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Establishment of Biological Nitrogen Removal Process for Drinking Water Treatment in Malaysia

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ABSTRACT

Eutrophication arises from human activities has been recognised globally as an environmental issue. Human activities have greatly increased the input of phosphorus and nitrogen into the water bodies which place a heavy pressure on clean water resources. There are limitations in the conventional water treatment processes, where phosphorus and nitrogen could not be removed efficiently. For nitrogen removal, biological method emerges as a sustainable and economically feasible alternative in recent years. Though the biological nitrogen removal (BNR) method is widely recognised in developed countries with temperate climate, knowledge and expertise on the application of this treatment process is relatively limited in developing countries, so as Malaysia in tropical region. Thus, this research aims to provide a preliminary view on the establishment of the BNR process for drinking water treatment in the local context. A lab scale sequencing batch reactor (SBR) seeded with fish pond sludge and activated sludge in the volume ratio of 1:1 was operated for the BNR process. After four weeks of sludge cultivation, the characteristic of simultaneous nitrification-denitrification was observed. The removal of total nitrogen (TN) was found exceeding 60% and little nitrate-nitrogen (NO₃⁻-N) concentration was observed at the end of the process. Besides, the reduction in orthophosphate (PO₄³⁻-P) concentration in a BNR system has indicated the process was able to perform nitrogen and phosphorus removal simultaneously. These findings suggest that the establishment of BNR is feasible in Malaysia.

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Abbreviations

<i>AOB</i>	Ammonium-Oxidizing Bacteria
<i>BNR</i>	Biological Nitrogen Removal
<i>COD</i>	Chemical Oxygen Demand
<i>DWTP</i>	Drinking Water Treatment Plants
<i>MLSS</i>	Mixed Liquor Suspended Solid
<i>MLVSS</i>	Mixed Liquor Volatile Suspended Solid
<i>OLAND</i>	Oxygen-limited Autotrophic Nitrification-Denitrification
<i>PAO</i>	Polyphosphate Accumulating Organisms
<i>SBR</i>	Sequencing Batch Reactor
<i>SND</i>	Simultaneous Nitrification-Denitrification
<i>TN</i>	Total Nitrogen

1. Introduction

Water is essential in our daily life. However, poor water quality has recently brought out the concerns to society especially in the developing countries (Edokpayi, Odiyo, & Durowoju, 2017). Human activities have affected the water quality by introducing nitrogen-based contaminants such as ammonia, nitrite and nitrate which can lead to serious water pollution such as eutrophication and the development of harmful N-nitroso carcinogen compounds to human (Ward et al., 2018; Xue et al., 2016). In order to overcome this, different regulations have been implied and enforced across the regions to control the N discharged into the water bodies. According to Drinking Water Quality Standards for Malaysia, the maximum allowable limit of ammonia-nitrogen and nitrate-nitrogen are 1.5 mg/L and 10 mg/L respectively (Ministry of Health Malaysia, 2020; United States Environmental Protection Agency, 2020). Although conventional Drinking Water Treatment Plants (DWTPs) have been practicing nitrogen removal through physical and chemical methods, these plants are having difficulties in removing nitrogen-based contaminants effectively (Amin, Alazba, & Manzoor, 2014). Therefore, a reliable and efficient treatment process is needed.

Biological Nitrogen Removal (BNR) process is lately becoming an alternative in reducing nitrogen content in water because of its high removal performance, low cost, environmental friendly and the ability in converting the nitrogen-based contaminants into harmless products (Mohseni-Bandpi, Elliott, & Zazouli, 2013). BNR is a two-step process to remove harmful nitrogen compounds, originated from ammonia. In the first step of BNR process, nitrification is taken place to oxidise the ammonium ion into nitrite and nitrate in the presence of nitrifying bacteria such as *Nitrosomonas* and *Nitrobacter* (American Water Works Association, 2002). The process is followed by the denitrification, a reduction process of nitrate to harmless nitrogen gas with the help of the denitrifying bacteria, for instance *Azopira sp.* and *Pseudomonas* (Rossi, Motta, Matrella, Proto, & Vigliotta, 2014). The BNR process categorized into conventional and modern system. The conventional BNR is sufficient to

