Optimization of Biogas Production and *In-Situ* Upgrading via Hydrogenotrophic Methanogenesis in a Lab-Scale UASB Reactor

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Abstract The increasing demand for sustainable bioenergy has strengthened interest in biogas upgrading technologies that aim to produce pure CH_4 by separating CO_2 and all other impurities. This study investigates the *in situ* biomethanation of CO_2 by feeding H_2 into a 7 L UASB anaerobic digester treating synthetic and potato-processing wastewater. Initially, the reactor operated under varying HRT and OLR to assess baseline performance. Following stabilization, H_2 was introduced either into the feed line or the recirculation line via a microbubble generation system. Hydrogen addition to the feed increased biogas production by 18%, methane concentration to 70.9%, and reduced CO_2 to 5.2%. Transitioning hydrogen injection to the recirculation line improved methane purity (78.8%) and TOC removal (74.3%), while doubling hydrogen flow further enhanced biogas production (4.6 ± 0.7 L/day) and increased TOC removal to 83.97%, despite a slight drop in methane content (73.1%). Across all conditions, hydrogen feed minimized VFA accumulation and supported syntrophic stability. Overall, *in-situ* hydrogen injection proved effective for enhancing methane yield and biogas quality, highlighting its potential for sustainable biogas biomethanation from industrial wastewaters.

Keywords: Hydrogenotrophic methanogenesis; *In-situ* biogas upgrade & biomethanation; Microbubbles; UASB

1. Introduction

Anaerobic digestion offers a sustainable method for converting organic waste into bioenergy. However, the raw biogas produced from the process, contains significant CO_2, reducing its energy value. Hydrogenotrophic methanogenesis, where CO_2 is biologically converted to CH_4 using hydrogen, presents an eco-friendly upgrading alternative. The hydrogen can be produced renewably through water electrolysis. While *ex-situ* systems work well, they're only cost-effective at large biogas volumes. *In-situ* upgrading, particularly via H_2 addition, is gaining significant attention. Among reactor configurations, the Upflow Anaerobic Sludge Blunket (UASB) reactor stands out, as its granular sludge promotes superior microbe-wastewater interactions, thereby enhancing methane yields. A key challenge, however, lies in achieving efficient hydrogen transfer and distribution within the system. Microbubble injection could improve hydrogen transfer, potentially raising methane output by 36% (Cuff et al., 2021). This study focuses on optimizing methane production via *in-situ* biomethanation of CO_2 in a lab-scale 7L UASB reactor feeding H_2 via a microbubble generation system. It evaluates system performance under varying operating conditions using both synthetic and real industrial wastewater, aiming to optimize efficiency and scalability.

2. Materials and methods

2.1. Characterization of feedstock

The UASB reactor was inoculated with 2 L anaerobic granular sludge obtained from a potato processing industry (PepsiCo Hellas S.A. & Tasty Foods S.A., located in Agios Stefanos, Attica, Greece). The sludge exhibited a pH of 7.9, alkalinity of 675 mg/L, TS of 13.54%, VS of 3.46% and ash content of 10.08%. The potato processing wastewater (PPW) was obtained from the same potato processing industry and exhibited a pH of 6.8, conductivity of 2.3 mS/cm, alkalinity of 750 mg $CaCO_3$/L, TS of 4.15 g/L, VS of 2.45 g/L, TSS of 1320 mg/L, VSS of 1000 mg/L, COD of 3189.11 mg/L, sTOC of 1061.4 mg/L, sTN of 330.9 mg/L, and NH_4^+ of 174 mg/L. Synthetic wastewater (SW) that was used as substrate for the first 72 days consists of 36 g of D-glucose, 2 g of urea, and 2 g of molasses in a final volume of 1 L of water containing 3 g COD/L, 1.18 g TOC/L, and 0.2 g TN/L.

2.2 Operational conditions of the UASB reactor

The 7 L Plexiglas cylindrical UASB reactor receives influent from the bottom and separates biogas at the top via a gas-liquid separator. Anaerobic sludge and wastewater are recirculated at 0.9 m/h, with temperature maintained at 35 °C through an external water bath. ORP and pH are continuously monitored by a PLC system. H_2, generated from renewable-powered electrolysis, was introduced either into the influent or the recirculation line using a porous stainless-steel diffuser, promoting CO_2-to-CH_4 conversion through microbubble enhanced mass transfer. Biogas volume was measured via water displacement, and gas composition was assessed using a portable gas analyzer (GFM 406 series).

Influent and effluent samples were analyzed for COD, Alkalinity, NH_4^+-N, TSS, and VSS following Standard Methods. VFAs were quantified by GC-FID (Shimadzu GC-2010 Plus). TOC and TN were measured using a Shimadzu TOC analyzer (TOC-VCHS with SSM-5000).

