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## Treatment of industrial wastewaters by microalgal bacterial flocs in sequencing batch reactors



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## HIGHLIGHTS

• Industrial wastewaters were screened for treatment in MaB-floc SBRs.

- Settling MaB-flocs were developed by bio-flocculation of microalgae.
- The nutrient removal rates and effluent qualities were wastewater dependent.

• MaB-flocs were dewatered by filter press with 79–99% recovery to 12–21% DM.

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## ABSTRACT

Microalgal bacterial flocs in sequencing batch reactors (MaB-floc SBRs) represent a novel approach to wastewater treatment. In this approach, mechanical aeration is replaced by photosynthetic aeration and MaB-floc settling separates the treated wastewater from the produced biomass. However, its technical potential for industrial wastewaters needs to be shown. Therefore, wastewaters of aquaculture, manure treatment, food-processing and chemical industry were treated in MaB-floc SBRs. This treatment resulted in significantly different nutrient removal rates and effluent qualities among wastewaters. A high MaB-floc production was obtained for all wastewaters, ranging from 0.14 to 0.26 g total suspended solids  $L_{reactor}^{-1}$  day<sup>-1</sup>. A major advantage of MaB-floc SBRs is the harvesting via a filter press with a large pore size of 200 µm, resulting in MaB-floc recoveries of 79–99% and cakes containing 12–21% dry matter. These results may contribute to evolving MaB-floc SBRs as a valuable remediation strategy, especially for aquaculture and food-processing wastewaters.

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### 1. Introduction

During the past decade, there has been a true renaissance of interest in dual-purpose microalgae technology coupling wastewa-

ter treatment via photosynthetic aeration and nutrient scavenging with the production of microalgal biomass (Olguín et al., 2012; Van der Ha et al., 2012; Su et al., 2012a; Park et al., 2011b; Van Den Hende et al., 2011a; Muñoz and Guieysse, 2006; Gutzeit, 2006). The produced microalgal biomass can be a bioresource for the production of valuable products, for example, for biogas production (Zamalloa et al., 2012), feed ingredients (Natrah et al., 2013) and fine chemicals (Olguín et al., 2012; Van der Ha et al., 2012). Although potentially beneficial, however, dual-purpose microalgal wastewater treatment systems are rarely used in an industrial scale. A major challenge is the separation of the microalgal biomass from the treated wastewater (Udom et al., 2013; Uduman et al., 2010). Indeed, microalgae harvesting may represent 20-60% of the total microalgae production costs (Richmond, 2004; Molina Grima et al., 2003). Bio-flocculation of microalgae and bacteria into microalgal bacterial flocs is a novel concept which addresses this harvesting challenge. Indeed, MaB-flocs settle by gravity without



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the addition of flocculants and this enables the discharge of biomass-free effluent (Su et al., 2012a; Van Den Hende et al., 2011a; Medina and Neis, 2007; Gutzeit, 2006). Moreover, in MaB-floc systems the microalgae retention time can differ from the hydraulic retention time (HRT) of the wastewater, or -in algae culture terminology-, the microalgae growth rate and culture dilution rate can be uncoupled (Van Den Hende et al., 2011a; Medina and Neis, 2007; Gutzeit, 2006). In this way, low-strength wastewaters can also be treated by high microalgae biomass densities. This is a large advantage compared to wastewater treatment systems with suspended microalgae.

To date, published research studies on the *in situ* bio-flocculation of microalgae for wastewater treatment in (semi-)continuous reactors are still scarce. Indeed, the reported research is limited to the treatment of a few wastewaters, specifically sewage (Su et al., 2012b: Park et al. 2011a: Gutzeit, 2006) and paper mill wastewater (Weinberger et al., 2012) in continuous stirred reactors with settling tank; and synthetic wastewaters (Van Den Hende et al., 2011b), sewage (Van Den Hende et al., 2011a) and paper mill effluent (Van Den Hende et al., 2012b) in sequencing batch reactors (SBRs). Industrial wastewaters largely differ in their chemical composition, turbidity and colour (Tchobanoglous et al., 2003). Moreover, an unbalanced C:N:P ratio of wastewater and the presence of colour compounds and suspended solids in wastewater have been shown to limit wastewater treatment by microalgae (Depraetere et al., 2013; Markou and Georgakakis, 2011). The question remains for which wastewaters this MaB-floc concept has potential, that is settling MaB-flocs can be developed and maintained, wastewater can be treated efficiently to discharge norms and the produced biomass can be easily harvested. Therefore, more industrial wastewaters need to be screened for treatment in MaB-floc reactors.

As an important first step towards their industrial implementation, this study further explores the technical potential of MaB-floc SBRs to treat wastewaters from aquaculture, manure treatment, food-processing and chemical industry and to simultaneously produce biomass. Batch experiments are set up to develop MaB-inoculum and as a first screening of different wastewaters from each industry to select one wastewater per industry for treatment in MaB-floc SBRs. To assess the potential of MaB-floc SBRs for wastewater treatment, their nutrient (C, N, P) removal and effluent quality (pH, C, N, P) are comparatively evaluated. Moreover, MaB-floc properties and biomass production are examined. Aiming at lowering the costs to harvest the produced biomass, the potential for MaB-floc dewatering with a filter press with large pores (200  $\mu$ m) is examined.

