RESEARCH ARTICLE

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The impact of hematite on the anaerobic digestion of cattle manure

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Abstract

A metal-based conductive material, hematite (Fe_2O_3), was used as an amendment in the anaerobic digestion process to determine the effects on the performance of anaerobic digestion of cattle manure (CM) at mesophilic temperature (35 °C). The first set of experiments (Set 1) was designed to assess whether there is a need to supplement nutrients for the effective digestion of CM. To this purpose, basal medium (BM) composed of macro nutrients, micro nutrients, reducing agent, and buffer was added to the reactors and a biochemical methane production assay was conducted. The presence of BM showed negative impacts on the anaerobic digestion of CM and its absence caused up to 40% higher methane production yield. In Set 2 experiments, the impact of hematite addition on methane production performance was determined. Two different dosages as 20 mM Fe (Fe20) and 50 mM Fe (Fe50) were applied to the batch reactors. Hematite amendments increased methane yield; at Fe20 (131 ± 2.6 mL CH_4/g VS_{added}) the increase was around 8% and at Fe50 (135 ± mL 0.2 CH_4/g VS_{added}) the increase was around 12% as compared to the control. Further, up to 36% increase in the methane production rate was calculated via Modified Gompertz fitting.

Keywords: Anaerobic digestion, Cattle manure, Methane production, Conductive materials, Hematite

INTRODUCTION

Conventional anaerobic digestion is an effective technology for the treatment of organic wastes and bioenergy production in the form of biogas (Anukam et al., 2019). During anaerobic digestion, biogas containing 60-70% methane (CH_4) and 30-40% carbon dioxide (CO_2) can be produced (Speece, 1983). Different feedstocks such as animal manure and wastewater treatment plant sludge may be used in anaerobic digestion for methane production. Especially, cattle manure due to its high organic content, and high level of microbial activity has been commonly preferred as a feed in anaerobic digestion (Zheng et al., 2015). Handling of cattle manure via digestion can also decrease its adverse environmental impacts. Although anaerobic digestion is a well-known and effective technology for organic waste disposal and simultaneous renewable energy production, it has some limitations. These limitations can be counted as low methane production rate due to slow reaction kinetics, high sensitivity to inhibitory compounds such as ammonia, volatile fatty acids (VFAs), and unstable operations with changing conditions due to accumulation of VFAs (Park et al., 2018; Yin et al., 2020). The slow processing of wastes is a result of little energy gained by anaerobic microbes during process, and the slow growth rate of the microorganisms involved in the process (Yin et al., 2020). These drawbacks are important for effective process operation, and they should be properly managed.

During anaerobic digestion, production of methane occurs via a series of reactions; hydrolysis, acidogenesis, acetogenesis, and methanogenesis (Park et al., 2018). Hydrolytic bacteria, acidogenic bacteria, and acetogenic bacteria are responsible for hydrolysis, acidogenesis and acetogenesis. Methanogenic archaea, on the other hand, are responsible for methanogenesis step (Kumar et al., 2021). The syntrophic interactions between bacteria and methanogens are the key to effective process performance and this interaction is based on electron transfer between different microbial communities (Park et al., 2018). Recent studies suggest that bacteria and methanogenic archaea perform direct interspecies electron transfer (DIET) via the use of conductive materials, leading to higher efficiency digestion process (Kutlar et al., 2022; Liu et al., 2012). Studies showed that DIET via conductive materials enables faster electron transfer than electron transfer via intermediates such as acetate and hydrogen as in conventional systems (He et al., 2021). This in turn is related to the faster utilization of feed and enhanced process performance. Further, amendment of conductive materials may lower the impact of inhibitory compounds, and decrease the oxidation-reduction potential of the medium, hence offering a more suitable environment for methanogenic activity (Kutlar et al., 2022). Therefore, this study aims to investigate the effect of conductive material amendment on biomethane production from cattle manure. To this purpose, a metal-based conductive material, hematite (Fe_2O_3) was used in the experiments. The hematite-amended reactor performances were evaluated based on the comparison of lag time, biomethane production rate, and biomethane production yield with a control reactor.

MATERIALS AND METHODS

Waste and Inoculum Characteristics

Cattle manure was taken from the inlet of a full-scale biogas plant located in Ankara, Turkey. The sample was blended for 1 hour for homogenization and then characterization analysis was conducted (Table 1). The inoculum used in this study has been taken from a municipal wastewater treatment plant located in Eskisehir, Turkey. Due to the waiting period, for two different sets of experiments two different samples of inoculum and manure were collected and used in the experiments.

Basal Medium and Conductive Material

To provide nutrients and other necessary compounds such as reducing agents a cocktail named basal medium (BM) was prepared. The composition of BM used in the experiments is as follows (concentrations are given in parenthesis as mg/L): NH_4Cl (1200), $MgSO_4.7H_2O$ (400), KCl (400), $Na_2S.9H_2O$ (300), $CaCl_2.2H_2O$ (50), $(NH_4)_2HPO_4$ (80), $FeCl_2.4H_2O$ (40), $CoCl_2.6H_2O$ (10), KI (10), $MnCl_2.4H_2O$ (0.5), $CuCl_2.2H_2O$ (0.5), $ZnCl_2$ (0.5), $AlCl_3.6H_2O$ (0.5), $NaMoO_4.2H_2O$ (0.5), H_3BO_3 (0.5), $NiCl_2.6H_2O$ (0.5), $Na_2WO_4.2H_2O$ (0.54), Na_2SeO_3 (0.5), cysteine (10), $NaHCO_3$ (6000) (Demirer et al., 2000). In this cocktail, cysteine acts as a reducing agent, and bicarbonate solution works as a buffer, while the others provide the macronutrients and micronutrients.

