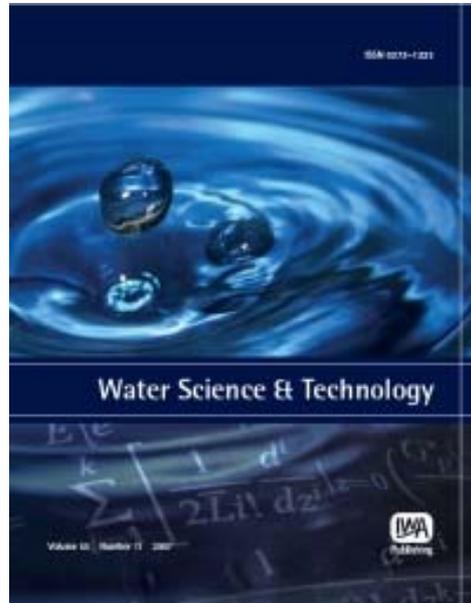


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## Anaerobic treatment of coconut husk liquor for biogas production

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### ABSTRACT

The market for coconut water causes environmental problems as it is one of the major agro-industrial solid wastes in some developing countries. With the aim of reusing the coconut husk, Embrapa developed a system for processing this raw material. During the dewatering stage Coconut Husk Liquor (CHL) is generated with chemical oxygen demand (COD) varying from 60 to 70 g/L due to high concentrations of sugars and tannins. The present study evaluated the feasibility of anaerobic treatment of CHL through Anaerobic Toxicity Assay and the operation of a lab-scale Upflow Anaerobic Sludge Blanket (UASB) reactor. Results showed that CHL can be treated through a UASB reactor operating with an OLR that reaches up to 10 kg/m<sup>3</sup>·d and that is maintained stable during the whole operation. With this operational condition, the removal efficiency was higher than 80% for COD and approximately 78% for total tannins, and biogas production was 20 m<sup>3</sup> of biogas or 130 kWh per m<sup>3</sup> of CHL. Seventy-five percent of the biogas composition was methane and toxicity tests demonstrated that CHL was not toxic to the methanogenic consortia. Conversely, increasing the concentration of CHL leads to increased methanogenic activity.

**Key words** | anaerobic treatment, coconut husk liquor, tannin, toxicity, UASB

### INTRODUCTION

The coconut palm (*Cocos nucifera*) is a tropical tree that is cultivated in nearly 90 different countries (Pires *et al.* 2004). The water of tender coconut, technically called the liquid endosperm, is enjoyed by people in tropical regions of the world, especially in Tropical Asia as well as Central and South America. It was discovered that this water has medicinal characteristics such as the capacity to act as an antioxidant, replenish fluid hydration, provide parenteral nutrition, etc. Despite its benefits, the market for coconut water causes solid waste environmental problems and is one of the major agro-industrial waste generators in some developing countries. For example, in 2007 Brazil had over 57,000 ha of coconut tree plantations which will annually produce about 300 billion fruits and will generate approximately 360,000 tons of husk (Pires *et al.* 2004). The coconut

husk is generally disposed of in landfills whose lifetime is then decreased as a result of the long period required for husk degradation.

With the objective of totally reusing the coconut husk and reducing the environmental impact of its disposal, Embrapa (the Brazilian Agricultural Research Corporation) supported the development of a technology for processing the husk to obtain powder and fibres that have a high aggregate value. As a result, a full-scale coconut husk processing unit was built in Fortaleza, Brazil in 2005. This unit is capable of producing 30 tons of powder and fibres per day. However this new industry generates a high strength wastewater called Coconut Husk Liquor (CHL) at a rate of 0.67 m<sup>3</sup> per ton of powder and fibres. The characterization of this effluent resulted in a chemical

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oxygen demand (COD) in the range of 60 to 70 g/L, an electrical conductivity of approximately 9 mS/cm, and total tannins of approximately 6 g/L (Teixeira 2007). Despite the high concentration of organic matter, which is desirable for anaerobic treatment systems, the primary concern is the tannin content which can inhibit the metabolism of microorganisms involved in the degradation process (Scalbert 1991; Bhat *et al.* 1998). Nevertheless, several fungi, yeast, as well as bacteria are resistant and can grow in medium containing these substances, demonstrating that biological degradation of tannin is feasible (Bhat *et al.* 1998; López-Fiuza *et al.* 2003).

