



Zero-valent iron enhanced anaerobic digestion of pre-concentrated domestic wastewater for bioenergy recovery: Characteristics and mechanisms



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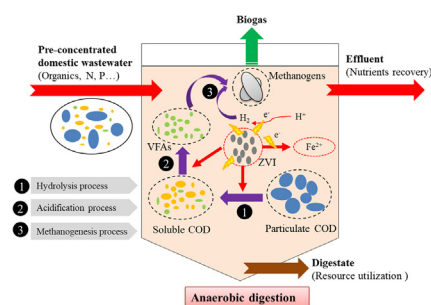
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GRAPHICAL ABSTRACT



ARTICLE INFO

Keywords:

Zero-valent iron
Anaerobic digestion
Domestic wastewater
Biochemical methane production
Bioenergy recovery

ABSTRACT

Pre-concentrated domestic wastewater (PDWW) rich in organic matters can be a suitable substrate for anaerobic digestion (AD) towards holistic resource and bioenergy recovery. Micron zero-valent iron (ZVI) was applied in designed batch experiments during anaerobic treatment of PDWW to verify its roles in performance enhancement and associated mechanisms. In the selected range of food to microorganism (F/M) ratio, 0.5 gCOD/gMLVSS was most appropriate as biomethane production potential (BMP) of 0.275 L CH₄/gCOD was obtained. The optimal ZVI dosage at fixed F/M of 0.5 was 6 g/L, further enhancing the BMP by 15.2%. Furthermore, ZVI improved the hydrolysis process (producing more soluble organics) and regulated acidification process (affecting volatile fatty acids distribution). No obvious impact on acetoclastic and hydrogenotrophic methanogenesis processes was noted with ZVI addition. ZVI based AD of the PDWW is promising for promoting the practical application of advanced domestic wastewater treatment strategy (pre-concentration plus anaerobic digestion).

1. Introduction

Wastewater treatment using conventional aerobic biological processes can solve the concerned environmental pollution issue by

effectively removing various pollutants. However, large amount of non-renewable energy is consumed without serious consideration on recycling available resources in wastewater. Nowadays, increasing attention is thus paid to more sustainable alternatives for wastewater

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<https://doi.org/10.1016/j.biortech.2020.123441>

Received 1 March 2020; Received in revised form 21 April 2020; Accepted 22 April 2020

Available online 24 April 2020

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treatment such as anaerobic digestion (AD). The advantages of AD process over its aerobic counterpart are lower energy consumption, less sludge production and smaller process footprint as well as biogas generation (Li et al., 2016). It has been estimated that applying AD-oriented system can even realize net energy recovery for domestic wastewater treatment (McCarty et al., 2011). However, at psychrophilic or cold temperature conditions using low-strength domestic wastewater for direct AD is challenging, which is more applicable for treating industrial wastewater or solid wastes with high organics content (Ekama et al., 2007).

To realize effective AD of domestic wastewater, pre-concentration technology has gained extensive attention to capture organic matters from the wastewater. One of the promising wastewater pre-concentration processes is membrane filtration, including high-loaded membrane bioreactor (Akanyeti et al., 2010), ultrafiltration or microfiltration (Kimura et al., 2017), forward osmosis filtration (Ansari et al., 2018) and dynamic membrane filtration (Li et al., 2017). Membrane-based pre-concentration processes can separate wastewater into two parts, the concentrate containing sufficient organic carbon source and the filtrate rich in nutrients (nitrogen and phosphorus). With influent chemical oxygen demand (COD) of 270–520 mg/L, pre-concentrated domestic wastewater (PDWW) containing COD more than 2000 mg/L is expected to be an effective substrate for AD (Mezohegyi et al., 2012; Gao et al., 2018). However, PDWW generally showed an altered property compared to the raw wastewater, because SCOD only accounted for 5%–10% of the total COD in PDWW and substantial retention of nutrients, suspended particles and even salts was also noted, showing inhibition effects on biogas production in some cases due to ammonia and salt accumulation (Gao et al., 2019). In biochemical methane potential (BMP) batch tests, PDWW achieved by membrane filtration could reach the methane yield of 200 mLCH₄/g COD or even more (Xiong et al., 2019; Gao et al., 2019). The results illustrate that PDWW may be partially utilized by anaerobes, so methane production is substantially lower than 350 mLCH₄/g COD, which is the theoretical methane yield at standard conditions. Thus, how to enhance the methane production of PDWW should be concerned, as little attention has been paid to the application feasibility of PDWW in AD process.

As documented, the membrane pre-concentration showed preferred retention of particulate and colloidal organic matter (70%–90% of TCOD) in the concentrate (Akanyeti et al., 2010; Li et al., 2017; Gao et al., 2018), while challenging the bio-methanation process due to low organics conversion rates. Though some pretreatment measures, such as thermal, chemical and mechanical methods (Zhen et al., 2017), have been proposed for improving AD process for organic wastes, however high energy consumption or chemical inputs made them costly (Wei et al., 2018a,b) and reduced effectiveness to treat organic substrates with high water content (like the PDWW) might limit their applications. Recently, zero-valent iron (ZVI) has attracted increasing attention to degrade organic pollutants (Wang and Tseng, 2009; He et al., 2015) and enhance methane generation during AD (Wei et al., 2018a,b), due to its low input energy compared with other techniques. ZVI can play important roles in enhancing AD performance with major reasons listed as follows: (1) ZVI affects solution chemistry and anaerobic environment by regulating pH and ORP that promotes methanogenesis (Carpenter et al., 2015); (2) ZVI can effectively accelerate the hydrolysis–acidification process (Feng et al., 2014), and higher propionate conversion rate is also noted after ZVI addition (Meng et al., 2013); (3)

adding ZVI can alter the microbial community structure evidenced by enriched syntrophic-methanogenic associations (Pan et al., 2019).

