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Performance of flocs and biofilms in integrated fixed-film activated sludge (IFAS) systems for the treatment of oil sands process-affected water (OSPW)

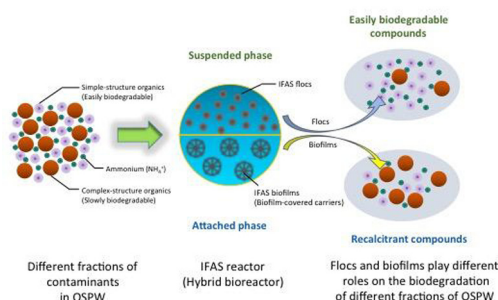
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HIGHLIGHTS

- IFAS-flocs and IFAS-biofilms were compared for the treatment of OSPW.
- IFAS-biofilms showed high AEF removal, IFAS-flocs showed high COD and $\text{NH}_4\text{-N}$ removal.
- Biodegradation was principal removal mechanism for the OSPW remediation in the IFAS systems.
- Nitrifiers and denitrifiers genes were more abundant in flocs than biofilms in IFAS systems.

GRAPHICAL ABSTRACT



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ABSTRACT

This study evaluated the roles of suspended flocs and attached biofilms from integrated fixed-film activated sludge (IFAS) systems in their overall contribution toward organic compound removal in oil sands process-affected water (OSPW). The batch experiments were operated with suspended flocs and biofilm carriers from our continuous laboratory-scale IFAS reactors for the OSPW treatment. A distinct difference between IFAS flocs and biofilms for the chemical oxygen demand (COD), ammonium and acid extractable fraction (AEF) removal was demonstrated. Compared to the biofilms, the flocs demonstrated considerably higher removal rates for COD and ammonium, whereas, biofilms had better performance on the AEF removal than flocs. Meanwhile, the results also revealed that the biodegradation was the principal removal mechanism whereas, the biosorption contributed little to the OSPW organic compounds and the ammonium removals in the IFAS system. Microbial analysis from *q*-PCR revealed that the abundances of nitrifiers and denitrifiers genes were significantly higher in flocs than in biofilms in both raw and ozonated-OSPW IFAS reactors. The microbial communities analysis from *MiSeq* sequencing showed that *Proteobacteria*, *Acidobacteria*, *Nitrospirae* and *Bacteroidetes* were dominant phyla in both flocs and biofilms.

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1. Introduction

The development and expand of the Athabasca oil sands industry in Northern Alberta, Canada, was rapid in the past few decades

[1]. The Athabasca oil sands in Northern Alberta is one of the world's largest oil reserves, containing over 168 billion barrels of recoverable bitumen [2]. The mined bitumen is separated from associated sands and clays using the caustic hot water extraction process [3], resulting in significant volume of oil sands process-affected water (OSPW), which is current stored in tailing ponds following a “zero discharge approach” maintained by the Alberta's regulatory framework [4]. Oil sands tailings contain a mixture of

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sand, clay fines, slits, dissolved salts, heavy metals and organic compounds, which have demonstrated toxicity to a number of aquatic organisms such as algae [5], fish [6], benthic invertebrates [7] and mammalian species [8]. The primary toxic constituents of OSPW are a group of organics, collectively known as naphthenic acids (NAs) [9]. Reported concentrations of NAs in OSPW are in the range of 20–120 mg/L [10]. Appropriate OSPW treatment methods that allow maximum reuse or safe discharge of treated OSPW will decrease the amount of freshwater withdrawn from the Athabasca River and reduce the potential environmental threat of the OSPW.

Biodegradation utilizing bioreactor technology is an economical, energy-efficient and environmentally sound approach for the OSPW reclamation [11]. Microbial aggregates are employed in biological wastewater treatment processes to remove organic compounds and nutrients (nitrogen and phosphorus). Microbial aggregates are complex ecosystems consisting of highly stratified microbial communities embedded in a matrix of extracellular polymeric substances (EPS). The two types of aggregates are suspended flocs (e.g., in the activated sludge process) and attached biofilms. Promise has been shown from both suspended and attached growth processes for their application on OSPW treatment [1,3,12–16].

Suspended and attached biomass differs in many aspects in bioreactors. Attached growth systems (i.e., biofilms) maintain a high microbial retention time, eliminating microbial wash out and encouraging the growth of slow growing microbes such as nitrifying bacteria and recalcitrant organics degraders. The improved endogenous metabolisms of the biofilm biomass under longer sludge retention time (SRT) allow the development of microbial communities, which are efficient for the biodegradation of poorly degradable contaminants [17]. Suspended growth systems, on the other hand, are characterized by greater substrate uptake rate and specific microbial growth rate as compared to attached growth systems. Such suspended growth systems are suitable for the easily biodegradable organic substrates removal. The difference of microbial population and hydrodynamic conditions in suspended and attached growth bioreactors may also lead to a significant difference in the biomass physical structure (e.g., biomass surface areas and density) and in the physicochemical characteristics (e.g., hydrophobicity and surface charge) between suspended and attached biomass. Under identical substrate loading condition, the biofilms structure tends to be smooth, dense and less porous, while the flocs have highly porous structure [18]. This observation is often attributed to the faster suspended biomass growth rate and higher EPS production rates as compared to the attached biomass [19]. Previous studies also showed that suspended biomass has greater negativity and higher hydrophobicity than attached biomass [20]. The difference of the physical structure and the surface properties between attached and suspended biomass may thus result in difference in substrate and oxygen diffusion rates in these structures.