In the experiments, hematite (Fe_2O_3) was used as a metal-based conductive material and was used at two different concentrations as 20 mM Fe and 50 mM Fe. The particle size distribution of the hematite sample was determined through sieving and given in Figure 1. The particle size was mostly around 2.5 mm. Before use, the solids content of the hematite sample was measured and the results are as follows: 99.2 ± 0.0% of TS, 0.4 ± 0.0 VS corresponding to 0.4% of VS/TS.

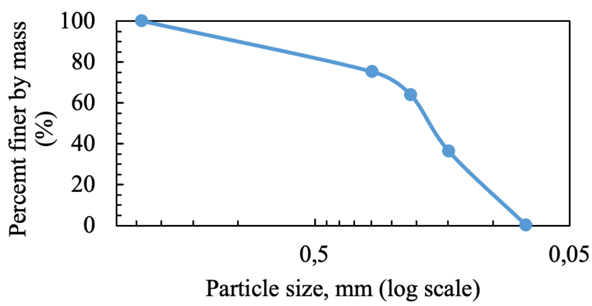

Figure 1. Particle Size Distribution of Hematite used in the Experiments.

Analytical Methods

For TS (Method 2540B), VS (Method 2540E), and chemical oxygen demand (COD) standard methods were used (Standard Methods for the Examination of Water and Wastewater, 1999). For COD measurements of manure sample open reflux method (Method 5220 B) was used and for inoculum closed reflux method (Method 5220C) was used (Standard Methods for the Examination of Water and Wastewater, 1999). For nitrogen (Hach Method 8038) and phosphorus (Hach Method 8178) measurements, colorimetric methods were used via a spectrophotometer (Hach DR9200, USA). pH measurements were conducted with a pH meter (St300, OHAUS, USA). Biogas production in the reactors was measured periodically during the operation via a liquid displacement device. The composition of biogas produced in the reactors was determined by a gas chromatography device (Trace GC Ultra, Thermo Scientific) equipped with a thermal conductivity detector (TCD) and columns connected series in (CP-Moliseve 5A and CP-Porabond Q). The temperature of the oven, injector and detector were

Table 1. Waste and inoculum characteristics.

Parameter	Set 1		Set 2	
	Cattle manure	**Inoculum**	**Cattle manure**	**Inoculum**
Density, (g/mL)	0.997	0.997	0.881	0.974
pH	7.8	7.6	7.8	7.5
Total solids (TS), (%)	12.2± 0.1	4.7 ± 0.0	12.2 ± 0.1	3.3 ± 0.0
Volatile Solids (VS), (%)	9.2 ± 0.1	1.8 ± 0.0	9.5 ± 0.1	1.8 ± 0.0
VS/TS (%)	76.0 ± 0.7	39 ± 0.5	77.6 ± 0.0	52.7 ± 0.4
Chemical oxygen demand (COD), (mg/L)	88,000± 11,000	30,500 ± 1,050	151,743 ± 6,446	30,027 ± 610
Nitrogen, (mg NH_4-N/L)	4,380 ± 390	nd	1,897 ± 116.8	nd
Phosphorus, (mg PO_4-P/L)	nd	nd	35.3 ± 1.6	nd

nd: not determined

35 °C, 50 °C, and 80 °C, respectively. The carrier gas was helium at a constant pressure of 75 kPa.

Daily produced methane was calculated from methane content and total produced biogas (Filer et al., 2019) using the following Equation (1):

$$V_{(CH4)} = \left(\frac{\%\,CH4,t}{100} * Vbiogas + \frac{\%CH4,t - \%CH4,t-1}{100}\right) * Vheadspace \quad (1)$$

where V_{CH4}, $V_{biogas,}$ and $V_{headspace}$ represent daily produced methane volume (mL), daily produced total gas (mL), and volume of reactor headspace (mL), respectively. $\%CH_{4,t}$ and $\%CH_{4,t-1}$ are the methane percentages of total biogas production on the corresponding day and the previous day, respectively.

Experimental Design

There were two sets in this study. In the first set, our objectives were two-fold: to determine (i) the impact of BM on the anaerobic digestibility of cattle manure, and (ii) the effect of initial COD on digestion performance. Two different initial COD concentrations representing higher (~ 30,000 mg/L) and lower (~20,000 mg/L) COD concentrations were adjusted. After setting up the reactors initial CODs were measured and in the reactors with higher COD, the COD concentrations ranged between 30,240 mg COD/L and 31,240 mg COD/L, and in the reactors with lower COD, the initial COD concentrations ranged between 21,760 mg COD/L and 22,240 mg COD/L. These additions established different food-to-microorganisms (F/M) ratios (in mg/L VS basis) in the reactors. F/M ratios of HCOD reactors were around 1.9 and LCOD reactors were around 1. The experimental design for Set 1 is shown in Table 2.

When needed, 10 mL of BM was added into the reactors according to the experimental design to determine the effect of BM on AD performance. Blank reactors having only inoculum in the absence of BM (B1) and the presence of BM (B2) were also operated to find out the background methane production from the inoculum.

In the second set, we run experiments to investigate the effect of hematite amendment on the performance of anaerobic digestion without any addition of BM. Two different dosages of hematite; 20 mM Fe containing hematite (Fe20) and 50 mM Fe containing hematite (Fe50) were used in the experiments. The experimental design of Set 2 is given in Table 3. For the determination of the performance of anaerobic digestion of cattle manure and comparing it with the hematite-amended reactors, we set the control without any hematite (AD). Also, we set a blank reactor (B) having only inoculum as another control to determine the methane production coming only from the inoculum.

