Int.J.Curr.Microbiol.App.Sci (2017) *6*(11): 1690-1694

International Journal of Current Microbiology and Applied Sciences
***ISSN: 2319-7706* Volume 6 Number 11 (2017) pp. 1690-1694**
Journal homepage: http://www.ijcmas.com

Original Research Article https://doi.org/10.20546/ijcmas.2017.611.203

Denitrification Evaluation in Chemostat Fermentation Utilizing H_2/CO_2 Gas Recirculation at Mesophilic Temperature

Olga Yolande Aké AKO[1,4*], Sahoré Alexis Drogba[2,4] and Boko AKA[3,4]

[1]Faculty of Sciences and Environment Management, University NANGUI ABROGOUA, 02 BP 801 Abidjan 02 Côte d'Ivoire
[2]Faculty of Sciences and Food Techniques, University NANGUI ABROGOUA, 02 BP 801 Abidjan 02 Côte d'Ivoire
[3]Biomass and Energy Laboratory, Research Institute for New Energies (IREN), University NANGUI ABROGOUA, 02 BP 801 Abidjan 02 Côte d'Ivoire
[4]Faculty of Fundamental and applied Science, University NANGUI ABROGOUA, 02 BP 801 Abidjan 02 Côte d'Ivoire
**Corresponding author*

Keywords

Denitrification, H_2/CO_2 gas, Mixing duration, Chemostat reactor, Fermentation, Hydrogenotrophic Methanogens.

Article Info

Accepted:
15 September 2017
Available Online:
10 November 2017

A B S T R A C T

The aim of this study was to evaluate the performance of an autotrophic denitrification during an anaerobic fermentation process usingH_2/CO_2 mixture gas (80:20, v/v) as sole energy source. Chemostat reactors were operated with anaerobically acclimated hydrogenotrophic methanogens culture broth. Reactors were connected with different volume of H_2/CO_2 gas bag (7L; 9L; 10L and 12L/d). Results showed that environmental factors such as gas mixing duration and H_2/CO_2 volume had effect on denitrification. The nitrogen gas (N_2) volume increased with the mixing duration and the H_2/CO_2 gas volume. The presence of carbon dioxide in the mixture feeding gas maintained the pH stable within all experiment duration. This should help to appreciate the autotrophic denitrification with H_2 as electron donor when the system is utilized for nitrogen removal in a biodegradation process.

Introduction

Biogas generated from anaerobic fermentation remained an important form of renewable energy easy to get in agricultural areas (Bussabong *et al.*, 2013); FFTC (2008) had suggested the utilization of agricultural wastes in anaerobic digestion as source of significant income for the farmers. During the anaerobic degradation process for biogas production, authors were considering the nitrogen as major factor and source for fermentations (Allison and Macfarlane, 1988; Calli *et al.*, 2005; Wagner *et al.*, 2012). Denitrification, as final process in the nitrogen cycle, was carried out simultaneously with methanogenesis in order to investigate anaerobic digestion using organic carbon with or without nitrate addition (Akizuki *et al.*, 2013; Yi *et al.*,

2017). Unfortunately, the environment factors affecting the autotrophic denitrification with hydrogen and employed technologies for its application had not yet been studied. Hence, the present paperaims to evaluate the capacity of denitrification by means of nitrogen gas (N_2) production during chemostat anaerobic process with hydrogenotrophic methanogens using H_2/CO_2 mixture gas as inorganic substrate and electron donor.

Materials and Methods

Inoculum preparation

Detailed by Ako *et al.,* (2008), anaerobic activated sludge (10L) was acclimated until the H_2/CO_2 dependents methanogens were dominant in the culture broth and the ultra-microscopy identified only blue auto fluorescence strain, long rod-shaped cells and cocci (data not shown).

Media

Mineral nutrients and trace metals with no traces of oxygen as conform to the ones prepared previously by Yang *et al.,* (2004) were stocked in a Duran vial after flushing the headspace with H_2/CO_2 (80:20, v/v) for 15 min at pressure of 0.1 MPa. The measured pH was 7.44.

Experimental procedure

Sixteen chemostat reactors (Fig. 1) were fed with 500 mL acclimated hydrogenotrophic methanogens broth culture. The reactors are connected to aluminum tedlar gas bag CCK with H_2/CO_2 (80:20,v/v) at supply the rate of 7L; 9L; 10L and12L/d, by means of 4 reactors per supply rate. Mixing was realized with sixteen small pumps at 0.08 MPa.

Four different mixing durations (continuously; 45 min/h; 30 min/h; 15 min/h) were applied to each supply rate. Media was added every day and the dilution was set up to 0.1/d.

