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# Sludge reduction and microbial community structure in an anaerobic/ anoxic/oxic process coupled with potassium ferrate disintegration



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## ABSTRACT

An anaerobic/anoxic/oxic (AAO) wastewater treatment system combining with a potassium ferrate  $(K_2FeO_4)$ oxidation side-stream reactor (SSR) was proposed for sludge reduction. Batch experiments showed that optimal  $K_2FeO_4$  dosage and reaction time for sludge disintegration was 100 mg/g suspended solids (SS) and 24 h, respectively. Subsequently, an AAO-SSR and a conventional AAO were operated in parallel to investigate effects of K2FeO4 oxidation on process performance, sludge characteristics and microbial community structures. The AAO-SSR process operated under the optimized condition achieved efficient COD and NH<sub>4</sub><sup>+</sup>-N removal, and reduced sludge by 47.5% with observed yield coefficient of 0.21 g SS/g COD.  $K_2FeO_4$  addition broke sludge particles, increased dissolved organic matters in the mixed liquor, and improved sludge dewaterability. Illumina-MiSeq sequencing results showed that  $K_2FeO_4$  oxidation in the AAO-SSR decreased microbial richness and diversity, enriched slow growers (Dechloromonas), anaerobic fermentative bacteria (Azospira) and Fe(III)-reducing bacteria (Ferribacterium), but limited the growth of phosphate-accumulating organisms.

### 1. Introduction

Activated sludge (AS) process is extensively applied to both municipal and industrial wastewater treatment by transferring particulate and soluble organic and nutrient matters into bacterial mass (active and inactive) and gaseous substances ( $CO<sub>2</sub>$ ,  $N<sub>2</sub>$ ,  $CH<sub>4</sub>$ , etc) [\(Mohammadi](#page--1-0) [et al., 2011\)](#page--1-0). With an observed sludge yield coefficient ( $Y_{\text{obs}}$ ) of 0.5 g biomass/g chemical oxygen demand (COD) removed [\(Tchobanoglous](#page--1-1) [et al., 2003](#page--1-1)), the large amount of waste activated sludge (WAS) generated imposed a heavy burden on wastewater treatment plants (WWTP), both operationally and economically. The increased stringent restrictions in disposal and requirements in reuse have made sludge treatment more technically challenging and costly ([Niu et al., 2016a;](#page--1-2) [Yang et al., 2015](#page--1-2)). Considering the environmental and economic burden, a profound research effort on exploiting and developing new technologies for WAS minimization within the AS process is urgently needed in wastewater treatment field.

Recently, many biological, chemical and physical approaches have been developed to minimize WAS production via sludge hydrolysis and/or disintegration. Among these technologies, chemical oxidation technologies by using oxidant, such as ozone [\(Qiang et al., 2015; Yang](#page--1-3) [et al., 2015](#page--1-3)), chlorine dioxide [\(Liu, 2016\)](#page--1-4), potassium ferrate ([Wu et al.,](#page--1-5) [2015; Ye et al., 2012\)](#page--1-5) and sulfate radicals ([Niu et al., 2016b](#page--1-6)), have been proved to be an efficient way for WAS disintegration. As an environmental-friendly strong oxidant, potassium ferrate  $(K_2FeO_4)$  can effectively disintegrate sludge, improve sludge dewaterability and enhance sludge biodegradability ([Wu et al., 2015; Ye et al., 2012; Zhang et al.,](#page--1-5) [2012\)](#page--1-5). [Ye et al. \(2012\)](#page--1-7) reported that  $K_2FeO_4$  oxidation was effective for WAS disintegration with the ratio of soluble COD (SCOD) to total COD of sludge reached 0.32 at dosage of 0.81 g/g SS. [Zhang et al. \(2012\)](#page--1-8) found that a  $K_2FeO_4$  dosage of 0.085 g/g SS at pH 3 was an ideal condition with maximum sludge dewaterability characteristics and sludge disintegration. [Wu et al. \(2015\)](#page--1-5) obtained an optimal  $K_2FeO<sub>4</sub>$ dosage of 0.052 g/g SS with 69% sludge disintegration, 85% reduction

