The Addition of Hatchery Liquid Waste to Dairy Manure Improves Anaerobic Digestion

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ABSTRACT

The objective of this study was to determine the optimal inclusion level of liquid egg hatchery waste for the anaerobic co-digestion of dairy cattle manure. A completely randomized experimental was applied, with seven treatments (liquid hatchery waste to cattle manure ratios of 0: 100, 5:95, 10:90, 15:85, 20:80, 25:75 and 30:70), with five replicates (batch digester model) each. The evaluated variables were disappearance of total solids (TS), volatile solids (VS), and neutral detergent fiber (NDF), and specific production of biogas and of methane. Maximum TS and VS disappearance of 41.3% and 49.6%, were obtained at 15.5% and 16.0% liquid hatchery waste inclusion levels. The addition of 22.3% liquid hatchery considerably reduced NDF substrate content (53.2%). Maximum specific biogas production was obtained with 17% liquid hatchery waste, with the addition of 181.7 and 229.5 L kg^{-1}TS and VS, respectively. The highest methane production, at 120.1 and 151.8 L CH_4 kg^{-1}TS and VS, was obtained with the inclusion of 17.5 and 18.0% liquid hatchery waste, respectively. The addition of liquid hatchery waste at ratios of up to 15.5% in co-digestion with cattle manure reduced solid and fiber levels in the effluent, and improved biogas and methane production.

INTRODUCTION

The global demand for food is increasing every year under the pressure of the worldwide population growth. Food supply has been possible due to the expansion and intensification of the meat production systems. However, these systems also produce a significant amount of waste, which is characterized by high organic and nutrient load, and therefore, it is highly polluting. On the other hand, these components may also favor biological treatment processes, recycling energy and nutrients, thereby resulting in lower environmental impact.

Thousands of eggs are incubated in Brazil, and the average efficiency of incubation is near 85%, producing a large volume of waste (Kobashigawa *et al.*, 2008). Carvalho *et al.* (2013) estimated that the daily incubation waste is around 1.6 and 1.3 tons per 100,000 incubated eggs, both in broiler and laying hen hatcheries.

Hatchery waste can be divided into two different fractions: solid and liquid. Solid waste consists of infertile eggs, unhatched eggs, dead chicks, dead or culled chicks, and eggshells, whereas liquid waste includes water used for cleaning the setters, hatchers, and hatchling handling areas (Araújo & Albino, 2011). This waste material has a high organic load, and contains, on average, 33.1% crude protein, 29% ether extract, 21.5% ash, and 28.8 MJ kg^{-1} gross energy (Glatz *et al.*, 2011). The inappropriate disposal of this waste can cause severe environmental issues.

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Anaerobic digestion is one of the most common techniques for the treatment of effluents with high concentrations of organic matter (Nuchdang & Phalakornkule, 2012; Aggarangsi *et al.*, 2013). During this process, the organic matter in waste is broken down in the absence of oxygen, yielding biogas, which contains high levels of methane. Also, the remaining digested and stabilized material can be used as biofertilizer.

Hatchery liquid waste can be submitted to anaerobic digestion because it can be easily added to the digesters. However, it cannot be individually digested because it contains high nitrogen (expressed as crude protein content) and ether extract levels. Therefore, anaerobic co-digestion, associating two or more types of waste, may be used as an alternative method for hatchery waste treatment. The anaerobic digestion of hatchery liquid waste with other waste materials may supplement deficiencies, and result in more efficient breakdown of the organic fractions and increase biogas and methane production.

According to Mata-Alvarez *et al.* (2014), the co-digestion of dairy cattle manure with energy-rich agroindustry waste provides better carbon (C) to nitrogen (N) ratio, resistance to acidification of the medium, and higher levels of nutrients available for digestion. An alternative for the digestion of liquid hatchery waste is the association with cattle manure. Although cattle waste can be individually biodigested and its anaerobic digestion is widely used, its high fiber content and limited N levels maybe balanced by the high lipid and N concentrations found in hatchery waste. There is a considerable body of research (Orrico Jr. *et al.*, 2012) on the anaerobic digestion of cattle manure; however, there are few studies on the anaerobic digestion of hatchery waste, and often discuss only the limitations of the use of its solid and liquid fractions.

