



Application of photosynthetic oxygen of microalgae for reducing energy in single algae–nitrification reactor

Jiwon Lee, Jiyeol Im, Kyungik Gil*

Department of Civil Engineering, Seoul National University of Science and Technology, Nowon-gu, Seoul, South Korea, email: kgil@seoultech.ac.kr (K. Gil)

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ABSTRACT

The paradigm of wastewater treatment system is changing from viewing the municipal wastewater treatment plants (MWTPs) as a center of energy consumption to viewing it as a center of energy production. Recently, many studies have focused on utilizing algae photosynthetic oxygen as a method for energy saving and production in MWTPs. However, such algae are still viewed as a target for treatment and research has been conducted completely on application in Korea. In this research, laboratory-scale algal-nitrification reactors were operated to investigate the practicality of using photosynthesis by algae to increase the energy efficiencies of nitrification in MWTPs. Three operating parameters were mainly examined: hydraulic retention time (HRT), mixed liquor suspended solids (MLSS):algae ratio and light. As a result, stable ammonia nitrogen removal at high efficiency was demonstrated in line with the HRT. When the algae concentration was held constant, the nitrification efficiency increased as the MLSS concentration was reduced. The results of this research could be used as a useful basic data for the algae-nitrification process.

Keywords: Micro-algae; Nitrogen; Nitrification; Energy save; Energy production

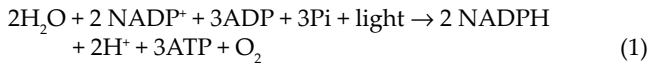
1. Introduction

In Korea, the municipal wastewater treatment plants (MWTPs) are recognized as an essential element in the social infrastructure but are assumed to require large amounts of electric power to operate aerator, pump, and sludge dewatering equipment. Meanwhile, the paradigm of wastewater treatment is changing from viewing the MWTPs as energy consumption facility to energy production facility in developed countries such as Netherlands, the United States, Germany, Spain, etc. [1–3]. At the 2017 International Water Association special conference held in the Netherlands, the MWTP was announced to as a “water and resource recovery facility (WRRF)”, which means future MWTPs will conduct producing energy such as methane gas, environmentally friendly plastics and biofuels as well as

treating wastewater. This paradigm change in MWTPs seems to be highly related with an application of energy conserving processes such as single reactor for high ammonium removal over nitrite (SHARON) or anaerobic ammonium oxidation (ANAMMOX). Accordingly, the interests in microalgae photosynthetic oxygen have been also increased due to its new role by conserving and generating energy in MWTPs. To date, algae have been viewed in Korea mainly as a source of eutrophication in lakes and rivers, and an increase in the algal population has been seen as a threat to the supply of clean water. So, many policies have been implemented to suppress algae [4]. In recent years, however, researchers have begun to investigate the role of algae in producing biofuels such as biodiesel. In particular, the use of oxygen produced by algae (photosynthesis) is being investigated as a way

* Corresponding author.

of utilizing microalgae in an MWTP [5–7]. Eq. (1) shows a basic photosynthesis formula of light-dependent reaction.



However, some limitations remain for applying microalgae to MWTPs. A key problem is that the hydraulic retention time (HRT) is longer than that of conventional activated sludge process. The growth and photosynthesis of algae are also significantly affected by weather and temperature [2]. To address these problems, studies have reported that cultivation of microalgae and bacteria in a fluid carrier [8], the effect of salinity and temperature on the growth of microalgae [9], and the effect of lighting condition such as kind of LED, wavelength, intensity of illumination, etc. [8,10]. The goal of this research was to analyze the nitrogen removal mechanism using photosynthetic oxygen of microalgae in nitrification and to identify key operating factors that influence the nitrogen removal mechanism. The experiment was conducted over a period of 130 d. The effects of HRT on nitrification, mixed liquor suspended solids (MLSS):algae ratio, and LED wavelength were analyzed. The results of this study provide basic data on the key operating factors in algae nitrification.