achieve 95% nitrogen removal efficiency, but the discharging effluent still contains nitrate (NO_3^-) and nitrite (NO_2^-). In the context to improve that, the process has been modified to a modern BNR. Several methods are implied in the modern BNR such as Simultaneous Nitrification-Denitrification (SND), Aerobic Deammonification and Oxygen-limited Autotrophic Nitrification-Denitrification (OLAND). These methods share some common features in which only one reactor is required for the process and harmless nitrogen gas (N_2) is the only discharged product. Moreover, SND and Aerobic Deammonification require aerobic condition while OLAND require oxygen limited condition. These methods were reported to achieve 60% - 100% of the nitrogen removal efficiency with N_2 as the only discharged element when they were operated using Sequencing Batch Reactor (SBR) (Zhu et al., 2008).

Full scale BNR processes are common in developed countries at temperate region. However, BNR process is relatively limited in developing countries, so as Malaysia due to the lacking of expertise and knowledge in operating BNR at tropical climate. For an effective and efficient BNR process, microbial growth is important to achieve the maximum activity of nitrification and denitrification. It is worth noting that the process removal efficiency is highly affected by the microbial population in the system. Studies found that the growth temperature for nitrifying bacteria and denitrifying bacteria is between 20°C to 40°C (Gerardi, 2003). For every 10°C increase, the microbial growth rate is almost doubled. This has then increased the rate of nitrification and denitrification. In addition, the process removal efficiency is also dependent to the seeding source. Study reported that stabilised aerobic granular sludge achieved about 60% of nitrogen removal efficiency (Wagner, Guimarães, Akaboci, & Costa, 2015). Moreover, the anaerobic open pond has higher nitrifying bacteria and denitrifying bacteria activities in comparison to anaerobic close system (Thongnueakhaeng & Chairasert, 2016). These reports suggested that the use of different types of seeding source will give a different performance to the BNR process. Therefore, the objective of this study is to establish the BNR process for water treatment in local context by using fish pond sludge and activated sludge.

The BNR performance is evaluated from the view of ammonium-nitrogen ($\text{NH}_4^+\text{-N}$) concentration, nitrite-nitrogen ($\text{NO}_2^-\text{-N}$) concentration, nitrate-nitrogen ($\text{NO}_3^-\text{-N}$) concentration, orthophosphate ($\text{PO}_4^{3-}\text{-P}$) concentration and Chemical Oxygen Demand (COD). Besides that, mixed liquor suspended solid (MLSS) and mixed liquor volatile suspended solid (MLVSS) are tested in order to support concentration profiles analysis.

2. Materials and methods**2.1 Set up of Sequencing Batch Reactor (SBR)**

In this study, modern BNR process is used whereby a 2L lab scale SBR was set up for the BNR process. The SBR was fed with the synthetic river water and seeded with fish pond sludge and activated sludge in volume ratio of 1:1. The initial MLSS was set at 1,500 mg/L. The SBR was operated in 4 cycles per day with 6 hours each cycle at room temperature of around 28°C. The pH was adjusted manually ranges 6.5 to 7.5 by adding either 0.5 M hydrochloric acid or 0.5 M sodium hydroxide. The SBR

consists of 5 major stages, 7 minutes of filling, 4.5 hours of aeration, followed by 1 hour of settling, 15 mins of decanting and 8 minutes of idling. The schematic diagram of the SBR is as shown in Figure 1.

2.2 Preparation of synthetic river water

Figure 1: Schematic diagram of the SBR

Synthetic river water was used in the SBR operation as the feed. The nutrient concentrations in the feed were as follow (per litre): 0.7 mg/L sodium acetate, 0.1 mg/L yeast extract, 34.0 mg/L calcium chloride, 30.8 mg/L magnesium sulphate heptahydrate, 0.49 mg/L iron (III) chloride, 19.0 mg/L ammonium chloride and 0.09 mg/L monopotassium phosphate (Smith, Davison, & Hamilton-Taylor, 2002). The chemicals used are supplied by Merck, Germany.

