Chemical Engineering Journal 330 (2017) 63–70

Contents lists available at ScienceDirect

Chemical Engineering Journal

journal homepage: www.elsevier.com/locate/cej

Performance and microbial community analysis of a microaerophilic sulfate and nitrate co-reduction system

Xi-Jun Xu[a], Chuan Chen[a,*], Xun Guan[e], Ye Yuan[d], Ai-Jie Wang[a], Duu-Jong Lee[a,b,c], Zi-Feng Zhang[a], Jian Zhang[f], Ying-Juan Zhong[f], Nan-Qi Ren[a,*]

[a] State Key Laboratory of Urban Water Resource and Environment, School of the Environment, Harbin Institute of Technology, Harbin, HeiLongjiang Province 150090, China
[b] Department of Chemical Engineering, National Taiwan University, Taipei 106, Taiwan
[c] Department of Chemical Engineering, National Taiwan University of Science and Technology, Taipei 106, Taiwan
[d] School of Environmental Science and Engineering, Yancheng Institute of Technology, Yancheng 224051, China
[e] Northeast Agriculture University, 59 Wood street, Xiangfang District, HeiLongjiang Province 150030, China
[f] Shenzhen Greenster Environmental Technology Co., Ltd., 142 Zhuguangbei Road, Taoyuan Street, Nanshan District, Shenzhen, Guangdong Province 518055, China

ARTICLE INFO

Keywords:
Bacterial community structure
Sulfide oxidation
Microaerophilic
Sulfate and nitrate co-reduction

ABSTRACT

The succession of complex internal sulfur cycles and sulfide-oxidizing bacteria community has been observed during wastewater treatment under microaerophilic conditions or denitrifying conditions. However, research on the microbial community involved in sulfur cycles under microaerophilic denitrifying conditions is scarce. In this study we characterized the dominant bacteria and microbial community structure stimulated by microaerophilic conditions in a sulfate and nitrate co-reduction system. Full denitrification was accomplished and the sulfate removal efficiency ranged from 79.93% to 96.81% for all the tested scenarios, with the degree of sulfate reduction slightly decreased with higher O_2 feeding rate. The proportion of S^0 to influent SO_4^{2-} was much greater at microaerophilic stages (27.5–69.2%) versus the anaerobic stage (11.1%). The peak S^0 recovery (69.2%) was achieved at O_2 = 4.0 mL/min. Illumina sequencing technology was used to characterize the bacterial community and the results indicated that *Bacteroidetes*, *Firmicutes*, *Proteobacteria*, *Spirochaetae* and *Synergistetes* members were dominant in microbial communities, however the variation of these dominant members across all the operating conditions did not well respond to reactor performance. Further analysis revealed that the structure of the microbial community, including community richness and evenness, might better respond to reactor performance, which deserves more research in future.

1. Introduction

Sulfide (S^{2-}), metabolite of biological sulfate reduction by sulfate reducing bacteria (SRB), is a corrosive, odorous and toxic substance. Ingestion of sulfide imposes a health risk for workers in oil refinery and wastewater treatment plant. Also sulfide could induce pipeline leaks and thus it is environmental and economically costly in diverse industrial ecosystems. Therefore, it's essential to develop innovative biotechnology to prevent sulfide emission and maintain people health and biodiversity of fragile ecosystems [1]. Mediated by the low dissolved oxygen (DO) concentration in the wastewater, a sulfur cycle, including sulfate reduction to sulfide ($SO_4^{2-} \rightarrow S^{2-}$) and subsequent sulfide oxidation to elemental sulfur ($S^{2-} \rightarrow S^0$), can be developed in wastewater biofilm or activated sludge [2,3]. Thus the sulfur cycle could simultaneously realize the biodegradation of organic matter, consumption of dissolved oxygen and detoxification of sulfide.

Nitrate (NO_3^-) is a common co-constituent in groundwater/wastewater with sulfate [4,5], and its reduction to dinitrogen gas leads to a variety of pathways to prevent sulfide emission [6–11]. NO_3^- has proven to be capable of inhibiting SRB growth through bio-competition with nitrate reducing bacteria (NRB), and eliminating sulfide production or sulfidogenesis by denitrification intermediate, nitrite (NO_2^-) through bio-inhibition on functional enzyme, dissimilatory sulfite reductase (Dsr) [8–10]. Other studies have also demonstrated that NO_3^- addition stimulated indigenous sulfide-oxidizing, nitrate-reducing bacteria instead of an inhibition of the sulfate reduction activity [11].

Although both oxygen (O_2) and NO_3^- are potential sulfide detoxifiers, to the best of our knowledge few studies have systematically evaluated detoxification of sulfide with both O_2 and NO_3^- as electron acceptors which are often simultaneously present in the waste stream.

