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Oxygen transfer dynamics and nitrification in a novel rotational sponge reactor

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ABSTRACT

The nitrification process demands higher oxygen supply, which is fulfilled through external aeration in wastewater treatment. To perform nitrification without external aeration, this study developed a novel rotating sponge (RS) reactor and estimated its oxygen transfer capability. The reactor obtained an oxygen transfer coefficient (K_La) value of over 100 d^{-1} at 40 rounds per hour (rph), which is one-eighth of the rotational speed of conventional rotational biological contactors. Within 53 days of operation, reactor achieved 97% nitrification efficiency with an effluent NH_4^+-N concentration of 1 mgL^{-1}. On the other hand, the volumetric oxygen transfer rate in the reactor was 245 mg-$O_2L^{-1}d^{-1}$ at 10 rph during the experimental period. However, the average volumetric oxygen consumption rate during nitrification was 187 mg-$O_2L^{-1}d^{-1}$, which was below the oxygen transfer rate. Microbial community analysis revealed high relative abundance of bacteria such as *Nitrosomonas*, *Nitrospira*, and *Nitrobacter* in the RS reactor, which contributed to nitrification.

1. Introduction

Wastewater contains ammonium nitrogen, which is toxic to aquatic organisms, causes environmental problems such as eutrophication, and depletes dissolved oxygen in water bodies [1,2]; therefore, it is necessary to remove nitrogen compounds from wastewater. In conventional biological nitrogen removal processes, ammonium nitrogen is oxidized to nitrate through nitrification, and nitrate is then reduced to nitrogen gas through denitrification [3]. Nitrification is the crucial step in this process, as nitrifying bacteria show lower growth rates compared to heterotrophic bacteria. Dissolved oxygen (DO) concentration is an important factor that affects the nitrification process. Park and Noguera [4] demonstrated that a low DO concentration inhibits the growth rate of nitrifying bacteria, which eventually reduces nitrification. Therefore, to maintain good nitrification efficiency, a higher oxygen supply is required; hence, external aeration is the main factor contributing to the cost of the process [5]. This research focuses on the development of a wastewater treatment system that could achieve nitrification without external aeration.

With the advantages of compactness, low energy consumption, easy operation, and a small ecological footprint, rotating biological contactors (RBCs) have gained popularity over conventional systems for wastewater treatment. However, conventional RBCs have a few shortcomings such as limited attachment area and a low oxygenation capacity, arising mainly from the use of disc media [6]. RBCs that are operated at rotational speeds of 100–300 rounds per hour (rph) could achieve oxygen transfer coefficients (K_La) of 30–100 d^{-1} [7,8]. To achieve high substrate degradation rates, increased oxygenation capacity is necessary, which is achieved by increasing the rotational speed [9]. However, high rotational speeds lead to high power consumption, which may not be economical for wastewater treatment applications [10]. As an alternative, this study proposes a modified novel rotating-type system, called a rotational sponge (RS) reactor, which will achieve higher oxygen transferring capabilities at lower rotational speeds.

To enhance the oxygen transfer capability in RBCs, this study proposes a modified novel rotating-type system, called a rotational

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sponge (RS) reactor. The use of sponges as microbial attach medium in down flow type reactors have been well documented [11–13]. Uemura et al. [14] have reported superior oxygen transferring capability in sponges used in the down flow type reactors. Unlike discs in conventional RBCs, sponges consist of more than 90% void space, resulting in a significant increase in entrapped biomass, which leads to longer solid retention time (SRT) [15]. These characteristics of sponge media may be important for obtaining high performance and oxygenation capability at lower rotational speeds in the RS reactor.

This study estimated the oxygen transfer capability in the RS reactor for achieving good nitrification efficiency without external aeration. To elucidate the nitrification characteristics in the RS reactor, inorganic synthetic wastewater containing ammonia was used for experiments. The structure of microbial community responsible for nitrification in the RS reactor was analyzed using sequence analyses based on the 16S rRNA gene.

2. Material and methods

2.1. Reactor design

The concept of the RS reactor is similar to that of a water wheel mounted vertically on a horizontal axle. Buckets arranged on the outside rim lift the influent with each rotation and allow wastewater to flow through randomly packed polyurethane sponges. In this study pilot-scale RS reactor was used with a rotating wheel, polyurethane sponges and a water tank (Fig. 1). The rotating wheel with a diameter of 2 m and width of 0.25 m was separated in to 8 partitions. Each partition was further compartmentalized in to two layers, named as outer and inner layers. 32 buckets were built-in to the outside rim of the wheel and each bucket could approximately lift 0.3 L of wastewater per one rotation. Each partition was packed with G3- type polyurethane sponges [16] with sponge volume of 110 L. The diameter and width of the water tank were 2.4 m and 0.5 m, respectively. The immersion level was set as 15% with 90 L wastewater volume in the tank. In this study, rotational speed of the reactor was controlled through a speed control motor.

