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Cattle manure, dewatered pig manure and chicken manure were pre-treated in a high-temperature reactor under isochoric conditions for 15 min at temperatures between 100 and 225°C with 25°C intervals to study the effect on their methane yield. After 27 days of batch incubation, cattle manure showed a significant improvement in its biochemical methane potential (BMP) of 13% at 175°C and 21% at 200°C. Pig manure showed improvements at temperatures of 125°C and above, with a maximum 29% increase in yield at 200°C. The BMP of chicken manure was reduced by 18% at 225°C, but at lower temperatures there were no significant changes. It was found that this method of pre-treatment could be feasible if sufficient surplus energy was available or if the energy used in the pre-treatment could be recovered.

Keywords: BMP; manure; biogas; thermal; pre-treatment; energy requirements

1. Introduction

Anaerobic digestion of manure is a means of producing carbon-neutral energy while decreasing its biodegradable content. In addition, this approach reduces methane emissions associated with raw manure storage [1], and the resulting digestate can be used as a soil amendment. However, one of the main issues with manure when used for biogas production is its recalcitrance to microbial degradation. Anaerobic digestion of substrates that contain complex insoluble organic material is limited by hydrolysis [2,3]. Manure contains up to 40–50% bio-fibres [4]. The fibre fraction of manure consists of lignocellulosic material containing mainly undigested plant material and often bedding material, as well as fats and nutrients [5]. The lignocellulosic complex consists of lignin, hemicellulose and cellulose, of which the cellulose and hemicellulose fractions are degradable anaerobically. These three components are closely associated and form a tight three-dimensional complex that restricts the access of hydrolytic enzymes, effectively resisting microbial degradation. Triolo *et al.* [6] have shown that lignin content has a major negative effect on the biochemical methane potential (BMP) of various types of manure. Improving the biodegradability of the lignocellulosic part of the bio-fibres will improve the BMP of the substrate. One way of achieving this is by pre-treating the substrate prior to anaerobic digestion. The pre-treatment step is aimed mainly at changing the structure or the composition of the substrate to improve the hydrolysis rate by breaking down the lignocellulose and increasing access to enzymatic attack [7,8]. High-temperature pre-treatment of pig manure and steam treatment of bio-fibres obtained from substrates that have previously undergone anaerobic digestion have been shown to increase the BMP by 25–64% [9–11]. These studies showed the potential of thermal pre-treatment in improving in the BMP of manure.

Although there have been a few studies on the effect of high temperature on manure, few have focused on a wide range of temperatures, particularly on temperatures of 200°C and above, and very little has been done to see the effect of such pre-treatments on chicken manure. Thus, the aim of the present study was to determine the effect of thermal pre-treatment at different temperatures on the methane yields of cattle, pig and chicken manure and observe the trend between the pre-treatment temperature and the BMP, if any.

2. Materials and methods

2.1. Experimental design

Cattle, pig and chicken manure samples were pre-treated at 100°C, 125°C, 150°C, 175°C, 200°C and 225°C. The pre-treatments carried out on cattle manure at 100°C, 150°C and 200°C, and were repeated twice to serve as treatment replicates (three independent trials in total) and to confirm the repeatability of the pre-treatment process. The acid detergent fibre (ADF), acid detergent lignin (ADL) and the neutral detergent fibre (NDF) contents were measured for all the samples.

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2.2. Materials

Cattle manure was collected from stables housing dairy cows located at Aarhus University, Research Centre – Foulum, Tjele (Denmark). There was no bedding used, and the cattle manure on the floor was scraped twice every day. The samples were collected directly from the scraped heap. The dry matter (DM) content of the cattle manure was 11.6% and the volatile solids (VS) content was 80.9% of the DM.

