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Biohydrogen production from pig slurry in a CSTR reactor system with mixed cultures under hyper-thermophilic temperature (70 °C)

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ABSTRACT

A continuous stirred tank reactor (CSTR) (750 cm^3 working volume) was operated with pig slurry under hyper-thermophilic (70 °C) temperature for hydrogen production. The hydraulic retention time (HRT) was 24 h and the organic loading rate was 24.9 $g\,d^{-1}$ of volatile solid (VS). The inoculum used in the hyper-thermophilic reactor was sludge obtained from a mesophilic methanogenic reactor. The continuous feeding with active biomass (inoculum) from the mesophilic methanogenic reactor was necessary in order to achieve hydrogen production. The hyper-thermophilic reactor started to produce hydrogen after a short adapted period of 4 days. During the steady state period the mean hydrogen yield was 3.65 $cm^3\,g^{-1}$ of volatile solid added. The high operation temperature of the reactor enhanced the hydrolytic activity in pig slurry and increased the volatile fatty acids (VFA) production. The short HRT (24 h) and the hyper-thermophilic temperature applied in the reactor were enough to prevent methanogenesis. No pre-treatment methods or other control methods for preventing methanogenesis were necessary. Hyper-thermophilic hydrogen production was demonstrated for the first time in a CSTR system, fed with pig slurry, using mixed culture. The results indicate that this system is a promising one for biohydrogen production from pig slurry.

1. Introduction

Hydrogen is an ideal, clean and alternative energy source with high energy value 122 $kJ\,g^{-1}$ [1]. During the last years, the necessity to exploit alternative energy sources in order to replace fossil fuels and reduce carbon dioxide emissions to the atmosphere, triggered the research for innovative hydrogen production systems. The use of hydrogen as a fuel does not aggravate the environment with gasses that make the global warming phenomenon more intense. Anaerobic fermentation and photosynthesis are the two main biological methods to produce hydrogen from organic wastes [2]. Dark fermentation is a more promising and environmentally friendly method to produce hydrogen from wastes compared to photosynthesis, especially when they contain high concentration of organic matter as the pig wastes [3]. It is difficult for light to penetrate into a rich in organic matter medium and thus photosynthetic microorganisms cannot grow [4]. On top of this, anaerobic

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fermentation gives higher hydrogen production rates. Therefore, fermentative hydrogen production from organic waste may be the most feasible process.

Anaerobic digestion consists of three major steps: hydrolysis/acidogenesis, acetogenesis and methanogenesis. Hydrogen is produced during the first two steps. The hydrogen produced, is rapidly consumed by the methanogenic bacteria in the third step. Therefore, it is important to prevent methanogenesis for successful biohydrogen production.

Studies on biohydrogen production have mainly focused on mesophilic [5] and thermophilic conditions [6]. However, biohydrogen production under hyper-thermophilic conditions has evident advantages such as: better pathogenic destruction [7], makes the system less prone to be contaminated with methanogenic bacteria [8], better thermodynamics condition for biohydrogen production [9–11] and gives high hydrogen yield [12,13]. Therefore, hyper-thermophilic hydrogen production starts to attract the interest of researchers. Furthermore, hyper-thermophilic process is a preferred choice to treat wastes with high concentration of protein [14], as the pig wastes used in this study.

Most studies focused on hydrogen production from industrial wastes [6,15–19], household wastes [20,21] and synthetic media [22–24] and are carried out in different types of reactors CSTR, upflow anaerobic sludge blanket (UASB), etc. [2,25]. Each of the reactor's type has its own advantage. In CSTR system it is more easy to wash out methanogenic bacteria by choosing short HRT (HRT is equal to solid retention time (SRT) in CSTR system), while UASB reactors are less sensitive to the fluctuation of environmental parameters.

Although animal wastes can be considered as potential sources for biohydrogen production [26], only a few studies are available on this area. This is mainly due to the creation of favourable conditions for methane production from animal waste, which will eventually reduce or even eliminate hydrogen production.

