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Biogas production from brewery spent grain as a mono-substrate in a two-stage process composed of solid-state anaerobic digestion and granular biomass reactors

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ABSTRACT

Anaerobic digestion of brewery spent grain as a mono-substrate was studied. Brewery spent grain is a substrate consisting largely of cellulose, hemicellulose and lignin, which are difficult to degrade anaerobically, mostly due to the presence of degradation products, such as phenolic compounds, which cause process inhibition. Therefore, a two-stage system was used for anaerobic digestion. Anaerobic digestion was phase separated in a solid-state anaerobic digestion reactor, where microbiological hydrolysis and acidogenesis occurred and in a granular biomass reactor where mostly methanogenesis was performed. The overall process exhibited total solids degradation efficiency between 75.9 and 83.0%. Average specific biogas production was 414 ± 32 L/kg, whereas biomethane production was 224 ± 34 L/kg of added total solids. Granular biomass after adaptation exhibited stable operation at substrate C/N ratios in range 0.16 −4.68. *p*-cresol was present in concentrations up to 45 mg/L and during the process was successfully degraded by granular biomass. The excellent adaptability of granular biomass was confirmed by 68.2% shift in bacterial and a 31.8% shift in archaeal community structure in a granular biomass reactor. The structure of the bacterial community from granular biomass reactor and solid-state anaerobic digestion reactor remained 79.4% similar at the end of the experiment, whereas archaeal community was only 31.6% similar. The process exhibited stable operation for 198 days, which shows that brewery spent grain can be successfully anaerobically digested and used for biogas production.

1. Introduction

Breweries as a significant user of fossil fuels are under constant economic as well as environmental pressure to reduce energy and residue disposal costs. Anaerobic digestion and biogas production from brewing residues offers a viable option for achieving a considerable reduction in fossil fuel use, consequently reducing energy and residue disposal costs while reducing the carbon footprint as well. Brewing residues include brewery wastewater, surplus yeast and brewery spent grain (BSG). Brewing wastewater treatment has been thoroughly addressed and well investigated (Connaughton et al., 2006) with granular biomass technology (such as UASB – Up-flow Anaerobic Sludge Blanket) as the prevalent technological approach (Baloch et al., 2007). The conditions for surplus yeast anaerobic digestion have also been thoroughly determined (Zupančič et al., 2012) and applied in full scale (Zupančič et al., 2016). Therefore, only BSG remains largely untapped resource for anaerobic digestion. There are many possible applications for BSG use (Mussatto et al., 2006); of which currently food supplement and cattle feed are the most often used (Fillaudeau et al., 2006). BSG are only in recent years considered as an energy substrate, due to decreasing revenues from cattle feed applications, higher energy costs and renewable energy production incentives. Already the earliest studies have revealed that even though BSG have a biogas potential, conventional anaerobic

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digestion is an economically unattractive option (Rieker et al., 1992; Behmel et al., 1993) because of long retention times and slow biodegradability. It has already then been pointed out that in order to successfully digest BSG, hydrolysis stage of the anaerobic process has to be separated from other stages because it is the rate-limiting step. Recent studies have also confirmed that using thermo-chemical pre-treatment (Panjičko et al., 2015) or just thermal pre-treatment (Bochmann et al., 2015), significantly reduces retention time and consequently improves the economics of the anaerobic process. This is due to the complex lignocellulosic BSG composition. BSG is comprised of cellulose (16.8–25.4%), hemicellulose (21.8–28.4%), lignin (11.9–27.8%), protein (15.2–24.0%), lipid (up to 10%), and ash (2.4–4.6%) (Mussatto et al., 2006). Behmel et al. (1993) were the first to show that separated hydrolysis stage from anaerobic digestion can provide as much as 86% organic solids degradation in the overall process. Moreover, Sežun et al. (2011) have shown that even with the separated hydrolysis stage, it is difficult to achieve a long-term stable anaerobic digestion process. The use of different thermal, mechanical and chemical pre-treatments, followed by conventional anaerobic digestion, resulted in process inhibition by phenolic degradation products, mainly *p*-cresol at the concentration of 200 mg/L. Inhibition always occurred in prolonged operation after several retention times, even up to 120 days of operation, depending on the efficiency of pre-treatment. The mechanism of *p*-cresol inhibition is based on the inhibition of acetogenesis, resulting in the accumulation of VFA (excluding acetic acid) and consequently indirect inhibition of methanogenic archaea (Rétfalvi et al., 2013). BSG also have a high protein content (C/N ratio of 3–5 and total nitrogen (TN) of 11–13 g/kg of wet weight), which may lead to ammonia inhibition if BSG are used as a mono substrate. Sung and Liu (2003) reported inhibition limit already at 4.92 g/kg of TN. Therefore, dilution of the substrate is necessary or using additional substrates to perform co-digestion, where BSG is diluted with a more carbon-rich substrate. Many studies have therefore been conducted, where BSG is used as a co-substrate. Malakhova et al. (2015) used Jerusalem artichoke phytomass as a co-substrate with BSG (5:1 total solids (TS) ratio of BSG vs. Jerusalem artichoke) and achieved specific methane production of 108 L/kg of wet weight BSG added in a batch experiment. Dido et al. (2014) used sewage sludge and digested maize as a co-substrates with BSG (mixtures not specified) in batch experiments, and achieved specific biogas production of 150 L/kg of volatile solids (VS) and 769 L/kg VS of added mixture, respectively. Tewelde et al. (2012) used cattle dung as a co-substrate with BSG (70:30 wet weight ratio) achieving a specific biogas production of 410 L/kg VS of added mixture in a continuous experiment with a hydraulic retention time of 40 days. Nevertheless, our goal was to use only BSG as a mono substrate, to overcome all difficulties and achieve a long-term stable operation.