2. Materials and methods

2.1. Characteristics of influent

In this study, synthetic wastewater was used as the influent to ensure a stable algae-nitrification. Table 1 shows the characteristics of the wastewater, which contained

Table 1
Characteristics of influent

Synthetic wastewater			
Parameter	Value	Parameter	Value
COD	30 mg/L	NH ₄ ⁺ -N	50 mg/L
BOD	20 mg/L	TP	5 mg/L
TN	60 mg/L	Alkalinity	360 mg/L

BOD (Biochemical oxygen demand); COD (Chemical oxygen demand); TP (Total phosphorus), TN (Total nitrogen).

ammonium nitrogen (ammonium sulfate), phosphorus (potassium phosphate) and alkalinity (sodium bicarbonate). These were dissolved in distilled water to NH₄⁺-N of 50 mg/L and TP of 5 mg/L. The optimal pH for nitrifying microorganisms has been reported to be between 7.5 and 8.5 [11]. To minimize the effect of alkalinity from nitrification and algae photosynthesis, the pH was monitored and adjusted as needed. The analysis of water quality followed the standard method [12].

2.2. Laboratory-scale reactors

The laboratory-scale reactors were acrylic cylinder with an operational volume of 3 L. Fig. 1 shows a schematic of the reactor by stage of operation. Under operating condition E, an external pump supplied additional oxygen to only one of the reactors (R2), allowing the supply of oxygen intermittently. This is because photosynthesis of the algae produced oxygen that is insufficient to support nitrification. To ensure complete mixing, a magnetic bar was placed inside the reactor and a stirrer was installed under the constant temperature bath.

2.3. Materials

Fig. 2 shows the LED and media, which can immobilize microorganisms. Bar-shaped red, white and blue LEDs were used and Table 2 shows the physical properties of the LEDs. The white LED was used for the operating conditions A to F, whereas blue and red LEDs only were used in the operating condition G. In this study, two types of media were used. First, media is named K-3, which is manufactured by a company “Veolia Water Technology” in France. It is shaped like a toothed wheel and used mainly for the cultivation of algae. The second was a polyurethane media with a sponge-like shape and structure and was operated with nitrifier attached. The physical properties of the media are shown in Table 3.

2.4. Analysis of chlorophyll-a

Various microorganisms such as microalgae and nitrifying bacteria were included in microalgae-nitrification reactor. So, it is difficult to independently quantify the

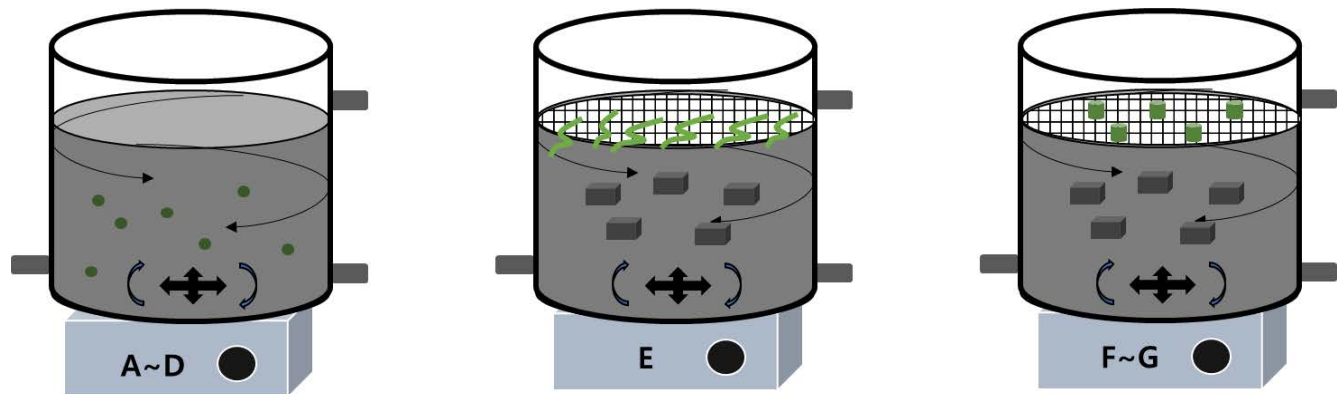


Fig. 1. Schematic diagram of laboratory-scale reactors by operation condition.

amount of microalgae. The microalgae biomass concentration was, therefore, estimated by measuring chlorophyll-a, which is present in all photosynthesizing organisms except bacteria and in the phytoplankton of water bodies. Raschke [13] estimated that it accounts for about 1%–2% of the dry weight of organic algal matter. For deriving the concentration of microalgae, Raschke [13] and Kang [14] proposed Eq. (2).

$$\text{Algae biomass (mg/L)} = 100 \div 1.5 \times \text{Chl-a(mg/L)} \quad (2)$$