Anaerobic degradation of gallotannic acid, a hydrolysable tannin, was first investigated by Field & Lettinga (1987). The authors observed that this tannin is highly toxic to methanogenic *archaea*. Vidal *et al.* (2001) tested the anaerobic degradation of kraft mill effluent using a lab-scale Upflow Anaerobic Sludge Blanket (UASB) reactor that was operated with influent concentrations varying from 800 to 1,900 mgCOD/L and tannins in the range of 44 to 64 mg/L. The results show that a mere 29% of tannin and lignin were removed. This was most likely due to the inhibition of anaerobic metabolism. However, other researchers reported the feasibility of anaerobic degradation of tannin. López-Fiuza *et al.* (2003) investigated anaerobic degradation of natural tannin extract using three lab-scale UASB reactors. They used three different synthetic influents comprised of glucose as the primary carbon source, nutrients, and tannins. Each influent contained a distinct tannin (quebracho, wattle and chestnut), at concentrations varying from 100 to 1,000 mg/L. Hydrolysable tannin (chestnut) removal efficiency achieved 90% at the beginning of the experiments and decreased to 60% when the maximum concentration of tannin extract was used. Condensed tannin was more recalcitrant and only 20% of this compound was degraded when 1,000 mg/L was imposed. Vijayaraghavan & Ramanujam (1999) investigated anaerobic degradation of tanning industry effluent using two lab-scale filters in series. The reactors were operated at a hydraulic retention time (HRT) which varied from 24 to 60 h and influent COD ranging from 1.5 to 13.5 g/L. They found that at an influent tannin concentration of 1.0 g/L and an HRT of 60 h, the system could remove approximately 80% of the compound. However, decreasing HRT to

24 h and increasing tanning concentration to 1.5 g/L, tannin removal efficiency dropped to approximately 30%.

The aim of this study is to evaluate the feasibility of anaerobic treatment of the tender coconut husk processing unit's wastewater (CHL), which possesses high tannin content, via anaerobic toxicity assays (ATA) and the operation of a lab-scale UASB reactor.

## MATERIALS AND METHODS

### Coconut husk liquor

CHL is a result of dewatering (by compress cylinders) following the trituration of coconut husks. The main characteristics of this effluent are: BOD (41.2 g/L), COD ( $63.5 \pm 3.0$  g/L), total tannins ( $5.95 \pm 0.51$  g/L), total sugars (45.1 g/L), total alkalinity (1.01 g/L), ammonia (0.75 g/L), nitrite (0.04 g/L), nitrate (0.07 g/L), total phosphorus (0.13 g/L), total solids ( $65.3 \pm 0.6$  g/L), total inorganic solids ( $6.2 \pm 0.2$  g/L), total volatile solids ( $59.1 \pm 0.5$  g/L), pH ( $4.9 \pm 0.1$ ), and conductivity (8.75 mS/cm). Values after  $\pm$  represent confidence interval for confidence level of 95%. Total tannin were determined as total phenols.

### Sludge methanogenic activity and anaerobic toxicity assay

The toxicity tests used in this investigation are a variation of specific methanogenic activity (SMA) tests which follows the procedure according to Jawed & Tare (1999). The main difference between toxicity and SMA tests is the replacement of part (or all) of volatile fatty acids (VFA) used as substrate by the substance to be tested (CHL for this experiment). In short, toxicity tests were carried out in 0.6 litre serum bottles sealed with a rubber septum and kept in place by a screw cap. Sludge was added to each bottle, then the liquid volume was completed to 0.4 litre with a solution comprised of distilled water, pH buffer, nutrients, trace elements and the substrate (VFA and/or CHL). The final concentrations of sludge (in terms of total solids) and substrate were 5 gTS/L and 2.5 gCOD/L respectively. Sodium acetate, sodium propionate, and sodium butyrate