Furthermore, ZVI enhancing methane production was observed for diverse substrates at varied dosage. It was pointed out the methane yield of excess sludge increased by 131.6% in the presence of micro-scale ZVI with the dosage of 10 g/L (Yu et al., 2016). When 20 g/L of ZVI added to pig manure, methane yield was enhanced by 20% (Liang et al., 2017). Dosing 12 g/L of ZVI also enhanced methane yield of municipal solid waste by 41.7% (Kong et al., 2018). It is recognized that within the suitable dosage range ZVI plays positive roles in anaerobic degradation of organic wastes. However, limited investigation has been conducted for PDWW treatment, thus it is of great interest to verify the influence of ZVI on methane production of PDWW and also the potential mechanisms.

Therefore, in this study the objectives are: (1) to examine the methane production potential of PDWW at varied F/M ratios; (2) to investigate the effect of ZVI addition on enhancing methane production of PDWW; (3) to clarify the major mechanisms involved. Bath tests, including BMP and specific methanogenic activity (SMA), were conducted with some critical parameters determined, including the biogas production and composition, soluble chemical oxygen demand as well as volatile fatty acids. To date, quite limited investigations have been done to verify the effect of ZVI on anaerobic digestion of pre-concentrated domestic wastewater. The results will provide a better understanding of ZVI-based anaerobic wastewater treatment process towards bioenergy and resource recovery.

2. Materials and methods

2.1. Zero-valent iron

The ZVI in micron size (purity greater than 98%, 100 mesh) purchased from Aladdin Corp., China was used in the experiments. High purity nitrogen gas has been used to protect the ZVI (sealed in bottles) from contact with air.

2.2. Substrate and seed sludge

The substrates (PDWW) used for a series of batch experiments (except methanogenic process) during anaerobic digestion were the concentrate of real domestic wastewater. As for examining the effect of adding ZVI on methanogenic process, synthetic substrates were applied, including various VFAs and H₂/CO₂ with details shown in Section 2.3.4. In this work, the designed batch tests included the influence of food to microorganism (F/M) ratio, the selection of appropriate ZVI dosage to enhance methane production as well as the effect of adding ZVI on the hydrolysis, acidification and methanogenic processes. The PDWW was obtained from a dynamic membrane filtration (DMF) reactor with the raw wastewater collected from a wastewater treatment plant as described elsewhere (Xiong et al., 2019). The quality of the PDWW is summarized in Table 1. Although the COD and SCOD of tested substrates showed some fluctuations, the ratios of SCOD to COD are between 9% and 17%, while the ratios of volatile solids (VS) to total solids (TS) are between 72% and 77%. In addition, PDWW contains a substantial amount of soluble proteins (52.72 ± 10.95 mg/L) and polysaccharides (8.48 ± 4.74 mg/L).

Seed sludge used in the experiments was obtained from a local full-

Table 1

Properties of the PDWW used in batch tests.

Batch tests	COD (mg/L)	SCOD (mg/L)	SCOD/COD (%)	TS (g/L)	VS (g/L)	VS/TS (%)	Polysaccharide (mg/L)	Protein (mg/L)
F/M ratio	2483 ± 155	228.7 ± 21.5	9.2	2.15 ± 0.05	1.66 ± 0.05	77.2	–	–
ZVI dosage	2528 ± 147	439.6 ± 32.7	17.4	1.55 ± 0.02	1.13 ± 0.01	72.9	–	–
Hydrolysis - acidification	1207.5 ± 82.5	137.1 ± 11.6	11.4	1.45 ± 0.04	1.05 ± 0.03	72.4	8.48 ± 4.74	52.72 ± 10.95

scale mesophilic anaerobic digester (Yang et al., 2020). The TS and VS of seed sludge used in all tests were 11.09 ± 0.89 g/L and 3.44 ± 0.43 g/L, respectively.

2.3. Design of batch experiments

2.3.1. Influence of F/M ratio on AD of the PDWW

To examine the effect of F/M ratio (gCOD/gMLVSS) on anaerobic digestion of PDWW, 20 mL seed sludge and certain amount of substrate (PDWW) were added into 120 mL serum bottles with a working volume of 80 mL by diluting with deionized water if necessary, and the F/M ratios of 0.2, 0.5, 0.8 and 1.0 were chosen according to the preliminary experiments with F/M ratios ranging from 0.2 to 1.8. Blank control was conducted using deionized water to replace the substrate with other conditions the same as aforementioned. Duplicate tests were conducted for each F/M condition. All of the bottles were sealed and then purged with high purity nitrogen gas according to previous study (Yang et al., 2020). Afterwards, the bottles were placed in a water bath device under continuous shaking speed of 120 rpm at mesophilic temperature (35 °C).

2.3.2. Influence of ZVI dosage on AD of the PDWW

After the determination of the optimal F/M ratio in the previous section, batch tests were adopted to examine the influence of ZVI dosage on anaerobic digestion at the optimal F/M ratio of 0.5. The ZVI was directly added into serum bottles (120 mL) at the dosages of 0, 2, 4, 6 and 8 g/L. Before starting batch tests, the same pre-treatment procedures including sealing and nitrogen gas flushing were conducted. Other operational conditions were the same as described in Section 2.3.1.

