Field study on N\textsubscript{2}O emission from subsurface wastewater infiltration system under variable loading rates and drying-wetting cycles

Ying-Hua Li, Hai-Bo Li, Xin-Yang Xu, Si-Yao Xiao, Si-Qi Wang and Shu-Cong Xu

**ABSTRACT**

In this field study, the impacts of influent loadings and drying-wetting cycles on N\textsubscript{2}O emission in a subsurface wastewater infiltration (SWI) system were investigated. N\textsubscript{2}O emitted under different operation conditions were quantified using static chamber and gas chromatograph techniques. N\textsubscript{2}O conversion rate decreased from 6.6 \textpm{} 0.1\% to 2.7 \textpm{} 0.1\% with an increase in hydraulic loading (HL) from 0.08 to 0.24 m\textsuperscript{3}/m\textsuperscript{2}\cdot d. By contrast, N\textsubscript{2}O conversion rate increased with increasing pollutant loading (PL) up to 8.2 \textpm{} 0.5\% (PL 4.2 g N/m\textsuperscript{2}\cdot d) above which conversion rate decreased, confirming that N\textsubscript{2}O production was under the interaction of nitrification and denitrification. Taking into consideration the pollutants (chemical oxygen demand (COD), NH\textsubscript{4}\textsuperscript{+}-N, NO\textsubscript{3}-N and total nitrogen (TN)) removal ratio and N\textsubscript{2}O emission, optimal loading ranges and drying-wetting modes were suggested as HL 0.08–0.12 m\textsuperscript{3}/m\textsuperscript{2}\cdot d, PL 3.2–3.7 g N/m\textsuperscript{2}\cdot d and 12 h:12 h, respectively. The results revealed that in SWI systems, conversion ratio of influent nitrogen to N\textsubscript{2}O could be between 4.5\% and a maximum of 7.0\%.

**Key words** | conversion rate, drying-wetting cycle, influent loading, N\textsubscript{2}O emission, subsurface wastewater infiltration system

**INTRODUCTION**

The discharge of effluent from domestic sources has detrimental effects on the aquatic ecosystem as this can deposit a large amount of organic matter, nutrients and pollutants leading to eutrophication, temporary oxygen deficits and accumulation of pollutants into receiving waterways (Kong et al. 2002). As a result, some advanced treatment technologies are generally deemed necessary to decrease these constituents prior to reuse. Among the many treatment technologies developed to improve effluent water quality, subsurface wastewater infiltration (SWI) approach is considered to be a promising alternative due to lower investment and operational costs, simple management requirement, prolonged reliability and durability.

As a kind of natural wastewater purification system, SWI is an effective way to treat wastewater according to integrated mechanisms of chemical, physical and biological reactions as it passes through the unsaturated soil in infiltration system (Pan et al. 2015). SWI is different from other well-known natural treatment systems, such as subsurface wetland and surface wetland, especially in nitrogen removal mechanisms. Constructed wetlands have higher NH\textsubscript{4}\textsuperscript{+}-N removal efficiency because of aerobic conditions (by the oxygenation of rhizosphere and the aeration of top layer) developed within the system (Zapater-Pereura et al. 2016). In addition, plant uptake plays an important role in nitrogen removal (Zapater-Pereura et al. 2016).

In SWI treatment, organic nitrogen is easily converted to ammonia nitrogen (NH\textsubscript{4}\textsuperscript{+}-N) by ammonifying bacteria and then the NH\textsubscript{4}\textsuperscript{+}-N can be adsorbed onto the soil because zeta potential of the soil particles are negative, and adsorption capacity of the soil can be quickly recovered to the initial state by nitrifying NH\textsubscript{4}\textsuperscript{+}-N to NO\textsubscript{3}-N, which is subsequently denitrified to N\textsubscript{2} or N\textsubscript{2}O by the denitrifying biomass under the anoxic condition (Li et al. 2014). When one of the following conditions occurs, such as low dissolved oxygen level or nitrite nitrogen (NO\textsubscript{2}-N) accumulation or limited nitric oxide reductase (Nos) activity, N\textsubscript{2}O
will be generated via nitrification or denitrification process (Sun et al. 2015). Mechanisms for N₂O emission are shown in Figure 1.