were used as a source of VFA, in a rate of 1:0.61:0.16 in terms of COD. Both nutrients and trace elements were added to the bottles to prevent deficiency during the test. The following nutrients (g/L) were added:  $\text{NH}_4\text{Cl}$  (0.28),  $\text{K}_2\text{HPO}_4$  (0.25),  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$  (0.10),  $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$  (0.01), and  $\text{CaCO}_3$  (0.60). For trace elements (1 mL/L) the following substances (mg/L) were added:  $\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$  (2000),  $\text{H}_3\text{BO}_3$  (50),  $\text{ZnCl}_2$  (50),  $\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$  (38),  $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$  (500),  $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$  (50),  $\text{AlCl}_3 \cdot 6\text{H}_2\text{O}$  (90), and  $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$  (2000). The activity tests were performed at a temperature of  $35 \pm 2^\circ\text{C}$ . Total volatile solids (VS) content of the sludge was determined to calculate the specific activity. During the test, methane production was monitored daily using a NaOH solution (5% w/w) displacement system (a type of Mariotte bottle). After 75% of the imposed COD load was recovered by  $\text{CH}_4$  production, a second feed of pH buffer, nutrients, trace elements, and VFA was applied to detect the maximum methanogenic activity. The percentages of CHL that replaced VFA were 0% (control), 25%, 50%, 75%, and 100%, which corresponded to 0, 80.2, 160.4, 240.6, and 320.8 mg/L of total tannins. All experiments were performed in duplicate.

### Lab-scale UASB reactor

The experimental investigation was carried out using a pilot-scale UASB reactor, built with PVC tubes, with a working volume of 16.8 L, height of 1.95 m, and internal diameter of 0.1 m. It had a modified gas-solid-liquid separator as described by Cavalcanti *et al.* (1999), and was equipped with dosing pumps, gas samplers, and eight sludge collection points. Methane production was monitored using

a gas meter (Ritter, model MGC-1). A very slow stirrer (1 rpm) was installed in the reactor to avoid channelling and “piston” formation (rising sludge due to entrapped biogas in the sludge layer). The reactor was first inoculated with an anaerobic sludge discharged from a brewery UASB reactor. Next, it was fed for 33 days with sucrose and nutrients (the same as used in SMA and toxicity tests). Dilution was done with tap water. Following the initial period, the influent was changed by replacing sucrose for an increasing concentration of CHL from 10% until the influent completely substituted the wastewater. The next step was to incrementally raise organic loading rate (OLR) from 2.2 to 10.0 kgCOD/m<sup>3</sup>·day. Operational parameters are presented in Table 1. Sodium bicarbonate was used as a buffer at the rate of 1 gram  $\text{NaHCO}_3$  per gram of influent COD. Nutrients and trace elements were only used during Phases I, II, and III.

### Analytical methods

All physical-chemical analyses were performed as recommended by Standard Methods (2005). Raw samples were used for Total COD. The micro-COD method was used for all COD analysis. Total VFA followed the procedure described in Buchauer (1998). Total tannins followed procedures described in Lowry *et al.* (1947) which determine total phenolic compounds using tannic acid as a standard. Biogas composition was determined by using a Varian CP-4900 micro gas chromatograph equipped with a thermal conductivity detector. Methane and carbon dioxide were analyzed using a Propac Q column with helium as carrier gas at 60°C.

**Table 1** | Operational parameters

Phase	Duration (days)	Flow Rate (L/day)			Total	COD* (g/L)	Total tannins* (g/L)	OLR* (kgCOD/m <sup>3</sup> ·d)	HRT (h)
		Sucrose solution	CHL	Tap water					
I	33	25.2	–	–	25.2	1.3–1.5	–	1.5–2.2	16.1
II	40	25.2–24.6	0.0–0.6	–	25.2	1.5	0.0–0.1	2.2	16.1
III	104	–	0.6–2.8	24.6–22.4	25.2	1.5–6.7	0.2–0.9	2.2–10.0	16.1
IV	12	–	2.8	9.7	12.5	13.5	1.8	10.0	32.3
V	19	–	2.8	3.2	6.0	28.0	3.7	10.0	67.2
VI	14	–	2.8	–	2.8	60.0	8.0	10.0	144.0

\*Theoretical values based on the assumption that CHL has 60 g/L of COD and 8 g/L of total tannins.

## RESULTS AND DISCUSSION

### Anaerobic toxicity assays

Toxicity tests were performed after 33 days of operation when the reactor achieved the pseudo steady state and prior to operation with CHL feeding. The results demonstrated that CHL was not toxic to the methanogenic consortia (even considering the fact that using 100% of CHL implies approximately 250 mg/L of total tannins). It was expected that increasing the CHL concentration would lead to a decreasing SMA as tannins form stable complexes with protein, bound cell membranes, and cause substrate and metal ion deprivation (Bhat *et al.* 1998). However, it was observed that values of methanogenic activity increased from 0.12 to 0.20 kgDQO/kgVS·d as CHL concentration increased from 0% to 100% respectively. This may be due to a high concentration of glucose in the wastewater which may have overcome the deleterious effect of the tannins. This phenomenon was also observed by Hwang & Cheng (1991) and Subramanyam & Mishra (2008) who found a significant increase on values of AME when glucose was used as a co-substrate for the degradation of recalcitrant phenols (resorcinol and catechol). According to Hwang & Cheng (1991), this phenomenon occurs due to proton transport of hydrogen through NADH or NADPH formation that is promoted by acidogenic reaction of glucose in an anaerobic environment.