The UASB underwent a 10-day start-up with 0.4 L/day synthetic wastewater, followed by 168 days of operation under varying conditions (Table 1). Hydrogen injection into the feed tank was initiated on day 106 and redirected to the recirculation line on day 143, initially once per day for 30 minutes at 35 mL/min, and from day 156, twice daily at the same rate.

Table 1. Operational parameters in each of the 8 phases.

Phase	**Days**	**HRT (d)**	**OLR (g/L d)**	**H_2 injection**	**Feed**
1	1-5	1.8	1.6	-	SW
2	6-21	0.9	3.2	-	SW
3	22-28	1.3	2.3	-	SW
4	29-72	1.3	2.3	-	PPW
5	73-105	1.8	1.8	-	PPW
6	106-142	1.8	1.8	Feed tank (32 mL/min)	PPW
7	143-152	1.8	1.8	Recirc. (1 L/d)	PPW
8	153-168	1.8	1.8	Recirc. (2 L/d)	PPW

3. Results and discussion

The 168-day operation of the UASB reactor showed distinct performance phases. TOC removal efficiency was initially 86.2 ± 2.1% but dropped to 61.1 ± 6.2% following an OLR increase and HRT reduction, indicating system overload (Phase 2). Adjustments of HRT and OLR, recovered TOC removal to 68.5 ± 1.7% (Phase 3). Transitioning to real PPW achieved TOC removal of 78.1 ± 11.2%, though still below typical UASB efficiencies (Phase 4). In Phase 5, under extended HRT, TOC removal improved to 88.8 ± 4.5%. H_2 injection (Phase 6) had minimal impact on TOC removal (88.1 ± 5.3%). Upon shifting H_2 to the recirculation line (Phase 7), TOC removal slightly decreased (74.3 ± 6.5%), but significantly improved in Phase 8, reaching 83.97 ± 5.78% where H_2 was doubled from Phase 7.

Influent TN initially increased during Phase 2, leading to elevated organic nitrogen levels in the effluent. Transition to PPW (Phase 4) improved nitrogen removal. After hydrogen injection (Phases 6–8), further TN and NH_4^+-N reductions were recorded, likely due to pH-driven shifts in nitrogen speciation and possible ammonia volatilization.

The pH remained stable around 7.7 during Phase 1 but dropped to 6.55 in Phase 2 due to organic overloading. Recovery was achieved through bicarbonate addition (Phases 3–4). During H_2 injection phases, pH stabilized near 8.1 (Phases 6–8), with alkalinity maintained above 2000 mg $CaCO_3$/L. ORP consistently remained within –450 to –580 mV throughout all phases, confirming stable anaerobic conditions.

Biogas production started at 1 L/day (Phase 1) and increased to 2.07 ± 0.75 L/day (Phase 2), despite VFAs accumulation. Recovery in Phase 3 (2.3 ± 0.36 L/day) and further stabilization in Phase 4 (2.5 ± 0.6 L/day) were observed. Under stabilized conditions (Phase 5), production rose to 2.8 ± 0.7 L/day. Hydrogen injection into the feed (Phase 6) boosted production by 18% (3.3 ± 0.7 L/day), and injection into recirculation (Phase 7) further increased it to 3.4 ± 1.1 L/day. Doubling hydrogen flow in Phase 8 resulted in the highest biogas production, 4.6 ± 0.7 L/day. Methane content improved from 47% (Phase 1) to 70.9% (Phase 6), peaked at 78.8% (Phase 7), and slightly declined to 73.1% in Phase 8. The CH_4/CO_2 ratio increased from 9.06 (Phase 5) to 13.63 with hydrogen injection into the feed line (Phase 6), though with moderate stability. Hydrogen recirculation (Phases 7–8) yielded slightly lower ratios (10.37–9.82) but markedly enhanced biogas volume, TOC removal, and reactor resilience.

A significant VFA buildup was observed in Phase 2 (1122.4 ± 372.8 mg/L), attributed to increased OLR. VFAs gradually decreased in Phases 3-5, reflecting system recovery. During hydrogen injection (Phases 6–8), VFAs remained at low concentrations (<200 mg/L), with propionic acid nearly eliminated, indicating enhanced syntrophic activity and process stability.

4. Conclusions

H_2 microbubble injection improved methane production, biogas quality, and TOC removal in a UASB reactor treating PPW. Shifting hydrogen addition to the recirculation line further enhanced system stability, confirming the potential of *in-situ* hydrogenotrophic methanogenesis for efficient biogas methanation.

References

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