



Effect of plant species on nitrogen recovery in aquaponics



Zhen Hu^a, Jae Woo Lee^b, Kartik Chandran^c, Sungpyo Kim^b, Ariane Coelho Brotto^c, Samir Kumar Khanal^{d,*}

^aShandong Key Laboratory of Water Pollution Control and Resource Reuse, School of Environmental Science and Engineering, Shandong University, Jinan 250100, China

^bDepartment of Environmental Engineering, College of Science and Technology, Korea University, Sejong-ro 2511, Sejong 339-700, South Korea

^cDepartment of Earth and Environmental Engineering, Columbia University, 500 West 120th Street, New York, NY 10027, USA

^dDepartment of Molecular Biosciences and Bioengineering, University of Hawaii at Manoa, Honolulu, HI 96822, USA

HIGHLIGHTS

- Plant species had significant influence on nitrogen transformations in aquaponics.
- Aquaponics could be important anthropogenic N₂O emission source.
- High root surface area in aquaponics favored the growth of nitrifying bacteria.

ARTICLE INFO

Article history:

Received 13 November 2014

Received in revised form 3 January 2015

Accepted 6 January 2015

Available online 20 January 2015

Keywords:

Aquaponics

Nitrogen transformations

Plant species

Nitrous oxide

Ammonia oxidizing bacteria

ABSTRACT

Nitrogen transformations in aquaponics with different edible plant species, i.e., tomato (*Lycopersicon esculentum*) and pak choi (*Brassica campestris L. subsp. chinensis*) were systematically examined and compared. Results showed that nitrogen utilization efficiencies (NUE) of tomato- and pak choi-based aquaponic systems were 41.3% and 34.4%, respectively. The abundance of nitrifying bacteria in tomato-based aquaponics was 4.2-folds higher than that in pak choi-based aquaponics, primarily due to its higher root surface area. In addition, tomato-based aquaponics had better water quality than that of pak choi-based aquaponics. About 1.5–1.9% of nitrogen input were emitted to atmosphere as nitrous oxide (N₂O) in tomato- and pak choi-based aquaponic systems, respectively, suggesting that aquaponics is a potential anthropogenic source of N₂O emission. Overall, this is the first intensive study that examined the role plant species played in aquaponics, which could provide new strategy in designing and operating an aquaponic system.

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1. Introduction

During 2000–2012, global aquaculture production expanded at an average annual rate of 6.2%, with world aquaculture production of 66.7 million tons in 2012, outpacing world population growth rate of 1.6%. It is estimated that aquaculture would account for 62% of world's fish supply for human consumption by 2030 (FAO, 2014). The large-scale application of aquaculture, however, is restricted by land and water utilization as well as by environmental concerns (Islam, 2005; Schwitzguébel and Wang, 2007). Thus, the further expansion of aquaculture now depends on development and application of new technologies to intensify fish cultivation while maximizing water and nutrients reuse, and minimizing environmental impacts. Aquaponics, which is an integrated system

that links recirculating aquaculture with hydroponic production, is considered to be an innovative and sustainable solution (Tyson et al., 2011). A well-managed aquaponics could improve nutrient retention efficiency, reduce water usage and waste (mostly nutrients) discharge to the environment, and improve profitability by simultaneously producing two cash crops (Diver, 2006; Tyson et al., 2011). It is generally believed that aquaponics, with concomitant nutrients recovery, will become one of the widely accepted methods of sustainable food production in the near future (Hu et al., 2012).

One major advantage of aquaponics is its high nitrogen utilization efficiency (NUE). Nitrogen is a vital element for all living organisms, and protein-rich fish feed which is the major source of nitrogen for fish cultivation, representing 50–70% of fish production costs (Valente et al., 2011). In aquaculture system, only about 25% of the nitrogen input is harvested through fish biomass, and over 70% is excreted into the surrounding environment in the form

* Corresponding author. Tel.: +1 808 956 3812; fax: +1 808 956 3542.

E-mail address: khanal@hawaii.edu (S.K. Khanal).

of ammonia (Hargreaves, 1998). To maintain a good water quality for fish growth, the fish cultivation water which is rich of nitrogen compounds, has to be exchanged with fresh water at a recirculation rate of 5–10% per day. This is a waste of natural resource. Furthermore, the nitrogen-rich effluent which is discharged to the surrounding ecosystem may lead to eutrophication and other environmental concerns. In aquaponics, an interesting nitrogen cycle is established to avoid the above mentioned problems. The cycle begins with the introduction of protein in fish feed, which is ingested by fish and then excreted to the aqueous phase in the form of total ammonia nitrogen (TAN, i.e., NH_3 and NH_4^+). Ammonia is firstly oxidized to nitrite (NO_2^-) by ammonia oxidizing bacteria (AOB), and then converted to nitrate (NO_3^-) by nitrite oxidizing bacteria (NOB, mainly *Nitrobacter* spp. and *Nitrospira* spp.). Instead of being discharged to the surrounding ecosystem, NO_3^- and residual TAN and NO_2^- could be taken up by plants. Thus, two crops (plants and fish) are harvested from the system by utilizing nutrient in a sustainable way. Although a wide variety of aquaponics designs have been examined over the past several decades; most of the studies were focused on the improvement of cash crop yield (Lennard and Leonard, 2006; Sace and Fitzsimmons, 2013). An in-depth study on investigation of nitrogen transformations in aquaponics is still lacking.

