PRETREATMENT OF ROBUSTA COFFEE HULLS AND CO-DIGESTION WITH COW-DUNG FOR ENHANCED ANAEROBIC DIGESTION

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ABSTRACT

The effect of alkali pre-treatment on anaerobic digestion of Robusta coffee hulls with a lignin content of 29% was studied in batch cultures at 55°C. In order to investigate the effect of cow dung in minimising the effect of coffee-based inhibitory compounds, digestion experiments were performed with blends of pre-treated and non-treated coffee hulls and cow dung with 0,5,10,20,30,50,75 and 100% coffee hulls at a total volatile solids concentration of 1.8g/l fermentation volume. Methane production increased with increase in coffee hulls concentration up to 30 % for both substrates. The lowest production was observed at 100 % non-treated coffee hulls. Methane production was significantly higher (13-164 %) for the pre-treated substrates compared to the non-treated one. The highest increase was obtained at 100 % coffee hulls. Degradation of volatile solids decreased with increase in coffee hulls concentration for both substrates. The extent of degradation of the pre-treated substrate (59-94 %) was significantly higher than that obtained for the non-treated substrate (30-91%). The lowest extent of degradation was measured for the non-treated hulls at 100 %. It is possible that there was an inhibition of methanogenesis by coffee based compounds with increase in the coffee hulls concentration in the digestion mixture.

INTRODUCTION

Coffee is one of the important cash crops in East Africa. Its processing both by dry and wet methods generates large amounts of wastewater and solid residues. The latter include pulp and parchment husks from wet processing, and hulls from dry processing. Wastewater is disposed of untreated and big

fractions of the solid residues are either left at the factory site to rot or are burnt, thus contributing to environmental pollution (Jungersen *et al.* 1997, Bressani 1979). Some of the coffee processing residues has high volatile solids and sugar content and hence potentially good feedstocks for biogas digesters (Kivaisi & Rubindamayugi 1996, Pandey *et al.* 2000). However, the solid residues are also highly lignified, making them difficult to degrade anaerobically without pre-treatment. Bioassays performed to degrade composite coffee samples anaerobically have revealed mixed results. While the liquid fractions of the samples can be degraded by 60%, the lignocellulosic fraction may be degraded by only 9% (Azhar & Stuckey 1994).

Anaerobic bioconversion of lignocellulosic materials has been previously enhanced through physical, chemical and biological pre-treatment. The latter method has been found to be very effective but its economic feasibility is yet to be determined (Hatakka 1983, Pielecki & Zdzislaw 2001). Similarly, use of concentrated chemicals on a large scale is expensive. However, use of dilute acids and alkalis that are relatively cheap has been shown to be effective in improving the rate of hydrolysis (Kivaisi & Eliapenda 1994, Amathussatam & Abubacker 1999) which is the limiting step in biomethanation of lignocellulosic residues (Noike *et al.* 1995). Apart from high lignin content, coffee residues have been found to contain inhibitory compounds such as caffeine, tannins and polyphenols that appear to inhibit methanogenesis. Previous studies on anaerobic digestion of instant coffee waste and spent coffee grounds at mesophillic conditions reported a decline in biogas production and an accumulation of volatile fatty acids (VFA) with loading rates higher than 1.3 g/l in Continuous Stirred Tank Reactors (CSTRs) and attributed this decline to the presence of unidentified inhibitory compounds after 80 days of operation (Lane 1983, Kida *et al.* 1992). On the contrary, under thermophillic conditions, Konstenberg and Marcham (1993) achieved a more stable digestion at a loading (LR) of 8.6gVS/l/d in a continuous reactor. However, they too observed an accumulation of VFAs with prolonged operation. In order to improve on the process, Dinsdale *et al.* (1996), applying thermophilic digestion, used feed supplemented with $Ca(OH)_2$, N,P, and trace elements. They achieved a stable degradation of 60% of coffee grounds at a LR of 1.6 g COD/l/d but observed process instability due to VFA accumulation beyond 50 days of operation. The present study was therefore undertaken to assess the effect of co-digestion of coffee hulls with cow dung, the latter being the diluting

substrate of the coffee based inhibitory compounds. To enhance hydrolysis, alkali pre-treatment of the coffee hulls was employed followed by thermophilic digestion.