### Performance of the UASB reactor treating CHL

Figure 1 presents the reactor response obtained throughout the entire research. The lab-scale UASB reactor was operated with increased OLR which reached up to 10.0 kgCOD/m<sup>3</sup>·d and managed to maintain its stability during the whole period. COD removal efficiency and gas production maintained relatively stable and showed only slight fluctuations during the first days after the operational parameter was changed. With this operational condition, the reactor produced approximately 50 L/d of biogas which consisted of 75% of methane in its composition. This efficiency and biogas production was used to verify the COD mass balance and is in accordance

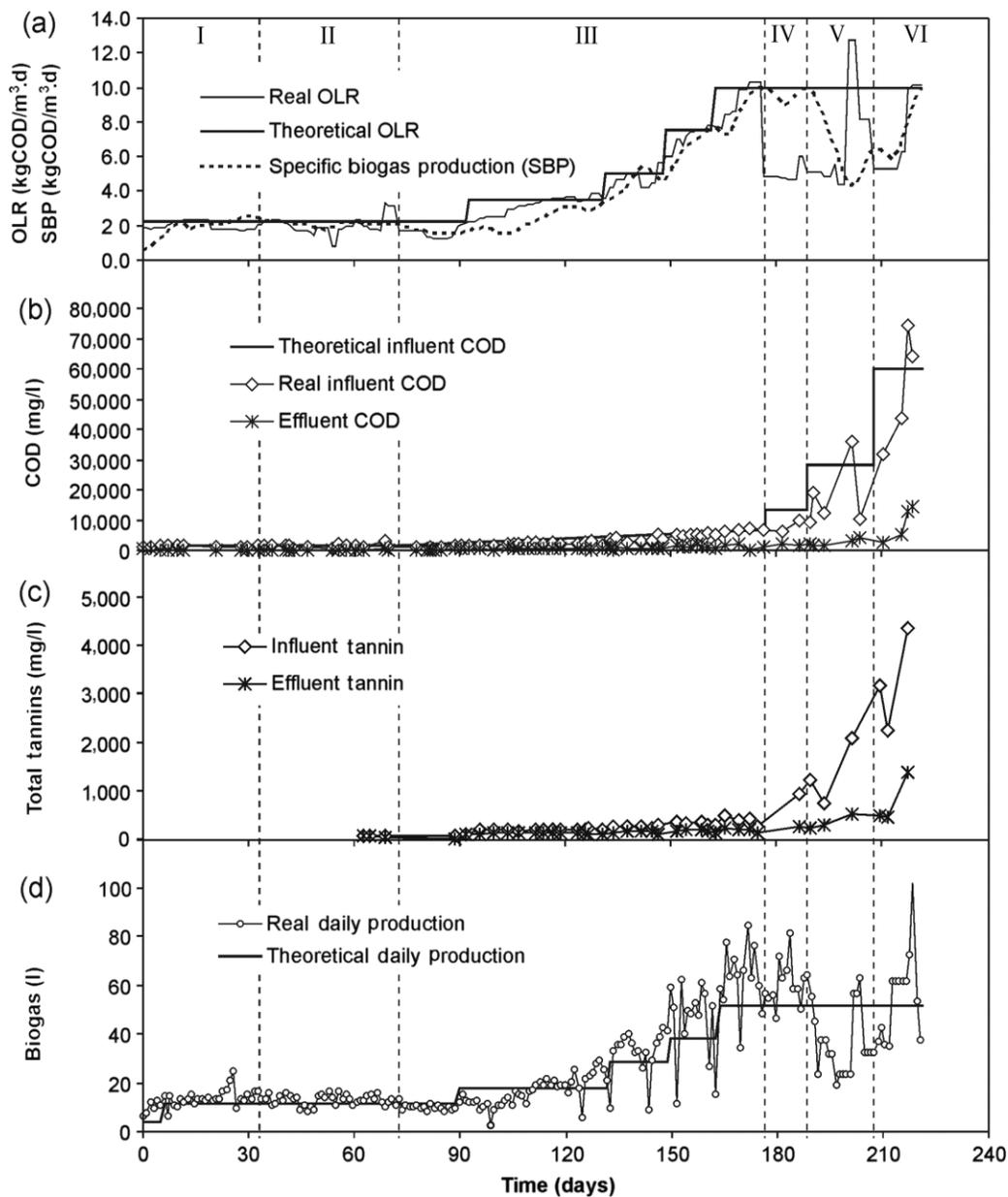
with van Haandel & Lettinga's model (1994) represented in Equation (1).