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Abbreviations: AAO, anaerobic/anoxic/oxic; ANOVA, analysis of variance; AOB, ammonia oxidizing bacteria; AS, activated sludge; COD, chemical oxygen demand; CST, capillary suction time; DO, dissolved oxygen; DOM, dissolved organic matte; EEM, excitation-emission matrix; EPS, extracellular polymeric substances; Ex/Em, excitation/emission wavelength;  $F_{\rm max}$ , maximum fluorescent intensity; HRT, hydraulic retention time; IC, inorganic carbon; K<sub>2</sub>FeO<sub>4</sub>, potassium ferrate; MLSS, mixed liquid SS; MLVSS, mixed liquid VSS; NH<sub>4</sub><sup>+</sup>-N<sub>2</sub> ammonium nitrogen; NO<sub>3</sub><sup>-</sup>-N, nitrate nitrogen; NOB, nitrite oxidizing bacteria; OSA, oxic/settling/anaerobic; OTUs, operational taxonomic units; PAOs, phosphate-accumulating organisms; PARAFAC, parallel factor; RAS, returned activated sludge; SAUR, specific ammonium uptake rate; SBR, sequencing batch reactor; SCOD, soluble COD; SOUR, specific oxygen uptake rate; SRE, sludge reduction efficiency; SRT, solid retention time; SS, suspended solid; SSR, side-stream reactor; TC, total carbon; TN, total nitrogen; TOC, total organic carbon; TP, total phosphorus; VSS, volatile SS; WAS, waste activated sludge; WWTP, wastewater treatment plant; Y<sub>obs</sub>, observed sludge yield coefficient

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of dewaterability and 44% enhancement of cumulative biogas production for anaerobic biodegradation. Those intensive efforts mentioned above are very helpful to understand effects of  $K_2FeO_4$  oxidation on sludge disintegration and dewaterability improvement, but most of the previous studies were batch tests. More detailed research is needed to coupling  $K_2FeO_4$  oxidation with bioreactors for long-term operation to better understand its effects on pollutants removal, microbial community structure and sludge characteristics.

Side-stream reactor (SSR) for sludge reduction inserting in the returned activated sludge (RAS) line of bioreactors is considered as the most promising way potentially employed in WWTPs [\(Cheng et al.,](#page--1-9) [2017; Khursheed & Kazmi, 2011; Yang et al., 2015](#page--1-9)). Maintaining anaerobic conditions in the SSR was the most commonly used strategy for sludge reduction via sludge decay and uncoupling mechanism through the shift between aerobic and anaerobic conditions ([Khursheed & Kazmi, 2011; Semblante et al., 2016a,b; Zhou et al.,](#page--1-10) [2015\)](#page--1-10). The SSR process based on cell lysis – cryptic growth was also realized by combining ozone, chlorine dioxide, ultrasonic treatment, ozonation/ultrasound with various existing bioreactors at lab and pilot scales, and has already yielded good sludge reduction performance [\(Liu,](#page--1-4) [2016; Mohammadi et al., 2011; Qiang et al., 2015; Yang et al., 2015](#page--1-4)). Compared to above chemical and physical measures,  $K_2FeO_4$  oxidation produces iron salt, which is an excellent flocculant for the improvement of sludge settleability and dewaterability, and probably has a significant influence on microbial community composition. To our knowledge, effects of high valence salt with strong oxidability, such as  $K_2FeO_4$ , KMnO<sub>4</sub> and KHSO<sub>5</sub>, on process performance and microbial community structures of bioreactors were scarcely reported. Moreover, K2FeO4 oxidation enhanced sludge biodegradability ([Wu et al., 2015](#page--1-5)), and thus a subsequent hydrolysis after sludge disintegration would further improve sludge reduction. In order to construct a bridge between lab-scale investigations and full-scale applications, comprehensive studies on process performance of combining  $K_2FeO_4$  oxidation pretreatment with bioreactors are necessary.

In this study, an anaerobic/anoxic/oxic (AAO) system combining with a  $K_2FeO_4$  oxidation SSR (AAO-SSR) was established for sludge reduction. The optimal parameters for  $K_2FeO_4$  oxidation and subsequent hydrolysis were determined using batch tests. Then pilot-scale AAO-SSR and AAO systems were operated in parallel to investigate effects of  $K_2FeO_4$  oxidation on pollutants removal, sludge characteristics and microbial community structures. The migration and transformation of dissolved organic matters (DOM) were also analyzed to expound mechanisms of secondary substrate release. Illumina-MiSeq sequencing was applied to compare microbial community structures in AAO-SSR and AAO.

