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Anaerobic digestion of solid waste in RAS: effect of reactor type on the biochemical acidogenic potential (BAP) and assessment of the biochemical methane potential (BMP) by a batch assay

K.I. Suhr[a,*], C.O. Letelier-Gordo[b,1], I. Lund[a]

[a] National Institute of Aquatic Resources, DTU Aqua, Section for Aquaculture, Technical University of Denmark, 9850 Hirtshals, Denmark
[b] DTU Environment, Department of Environmental Engineering, Bioenergy Group, Technical University of Denmark, Anker Engelunds Vej 1 – Building 101A, 2800 Kgs. Lyngby, Denmark

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ABSTRACT

Anaerobic digestion is a way to utilize the potential energy contained in solid waste produced in recirculating aquaculture systems (RASs), either by providing acidogenic products for driving heterotrophic denitrification on site or by directly producing combustive methane. In this study the biochemical acidogenic potential of solid waste from juvenile rainbow trout was evaluated by measuring the yield of volatile fatty acids (VFA) during anaerobic digestion by batch or fed-batch reactor operation at hydrolysis time (HT)/hydraulic retention time (HRT) of 1, 5, or 10 days (and for batch additional 14 and 20 days) in continuously stirred tank reactors. Generally, the VFA yield increased with time and no effect of the reactor type used was found within the time frame of the experiment. At 10 days HT or 10 days HRT the VFA yield reached 222.3 ± 30.5 and $203.4 \pm 11.2\,\mathrm{mg\,VFA\,g^{-1}\,TVS_0}$ (total volatile solids at day 0) in batch and fed-batch reactor, respectively. For the fed-batch reactor, increasing HRT from 5 to 10 days gained no significant additional VFA yield. Prolonging the batch reactor experiment to 20 days increased VFA production further ($273.9 \pm 1.6\,\mathrm{mg\,VFA\,g^{-1}\,TVS_0}$, $n = 2$). After 10 days HT/HRT, 16.8–23.5% of total Kjeldahl N was found as TAN and 44.3–53.0% of total P was found as ortho-phosphate. A significant difference between reactor types was detected for the phosphorous dissolution at 5 days HT/HRT as a relatively steep increase (of a factor 2–3) in ortho-P content occurred in fed-batch reactors but similar steep increase was only notable after 10 days HT for batch reactors. No differences between reactor types at the other HT/HRT were recorded for P as well as (for all HT/HRT for) N. Based on this study a HRT of approximately 5 days would be recommended for the design of an acidogenic continuously stirred reactor tank in a RAS single-sludge denitrification set-up. The biochemical methane potential of the sludge was estimated to $318 \pm 29\,\mathrm{g\,CH_4\,g^{-1}\,TVS_0}$ by a batch assay and represented a higher utility of the solid waste when comparing the methane yield with the VFA yield (in COD units). This points toward a technological challenge of ultimately increase the acidogenic output to match the methane yield as both products are formed from the same source.

1. Introduction

In countries with a strict environmental legislation the continuous success of land-based aquaculture will rely on the ability to treat and reduce the nutrient pollution load cost-efficiently. A distinct feature of recirculating aquaculture systems (RASs) compared to earthen ponds aquaculture and open net-pens is the considerable amount of solid fish waste/fecal matter collected in RASs. This makes sludge management an important part of RAS operation (Cripps and Bergheim, 2000; Sharrer et al., 2010) for mitigating potential pollution problems. Currently, a common way of

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Abbreviations: ANOVA, analysis of variance; B, batch; BAP, biochemical acidogenic potential; BMP, biochemical methane potential; COD, chemical oxygen demand; FB, fed-batch; HRT, hydraulic retention time (fed-batch experiment); HT, hydrolysis time (batch experiment); RAS, recirculating aquaculture systems; SRT, sludge retention time; SS, suspended solids; STP, standard temperature and pressure (20 °C, 1 atm.); TAN, total ammonia nitrogen; TKN, total Kjeldahl nitrogen; TP, total phosphorus; TS, total solids; TVS, total volatile solids; VFA, volatile fatty acid.

[*] Corresponding author. Tel.: +45 3588 3273; fax: +45 3396 3260.
E-mail addresses: ksu@aqua.dtu.dk (K.I. Suhr), colg@aqua.dtu.dk (C.O. Letelier-Gordo), il@aqua.dtu.dk (I. Lund).

[1] Present address: National Institute of Aquatic Resources, DTU Aqua, Section for Aquaculture, Technical University of Denmark, 9850 Hirtshals, Denmark.

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treating the solid fish waste is by storage in sludge basins or Geotextile bags where the liquid part is drained off prior to spreading the sludge on agricultural land as fertilizer. An alternative use of the solid fish waste is utilization as a carbon source for denitrification (Aboutboul et al., 1995) or/and in anaerobic digestion for methane generation (Lanari and Franci, 1998). From an environmental point of view controlled digesters are preferred to sludge basins due to the diffuse greenhouse gas emissions released from the basins (Mirzoyan et al., 2012).