2.3.3. Influence of ZVI on hydrolysis-acidification of the PDWW

In order to further explore the mechanisms of ZVI addition enhancing AD process, the influences of ZVI on the hydrolysis-acidification process were firstly investigated under well-controlled experiments. To ensure the AD process terminated at hydrolysis-acidification stage, a methanogenic inhibitor, sodium 2-bromoethanesulfonate ($C_2H_4BrO_3S$, BES), was used to inhibit methanogenic process at a BES concentration of 50 mmol/L (Lu et al., 2018).

The detailed design of batch experiments is shown in Table 2. Specifically, two sets of batch experiments were carried out based on with or without BES addition. Each set covered three groups of batch test, namely negative control group (with seed sludge addition only), positive control group (with seed sludge and substrate addition), and ZVI addition group (with seed sludge, substrate and 6 g/L ZVI addition). 60 mL of seed sludge was added into anaerobic bottles with total working volume of 210 mL (replenished by deionized water if necessary). Duplicate tests were done for each condition. The pre-treatment methods and operation conditions were the same as those mentioned in Section 2.3.1.

2.3.4. Influence of ZVI on methanogenesis of the PDWW

To explore the effects of ZVI on acetoclastic methanogenesis and hydrogenotrophic methanogenesis processes, two kinds of substrate were adopted, namely, volatile fatty acids (VFAs) and H_2/CO_2 (Zhao

et al., 2018). The VFAs used as the substrate were further divided into three categories: acetic acid alone, propionic acid alone, as well as a mixture containing acetic acid and propionic acid (COD ratio of 5: 1). Various VFAs (at fixed F/M = 0.5), 6 g/L ZVI and 20 mL seed sludge were added into 120 mL anaerobic bottles (80 mL effective volume). As for experiment utilizing H_2 and CO_2 as the substrates, 20 mL seed sludge and 6 g/L ZVI were put into the anaerobic bottle, then the bottle was purged with the hydrogen and carbon dioxide gas mixture (volume ratio of 8: 2) for 5 min. The gas component was measured every 2 h and then filled 40 mL mixed gas into the bottles to keep the 1.4 times of standard atmospheric pressure. Blank control was conducted without ZVI addition, and duplicate tests were carried out.

2.4. Data analysis

Batch tests results regarding the impact of ZVI on the methanogenic process were simulated by the modified Gompertz equation (Eq. (1)) using Origin 8.0 software (Origin Cop., USA).

$$P = P_0 \cdot \exp \left\{ -\exp \frac{R_{\max} \cdot e}{P_0} \cdot [(t_0 - t) + 1] \right\} \quad (1)$$

where P is cumulative methane production (mL CH_4 /gCOD), P_0 is the maximum potential of methane production (mL CH_4 /gCOD), R_{\max} is the maximum methane production rate (mL CH_4 /gCOD-d), $e = 2.7183$, t_0 is the lag time (day) and t is the cultivation time (day).

2.5. Analytical methods

Biogas production was tested manually by using a glass syringe, and biogas composition (mainly CH_4 , CO_2 , H_2 and N_2) was detected using a gas chromatograph (GC) (GC7900, Tianmei, China) (Yang et al., 2020). To analyze the content of VFAs and lactate acid, a liquid chromatograph (LC-20A, Shimadzu, Japan) was used, which was equipped with an ultraviolet detector and a column (Aminex HPX-87H Column, Bio-Rad, USA). The mobile phase was 0.05 mol/L sulfuric acid buffer liquid at a flow velocity of 0.3 mL/min. The detector temperature was set at 40 °C with UV wavelength of 210 nm. The modified Lowry method was used to measure proteins (Lowry et al., 1951), and Anthrone-sulfuric acid method was adopted to measure polysaccharides (Gaudy, 1962). Measurements for SCOD, COD, TS and VS were based on the standard methods (APHA, 1998).

3. Results and discussion

3.1. Effect of F/M ratio on anaerobic digestion of PDWW

The effect of F/M ratio (0.2–1.0) on accumulative methane production using PDWW as the substrate is shown in Fig. 1(a) according to batch BMP tests lasting for 36 d, with the final cumulative methane production presented in Fig. 1(b). The maximum cumulative methane production reached 0.275 L CH_4 /gCOD at F/M ratio of 0.5, while the values were 0.265, 0.266 and 0.263 L CH_4 /gCOD when the F/M ratios were 0.2, 0.8 and 1.0. Thus, F/M ratio of 0.5 is considered to be optimal for anaerobic digestion of PDWW at least under the test conditions applied. However, the accumulative methane production of PDWW was

Table 2

Tests designed to verify the effect of ZVI addition on hydrolysis-acidification process.

Tested group	Seed sludge(mL)	Substrate (mL)	BES (mmol/L)	ZVI (g/L)
Negative control without BES	60	–	–	–
Positive control without BES	60	90	–	–
6 g/L ZVI addition without BES	60	90	–	6
Negative control with BES	60	–	50	–
Positive control with BES	60	90	50	–
6 g/L ZVI addition with BES	60	90	50	6