The pig manure was collected from a farm located in Skanderborg (Denmark). The manure had been dewatered using a decanting centrifuge (GEA Westfalia, UCD 305, Germany). The DM content of the pig manure was 30% of which 84.2% was VS. Chicken manure was collected from a broiler farm located in Hammel (Denmark) and was from chicken houses that used sawdust as bedding material. The DM content of the chicken manure was 66.9% and had a VS content of 88.4%.

The pig and chicken manures were diluted to 20% DM using deionized water to facilitate mixing during pre-treatment.

2.3. Methods

2.3.1. Pre-treatment

The pre-treatments were performed in a bench-scale high-temperature and pressure reactor (Parr Instrument Company, USA, model Parr 4524). The pre-treatment can be considered isochoric as the reactor is inelastic (i.e. the volume remains constant) and consequently the pressure inside the reactor changes according to the changes in temperature. The stainless steel reactor had a total volume of 2 L. The reactor was stirred mechanically and was heated by convection using an external electric heating coil. The reactor was also fitted with a thermo-well and a pressure gauge. The temperature was monitored and controlled using a temperature probe that was inserted into the thermo-well and connected to a proportional integral derivative (PID) temperature controller. The reactor was completely sealed during the pre-treatment process. Once the material inside reached the selected temperature it was held at that particular temperature for 15 min, after which the reactor was cooled down to about 35°C using a water bath (with water at room temperature) before opening the reactor. The material was stored in a freezer immediately after the pre-treatment, until the start of the BMP assay. The effect of the freezing and subsequent thawing of solid fractions of digestate were studied [12] and it was found that the effect of this on the methane yield depended on the incubation temperature used during the BMP assay. Since all the samples used in the study were frozen and the incubation temperature for all the samples were the same it was assumed that effect of the freezing and thawing was the same on all the samples and hence the difference in the BMP yields if any was due to the pre-treatments applied.

2.3.2. BMP assays

The BMP assays included all the pre-treated manure samples along with the untreated manure samples (raw manure) as controls. The assays were based on the method proposed by Owen *et al.* [13] and done in triplicate for a period of 90 days at a mesophilic temperature of 35°C ± 1°C. The anaerobic sludge used as inoculum was obtained from a full-scale anaerobic digester located at the Research Centre – Foulum, Tjele (Denmark). This digester is fed with a mixture consisting of 70% manure, 20% maize and 10% grass silage (on a wet weight basis). The inoculum was pre-incubated as suggested by Angelidaki *et al.* [14] for about 28 days to ensure that the amount of biogas produced by the inoculum itself was minimal. Bottles containing only the inoculum were included in the batch assay to determine the BMP produced by the inoculum itself. No additional nutrients were added as the inoculum was manure-based and hence already included the nutrients required [15]. Each of the manure samples were digested in batch reactors, which were 500 mL glass bottles, each containing 200 g ± 5 g of inoculum. The batch reactors were sealed and the headspace of each was flushed with 99.9% nitrogen gas to remove gaseous oxygen from the system before being placed into the incubator. The volume of the biogas produced in each of the reactors was measured using an acidified water displacement method (pH < 2) and a small gas sample was taken for analyzing the composition. The amount of methane produced in terms of litres of methane per kilogram of volatile solids added was calculated (L/kg VS). The results were analysed using the *t*-test function available in MicroSoft Excel 2007, at a 95% confidence level.

2.3.3. Analysis

The ADF and ADL were performed based on the method proposed by Van Soest [16]. The NDF was measured according to the method proposed by Van Soest and Wine [17]. The cellulose content was calculated by subtracting the ADL from the ADF (ADF – ADL) and the hemicellulose was calculated by NDF – ADF [18]. The composition of the biogas was determined by collecting 300 mL of the biogas sample, flushing a 22 mL glass vial with it. The sample was analysed on a Varian 3600 gas chromatograph equipped with a thermal conductivity detector. The DM and VS were measured according to APHA [19]. The pH of the manure samples were measured by using a pH meter (Metrohm AG, Herisau, Switzerland).