Chlorophyll-a was estimated using the standard methods [12]. First, the sample were filtered with GF/C, crushed using acetone (9 + 1), and then stored in a cold dark place at 4°C for 24 h. After centrifugation (500 g for 20 min), the amount and absorbance of supernatant at wavelengths of 663, 645, and 630 nm were measured. The chlorophyll-a concentration was analyzed using Eq. (3).

$$\text{Chlorophyll-a (mg/mL)} = 11.64X1 - 2.16X2 + 0.10X3 \quad (3)$$

X1 = OD663 – OD750
X2 = OD645 – OD750
X3 = OD630 – OD750 (OD = optical density)

3. Results and discussion

3.1. Experiment results of reactor

The reactors were operated for about 130 d, under the experimental operating conditions A–G. Fig. 3 shows the nitrogen concentration in each reactor. Basically, R1 and R2

were operated from the beginning to end, but in the experiment where more reactors were needed in accordance with the operating conditions, R3 or R4 was added flexibly. In all four reactors, nitrate was produced and ammonium nitrogen was removed, confirming that nitrification proceeded normally. The ammonium nitrogen removal efficiency (ARE) was estimated from the difference in ammonium nitrogen concentration of the influent and effluent.

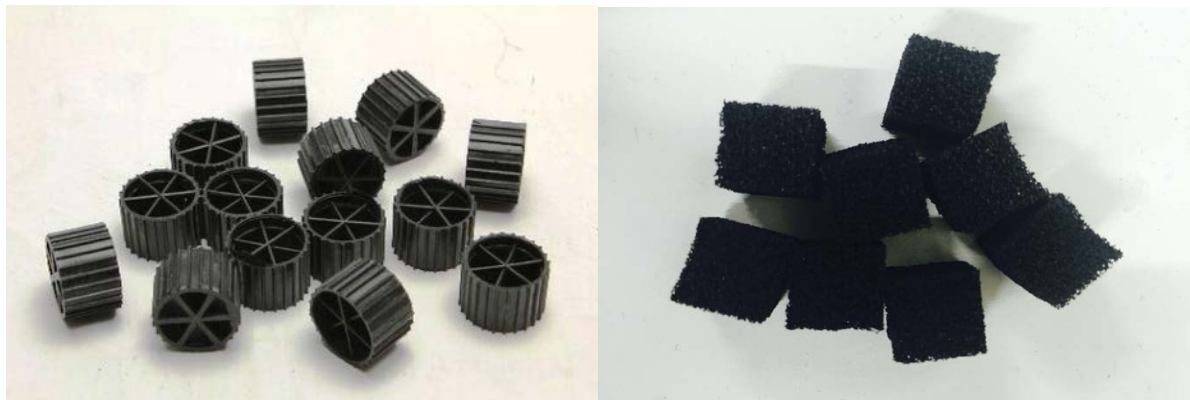
Table 4 shows AREs by operating periods at each reactor. First, R1 was fully operated during period A~G and following results appeared. 15.8%~21.2% for A, 79.6%~90.8% for B, 9.6%~20.8% for C, 21.0%~35.2% for D, 28.6%~43.0% for E, 27.6%~59.2% for F and 58.4%~67.2% for G. Second, R2 was also fully operated all periods that shows 9.8%~30.4% for A, 67.8%~79.8% for B, 15.8%~38.4% for C, 14.6%~31.8% for D, 27.8%~52.8% for E, 50.6%~81.0% for F and 77.2%~90.6% for G. Third, R3 was partially operated that 16.4%~24.8% in A, 27.0%~71.0% in C, 27.8%~49.2% in D. Finally, R4 was operated only in period D of which values were 46.4%~57.2%.