* Corresponding authors.
E-mail addresses: echo110244@126.com (C. Chen), rnq@hit.edu.cn (N.-Q. Ren).

http://dx.doi.org/10.1016/j.cej.2017.07.136
Received 7 June 2017; Received in revised form 20 July 2017; Accepted 22 July 2017
Available online 24 July 2017
1385-8947/

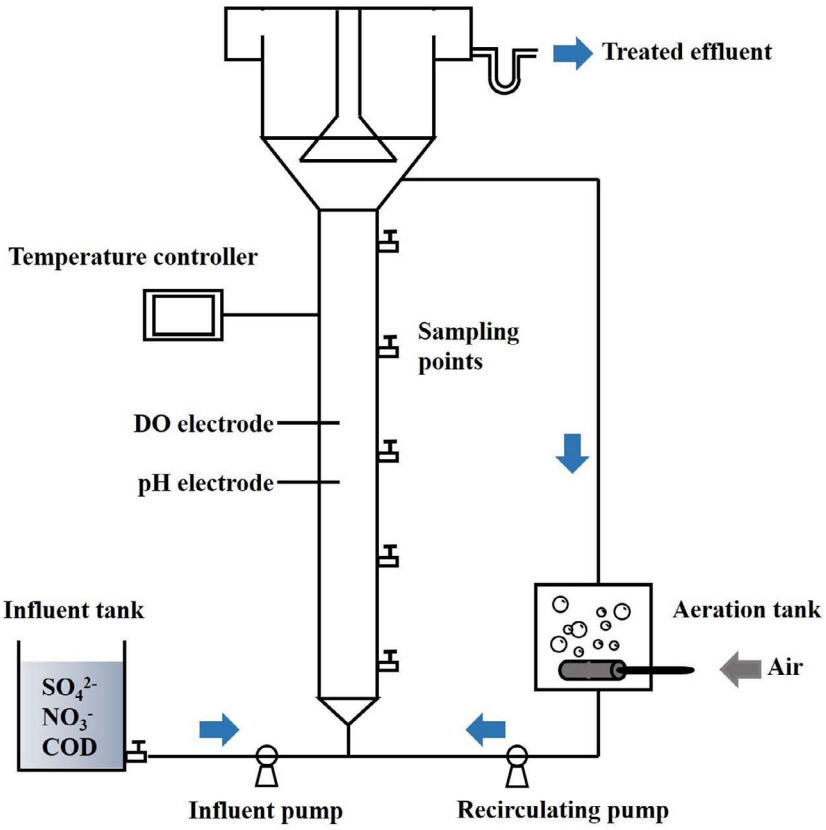

Fig. 1. Schematic of the reactor in present study.

Table 1
Operating conditions and performance of the reactor.

Stages	1	2	3	4	5	6	7
Time (d)	1–52	53–107	108–143	144–211	212–232	233–275	276–322
HRT (h)	18	18	18	18	18	18	18
AR (mL/min)	0	1	4	8	16	25	0
DO (mg/L)	0.02	0.07–0.10	0.10–0.14	0.14–0.20	0.25–0.30	0.30–0.35	0.02
Inf-sulfate (mg/L)	998.8	1005.2	996.3	990.4	997.6	1008.4	1000.3
Sulfate removal (%)	96.35	96.81	91.00	93.20	86.35	79.93	84.08
Inf-nitrate (mg/L)	500.9	506.3	502.0	506.7	503.3	503.3	496.7
Nitrate removal (%)	~100	~100	~100	~100	~100	~100	~100
COD removal (%)	52.0	61.8	89.6	94.7	91.0	88.7	85.4%
Sulfide (mg/L)	273.5	239.4	139.8	217.0	150.2	87.5	207.4
Sulfur recovery (%)	11.1	32.1	69.2	27.5	41.2	40.6	19.6
Samples	a_1	a_2	a_3	c_2	c_3	c_4	c_6

AR: aeration rate.
Inf-sulfate/nitrate: sulfate/nitrate in the influent.

Therefore, in present study the first aim is to investigate the performance of sulfide oxidation and sulfur recovery when these two electron acceptors simultaneously occur and to what extent might each one contribute to sulfide oxidation during bioremediation. The micro-aerobic technology would be feasible to improve the effluent quality in wastewater treatment plant if it could eliminate sulfide emission and meanwhile notably enhance S^0 production. Furthermore, compared to the conventional aerobic process, the micro-aerobic process could greatly reduce the cost of aeration.

Biological sulfide oxidation is achieved by sulfide-oxidizing bacteria (SOB). Most SOB belong to *Proteobacteria* phylum, and are capable of oxidizing various sulfur compounds (e.g. S^0, thiosulfate ($S_2O_3^{2-}$), sulfite (SO_3^{2-})) with O_2 or NO_3^- as electron acceptor [12]. However, current study that examined microbial communities in bacteria oxidizing sulfide powered by electrons from both O_2 and NO_3^- is limited. Therefore, the second objective of this study is to clarify the microbial community dynamics responding to the two electron acceptors, especially community dynamics associated with sulfide-oxidation by a high throughput biotechnology, Illumina sequencing. Importantly, we also investigated whether the enriched dominant microorganisms in the microbial community or the microbial community structure (e.g. richness and evenness) best contributed to the enhanced S^0 production in a microaerophilic sulfate and nitrate co-reduction system. Understanding the precise microbial response to the two electron acceptors can help us better regulate and control the developed sulfide detoxification biotechnology in the future.

2. Materials and methods

2.1. Bioreactor design and operation

A lab-scale expanded granular sludge bed (EGSB) with working volume of 4 L (height of 120 cm and internal diameter of 50 mm) was built. Five sampling points were evenly distributed along reactor wall