This study aimed at determining the optimal addition level of liquid hatchery waste to dairy cattle manure submitted to anaerobic co-digestion to reduce solid and fiber content and to increase biogas and methane production.

MATERIAL AND METHODS

The study was carried out in Dourados, state of Mato Grosso, Brazil (latitude 22°11'55" S, longitude 54°56'7" W, and altitude of 452 m). The climate of this region is wet mesothermal (Cwa, according to Köppen climate classification), with 20-24 °C average environmental temperature and 1250-1500 mm annual rainfall.

A completely randomized experimental design was adopted, with seven treatments. Treatments consisted of the anaerobic digestion of different ratios of liquid hatchery waste to dairy-cattle manure (0:100; 5:95; 10:90; 15:85; 20:80; 25:85; and 30:70, respectively), on total solid (TS) content basis, in five different biodigesters (replicates), totaling 35 experimental units.

Cattle waste was collected on a dairy farm by scraping the floor where the cows were housed, prioritizing fresh manure and trying to avoid contamination from the floor. A commercial hatchery provided the liquid hatchery waste, which primarily consisted of infertile and abnormal eggs of the last egg batch transferred on day 18 of incubation from the setter to the hatcher. During disposal at the hatchery, the waste was separated in two fractions: one consisting of eggshells (solid fraction), and the other of egg contents (liquid fraction), which was used for the anaerobic co-digestion process in the present experiment. After collection, both cattle manure and hatchery waste were submitted to the Agricultural Waste Management Laboratory of the Federal University of Grande Dourados for chemical analyses (Table 1).

Table 1 – The chemical composition of the wastes and inoculum used for the preparation of the substrates.

Components	Manure	Liquid hatchery waste	Inoculum
TS (%)	19,00	26,70	1,44
VS (% of TS)	79,23	96,20	67,4
EE (% of TS)	2,00	37,60	-
N (% of TS)	2,25	7,90	-
C (% of TS)	23,40	40,27	-
NDF (% of TS)	58,93	-	-

Before the substrates were prepared for digestion, the inoculum was produced by feeding batch digesters with dairy manure diluted in water at a concentration of 3% TS. The inoculum was considered ready when maximum methane concentration was achieved and maintained at 82.3%. The inoculum was added at 10% TS to each digester at the beginning of the experimental period.

The digestion substrates, containing different ratios of dairy cattle manure, hatchery residue, inoculum, and dilution water were prepared to contain $5 \pm 0.5\%$ TS (Table 2). The biodigesters were evaluated during the period of biogas production (105 days). The biodigesters were kept under environmental conditions, and protected from sunlight and rain throughout the experimental period.

Table 2 – Chemical composition and pH of the evaluated substrates and effluents, according to treatment.

Variables	Experimental treatments (%liquid hatchery waste)						
	0	5	10	15	20	25	30
TS (%)	4.63	5.21	5.05	5.03	4.83	5.53	5.89
VS (% of TS)	78.05	78.90	79.74	80.59	81.44	82.29	83.14
EE (% of TS)	1.80	3.58	5.36	7.14	8.92	10.70	12.48
N (% of TS)	2.03	2.31	2.59	2.87	3.16	3.44	3.72
C (% of TS)	21.06	21.90	22.75	23.59	24.43	25.28	26.12
NDF (% of TS)	53.04	50.09	47.14	44.20	41.25	38.30	35.36
Initial pH	7.71	7.77	7.88	7.93	7.77	7.89	7.88
Final pH	7.30	7.61	7.61	7.57	7.70	7.74	7.74