Table 2. Experimental Design of Set 1.

Reactor	Substrate	BM	Initial COD (mg/L)	Initial VS (mg/L)	F/M ratio
B1 w/o BM	-	-	-	-	-
B2 w/ BM	-	+	-	-	-
AD1, HCOD w/o BM	+	-	30,240 ± 1,020	21,950 ± 317	1.9
AD2, HCOD w/ BM	+	+	31,200 ± 880	23,650 ± 450	1.9
AD3, LCOD w/o BM	+	-	21,760 ± 640	16,500 ± 167	1.0
AD4, LCOD w/ BM	+	+	22,240 ± 720	17,067 ± 567	1.0

HCOD: higher COD; LCOD: lower COD; BM: basal medium; F/M: food to microorganisms ratio

Table 3. Experimental Design of Set 2.

Reactor	Substrate	Hematite	Initial COD (mg/L)	Initial VS (mg/L)	F/M ratio
Blank	-	-	-	-	
Control	+	-	52,400 ± 640	32,066 ± 505	1.2
Fe20	+	+ (20 mM Fe)	49,200 ± 940	31,964 ± 209	1.2
Fe50	+	+ (50 mM Fe)	48,400 ± 820	31,580 ± 104	1.1

Reactor Operation

Set 1 experiments were conducted in 110 mL serum bottles with an active volume of 60 mL. All reactors were inoculated with 30 mL of AD seed. Then, 20 mL of cattle manure was added to the reactors. All reactors were covered with aluminum foil and incubated at 35±1 °C in the temperature-controlled room. In Set 1, all reactors were operated in triplicate without mixing.

In Set 2, the reactor volumes were increased slightly in comparison to Set 1. In Set 2, 300 mL serum borosilicate bottles were used with a working volume of 150 mL. The initial COD of the reactors was aimed to be around 50 g/L and initial COD measurements ranged between 48,400-52,400 mg COD/L with an F/M ratio of approximately 1. Similar to Set 1, all reactors were operated in triplicate. In Set 2, the reactors were mixed with a shaker at 150 rpm to prevent the settlement of hematite particles.

When reactors were filled according to the experimental design before the incubation, all reactors were sparged with 70% nitrogen (N_2) and 30% carbon dioxide (CO_2) for 3 mins to maintain anaerobic conditions. After sparging, the reactors were immediately sealed with rubber stoppers that are tied with plastic cable. For the removal of oxygen in the headspace and providing an anaerobic environment, the headspaces of the reactors were purged with the same gas for 2 mins.

During the incubation period, produced biogas amount and its composition were monitored periodically. When cumulative methane production as compared to previous measurement was less than 10% for two times in a row, the operation of Set 1 reactors was stopped. After the completion of the batch test, all reactors were stored at 4 °C until the final analysis of composition was complete. TS, VS, and COD analysis for the reactor effluents were conducted. For the comparison of the reactor performances, cumulative methane productions, methane yields (based on the amount of added VS), and organic removals were calculated. In Set 2, after the reactor operation, pH, conductivity, and ORP measurements of the effluents were conducted. Also, the final phosphorus and ammonium concentrations of the effluents were analyzed.

Modeling of Biomethane Production

Modified Gompertz fitting to cumulative methane production data was conducted for comparing the performances of hematite-added reactors and conventional AD in Set 2 (Zwietering et al., 1990). The kinetic parameters such as specific methane production potential (mL, B_t), maximum methane production potential (mL, B_0), methane production rate (mL CH_4/day, R_m), and lag time for the reactor (day, λ) were determined using the modified Gompertz model provided in Equation (2) (Zwietering et al., 1990):

$$B_{(t)} = B_0 * \exp\{[\frac{Rm*e}{B0} * (\lambda - t) + 1]\}, t \geq 0 \qquad (2)$$

In Equation (2), t is incubation time (day) and e is 2.718.

RESULTS AND DISCUSSION

Impact of Basal Medium

In terms of cumulative methane production, the highest production was observed in AD1 w/o BM (141 ± 5 mL CH_4) which is 25% higher than the production in AD2 w/ BM (113 ± 6 mL CH_4) among HCOD reactors (Figure 2). Similarly, for LCOD reactors, AD3 w/o BM (79 ± 8 mL CH_4) produced 39% higher cumulative methane than AD4 w/ BM (57 ± 4 mL CH_4). AD1 and AD2 reactors produced 78% and 98% higher methane as compared to AD3 and AD4, respectively. This is due to higher COD levels in AD1 and AD2 in comparison to AD3 and AD4.

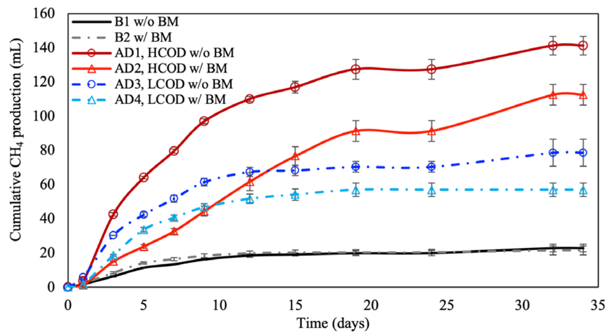

Figure 2. Cumulative Methane Production in Set 1 Reactors. (Error bars may be smaller than the symbols).