Analysis methods

The pH was measured in situ with pH meter TPX-90 (Toko chemical Laboratories Co. Ltd). Gas bag are changed daily and the nitrogen gas (TN) in the headspace measured by GC-14B (Shimadzu) gas chromatograph equipped with a thermal conductivity detector, connected to a C-R8A data analyzer. TN was determined with 5 duplicates analyses per gas pack to ensure accuracy of the results obtained according to the Standard Methods for Examination of Water and Wastewater (APHA, 2005).

The data were subsequently averaged and the obtained deviations were found to be less than 4%.

Results and Discussion

pH factor in autotrophic denitrification with H_2/CO_2

The pH, generally, acts as an important external inhibitor for bacterial activity within the continuously flowing reactor (Keshtkar *et al.,* 2003). In the present study, the pH plot shows a range from 7.39 to 7.9 (Fig. 2).

Authors variously report the optimum pH range to perform the Hydrogenotrophic denitrification, from 7.6 to 8.6 (Ghafari *et al.,* 2009) or from 7.5 to 7.6 (Zhou *et al.,* 2007).

To overcome the effect of pH on denitrification (Karanasios *et al.,* 2010), the utilization of a pressure of 0.08 MPa and a mixture gas H_2/CO_2in experiments is suitable to maintain pH due to the tampon effect of CO_2 as Tang *et al.,* (2011) advise the application of CO_2 to stabilize the pH.

Fig 1: One of the 4 Incubators compose with 4 Duran vials connected to 4 gas bags

Incubator

Fig 2: pH variation during 5 days experiments

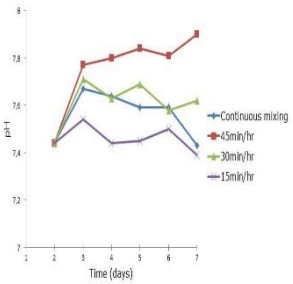

Figure 2

Fig 3: N_2 gas production during continuous mixing

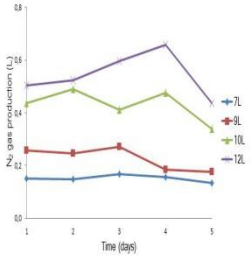

Figure 3

Fig 4: N_2 gas production during intermittent at 45min/hr

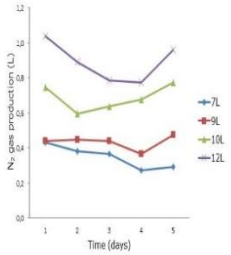

Figure 4

Fig 5: N_2 gas production during mixing at 30 min/hr

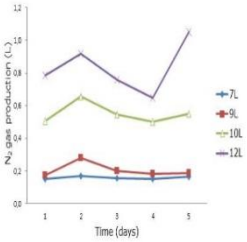

Figure 5

Fig 6: N_2 gas production during mixing at 15 min/hr

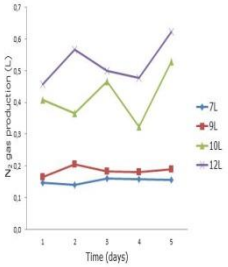

Figure 6

Fig 7: Effect of mixing duration on nitrogen gas production

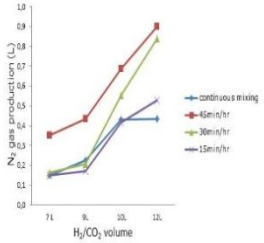

Figure 7

In General, less than 1% of N_2 gas content in the biogas resulted from the degradation of organic materials and 5 to 15% in the landfills produced biogas (Jönsson *et al.*, 2003). During all experiments no external N_2 gas was sparge and no organic material that might contain nitrogen in their complex molecules was added to the growth culture. The autotrophic denitrification with hydrogen as electron donor indicates a production of Nitrogen gas comprises between 2.98 to 5.8%, albeit we operate in hydrogenotrophic methanogens culture broth environment.

Effect of mixing duration on nitrogen gas production

The monitoring of mixing duration shows in Figure 7 a regular effect of intermittent mixing duration on the nitrogen production. The highest nitrogen is produced at 45 min/hr, results assert that a high mixing duration increases the nitrogen gas production and the continuous mixing does not permit a high activity. Kaparaju *et al.*, (2008) have shown an improvement of methane production during intermittent mixing on comparison to continuous mixing at high temperature. Continuous mixing seems to have negative effect on bacterial activity.

During the conversion of H_2/CO_2 gas to methane, nitrogen is formed as result of the process by-product. In the present paper, using a chemostat fermentation technology, the mixing duration and the volume of gas by means of H_2/CO_2 used as sole source of carbon influence the nitrogen production. The result can help to appreciate and/or simulate when autotrophic denitrification with H_2 as electron donor is utilize for nitrogen removal in a biodegradation process. Belmonte *et al.*, (2016) indicate the use of compounds such H_2avoids the appearance of secondary pollution in comparison with sulfur autotrophic denitrification; the technology can be a sustainable alternative for nitrate removal.