3. Results and discussion

The results of the analyses performed on the manure samples are shown in Table 1. The BMP results, in L/kg VS, are shown in Table 2 along with their standard deviations, at 27, 60 and 90 days to see the change in the BMP through the entire period. Table 3 shows the energy considerations. It includes the energy gain due to pre-treatment (only values

Table 1. Results of the analyses performed on the pre-treated and untreated manures.

		NDF (%)	ADF (%)	ADL (%)	Cellulose (%)	Hemicellulose (%)
Cattle	Untreated	51	32	11	21	19
	100°C	51	32	11	21	19
	125°C	52	32	12	20	20
	150°C	55	34	11	23	20
	175°C	na	na	na	na	na
	200°C	52	36	14	22	16
	225°C	na	na	na	na	na
Pig	Untreated	70	37	7	30	32
	100°C	71	38	7	31	32
	125°C	73	40	8	32	33
	150°C	69	38	9	29	31
	175°C	71	39	9	31	32
	200°C	71	41	9	32	30
	225°C	na	na	na	na	na
Chicken	Untreated	44	24	4	19	20
	100°C	46	23	4	19	23
	125°C	47	23	4	19	23
	150°C	46	23	5	19	23
	175°C	44	25	5	19	20
	200°C	40	25	5	20	15
	225°C	36	29	9	20	8

Note: NDF, ADF and ADL and hence the cellulose and the hemicellulose are in percentage of dry matter. na = not available.

Table 2. The BMPs of all the manure samples.

	Day	Untreated (L/kg VS)	100°C (L/kg VS)	125°C (L/kg VS)	150°C (L/kg VS)	175°C (L/kg VS)	200°C (L/kg VS)	225°C (L/kg VS)
Cattle	27	244 (32)	222 (9)*	242 (9)	244 (5)	275 (3)*	296 (1)*	259 (8)
	60	268 (36)	247 (8)*	270 (9)	267 (7)	298 (3)	317 (4)*	275 (8)
	90	281 (35)	264 (11)	287 (9)	281 (9)	311 (4)*	331 (4)*	292 (9)
Pig	27	215 (11)	208 (12)*	234 (15)*	232 (9)*	240 (3)*	277 (9)*	272 (18)*
	60	266 (12)	263 (13)	290 (17)*	268 (8)*	293 (3)*	325 (11)*	314 (17)*
	90	294 (12)	289 (15)	315 (16)*	311 (9)	315 (5)*	344 (14)*	328 (10)*
Chicken	27	334 (3)	317 (2)	314 (7)	264 (6)	299 (5)	312 (9)	273 (7)*
	60	357 (4)	340 (1)	336 (7)	306 (8)	322 (5)	331 (9)	304 (7)
	90	369 (9)	354 (2)	346 (7)	316 (9)	333 (5)	340 (8)	312 (8)

Note: Standard deviations in brackets. *Statistically significant difference when compared to the untreated manure ($\alpha = 0.05$).

that showed a significant increase have been included) and the energy inputs required for such a pre-treatment. The change in the BMP of manure (as a percentage) due to the pre-treatments when compared to the untreated manure is shown in Figure 1. The curves representing the cumulative methane yield for each of the untreated manure sample are shown in Figure 2. The pH of the untreated cattle, pig and chicken manure were 7.4, 7.9 and 6.9, respectively.

Literature values for cattle manure, solid fraction of pig manure and chicken manure show a wide range of methane yields; 100 to 300 L/kg VS, 159 to 506 L/kg VS and 300–600 L CH_4/kg VS, respectively [20–22]. The methane yields of the untreated manure samples obtained in our study are well within this range. The increase in the BMP of lignocellulosic material due to high-temperature pre-treatment can be attributed to many factors: the release of easily degradable material such as sugars due to dissolution of the hemicellulose and a decreased degree of polymerization of cellulose [23], or a change in the structure of the substrate increasing the accessibility of material to the microbial enzymes [24].