VS removals in the reactors were similar. VS removals were 34% and 37% for HCOD and LCOD reactors, respectively, without showing any significant change due to BM addition (Figure 3). Among HCOD reactors, COD removals were 44% and 40% in AD1 and AD2, respectively. For the reactors with LCOD, the removal of COD did not change with the addition of BM, and it was 36% for AD3 and AD4.

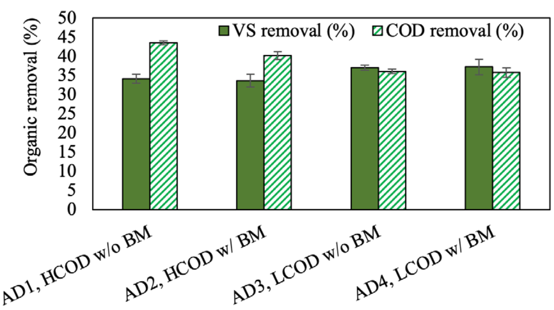

Figure 3. Organic Removal in Set 1 Reactors. (Error bars may be smaller than the lines).

Methane yields were also evaluated based on the added amount of VS and COD (Figure 4). HCOD reactors, AD1 (107 ± 4 mL CH_4/g VS_{added}) showed 35% higher yield as compared to AD2 (79 ± 4 mL CH_4/g VS_{added}). Similarly, for LCOD, the absence of BM in AD3 (79 mL ± 8 CH_4/g VS_{added}) resulted in 41% increase in methane yield over AD4 (56 ± 4 mL CH_4/g VS_{added}) with BM addition. In addition to this, the application of HCOD resulted in higher methane yield in AD1 and AD2 as compared to AD3 and AD4. 35% higher methane yield was observed in AD1 in comparison to AD3 which is due to higher organic content. When BM present reactors were compared a similar result was attained; 43% higher methane yield was obtained in AD2 as compared to AD4. Methane yields based on added COD had also similar trends. The absence of BM increased the yield 30% in AD1 (78 mL ± 3 CH_4/g COD_{added}) over AD2 (60 ± 3 mL CH_4/g COD_{added}) for HCOD reactors and 40% enhancement was observed in AD3 (60 ± 6 mL CH_4/g COD_{added}) without BM addition as compared to AD4 (43 ± 3 mL CH_4/g COD_{added}) with BM among LCOD reactors.

Methane yields suggested that the presence of BM on anaerobic digestion of cattle manure causes inhibition in both LCOD and HCOD reactors. This can be attributed to the nutrient content of cattle manure. Similarly, it was reported that the nutrient in cattle manure was already sufficient for anaerobic microbial growth without the need for an extra addition and BM addition resulted in lower performance (Güngör-Demirci & Demirer, 2004). In other words, since available nutrients in cattle manure is sufficient for anaerobic digestion, the digestion is inhibited with the extra nutrient addition via BM. In terms of the amount of organic loading, the HCOD reactors showed higher methane productions and methane yields than the LCOD reactors, and this may result from an amount of higher available carbon source for microbial growth and methane production.

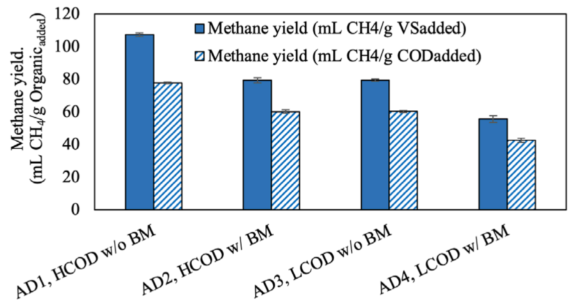

Figure 4. Methane Yield in Set 1 Reactors. (Error bars may be smaller than the lines).

As given in Table 4, a wide range (89-267 mL CH_4/g VS_{added}) in terms of methane yield of cattle manure digestion was reported in the literature. The methane yields attained in HCOD reactors of this study are within this range. There

Table 4. Comparison of Methane Yields with Other Studies where Cattle Manure was used as Feed.

Inoculum	T (°C)	Operation	Organic Load	Methane Yield* (mL/g VS_{added})	Reference
AD seed from WWTP	36	Batch	32,904 mg/L initial COD	266	(Huang et al., 2016)
Laboratory-scale anaerobic digester	37	Batch	6% initial TS	89	(Zheng et al., 2015)
Cattle manure anaerobic digester	37	Batch		150	(Song & Zhang, 2015)
Mixed anaerobic culture	35	Batch	a) 12,000 mg/L initial COD b) 53,500 mg/L initial COD	a) (155) b) (195)	(Güngör-Demirci & Demirer, 2004)
Dairy farm			7% initial TS	124	(Rosenberg & Kornelius, 2017)
Laboratory-scale anaerobic digester	35	Batch	15% initial VS	231	(Wei et al., 2019)
Digested slurry	35	Batch	15% initial TS	251 (210)	(Li et al., 2011)
AD seed from WWTP	35	Batch	a) 22,000 mg/L initial COD b) 30,720 mg/L initial COD	a) 79 (60) b) 107 (78)	This study

AD: anaerobic digester; WWTP: wastewater treatment plant
*Number in parenthesis show methane yield based on added COD: mL/g COD_{added}

can be several reasons for obtaining different methane yield in different studies, such as the lignin content of manure used, the initial organic loading, F/M ratio, and the use of different inoculums.