2.3 Monitoring of BNR performance

The cyclic profile of the process was monitored weekly by measuring the chemical oxygen demand (COD), inorganic ion concentrations ($\text{NH}_4^+\text{-N}$, $\text{NO}_2^-\text{-N}$, $\text{NO}_3^-\text{-N}$, $\text{PO}_4^{3-}\text{-P}$), mixed liquor suspended solid (MLSS) and mixed liquor volatile suspended solid (MLVSS). The COD was identified using HACH Spectrophotometer (DR 3900, Hach). The ion concentrations of $\text{NH}_4^+\text{-N}$, $\text{NO}_2^-\text{-N}$, $\text{NO}_3^-\text{-N}$ and $\text{PO}_4^{3-}\text{-P}$ were analysed using Ion Chromatography (861 Advanced Compact IC, Metrohm). The MLSS and MLVSS were determined using APHA standard method (Clesceri, Greenberg, & Eaton, 1998). The mixed liquor samples from the SBR were filtered with 0.45 μm regenerated cellulose syringe filter prior to the COD analysis and were further filtered with 0.2 μm PTFE syringe filter prior to the Ion Chromatography analysis.

Figure 2: Concentration profiles of $\text{NH}_4^+\text{-N}$, $\text{NO}_2^-\text{-N}$, $\text{NO}_3^-\text{-N}$, $\text{PO}_4^{3-}\text{-P}$ and COD in one SBR cycle on (a) Week 1; (b) Week 2; (c) Week 3; (d) Week 4 for fish pond sludge-activated sludge cultivation; (hatched bars) $\text{NH}_4^+\text{-N}$; (solid black bars) $\text{NO}_2^-\text{-N}$; (dotted bars) $\text{NO}_3^-\text{-N}$; (solid grey bars) $\text{PO}_4^{3-}\text{-P}$; (blue line with circles) COD.

2.4 Removal efficiency of total Nitrogen

The removal efficiency of TN, η can be calculated using Equation 1, where C_o is the initial TN concentration and C_t is the final TN concentration.

$$\eta = \left[1 - \left(\frac{C_t}{C_o} \right) \right] \times 100\% \quad (1)$$

3. Results and Discussion

3.1 BNR sludge acclimatisation

The COD uptake, ammonium reduction, nitrate formation and phosphate uptake patterns in the selected cycles of the SBR operation were monitored for the emergence of BNR characteristics. The concentration profiles of $\text{NH}_4^+\text{-N}$, $\text{NO}_2\text{-N}$, $\text{NO}_3\text{-N}$, $\text{PO}_4^{3-}\text{-P}$ and COD for a period of 4 weeks are as shown in Figure 2.

As shown in Figure 2(a), the decrease of $\text{NH}_4^+\text{-N}$ from 21.9 mg/L to 4.6 mg/L and gradual increase of $\text{NO}_3\text{-N}$ from 4.8 mg/L to 7.1 mg/L has indicated the occurrence of nitrification in the system. The increase of $\text{NO}_3\text{-N}$ concentration was not corresponding to the drastic dropped of $\text{NH}_4^+\text{-N}$ concentration which could possibly due to the simultaneous occurrence of denitrification with nitrification, where a portion of $\text{NO}_3\text{-N}$ had been converted into N_2 . This simultaneous nitrification and denitrification (SND) could be caused by the high oxygen content, high $\text{NO}_2\text{-N}$ concentration and low organic carbon contents (Khanitchaidecha, Nakaruk, Koshy, & Futaba, 2015).