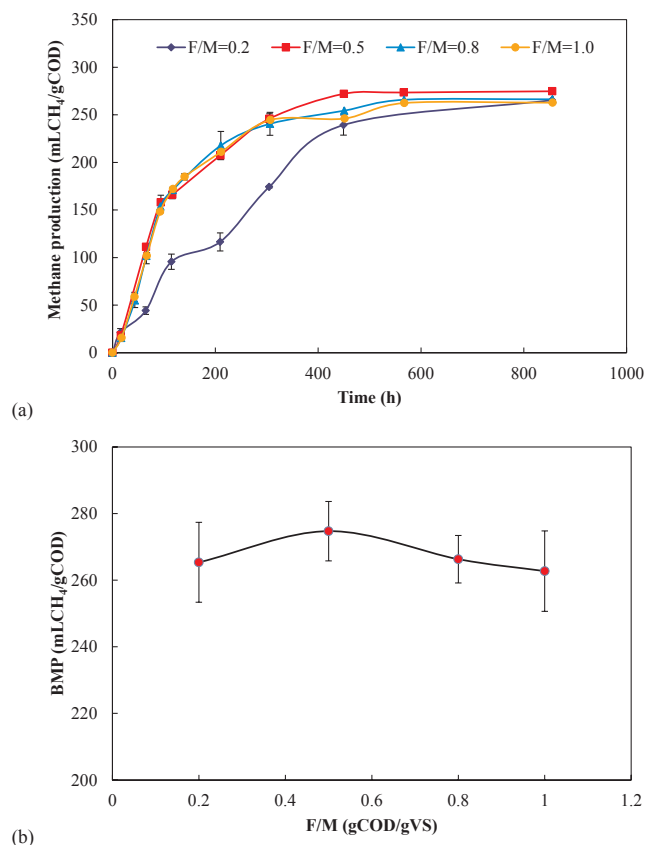


Fig. 1. The effect of F/M ratio on PDWW during AD: (a) variation of accumulative methane production and (b) BMP at different F/M ratios.

approximately 0.26–0.28 L CH₄/g COD when the F/M ranged from 0.5 to 1.0, still lower than theoretical methane yield. On the other hand, the reported methane yields during anaerobic treatment of real domestic wastewater were from 0.13 to 0.27 L CH₄/g COD (Ozgun et al., 2013; Gouveia et al., 2015). It meant that methane production seemed to be unaffected after pre-concentration treatment of domestic wastewater. Besides, compared with reported BMP values of sewage sludge (0.18–0.21 L CH₄/g COD) (Ding et al., 2017), PDWW showed higher methane production potential.

It has been reported that the duration for biogas production to be stable in BMP tests was less than 15 d when using food waste, sewage sludge and maize as the substrates (Koch et al., 2017; Zhang et al., 2007). However, the duration using PDWW as the substrate was between 17 and 23 d. Anaerobic digestion of PDWW appeared to be slower, possibly due to the difference in composition between organic solid wastes and PDWW. The SCOD/COD of PDWW used was 9.2%, illustrating that high content of particulate organic matters existed in PDWW, therefore AD process might be limited at the hydrolysis stage. Overall, the results indicated that PDWW is suitable for methane production and the most appropriate F/M ratio is 0.5, though the methane production needs to be improved and the duration of AD process should be reduced.

3.2. Effect of ZVI dosage on anaerobic digestion of PDWW

To verify the effect of ZVI addition on AD of the PDWW, batch experiments results at F/M ratio of 0.5 lasting for 20 d with a series of ZVI dosage are shown in Fig. 2(a). Firstly, the final accumulative methane production of the control is 0.254 L CH₄/g COD, which reaches 72.6% of the theoretical methane production and is approximately equal to VS/TS ratio (72.9%) of the substrate used in batch tests. It is

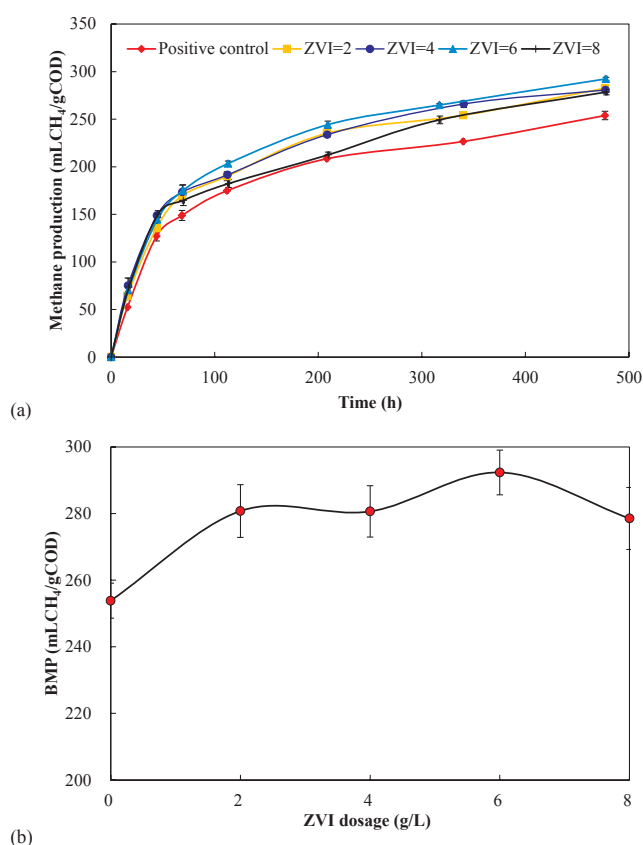
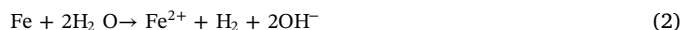


Fig. 2. The effect of ZVI dosage on PDWW during AD at F/M of 0.5: (a) variation of accumulative methane production and (b) BMP at different ZVI dosages.

supposed that the available organics in PDWW for methane production is related to the content of volatile organic substances.

Compared with the control, BMP in the test group with 6 g/L ZVI addition was increased by 15.2%, while BMP was improved by 10.6%, 10.4% and 9.7% with 2, 4 and 8 g/L of ZVI addition (Fig. 2(b)). Therefore, the most appropriate dosage of ZVI enhancing methane production was regarded as 6 g/L at F/M of 0.5. It has been reported that simulated brewery wastewater exposed to ZVI showed a methane production increasing rate ranging from 5% to 28.3% (Carpenter et al., 2015). In other studies, ZVI has enhanced methane production by at least 20% using sewage sludge as the substrate (Hu et al., 2019; Wei et al., 2018a,b). Thus, how methane production of PDWW was enhanced by ZVI addition deserves further investigation to clarify the underlying mechanisms.