$$Q_{\text{Inf}} \times \text{COD}_{\text{Inf}} = (Q_{\text{Eff}} \times \text{COD}_{\text{Eff}}) + (Y \times Q_{\text{Inf}} \times \text{COD}_{\text{Inf}}) + \frac{Q_{\text{Biogas}} \times \% \text{CH}_4}{\text{COD}_{\text{CH}_4}^{\text{CNTPT}} \times (273 + T) / (273 \times \text{Atm})} \quad (1)$$

where:  $Q_{\text{Inf}}$  = Influent flow rate (m<sup>3</sup>/d);  $\text{COD}_{\text{Inf}}$  = Influent COD concentration;  $Y$  = cellular yield (adopted 0.21 kgCOD/kgCOD imposed);  $Q_{\text{Biogas}}$  = Biogas flow rate;  $\% \text{CH}_4$  = Methane concentration in biogas;  $\text{COD}_{\text{CH}_4}^{\text{NPT}}$  = Methane in terms of COD under Normal Temperature and Pressure (0.35 L CH<sub>4</sub>/gCOD);  $T$  = temperature in Celsius; and  $\text{Atm}$  = atmospheric pressure.

A tender coconut husk processing unit, similar to the one created by Embrapa, produces about 15 m<sup>3</sup> CHL/d. Based on the results of this research and the model in Equation (1), it is estimated that 290 m<sup>3</sup>/d of biogas can be produced in a 97.5 m<sup>3</sup> UASB reactor that operates with OLR of 10 kgCOD/m<sup>3</sup>·d and HRT of 156 h. The calorific power of the biogas is 5,850 Kcal/m<sup>3</sup>; therefore, 290 m<sup>3</sup>/d of biogas is enough to theoretically generate 1,979 kWh. Considering that the overall efficiency of an electrical generator powered by a biogas engine is approximately 25%, then anaerobic reactors can supply the coconut processing unit with 62 kW for eight hours per day, which is enough to run all of the electrical motors.

The total tannins removal efficiency increased steadily during the operation and achieved 78% without any long-term inhibition effect. This was confirmed by eight SMA tests performed throughout the operational period using VFA as a substrate (ranging from 0.11 to 0.16 kgCOD/kgVS·d). Interestingly enough, raising OLR, and consequently the tannin loading rate, resulted in sludge with higher SMA. When CHL was used as a substrate for the activity tests, SMA reached up to 0.27 kgCOD/kgVS·d by the end of the operational period. These are all indicators that, despite high tannin concentration (approximately 6 g/L), the reactor could easily adapt to CHL even if high OLR is imposed. This phenomenon is the opposite to what was expected, as results of several researchers shows that a high concentration of tannins can cause deleterious effect on anaerobic



**Figure 1** | Reactor behaviour during the operational period. Vertical dashed lines divide operational phases.

sludge (Field & Lettinga 1987; Vidal *et al.* 2001), even when using glucose as a co-substrate (López-Fiuza *et al.* 2003). The high tannins removal efficiency can most likely be attributed to the very high concentration of glucose in the influent (approximately 40 g/L), which may have acted as an electron donor during anaerobic degradation of recalcitrant compounds (Hwang & Cheng 1991). Furthermore, easily degradable glucose may have induced cellular

growth at a higher rate than decay due to the harmful effects of tannins.

## CONCLUSIONS

- CHL with high tannin content is not toxic to methanogenic consortia. On the contrary, it can

increase the methanogenic activity due to high glucose concentration;

- A UASB reactor, operated at an OLR of 10.0 kgCOD/m<sup>3</sup>·d and an HRT of 156 h, can remove 80% of the COD and 78% of total tannins from CHL and can produce 19.4 m<sup>3</sup> of biogas per cubic meter of CHL per day;
- The sludge presented SMA ranging from 0.11 to 0.16 kgCOD/kgVS·d throughout the operational period. This indicates that there is no long-term inhibition due to CHL.

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