### 2. Methods

#### 2.1. Wastewaters

Several wastewaters from four industrial sites were collected (Fig. 1; Table 1). Aquaculture wastewaters ( $A_{quar}$ ,  $A_{small}$  and  $A_{outgrow}$ ) were drum filter effluents from pikeperch cultures (Aquaculture Practice Centre of Inagro, Roeselare, Belgium).  $A_{quar}$  originated from quarantine pikeperch cultures,  $A_{small}$  originated from cultures of pikeperches smaller than 500 g and  $A_{outgrow}$  originated from outgrowth trails with pikeperches larger than 500 g. Manure treatment wastewaters ( $M_{pond1}$  and  $M_{pond2}$ ) were effluents collected from buffer ponds (Innova Manure, Gistel, Belgium). Food processing wastewater ( $F_{UASB}$  and  $F_{CAS}$ ) was upflow anaerobic sludge blanket reactor (UASB) effluent and conventional activated sludge (CAS) effluent from a soy-processing company (Alpro, Wevelgem, Belgium). Chemical production wastewaters were mixtures of influent and effluent from a CAS

reactor (BASF, Antwerp, Belgium). Influent:effluent mixtures (v:v%) of 100:0, 25:75 and 10:90 were used in the batch reactors and 50:50 was used in SBR ( $C_{-1001:0E}$ ,  $C_{-251:75E}$ ,  $C_{-101:90E}$  and  $C_{-501:50E}$ , respectively). Prior to feeding, all wastewaters were sieved through a 1–3 mm sieve to avoid tube clogging and stored at 4 °C.

### 2.2. MaB-floc preculture in batch reactors

MaB-flocs were precultured in batch reactors in two steps. For each wastewater type, 800 mL wastewater, 300 mL of a consortium of microalgae/cyanobacteria collected on industrial sites (<0.400 volatile suspended solids (VSS) L<sup>-1</sup> and 80 mL of MaB-flocs from previous cultures (0.090 g VSS  $L^{-1}$ ) were mixed. For each wastewater type, two 500 mL Erlenmever flasks were filled with the algae/ wastewater mixes and operated in batches with a 17 h light period with cycles of 0.5 h not stirring and 0.5 h stirring (210 rpm: Heidolph, UK), followed by a 7 h dark period without stirring. After 2.5 days, the reactor liquor was transferred to 5 L Erlenmeyers flasks and wastewater was added to obtain a working volume of 4 L. Over 6–7 days, a 14 h light:10 h dark cycle was applied with continuous stirring during the light phase and no stirring during the dark phase. Fluorescent lamps (36/840, Philips, Belgium) provided a photosynthetic active photon flux density (PPFD) of around 100  $\mu$ mol photons m<sup>-2</sup> s<sup>-1</sup> at the water surface in the Erlenmeyer flask. Dissolved oxygen (DO), pH and temperature (T) were measured at the start and at the end of each light cycle. The A664b/ A665a ratio of MaB-flocs was determined daily. The diluted sludge volume index (dSVI) was determined at the end of each batch.

#### 2.3. Sequencing batch reactors

Wastewaters  $A_{small}$ ,  $M_{pond1}$ ,  $F_{UASB}$  and  $C_{501:50E}$  were treated in SBRs, referred to as SBR<sub>aquaculture</sub>, SBR<sub>manure</sub>, SBR<sub>food</sub> and SBR<sub>chemical</sub>, respectively. SBRs were inoculated with 4 L reactor liquor of the batch reactors. SBRs were photobioreactors of 5 L with a working volume of 4 L, as previous described (Van Den Hende et al., 2011a). The DO (Hanna Instruments, Belgium), pH and T (Jumo, Belgium) were logged every 30 s (only in SBRaguaculture and SBR<sub>manure</sub>). Illumination by one halogen lamp (500 W, Silon CE-82-Y, Hong Kong) for each SBR provided an average PPFD at the inner reactor wall (in  $\mu$ mol photons m<sup>-2</sup> s<sup>-1</sup>) of 152 in SBR<sub>aquaculture</sub> and SBR<sub>manure</sub>, 171 in SBR<sub>food</sub> and 174 in SBR<sub>chemical</sub>. Average reactor temperature in the SBRs was  $31.2 \pm 4.0$  °C. Each SBR was equipped by a diaphragm pump for influent feeding (Blackstone, USA), a peristaltic pump for effluent withdrawal (Watson Marlow, USA) and an overhead stirrer (210 rpm; Heidolph RZR 2020, Germany). Influents were stored at 4 °C and pumped into each SBR while being magnetically stirred (150-200 rpm; Heidolph, UK).

Two operation SBR modes were applied. The first consisted of a 7.75 h stirring phase in the light, 12 h settling phase in the dark, 3 h stirring phase in the light, 0.5 h settling phase in the dark, 0.25 h effluent withdrawal in the dark and 0.5 h influent feeding in the light while reactor stirring was carried out. The 3 h light phase before effluent withdrawal and influent feeding was introduced to aim for aerobic conditions while influent feeding. However, this resulted in gas bubbles containing floating MaB-flocs, especially in SBR<sub>aquaculture</sub>. Therefore, after the first 14 days for SBR<sub>aquaculture</sub> and SBR<sub>manure</sub>, and after 10 days for SBR<sub>food</sub> and SBR<sub>chemical</sub>, a second SBR modus was applied consisting of a 11.5 h stirring phase in the light, a 8.75 h phase in the dark with 0.25 h stirring every 1-1.25 h, a 3 h settling phase in the dark, 0.25 h effluent withdrawal in the dark, and 0.5 h influent feeding in the light while reactor stirring was carried out. During the first 7 days in SBR<sub>food</sub>, the HRT was 4 days to adapt to high TCOD concentrations, and

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