3.2. Operating condition

Table 5 and Fig. 4 show summarization on experiment contents and ARE results by operating condition. In section A, to investigate appropriate N:P ratio, different ratios were applied to each reactor. In section B, to see a stable nitrification, extended HRT and aeration were applied to R1 (4 d + Air), and R2 (8 d). Under operating period C, solid retention time (SRT) of 20 d was applied to minimize leakage through wash-out of algae, and each reactor

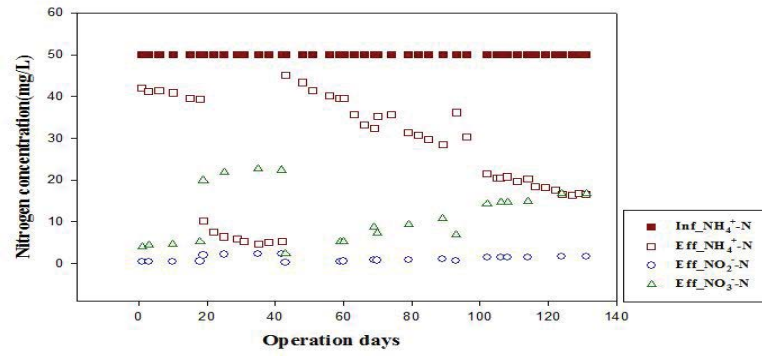


(a)

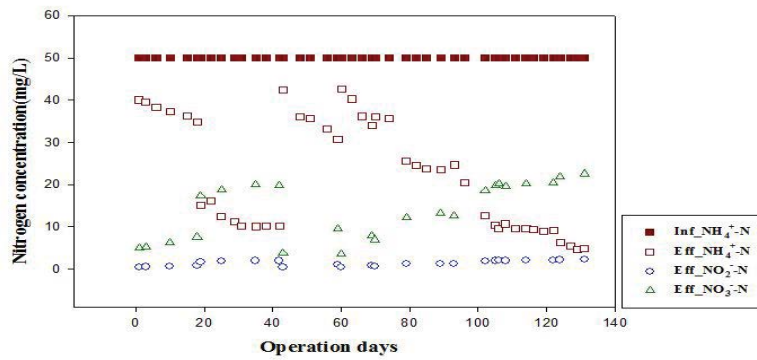


(b)

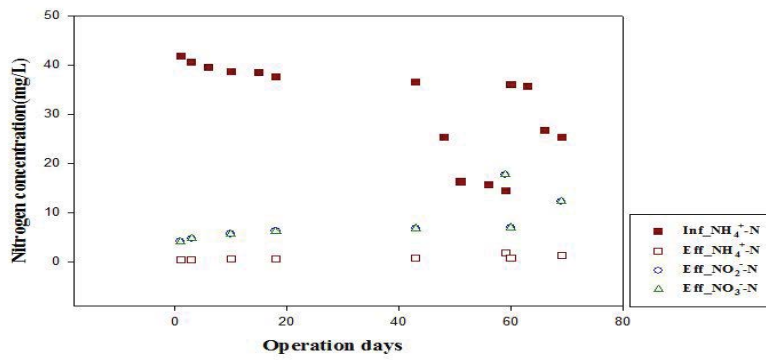
Fig. 2. Used (a) LED and (b) medias in this study.



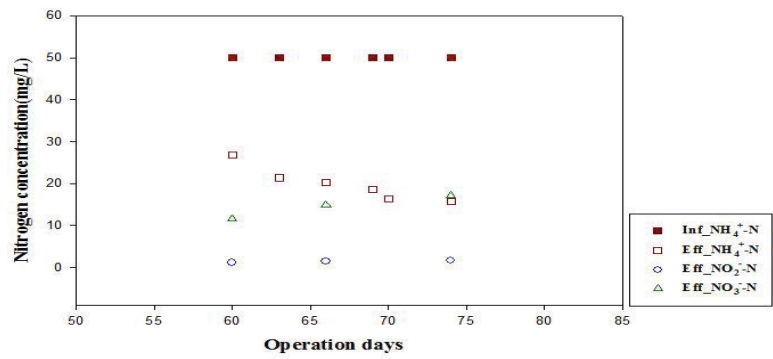
(a) R1



(b) R2



(c) R3



(d) R4

Fig. 3. Nitrogen concentration variation of each reactor.

Table 2
Physical characteristics of LED

Type	Bar
Lux	15,000–30,000 lx
PPFD (photosynthetic photon flux density)	195–390 $\mu\text{mol}/\text{m}^2 \text{ s}$
Dimension	1,000 mm (L) \times 31 mm (W) \times 32 mm (H)

Table 3
Physical characteristics of media

Base material of media	K-3	Polyurethane
S.G. of media	0.95	0.07
Shape and size (mm)	Round (12–14)	Cube (15 mm \times 15 mm \times 15 mm)
Specific surface area of media (m^2/m^3)	500	2,000

S.G.: Specific gravity.