Furthermore, it is not the fact that the higher the ZVI dosage is, the better the methane production performance will be. ZVI can release of H₂ and OH⁻ in anaerobic conditions as expressed by the following equation (Eq. (2)).



Consequently, why the high dosage of ZVI may inhibit methanogenesis can be explained as follows. High H₂ partial pressures due to rapid H₂ production at excess ZVI addition will lead to several negative effects on methanogenesis process (Yang et al., 2013). For instance, high partial pressures of H₂ bring about CO production ($\text{CH}_3\text{COO}^- + \text{H}^+ + \text{H}_2 \rightarrow \text{CH}_4 + \text{CO} + \text{H}_2\text{O}$, $\Delta G^0 = -15.8 \text{ kJ/mol}$) breaking the balance among CH₄, CO₂, and CO thus inhibiting acetoclastic methanogenesis (Zinder and Anguish, 1992). Moreover, high hydrogen partial pressure also makes the methanogenic process thermodynamically unfavorable (Yang et al., 2013). On the other hand, strong reduction action of ZVI is harmful to bacteria and methanogens

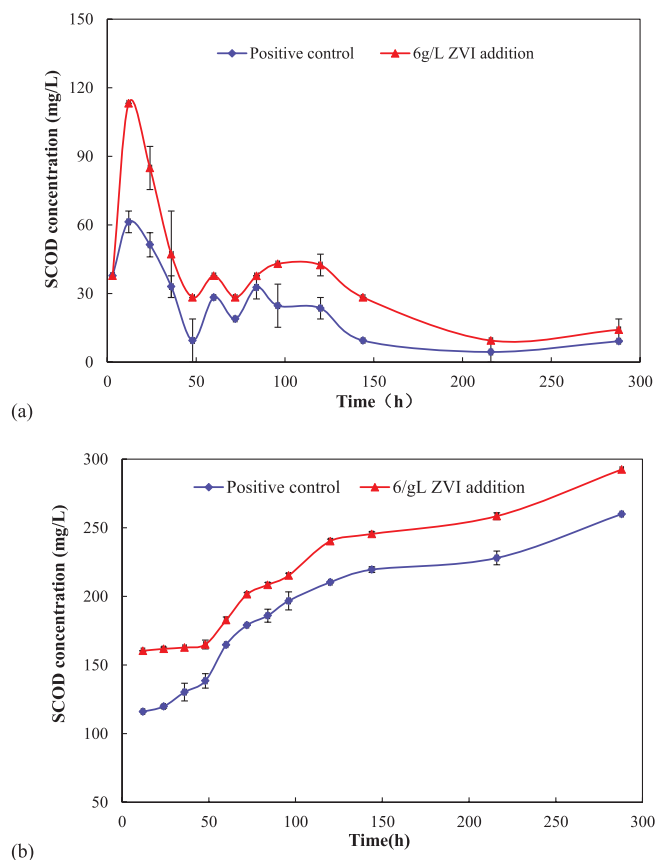


Fig. 3. The effect of ZVI on hydrolysis-acidification process: (a) variation of SCOD without BES addition and (b) variation of SCOD with BES addition.

extremely by decomposing functional groups of proteins on cell membranes and destructing respiration occurring on the surface of the cell membranes (Xu et al., 2019a,b).

3.3. Effect of ZVI on hydrolysis-acidification process during AD

3.3.1. Effect of ZVI on hydrolysis process

During hydrolysis stage (the first and commonly rate-limiting stage of AD process), complex organic substances were decomposed into monosaccharides and disaccharides, amino acids, peptides and others under the action of extracellular hydrolase of hydrolytic bacteria. SCOD chosen as an indicator was measured to investigate the influence of ZVI on PDWW hydrolysis process.

Substrates were hydrolyzed and eventually converted into methane in the two groups without BES (methanogenic inhibitor) as mentioned in Section 2.3. As shown in Fig. 3(a), SCOD in both tests groups without BES addition (positive control without ZVI addition, the other with 6 g/L ZVI addition) showed an increasing trend firstly (within 12 h) and then decreased to around or below 30 mg/L. It was presumed that easily biodegradable organics in the substrate were substantially hydrolyzed (within 12 h), causing the rate of hydrolysis process higher than the rate of methanogenesis process, thus SCOD concentrations increased at the initial stage. However, along with limited available organics being degraded, the rate of hydrolysis process gradually reduced, causing SCOD concentrations to be reduced and stable. Compared with the positive control, ZVI addition group showed the largest SCOD increment of 51.88 mg/L at 12 h, and also presented higher SCOD continuously during the remaining cultivation time. It is preliminarily verified that ZVI promotes the hydrolysis process during anaerobic digestion of PDWW.