In wastewater literature quantification of the fermentable fraction of the organic matter (corresponding to the maximum VFA concentration obtained at anaerobic conditions) is called the biochemical acidogenic potential (BAP) (Ruel et al., 2002; Lie and Welander, 1997). This fraction represents the majority of the easily biodegradable carbon compounds produced by anaerobic digestion. In a complete anaerobic digestion process, this fraction of biodegradable organic matter is further degraded to methane (Appels et al., 2008). Anaerobic digestion studies of solid fish waste have shown BAP in the range of 0.13 g VFA g^{-1} TVS for salmon smolt (Conroy and Couturier, 2010) and 0.15–0.21 g VFA g^{-1} TVS for rainbow trout (Suhr et al., 2013). When using the biodegradable organic matter present in the system as electron donors in heterotrophic denitrification the term "single-sludge denitrification" is used. This process has the advantage of reducing both the organic matter and nitrate discharge from RAS (van Rijn et al., 2006). In experimental zero-discharge RASs, single-sludge denitrification is integrated in the RAS and only the surplus solid waste not used for denitrification is ultimately subjected to biogas reactor digestion (Tal et al., 2009). Mirzoyan et al. (2010) reviewed the field of solid fish waste use in anaerobic biogas digesters and found organic matter (COD and VS) digestion efficiencies of up to nearly 100%. The methane output, however, varied considerably (from 4 to >80% methane of total gas produced) due to differences in operating conditions in the various studies (e.g. reactor types, feed type, loading rate, hydraulic retention time, pH, salinity, temperature, composition, dilutions, etc.) (Mirzoyan et al., 2010).

The boost in denitrification rate by maximizing the dissolved and easily biodegradable part of the carbon source has been recognized for years (Barnard, 1974) and thus sludge pre-fermentors or side-stream hydrolysis operation is well established in conventional wastewater treatment plants (Pitman et al., 1992; Vollertsen et al., 2006). In the first part of the present study, we focused on acidogenic production in RAS sludge pre-fermentors for potential use in single-sludge denitrification. Such tanks should in general operate at SRT < 10 days to avoid competition and methane generation from the slow growing methanogens generally requiring a SRT of 10–28 days in anaerobic digesters (Metcalf and Eddy, 2003). In a second separate part of this study, the methane yield of the solid fish waste was assessed. The first step of anaerobic digestion (common for both acidogenic and methane production) is the hydrolysis of the suspended organic matter, which also commonly is the rate limiting step for the whole process (Eastman and Ferguson, 1981; van Lier et al., 2001). It has been shown that the electron acceptor conditions affect the development of the bacterial hydrolytic enzyme system (aerobic > anoxic > anaerobic) (Henze and Mladenovski, 1991) producing an apparent relationship between the specific growth rate and the specific enzyme production rate. The relative changes to this, however, are affected by the constituent microorganisms in the sludge and found to be more pronounced for pure culture systems than in activated sludge (Goel et al., 1997, 1998).

Anaerobic RAS sludge digestion studies are often performed in batch experiments (van Rijn et al., 1995; Conroy and Couturier, 2010) as this is most convenient. However, sludge hydrolysis and fermentation in a RAS facility will likely be operated in a kind of continuous or fed-batch mode due to the intermittent nature of the discharge from swirl separators and/or drum-filters. Theoretically, differences in environmental factors influence the microbial growth and thereby implicit their enzyme production. The objective of this study was to examine the effect of using a batch or fed-batch operational mode on BAP of RAS waste sludge in continuously stirred tank reactors. The simultaneous remineralization of N and P was also recorded in the study. Furthermore, in a separate trial the biochemical methane potential (BMP) of the same type of solid waste was assessed by a standardized (batch) assay for evaluating the yield at complete anaerobic digestion.

2. Materials and methods

2.1. The fish production unit supplying solid waste

The RAS consisted of 12 fish tanks each with a volume of 600 L and holding approximately 27 kg juvenile rainbow trout (*Oncorhynchus mykiss*) (i.e. 327 kg in total). Fish were daily fed 3925 g (Biomar Enviro 920, 3 mm) in total. The feeding was conducted during a 6 h period per day, and light was on 24 h a day. Water temperature was 15 °C and freshwater intake was 275 L h^{-1} in the system with a total volume of approximately 17–18 m^3. A 2 L container was mounted on the bottom of the outlet swirl-separator in each tank collecting the discharged solid waste and providing a means to observe if any feed waste occurred. The containers were discharged daily together with an additional refilling with RAS water. The discharged solid waste and additional flushing water from the 12 tanks were pooled together (44 L) before further distribution.

All experiments were performed during a period of 32 days.

2.2. Anaerobic hydrolysis and fermentation in batch (B) reactors

Subsamples from the pooled solid waste were incubated in 2 L Blue Cap bottles (in triplicate) at completely mixed conditions:mixing was achieved by magnetic stirrers. The bottles were closed (for maintaining anaerobic conditions) with 2-port screw caps; one port connected to a one-way valve allowing excess gas production to escape and the second port was used for sampling. Sampling (100 mL liquid $bottle^{-1}$) was performed at day; 0, 1, 2, 5, 10, 14, and 20, and will be referred to as hydrolysis time (HT). Nitrogen gas was used to reflux the liquid hydrolysate mixture from the sampling tube immediately after sample retrieval. These batch reactors were kept at room temperature (in the same room as the fed-batch reactors below). However, due to more vigorous stirring and better isolated container walls, the batch reactors experienced slightly higher temperature (17.3 ± 0.7 °C) than fed-batch reactors (15.4 ± 0.7 °C).

2.3. Anaerobic hydrolysis and fermentation in fed-batch (FB) reactors

The pooled solid waste was distributed while mechanically stirred (by a drilling machine with a whisk inserted) into 20 L reactors in quantities of 16 L day^{-1}, 4 L day^{-1} and of 2 L day^{-1}, corresponding to average hydraulic retention times (HRT) of 1.25 days, 5 days and 10 days in the reactors. Each HRT treatment was performed in duplicate (6 reactors in total).

The reactors were constantly stirred by a motor-driven propeller-whisk, at approximate 32 rpm. An equal amount of the reactor volume to be renewed daily by fresh substrate addition was discharged from the reactors immediately prior to the loading of new substrate. Temperature, pH and oxygen were measured in the daily discharge from the reactors.

Laboratory samples for analysis (in triplicate) from the reactors and the 'raw sludge' (i.e. the pooled and stirred discharged solid