

# Effect of high ammonia loads emitted from poultry-manure digestion on nitrification activity and nitrifier-community structure in a compost biofilter



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## ABSTRACT

Ammonia emissions from poultry-manure disposal and agricultural applications pose a global environmental challenge that requires the development of proper management practices. Recently, a scheme comprised of a compost-based biofilter for the treatment of high loads of ammonia emitted from poultry-manure digestion (up to  $500 \text{ g NH}_3 \text{ m}^{-3} \text{ biofilter day}^{-1}$ ) was suggested. In this study, we hypothesized that the high ammonia-adsorption capacity of the compost matrix creates an ammonia gradient along the biofilter that is occupied by adapted nitrifying communities, thus allowing high nitrification rates and ammonia removal. Accordingly, pilot-scale compost biofilters were constructed and batch-fed with ammonia emitted from digested poultry manure for over a year. The operation cycle included a nitrate washing step from the biofilter. Compost samples withdrawn at 20, 40 and 60 cm distance from the gas inlet were chemically characterized and analyzed for nitrification activity and nitrifier abundance and diversity. The number of ammonia-oxidizing bacteria (AOB) was 0.5–1 order of magnitude lower in the bottom section (0–20 cm), which was dominated by *Nitrosomonas* species, compared to the top layers which were occupied by a mixed *Nitrosomonas* and *Nitrospira* population. In addition, ammonia-oxidizing archaea (AOA) were homogeneously distributed along the biofilter profile and their numbers were at least one order of magnitude higher than those of the AOB. Significantly lower potential nitrification activity was detected in the bottom layer, and correlated with AOB abundance. Together, the results indicate that with proper operation of compost biofilters, suitable communities of nitrifying microorganisms that are able to cope with a wide range of ammonia concentrations, sourced from manure digestion, will develop.

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## 1. Introduction

Agro-biowaste, such as animal manure, poses global environmental challenges, necessitating the development of proper management schemes for safe disposal. Among animal wastes, poultry manure contributes significantly to the emission of gaseous ammonia ( $\text{NH}_3$ ) to the atmosphere as a by-product of the microbial decomposition of proteins, amino acids, uric acid and other organic compounds (Lahav et al., 2008; Ni et al., 2010). Biofiltration using a mixture of substances as packing materials (e.g. compost, peat, sludge, woodchips, gravel, sand, etc.) has been reported to efficiently remove  $\text{NH}_3$  by adsorption and nitrification, even at

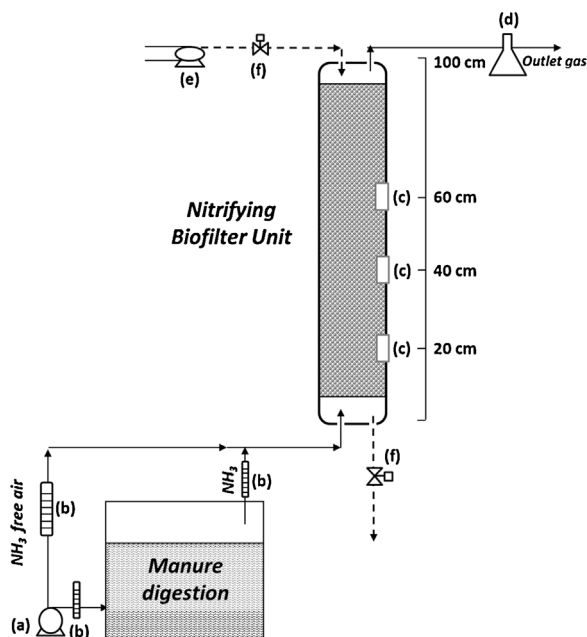
concentrations of up to  $550 \text{ mg NH}_3 \text{ m}^{-3}$  (Gregory et al., 2010; Jun and Wenfeng, 2009; Smet et al., 2000; Wang et al., 2013).