METHODS

Preparation of substrates

Robusta coffee hulls (RCH) were obtained from a processing factory in Bukoba, Tanzania and stored at room temperature (25-30°C) until required. The RCH were milled to pass through a 0.85 mm mesh sieve. A portion of the milled material was treated by immersion in 1M CaOH solution for 30 days at 55°C. The mixture was then neutralized with HCl and the liquid decanted. The treated RCH were then washed with tap water and sun dried. The dry material was stored at room temperature. The composition of RCH before treatment is shown in Table 1. Cow dung was obtained from a dairy farm in Dar es Salaam.

Table 1. Composition of RCH (mean ± standard deviation[c]) from a processing factory in Bukoba, Tanzania, 1999

Determination	% dry weight
Dry weight	84.03 ± 0.5
Volatile solids	68.27 ± 0.3
Ash	11.48 ± 0.5
NDF[a]	64.70 ± 0.4
ADF[b]	57.00 ± 0.4
Hemicellulose	7.70 ± 0.4
Cellulose	28.00 ± 0.2
Lignin	29.00 ± 0.5
Total nitrogen	1.36 ± 0.3
Reducing sugars:	
D-fructose	12.46 g/l
sucrose	15.97 g/l
D-glucose	7.77 g/l

[a] NDF = neutral detergent fibre
[b] ADF = acid detergent fibre
[c] n= 4

Inoculum

The inoculum used in this study was active sludge from sisal anaerobic digesters run at the Danish Technological Institute in Denmark. It was transported and stored at 4°C.

Fermentation experiments

Treated and non-treated RCH were fermented singly and in combination with varying amounts of cow dung and 30 ml of inoculum (Table 2) in 120 ml serum bottles closed with butyl rubber stoppers and aluminium caps. After closing the bottles were flushed with N_2 gas for 10 minutes in order to create anaerobic conditions. Incubation was done at 55°C in a shaking incubator (100 rpm) for 168 hours. Samples for methane and volatile fatty acids (VFA) determinations were taken at 6,12, 24, 48, 72, 96, 144 and 168 hours. At each of the sampling times, liquid samples were drawn from three bottles for each set of the experiment (Table 2) with a plastic syringe and stored at —20°C in eppendorf vials until analysis. The bottles were then flushed with oxygen free N_2 gas for 10 minutes followed by further incubation. Incubations with only treated and non-treated RCH and manure were included as controls. The overall substrate concentrations in all the experiments was 2 % VS. All the experiments were repeated three times.

Table 2. Distribution of substrate in experimental[a] vials for coffee husks obtained from a Bukoba factory and cow dung from a diary farm in Dar es Salaam, Tanzania

Experiment	Coffee composition % (gVS*)	Cow dung (gVS)
1	0	0.06
2	5 (0.003)	0.057
3	10 (0.006)	0.054
4	20 (0.012)	0.048
5	30 (0.018)	0.042
6	50 (0.03)	0.03
7	75 (0.045)	0.015
8	100 (0.06)	0
9	Control[b]	0

[a] inoculum was 30 ml anaerobic thermophilic sludge
[b] inoculum only, * VS amounts are shown in brackets

Analytical procedures

Total solids (TS), and volatile solids (VS) were determined by standard methods (Anon 1995). Fibre content was analyzed according to Goering and van Soest (1970). Total nitrogen and reducing sugars were analyzed by the Kjedahl method (Egn *et al.* 1981) and Dubois method (Dubois *et al.* 1956) respectively. Methane and VFA were measured as previously described (Kivaisi & Eliapenda 1995).

Statistical analyses

To evaluate the effect of RCH concentration and alkali pre-treatment of the coffee hulls, two-way analysis of variance tests were performed. The difference between the treatments was considered significant if P was < 0.05.