In this study, the objective was to omit the conventional anaerobic digestion of BSG and to use a novel approach. The studied process was composed of separated hydrolysis stage with a two-stage process, where microbiological hydrolysis and most of the acidogenesis is performed in a solid-state anaerobic digestion reactor (SS-AD), while methane production is mainly performed in the UASB similar granular biomass reactor (GBR). Solid-state anaerobic digestion (SS-AD), also commonly known as “dry digestion” or “solid-phase digestion”, an anaerobic digestion system that operates at a total solids (TS) content higher than 15%, is a promising technology for converting the lignocellulosic biomass to renewable energy in the form of biogas (Sawatdeenarunat et al., 2015). The SS-AD (either single or two stage) is considered more suitable for the treatment of lignocellulosic biomass than conventional AD due to the reduced floating and stratification problems associated with fibrous materials (Karthikeyan and Visvanathan, 2013). Furthermore, the SS-AD has several other advantages compared to conventional anaerobic digestion: smaller reactor volume, lower energy requirements for heating, minimal material handling requirements, and lower total parasitic energy loss (Li et al., 2011). The biogas production from SS-AD is comparable to the output of conventional AD (Brown et al., 2012); however, it performs more effectively at higher organic loading rates. Even more, a two-stage SS-AD system with separated biological hydrolysis stage offers significant improvement of degradation rate and biogas production (Panjičko et al., 2015). Authors researching SS-AD agree that due to previously mentioned advantages, it can be a suitable technology for lignocellulosic biomass digestion. Therefore, it is our assessment that it could be suitable for BSG digestion as well. We have set out to continue the research on BSG anaerobic digestion considering previous studies in this field (Rieker et al., 1992; Behmel et al., 1993; Bochmann et al., 2015), where authors clearly showed a positive impact of a separated hydrolytic pre-treatment as well as findings from Sežun et al. (2011) about *p*-cresol inhibition. In essence, granular biomass structure protects bacteria and archaea from exposure to toxic compounds by providing better mass transfer resistance to different substances diffusion inside the granules. It has long been known that granular sludge based technology is effective for the anaerobic digestion of industrial wastewaters containing toxic phenolic compounds either in a single stage system (Scully et al., 2007) or in a two-stage system (Wang et al., 2011). Therefore, we assumed that two-stage process that combines a granular biomass reactor for successful digestion of possible phenolic-type degradation compounds, and a solid-state anaerobic digestion reactor to assure good microbiological hydrolysis decomposition, should suffice to design an efficient and long-term stable operation of anaerobic digestion process using BSG as a mono substrate.

2. Materials and methods

2.1. Seed sludge - inoculum

The seed sludge was taken from an operational EGSB (Expanded Granular Sludge Bed) reactor of the same brewery where the substrates were collected. The seed sludge was put in operation quickly after collection without any conditioning. Samples were taken for TS, VS and microbial biomass analysis. The sludge was typically granular with TS concentration of 80–100 g/L, of which 90% are VS and the cross-section of the sludge granules ranged from 1 to 4 mm.

2.2. Substrates

The brewery spent grain (BSG) and anaerobically digested brewery wastewater (BWW) were collected from a local brewery with annual BSG production of 17,000 tonnes and brewery wastewater production of approx. 400,000 m^3 annually. BSG are the main waste fraction of beer production. It is a residue of the final mash process of malting and includes the lignocellulosic malt hulls of 2–4 mm in size. BSG used in the experiments had total solids (TS) concentration between 211 and 263 g/kg. Of those, on average, 96.1% were volatile solids (VS). Total organic carbon (TOC) was in the range of 65–70% of TS and total nitrogen (TN) concentration in the range of 11–13 g/kg of wet weight. The prevailing compounds in BSG (average of 6 used samples) were hemicellulose (24.7 ± 0.5% of TS), cellulose (23.7 ± 0.5% of TS), lignin (24.6 ± 0.4% of TS) and protein, determined as crude protein (21.4 ± 0.3% of TS).

To ensure proper hydraulic handling, BWW was used in BSG:BWW wet weight ratio of 1:2. BWW had the pH between 6.8 and 7.1, TS concentration in the range of 90–280 mg/L (average VS were 81% of TS), TOC concentration in the range of 100–120 mg/L