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Review

Efficiency and bacterial populations related to pollutant removal in an upflow microaerobic sludge reactor treating manure-free piggery wastewater with low COD/TN ratio

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HIGHLIGHTS

- An upflow microaerobic sludge reactor used to treat manure-free piggery wastewater.
- NH_4^+-N and COD/TN ratio of the wastewater being 299 mg/L and 0.84, respectively.
- Average load removal of COD and TN as high as 0.72 and 0.94 kg/(m^3 d), respectively.
- Aerobes, anaerobes and facultative aerobes thriving in the microaerobic system.
- Populations related to COD, NH_4^+-N, TN and phosphate removal being identified.

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ABSTRACT

A novel upflow microaerobic sludge reactor (UMSR) had proved excellent in nitrogen removal from manure-free piggery wastewater characterized by high concentration of ammonium (NH_4^+-N) and low chemical oxygen demand (COD) to total nitrogen (TN) ratio, but the biological mechanism in the UMSR was still indeterminate. With a constant nitrogen loading rate of 1.10 kg/(m^3 d) at hydraulic retention time 8 h, the UMSR was kept performing for 67 days in the present research and the average load removal of COD, NH_4^+-N and TN was as high as 0.72, 0.76 and 0.94 kg/(m^3 d), respectively. Compared with the inoculated sludge, the acclimated sludge was richer in genera responsible for the biological removal of carbon, nitrogen and phosphorus. Ammonium oxidation bacteria, heterotrophic denitrifiers, autotrophic denitrifiers and phosphate accumulating organisms coexisted perfectly in the microaerobic system, and their synergistic action made the UMSR perform well in COD, NH_4^+-N, TN and phosphate removal.

1. Introduction

In the past two decades, there has been a considerable increase in pig breeding in China and many other developing countries. The National Bureau of Statistics of the People's Republic of China reported that 7.35×10^9 pigs had been slaughtered in 2014, indicating a 2.7% increase with respect to the previous year. Statistically, recorded piggery wastewater discharged to environment was above 1.1×10^7 tons in 2014. Practicing of "open dumping" of piggery wastewater has a high possibility in leading to significant health and environmental consequences due to the uncontrolled decomposition of waste which may eventually result in the outbreak of diseases, proliferation of foul odors, eutrophication of water body and climate change (Yu and Cao, 2014). Given the holophytic nutrition, piggery wastewater has been used as fertilizer directly (Ra et al., 2000). However, this management has a poor feasibility in many countries due to the limited field resources, especially in China with a large population. Piggery waste is commonly consisted of high-strength suspended solids (SS), organisms and nutrient (Lim et al., 2012), but the quality of piggery wastewater discharged from pig farms vary greatly from each other mainly due to the different collection mode of manure, which includes fermented bed, urine-free manure, combined

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manure with urine, and soaked manure with urine (Wang and Guo, 2009). Per the traditional manure collection mode in China, removal of particulate solids (manure) before piggery being flushed with water is still widely used in commercial scaled pig farms whereby water effluent after flushing is termed manure-free piggery wastewater. Manure-free piggery wastewater is a typical high-strength ammonium (NH_4^+-N) wastewater with low carbon/nitrogen (C/N) ratio, which initiates difficulty in an attempt to remove nitrogen (Zhao et al., 2014).

Anaerobic–aerobic combined process, as an engineered treatment technology with robust adaptability, is commonly used to treat piggery wastewater because of its simultaneous removal of carbon, nitrogen and phosphorous. The combined process is generally carried out in two reactors in series or in a single reactor operated in sequencing process, resulting in a high treatment cost or low treatment efficiency. Furthermore, it is more difficult to remove nitrogen under low C/N ratio (about 0.5–1.8 in the manure-free piggery wastewater) (Yiğit and Akbal, 2014). The beneficial C/N ratio of 6–8 has been suggested for denitrification (Obaja et al., 2003). And when C/N ratio was less than 3.4, the denitrifying bacteria would be inhibited because of the lack of carbon source (Kuba et al., 1996). Dosing carbon source or decreasing NH_4^+-N concentration through physical chemistry methods have been used to gain a feasible C/N ratio for biological nitrogen removal, but extra treatment cost and complexity of equipment management often results in the process (Guštin and Marinšek-Logar, 2011).