3.1. Changes in the BMP of manure

In the case of cattle manure, as seen in Table 2, significant improvement in the BMP resulted when temperatures of 175°C and 200°C were used for pre-treatment. At those temperatures, the increase in the BMP is seen throughout the 90 days. The increase in the BMP at higher pre-treatment temperatures could be due to dissolution of hemicellulose, as seen in the slight reduction of the hemicellulose content at a pre-treatment temperature of 200°C in Table 1. The dissolution of the hemicellulose would have provided

Table 3. Energy considerations (per tonne of manure).

	I (°C)	II* (kW h)	III* (kW h)	IV* (kW h)	V* (kW h)
Cattle	175	177	0.11	30	29
	200	204	0.12	49	49
Dewatered pig	125	116	0.08	36	27
	150	142	0.09	31	21
	175	167	0.11	47	37
	200	192	0.12	115	106
	225	217	0.14	107	97

Notes: I: Pre-treatment temperature, II: Required energy input to raise the temperature, III: Heat losses from the pre-treatment unit, IV: Energy gain obtained due to pre-treatment of the manure, V: Net energy gain without considering the energy required for heating the manure to the pre-treatment temperature and after subtracting the heat losses and, in the case of pig manure, subtracting both heat losses and energy required for the dewatering process from the increased energy. *Calculations for the values mentioned in these columns are explained in detail in the supplementary section.

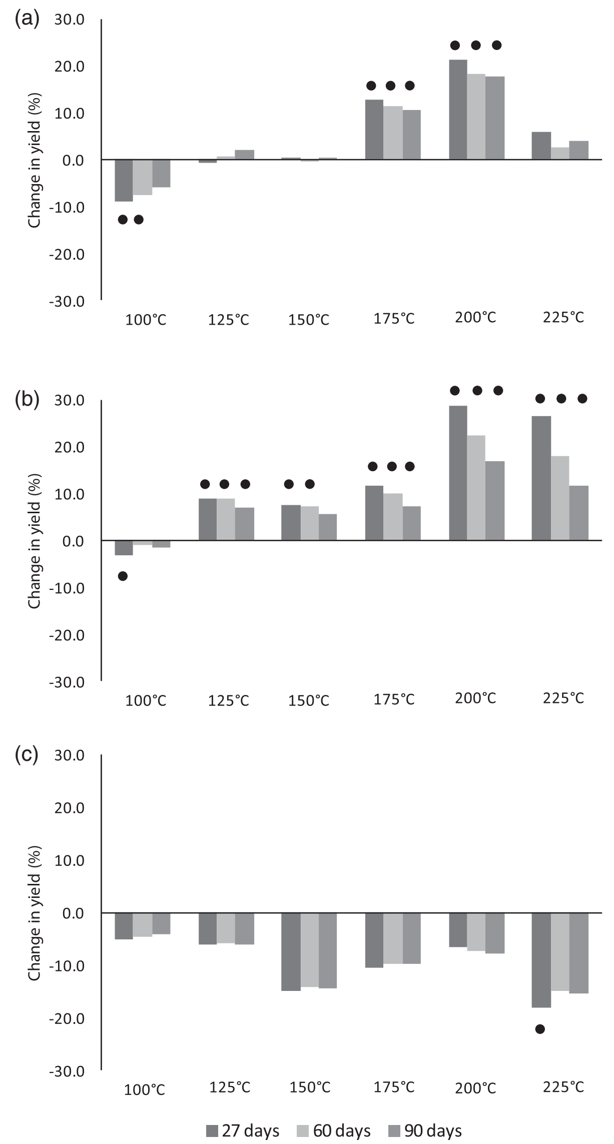

Figure 1. Change in BMP yields of pre-treated (a) cow manure, (b) pig manure and (c) chicken manure when compared to the respective untreated manure samples, as percentages. The percentage change is shown for each pre-treatment temperature at 27, 60 and 90 days. Circles on top indicate that the value is significant at $\alpha = 0.05$.