The $\text{NO}_3\text{-N}$ concentration in Week 1 was higher than that in Week 2 (Figure 2(b)) and Week 3 (Figure 2(c)). This was caused by the intermediate NO_2^- accumulation from partial nitrification. The $\text{NO}_2\text{-N}$ was seen increasing over the reaction time (Ruiz, Jeison, & Chamy, 2003). This could happen with the pH change in system. The nitrifier and denitrifier require a pH range from neutral to slightly alkaline. Low pH could lead to the inhibition of enzymatic activity. In contrast, high pH would affect organotrophs microbial activity (Gerardi, 2003). The $\text{NO}_3\text{-N}$ was not detected from 0 h to 2.5 h in Week 4 (Figure 2(d)). However, it showed a relatively constant concentration at 3.5 h and 4.5 h. Accumulation of $\text{NO}_2\text{-N}$ was observed too.

As for the $\text{PO}_4^{3-}\text{-P}$ concentration profile, there was neither increment nor reduction in Week 1 (Figure 2(a)). The constant orthophosphate concentration of around 2 mg $\text{PO}_4^{3-}\text{-P/L}$ indicated that the SBR was not acclimatised for the P uptake. However, the $\text{PO}_4^{3-}\text{-P}$ concentration dropped to approximately 0.2 mg/L in Week 2 (Figure 2(b)) and this could be due to the bacterial metabolism, specifically polyphosphate accumulating organisms (PAOs) (Tarayre et al., 2016). Other than that, one interesting result had been observed, where the $\text{PO}_4^{3-}\text{-P}$ concentration decreased gradually from 0.8 mg/L to 0.0 mg/L in Week 3 of BNR acclimatisation, as shown in Figure 2(c). This indicated that the BNR system has the ability to perform nitrogen and phosphate removal simultaneously (Yin, Zhang, Li, Li, & Hai, 2015). Similarly in Week 4 (Figure 2(d)), the reduction of orthophosphate concentration from 0.2 mg/L to 0.0 mg/L in the aerobic phase has further clarified that the system has developed a characteristic of simultaneous biological nitrogen and phosphate removal.

In terms of COD profile, the values were fluctuating over the four weeks. The decreasing trend of COD was observed. The decrease of $\text{NH}_4^+\text{-N}$ concentration

and orthophosphate uptake corresponded to the decrease COD concentration indicated that the microorganisms had consumed the carbon source to perform microbial growth and metabolism (Fang, Xu, Xia, & Pan, 2018).

After 4 weeks of sludge acclimatisation, it was found that the MLSS and MLVSS were decreasing. This could be caused by the poor sludge settleability, and further caused the sludge washout (Lu et al., 2015).

3.2 TN removal efficiency

Figure 3: Removal efficiency of TN throughout four weeks; (■) Initial TN; (▨) Final TN; (▲) Removal Efficiency.

Figure 3 showed that the removal efficiency of TN was increased from 45.8% to 62.1% in the first two weeks. The relatively high initial $\text{NH}_4^+\text{-N}$ concentration at Week 1 can be caused by the presence of NH_4^+ in the fresh sludge. After one week of adaptation period, the initial $\text{NH}_4^+\text{-N}$ concentration seems to be constant in Week 2 and Week 3. However, it was found that the TN removal efficiency was dropped from 62.1% to 54.0% which may be caused by the inhibition of ammonium-oxidizing bacteria (AOB) during that period. A drastic drop in TN removal efficiency to -7.2% was noticed in Week 4 which was caused by the high final $\text{NH}_4^+\text{-N}$ concentration remained in the system. This could be resulted from the low microbial activity from AOB. Furthermore, the NO_2^- accumulation in the system also led to high N content at the final concentration.

4. Conclusion

The BNR characteristics were developed in the SBR operated in this study, which further indicates the establishment of BNR process using a mixture local activated sludge and fish pond sludge is feasible. Throughout the sludge acclimatization period, the characteristic of simultaneous nitrification-denitrification (SND) was observed in the SBR. In addition, the phosphorus removal ability was emerged too in the system after two weeks of sludge cultivation. Overall, the removal efficiency of TN was achieved as high as 62.1%. Further studies in the correlation of microbial population and the process performance will definitely provide more in-depth information for the operation of BNR process in the real drinking water treatment plant in Malaysia.

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