International Biodeterioration & Biodegradation 133 (2018) 116–123

Contents lists available at ScienceDirect

International Biodeterioration & Biodegradation

journal homepage: www.elsevier.com/locate/ibiod

Developing a mathematical modeling method for determining the potential rates of microbial ammonia oxidation and nitrite oxidation in environmental samples

Yiguo Hong[a,*], Yan Wang[b], Jiapeng Wu[b,c], Lijing Jiao[b,c], Xiang He[a], Xiaomei Wen[a], Hongguo Zhang[a], Xiangyang Chang[a]

[a] School of Environmental Science and Engineering, University of Guangzhou, Guangzhou, 510006, PR China
[b] State Key Laboratory of Tropical Oceanography (LTO), South China Sea Institute of Oceanology, Chinese Academy of Sciences, Guangzhou, 510301, PR China
[c] University of Chinese Academy of Sciences, Beijing, 100049, PR China

ARTICLE INFO

Keywords:
Nitrification
Ammonia oxidation
Nitrite oxidation
Mathematical modeling
Instantaneous rate
Average rate

ABSTRACT

This study report a new method for simultaneously determining microbial ammonia oxidation rate (V_a) and nitrite oxidation rate (V_n) in water and sediment. This method is based on mathematical modeling analysis for the time series variation of DIN (nitrite, nitrate, and ammonium) in an incubation system without an inhibitor. Compared to the traditional methods with specific inhibitors, this method is not affected by inhibitors and can determine both instantaneous and average V_a and V_n accurately and simultaneously. Practical application indicated that this method can well determine the dynamic variations in V_a and V_n in both water and sediment samples.

1. Introduction

Nitrification, the biological oxidation of ammonia or ammonium to nitrite followed by the oxidation of the nitrite to nitrate, plays a key role in the global nitrogen cycle (Klotz and Stein, 2008; Lehtovirta-Morley et al., 2013; Ward et al., 2007), linking organic matter mineralization to microbial denitrification and anammox in water, soil, and sediment (Martens-Habbena et al., 2015). Nitrification in agricultural systems can lead to fertilizer loss (Gubry-Rangin et al., 2010) and nitrogen pollution in receiving waters. In wastewater treatment, nitrification is a critical process and a rate-limiting step (Cortés-Lorenzo et al., 2015) in nitrogen removal (Wang et al., 2015) that is linked to denitrification and fixed nitrogen loss and also has an important climate effect due to nitrous oxide emission (Smith et al., 2016), i.e., the release of a potent greenhouse gas to the atmosphere (Freing et al., 2012). In the ocean, significant nitrification may take place in the euphotic zone (Feliatra and Bianchi, 1993; Peng et al., 2016) and supply substrates for denitrification directly.

Since the discovery of autotrophic nitrifying bacteria about 100 years ago, nitrification has always been regarded as a two-step separated process wherein ammonium is first oxidized to nitrite by ammonia-oxidizing bacteria (AOB), then the nitrite is oxidized to nitrate by nitrite-oxidizing bacteria (NOB) (Ward et al., 2011). Two discoveries in the last 15 years have changed our traditional views regarding nitrification. First is the description of ammonia-oxidizing archaea (AOA), of which *Nitrosopmilus maritimus* strain SCMl was the first species isolated in pure culture (Konneke et al., 2005), which changed the traditional understanding of the ammonia oxidation process. Numerous studies have suggested that AOA play a significant role in the global nitrogen cycle (Biller et al., 2012; Meng et al., 2017; Meng et al., 2016; Stahl and de la Torre, 2012; Zhang et al., 2012), as evidenced by their numerical dominance as ammonia oxidizers in many cases, greatly exceeding the abundance of AOB in the ocean (Löscher et al., 2012), and their more widespread presence than AOB in sediments (Zheng et al., 2013) and soils (Leininger et al., 2006). Moreover, genome analysis revealed that strain SCM1 primarily utilizes copper-dependent mechanisms of electron transfer, which are distinctly different from the iron-based bacterial mechanisms for ammonia oxidation (Walker et al., 2010). A putative nitroxyl oxidoreductase (NXOR) and a copper hydroxylamine oxidoreductase (CuHAO) (Schalk et al., 2000) were proposed as potentially important electron transport components. Although AOA and AOB can grow chemolithoautotrophically on ammonium, AOA use the 3-hydroxypropionate/4-hydroxybutyrate pathway for carbon fixing instead of the Calvin cycle used by AOB (Stahl and de la Torre, 2012). The second important breakthrough concerning microbial nitrification is the recent pioneering studies.