References

Akizuki, S., Izumi, K., Nagao, N., Shiotani, T., Niwa, C., and Toda, T. 2013. Effect of ratio and seed sludge on simultaneous methanogenesis and denitrification in intermittent organic solid waste treatment. International Biodeterioration and Biodegradation. 84: 8-13.

Ako, O., Kitamura, Y., Intabon, K., Takaaki,S. 2008.Steady state characteristics of acclimated hydrogenotrophic methanogens on inorganic substrate in continuous chemostat reactors. Bioresource Technology. 99(14): 6305-6310

Allison, C., Macfarlane, G.T. 1988. Effect of nitrate on methane production and fermentation by slurries of human faecal bacteria. Journal of General Microbiology. 134: 1397-1405.

APHA 2005. Standard methods for the examination of water and wastewater (APHA, AWWA and WEF), 21 st Ed., Washington, DC.

Belmonte, M., Fajardo, C., Toledo-Alarcón, J. B., Heredia, D. V., Jorquera, L., Méndez, R., Ruiz-Filippi, G. 2016. Autotrophic Denitrification processes technologies for the treatment and recovery of nutrients from industrial wastewater. 147.

Bussabong, N., Watcharenwong, A., Dararat, S. 2013. Biogas production from Ruzi grass in the continuous stirred tank reactor (CSTR). International Journal of Chemistry Environment and Biological Science. 1(2): 348–351.

Calli, B., Mertoglu, B., Inanc, B., Yenigun O. 2005. Effects of high free ammonia concentration on the performances of anaerobic bioreactors. Process Biochemistry. 40: 1285–1292.

Foods and Fertilizer Technology Centre, FFTC 2008. Sustainable production and

utilization of biomass as renewable energy source in the Asian and Pacific region. FFTC meeting, November.

Ghafari, S., Hasan, M., Aroua, M. K. 2009.A kinetic study of autohydrogenotrophic denitrification at the optimum pH and sodium bicarbonate dose. Bioresource Technology. 101(2010): 2236-2242.

Jönsson, S., Ejlertsson, J., Svensson, B. H. 2003. Behaviour of mono- and diesters of *o*-phtalic acid in leachates released during digestion of municipal solid waste under landfill conditions. Advances in Environmental Research. 7(2): 429-440.

Kaparaju, P., Buendia, I., Ellegaard, L., Angelidakia, I. 2008. Effects of mixing on methane production during thermophilic anaerobic digestion of manure: Lab-scale and pilot-scale studies. Bioresource Technology. 99(11): 4919-4928.

Karanasios, K.A., Vasiliadou, I.A., Pavlou S., Vayenas D.V. 2010. Hydrogenotrophic denitrification of potable water A review. Journal of Hazardous Materials. 180(1-3): 20-37.

Keshtkar A., Meyssami B., Abolhamd G., Ghaforian H., Khalagi Asadi M. 2003. Mathematical modeling of non-ideal mixing continuous flow reactors for anaerobic digestion of cattle manure. Bioresource Technology. 87(1): 113-124.

Ryckebosch, E., Drouillon, M., Vervaeren, H. 2011.Techniques for transformation of biogas to Biomethane. Review 35(5): 1633-1645.

Wagner, A. O., Holhbrugger, P., Lins, P., Illmer, P. 2012. Effect of different nitrogen sources on biogas production – A lab scale investigation. Microbiological Research. 167: 630-636.

Yang, Y., Zhang, Z., Lu, J., Maekawa, T. 2004. Continuous methane fermentation and the production of vitamin B_{12} in a fixed-bed reactor with loofah. Bioresource technology. 92(3): 285-290.

Yi, X.H., Wan, J., Ma, Y., Wang, Y., Guan, Z., Jin, D.D. 2017. Structure and succession of bacterial communities of the granular sludge during the initial stage of the simultaneous denitrification and methanogenesis Process. Water Air and Soil Pollution. 228:121.

Zhou, M., Fu, W., Gu. H., Lei L.2007. Nitrate removal from groundwater by a novel three-dimensional electrode biofilm reactor. Electrochimica Acta. 52(19): 6052-6059.

How to cite this article:

Olga Yolande Aké, AKO, Sahoré Alexis Drogba and Boko, AKA. 2017. Denitrification Evaluation in Chemostat Fermentation Utilizing H_2/CO_2 Gas Recirculation at Mesophilic Temperature. *Int.J.Curr.Microbiol.App.Sci.* 6(11): 1690-1694.
doi: https://doi.org/10.20546/ijcmas.2017.611.203