2.2. Determining of physical oxygen transfer capability

Physical oxygen transfer rate (P-OTR) was measured in the RS reactor using tap water with clean sponge media. The water tank was filled with tap water and sparged with nitrogen gas to lower the DO level to 1 mg-O_2L^{-1}. Subsequently, rotation of the reactor was started, and increases in DO level were monitored. P-OTR was estimated using the slope of linear DO increase with time. Volumetric oxygen mass transfer coefficient water (K_La) was calculated using equation 1 [17].

$$dc/dt = K_La\cdot(C_{st}-C_t)$$

dc/dt: transfer rate of oxygen to the water ($mgL^{-1}d^{-1}$)

C_{st}: saturation dissolved oxygen concentration (mgL^{-1})

C_t: dissolved oxygen concentration in the effluent at time t (mgL^{-1})

K_La: volumetric oxygen mass transfer coefficient water, (d^{-1})

Since this study maintained the ambient temperature at 30 °C, C_{st} value was taken as 7.53 mgL^{-1} for the calculation purposes [17]. The experiment was carried out at rotational speeds of 5, 10, 15, 20, 25, 30, 35 and 40 rph at an immersion level of 15%. The K_La values obtained from this study were compared with those for conventional RBCs to identify the physical oxygen transfer capability in the RS reactor.

2.3. Operational conditions for nitrification

Prior to 30 days of operation of the reactor, nitrifying bacteria were pre-cultured in a separate tank using seed sludge obtained from conventional activated sludge process (ASP) located at sewage treatment center in Nagaoka, Japan. During this period, NH_4Cl was used as nitrogen source to achieve the NH_4^+-N concentration of 35 mgL^{-1} in the influent and also $NaHCO_3$ to maintain pH for growth of nitrifying bacteria.

Clean sponges that were earlier used for the determination of P-OTR were replaced with these pre-cultured sponges in the reactor. The rotational speed for the operation was set to 10 rph. During the operation of the reactor inorganic synthetic wastewater was fed to the reactor. The composition of synthetic influent was varied as follows: NH_4Cl (0.12 gL^{-1}), $NaHCO_3$ (1.44 gL^{-1}), KH_2PO_4 (0.14 gL^{-1}), K_2HPO_4 (3.5 gL^{-1}), Na_2SO_4 (0.43 gL^{-1}), $CaCl_2{\cdot}2H_2O$ (0.07 gL^{-1}), $MgCl_2.6H_2O$ (0.1 gL^{-1}). Influent was supplied at a flow rate of 300 Ld^{-1} resulting in a hydraulic retention time (HRT) of 16 h. Temperature and pH were maintained at 30 °C and 8.5 respectively, which were within the optimum range for nitrification[18]. During the operation NH_4^+-N, NO_3^--N, and NO_2^--N of both influents and effluents were measured frequently. However, the values of same parameters at outer and inner layers of the reactor were measured on day 30 and 53 of the operation.

2.4. Analysis

To monitor the nitrification performance, NH_4^+-N, NO_3^--N, and NO_2^--N in the influent and effluent were regularly measured by high-performance liquid chromatography (Shimadzu, Japan). pH and DO were measured on-site using a portable pH meter (TPX-999Si- TOKO, Japan) and a DO meter (DKK-TOA, Japan). All other analytical procedures were performed as described in APHA [19].

2.5. DNA extraction and polymerase chain reaction (PCR)

On day 53, three samples were obtained from different locations at same height on the outer layer. Two samples were also collected in a similar manner from the inner layer. 200 mL of each sample was collected by squeezing out 20 sponges. The samples were then washed with a PBS solution and centrifuged 3 times at 15,000 rpm at 4 °C for 10 min, and stored at −20 °C until DNA extraction. DNA was extracted from the samples using a FastDNA SPIN Kit for Soil (MP Biomedicals). The 16S rRNA genes were amplified using the universal forward primer Uni515F and the universal reverse primer Uni806R [20]. PCR amplification was performed using Premix Ex Taq Hot Start Version (Takara Bio Inc., Shiga, Japan) with a Veriti Thermal Cycler (Applied Biosystems, California, USA). The conditions for the PCR amplification were as follows; 1 cycle of initial complete denaturation at 95 °C (3 min) 25 cycles at 95 °C (45 s), 50 °C (60 s), 72 °C (90 s), and a final extension at 72 °C (10 min). The PCR products were purified using MinElute PCR Purification Kit (QIAGEN, Hilden, Germany) according to manufacturer's protocols.

2.6. DNA sequencing and analysis

PCR products were sequenced using MiSeq Reagent Kit v2 Nano with MiSeq System (Illumina INC., San Diego, CA, USA). Raw sequence reads were quality trimmed and filtered using Trimmomatic version 0.33 [21]. Trimmed reads from the paired-end library were merged with FLASH version 1.2.11 [22]. The merged sequences were analyzed using UPARSE pipeline [23]. High-quality reads were clustered at 97% sequence identity into operational taxonomic units (OTUs). The representative sequences were assigned