The production of N₂O during nitrification process mainly occurs in the oxidation of NH₄-N and hydroxylamine (NH₂OH). When dissolved oxygen level is limited, NO₂⁻-N will accumulate and extend its toxicity to microbial cells. Thus, heterogeneous nitrite reductase will be produced to reduce NO₂⁻-N to N₂O. On the other hand, denitrification process can promote N₂O emission in two ways: N₉S activity is restrained and NO₂⁻-N cannot be reduced completely, resulting in the NO₂⁻-N accumulation and N₂O production. In addition, under acid condition, accumulated NO₂⁻-N reacts with soil humic acid and generates N₂O. Therefore, there is a hypothesis that N₂O generation is the interaction of nitrification and denitrification.

Kong et al. (2002) compared the N₂O emission of SWI in Japan with that in China by fact-finding survey, and found a positive correlation between nitrogen removal, N₂O emission and oxidation reduction potential. Studies conducted by Ahn et al. (2011), Rassamee et al. (2011) and Li et al. (2016) confirmed that loading rate and drying-wetting regimes could affect nitrogen circulation in the following ways: (1) increasing hydraulic loading (HL) means shortening hydraulic retention time (HRT), so organic nitrogen is not fully degraded before discharged from the SWI system; (2) increasing loading leads to stronger shock for media surfaces, which is also responsible for NO₂⁻-N accumulation and N₂O production, as shown in Figure 1. Therefore, it is clear that loading and drying-wetting cycles can have a significant effect on the N₂O emission. This field study was carried out to (1) explore the influence of loading rate and drying-wetting cycle on N₂O emission and (2) suggest optimal loading and cycle ranges taking into consideration of treatment efficiency and N₂O emission abatement.

### MATERIALS AND METHODS

#### Site description

The experiment was carried out at the campus wastewater treatment plant in Shenyang, China. The SWI system has a design capacity of 3,000 people equivalent and covers 3,000 m², dividing equally into eight independent cells. The inflow rate was 300 m³/day. The primary effluent flows by gravity into the primary and secondary distribution tanks. Then, the wastewater flows through the SWI systems, and the effluent is finally collected into the collection tank (Figure 2). The SWI system is planted with turf type ryegrass (Lolium perenne L.).

Static chambers were used to monitor N₂O production (Tsushima et al. 2014). Four gas chambers (Figure 2, stainless steel, length 50 cm, width 50 cm, height 40 cm, sealed with a water-filled U trench, painted white to avoid heating during the application) were installed at four cells.

#### Matrix

The filling materials were meadow brown soil, dry activated sludge and slag mixed evenly in volume ratio 7:2:1. The meadow brown soil was sampled from the top 20 cm from Shenyang Ecological Station. The activated sludge was obtained from the aeration tanks in Shenyang Northern Municipal Sewage Treatment Plant, China, air dried after being centrifuged for 15 min at 1,500 rpm. Other materials (gravel and coal slag) were purchased from a local market (particle size: gravel 10–25 mm and coal slag 4–8 mm). The infiltration rate, porosity and surface area of the matrix were 0.37 m³/m²·d, 59% and 5.21 m²/g, respectively. The maximum adsorption capacity for NH₄-N was 0.724 mg/g.

#### Sampling and analytical methods

#### Water quality

Water samples were taken between 9:00–10:00 a.m. once a week from the secondary distribution tank and collection well, respectively. Influent and effluent samples were analyzed immediately for pH, chemical oxygen demand (COD), biochemical oxygen demand (BOD), NH₄-N, NO₃-N and total nitrogen (TN) using Chinese EPA standard methods (Chinese EPA 2002). From May 5th to September 15th, 2015, the variations of influent were 7.0–7.4 for pH, 210–361 mg/L for COD, 125–270 mg/L for BOD₅, 17–25 mg/L for NH₄-N, 27–43 mg/L for TN and 2–8 mg/L for NO₃-N.