Table 4
Ammonium nitrogen removal efficiency by operating periods at each reactor

Reactors	Ammonium nitrogen removal efficiency by operating period (%)						
	A	B	C	D	E	F	G
R1	15.8–21.2	79.6–90.8	9.6–20.8	21.0–35.2	28.6–43.0	27.6–59.2	58.4–67.2
R2	19.8–30.4	67.8–79.8	15.8–38.4	14.6–31.8	27.8–52.8	50.6–81.0	77.2–90.6
R3	16.4–24.8	–	27.0–71.0	27.8–49.2	–	–	–
R4	–	–	–	46.4–57.2	–	–	–

was used a different HRT (2, 3, and 4 d). In section D, ARE variation was investigated at different MLSS:algae concentration. R1 was operated at 3,000:200, R2 was operated at 2,000:200, R3 was operated at 1,000:200 and R4 was operated at 500:200. In operating period E, mesh filter was used to R1 and R2 to reduce wash-out of algae biomass and media used for preventing light blindness by nitrifier in MLSS. In section F, the algae biomass also was attached to media (K-3), and additional aeration (6 h) of total HRT (12 h) was applied to R2, because photosynthetic oxygen of pure algae was deemed to be insufficient to produce high-efficiency nitrification. Lastly, in period G, light condition was changed from white light to a mix of blue and red light according to the dissertations [8,15] which refer. The amount of photosynthetic oxygen production in algae increases at mixed light than at single light.

3.3. Effect of HRT

In Fig. 5, the effect of HRT on nitrification and algal biomass production was analyzed. When HRT was 2 d, ARE was from 10% to 21% and average value of algae biomass was about 145 mg/L. In HRT 3 d, ARE was from 15% to 40% and average value of algae biomass was about 190 mg/L. When HRT was 4 d, ARE was from 27% to 71% and average value of algae biomass was about 212 mg/L. In reactor, algae biomass was maintained by wash-out at a constant concentration. But when the HRT is shortened, because of increased

effluent flow, wash-out volume becomes increased that makes algae biomass low inside reactor. For this reason, amount of oxygen generated by the algae become reduced, which means that sufficient oxygen supply for nitrification is difficult. On the other hand, when the HRT is extended, both ARE and algae biomass also increased. Especially, ARE increased significantly when the HRT was changed from 3 to 4 d. In other words, as though ARE seems to be related to HRT, ARE is considered to be more related to algal biomass, which generate oxygen by photosynthesis.

3.4. Effect of MLSS:algae ratio

Fig. 6 shows the changes of ARE with various MLSS:algae ratio in suspended growth process. MLSS:algae ratio was operated 15:1 (3,000:200), 10:1 (2,000:200), 5:1 (1,000:200) and 2.5:1 (500:200). The ratio was controlled by change of MLSS. As can be observed through Fig. 6, the highest ARE (over 50%) was observed during operational period of 2.5:1 (500:200). In high MLSS:algae ratio condition (over 5:1) shows lower ARE than low MLSS:algae ratio (2.5:1) in this research. ARE was decreased about 20% when changes of MLSS:algae ratio 2.5:1 to 5:1 and the lowest ARE was showed up during 10:1. Relationship of MLSS:algae ratio and ARE was not observed in over 10:1 MLSS:algae condition. According to Fig. 6, low MLSS:algae ratio is an effective condition for nitrification in suspended growth process, because MLSS in suspended growth process is

