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# Treatment of low-strength wastewater at mesophilic and psychrophilic conditions using immobilized anaerobic biomass

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#### HIGHLIGHTS

• Immobilized biomass in AnIBPR removes COD at low temperature and short HRT.

• COD removal by AnIBPR was achieved at 15-35 °C.

• Dissolved CH<sub>4</sub> were oversaturated at saturation ratio of 1.02–1.21.

• Low K<sub>L</sub>a limited liquid-to-gas mass transfer of dissolved CH<sub>4</sub> at cold temperature.

• AnIBPR performance was not impeded at low VFA contents.

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#### ABSTRACT

An anaerobic immobilized bio-plates reactor (AnIBPR) of effective volume of 14.5 L and bio-plate packing ratio of 27.5% was used to treat low-strength synthetic wastewater (COD of 400 mg/L) at a hydraulic retention time (HRT) of 16 h. Treatment performance was investigated at different temperatures. The results showed that the reactor AnIBPR achieved COD removal of 86–92% at mesophilic (35 °C and 25 °C) and psychrophilic (15 °C) conditions. Dissolved methane (CH<sub>4</sub>) was found to be oversaturated in the reactor (saturation factor of 1.03–1.21), increasing with decreasing temperature. Increased dissolved CH<sub>4</sub> content at 15 °C, albeit at decreased production, was attributed to significant mass transfer limitation due to lower  $K_{La}$  at low temperature, resulting in oversaturation of dissolved CH<sub>4</sub>. Increasing treatment temperature to mesophilic conditions led to increased reaction rates and yields. At steady state, gasphase CH<sub>4</sub> content was 75–83% and the overall yield was 0.22–0.26 L/g COD removed. Steady-state volatile fatty acids (VFAs) were measured at 3.3–12 mg COD/L, small amounts that were unlikely to hamper treatment performance.

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Abbreviations: A, acidification; AA, acetic Acid; ABR, anaerobic baffled reactor; AF, anaerobic Filter; AnBEMR, anaerobic bio-entrapped membrane reactor; AnCMBR, anaerobic ceramic membrane bioreactor; AnIBPR, anaerobic immobilized bio-plates reactor; AnMBR, anaerobic membrane bioreactor; ASBR, anaerobic sequencing batch reactor; BA, butyric acid; BID, barrier ionization discharge; CH<sub>4</sub>, methane; COD, chemical oxygen demand; DO, dissolved oxygen; EGSB, expanded granular sludge bed; EPS, extracellular polymeric substance; FA, formic acid; GC, gas chromatograph; HRT, hydraulic retention time; M, methanogenesis; MLSS, mixed liquor suspended solids; NFDM, non-fat dry milk; OLR, organic loading rate; ORP, oxidation-reduction potential; PA, propionic acid; SAF-MBR, staged anaerobic fluidized membrane bioreactor; SCOD, soluble chemical oxygen demand; SMP, soluble microbial product; SRT, sludge retention time; SS, suspended solid; TCD, thermal conductivity detector; TCOD, total chemical oxygen demand; UASB, upflow anaerobic sludge blanket; VFA, volatile fatty acid.

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#### 1. Introduction

Anaerobic treatment of wastewaters offers advantages in low energy expenditure, low sludge volume, and methane generation relative to conventional aerobic activated sludge processes. However, issues such as slow growth rates of the anaerobic bacteria, washing out of biomass, and biochemical reaction kinetics varying with time and temperature have prevented the wide application of anaerobic treatment for low-strength wastewaters. High-rate anaerobic reactors, such as anaerobic filter (AF) [1], expanded granular sludge bed (EGSB) [2], upflow anaerobic sludge blanket (UASB) [3,4], and anaerobic baffled reactor (ABR) [5–9] that used packing materials, granular sludge, or multiple baffles have made

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possible biomass retention at relatively short HRTs while maintaining high treatment performance for wastewater. Recent developments of anaerobic membrane bioreactor (AnMBR) [10–15] and staged anaerobic fluidized membrane bioreactor (SAF-MBR) [16– 19] deployed membrane to significantly widen the separation of HRT from sludge retention time (SRT). The AnMBR could also be operated at short HRT with efficient solid-liquid separation to maintain high treatment performance for low-strength wastewaters, demonstrating a great potential of anaerobic treatment to improve environmental sustainability. However, washing out of biomass from reactors remains a critical issue for highperformance reactors AF, EGSB, UASB, and ABR, where a concomitant high biomass content contributes significantly to membrane fouling in the AnMBR and SAF-MBR.

In light of these issues, we developed the AnIBPR as an alternative to treat low-strength wastewaters. Our previous applications of immobilized biomass focused on aerobic treatment processes [20,21] and aerobic MBR [22-24]. Treatment evaluation of immobilized anaerobic process for low-strength wastewaters has been scarcely available in the literature. The objectives of the immobilized biomass approach to wastewater treatment have been to deploy high biomass concentration, reduce washout of biomass from the reactor, accelerate slow-growing bacteria to tackle complex organic compounds, and achieve simultaneous removal of carbon and nitrogen from a high load of dissolved organics in a single-pass process [20,21,24]. In recent years, we applied the immobilized anaerobic process in the anaerobic bio-entrapped membrane reactor (AnBEMR) packed with bio-ball carriers that treated high-strength pharmaceutical wastewater and found that lower suspended biomass resulted in lower extracellular polymeric substances (EPS) and soluble microbial products (SMP) in the AnBEMR, thus reducing membrane fouling and extending the membrane filtration time relative to conventional AnMBR [25]. Such coupling of aerobic immobilized biomass and membrane maintained longer SRT, produced little suspended solids in the effluent, enhanced contact of nutrients with biomass, and reduced EPS and SMP production [22–24]. However, the use of anaerobic immobilized process for treatment of low-strength wastewaters. as is the main focus of this work, has not been documented.