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**Figure 1** | N₂O production mechanisms in SWI system.

\[\text{NH}_4^+ \rightarrow \text{NH}_3\text{OH} \rightarrow [\text{HNO}] \rightarrow \text{NO} \rightarrow \text{NO}_2^- \rightarrow \text{N}_2\text{O} \rightarrow \text{N}_2\]

\[\text{NO}_2^- \rightarrow \text{NO}_3^- \rightarrow \text{NO} \rightarrow \text{N}_2\text{O} \rightarrow \text{N}_2\]

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Gas collection and analytical method

Gas sampling was carried out once a week simultaneously with that of wastewater. The first air sample was collected immediately after the chamber was sealed. According to Kong et al. (2002), saturation time for field scale static chamber would not exceed 60 min. Thus, five air samples were taken from the roof of the chamber at 15 min intervals (0, 15, 30, 45 and 60 min) using a 50-mL plastic syringe with a three-way stopcock and a Teflon tube. The final accumulation value minus the initial one served as the basis for the calculation of N$_2$O production rates.

Gas samples were analyzed using a gas chromatograph (GC, 7890A, Agilent Technologies, USA) equipped with micro-electron capture detector running at 350°C. High-purity dinitrogen (N$_2$ 99.999%) was used as the carrier gas for N$_2$O analysis. The system was calibrated during each measurement cycle using known concentrations of gas (0.390 ppm N$_2$O in pure N$_2$). The calibration line used to analyze N$_2$O air samples by GC had a precision of 96.8%. Therefore, an error of ±5.2% may be assumed. Air samples were analyzed three times to quantify the human error that can be introduced by manual injection of the samples into the GC. Correlation analysis was carried out using SPSS 18.0.

Figure 2 | Sketch showing of the SWI system and static chambers. (a) Box. (b) Foundation support.

Calculation methods

The production rate of N$_2$O was estimated as the production of N$_2$O per unit area and unit interval. It was calculated by formula (1) (Sun et al. 2015), where $F$ was N$_2$O production rate (mg/m$^2$·h), $\rho$ was the density of N$_2$O at 0°C and 760 mm Hg (kg/m$^3$), $\Delta$C/$\Delta$t was the rate of N$_2$O concentration increased in the chamber (ppmv N$_2$O/h), $H$ was the height of the chamber headspace (m) and $T$ was the ambient air temperature (°C).

$$ F = \rho \times \frac{\Delta C}{\Delta t} \times H \times \frac{273}{(273 + T)} $$

(1)

Conversion ratio was the percentage of N$_2$O removal in influent TN. Its calculation was based on formula (2), in which $P$ was the conversion ratio of N$_2$O emission (%), $m$ was the quantity of nitrogen in N$_2$O (g) and $M$ was the quantity of total nitrogen in influent (g).

$$ P = \frac{m}{M} \times 100\% $$

(2)

Statistical analyses were carried out with MicroCal Origin 8.0 (OriginLab) and SPSS 18.0.
Experimental operation

During the whole experimental period, SWI systems were operated in alternation of wetting and drying cycles. Each cycle of the intermittent operation included a continuous flow period of 12 h and a drying period of 0, 4 h, 6 h, 12 h, 24 h and 36 h, indicating that the drying-wetting cycles were 0, 1:3, 1:2, 1:1, 2:1 and 3:1, respectively.

RESULTS AND DISCUSSION

N<sub>2</sub>O emission and conversion under variable HLs

During the first study period, HL (wastewater load per unit area per day) was set at 0.08, 0.12, 0.16, 0.20 and 0.24 m<sup>3</sup>/m<sup>2</sup>·d. Effluent quality, N<sub>2</sub>O production and conversion ratio were analyzed, as shown in Figures 3 and 4(a), respectively.
With the increase of HL, the removal rates of BOD$_5$ and NH$_4^+$-N decreased gradually, while that of NO$_3^-$-N presented an increasing trend. When the HL was lower than 0.12 m$^3$/m$^2$·d, removal rates for COD and TN increased with HL increase. However, when HL was higher than 0.12 m$^3$/m$^2$·d, the removal rates decreased dramatically. Correlation analysis for Figure 3 demonstrated that BOD$_5$ and NH$_4^+$-N removal rates had negative correlations with HL ($P < 0.05$). A high loading rate of carbon (BOD$_5$) leads to increased denitrification rates both by increasing the oxygen consumption making conditions more suitable for denitrification and providing organic carbon in the denitrification process (Li et al. 2013). In general, the COD requirement for denitrification is 3.0–4.5 mg COD/mg TN (Desloover et al. 2013). Thus, COD and TN removal presented similar trends. With respect to NO$_3^-$-N removal rate, HL had a positive effect on it. The N$_2$O production rate demonstrated an increasing trend firstly and then fell. The production rate, as high as 4.1 ± 0.2 mg/m$^2$·d, was achieved when the hydraulic was 0.16 m$^3$/m$^2$·d. On the other hand, N$_2$O conversion ratio decreased from 6.6 ± 0.1% to 2.7 ± 0.1% with an increase of HL from 0.08 to 0.24 m$^3$/m$^2$·d.