Impact of Hematite

The amendment of hematite slightly increased cumulative methane production in comparison to control reactor (Figure 5). There was a slight difference in the cumulative methane production of different dosage reactors (Fe20 and Fe50). Fe20 reactors produced a total of 626 ± 13 mL of CH_4 and Fe50 reactors produced 641 ± 1 mL of CH_4 corresponding to 7% and 10% increase over control reactor, respectively (Figure 5).

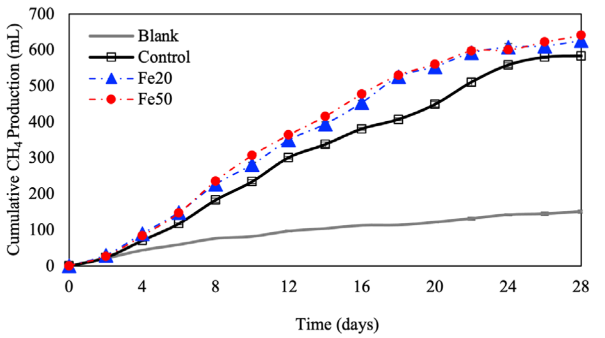

Figure 5. Cumulative Methane Production in Set 2 Reactors. (Error bars may be smaller than the symbols).

Through the addition of hematite, there was no significant change in VS or COD removal as compared to the control was not observed (Figure 6). This is similar to other studies in the literature. As reported in the literature, the amendment of hematite did not enhance the organic removal in anaerobic digestion of swine manure (Lu et al., 2019). A similar trend was observed with the application of another iron-based conductive material, magnetite (Fe_3O_4) (Yin et al., 2017). There was no significant change in organic removal via magnetite application over the control (Yin et al., 2017).

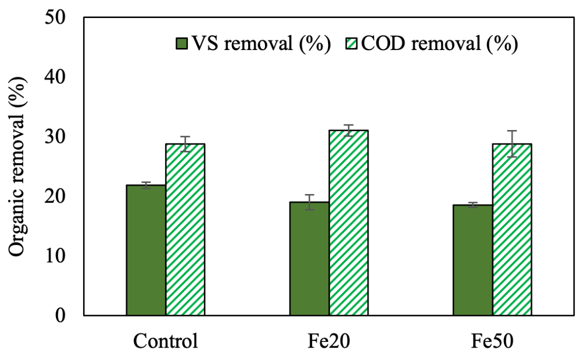

Figure 6. Organic Removal in Set 2 Reactors. (Error bars may be smaller than the lines).

Methane production yield based on added VS and added COD were also calculated (Figure 7). Fe20 (131 ± 2.6 mL CH_4/g VS_{added}) and Fe50 (135 ± mL 0.2 CH_4/g VS_{added}) enhanced methane yield by 8% and 12% as compared to the control, respectively (Figure 7). In another recent work, authors reported that the addition of hematite improved the methane production yield by 7% during anaerobic digestion of swine manure (Lu et al., 2019). Similarly, Ye et al., (2018) observed 36% increase in methane yield via hematite application on anaerobic digestion of activated sludge. In our work, although there is a 2.5 times difference in dosage values of Fe20 and Fe50, the enhancement in methane yield in Fe50 as compared to Fe20 was not as significant.

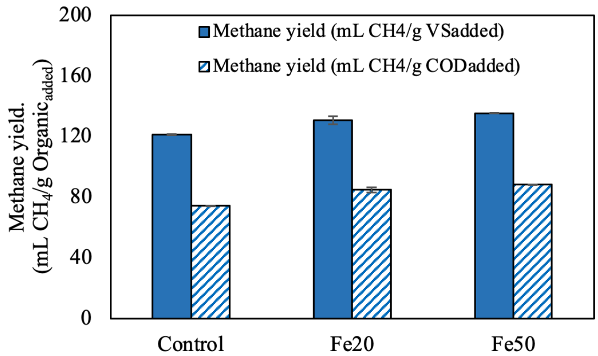

Figure 7. Methane Yields in Set 2 Reactors. (Error bars may be smaller than the lines).

For quantification of the change in the methane production rate and the lag time, modified Gompertz model was fitted to the cumulative methane production graphs of Set 2 reactors (Figure 8). Based on this fitting the increase in methane production rate (R_m) and the decrease in the lag time for the production of methane were calculated (Table 5). The application of hematite enhanced methane production rate as given in Table 5. Fe20 (35.9 ± 0.7 mL CH_4/day) and Fe50 (38.2 ± 0.6 mL CH_4/day) improved the rate 26% and 34% over the control, respectively (Table 5). Similarly, it was reported that the application of hematite enhanced methane production rate by 34% on AD of swine manure (Lu et al., 2019). On the other hand, there was no improvement in lag time with the application of hematite over the control.

Other studies using hematite for the enhancement of conventional anaerobic digestion are summarized in Table 6. There was only one study conducted with animal manure and the authors reported 11% increase in methane yield via hematite addition to AD reactors fed with swine manure (Lu et al.,2019). Comparison of enhancements in this study with others in the literature show that our results are consistent with the literature. Yet, it should be highlighted that in none of the studies cattle manure was used as a feed during hematite amendment. However, because of the presence of different microbial communities in different complex wastes an experimental study is required to investigate the impact of conductive material amendment.