Yokohama et al. [27] have used cow slurry to produce hydrogen under various temperature conditions. They found that it is possible to produce hydrogen in batch experiments only by incubating cow slurry without the presence of additives or seed bacteria. While, Tang et al. [28] have studied the effect of temperature and pH on H_2 production from cattle wastewater also in batch experiments. Venkata Mohan et al. [29] have examined the influence of chemical treatment, heat treatment and pH treatment as well as the combination of them on dairy wastes, to enhance biohydrogen production at the mesophilic range in batch mode. Zhu et al. [30] have used pig slurry mixed with a variety of nutrients and glucose to produce hydrogen at the mesophilic range in continuous digestion mode; they also used pH control to keep the pH at 5.3 in the system. Up to now biohydrogen production from animal wastes without any pre-treatment, pH control and additional feeding in continuous system is still lacking.

In the light of the above developments, this work uses mixed culture without any pre-treatment for hydrogen production in a continuous system from pig slurry under hyper-thermophilic temperature. The aim of this study is to investigate the performance of a hyper-thermophilic hydrogen production system which was designed in order to prevent methanogenesis and to produce hydrogen from pig slurry. In addition, this research provides valuable information towards the development of an effective anaerobic system for converting carbohydrates derived from animal wastes into hydrogen.

2. Materials and methods

2.1. Pig slurry and preparation of the feedstock

Raw slurry of 18.34% total solids (TS) was collected from a pig farm near by the laboratory (Thermi City, Greece). For the preparation of the experimental feedstock, the raw pig slurry was shredded into a blender for 5 min and then was screened using a 1 mm mesh size sieve to remove coarse materials so as to avoid potential clogging problems. At the end, it was diluted with distilled water to reach a solid content of 4.54% and stored at 4 °C. In order to create anaerobic conditions, the slurry was flushed for 5 min with 100% gas N_2 into the influent vessel. The characteristics of the feedstock used in the experiment are given in Table 1. No pre-treatment such as heating and acid treatments to prevent methanogenesis in the slurry, was performed.

2.2. Experimental set up and operation

The experiment was carried out in a hyper-thermophilic CSTR reactor R1 (750 cm^3 working volume) in combination with a mesophilic methanogenic reactor. The methanogenic reactor was connected with the R1 reactor in order to provide the inoculum. No pre-treatment of the inoculum was performed to inhibit methanogenic bacteria. The characteristics of the inoculum are shown in Table 2.

The experimental setup also consisted of a feed vessel, two peristaltic pumps, an effluent bottle (gas–liquid separator), a magnetic heating stirrer for the homogenization of the pig slurry and a gas meter. The schematic diagram of the system used in this study is shown in Fig. 1.

The reactor R1 was kept at 70 ± 1 °C with the assistance of the heating plate of the magnetic stirrer which was equipped with a temperature control unit. This reactor was continuously operated with HRT of 24 h. The inoculum was transferred from the methanogenic reactor to R1 with regulated flow of 46 $cm^3\,d^{-1}$, during the first 15 days and then it was stopped. The evolved biogas was collected into a gas bag (Aluminized Polyethylene Bag). The organic loading rate was

Table 1 – Characteristics of the feedstock used in the experiment.

Parameters	Feedstock ± SD[a]
Total solids (TS) (% w w^{-1})	4.54 ± 0.2
Volatile solids (VS) (% w w^{-1})	3.32 ± 0.2
Total kjeldahl nitrogen (TKN) (mg L^{-1})	2230 ± 123
Acetate (mmol L^{-1})	42.52 ± 0.851
Propionate (mmol L^{-1})	14.2 ± 1.125
n- and iso-Butyrate (mmol L^{-1})	3.87 ± 0.126
pH	6.7 ± 0.1

a SD, standard deviation.