These observations confirmed that HL influenced organics removal in SWI in the following ways: first, the increase of HL shortened HRT. The organics were not fully degraded before discharged from SWI (Tsushima et al. 2017). Second, the increase of HL led to stronger shock for the matrix, which was also responsible for the decrease of removal rates (Bednarek et al. 2014). For NH$_4^+$-N and NO$_3^-$-N removal rates, the results were a consequence of competition between heterotrophic and autotrophic bacteria (Li et al. 2011). As mentioned before, the increase of HL resulted in higher organic loadings, which could shift in favor for heterotrophic bacteria rather than autotrophic bacteria contribution. Taking pollutants removal and N$_2$O emission into consideration, the recommended range for HL was 0.08–0.12 m$^3$/m$^2$·d.

**N$_2$O emission and conversion under variable pollutant loadings**

During the second experimental period, HL was fixed at 0.10 m$^3$/m$^2$·d. pollutant loading (PL) gradually increased from 3.2 to 3.7, 4.2, 4.7 and 5.2 g N/m$^2$·d. Water quality, N$_2$O production and conversion ratio were analyzed and shown in Figures 4(b) and 5.

Correlation analysis suggested that PL exerted significant negative influence on the removal of COD ($r = -0.976$, $P = 0.04$) and NH$_4^+$-N ($r = -0.994$, $P = 0.01$). Average NO$_3^-$-N removal rates increased from 86.4% to 99.1% with the increase in PL. Correlation analysis revealed that PL positively correlated with NO$_3^-$-N removal ($r = 0.990$ and $P = 0.01$).

Increased denitrification with PL probably resulted from the decreased oxygen diffusion into microsites and decreased oxygen concentration due to increased O$_2$ consumption (Pan et al. 2015). Zhu et al. (2015) studied the effect of nitrogen application on N$_2$O emission and suggested that external nitrogen source can increase N$_2$O emission and the high peaks of N$_2$O emission were observed during external nitrogen source application. By contrast, the results from this study showed that when PL grew as high as 4.2 g N/m$^2$·d, N$_2$O emission reached its maximum followed by a dramatic decrease. In addition, correlation analysis showed that under loading rates of low to medium level
(HL 0.08–0.16 m³/m²·d, PL 3.2–4.2 g N/m²·d), N₂O production rate showed a positive correlation with loadings. Instead, loading rates had significant negative effects on N₂O production when the loadings were in high ranges (P < 0.05). Variations of N₂O emission along with loading rates verified that N₂O production was the interaction of nitrification and denitrification process (Rajagopal & Béline 2011; Ak & Gunduz 2013; Hernandez-Paniagua et al. 2014).

Considering the pollutant (COD, NH₄⁺-N, NO₃⁻-N and TN) removal rates and N₂O generation rate, influent PL with ranges of 3.2–3.7 g N/m²·d was recommended. Under the optimal conditions (HL 0.08–0.12 m³/m²·d and PL 3.2–3.7 g N/m²·d), effluent quality could meet the Water Quality Standard for Scenic Environmental Use in China (GB/T 18921–2002, BOD₅ ≤ 10 mg/L, NH₄⁺-N ≤ 5 mg/L, TN ≤ 15 mg/L and TP ≤ 0.5 mg/L). Conversion ratio for nitrogen to N₂O was between 4.5% and 7.0%.