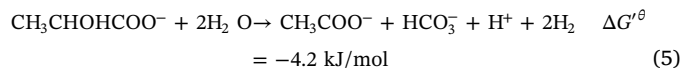
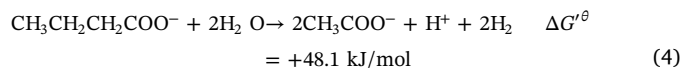
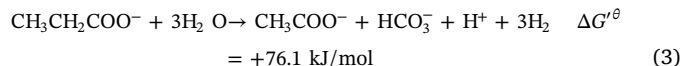
On the other hand, when BES is used for preventing methane

production SCOD will not be consumed for methanogenesis, but accumulated or converted as metabolic products (such as VFAs). Accumulative SCOD is measured and shown in Fig. 3(b). Similarly, the largest increment in SCOD of 44.34 mg/L at 12 h was noted for ZVI addition group compared to the control group. In addition, in both groups SCOD gradually increased with the fermentation time, while ZVI addition group always showed higher SCOD (33 mg/L after 120 h) than the control one. The results further verified that ZVI addition accelerated the hydrolysis process. It is mainly because ZVI as an electron donor promotes the breaking of unsaturated chemical bonds of organic substances in the hydrolysis stage. Moreover, iron is a necessary element for synthesizing related enzymes to fulfil extracellular hydrolysis process (Yang et al., 2013).

3.3.2. Effect of ZVI on acidification process

To investigate ZVI addition affecting acidification process (including acidogenesis and acetogenesis), the accumulation and behaviors of VFAs (mainly acetic acid) and lactic acid were analyzed since in the continuous anaerobic fermentation stage acidogenic and acetogenic processes can not be completely separated. In tested groups without BES addition VFAs and lactic acid were not accumulated with the total contents all below 15 mg/L due to their continuous conversion to biogas by the methanogens. As for the tested groups with BES addition, hydrolyzed organics are supposed to mainly convert into VFAs rather than methane production. The distribution of VFAs and lactic acid with time in three groups (negative control with BES addition, positive control with BES addition and 6 g/L ZVI addition) is shown in Fig. 4. The results of the positive control group (Fig. 4(b)) showed that acetic acid was the major product during PDWW acidification stage, with acetic acid accounting for more than 70% of the total products followed by other VFAs and lactic acid. Similar results regarding total VFAs content were noted for 6 g/L ZVI addition group (Fig. 4(c)). It was indicated that adding ZVI seemed to show little impact on the acidogenesis process as a similar level of organic acids accumulation was obtained. However, a minor difference in VFA distribution was noted as lactic acid content in ZVI addition group was lower than that in the positive control, showing potential impact on acetogenesis processes by regulating acetic acid production from other VFAs. This phenomenon was in agreement with previous studies reporting enhanced propionic and butyric acids conversion to acetic acid by ZVI addition (Meng et al., 2013; Kong et al., 2018).

Furthermore, it is well known that acetic acid can be directly used by acetoclastic methanogens for methane production. Therefore, the obtained results indicated that PDWW was a suitable substrate due to the high proportion of acetic acid. But a substantial accumulation of propionic acid followed by butyric acid and isovaleric acid was also noted. Propionic acid accounts for around 10% of the final fermentation products since it has been reported that protein-containing wastewater is prone to produce propionic acid.



As shown in Eqs. (3) and (4), the production of hydrogen and acetic acid from propionic acid and butyric acid cannot proceed spontaneously under standard conditions, unless at low partial pressure of H_2 (less than 10 Pa) with the actual free energy less than 0. Generation of acetic acid from lactic acid (Eq. (5)) seemed to be not easily proceeded as well. Moreover, approximate 30% of propionic acid and other VFAs existing in PDWW fermentation products is responsible for low methane

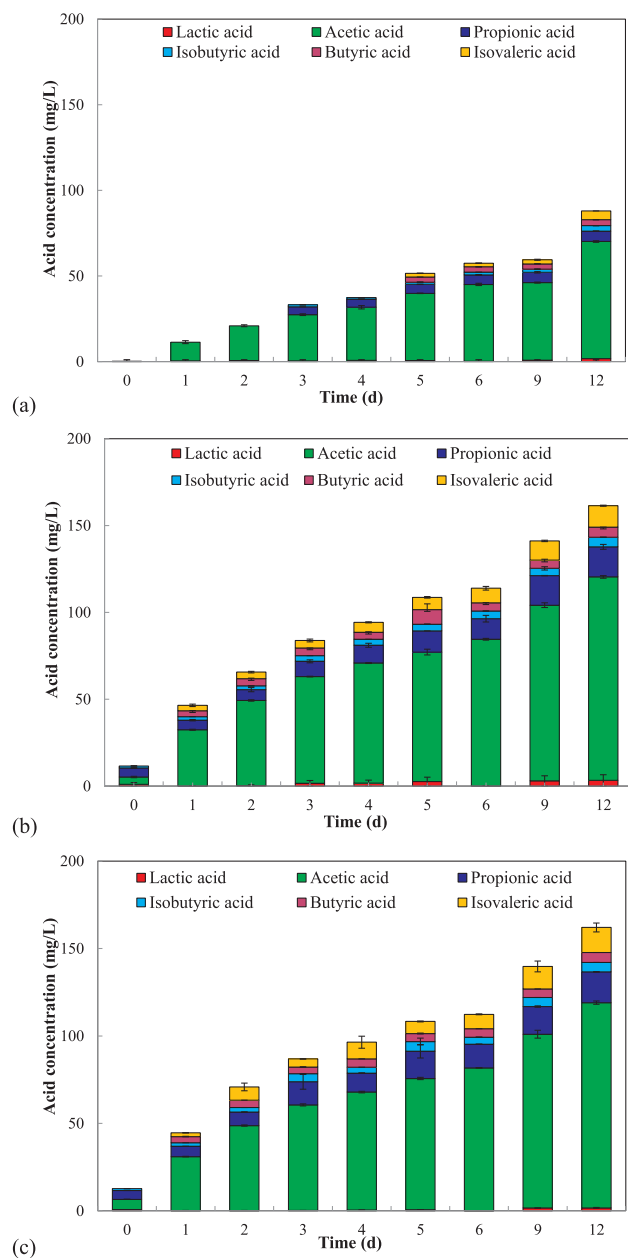


Fig. 4. Organic acids accumulation with BES addition: (a) negative control; (b) positive control and (c) 6 g/L ZVI addition.

production rate using PDWW as the substrate, possibly limited by the low conversion rate of these VFAs to acetic acid even with ZVI addition (Huang et al., 2019).

