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Effects of pressurized aeration on organic degradation efficiency and bacterial community structure of activated sludge treating saline wastewater

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HIGHLIGHTS

- Both treating efficiency and DHA improved at high salinity by pressurized aeration.
- Bacterial richness and diversity were significantly higher at each salinity.
- *Bacteroidetes* phylum kept abundant in pressurized reactor at high salinity.
- More species originating from fresh wastewater survive in pressurized reactor.

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ABSTRACT

This study was aimed to investigate the effect of moderate pressure on organic matter removal efficiency and microbial population of activated sludge treating saline wastewater. The activated sludge was cultivated with a gradual increase of salt concentrations under gage pressure of 0.3 MPa for 71 days. Microbial diversities of activated sludge sampled in different stages of domestication were investigated by Illumina sequencing technology. Results showed that pressurized aeration could improve the treatment efficiency and the dehydrogenase activity (DHA) of activated sludge, especially at high salinity (35, 50 g NaCl L^{-1}). Bacterial richness and community diversity of activated sludge in the pressurized reactor were significantly higher than those in the control reactor. Microbial population structures were quite different between the two reactors. More species originating from fresh wastewater biological treatment process would survive and remain in pressurized activated sludge.

1. Introduction

Activated sludge process is widely used as an effective and economical treatment for organic pollutants degradation in saline wastewater, which represents as much as 5% of worldwide effluent treatment requirements (Lefebvre et al., 2007). However, high salt concentration would strongly inhibit microorganisms in conventional biological treatment process (Zhao et al., 2013). Typical impact is the high osmotic pressure which leads to changes of microbial metabolism, cell plasmolysis, and even death of unacclimated microbes (Cortés-Lorenzo et al., 2014). Acclimation of the microbial system is a feasible technique to improve the treatment efficiency of saline wastewater by washing out nonhalophiles and promoting the abundance of halotolerants or halophiles in the system (Wang et al., 2014). Halophilic archaea and bacteria are two main groups of microorganisms adaptive in saline environments. Studies have been focused on the fundamental mechanisms of salt tolerance in recent years, including increase of the intracellular ion concentration (mainly potassium) and accumulation of organic solutes called "compatible solutes". Although the adaptation of activated sludge has already been proved to be possible, a major bottleneck is that the proper performance of such salt-adapted systems is usually limited to less than 5% salt concentration (Lay et al., 2010). Effective degradation of pollutants in saline wastewater remains one of the major challenges in organic wastewater biological treatment.

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Oxygen supply is an important factor in aerobic biodegradation. With regard to saline wastewater, high salt concentration results in increasing viscosity and reducing diffusivity and hence the oxygen transfer from air to water (Krampe and Krauth, 2003). Furthermore, it directly affects the oxygen solubility. DO concentration decreases with the increasing salinity, and the dependence appears to be linear in the salinity range of 0–50 g L^{-1} (Lay et al., 2010). Besides, it has been demonstrated that relatively more oxygen is used by cells grown in the presence of higher salt concentrations (Kincannon and Gaudy, 1966), which means that the adverse effect of high salt concentration on oxygen transfer may further deteriorate biological treating efficiency. Some biological processes enhance the solubility of oxygen by moderate pressure, e.g. pressurized activated sludge process by increasing total air pressure (Jin et al., 2010) and deep-shaft process by using hydrostatic pressure to increase oxygen transfer efficiency. The moderate pressures exerted in these processes have been demonstrated to be no damage to microorganism (Dufresne et al., 1990). Even though these technologies have been employed to treat industrial wastewaters always accompanied with high salinity (Irwin et al., 1989; Krauth and Staab, 1993; Krauth, 1996; Jin et al., 2010; Hait and Mazumder, 2011), little work has been conducted to study the salt resistant ability of activated sludge under moderate pressure.

The influence of salt on microbial community diversity in biological process has also been studied. Traditional molecular techniques have been successfully used to study microbial community composition and dominant microbial population of activated sludge treating saline wastewater, e.g. denaturing gradient gel electrophoresis (DGGE) combined with other molecular tools (Yoshie et al., 2001; Vyrides et al., 2010). High-throughput sequencing methods have also been applied to investigate changes of the microbial communities in activated sludge by providing enough sequencing depth to cover the complex microbial communities (Ma et al., 2013, 2015; Zhao et al., 2014). In general, communities always change obviously after an acclimation period in saline wastewater due to dying out of nonhalophiles and abundant growth of halophiles (Cortés-Lorenzo et al., 2014; Wang et al., 2014), which plays important role in biological treatment of saline wastewater. However, it is still unknown how pressurized aeration affects microbial population of activated sludge treating saline wastewater.