During the studied period, clogging did not occur owing to the proper construction and operation of the system. The results of infiltration rate indicated that HL and PL had negative effect on infiltration capacity. After some period of operation, SWI experienced a sufficient decline in the infiltration capacity under high loading rates. Especially in the 0.2–0.4 m depth interval, infiltration rates decreased gradually to 9.0 × 10⁻⁴ cm/s (60–80% of the initial value). This decline was attributed to the accumulation of biodegradable organic matter (measured by total solids, total suspended solids and BOD) at high loading rates. If the infiltration rate continues to decrease, the environmental conditions of upper layer would immediately begin to shift from aerobic to anaerobic, corresponding to the pollutant (COD, NH₄⁺-N, NO₃⁻-N and TN) removal rates decreasing and matrix clogging. Therefore, in order to extend the life-span and keep a reliable removal performance, field scale SWI should be operated under the optimized loadings. Moreover, water pH in the upper layers (0.2–0.4 m) declined from 7.2 (mean value of influent) to 6.9 (on average), indicating that, in this section, nitrification process played a dominant role in nitrogen cycle. Then, pH showed an increase to 7.1 (mean value of effluent) caused probably by denitrification process. As a result, pH and N₂O could be seen as indicative indexes for the nitrification and denitrification process.

Figure 5 | The variation of removal efficiency under different PLs.
N₂O emission under variable drying-wetting cycles

Table 1 describes pollutant removal efficiencies and effluent concentration under continuous and intermittent feeding modes. It is clear that under continuous operation (drying-wetting cycle 0), pollutant removals were relatively low, especially for COD and NH₄⁺-N. When the feeding days was extended, removal efficiency of NH₄⁺-N continuously declined to 88.4 ± 0.9% under drying-wetting cycle of 1:3. In contrast, TN removal efficiency increased with the declining of the drying-wetting cycle. The possible reasons may be: prolonged feeding days led to increased denitrification rates both by increasing the oxygen consumption making conditions more suitable for denitrification and providing organic carbon needed in the denitrification process.

When the running time was extended, N₂O emission showed periodic variations (Figure 6). During the wetting period (saturated conditions in lower layers), pollutants were removed due to the suspended solids deposition and bacterial growth in soil spaces. For one thing, prolonging the wetting

![Figure 6](https://iwaponline.com/wst/article-pdf/76/8/2158/449295/wst076082158.pdf)

**Figure 6** | N₂O production rate under variable drying-wetting cycles.

### Table 1  | Effluent quality under different drying-wetting cycles

<table>
<thead>
<tr>
<th>Drying-wetting cycle</th>
<th>COD Effluent mg/L</th>
<th>COD Removal %</th>
<th>BOD Effluent mg/L</th>
<th>BOD Removal %</th>
<th>NH₄⁺-N Effluent mg/L</th>
<th>NH₄⁺-N Removal %</th>
<th>NO₂⁻-N Effluent mg/L</th>
<th>NO₂⁻-N Removal %</th>
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</thead>
<tbody>
<tr>
<td>3:1</td>
<td>20.0 ± 8.0</td>
<td>93.3 ± 2.6</td>
<td>2.0 ± 1.0</td>
<td>99.1 ± 0.4</td>
<td>1.0 ± 0.2</td>
<td>95.3 ± 0.9</td>
<td>0.26 ± 0.05</td>
<td>13.3 ± 16.7</td>
</tr>
<tr>
<td>2:1</td>
<td>14.0 ± 4.0</td>
<td>95.3 ± 1.3</td>
<td>5.0 ± 1.0</td>
<td>97.8 ± 0.4</td>
<td>1.0 ± 0.15</td>
<td>95.3 ± 0.7</td>
<td>0.27 ± 0.07</td>
<td>10.0 ± 23.3</td>
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<td>1:1</td>
<td>26.0 ± 5.0</td>
<td>91.3 ± 1.6</td>
<td>5.0 ± 1.0</td>
<td>97.8 ± 0.4</td>
<td>1.8 ± 0.2</td>
<td>91.6 ± 0.9</td>
<td>0.48 ± 0.03</td>
<td>60.0 ± 10.0</td>
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<td>1:2</td>
<td>32.0 ± 6.0</td>
<td>89.5 ± 2.0</td>
<td>9.0 ± 2.0</td>
<td>96.1 ± 0.9</td>
<td>2.0 ± 0.3</td>
<td>90.7 ± 1.4</td>
<td>0.42 ± 0.04</td>
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<td>1:3</td>
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<td>85.7 ± 1.0</td>
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<td>94.8 ± 0.4</td>
<td>2.5 ± 0.2</td>
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<tr>
<td>0</td>
<td>60.0 ± 2.0</td>
<td>80.0 ± 0.7</td>
<td>15.0 ± 2.0</td>
<td>93.5 ± 0.9</td>
<td>5.0 ± 0.15</td>
<td>76.7 ± 0.7</td>
<td>0.05 ± 0.05</td>
<td>83.3 ± 16.7</td>
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</table>