3.4. Effect of ZVI on methanogenesis process during AD

3.4.1. Effect of ZVI on acetoclastic methanogenesis process

Generally, it is recognized that in anaerobic digestion methane produced by acetoclastic methanogenesis process (acetic acid as the substrate) accounts for 70%, with the rest attributed to hydrogenotrophic methanogenesis process (H_2/CO_2 as the substrate). Batch tests were conducted to explore the effects of ZVI on acetoclastic methanogenesis using various VFAs as the organic substrates, including acetic acid (Fig. 5(a)), propionic acid (Fig. 5(b)) and a mixture of acetic acid and propionic acid (Fig. 5(c)). The results regarding accumulative methane production were simulated by using Gompertz equation with detailed parameters presented in Table 3.

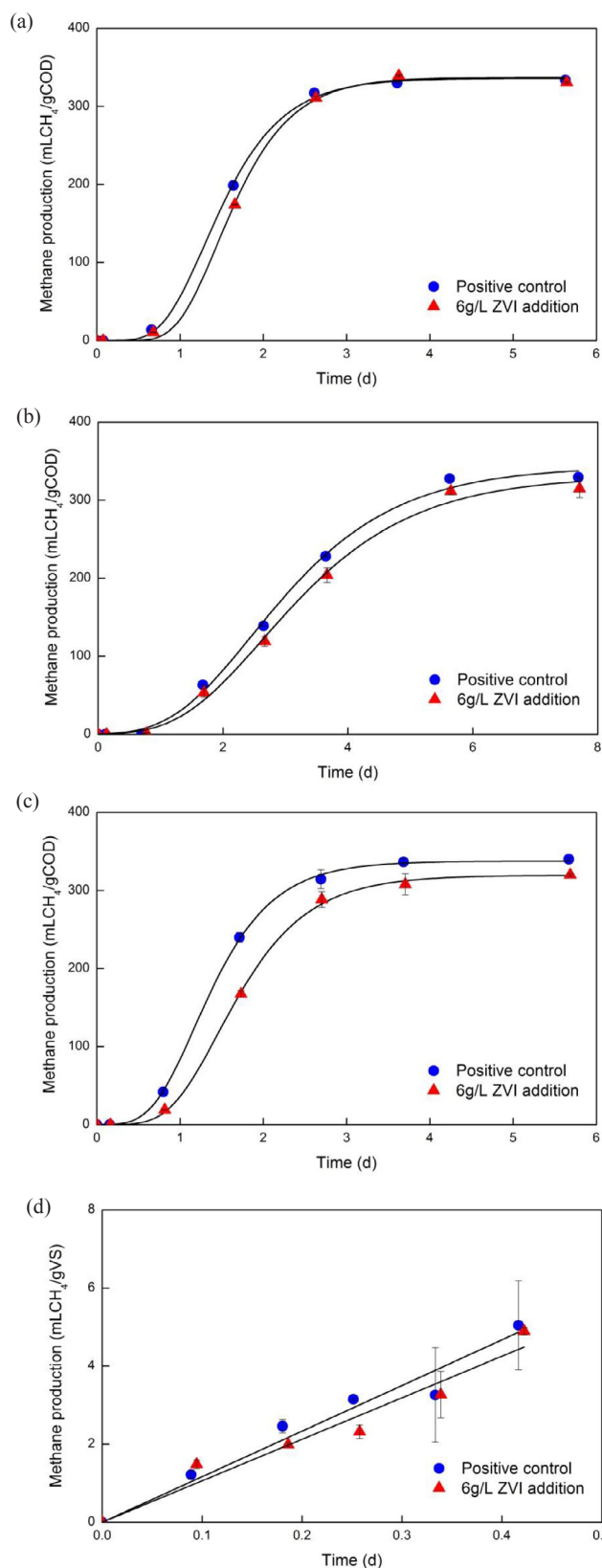


Fig. 5. The effect of ZVI on methanogenesis process with various substrates: (a) acetic acid; (b) propionic acid; (c) acetic acid and propionic acid mixture (5: 1) and (d) H_2 and CO_2 mixture (8: 2).

Table 3

Influence of ZVI addition (6 g/L) on acetoclastic methanogenesis process by simulating Gompertz equation.

Substrate	Sample	P ₀ (LCH ₄ /gCOD)	R _{max} (L CH ₄ /gCOD)	t ₀ (d)
Acetic acid	Positive control	0.336	0.241	0.8
	ZVI addition	0.337	0.255	1.0
Propionic acid	Positive control	0.344	0.098	1.2
	ZVI addition	0.331	0.091	1.3
Acetic acid and propionic acid	Positive control	0.338	0.240	0.7
	ZVI addition	0.319	0.199	0.9

Although there were no obvious differences in methane production potential (P₀) and lag time (t₀), the maximum methane production rate (R_{max}) with 6 g/L ZVI addition group was 6% greater than the positive control when the substrate was acetic acid. However, when the substrates were changed to propionic acid or the mixture of acetic acid and propionic acid, the methane production potential with 6 g/L ZVI addition group was 4%–6% lower than the positive control, while the maximum methane production rate with 6 g/L ZVI addition group was 7%–17% lower than the positive control. This can be explained that hydrogen production by ZVI dissolution (shown in Eq. (5)) will be thermodynamically detrimental for propionic acid conversion into acetic acid, and subsequently acetoclastic methanogenesis can be inhibited due to less available acetic acid (Capson-Tojo et al., 2017). However, the reduction of BMP with ZVI addition was insignificant, thus ZVI seemed to have no obvious effect on the acetoclastic methanogenesis process with the substrates dominated by acetic acid.