The digesters basically consisted of with two straight PVC cylinders, measuring 150 and 100 mm diameters each, and a 65-mm diameter container for the storage of the material to be fermented, with an average volumetric capacity of 1.3 L of substrate in fermentation each. The 100-mm cylinder was inserted into the 150-mm cylinder so allow the space between the outer wall of the inner cylinder and the inner wall of the outer cylinder to hold a volume of water ("water seal"). The top end of the 100-mm diameter cylinder was closed end and had a opening for the release of biogas. It was kept immersed in the water seal to provide anaerobic conditions and to store the biogas produced, as shown by Sunada *et al.* (2014). The volume of biogas produced per day was determined by measuring the vertical displacement of tubes according to their cross-sectional area. The gas volume was corrected according to standard conditions for temperature and pressure.

Biogas composition was analyzed weekly to determine the volume (L) of methane produced. Biogas composition was determined using a gas chromatograph (Finnigan GC-2001, equipped with Porapack Q molecular sieve columns), and a thermal conductivity detector gas analyzer (GA-21 Plus). The production of biogas and methane were calculated considering the volume (L) and the amount (kg) of TS and volatile solids (VS) added to the digesters.

TS and VS levels and hydrogen ionic potential (pH) were determined in the beginning (affluent) and end (effluent) of the biodigestion, according to the methodology described by APHA (2005). Neutral detergent fiber (NDF) content was measured according to the method proposed by Detmann *et al.* (2012). Organic carbon contents in the substrates and effluents were estimated by the method described by Kiehl (1985), and the ether extract was determined according to the recommendations of Silva & Queiroz (2006).

The results were submitted to analysis of variance (ANOVA), considering the amounts of addition of liquid hatchery waste as a source of variation. Orthogonal contrasts were applied to evaluate linear, quadratic, and cubic effects. Calculations and analyses were carried out using the R software (version 3.1.0 for Windows).

RESULTS AND DISCUSSION

The disappearance of TS and VS presented a quadratic behavior (Table 3 and Figure 1), with optimal doses of liquid hatchery waste addition to cattle ma-

Table 3 – Modelof regression, followed by r², P (probability), and CV (coefficient of variation) values, of the disappearance of total solids (TS), volatile solids (VS), and neutral detergent fiber (NDF), and biogas and methane (CH_4) production during the co-digestion of the substrates prepared with cattle manure and theaddition on increasingliquid hatchery waste levels.

Evaluated parameter	Regression model	r²	P	CV
TSdisappearance (%)	$y = -0.0728x^2 + 2.2601x + 23.731$	0.93	<0.001	4.2
VSdisappearance (%)	$y = -0.067x^2 + 2.1414x + 31.753$	0.91	<0.001	4.2
NDFdisappearance (%)	$y = -0.0614x^2 + 2.7385x + 22.649$	0.91	<0.001	4.3
Biogas (L kg^{-1}ofTS added)	$y = -0.2489x^2 + 8.5435x + 108.43$	0.62	<0.001	5.4
Biogas (L kg^{-1}ofVSadded)	$y = -0.2774x^2 + 9.8434x + 142.2$	0.79	<0.001	5.6
CH_4 (% in biogas)	$y = -0.049x^2 + 1.650x + 54.50$	0.87	<0.001	4.9
CH_4 (L kg^{-1}ofTSadded)	$y = -0.2004x^2 + 7.0378x + 58.354$	0.82	<0.001	5.1
CH_4 (L kg^{-1}ofVS added)	$y = -0.2295x^2 + 8.2973x + 76.818$	0.82	<0.001	5.4

nure of 15.5 and 16.0%, respectively, which resulted in 41.3% and 49.6% disappearance of TS and VS in the effluents relative to the substrates. These results indicate better degradation of solids during biodigestion with the inclusion of liquid hatchery waste, which increased TS disappearance in74.0%compared with the control treatment; while VS in 56.2%. This suggests that substrate quality improved with the addition of hatchery residue, increasing substrate nutrient levels, which allowed better degradation (Table 2).