RESULTS AND DISCUSSION

Effect of RCH concentration

Increasing VFA concentrations and methane production with increasing RCH concentration up to 30 % were observed for both the treated and non-treated substrates (Table 3, Fig. 1). The lowest VFA concentration and methane production were recorded at 5 and 100 % RCH, respectively. These results correspond with the observed decrease in the overall extent of volatile solids (VS) degradation with increasing RCH concentration (Fig. 2). This decrease was most likely due to inhibition by some coffee components including caffeine and brown melanoid pigments that increased with increasing coffee hull concentration in the digestion mixture. Most likely, the coffee based inhibitory components in the digestion mixture at RCH concentrations up to 30% could be tolerated by the microorganisms. This implies that there is a critical level of coffee based inhibitory compounds at which microorganisms can carry out methanogenesis optimally. A number of previous studies applying continuous thermophilic digestion of coffee wastes also reported inhibition of methanogenesis, characterised by accumulation of VFA and accompanied by low gas yields, with prolonged operations (Lane 1983, Fernandez & Foster 1993, Ikbal *et al.* 1994, Dinsdale *et al.* 1996). Although the levels of VFA were well below the inhibitory amounts, signs of a limited methanogenesis process were observed. The changed pattern of molar ratios of the acids with increasing coffee hull concentration above 30 % for the treated substrates whereby propionate and butyrate levels doubled, was characteristic of inhibited methanogenesis.

Table 3. Effect of Robusta Coffee Husks (RCH) concentration and pre-treatment on VFA production[a] form RCH collected from a factory in Bukoba, Tanzania,

RCH conc. (%)	VFA production (mmol/l)		VFA Molar ratios (%)					
	NT-RCH	T-RCH[b]	NT-RCH			T-RCH		
			A	P	B	A	P	B
5	49.97 ± 1.54	61.46 ± 1.32	81	11	8	82	15	3
10	61.49 ± 1.32	82.78 ± 1.48	81	11	8	79	15	6
20	76.10 ± 1.25	95.33 ± 1.27	77	15	7	78	15	7
30	81.18 ± 1.46	129.50 ± 1.67[c]	79	15	6	81	14	6
50	78.34 ± 1.20	106.23 ± 1.36	75	17	8	61	27	12
75	65.62 ± 1.50	92.57 ± 1.50	81	13	6	54	35	11
100	53.18 ± 1.12	100.40 ± 1.78	87	9	4	53	34	13

[a] : mean values ± SD, n = 6

NT-RCH= non-treated coffee hulls; T-RCH= treated coffee hulls

[b] : values are significantly different (P=<0.05) from the non-treated RCH.

[c] : the value is significantly higher (P=<0.05) than the rest of the values

A=acetate; P= propionate; B= butyrate

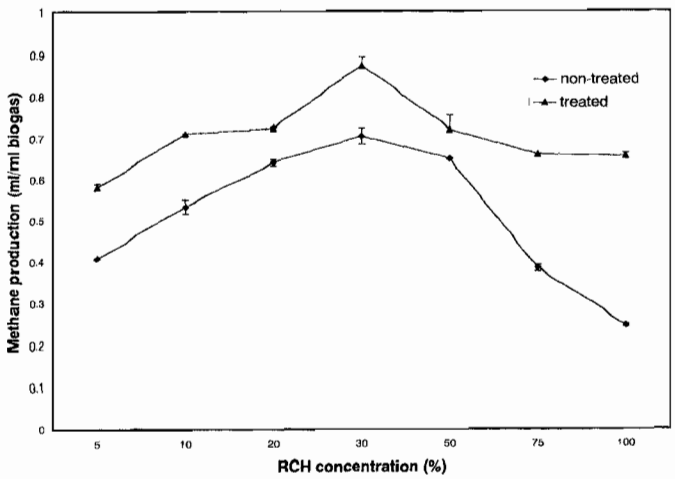

Fig. 1: Effect of concentration of RCH on methane production from the treated and non-treated substrate. (Bars shows SD, n = 6). RCH collected from a factory in Bukoba, Tanzania