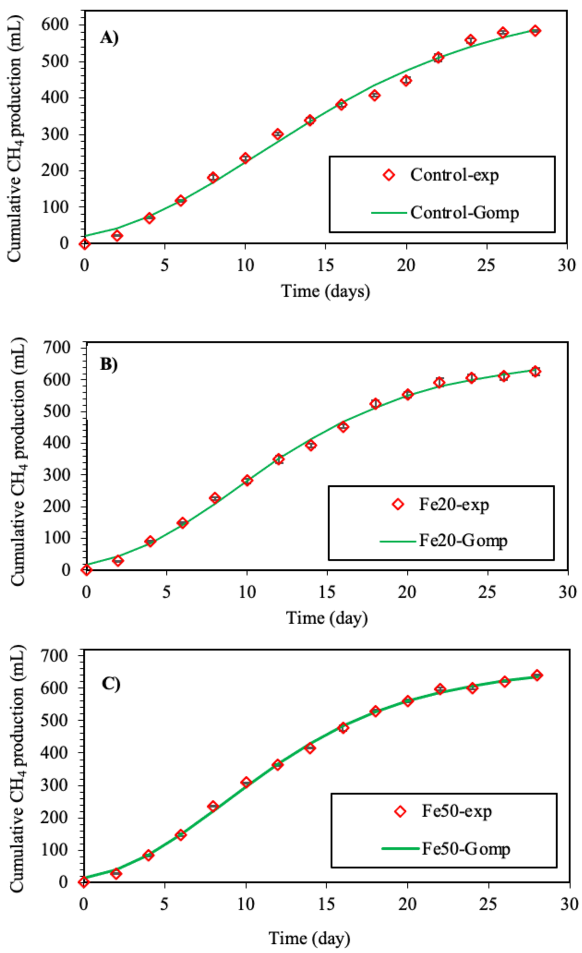

Figure 8. Cumulative Methane Production Curves of Set 2 Reactors fitted with Modified Gompertz.

Ammonia concentration is an important parameter for anaerobic digestion since the release of ammonia during protein degradation can cause inhibition of methanogenic activity (Rasapoor et al., 2020). In the literature, various ammonia concentrations were reported to cause inhibition in anaerobic digesters and this inhibitory level significantly depends on the feed and reactor conditions (Yenigün & Demirel, 2013). For example, it was reported that with unacclimated inoculum total ammonia concentrations of 1700 – 1800 mg/L were inhibitory, yet with acclimated inoculum when cattle manure was used as feed the inhibitory total ammonia concentration was raised to 6000 mg/L (Yenigün & Demirel, 2013). Nutrient (N and P) concentrations for each reactor of Set 2 were measured at the end of the operation (Figure 9). In Set 2 reactors ammonium nitrogen (NH_4-N) concentrations were around 1500 mg/L and pH in Set 2 reactors was measured as 7.8 in each reactor (Figure 10). pH level being below 8 when considered along with moderate NH_4-N concentrations it is concluded that there may only be a slight inhibition, if any.

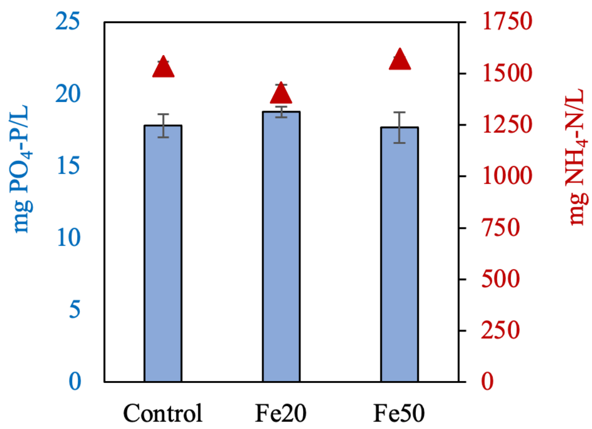

Figure 9. Nitrogen and Phosphorus Concentrations in Set 2 reactors. (Triangles show NH_4-N concentrations. Error bars may be smaller than the symbols).

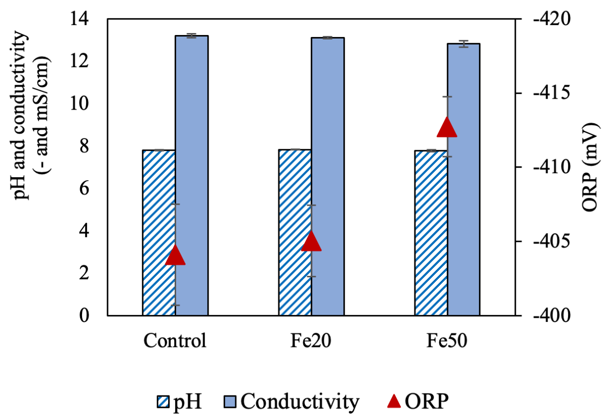

Figure 10. pH, conductivity and ORP of Set 2 reactors. (Error bars may be smaller than the lines).

The final pH, ORP, and conductivity of the reactors are shown in Figure 10. As reported in the literature, these

Table 5. Kinetic parameters calculated from the fitting with the modified Gompertz model in Set 2

Reactor	P (mL)	R_m (mL CH_4/day)*	λ (day)	R^2
Control	674.9 ± 6.8	28.5 ± 0.5	2.2 ± 0.1	0.9939
Fe20	674.2 ± 14.1	35.9 ± 0.7 (26%)	2.1 ± 0.1	0.9973
Fe50	666.3 ± 4.3	38.2 ± 0.6 (34%)	2.2 ± 0.1	0.9982

*The number in the parenthesis indicates the enhancement in methane production rate as compared to the control.

Table 6. Comparison of Enhancements in Different Studies where Hematite was applied during Anaerobic Digestion.