Energy efficiency of treatment processes in domestic wastewater treatment plants and associated carbon footprint offer ample opportunities for improvement [26]. While low-strength wastewaters were routinely treated at ambient temperatures ranging 10– 35 °C, anaerobic processes were commonly operated at mesophilic conditions of 30–35 °C at high energy costs [16,27,28]. In general, the growth rate of methanogens is slow below 20 °C due to sluggish biochemical reaction kinetics at low temperature. Maximum substrate utilization rates of microorganisms were significantly influenced by temperature as well as by the wastewater properties including viscosity, particle settling velocities, and diffusion of soluble compounds at various temperatures [29–34].

Recent research showed relatively high concentrations of dissolved methane in the effluent containing 28-75% of total methane production at 11-26 °C. Dissolved CH<sub>4</sub> was oversaturated during treatment of low-strength wastewaters [3,17,35-39]. The degree of saturation (as C/C\* where C and C\* are calculated and saturation concentrations, respectively) ranged from 1.9 to 6.9 based on COD mass balance calculation [36]. Yeo et al. [38] investigated the dissolved CH<sub>4</sub> in an AnMBR when treating a low-strength wastewater (glucose as carbon source) with organic loading rate (OLR) of 0.39 to 1.1 g COD/L-d at 24-26 °C and HRT of 1 d, and found the dissolved CH<sub>4</sub> to be 2.2-2.5 folds higher than saturation values according to Henry's law accounting for 76% of total CH<sub>4</sub> production at OLR of 0.39 g COD/L-d. They concluded that mass transfer limitation due to low gas production rate in the AnMBR being operated at low OLRs and low influent COD as responsible for oversaturation in the reactor. However, mechanisms for oversaturated dissolved  $CH_4$  in anaerobic reactors and explanations have not been available.

We deployed in this study immobilized anaerobic biomass in the form of bio-plates as readily installed baffles to provide high anaerobic biomass content but low suspended biomass content in the reactor and to achieve long SRT for treatment of lowstrength wastewaters. Treatment performance of the immobilized anaerobic system according to mesophilic and psychrophilic conditions (15–35 °C) were investigated. The production of dissolved CH<sub>4</sub> and VFA were investigated in order to better understand the new AnIBPR.

#### 2. Materials and methods

#### 2.1. Synthetic wastewater composition

A low-strength synthetic wastewater was freshly prepared every 2 d with non-fat dry milk (NFDM) and sodium acetate as carbon source according to Table 1 and diluted to 400 mg COD/L with tap water for use. NFDM comprised of carbohydrate (mainly lactose, 54.1%), protein (33.4%), fat (0.8%), Na (0.39%), Ca (1.1%), P (0.95%), Mg (0.11%), Zn (0.0033%), K (1.76%), and trace elements (Fonterra, New Zealand). The synthetic wastewater contained sodium bicarbonate buffer to maintain pH at 6.8–7.1 and minerals for nitrogen and micronutrient sources.

#### 2.2. Reactor setup and operation

An AnIBPR was installed with hanging and standing baffles dividing the reactor into 7 compartments (14 slabs of immobilized bio-plates) with a total working volume of 14.5 L (Fig. 1). The baffles forced the wastewater to flow under the hanging and over the standing baffles from the inlet to the outlet; under the hanging baffles there were slanted (30°) flow-routing pieces to enhance contact of the substrate with biomass. The effluent line contained a glass U-tube for level control and collection of biogas. The immobilized bio-plates were prepared per Yang et al. [20], Wang et al. [40], Wang et al. [41], Liu et al. [42], Chao et al. [43] and Chao et al. [44], in which a dewatered (20% w/v) anaerobic sludge from a UASB treating food and beverage wastewater was formed into an immobilized bio-plate of 30 cm, 20 cm, and 0.8 cm, in length, width, and thickness, respectively. The reactor contained an equivalent mixed liquor suspended solids (MLSS) of 58 g/L at a packing ratio 27.5% (i.e. total bio-plate volume/total reactor volume), respectively, and operated at the SRT of 260-570 d per Qian et al. [45]. The wastewater was continuously fed into the AnIBPR by use of a peristaltic pump (Masterflex, L/S, USA) to achieve HRT of 16 h and OLR of 0.6 g/L-d.

During initial start-up, the reactor was fed with synthetic wastewater of  $425 \pm 7 \text{ mg COD/L}$  (including NFDM: 91.8 mg/L as 100 mg COD/L; sodium acetate: 439 mg/L as 300 mg COD/L) at

Table 1		
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Compositions of synthetic wastewater fed at different stages.

Component	Stage I Concentration (mg/L)	Stages II, III, IV Concentration (mg/L)
Non-fat dry milk (NFDM)	184	275
Sodium acetate (CH <sub>3</sub> COONa)	299	149
NH <sub>4</sub> Cl	108	108
KH <sub>2</sub> PO <sub>4</sub>	28.1	28.1
NaHCO <sub>3</sub>	600	600
CaCl <sub>2</sub>	14.6	14.6
FeCl <sub>3</sub>	13.5	13.5
MgCl <sub>2</sub> ·6H <sub>2</sub> O	5.00	5.00

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