



## Research paper

# Effect of increasing proportions of lignocellulosic cosubstrate on the single-phase and two-phase digestion of readily biodegradable substrate



Rangaraj Ganesh<sup>a</sup>, Michel Torrijos<sup>a,\*</sup>, Philippe Sousbie<sup>a</sup>, Aurelien Lugardon<sup>b</sup>,  
Jean Philippe Steyer<sup>a</sup>, Jean Philippe Delgenes<sup>a</sup>

<sup>a</sup> INRA, UR50, Laboratoire de Biotechnologie de l'Environnement, Avenue des Etangs, Narbonne, F-11100, France

<sup>b</sup> Naskeo Environment, 52 rue Paul Vaillant Couturier, F-92240 Malakoff, France

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## ABSTRACT

The influence of different proportions of lignocellulosic substrate (cow manure with straw, CM) on the single-phase (conventional reactor) and two-phase (acidification/methanation with solids and liquid recirculation) digestion of a readily biodegradable substrate (fruit and vegetable waste, FVW) was investigated in order to determine the optimum cosubstrate ratio and the process best suited for codigestion. Both processes were fed initially with FVW, followed by FVW and CM at 80%:20% and 60%:40% (on volatile solids, VS basis) during an experiment run over eleven months. For the single-phase process, energy yield and VS destruction decreased by 11% and 9% with the 80%:20% FVW and CM ratio and by 16% and 17% with the 60%:40% feed ratio when compared to 100% FVW feed. For the two-phase process, energy yield and VS destruction decreased by 21% and 14% with 80%:20% feed ratio and by 48% and 33% with 60%:40% feed ratio compared to 100% FVW. Substrate solubilization in the acidification reactor was very efficient for all the feed proportions but it resulted in compounds other than volatile fatty acid (non-VFA COD) which were not easily amenable to methane generation. This led to a lower energy yield per kg of VS fed in the two-phase process compared to the single-phase process for the respective waste combination. For single-phase digestion, both 80%:20% and 60%:40% ratios were effective co-substrate combinations due to their higher energy yield. The two-phase process can be used for these ratios if higher VS reduction and a higher loading rate are the objectives.

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## 1. Introduction

Anaerobic digestion of solid waste has become widespread in several European countries over recent decades, especially in the agricultural sector where it resulted in accrued economic benefits and an additional source of income for farmers [1,2]. In the agricultural sector, livestock manure represents a substantial source of readily-collectable biomass, however, improper disposal causes odour problems, release of pathogens, contamination of surface and groundwater as well as methane and ammonia emissions [3]. The anaerobic digestion of manure decreases pathogenic bacteria and enhances the agronomical value of the residue [4,5] thereby reducing the negative environmental impact it can have.

Straw, along with rice husk and saw dust, is the bedding material normally used to absorb manure and to eliminate the need for frequent cleaning of the sheds. Solid manure is removed and the soiled bedding material is disposed as spent straw [6,7] or disposed as such. Proper disposal of the soiled bedding material is imperative as it can cause environmental and hygiene concerns. While the solid manure in the bedding material can be digested anaerobically, straw degradation remains a challenge as it is composed of lignocellulosic compounds which affect the overall kinetics of biogas formation [8].

The carbon to nitrogen (C/N) ratio of cow manure is low for anaerobic digestion, hence addition of a readily biodegradable cosubstrate allows for a better nutrient balance and enhances bioreactor performance [3,9]. Among the types of biodegradable organic waste, fruit and vegetable waste (FVW) is an interesting option as such residues undergo rapid acidification at high loading rates [10]. Furthermore, the nitrogen and phosphorus content of

\* Corresponding author.

E-mail address: [michel.torrijos@supagro.inra.fr](mailto:michel.torrijos@supagro.inra.fr) (M. Torrijos).

FVW is often low and thus the addition of a cosubstrate can improve the nutrient balance [11]. The complementary characteristics of FVW and manure offer an advantage in utilizing such waste for co-digestion and for enhancing biogas productivity [12]. The codigestion of FVW and manure has been little studied [12–14] and these studies have focused on single-phase digestion.