Dosage (mM Fe)	Reactor	Inoculum	Substrate	Impovements as compared to control	Reference
20	Batch	Paddy soil	Acetate	Acceleration of methanogenesis and decrease in lag time	Kato et al., 2011
25	Batch	Paddy soil	Acetate	110% increase in cumulative methane production	Zhou et al., 2013
25	Batch	Paddy soil	Benzoate	25% increase in the methane production rate	Zhuang et al., 2015
187.5	Batch	Inoculum from swine farm	Swine Manure	11% increase in methane yield	Lu et al., 2019
250	Batch	Laboratory-scale UASB reactor	Activated Sludge	36% increase in methane yield	Ye et al., 2018
a) 20 b) 50	Batch	AD seed from WWTP	Cattle Manure	a) 10% increase in methane yield 26% increase in methane production rate b) 12% increase in methane yield 34% increase in methane production rate	This study

UASB: upflow anaerobic sludge blanket; AD: anaerobic digester; WWTP: wastewater treatment plant

pH values are between the optimum pH range which is 7.0-8.0 (Uçkun Kiran et al., 2016). Final pH values were around 7.8, which is approximately the neutral pH range indicating no significant acid accumulation.

Conductivity has also been measured in the reactor effluents (solid bars, Figure 10). The addition of hematite did not significantly change the conductivity levels as compared to the control. We also conducted the final ORP measurement, which is an important parameter for the microbial activity of methanogens. Methanogenesis ideally occurs at ORP range of -200 mV to -400 mV (Martins et al., 2018). ORP values in the reactors having hematite were -405 ± 2 mV and -413± 2 mV in Fe20 and Fe50, respectively, which are very close to ORP value in the control (-404 ± 3 mV).

CONCLUSION

In this study, the impacts of supplementation of an all-inclusive nutrient cocktail, BM, and a metal-based conductive material, hematite, on methane production from cattle manure were investigated

- At VS concentrations around 20 g/L the supplementation of BM has adverse effects on anaerobic digestion of cattle manure. Methane production yield was 35 – 41 % higher in the absence of BM.
- Addition of hematite is beneficial during anaerobic digestion of cattle manure, and up to 12% increase in the methane production yield and around 34% increase in the methane production rate were attained via supplementation of hematite at a dosage of 50 mM Fe.

These results are promising for effective anaerobic digestion of cattle manure and may be used for increasing the performance of a full-scale biogas plants.

COMPLIANCE WITH ETHICAL STANDARDS

Conflict of interest

The authors declared that for this research article, they have no actual, potential or perceived conflict of interest.

Author contribution

Yasin Odabas carried out experiments, data analysis and writing the original draft. Yasemin Dilsad Yilmazel designed the study, supervised, wrote, reviewed and edited the original draft. All authors read and approved the final manuscript. All authors verifies that the Text, Figures, and Tables are original and that they have not been published before.

Ethical approval

Ethics committee approval is not required.

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Data availability

Not applicable.

Consent for publication

Not applicable.

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REFERENCES

Anukam, A., Mohammadi, A., Naqvi, M., & Granström, K. (2019). A review of the chemistry of anaerobic digestion: Methods of accelerating and optimizing process efficiency. Processes, 7 (8), 1–19. https://doi.org/10.3390/PR7080504

Demirer, G. N., Duran, M., Ergüder, T. H., Güven, E., Ugurlu, Ö., & Tezel, U. (2000). Anaerobic treatability and biogas production potential studies of different agro-industrial wastewaters in Turkey. Biodegradation, 11, 401–405. https://doi.org/10.1023/A:1011659705369

Filer, J., Ding, H. H., & Chang, S. (2019). Biochemical methane potential (BMP) assay method for anaerobic digestion research. Water, 11(5), 921, 1-29. https://doi.org/10.3390/w11050921

Güngör-Demirci, G., & Demirer, G. N. (2004). Effect of initial COD concentration, nutrient addition, temperature and microbial acclimation on anaerobic treatability of broiler and cattle manure. Bioresource Technology, 93(2), 109–117. https://doi.org/10.1016/j.biortech.2003.10.019

He, X., Guo, Z., Lu, J., & Zhang, P. (2021). Carbon-based conductive materials accelerated methane production in anaerobic digestion of waste fat, oil and grease. Bioresource Technology, 329. https://doi.org/10.1016/j.biortech.2021.124871

Huang, X., Yun, S., Zhu, J., Du, T., Zhang, C., & Li, X. (2016). Mesophilic anaerobic co-digestion of aloe peel waste with dairy manure in the batch digester: Focusing on mixing ratios and digestate stability. Bioresource Technology, 218, 62–68. https://doi.org/10.1016/j.biortech.2016.06.070

Kato, S., Hashimoto, K., & Watanabe, K. (2012). Methanogenesis facilitated by electric syntrophy via (semi)conductive iron-oxide minerals. Environmental Microbiology, 14(7), 1646–1654. https://doi.org/10.1111/j.1462-2920.2011.02611.x

Kumar, V., Nabaterega, R., Khoei, S., & Eskicioglu, C. (2021). Insight into interactions between syntrophic bacteria and archaea in anaerobic digestion amended with conductive materials. Renewable and Sustainable Energy Reviews, 144, 110965. https://doi.org/10.1016/j.rser.2021.110965

Kutlar, F. E., Tunca, B., & Yilmazel, Y. D. (2022). Carbon-based conductive materials enhance biomethane recovery from organic wastes: A review of the impacts on anaerobic treatment. Chemosphere, 290. https://doi.org/10.1016/j.chemosphere.2021.133247