* Corresponding author. School of Environmental Science and Engineering, University of Guangzhou, Guangzhou, 510006, PR China.
E-mail address: yghong@gzhu.edu.cn (Y. Hong).

https://doi.org/10.1016/j.ibiod.2018.06.021
Received 31 May 2018; Received in revised form 24 June 2018; Accepted 25 June 2018
0964-8305/ © 2018 Published by Elsevier Ltd.

(Daims et al., 2015; van Kessel et al., 2015). Via cultivation, a completely nitrifying bacterium affiliated to the genus *Nitrospira* was discovered, which can completely oxidize ammonia to nitrate (Comammox) and has a competitive advantage in biofilms or other microbial aggregates with low substrate concentration (Kessel et al., 2015). The genome of the *Nitrospira* bacterium encodes the pathways for both ammonia and nitrite oxidation, and the genes for ammonia monooxygenase and hydroxylamine dehydrogenase of this bacterium are phylogenetically distinct from those identified from cultures and environmental samples.

At present, as no appropriate method exists for direct measurement of nitrification rates in situ, potential nitrification rates are generally determined using incubation system. Ward reviewed the methods for measuring nitrification rates in water and sediment (Ward, 2008). In general, methods with inhibitors have been commonly used for the measurement of nitrification rates. At present, several inhibitors, including acetylene (C_2H_2), nitrapyrin, and allylthiourea (ATU), have been applied to act upon the ammonia-oxidizing bacteria. Inhibition of ammonia oxidation by C_2H_2 was first used in soil systems (Henriksen, 1980; Walter et al., 1979), but C_2H_2 also inhibits denitrification (Kester et al., 1996; Yoshinari and Knowles, 1976). Nitrapyrin was a relatively common ammonia oxidation inhibitor used in sediment (Henriksen, 1980), but cannot dissolve in water and thus must be dissolved in a solvent, which would affect the microbial activities (Owens, 1986). Compared to the nitrapyrin and C_2H_2, ATU was the most persistent inhibitor over the maximum incubation time of 2.5 days (Hall, 1984). Until AOA were discovered, ATU has been always used as the only inhibitor to block the ammonia oxidation process because experiments showed several AOB pure culture strains were indeed sensitive to ATU (Li et al., 2015). However, experiments indicated that AOA were insensitive to ATU (Clark et al., 2007). Thus, if AOA play more important roles than AOB in samples, ATU would not inhibit the ammonia oxidation process completely and the nitrification rate could not be determined accurately (Walker et al., 2010). Recent studies showed that ammonia oxidation by AOA could be inhibited by 2-phenyl-4,4,5,5,-tetramethylimidazoline-1-oxyl-3-oxide (PTIO) (Martens-Habbena et al., 2015; Peng et al., 2015). Concerning the Comammox process, no study to date has reported on how to block it. Therefore, the discoveries of these new types of nitrifiers led to new challenges in the determination of nitrification rates with inhibitors. Furthermore, greater deviation may come from uncoupling the successive pathways of inhibitors.

Similar to the inhibitor methods, isotopic methods also have some disadvantages in determining nitrification rates. Although the isotopic method of using a labeled $^{15}N\text{-}NH_4^+$ isotope tracer in the samples is relatively more accurate than those using inhibitors, adding the $^{15}N\text{-}NH_4^+$ isotope tracer into sediment or suspension will weaken the isotope's signal or cause it to disappear owing to adsorption by the sediment (Dai et al., 2008), and in the process of cultivation of water or sediment samples, the isotope may gradually be released, leading to possible sources for experimental errors Furthermore, the rates of ammonia oxidation and nitrite oxidation were difficult to determine separately and simultaneously because the ^{15}N-nitrite and ^{15}N-nitrate were barely distinguishable via isotopic mass spectrometry.

