Bioresource Technology 114 (2012) 231–240

Contents lists available at SciVerse ScienceDirect

Bioresource Technology

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

# Anammox sludge immobilized in polyvinyl alcohol (PVA) cryogel carriers

Albert Magrí*, Matias B. Vanotti, Ariel A. Szögi

*USDA-ARS Coastal Plains Soil, Water and Plant Research Center. 2611 W. Lucas St., Florence, SC 29501, USA*

ARTICLE INFO

*Article history:*
Received 10 February 2012
Received in revised form 20 March 2012
Accepted 22 March 2012
Available online 2 April 2012

*Keywords:*
Anaerobic ammonium oxidation (anammox)
Immobilization
Gel cell entrapment
Nitrite inhibition
Swine wastewater

ABSTRACT

This study evaluated the use of PVA cryogels to encapsulate slow-growing anammox bacteria for deammonification treatment of wastewater. The cryogel pellets were prepared by freezing-thawing at −8 °C. On average, pellets contained 11.8 mg-TSS/g-pellet of enriched anammox sludge NRRL B-50286 (Candidatus *Brocadia caroliniensis*) in 4-mm cubes. They were tested with synthetic and partially nitrified swine wastewater using continuous stirred-tank reactors packed at 20% (w/v). The immobilized gel was retained inside the reactor by a screen that eliminated the need of sludge recycling. The stoichiometry of anammox reaction was maintained for more than 5 months under non-sterile conditions. The process was not limited by substrates availability unless quite low N concentration (<5 mg/L) achieving >93% removal efficiency. In mass balances, >80% of the potential N conversion activity was achieved (2920 mg-N/kg-pellet/d). In addition, the immobilized bacteria were resilient to inhibition at high nitrite concentrations (244–270 mg-N/L).

## 1. Introduction

Discovery in the early 1990's of the anaerobic ammonium oxidation (anammox) (Mulder et al., 1995) as a new pathway to biologically convert ammonium ($NH_4^+$) to dinitrogen gas ($N_2$) under absence of oxygen has arisen great expectations in the field of wastewater treatment. However, due to the low growth rate of anammox bacteria (doubling time in the range of 1.8–11 days; Isaka et al., 2006; Strous et al., 1998), one of the main challenges for implementing the anammox process is to ensure bacterial cells' retention inside the reactors. For this reason, immobilization of microbial cells has received increasing interest in wastewater treatment to minimize the risk of biomass wash-out from the reactors and provide stabilized treatment (Quan et al., 2011). These immobilization techniques include self-immobilization as granular biomass (Dapena-Mora et al., 2004; López et al., 2008), attachment on the surface of a carrier forming biofilm (Ni et al., 2010; Tsushima et al., 2007), and entrapment of the microbial biomass into gel pellets (Furukawa et al., 2009; Isaka et al., 2007).

The anammox process is especially suitable for the removal of nitrogen (N) from wastewaters containing high ammonium and low biodegradable organic carbon (Paredes et al., 2007). This process consists of a chemolithoautotrophic bioconversion mediated by Planctomycetes-like bacteria that under anoxic conditions oxidize $NH_4^+$ using nitrite ($NO_2^-$) as the electron acceptor. According to the anammox reaction proposed by Strous et al. (1998) (Eq. 1), $NH_4^+$ and $NO_2^-$ are converted to $N_2$ and nitrate ($NO_3^-$) under stoichiometric molar ratios of 1.00:1.32:0.26:1.02 for $NH_4^+$ consumption, $NO_2^-$ consumption, $NO_3^-$ production and $N_2$ production, respectively.

$$1.00NH_4^+ + 1.32NO_2^- + 0.066HCO_3^- + 0.13H^+ \rightarrow 1.02N_2 + 0.26NO_3^- + 0.066CH_2O_{0.5}N_{0.15} + 2.03H_2O \quad (1)$$

The use of synthetic polymers such as urethane, polyethylene glycol (PEG), and polyvinyl alcohol (PVA) for the entrapment of microorganisms was reported as advantageous in the field of wastewater treatment. These advantages include closely packed design of bioreactors, non-toxicity to microorganisms, mechanical strength and long life span of gels, enhanced process efficiency, and resilience to overloading rates as demonstrated in the studies of Sumino et al. (1992), Tanaka et al. (1996) and Vanotti and Hunt (2000) on immobilization of nitrifying biomass. Furukawa et al. (2009) demonstrated that both anammox and nitrifying biomass can be immobilized in PEG gel pellets and used in connected reactors for the successful deammonification treatment of anaerobic digested liquor containing high ammonium concentration (1400–1600 mg $NH_4^+$-N/L).

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*Abbreviations:* ACR, ammonium conversion rate; BOD, biological oxygen demand; COD, chemical oxygen demand; CSTR, continuous stirred-tank reactor; DO, dissolved oxygen; EC, electrical conductivity; HRT, hydraulic residence time; NCE, nitrogen conversion efficiency; NCR, nitrogen conversion rate; NLR, nitrogen loading rate; NRE, nitrogen removal efficiency; NRR, nitrogen removal rate; PEG, polyethylene glycol; PVA, polyvinyl alcohol; SBR, sequencing batch reactor; SNCR, specific nitrogen conversion rate; SNRR, specific nitrogen removal rate; TSS, total suspended solids; VSS, volatile suspended solids.

* Corresponding author. Permanent address: GIRO Technological Centre. Rambla Pompeu Fabra 1. 08100 Mollet del Vallès, Barcelona, Spain. Tel.: +34 93 579 67 80; fax: +34 93 579 67 85.

*E-mail addresses:* albert.magri@gmail.com (A. Magrí), matias.vanotti@ars.usda.gov (M.B. Vanotti), ariel.szogi@ars.usda.gov (A.A. Szögi).

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http://dx.doi.org/10.1016/j.biortech.2012.03.077