Our recent study applied integrated fixed film activated sludge (IFAS) reactors (i.e., combined suspended and attached growth systems) for OSPW treatment. This study showed that ozonation combined with IFAS is a promising approach for OSPW treatment [16]. The coexistence of suspended and attached biomass leads to the degradation of a wide range of contaminants. In IFAS systems, biofilm detachment occurs naturally, and can enable attached biomass to spread and get attached to the flocs, thus enriching flocs with slow growing biomass [21]. Suspended and attached biomass may also compete for available organic substrates and oxygen [22]. Moreover, bioreactor performance is determined by the diversity and density of the bacterial population present in activated sludge or biofilm. Therefore, the microbial diversity information and microbial biomass functionality in suspended and attached phases

are necessary for the further optimization of the bioreactor design and operation.

Our current study aims at comparing and elucidating the performance and mechanisms of suspended and attached biomass in IFAS systems for the OSPW treatment. Batch IFAS reactors were operated using flocs and biofilms, taken from stabilized flow-through IFAS systems, for both raw and ozonated OSPW treatment. The roles of the two main removal mechanisms (biodegradation and biosorption) on OSPW remediation were evaluated. Microbial community structures in suspended and attached biomass were also characterized. The results obtained from this study could benefit the modelling and optimization of IFAS systems.

2. Materials and methods

2.1. Source water information

The fresh OSPW was collected from oil sands tailing ponds in Fort McMurray, AB, Canada, in September 2013. OSPW samples were preserved in 200 L polyvinyl chloride barrels in a cold room (4 °C) and were well mixed prior to use.

The 0.1% (v/v) trace nutrient solution contained: $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$, 3 g L⁻¹; CaCl_2 , 1.5 g L⁻¹; $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, 0.28 g L⁻¹; $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, 0.13 g L⁻¹; $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$, 0.12 g L⁻¹; $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, 0.0074 g L⁻¹. The pH was 7.3 ± 0.3 . All chemicals and supplies were obtained from Thermo Fisher Scientific.

2.2. Batch IFAS reactor set-up and operation

2.2.1. Bench scale IFAS reactors

Two IFAS systems (raw and ozonated OSPW IFAS) were employed in our previous study [16], one with raw OSPW as influent and the second with ozonated OSPW (30 mg/L ozone dose) as influent. A 60% volume fraction of polyethylene (PE) carriers (Bioflow 9, Rauschert, Steinwiessen, Germany) with specific biofilm growth areas of 800 m²/m³ were employed in the IFAS reactors.

2.2.2. Batch reactor (biodegradation and biosorption) set-up and operation

After the two acclimated IFAS systems were stabilized, the batch reactors were performed using suspended activated sludge flocs and attached biofilm carriers from two continuous IFAS. To determine the biomass concentrations in the IFAS systems, mixed liquor suspended solids (MLSS) was used to quantify the suspended and attached biomass in the IFAS system. The detailed information of IFAS operational conditions is shown in Table 1. Three types of biomass inoculants were evaluated and compared in the batch tests, including: (1) suspended activated sludge (IFAS-flocs), (2) attached biofilm carriers (IFAS-biofilms), and (3) suspended activated sludge and attached biofilm carriers (IFAS-hybrid).

Suspended sludge with the concentration of ~1000 mg MLSS/L, and/or colonized biofilm carriers (volume fraction is 60%) (The same with that in continuous IFAS reactors) were applied. Amber bottles containing duplicates of individual treatments were applied. After raw or ozonated OSPW with nutrients and biomass were introduced, the bottles were shaken and sampling was done immediately to obtain initial pollutant concentrations at $t = 0$ h. Batch reactors were operated on a platform shaker at 150 rpm (Innova™ 2100, Platform Shaker, New Brunswick Scientific, USA) for 21 days at room temperature (20 ± 1 °C). During the entire batch reactors operation, external carbon source (sodium acetate, 300 COD mg/L) as well as nitrogen (NH_4Cl , 30.0 ± 3.0 mg N/L), phosphorus (KH_2PO_4 , 3.0 ± 0.2 mg P/L) and trace metals (mentioned in Section 2.1) were provided for the aggregated biomass

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