The addition of liquid hatchery waste at ratios higher than 16.0% limited TS and VS degradation (Figure 1), suggesting that the effectiveness of digestion was impaired. A possible explanation is that the lipid or nitrogen levels added to the substrate by the hatchery waste may have been excessive. The adverse effects of lipids on biodigestion were reported by Orrico *et al.* (2015), who used cooking oil waste for the co-digestion of pig manure, and found that oil ratios higher than 5.7% reduced the ST and SV degradation during anaerobic digestion due to the accumulation of long-chain fatty acids resulting from lipid digestion.

Markou (2015) emphasized the impact of waste containing high N levels, and consequently low C:N ratios, on biodigestion efficiency. That author recommends C:N ratios greater than 8:1 to prevent the accumulation of ammonia, which may limit the development of microbes, particularly of the methanogenic flora. Substrates containing liquid hatchery waste ratios higher than 15% of (Table 2) presented degradation rates, as shown by the lower TS disappearance.
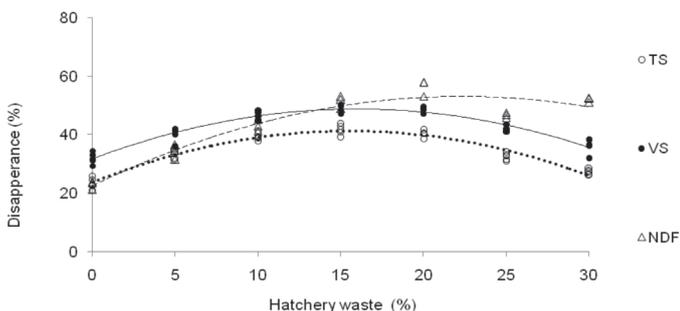

Figure 1 – Disappearance of total solids (TS), volatile solids (VS), and neutral detergent fiber (NDF) during the co-digestion of cattle manure with increasing liquid hatchery waste levels.

The maximum VS degradation rates obtained in the present study were lower than those reported by El-Mashad & Zhang (2010), of 68%, when evaluating the co-digestion of cattle manure with food waste. This difference may be associated with the concentration of solids being digested. In the study by El-Mashad & Zhang (2010), TS contents in the substrates were

only 2.1%, while in the present research the waste contained concentrations between 4.6 and 5.9% TS. In addition, the level of fibrous compounds may have limited the extension of VS breakdown, since in the present study NDF levels ranged from 35.3 to 53.0%, while those reported by El-Mashad & Zhang (2010) were not higher than 26.7%.

Substrate NDF disappearance (Table 3, Figure 2) increased up to 22.3% of liquid hatchery waste inclusion, reaching 53.2% maximum disappearance, which corresponds to 134.0% higher disappearance compared with the control treatment. This effective reduction of fiber in the substrates may be explained by the degradation of hemicellulose during biodigestion, because hemicellulose is the most available compound in NDF. In a similar study, Manarelli *et al.* (2014), evaluating the anaerobic co-digestion of cattle manure with cooking oil waste, obtained 49.5% NDF reduction in with an oil inclusion of 6.5%, and verified that hemicellulose fraction was the main component of the degraded fiber.