<table>
<thead>
<tr>
<th>Drying-wetting cycle</th>
<th>NO₂⁻-N Effluent mg/L</th>
<th>NO₂⁻-N Removal %</th>
<th>TN Effluent mg/L</th>
<th>TN Removal %</th>
<th>TP Effluent mg/L</th>
<th>TP Removal %</th>
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</thead>
<tbody>
<tr>
<td>3:1</td>
<td>1.5 ± 0.10</td>
<td>50.0 ± 3.3</td>
<td>4.8 ± 0.3</td>
<td>80.8 ± 1.2</td>
<td>0.0 ± 0.0</td>
<td>100.0 ± 0.0</td>
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<tr>
<td>2:1</td>
<td>1.2 ± 0.08</td>
<td>60.0 ± 2.7</td>
<td>4.3 ± 0.3</td>
<td>83.2 ± 1.2</td>
<td>0.0 ± 0.0</td>
<td>100.0 ± 0.0</td>
</tr>
<tr>
<td>1:1</td>
<td>1.0 ± 0.05</td>
<td>66.7 ± 1.7</td>
<td>2.8 ± 0.2</td>
<td>88.8 ± 0.8</td>
<td>0.2 ± 0.1</td>
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<td>1:2</td>
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<td>88.6 ± 0.4</td>
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<td>1:3</td>
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<td>66.7 ± 1.4</td>
<td>2.5 ± 0.4</td>
<td>89.6 ± 1.6</td>
<td>0.2 ± 0.1</td>
<td>95.0 ± 2.5</td>
</tr>
<tr>
<td>0</td>
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<td>6.4 ± 0.3</td>
<td>74.4 ± 1.2</td>
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<td>95.0 ± 2.5</td>
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period prevents the penetration of oxygen into the soil matrix, promoting denitrification smooth processes (as shown in Table 1, NO₂⁻, NO₃⁻ and TN removal rates increased by prolonging the wetting days). In addition, a longer wetting period ensured longer residence times, which, in essence, were observed to be a crucial factor in the removal of more complex organic matter present in water (Rassamee et al. 2011). Hence, lower drying-wetting cycle (0, 1:3 and 1:2) showed relatively stable N₂O emission rate.

By contrast, intermittent operation rather than continuous feeding is an encouraging method to ensure the dissolved oxygen availability for the growth of the nitrifying bacteria, especially in upper layers. Furthermore, periodic resting is a passive method for removing the microbial metabolites and restoring the hydraulic capacity and oxygen reduction potential (Desloover et al. 2011; Tsushima et al. 2014). Thus, intermittent operation was favorable for nitrifying bacteria growth and accumulation of NO₃⁻ (Table 1), which was ready to be denitrified to N₂O. However, when the drying time was extended, a more easily biodegradable portion of organic matter was quickly consumed by microorganisms in the first 10 cm of the layer where oxygen levels peaked (Li et al. 2011). Consequently, denitrification process would be suspended resulting from the absence of a carbon source.

The optimal drying-wetting cycle from the point of view of maximum infiltration along with the desired objective of quality improvement in the wastewater was found to be 12 h of flooding followed by 12 h of drying, i.e. drying-wetting cycle 1:1.

CONCLUSIONS

Influent loadings and drying-wetting regimes impacted N₂O generation and conversion by affecting HRT and nitrogen circulation indirectly. N₂O conversion ratio decreased with an increase in HL (decreased from 6.6 ± 0.1% to 2.7 ± 0.1% when HL increased from 0.08 to 0.24 m³/m²·d). N₂O conversion rate increased with an increase in PL up to 8.2 ± 0.5% followed by a decrease to 3.8 ± 0.2%. Optimal drying-wetting cycle from the point view of maximum infiltration along with the desired objective of quality improvement in the wastewater was found to be 12 h of flooding followed by 12 h of drying. In order to abate N₂O emission as well as improve effluent quality, optimal HL and PL were suggested as ranges of 0.08–0.12 m³/m²·d and 3.2–3.7 g N/m²·d, respectively. Under these conditions, conversion rate for influent nitrogen to N₂O would be between 4.5% and 7.0%.

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