In this paper, a complete description a protocol for the simultaneous analysis of ammonia oxidation and nitrite oxidation was provided via building a mathematical model independent of inhibitors, as well as background and additional information relevant to the application of this method. The model was validated through practical application, and procedures are proposed for application in future microbial nitrogen cycle studies.

2. Materials and methods

2.1. Sample collection and incubation setup

Samples used in this study were obtained from the Pearl River, the Pearl River Estuary and Daya Bay. Among them, the water sample obtained from the Pearl River (23°6′22.80″N 113°17′39.50″E, water depth = 4 m, salinity = 0 psμ) and the surface sediment sample collected in Daya Bay (114°57.4′N, 22°60.78′E, water depth = 15 m, salinity = 31.2 psμ) were used to build the mathematical model for determining the nitrification rate. The water and sediment samples collected from the Pearl River Estuary at five stations (P_1, P_3, P_7, P_{13}, and P_{18}, Fig. S1) along the mainstream were used in practical verification of the new method developed in this study. Water samples were collected using a Plexiglas water-harvesting device, and sediment samples were collected via an undisturbed box dredging device. All the samples were sealed and stored at 4 °C using sterile plastic bags and transported to the laboratory on ice within 4 h.

The incubations were set up with 250 mL sterile wide-mouth bottles (HDPE, Thermo). For the water sample, 125 mL in situ water (filtered by nylon mesh) was injected into each bottle. For the sediment samples, 10 g (wet weight) sediment mixed with 125 mL filtered in situ water (with a 0.22 μm filter) was added into each bottle. The headspace in the incubation bottle can ensure sufficient oxygen for nitrification. Then, prepared bottles with water or sediment samples were incubated at in situ temperature with gentle shaking (75 rpm) to maintain them in an aerated state. The samples were incubated in the dark to prevent DIN species assimilation by phytoplankton. The water or overlaying water in the incubations was sampled every 12 h and measured for ammonia, nitrate, and nitrate (DIN) concentrations. Incubations were sustained until the ammonium and nitrite were completely consumed and the nitrate concentration stabilized. The concentration of NO_2^- was quantified colorimetrically by adding Greiss reagents (Griess, 1879) and NO_3^- was measured by converting nitrate to nitrite with cadmium-copper reduction method followed by nitrite detection with Greiss reagents (Wu et al., 2016). NH_4^+ was determined by indophenol blue method (Aminot et al., 1997). The time series of DIN species can be used for further establishing the mathematical model of the nitrification reaction. To compare the difference between our mathematical modeling method and traditional inhibitor methods, we also designed incubations with the inhibitors ATU, PTIO or $NaClO_3$. These incubations were also monitored for their variation in DIN concentration.

2.2. Nitrification rate modeling

Simple box modeling (Fig. 1) was used to simultaneously calculate the rates of ammonia oxidation (Va) and nitrite oxidation (Vn), depending on the concentration variation of DIN species. The model was built based on two assumed conditions. First, the ammonium is unconstrained in the incubation system, allowing it to be produced via organic mineralization. Second, the incubation remains in an aerobic state. Thus, denitrification and Anammox processes would remain inactive, so no nitrogen would be lost from the incubation system. Under these conditions, nitrate should be the product of nitrification in the incubation system. Va can be expressed by the increment of the total concentration of nitrite and nitrate ($Va = \Delta[NO_x^-]/\Delta T$) per unit time, and Vn can be expressed by the increment of the nitrate concentration in the system per unit time ($Vn = \Delta[NO_3^-]/\Delta T$).

The DIN species point data were empirically smoothed with a second-order local smoothing filter to minimize the higher frequency noise resulting from sampling. Then, the DIN time series data were fitted reasonable well by such a sigmoidal curve (Boltzmann distribution pattern) and obtained using origin software with Eq. (1). A sigmoid function is a mathematical function having a characteristic “S"-shaped curve or sigmoid curve, which is often used to exhibit a natural processes progression from small beginnings that accelerates and approaches a climax over time (Gibbs and MacKay, 2000).

$$Y = Y_{max} + \frac{(Y_0 - Y_{max})}{1 + e^{(x-x_0)/dx}} \quad (1)$$

where Y, Y_0, and Y_{max} are the calculated, initial, and final

Download English Version:

https://daneshyari.com/en/article/8843693

Download Persian Version:

https://daneshyari.com/article/8843693

Daneshyari.com