3.4.2. Effect of ZVI on hydrogenotrophic methanogenesis process

As shown in Fig. 5(d), the effect of ZVI on hydrogenotrophic methanogenesis process is also investigated. It was noted that accumulative methane production increased linearly with batch test time, and the methanogenesis rate could be obtained accordingly. The hydrogenotrophic methanogenesis rates of positive control and 6 g/L ZVI addition group were 0.012 and 0.011 LCH₄/gMLVSS-d, indicating no enhancement of hydrogenotrophic methanogenesis by adding ZVI. It was different from one previous study reporting that methane production from hydrogenotrophic methanogenesis was approximately improved by 70% with ZVI addition (Zhao et al., 2018). In this work, the inoculums collected from a waste sludge treatment plant was used, and also it highlighted the importance of hydrogenotrophic methanogenesis and syntrophic methane production between syntrophic bacteria and methanogens enhanced by ZVI addition. Moreover, conducting fluorescence in situ hybridization (FISH) analysis indicated that adding waste iron scraps enhanced the abundance of acidifying bacteria, acetoclastic and hydrogenotrophic methanogens of the pre-cultured inoculum (Hao et al., 2017). The main reason might be that the activity of hydrogenotrophic methanogens in the seed sludge (from full-scale anaerobic digester treating industrial wastewater) was low, and also the inoculum was not acclimatized to the tested conditions (such as ZVI addition). Therefore, more efforts are suggested to verify this effect by using well-adapted inoculum in long-term operated anaerobic digesters. In addition, it will be of great interest to apply advanced analytical methods, such as high throughput sequencing and FISH, to provide more evidence for illuminating the exact effects of ZVI on microbial community properties.

3.5. Implications for practical application

Wastewater is increasingly recognized as a valuable resource rather than a waste, and this conceptual change has driven the shift of wastewater treatment paradigm from pollutant removal to maximum resource recovery. In this case, many exploration and practice have been made from varied aspects, such as proposing hypothetical treatment systems with future perspective (Verstraete et al., 2009; McCarty et al., 2011), evolving wastewater infrastructure paradigm based on

sustainability principles (Wang et al., 2018), wastewater pretreatment for carbon capture or redirection (Sancho et al., 2019; Guven et al., 2019) as well as nutrients recovery (Yan et al., 2018). However, most of the current efforts are paid to system development and modelling, the actual conditions of resource recovery and potential technical limitations are insufficiently understood and to be well investigated.

This study primarily verified the promising strategy (membrane filtration for organics capture plus anaerobic digestion for bioenergy generation) for useful resource recovery. However, due to high enrichment of particulate and colloidal organic substances from domestic wastewater, the efficiency of AD process seemed to be easily limited, due to low reaction rate of the hydrolysis-acidification process (Zhen et al., 2017). The proposed measure of ZVI addition to enhance AD performance was investigated, showing substantial enhancement on the hydrolysis-acidification of pre-concentrated domestic wastewater and overall methane production. A rough comparative estimation on economic benefit between conventional AD and ZVI-assisted AD for PDWW treatment is carried out with some important default values based on one previous study (Wei et al., 2018a,b). The initial conditions are set as follows: treatment capacity of anaerobic digester of 5000 m³/d, methane yield of 0.25 and 0.29 LCH₄/gCOD, and the produced biogas is considered to be combusted in a combined heat and power (CHP) system with the benefit from power generation firstly considered rather than heat production. The calculation results indicate that ZVI-based AD system can be more beneficial than its counterpart (AD alone) when taking power production benefit, ZVI cost and other expenditures (ZVI transportation and dosing) into comprehensive consideration. Furthermore, using low-cost ZVI (such as scrap iron) or optimizing ZVI usage, and considering phosphorous recovery and sulfur control during AD process will further increase the benefits by the enhanced methane production and resource recovery.

In all, the results obtained highlighted that further optimization of the AD process of PDWW is needed with the focuses on integrated low-cost pre-treatment method and the enhancement of ZVI utilization efficiency to realize the efficient recovery of renewable resource.

4. Conclusions

The viability of ZVI addition to improve PDWW treatment by AD process was investigated. The PDWW containing high content of particulate organics could be effectively digested at optimal F/M of 0.5 gCOD/gMLVSS while adding 6 g/L ZVI further enhanced the BMP by 15.2%. The major contribution of ZVI was attributed to substantially enhancing the hydrolysis process and regulating the acidification process rather than affecting methanogenesis process. The results indicate that integrating ZVI with pre-treatment measures to further enhance PDWW hydrolysis-acidification will be appreciated to realize the application of the proposed strategy (wastewater pre-concentration plus anaerobic digestion) towards holistic resource recovery.

CRediT authorship contribution statement

Ying Zang: Data curation, Investigation, Writing - original draft.
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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This study was supported by the Natural Science Foundation of Shaanxi Province (grant no. 2018JQ5054), the Japan Society for the Promotion of Science (JSPS) support for JSPS Fellows (no. P19745), the Science Foundation for Fostering Talents of Xi'an University of Architecture and Technology (grant no. RC1710), and the Shaanxi Provincial Program for Innovative Research Team (grant no. 2019TD-025 and IRT2013KCT- 13).

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.biortech.2020.123441>.

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