Microaerobic process has been applied to treat domestic wastewater in recent years (Zheng and Cui, 2012). Microaerobic condition is a transitional state between anaerobic and aerobic conditions. Generally, the dissolved oxygen (DO) concentration in microaerobic reactors is ranged from 0.3 to 1.0 mg/L (Zitomer, 1998). Aerobe, anaerobe and facultative aerobe could thrive in different depths of activated sludge flocs according to their demand for oxygen, which dedicates that the organics and nutrients would be removed simultaneously in a single reactor under microaerobic condition (Zheng and Cui, 2012). Aerobic bacteria like ammonium oxidation bacteria (AOB), nitrite oxidation bacteria (NOB) and heterotrophic aerobic bacteria can thrive on the outer layer of activated sludge floc resulted in the relative abundance of DO. Oxygen consumption of the aerobic bacteria can create anaerobic microenvironment deep in the sludge floc and allows anaerobic microorganism like denitrifiers and anaerobic fermentation bacteria to dwell in (Liu and Dong, 2011; Ergüder and Demirer, 2005). In addition, capacity of anti-impact load can be achieved in microaerobic processes, with lower yield of excess sludge, economical operating cost and higher chemical oxygen demand (COD) removal (Zitomer, 1998).

In previous research, an upflow microaerobic sludge reactor (UMSR) has been proved a feasible process to remove NH_4^+-N and total nitrogen (TN) from manure-free piggery wastewater with low COD/TN ratio (Meng et al., 2015a). Though the analysis of potential nitrogen removal pathways suggested that anaerobic ammonium oxidation (ANAMMOX) was the main mechanism for NH_4^+-N and TN removal in the UMSR, the biological mechanism was still indeterminate due to the lack of information about microbial community structure. As known, microbes related to pollutant removal play an essential role and the performance of a biological wastewater treatment process has a significant relationship with its microbial community structure (Carvalho et al., 2007). More often, the component and structure of the microbial community would be evidently affected by the operational condition of the treatment process. The microbial community diversity in traditional wastewater treatment plants has been widely investigated (Kuroda et al., 2015). However, the information on efficiency and microbial community structures in microaerobic treatment process is scanty and minimum, which may be due to the complexity of functional species. The high throughput sequencing technique was originated several years ago and has been applied widely to analyze the microbial communities in various environment (Yang et al., 2014). Compared to conventional molecular biology methods, pyrosequencing can profile exactly complex bacterial community by providing quantity of DNA reads, operational taxonomic units (OTUs) and relative abundances of the predominant bacteria (Glenn, 2011).

After sludge acclimation and a steady state reached at a nitrogen loading rate (NLR) of about 1.10 kg/(m^3 d), the UMSR was kept performing for another 67 days while the pollutant removal was evaluated in the present research. To get a comprehensive insight into the biological mechanism for removal of carbon, nitrogen and phosphorus simultaneously in the microaerobic treatment process, the microbial community structure and the relationship between the key microbial populations and the pollutant removal were investigated.

2. Methods

2.1. Microaerobic treatment system

The microaerobic treatment system was illustrated in Fig. 1. The UMSR in the microaerobic treatment system was constructed with a plexiglass column. The reactor's design features included a working volume of 4.9 L, height of 500 mm, a 0.5 L circular cone attached to the bottom of the column, a 3 L solid–liquid–gas separator fitted onto the top of the column, and a four-sampling ports at 100 mm intervals and away from bottom of the solid–liquid–gas separator. The off-gas was discharged after a water seal. The piggery wastewater was introduced into the reactor by a peristaltic pump from the bottom. The effluent was collected by a 10 L tank. Part of the effluent was aerated to a DO of about 3 mg/L and recirculated into the reactor at a reflux ratio of 45:1 to create a microaerobic environment with a DO less than 1 mg/L in UMSR. DO in the reactor was detected by an on-line monitoring instrument, which was used to control the aeration in the aerating tank. The temperature in the reactor was kept at 35 ± 1 °C constantly by a temperature controller.

2.2. Wastewater and inoculum

The raw manure-free piggery wastewater was collected from a local pig farm in Harbin, China. The quality of the wastewater fluctuated following breeding seasonality. The average concentration of COD, NH_4^+-N, nitrite (NO_2^--N), nitrate (NO_3^--N), TN, total phosphors (TP), total alkalinity (TA) and pH was 307, 299.7, 0.1, 1.0, 366.9, 10.2, 1150.92 mg/L and 8.0, respectively. It could be observed that NH_4^+-N dominated the TN and the COD/TN ratio averaged as low as 0.84. Thus, the primary mission for treating the wastewater is the removal of NH_4^+-N and TN.

The seed sludge inoculated the UMSR was anaerobic sludge collected from an upflow anaerobic sludge blanket (UASB) treating piggery wastewater in the State Key Laboratory of Urban Water Resource and Environment, Harbin Institute of Technology, China. The initial biomass in terms of mixed liquor suspended solids (MLSS) was 5.68 g/L.

2.3. Acclimation of the inoculated sludge

Before the presented performance of the UMSR, the inoculated sludge had been acclimated for 200 days based on a program of decreasing CON/TN ratio step by step. The COD/TN ratio were regulated using molasses. The feed, operating parameters and results