66.9 mg L^{-1} $MgCl_2{\cdot}6H_2O$, 33.3 mg L^{-1} $CaCl_2$, 3.56 mg L^{-1} $FeCl_2{\cdot}4H_2O$, 0.4 mg L^{-1} $CoCl_2{\cdot}6H_2O$, 0.81 mg L^{-1} $NiCl_2{\cdot}6H_2O$, 0.25 mg L^{-1} $Na_2MoO_4{\cdot}2H_2O$, 0.21 mg L^{-1} $ZnCl_2$, and 0.36 mg L^{-1} $MnCl_2{\cdot}4H_2O$, was added to the reactor at regular intervals. The influent pH was adjusted through the addition of NaOH and H_2SO_4.

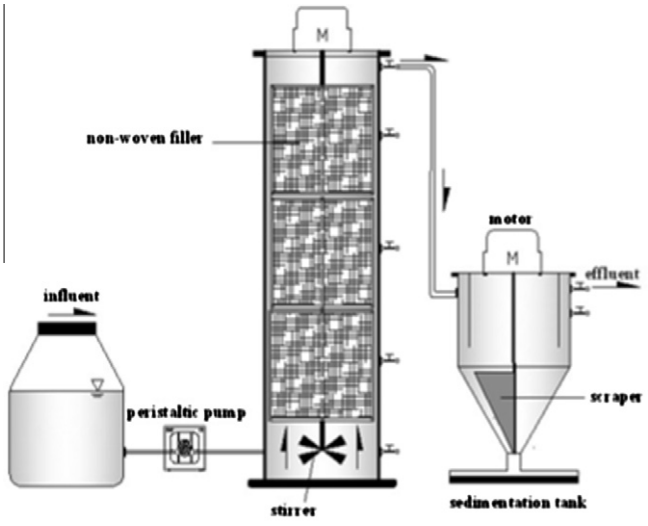

Fig. 1. Sketch of up-flow non-woven biofilm reactor.

2.2. Orthogonal tests

Taguchi orthogonal tests with an $L_9(3^4)$ array consisting of four factors and three levels are listed in Table 1. Appropriate levels of factors were designed according to the characteristics of domestic sewage and the operating conditions described in previous studies [15]. These tests were used to obtain the suitable operational conditions of acidogenesis, and each of the experimental conditions was maintained for a week. The experimental criteria in this study included the removal efficiency of COD, removal efficiency of NH_4^+—N, and the rate of VFAs/COD. T_1, T_2, and T_3 denote the average values at different levels of the same factor, and the range (R value) of each column, which represents the difference between the lowest and highest T values for a factor, was listed at the bottom of the table. According to the principle of the Taguchi orthogonal method, the induction sequence of factors could be determined by classifying the R values, that is, the corresponding factor with the largest R value was the most significant factor in the experimental criteria.

2.3. Analytical methods

The COD and NH_4^+—N concentrations were determined according to the standard methods [16]. pH was measured using a pH meter (Sartorius, PB-10, Germany). The filtrate obtained by nanofiltration was collected in a 1.5 mL gas chromatography vial for VFA analysis. Formic acid (3%) was added to adjust the pH to below 3.0. The concentration and composition of the VFAs were analyzed using a Shimadzu GC-2010 gas chromatograph equipped with the free fatty acid phase capillary column (30 m × 0.53 mm × 1.00 μm) and a flame ionization detector, according to the modified method [17]. Nitrogen was used as the carrier gas, and the flux was 50 mL min^{-1}. The injection port and the detector were maintained at 200 °C and 220 °C, respectively. The oven temperature was programmed to start at 110 °C, remain constant for 1 min, increase to 170 °C at a rate of 6 °C min^{-1}, and hold at 170 °C for an additional 5 min. The sample injection volume was 1.0 μL.

2.4. Characterization of microorganisms

The morphological characteristics of the biofilm were observed using scanning electron microscopy (SEM, JSM-5600LV, Tokyo, Japan). The samples were cut from the biofilm using a sterile blade, washed several times with phosphate buffer, and fixed with 2.5% glutaraldehyde for 24 h at 4 °C. Subsequently, the experimental samples were dehydrated through a graded series of tertiary butanol solution (50%, 70%, 80%, 95%, and 100%; 15 min for each concentration). Finally, the samples were evacuated for several hours and gold-coated using a sputter.

The composition of the microbial community in the biofilm was analyzed using fluorescent in situ hybridization (FISH). Details of the probes are listed in Table 2. Probes purchased from TaKaRa (Dalian, China) were fluorescence-labeled with fluorescein isothiocyanate dye (Cy3-red, FITC-green) and sulfoindocyanine dye (Cy5-