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Effect of inoculum on the anaerobic digestion of food waste accounting for the concentration of trace elements

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A B S T R A C T

The production of renewable energy in the form of methane from the anaerobic digestion (AD) of food waste (FW) varies depending on factors such as the quantity and quality of the inoculum. This research evaluated the influence of trace elements (Ca, K, Fe, Zn, Al, Mg, Co, Ni, and Mo) present in inoculum from different sources (wastewater treatment plants (WWTPs): 2 agro-industrial WWTPs and 1 municipal WWTP) on the AD of FW. This study found that the source of the inoculum determines the content of macronutrients and trace elements, which can alter the requirements of the AD process and therefore affect methane production. The inoculum obtained from municipal WWTPs contain potentially inhibitory concentrations of Zn and Al that negatively affect methane production (<70 mL CH_4·gVS^{-1}), the hydrolysis constant (<0.19 d^{-1}), and the lag-phase (>7 days). It was also found that high concentrations of trace elements such as Ni (35.2 mg kg^{-1}) and Mo (15.4 mg kg^{-1}) in the inoculum increase methane production (140.7 mL CH_4·gVS^{-1}) and hydrolysis constant (>0.18d^{-1}) in addition to presenting short lag-phase (<1 day) in the AD of food waste.

1. Introduction

Food waste (FW) constitutes 40% municipal solid waste (MSW) in developed countries and up to 75% MSW in developing countries (Cabeza et al., 2016). According to Dhamodharan et al. (2015), 1300 million tons of FW are generated each year. This FW is characterized by a high moisture content (70 to 90%), a high organic matter content (VS/TS > 80%), and C/N ratios ranging from 15 to 36 (Zhang et al., 2014; Thi et al., 2015).

Anaerobic digestion (AD) is a technological alternative that is gaining interest for the conversion of the organic matter present in FW into biogas. In general, biogas consists of 50–75% CH_4 and 25–50% CO_2, along with other trace components such as water vapor (H_2O), hydrogen sulfide (H_2S), and ammonia (NH_3) (Rasi, 2009); however, the composition of the biogas obtained varies depending on the type of raw material and the operating conditions of the digester (Verstraete et al., 2005; Matheri et al., 2017).

The start-up of an anaerobic digester is importantly affected by the quality of the inoculum and by the concentration of trace elements (Owen et al., 1979; Demirel and Scherer, 2011). Studies such as those of Raposo et al. (2006), Angelidaki et al. (2009) and Holliger et al. (2016) indicate that the quality of the inoculum is related to factors such as source, concentration, specific methanogenic activity (SMA), preincubation, acclimatization, adaptation, and storage conditions.

It has been recognized that micronutrients and trace elements play an important role in the startup and operation of anaerobic reactors (Schattauer et al., 2011; Nges and Björnsson, 2012; Facchin et al., 2013). Therefore, it is important to add these components to the reaction mixture. This has traditionally been done by using a co-substrate that supplies the deficit of these elements in the waste to be treated or by supplementation with commercial products. Few studies have analyzed the contribution of these elements when they are present in the inoculum. In that sense, Ishaq (2012) and Roussel (2012) affirmed that biomass used for the development of anaerobic processes can contain high concentrations of trace elements that do not limit methane production. On

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the other hand, certain levels of trace elements can cause inhibition of AD (Chen et al., 2008a, 2008b; Islam et al., 2017), hence the importance of further studying this aspect in AD. Therefore, the aim of this study is to evaluate the presence of trace elements (Ca, Fe, Mg, Zn, Al, Ni, Co and Mo) in three inoculum from different WWTPs and to characterize their influence on the methane production and on the kinetics of the process.

2. Materials and methods

2.1. Substrate and inoculums

The FW used as a substrate was collected from the restaurant of the University of Valle (Cali, Colombia), which serves more than 3000 students each day and generates 86.6 kg d^{-1} waste. This FW consists of banana and potato peels (56%), citrus fruits (25%), non-citrus fruits (8.2%), fibers and minerals (eggshells, celery, and herbs, among others), and other types of herbs (cilantro, chard, and citron, among others) (3.2%). Prior to conducting biochemical methane potential (BMP) testing, the FW was mechanically pretreated using a CB15 blender (*Waring Commercial, Connecticut, United States*) at a speed of 15,800 rpm for one minute (standard blender speed); this ensured a particle size of less than 30 mm (Izumi et al., 2010). Three inoculum were used, one from a municipal WWTP and two from agro-industrial WWTPs. Table 1 shows the source of each of the inoculum used.

2.2. Analytical methods

The FW was characterized according to ICONTEC (2004), ICONTEC (2009) and APHA (2005) in terms of moisture, pH, total alkalinity (TA) and bicarbonate alkalinity (BA, volatile fatty acids (VFAs), chemical oxygen demand (COD_{total}; $COD_{filtered}$), biochemical oxygen demand (BOD_5), total solids (TS) and volatile solids (VS), UV_{254}, total organic carbon (TOC), total nitrogen, total ammoniacal nitrogen, total phosphorus, calcium (Ca), potassium (K), iron (Fe), zinc (Zn), aluminum (Al), magnesium (Mg), cobalt (Co), nickel (Ni), and molybdenum (Mo).

The inoculums were analyzed for pH, TA, BA, VFAs, VS, TS, the same trace elements identified in the substrate, settleability (V_s), and specific methanogenic activity (SMA).