Table 5
Summary of experiment contents according to operating conditions

Operating days	Periods	Operating condition	Experiment contents
18(1~18)	A	R1 N:P = 5:1	Experiments were conducted to investigate the effects of different N:P ratio for each reactor Low nitrification rate due to insufficient oxygen production through algae photosynthesis
		R2 N:P = 10:1	
		R3 N:P = 20:1	
24(19~42)	B	R1 HRT 4 d + air	Stable nitrification in the algae-nitrification-tank operated through additional oxygen supply and HRT increase
		R2 HRT 8 d	
17(43~59)	C	R1 HRT 2 d	Effect of HRT was analyzed by different HRT for each reaction tank Drive SRT to 20 d to minimize runoff through algae wash-out
		R2 HRT 3 d	
		R3 HRT 4 d	
10(60~69)	D	R1 MLSS:algae = 3,000:200	Comparative analysis of photosynthetic oxygen production by algae through MLSS:algae ratio Prevention of algal growth inhibition due to high concentration MLSS and decrease of MLSS concentration induce increase of light transmittance and nitrification efficiency
		R2 MLSS:algae = 2,000:200	
		R3 MLSS:algae = 1,000:200	
		R4 MLSS:algae = 500:200	
20(70~89)	E	R1 Mesh	Operated using mesh filter to separate algae and nitrifier and to reduce wash-out of algae biomass A nitrifier was stored in media (fluid carrier), to prevent light blindness
		R2 Mesh + media(AOB)	
16(90~105)	F	R1 Media(algae) on the mesh + media(AOB)	Based on the operating condition E, both reactors are operated using algae media in addition to AOB media Reactor 2 has 6 h of additional aeration based on HRT 12 h
		R2 Media(algae) on the mesh + media(AOB)+air	
26(106~131)	G	R1 LED condition change: white → blue and red	LED condition changed from single white light condition to blue and red light condition Blue and red light mixed conditions have higher algal growth rates than single white light [14]
		R2	

SRT: solid retention time; MLSS: mixed liquor suspended solids.

related to permeability of light. Permeability of light is in turn related to oxygen production by algae directly [14]. Thus, MLSS is acted to inhibition parameter on permeability of light. The MLSS:algae ratio is an important parameter in this process.

3.5. Effect of light condition

Advanced research result, the light condition is highly correlated to algae biomass growth. [8,15]. The period G was operated to analyze the effect of light condition. Fig. 7 shows summarized operational result according to change of light condition. In this research, the operational condition of light was controlled single-light condition (white) and mixed-light condition (blue and red). In this paper, solar light (natural light) and artificial light were embodied the single light condition (white) and mixed light condition (blue and red) respectively. The artificial light was suggested to be one of the methods to increase ARE from advanced research result [8,14,15]. Single light and mixed light shows similar ARE, and ARE in each reactor reached about 60% when end of period G (operating day 9). The difference

of light condition was not shown clearly in end of period G. However, the initial of period G, ARE in mixed-light condition was higher about 30% than ARE in single-light condition. And operating day 5, ARE had been similar gradually. According to this result, the light condition affects algae biomass growth. Thus, mixed light condition has an advantage for fast stable algae-nitrification process.

4. Conclusions

The following conclusions can be drawn from the operation results of the algal-nitrification laboratory-scale reactor using synthetic wastewater.

- In this study, nitrification reactor based on microalgae photosynthetic operation was operated for about 130 d and operated to find optimal operating factors according to operating conditions from A to G. As a result, stable ammonia nitrogen removal efficiency was secured.
- Among the operating condition A~G, three important operating factors were considered: HRT, MLSS:algae ratio, and light condition. The nitrification efficiency

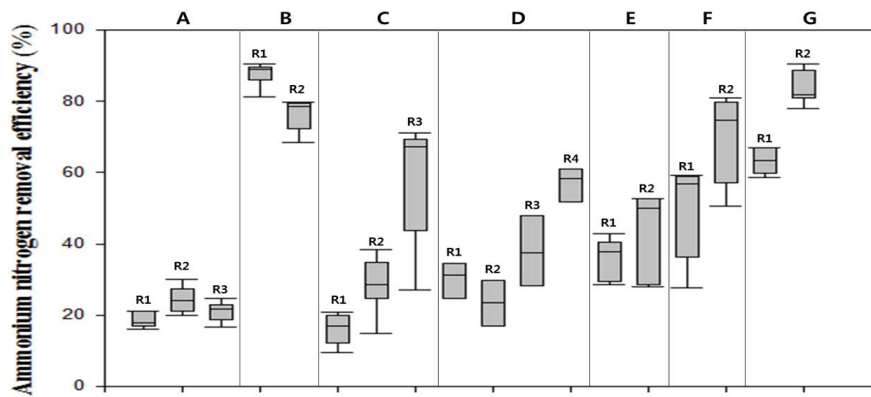


Fig. 4. Summarized ARE results by operating condition.