Li, J., Kumar Jha, A., He, J., Ban, Q., Chang, S., & Wang, P. (2011). Assessment of the effects of dry anaerobic co-digestion of cow dung with waste water sludge on biogas yield and biodegradability. International Journal of the Physical Sciences, 6(15), 3723–3732. https://doi.org/10.5897/IJPS11.753

Liu, F., Rotaru, A. E., Shrestha, P. M., Malvankar, N. S., Nevin, K. P., & Lovley, D. R. (2012). Promoting direct interspecies electron transfer with activated carbon. Energy and Environmental Science, 5(10), 8982–8989. https://doi.org/10.1039/c2ee22459c

Lu, T., Zhang, J., Wei, Y., & Shen, P. (2019). Effects of ferric oxide on the microbial community and functioning during anaerobic digestion of swine manure. Bioresource Technology, 287, 121393 https://doi.org/10.1016/j.biortech.2019.121393

Martins, G., Salvador, A. F., Pereira, L., & Alves, M. M. (2018). Methane Production and Conductive Materials: A Critical Review. Environmental Science and Technology, 52 (18), 10241–10253. https://doi.org/10.1021/acs.est.8b01913

Park, J. H., Kang, H. J., Park, K. H., & Park, H. D. (2018). Direct interspecies electron transfer via conductive materials: A perspective for anaerobic digestion applications. Bioresource Technology, 254,300–311. https://doi.org/10.1016/j.biortech.2018.01.095

Rasapoor, M., Young, B., Asadov, A., Brar, R., Sarmah, A. K., Zhuang, W. Q., & Baroutian, S. (2020). Effects of biochar and activated carbon on biogas generation: A thermogravimetric and chemical analysis approach. Energy Conversion and Management, 203, 112221. https://doi.org/10.1016/j.enconman.2019.112221

Rosenberg, L., & Kornelius, G. (2017). Experimental investigation of biogas production from feedlot cattle manure. Journal of Energy in Southern Africa, 28(4), 1–8. https://doi.org/10.17159/2413-3051/2017/v28i4a1753

Song, Z., & Zhang, C. (2015). Anaerobic codigestion of pretreated wheat straw with cattle manure and analysis of the microbial community. Bioresource Technology, 186, 128–135. https://doi.org/10.1016/j.biortech.2015.03.028

Speece, R. E. (1983). Anaerobic biotechnology for industrial wastewater treatment. Environmental Science and Technology, 17(9), 416A-427A. https://doi.org/10.1021/es00115a001

Standard Methods for the Examination of Water and Wastewater. (1999).

Uçkun Kiran, E., Stamatelatou, K., Antonopoulou, G., & Lyberatos, G. (2016). Production of biogas via anaerobic digestion. In Handbook of Biofuels Production: Processes and Technologies: Second Edition (pp. 259–301). Elsevier Inc. https://doi.org/10.1016/B978-0-08-100455-5.00010-2

Wei, L., Qin, K., Ding, J., Xue, M., Yang, C., Jiang, J., & Zhao, Q. (2019). Optimization of the co-digestion of sewage sludge, maize straw and cow manure: microbial responses and effect of fractional organic characteristics. Scientific Reports, 9(1). https://doi.org/10.1038/s41598-019-38829-8

Ye, J., Hu, A., Ren, G., Chen, M., Tang, J., Zhang, P., Zhou, S., & He, Z. (2018). Enhancing sludge methanogenesis with improved redox activity of extracellular polymeric substances by hematite in red mud. Water Research, 134, 54–62. https://doi.org/10.1016/j.watres.2018.01.062

Yenigün, O., & Demirel, B. (2013). Ammonia inhibition in anaerobic digestion: A review. In Process Biochemistry, 48(5-6), 901–911. https://doi.org/10.1016/j.procbio.2013.04.012

Yin, Q., Gu, M., & Wu, G. (2020). Inhibition mitigation of methanogenesis processes by conductive materials: A critical review. Bioresource Technology, 317, 123977. https://doi.org/10.1016/j.biortech.2020.123977

Yin, Q., Miao, J., Li, B., & Wu, G. (2017). Enhancing electron transfer by ferroferric oxide during the anaerobic treatment of synthetic wastewater with mixed organic carbon. International Biodeterioration and Biodegradation, 119, 104–110. https://doi.org/10.1016/j.ibiod.2016.09.023

Zheng, Z., Liu, J., Yuan, X., Wang, X., Zhu, W., Yang, F., & Cui, Z. (2015). Effect of dairy manure to switchgrass co-digestion ratio on methane production and the bacterial community in batch anaerobic digestion. Applied Energy, 151, 249–257. https://doi.org/10.1016/j.apenergy.2015.04.078

Zhou, S., Xu, J., Yang, G., & Zhuang, L. (2014). Methanogenesis affected by the co-occurrence of iron(III) oxides and humic substances. FEMS Microbiology Ecology, 88(1), 107–120. https://doi.org/10.1111/1574-6941.12274

Zhuang, L., Tang, J., Wang, Y., Hu, M., & Zhou, S. (2015). Conductive iron oxide minerals accelerate syntrophic cooperation in methanogenic benzoate degradation. Journal of Hazardous Materials, 293, 37–45. https://doi.org/10.1016/j.jhazmat.2015.03.039

Zwietering, M. H., Jongenburger, I., Rombouts, F. M., van ' K., & Riet, T. (1990). Modeling of the Bacterial Growth Curve. Applied and Envrionmental Microbiology, 56(6): 1875–1881. 10.1128/aem.56.6.1875-1881.1990