2.3. Experimental unit (BMP tests)

For the laboratory experiments (BMP tests), biogas quantification was performed by the manometric method using the Oxitop® system (WTW, Giessen, Gemany) with 250-mL reactors (Pabón et al., 2012). The working volume used in each of the tests was 200 mL, leaving 50 mL for biogas storage. To ensure that the biogas measured by manometry was primarily methane, carbon dioxide was captured on NaOH solution, and the composition of the captured gas was measured using a GC-2014 Shimadzu chromatograph. To measure CH_4, a flame ionization detector (FID) and an electron capture detector (ECD) were used with a Porapak Q precolumn and a column head (80–100 mesh), whereas for CO_2, an infrared analyzer with a NaOH trap and silica gel was used. The volume of CH_4 emitted under standard conditions (T = 273 K, P = 1 atm) was determined according to Parra-Orobio et al. (2015); this determination takes into account the amount of dissolved methane.

The tests were performed in a WTW TS 606-G/2-I (Giessen, Germany) incubator with intermittent manual stirring for 32 days at a temperature of 35 ± 0.5 °C. The pH was adjusted to 7.0 with 4% (v/v) $NaHCO_3$ solution. The substrate-inoculum ratio was 1.0 g $VS_{substrate}\cdot g\ VS_{inoculum}^{-1}$, according to the recommendations of Lesteur et al. (2010) and Cárdenas-Cleves et al. (2016). In all experimental units, the inoculum concentration was 1.5 g $VS_{inoculum}\cdot L^{-1}$, the recommended value for BMP tests without continuous agitation (Cárdenas-Cleves et al., 2016).

In the BMP tests of each of the inoculum used, two conditions, with regard to nutrient availability, were evaluated: i) with nutrients (WN: 1 mL for each working liter of the reactor) and ii) no nutrients (NN). The solutions were modified based on the recommendations of Aquino et al. (2007), Angelidaki et al. (2009), and Torres and Pérez (2010), who assert that the addition of macro and micronutrients in BMP test ensures the achievement of the highest biodegradation of the substrate in an anaerobic environment. Each condition was evaluated in triplicate in parallel with a control inoculum (distilled water plus inoculum) to allow endogenous correction for methane.

The macronutrient solution contained NH_4Cl (170 $g\cdot L^{-1}$), $NaHCO_3$ (1 $g\cdot L^{-1}$), KH_2PO_4 (37 $g\cdot L^{-1}$), $MgSO_4\cdot 4H_2O$ (9 $g\cdot L^{-1}$), and $CaCl_2\cdot 2H_2O$ (8 $g\cdot L^{-1}$); the micronutrient solution contained $FeCl_2\cdot 6H_2O$ (2 $g\cdot L^{-1}$), $ZnCl_2$ (0.05 $g\cdot L^{-1}$), $CuCl_2\cdot 2H_2O$ (0.03 $g\cdot L^{-1}$), $MnCl_2\cdot 4H_2O$ (0.5 $g\cdot L^{-1}$), $(NH_4)Mo_7O_{24}\cdot 4H_2O$ (0.09 $g\cdot L^{-1}$), $AlCl_3\cdot 6H_2O$ (0.05 $g\cdot L^{-1}$), $CoCl_2\cdot 6H_2O$ (2 $g\cdot L^{-1}$), $NiCl_2\cdot 6H_2O$ (0.05 $g\cdot L^{-1}$), H_3BO_3 (0.05 $g\cdot L^{-1}$), $Na_2SeO_3\cdot 5H_2O$ (0.1 $g\cdot L^{-1}$), EDTA (1 $g\cdot L^{-1}$), and HCl (1 $mL\cdot L^{-1}$). To ensure a total redox environment, resazurin (0.5 $g\cdot L^{-1}$) and $NaS\cdot 7H_2O$ (0.1 $g\cdot L^{-1}$) were added to each reactor.

2.4. Statistical analysis

To determine the effects of the inoculum and the trace elements on BMP, ANOVA and post-ANOVA (Tukey's test) analyses were performed using the statistical package R i386 3.0.2 (R Foundation®). The response variable was BMP, and the significance level was set at $p < 0.05$.

2.5. Determination of the theoretical methane potential

The theoretical BMP and the biodegradability index (%B) were determined from the stoichiometry of the substrate degradation reaction using Eqs. (1)–(3), as recommended by Buswell and Mueller (1952) and Sosnowski et al. (2003).

$$C_nH_aO_bN_c + \left(n - \frac{a}{4} - \frac{b}{2} + \frac{3c}{4}\right)H_2O \rightarrow \left(\frac{n}{2} - \frac{a}{8} - \frac{b}{4} + \frac{3z}{8}\right)CO_2 + \left(\frac{n}{2} + \frac{a}{8} - \frac{b}{4} - \frac{3z}{8}\right)CH_4 + cNH_3 \quad (1)$$

$$BMP_{theoretical}(\text{mL CH}_4\ \text{g VS}^{-1}) = \frac{22.4 * \left(\frac{n}{2} + \frac{a}{8} - \frac{b}{4} - \frac{3c}{8}\right) * 1000}{12n + a + 16b + 14c} \quad (2)$$

Table 1
Description of each inoculum used in the study.

Inoculum	Sludge type	Reactor type	Wastewater type	HRT (h)	Flow rate ($L\cdot s^{-1}$)	Temperature (°C)
Inoculum I	Granular	UASB	Sugar industry	6	8.3	34
Inoculum II	Granular	UASB	Slaughter of cattle and pigs	12	7	25
Inoculum III	Flocculent	UASB	Municipal	8.5	96	20

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