#### 2. Materials and methods

#### 2.1. Batch tests for  $K_2FeO_4$  oxidation and hydrolysis

The WAS used in this study was collected from RAS line of Bailonggang WWTP (Shanghai, China) with wastewater flow rate of  $2,000,000$  m $^{3}/d$  treated by AAO process. The main characteristics of WAS samples were as follows: suspended solid (SS) of 4.8–6.5 g/L, volatile SS (VSS) of 2.9–4.0 g/L, SCOD of 41.6–104.2 mg/L, filtrate total nitrogen (TN) of 4.75–23.08 mg/L and pH of 7.17–7.49.

Eight dosages (0, 6.5, 12.5, 25, 50, 75, 100 and 125 mg/g SS) of K2FeO4 (Aladdin Chemicals Co., USA) were used to investigate effects of  $K_2FeO_4$  dosages on sludge disintegration. The batch tests were conducted in a lab-scale reactor of 1 L equipped with thermostat and magnetic stirrer. The preseted dosage of  $K_2FeO_4$  was added into the WAS, and then the sludge was continuously mixed at 200 rpm for 2 h. All the experiments were carried out at 20.0  $\pm$  0.1 °C. A 50 mL sample was withdrawn and analyzed in duplicate after filtration by 0.45 μm vinyl cellulose membrane.

The hydrolysis tests subsequent to  $K_2FeO_4$  oxidation were carried

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Fig. 1. Schematic diagram of the AAO-SSR sludge reduction process.

out under the optimal dosage chosen from above batch tests. The sludge was firstly mixed with  $K_2FeO_4$  at 200 rpm, and then continuously stirred for 48 h at 20.0  $\pm$  0.1 °C. A 50 mL sample was withdrawn at intervals of 10 min each in the first 1 h, and then at 2, 4, 8, 12, 18, 24, 28, 32, 40 and 48 h. The samples were filtered through 0.45 μm membrane to analyze variations of DOM and nitrogen during  $K_2FeO<sub>4</sub>$ oxidation and hydrolysis process. A control test by using 2 L untreated WAS was also conducted in parallel.

#### 2.2. Experimental system and operating conditions

Two pilot-scale bioreactors, AAO and AAO-SSR processes (as shown in [Fig. 1\)](#page-1-0), were both employed to treat influent of the Bailonggang WWTP for 85 days. In the AAO system, the effective volume of anaerobic, anoxic and oxic tank was 12.0, 18.75 and 58.5 L, with hydraulic retention time (HRT) of 1.67, 2.60 and 8.13 h, respectively. Three peristaltic pumps were employed to continuously feed the influent, recycle the AS from the settler to the anaerobic tank and fulfill the mixed liquor recirculation, respectively. The RAS rate of AAO was controlled at 100% of the influent flow rate, and the mixed liquor recirculation ratio was maintained at 200% for denitrification. Agitation facility was set in the anaerobic and anoxic tanks to guarantee mass transferring requirements for reaction and suspension of AS. The average dissolved oxygen (DO) concentration in the oxic tank was controlled at 4.5 mg/L. The solid retention time (SRT) was remained at 18 d by discharging 5 L WAS from oxic tank every day. The temperature was  $15 \pm 2^{\circ}$ C during the operation time.

In the AAO-SSR process, the HRT, recirculation rate, DO and SRT were maintained the same as the AAO. An SSR module with effective volume of 10 L was inserted for sludge reduction by dosing  $K_2FeO_4$  at dosage of 100 mg/g SS obtained by batch tests. The SSR was divided into two equal compartments by one internal baffle. In the upper compartment of the SSR, 5 L WAS discharged from the oxic tank was added and reacted for 24 h, then flowed into the lower compartment and continuously pumped into the anaerobic tank in another 24 h. The emptied upper compartment was filled by WAS again and repeated the cycle.

#### 2.3. Analytical methods

#### 2.3.1. Preparation of samples for DOM analysis

The mixed liquor samples were collected from the oxic tank of the AAO-SSR and AAO processes at day 85 for DOM analysis. 30 mL collected samples were centrifuged at 12,000 rpm for 5 min, and then the Download English Version:

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