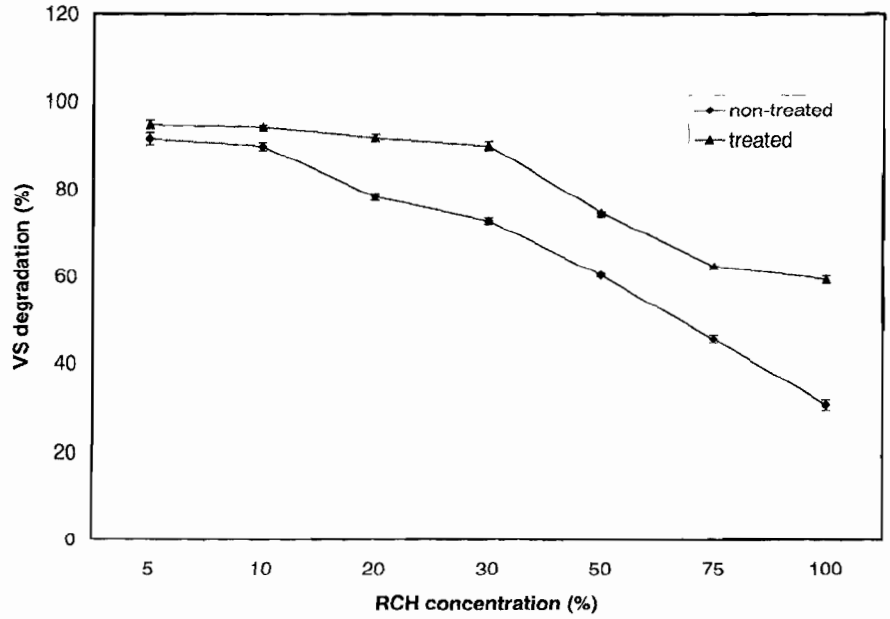

Fig. 2: Effect of concentration of RCH on VS degradation of the treated and non-treated substrate. (Bars show SD, n = 6). RCH collected in Bukoba, Tanzania

Apart from the problem of accumulation of volatile fatty acids, Dinsdale *et al.* (1996), attributed limited methanogenesis to inhibition by high lipid levels in the digesters with coffee grounds containing 26% lipid content. In other studies, high lipid levels also caused digester instability (Hanaki *et al.* 1981, Ahring *et al.* 1991). In this study therefore, the limited conversion of the substrate into methane above 30% coffee hulls concentration could have been partly due to high lipid levels. However, the lipid content of the hulls used in this study was not determined. Since this study was done in batch cultures with small volumes (50 ml), a successful conversion of the coffee hulls into methane at more than 30 % of the substrate and at a higher VS loading is anticipated in a continuous digester.

Effect of pre-treatment

The results on the effects of pre-treatment on the extent of acidification of the substrates and methane production are presented in Table 3 and Fig. 1, respectively. VFA concentrations in incubations with pre-treated substrate were 22-88% higher than those with the non-treated substrate and they were significantly different ($P < 0.05$). Similarly, methane production was significantly higher ($P < 0.05$) in the range of 13- 164 % for the treated

substrate than for the non-treated one. The highest increase was observed at 100 % RCH. The effect of pre-treatment is reflected in the overall conversion of the substrate (Fig. 2) whereby the extent of degradation of the pre-treated substrate (59-94 %) was significantly higher ($P < 0.05$) than that obtained for the non-treated substrate (30-91 %). The effect of pre-treatment on VS degradation was highest at 100 % RCH where an increase of 29 % was measured. The lowest extent of degradation of 30 % was measured for the non-treated substrate at 100 %, and the highest was measured for the pre-treated substrate at 5 %. These results indicate an improved hydrolysis of the degradable components of the pre-treated hulls and hence enhanced methanogenesis. Lignin is known to inhibit enzymatic hydrolysis of cellulose in lignocellulosic materials, a step which has been shown to be rate limiting (Noike *et al.* 1985). Enhanced methanogenesis of lignocellulose by alkali pre-treatment has been reported previously (Pavlostathis & Gosset 1985, Hashimoto 1986, Kivaisi & Eliapenda 1995).

CONCLUSION

It can be concluded that highly lignified coffee hulls can be used for biogas production by applying alkali pre-treatment and co-digestion with cow dung, and there appears to be an optimal coffee waste concentration in the digestion mixture for successful anaerobic conversion to biogas. Approximate amounts of coffee hulls suitable for digestion in continuous and large scale reactors need to be established.

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