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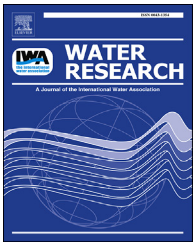

Digester performance and microbial community changes in thermophilic and mesophilic sequencing batch reactors fed with the fine sieved fraction of municipal sewage

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ABSTRACT

This study investigates the start-up and operation of bench-scale mesophilic (35 °C) and thermophilic (55 °C) anaerobic sequencing batch reactor (SBR) digesters treating the fine sieved fraction (FSF) from raw municipal sewage. FSF was sequestered from raw municipal wastewater, in the Netherlands, using a rotating belt filter equipped with a 350 micron mesh. For the given wastewater, the major component of FSF was toilet paper, which is estimated to be 10–14 kg per year per average person in the western European countries. A seven months adaptation time was allowed for the thermophilic and mesophilic digesters in order to adapt to FSF as the sole substrate with varying dry solids content of 10–25%. Different SBR cycle durations (14, 9 and 2 days) were applied for both temperature conditions to study methane production rates, volatile fatty acids (VFAs) dynamics, lag phases, as well as changes in microbial communities. The prevailing sludge in the two digesters consisted of very different bacterial and archaeal communities, with *OP9* lineage and *Methanothermobacter* being pre-dominant in the thermophilic digester and *Bacteroides* and *Methanosaeta* dominating the mesophilic one. Eventually, decreasing the SBR cycle period, thus increasing the FSF load, resulted in improved digester performances, particularly with regard to the thermophilic digester, i.e. shortened lag phases following the batch feedings, and reduced VFA peaks. Over time, the thermophilic digester outperformed the mesophilic one with 15% increased volatile solids (VS) destruction, irrespective to lower species diversity found at high temperature.

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1. Introduction

Cellulose makes up about 30–50% of the suspended solids in the sewage of western countries, mainly originating from the use of toilet paper which is estimated to be 10–14 kg per person per year (STOWA, 2010). This material can enter the aerobic sewage treatment, adding significant costs to sewage treatment due to energy input for aerobic degradation and incineration costs of the non-degraded fibres that end up in wet waste sludge after digestion (Ruiken et al., 2013). Ruiken et al. (2013) suggested the use of a fine sieve (mesh size 350 μm) to separate suspended solids from sewage before entering the biological treatment, instead of using conventional primary clarifiers. Based on thermographic measurements, the cellulose fraction found in the FSF was 79% of the total mass and 84% of the organic mass; the inorganic matter fraction was 6%. In comparison, the cellulose fraction of primary sludge only reaches a maximum of 32–38% of organic mass (Ruiken et al., 2013). Also, the total solids content in the FSF without additional dewatering is higher than that of primary sludge, i.e. 10–25% as found in our present study, compared to 4–12% as indicated in previous works (Inc et al., 2003; Tchobanoglous et al., 2003), resulting in a lower total sludge volume production. FSF can be reused as fibres in several processes where nowadays recycled paper is used; however, the origin of these fibres could limit these opportunities (STOWA and Grondstoffenfabriek, 2013). A more straightforward method to valorise the FSF on site is by anaerobic (dry) digestion. Anaerobic digestion is a carbon-neutral technology to produce biogas that can be used for heating, generating electricity, mechanical energy, or for supplementing the natural gas supply. The produced bioenergy such as methane can contribute to the goal of realising an energy neutral or energy producing sewage treatment plant (Roeleveld et al., 2010).

In nature, hydrolysis and fermentation of lignocellulosic biomass is done by cellulolytic microorganisms belonging to the phyla *Firmicutes*, *Actinobacteria*, *Bacteroidetes*, *Proteobacteria*, *Thermotogae* and *OP9* (Peacock et al., 2013; Kaoutari et al., 2013). These microorganisms can release fermentation products, such as various types of fatty acids, into natural environments and complete the carbon cycle via methane and/or CO_2 under anaerobic conditions (Minty et al., 2013; DeAngelis et al., 2012). Lignocellulosic biomass, which has similar characteristics to FSF, has been widely used for bio-methanation by coupling cellulolytic microorganisms, fermenting bacteria and methanogenic archaea in one or two-stage anaerobic bioreactors (Zhang et al., 2013; Merlino et al., 2013). Such process can be operated under mesophilic (35 °C) and thermophilic (55 °C) conditions.

Mesophilic anaerobic digestion of organic solids is often reported as the most convenient, stable and reliable form of substrate conversion leading to stable methane production rates. However, mesophilic hydrolysis rates are lower compared to thermophilic conversion rates (Lu et al., 2013). On the other hand, thermophilic digestion requires higher energy input, and is regarded more sensitive to changes in operational conditions, such as changes in temperature and the organic loading rate, as well as to changes in substrate characteristics (Kim et al., 2002; van Lier, 1996). The perceived poor process stability as well as the lack of experience in operating thermophilic processes are probably the main reasons that have prevented its wide-scale application. The higher vulnerability could be due to a less diverse microbial community (Raskin et al., 1994), persistence of propionate (Wilson et al., 2008) and increased toxicity of intermediates at the thermophilic temperature range (van Lier, 1996).

Thermophilic anaerobic digestion of lignocellulosic biomass, such as FSF, might be more effective than mesophilic digestion (De Baere, 2000). The hydrolysis of complex polysaccharides by thermophilic microorganisms establish higher rates compared to mesophiles; each 10 °C increase in temperature can increase enzymatic rates by two- to three-fold (Mozhaev, 1993). High temperatures can also increase substrate solubility (Mozhaev, 1993) and decrease the bulk liquid viscosity (Eshtiaghi et al., 2013), leading to improved mixing performance and thus an increased hydrolysis of (hemi-)cellulose to monomers (Eichorst et al., 2013).

At present, anaerobic digestion at the mesophilic temperature range is widely applied and well described in many publications, whereas the application of thermophilic digestion is still limited. With regard to lignocellulosic wastes, such as FSF, comparative studies conducted in parallel under both thermophilic and mesophilic conditions (Golkowska and Greger, 2013) are difficult to find. In this research, the feasibility and efficiency of one-step anaerobic digestion of FSF under thermophilic and mesophilic conditions in laboratory batch fed reactors (8 L) was compared. Digestion performance and microbial dynamics were followed in time under both conditions during reactor start-up and after extended adaptation times.

2. Materials and methods

2.1. *Digester*

Four water jacketed laboratory mixed digesters with a working volume of 8 L were used in duplicate to conduct the digestion of FSF under both thermophilic and mesophilic conditions, at 55 °C and 35 °C, respectively applying sequencing batch feeding conditions. The reactors were continuously mixed by stirring (60-80RPM, Maxon motor Benelux B.V., Switzerland) to achieve a more homogenized matrix. The system was equipped with a pH and temperature probe (CPS41D, Endress+Hauser B.V., Switzerland) and an on-line biogas measuring device (RITTER MilliGascounter MGC-1 PMMA, Germany). The temperature was controlled by circulating water from a programmable water bath (TC16, PMT TAMSON, the Netherlands). Temperature, pH, biogas flow rate were continuously monitored using Labview software.

2.2. *Substrate*

A rotating belt filter (Salsnes Filter, Norway) equipped with a 350 μm pore size fine sieve, was operated to treat the screened (mesh size 6 mm) sewage at WWTP Blaricum, the Netherlands (plant size: 30,000 pe, maximum hydraulic capacity 1600 m^3/h). The FSF coming from this sieve was collected once every