Nitrifying bacteria, which convert NH_4^+ to NO_3^- through nitrification, play an important role in efficient operation of aquaponics. During the nitrification process, nitrous oxide (N_2O) emission also takes place (Stief et al., 2009; Beaulieu et al., 2011). In addition, there exist anoxic zones where denitrification could occur. N_2O is an inevitable intermediate product of denitrification. N_2O is considered to be an important greenhouse gas with the global warming potential 296 times that of CO_2 (IPCC, 2007). Evidences have shown that aquaculture could be a notable source of anthropogenic N_2O emissions (Hu et al., 2013). Compared with conventional recirculating aquaculture system, Sfetcu et al. (2008) observed lower NH_4^+ , NO_2^- and NO_3^- concentrations in an aquaponic system, which might lead to lower N_2O emission (Ahn et al., 2010). More thorough study is needed to further examine this.

Both leafy plant (e.g., lettuce, spinach, and pak choi) and fruity plant (e.g., tomato and cucumber) can be used in aquaponics (Lennard and Leonard, 2006; Sace and Fitzsimmons, 2013). To date, the selection of plant species relies mainly on experience, depending on stocking density of fish tanks and subsequent nutrient concentrations of aquaculture effluent (Diver, 2006). Since different plants have different growth characteristics and nitrogen utilization capacity, nitrogen transformations in aquaponics can be directly influenced by plant species (Jin et al., 2010). In addition, the growth of nitrifying bacteria requires sufficient surface area where the nitrifies can develop a coating or shield of exopolymeric substances (EPS) that protects them. Thus, plants with larger root surface area might have unique merit in an aquaponic system. However, very limited studies have been conducted to systematically investigate the effect of plant species on the performance of an aquaponic system.

In this study, nitrogen transformations and N_2O emissions in aquaponics were investigated. Fish production, plant growth and water quality were measured and their dependence on plant species were examined. Furthermore, molecular techniques were employed to identify and quantify the abundance of microbes responsible for nitrogen transformations in the aquaponics.

2. Methods

2.1. Experimental setup and operation

Experiments were conducted in the greenhouse at Mauka campus of University of Hawaii at Manoa (21°18'22"N, 157°48'32"W).

Two aquaponics were operated side by side for nearly 5 months. For each aquaponics, 300-L oval plastic tanks filled with 200 L water, was operated as aquaculture unit. The tank was stocked with high density (30 kg/m³) of tilapia fish (*Oreochromis niloticus*), which were obtained from Windward Community College (Honolulu, Hawaii, USA) and had been cultivated in conventional aquaculture system for 4 months prior to the start of the experiment. Fish were fed once per day and the feeding amount was determined based on fish response, according to the method described in Casillas-Hernández et al. (2006). Air pump was used to provide sufficient oxygen for fish growth by aerating the tank water and dissolved oxygen (DO) concentrations were maintained above 5 mg/L. The tank was covered with wooden board to prevent algal growth.

Peristaltic pump was used to pump the fish tank water to a clarifier, which was built using 20-L sealed bucket filled with biomedica (Kaldnes @ media, Aquatic Eco-System, Apopka, FL, USA). The clarifier captured majority of the suspended solids from aquaculture tank to protect the plant roots in the grow bed. After passing through the clarifier, tank water flowed into the grow bed. Floating raft hydroponic system was employed in this study. Rectangle plastic tank with an effective volume of 400 L was used as the grow bed and plants were held up by a foam raft that floats on the water. Shade cloth (50%) was installed over the grow bed to prevent excessive sun exposure to the plants. Two different plant species, tomato (*Lycopersicon esculentum*), a fruity plant and pak choi (*Brassica campestris* L. subsp. *chinensis*), a leafy plant, were used simultaneously in different aquaponics. The plant seeds were germinated according to method in Ako and Baker (2009). After 2 weeks, healthy plant seedlings were transplanted to the grow bed at their optimum planting density as per the seed supplier's guidelines. The plant density of tomato and pak choi were 13 and 20 plants/m², respectively. Nutrients present in aquaculture effluent were absorbed by plants in grow bed and the reclaimed water was then recirculated into the fish tank. For schematic diagram of the aquaponics, please refer to Fig. S1 (see Supporting information).

The water was recirculated continuously between fish tank and grow bed in aquaponics and no water exchange was conducted during the study period except for replenishing evapotranspiration losses. Calcium hydroxide and potassium hydroxide were periodically dosed into the aquaculture tank to maintain the pH at a neutral range. Since fish water is deficient in iron, Fe-EDTA was added to the system on a weekly basis to maintain Fe^{2+} concentration above 2 mg/L (Rakocy et al., 2006).

2.2. Sampling and analytical methods

Water samples were obtained from fish tank every other day after feeding, and were analyzed immediately for TAN, NO_2^- , and NO_3^- concentrations, using HACH reaction kits (Loveland, CO, USA), namely Ammonia TNTplus (TNT 830), Nitrite TNTplus (TNT 839), and Nitrate TNTplus (TNT 836), respectively. DO concentrations, temperature, and pH were measured *in situ* daily using the HQ40d Portable Water Quality Lab Package (HACH, Loveland, CO, USA). The total suspended solids (TSS) concentration was determined according to the Standard Methods (APHA, 2005). Daily fish feed consumption was recorded; fish and plant biomass increase were accurately weighed using analytical balance. At the end of the experiment, samples of fish, plants, fish feed, and suspended solids (microbial biomass) were dried, and their total nitrogen contents were determined by using LECO TruSpec C/N analyzer (LECO Corp., St. Joseph, MI, USA). Plant root surface area was determined using WinRHIZO root-scanning methods (WinRhizo Pro v.2005b, Régent Instruments, Québec, Canada) (Pang et al., 2011).

Diurnal variations of N_2O emission from aquaculture tank was measured every other week after stable nitrite concentration was

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