The first and rate-limiting step of nitrification is the oxidation of  $\text{NH}_3$  to nitrite ( $\text{NO}_2^-$ ) (Kowalchuk and Stephen, 2001), and it may determine the progression of the biofiltration process (Jun and Wenfeng, 2009). Moreover, it is now well established that  $\text{NH}_3$  oxidation is performed by both bacterial and archaeal species which have different sensitivities to  $\text{NH}_3$  concentrations and pH (Hatzenpichler, 2012; Prosser and Nicol, 2012). The presence of ammonia-oxidizing bacteria (AOB) and ammonia-oxidizing archaea (AOA) in composting material such as swine, poultry and cattle manure has been previously reported (Kowalchuk et al., 1997; Yamamoto et al., 2010). Furthermore, Jun and Wenfeng (2009) showed the dominance of *Nitrospira* spp. over *Nitrosomonas* spp. in a compost biofilter receiving low concentrations of  $\text{NH}_3$  ( $20\text{--}100 \text{ mg NH}_3 \text{ m}^{-3}$ ).

A significant reduction in nitrification efficiency by compost biofilters receiving inlet concentrations of over  $110 \text{ mg NH}_3 \text{ m}^{-3}$

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**Fig. 1.** Schematic drawing of the compost biofilter: (a) blower; (b) flow meter and controller; (c) filter-bed sampling ports; (d) ammonia trap; (e) peristaltic water pump; (f) normally closed water valve.

was demonstrated by Chen et al. (2005). Recently, a scheme that includes a compost biofilter for the capture of  $\text{NH}_3$  emitted during manure digestion and its conversion to nitrate ( $\text{NO}_3^-$ ) has been suggested (Gross et al., 2012; Posmanik et al., 2013). The manure is added in batches, and the produced  $\text{NO}_3^-$  is washed away after each cycle, thus regenerating the filter bed and improving its  $\text{NH}_3$ -removal efficiency. The produced  $\text{NO}_3^-$ -rich effluent can be applied as a liquid fertilizer in agricultural applications. Additional outputs of the suggested biofilter are effluent gases that have been characterized to be free of  $\text{NH}_3$  and other hazardous gases such as hydrogen sulfide ( $\text{H}_2\text{S}$ ), methane ( $\text{CH}_4$ ) and nitrous oxide ( $\text{N}_2\text{O}$ ) (Posmanik et al., 2013). This system exhibited high nitrification activity under very high  $\text{NH}_3$  concentrations ( $>1700 \text{ mg m}^{-3}$ ) emitted from digested manure. Based on this, we hypothesized that these high nitrification rates, despite the high  $\text{NH}_3$  loads, were made possible by the heterogeneous distribution (according to  $\text{NH}_3$  gradient) of the nitrifying microorganisms (abundance and community structure) in the biofilter layers. Nitrification activity, abundance and community structure of dominant AOB, AOA and nitrite-oxidizing bacteria (NOB) were therefore characterized in the different layers of a compost biofilter receiving high  $\text{NH}_3$  loads from digested manure.

## 2. Materials and methods

### 2.1. Experimental setup and sampling

A pilot-scale anaerobic manure digester (as a source of  $\text{NH}_3$ ) attached to a nitrifying compost biofilter was used as detailed by Posmanik et al. (2013) and illustrated in Fig. 1. Briefly, the manure digester consisted of a 120-L tank (Netafim, Hatzerim, Israel) into which fresh poultry manure was introduced at a 1:10 (w/w) manure-to-water ratio. The resulting slurry was mixed for 10 days by gentle air jet which also carried the  $\text{NH}_3$  produced during manure digestion into the attached nitrifying biofilter (Fig. 1). To enhance  $\text{NH}_3$  emission from the digester into the biofilter, the slurry pH was gradually increased to 10 by addition of lime after 7