The degradation of organic material produces biogas, which volume depends on the degradation of solids. Hatchery liquid waste ratios of up to 17% increased biogas production (Figure 2), corresponding to 181.7 and 229.5 L kg^{-1} of TS and VS addition. These values were 67.5 and 61.39% higher than those obtained in biodigester containing no hatchery liquid waste.
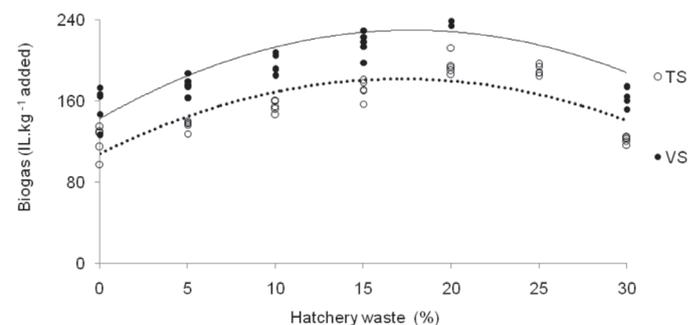

Figure 2 – Specific biogas production as a function ofTSandVS addition tosubstrates-consisting of cattle manure and increasing liquid hatchery waste levels.

This result may be explained by the findings of Long Hunter *et al.* (2012), who observed that the addition of carbon-rich compounds, such as fats, greases and oils, for the co-digestion of animal waste increased biogas production by 30% or more, according to N substrate levels. These results also explain the decrease in biogas production at liquid hatchery waste inclusions greater than 17%, which resulted in C: N ratios below 8:1, as previously mentioned. In a study conducted by Zhang *et al.* (2013), cattle manure was co-digested with food waste, and yielded up to 420 L of biogas kg^{-1}of VS, which is higher than the yield obtained in

the present study. This difference is may be due to the lower proportion of cattle manure used by Zhang *et al.* (2013), rendering the substrate more accessible to degradation.

The time biogas starts to be produced determines the substrate residence time in the biodigesters. Its determination is essential to plan the use of biodigesters under field conditions, with the aim of optimizing the volume of waste treated as a function of time. The experimental results (Figure 3) show that the time of biogas started to be produced was anticipated and biogas production increased with the increase of liquid hatchery waste inclusion levels in the substrate. In the 5th week of the retention of the substrates in the biodigesters, total biogas production was 39.2% for the substrate with no liquid hatchery waste inclusion and 46.4, 52.2, 54.3, 58.5, 58.8, and 59.9% in the biodigester with substrates containing 5, 10, 15, 20, 25, and 30% of liquid hatchery waste, respectively.
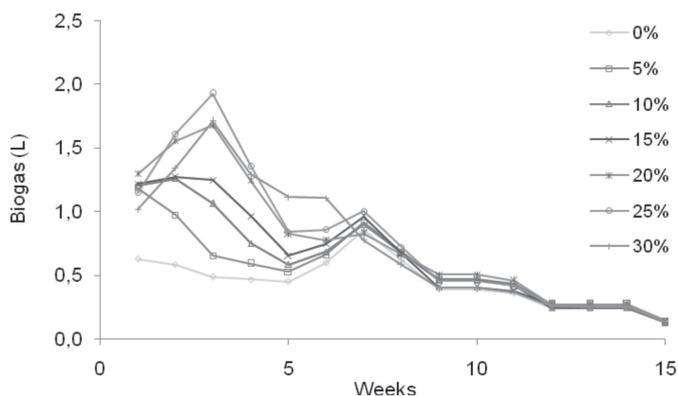

Figure 3 – Time distribution of biogas production by substratesconsisting of cattle manure and increasing liquid hatchery waste levels.

In addition to biogas production, methane concentration is also an important factor to be considered due to the energy potential of this gas. The highest specific methane productions (Figure 4) were obtained with17.5 and 18% liquid hatchery waste inclusion ratios at120.1 and 151.8 L. CH_4 kg^{-1} of TS and VS addition, respectively, corresponding to 105 and 97.6% higher methane production compared with the control treatment (with no liquid hatchery waste inclusion). This indicates that the inclusion of more available nutrients in the digestion environment by the addition of the liquid hatchery waste favored the fermentation of the substrate.