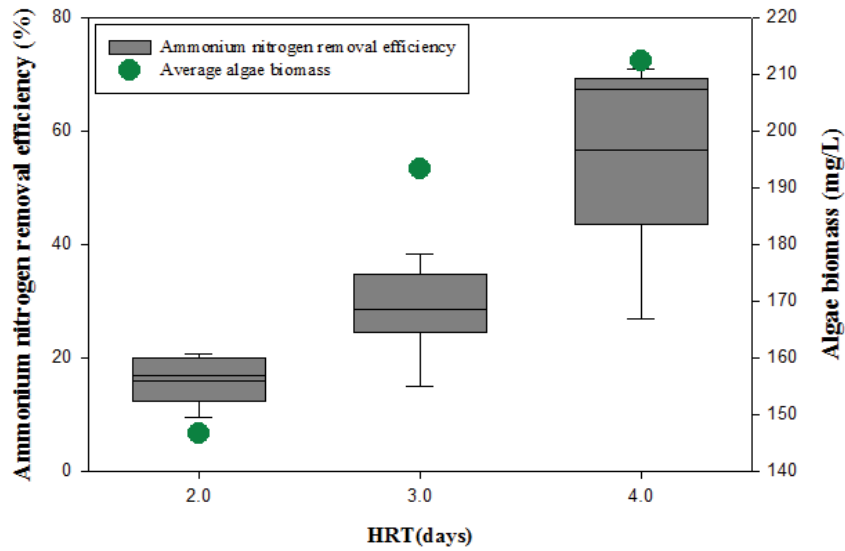


Fig. 5. Ammonium nitrogen removal efficiency and algae biomass as HRT.

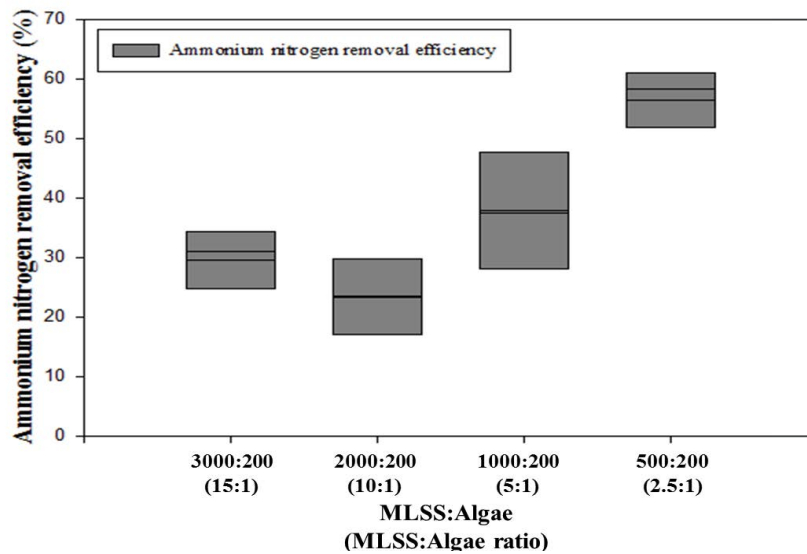


Fig. 6. Ammonium nitrogen removal efficiency as MLSS:algae ratio.

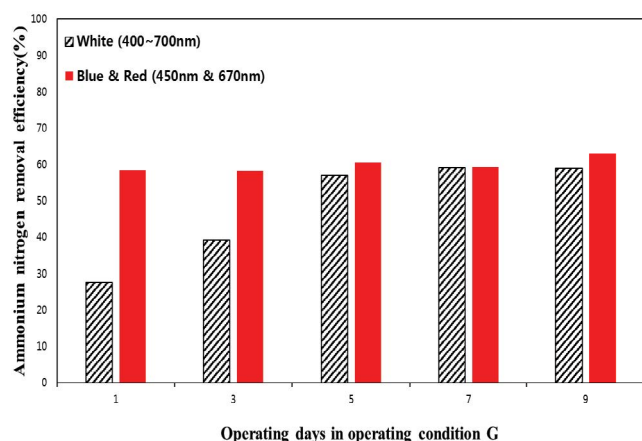


Fig. 7. Ammonium nitrogen removal efficiency as wavelength.

increased in line with the HRT. When the algae concentration was held constant, the nitrification efficiency increased as the MLSS level was reduced. The nitrification efficiency was slightly higher when the illumination was supplied by a mix of blue and red LEDs than by a white LED.

- In this study, for comparison with a full-scale MWTP, a HRT of 12 h was used. When relying on only oxygen from algal photosynthesis, ammonia nitrogen removal was limited. However, when additional aeration was supplied for 6–12 h, stable ammonia nitrogen removal was achieved. The results provided basic data for studies of the key operating factors in algae nitrification.

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