days of digestion. After 10 days the digester was empty and another batch of poultry manure was introduced. Overall, the system was operated through this cycle for 12 months. Operational conditions,  $\text{NH}_3$  load and concentration into the nitrifying biofilters (duplicates) are summarized in Table 1. Each biofilter was constructed from a 1-m long opaque PVC column with an inner diameter of 200 mm, packed with mature dairy-manure compost (Tuff Merom-Golan, Merom-Golan, Israel) and plastic beads (10 mm diameter, Aridal bio-balls, Kefar Hasidim, Israel) serving as both a biomass carrier and a nutrient source. Densities of the compost and plastic beads were  $250$  and  $160 \text{ kg m}^{-3}$ , respectively. Three sampling ports were located along the biofilter to allow sampling at 20, 40 and 60 cm distance from the gas inlet (Fig. 1). Filter-bed samples (20 g) were collected from each sampling port three times during the year (6, 8 and 10 months from initiation of the experiment). Samples were sieved and homogenized before analyses. Subsamples were taken for chemical characterization, and the rest was stored frozen ( $-20^\circ\text{C}$ ) for later extraction of total genomic DNA.

### 2.2. Profile of gaseous $\text{NH}_3$ in the biofilter

The profile of gaseous  $\text{NH}_3$  along the filter bed was quantified by introducing a constant  $\text{NH}_3$  flow ( $3750 \text{ mg NH}_3 \text{ m}^{-3}$ ) from a cylinder. The  $\text{NH}_3$  flow was continuously supplied for 24 h at the bottom of the biofilter (duplicated) and routine gas sampling was performed from the three sampling ports (20, 40, and 60 cm from the gas inlet). This experiment was done at the end of the 12 months of biofilter operation. To reach the required concentration, 0.5 L of  $\text{NH}_3$  from a concentrated cylinder (10%  $\text{NH}_3$  in  $\text{N}_2$ , Maxima Air Separation Center, Israel) was mixed with a jet of  $\text{NH}_3$ -free humid air ( $10 \text{ L air min}^{-1}$ ). Gas samples (30 ml) were collected by a gas-tight syringe and introduced into sealed 40-ml vials filled with 10 ml of 1 N HCl. Vials were mixed, allowing capture of the  $\text{NH}_3$  by the acid. The HCl was then analyzed for total N by a multi N/C 2100s analyzer (Analytic-Jena, Jena, Germany). Finally, the ratios between the outlet  $\text{NH}_3$  concentration of each sampling port and its inlet concentration ( $C/C_0$ ) were calculated and plotted over time.

### 2.3. Chemical analyses

Chemical analyses of the compost followed standard methods (Soil and Plant Analysis Council Inc., 1999). Total Kjeldahl N (TKN) was determined by the Kjeldahl method and inorganic N species were determined following extraction of 4 g air-dried compost in 20 ml 1 M KCl. The filtered extract was then analyzed for total ammonia N (TAN) by the Nessler method (APHA, 1989),  $\text{NO}_3^-$  by the second-derivative method, and  $\text{NO}_2^-$  by the diazo color method (APHA, 2005). Organic N was calculated as the difference between TKN and inorganic N species. Total organic carbon (TOC) was determined using the Walkley–Black method and total organic matter by the loss-on-ignition method (Soil and Plant Analysis Council Inc., 1999). The C:N ratio was calculated from the ratio between TOC and TKN. Electrical conductivity (EC) and pH were measured by multimeter (WTW, Multimeter 420i, Weilheim, Germany) in a 1:5 (w/w) mixture of compost and distilled water. Water content of the compost samples was determined by the gravimetric method. The biofilter's liquid effluent was characterized for total N by the persulfate method (APHA, 2005) and for TAN,  $\text{NO}_3^-$  and  $\text{NO}_2^-$  as described above.

### 2.4. Ammonia-oxidation potential (AOP)

The potential activity of the nitrifying microorganisms was assessed by testing the aerobic AOP: 5 g of fresh compost sample was mixed with 50 ml medium consisting of 25 mM  $\text{K}_2\text{HPO}_4$

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