The pH values of the substrates (Table 2) show that the pH of the effluents were slightly alkaline, and consequently did not limit the activity of the methanogenic microorganisms.
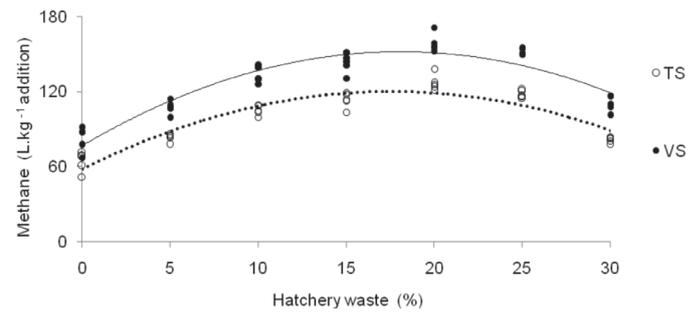

Figure 4 – Methane specific production as a function of TS and VS addition to substratesconsisting of cattle manure and increasing liquid hatchery waste levels.

Orrico Junior *et al.* (2015) evaluated the anaerobic co-digestion of sheep manure with crude glycerin, which is a readily-available energy source. Those authors obtained 440.0L of methane/kg VS addtion with 6% glycerin inclusion in the substrate. This methane volume is higher than that verified in the current study, probably due to the high energy levels of crude glycerin and of the manure, because the sheep were fed high levels of a concentrate. In the study of Orrico Junior *et al.* (2015), a maximum methane content of 83.8% was achieved the inclusion of 11.73% crude glycerin, with a calorific value of 30,960 KJ m^{-3} of biogas, which was 13.16% higher than that of the biogas generated by the substrate with no crude glycerin inclusion. That calorific value was calculated using the equations proposed by Mitzaff (1998), which can be applied to the results obtained in the present study. Considering the methane ratio in the biogas obtained in the control treatment (54.5%), and the maximum methane ratio of 68.4% obtained with 16.8% hatchery liquid waste inclusion (regression model shown in Table 3), calorific values of 18,519 and 23,242 kJ m^{-3} of biogas were calculated for inclusions of 0 and 16.8% of liquid hatchery waste, respectively. This result indicates that the substrate with 16.8% of liquid hatchery waste produced 25.5% more methane.

The distribution of methane production (Figure 5) as a function of time is consistent with the distribution of biogas production, indicating that the addition of liquid hatchery waste to cattle manure allowed higher methane production.

Based on the results of the present study, the anaerobic co-digestion of dairy manure with hatchery liquid waste may be recommended as an effective treatment of both wastes, thereby reducing the organic load of biofertilizers. In addition to higher degradation of solids, higher biogas and methane production and calorific biogas power obtained.
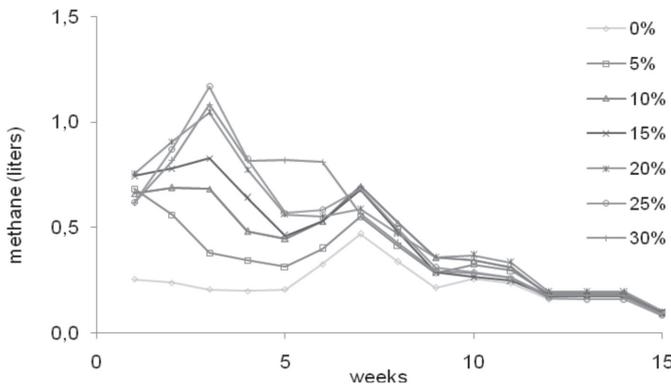
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Figure 5 – Time distribution of methane production by substrates consisting of cattle manure and increasing liquid hatchery waste levels

CONCLUSIONS

The anaerobic co-digestion cattle manure with inclusion of up to 15.5% liquid hatchery waste increases the degradation of solids and fibrous compounds in the substrate, and the production of biogas and methane. The inclusion of any dose of liquid hatchery waste to cattle manure submitted to anaerobic digestion anticipated the production of biogas and methane, which achieved higher volumes at shorter digestion times.

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