The aim of this work was to evaluate the influence of pressurized aeration on salt resistant ability of activated sludge. The activated sludge has been cultivated with a gradual increase of salt concentration in a sequencing batch reactor for 71 days under 0.3 MPa gage pressure. Another sequencing batch reactor was running in atmospheric environment as control reactor, with the same operation parameters except for the pressure. Samples were collected routinely and bacterial diversity was investigated by Illumina sequencing technology. The salt resistant ability of activated sludge was compared and evaluated by both the treating efficiency and the microbial diversity.

2. Materials and methods

2.1. Wastewater and activated sludge

The composition of the synthetic fresh wastewater was as follows: CH_3COONa, 500 mg L^{-1}; Glucose: 250 mg L^{-1}; NH_4Cl: 170 mg L^{-1}; KH_2PO_4: 20 mg L^{-1}; $NaHCO_3$: 40 mg L^{-1}; $CaCl_2$: 40 mg L^{-1}; $MgSO_4{\cdot}7H_2O$: 164 mg L^{-1}. The wastewater was characterized with total organic carbon (TOC) concentration of 200–300 mg L^{-1}, ammonia nitrogen (NH_3-N) 40–50 mg L^{-1}, total phosphorus (TP) 4–5 mg L^{-1} and pH 6.5–7.5. The saline wastewater was the solution of the synthetic fresh wastewater containing different dosages of sodium chloride according to the experimental requirements.

Seed sludge was obtained from a municipal wastewater treatment plant in Nanjing, China. The seed sludge was then fed with the synthetic fresh wastewater. The sequencing batch culture process ran four cycles a day in a 150 L plastic container under normal environment. Each cycle (5 h) with the exchange volume ratio of 75 L/cycle consisted of the following steps: feeding (0.5 h), aeration (3 h), settling (1 h), and decanting (0.5 h). Aeration rate and sludge retention time (SRT) were controlled at 500–600 L h^{-1} and 15 d, respectively. The synthetic fresh wastewater culture process ran over 3 months until it reached steady state.

2.2. Reactors and experimental design

The experiments were conducted in two parallel bench-scale bioreactors. The pressurized bioreactor, running under gage pressure of 0.3 MPa by an air compressor, was made of steel with 300 mm in diameter, 900 mm in height and 50 L in working volume. The atmospheric contrast bioreactor as control reactor was transparent plexiglas column covered with black craft paper in order to avoid influence of transmitted light on microbial population. Both reactors were in the same size and shape except for the pressure.

The experimental reactors were inoculated by the activated sludge which has been fed with the synthetic fresh wastewater. The initial mixed liquor suspended solids (MLSS) concentration was approximately 2500 mg L^{-1}. Then salt-resistant acclimation was conducted in sequencing batch mode with gradual increase of salt concentrations. The SBR had four cycles a day with an exchange volume ratio of 25 L/cycle of each cycle (5 h), consisting the same steps as those of synthetic fresh wastewater culture process. Initially, the two reactors ran for 5 days without salt. Then, 5 g L^{-1} of NaCl solution was added and the reactors ran for 7 days. The salt concentration was subsequently increased in sequence with an elapsed time of 11–18 days between each addition of salt, depending upon the time needed for each salinity. Table 1 summarizes the main operating conditions of the acclimation process.

2.3. Activated sludge samples collection

The sludge samples were collected at regular intervals. Nine sludge samples were collected totally. Sample IN was collected from inoculated sludge on the first day of the experiment, which represented the initial status of the activated sludge of both control reactor and pressurized reactor. Samples A1, A2, A3 and A4 were all collected from the control reactor in different periods, representing the microbial community structures of activated sludge adapted to different salt concentrations. P1, P2, P3 and P4 were collected from the pressurized reactor in the corresponding periods, respectively. The sampling date and the corresponding salinity are described in Table 2.

Table 1
Operation conditions of the salt-resistant acclimation process.

	Unit	Pressurized reactor	Control reactor
Gage pressure	MPa	0.3	0
Aeration rate	L h^{-1}	300	300
DO	mg L^{-1}	~8.2	~4.3
SRT	d	13–20	13–20
Temperature	°C	25 ± 2	25 ± 2
MLSS	mg L^{-1}	2800 ± 300	2800 ± 300
Influent TOC	mg L^{-1}	200–300	200–300
Gradual salinities	g L^{-1}	0, 5, 15, 25, 35, 50	0, 5, 15, 25, 35, 50