more access to the cellulosic content in the lignocellulosic complex, thereby leading to an improvement in the BMP. Hemicellulose under neutral conditions solubilizes at temperatures above 150°C [25]. Acidic conditions catalyse the solubilization of hemicellulose and could reduce the required pre-treatment temperature [25], but the pH of the cattle manure used in this study was 7.4, which is nearly neutral and could be a reason for the dissolution of the hemicellulose not occurring at lower pre-treatment temperatures. There was no significant difference in the pre-treatment replicates throughout the 90-day period for all three pre-treatment temperatures, which confirms the repeatability of the pre-treatment process (refer to the supplementary section).

For pig manure, however, an increase in BMP was seen at all temperatures from 125–225°C. The BMP reached the highest yield at 200°C and then decreased slightly at 225°C. This is consistent with other research, which has showed the formation of inhibitors at higher temperatures [25]. As seen in Table 1, the cellulose and hemicellulose fractions change very little in pig manure after pre-treatment. The increase in BMP in dewatered pig manure could be attributed to structural changes in the lignocellulosic fractions. Kristensen *et al.* [26] have suggested that pre-treating straw hydrothermally caused hemicellulose removal and lignin relocation.Since there is little hemicellulose or lignin dissolution seen in our study, lignin relocation (therefore higher accessibility to cellulose) could be responsible for the improved BMP.

In our study a 29% increase in the BMP of pig manure was seen at a pre-treatment temperature of 200°C for a treatment period of 15 min. The study by Rafique *et al.* [10] showed that thermal pre-treatment of dewatered pig manure at 100°C for 1 h improved its BMP by 25%. This indicates that the length of the pre-treatment period is an

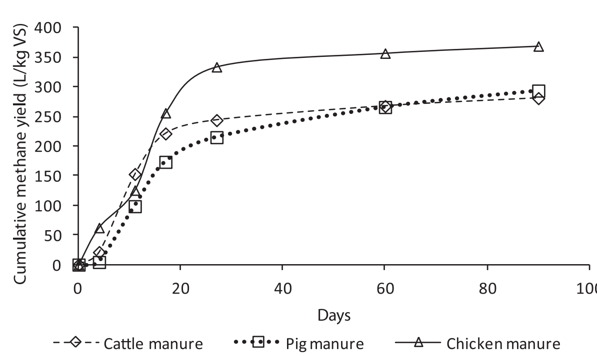

Figure 2. Cumulative methane production (L/kg VS) as a function of days for all the untreated manure samples.

important factor. This is especially important for practical applications.

There was no significant change in the BMP of the chicken manure after pre-treatment at temperatures up to 200°C, but there was a significant reduction in the BMP (18%) of the manure pre-treated at 225°C after 27 days of batch digestion. This reduction, however, was not significant after 60 days. The chicken manure (with no bedding material) in the study performed by Ardic and Taner [27] nevertheless showed improved BMP when pre-treated for 2 h at 100°C, indicating that an increase in the pre-treatment period could improve the BMP. The lack of significant changes in the BMP at temperatures below 225°C might be explained by the high degradability of the chicken manure in this study, giving a limited possibility for improvement. The reduction at 225°C could be due to the formation of inhibitors at high pre-treatment temperatures [25] and because there could be a loss of material due to disintegration to other organic compounds at pre-treatment temperatures above 200°C. But since the change in the BMP is not significant after 60 days, it is more likely that the reduction was primarily due to inhibition during the initial days of batch digestion, after which the inhibition was overcome.

3.2. Energy considerations

When considering thermal pre-treatment it is important to account for the energy that is gained as a result of the pre-treatment and the extra energy input that is required for the pre-treatment process. Table 3 shows the energy gains and the energy inputs required for pre-treating 1 tonne of manure, and the corresponding details of the calculations and assumptions are given in the supplementary section. The energy gain is the amount of extra energy that is obtained due to the pre-treatment in comparison with the untreated manure (Table 3, column IV). The energy input required includes the energy required to heat the material to the desired pre-treatment temperature (Table 3, column II), along with the energy required to compensate for the heat losses during the pre-treatment process (Table 3, column III) and, since we used dewatered pig manure, in the case of pig manure the energy input would also include the energy required for dewatering. The net energy gain is given by: energy gain – energy input. Only the cases where the pre-treatment significantly increased the BMP at 27 days in comparison to the untreated manure are shown in Table 3. The energy inputs required to pre-treat 1 tonne of manure at the selected pre-treatment temperatures were calculated using the basic heat transfer formula

$$Q = m \times Cp \times \Delta T \quad (1)$$

where Q is the energy required to raise the temperature of material with a specific heat capacity (Cp) and mass (m), from an initial temperature to the required pre-treatment temperature (the difference between these two temperatures being ΔT). The Cp of the manure samples was calculated based on the dry matter (DM) contents of the manure using the equation suggested by Chen [28,29],

$$Cp = 4.19 - (0.00275 \times \text{DM}) \quad (2)$$

The heat losses during the pre-treatment process were calculated based on a hypothetical pre-treatment unit with a capacity to pre-treat 1 tonne of manure.

In case of the pig manure, the amount of energy required for the dewatering process was also deducted from the energy gain due to pre-treatment and, based on the values from Møller *et al.* [30], the energy required to obtain 1 tonne of solids (at 30% DM) after dewatering is approximately 9.6 kW h.

From Table 3 it is obvious that the energy input required is greater than the energy gain that can be obtained via pre-treatment. There are two ways of solving this issue:

(1) Using surplus energy that would otherwise have been wasted for pre-treatment,
(2) Recovering the heat used for pre-treatment.

If these two scenarios can be applied then the net energy gained due to the pre-treatment might more than offset the energy input required to heat the material to the pre-treatment temperature, and so would only need to account for the heat losses in the case of cattle manure, and the heat losses and the energy for dewatering in the case of pig manure (Table 3, column V).

Full-scale biogas plants are usually associated with a combined heat and power unit (CHP). A CHP unit ideally converts 35–40% of biogas into electricity, 45–50% to heat that can be extracted and the remaining 15% are losses [31]. The heat generated by the CHP, if unused for other purposes such as district heating, is a source of low-cost energy that can be used for pre-treatment. This process can be made more energy-efficient by recycling the heat used to pre-treat the substrate for heating the anaerobic digester or the incoming material. The second option is where the pre-treated material is directly mixed with other incoming feed material streams to bring the temperature of the mixture to the operating temperature of the anaerobic digester, hence recovering nearly all the energy used for the pre-treatment.

A few studies have already demonstrated that thermal pre-treatment can be implemented in full-scale operations with a net energy gain. Pickworth *et al.* [32] describe a full-scale biogas plant that uses surplus heat from gas engines to thermally pre-treat their substrate, while Kepp *et al.* [33] describe a full-scale plant that uses the heat from the flue gases to produce steam for their pre-treatment plant. Thus, in a situation where surplus heat energy is available, or where the heat used for pre-treatment can be recovered, the addition of a pre-treatment step before the actual anaerobic digestion process can be justified.

4. Conclusion

The influence of thermal pre-treatment was different on different types of manure. The BMP of cattle manure improves at pre-treatment temperatures in the range 175–200°C. The pig manure BMP shows a positive effect from 125–225°C. Both cattle manure and pig manure have the highest improvement in methane yields at 200°C. There was no significant increase in the BMP of the chicken manure. This study shows that thermal pre-treatment has a positive effect on the BMP of both pig manure and cattle manure. The study also concludes that this method of pre-treatment is practical when surplus energy is available or when the heat used for the pre-treatment process can be recovered.

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