Among the various aspects of anaerobic digestion (AD) research, shortening the digestion time with enhanced process efficiency has been a central concern [15]. Other basic requirements of anaerobic digester design include maximizing volatile solids degradation, allowing for a continuously high and sustainable organic loading rate (OLR) and thorough mixing for effective transfer of organics and gas bubble release [3]. Single-stage anaerobic systems in which all three phases of hydrolysis, acetogenesis and methanogenesis take place simultaneously in a single reactor remains the preferred option [16] for majority of waste. However, operation of systems at high OLRs and for waste with a high biodegradable organic content (e.g. fruit and vegetable waste) becomes difficult; as such waste undergoes rapid acidification, decreasing the pH if the system is not adequately buffered and inhibiting methanogenic activity [10]. Two-phase systems on the other hand have the advantage of buffering the OLR in the first stage, allowing a more constant feeding rate to the methanogenic second stage [17]. A two-stage system limits the imbalance between acidogenesis and methanogenesis while bringing robustness to the digestion process [18].

Studies to optimize the proportions of cosubstrate mixtures along with the choice of the codigestion process (whether single- or two-phase) need to be carried out through experimentation. Hence, the present investigation was undertaken to compare the single-phase and two-phase anaerobic digestion of different proportions of easily degradable substrate (FVW) and slowly degradable lignocellulosic cosubstrate (cow manure with straw). A conventional reactor was used for the single-phase process while the two-phase process comprised of acidification and methanation reactors with the recirculation of solids and liquid. Methane yield, VS destruction, reactor stability and energy yield with the different feed proportions were compared for the two processes.

## 2. Materials and methods

### 2.1. Feed substrates

Fruit and vegetable waste (FVW) and cow manure with straw (CM) were used as the feed substrates for the experiments. CM was crushed to a size of approximately 1 cm in a Blik BB 230 crusher equipped with stainless steel rotating blades and then stored at  $-20\text{ }^{\circ}\text{C}$ . For the preparation of FVW feed, equal quantities of apple, banana, carrot, potato and lettuce were ground to approximately 1 cm size in the crusher, mixed thoroughly and then stored at  $-20\text{ }^{\circ}\text{C}$ . The composition of the FVW and CM are shown in Table 1. Batches of the mixture of the feed substrates were pre-weighed and stored, then brought to room temperature and fed into the reactors. The average total solids (TS) and volatile solids/total solids (VS/TS) with the 80% FVW - 20% CM feed mixture were

14.3% and 93.2%, respectively, and with the 60%:40% feed mixture, 16.3% and 92.1%.

### 2.2. Reactor set-up

Single-phase and two-phase configurations were studied. In the single-phase configuration, one reactor referred to as a single-phase reactor (SPR) was used and operated in conventional mode. In the two-phase configuration, two reactors were operated in series: the first was the acidification reactor, referred to as the two-phase acidification reactor (TPAR); the second was the methanogenic reactor, referred to as the two-phase methanogenic reactor (TPMR).

The same type of reactor was used for SPR and TPAR, the schematic diagram of which is shown in Fig. 1. These stainless steel reactors were double-walled and maintained at  $35\text{ }^{\circ}\text{C}$  by a regulated water bath. The total volume of the reactors was 15 L with an effective sludge weight of 10 kg. Feeding and draining were carried out manually by opening the top cover of the reactors. The reactors were equipped with a paddle-shaped stirrer powered by a 1 HP motor. Mixing times were programmed through a process controller. For the TPMR, a double-walled glass reactor of 6 L volume was used and was maintained in mesophilic conditions at  $35\text{ }^{\circ}\text{C}$ . The biogas produced passed first through a moisture trap then through a milligas meter fitted with a 4–20 mA output (MGC-1 gas flow meter, Ritter). The data was recorded and displayed online. Software (Modular SPC) developed at the INRA laboratory was used to log the data. The SPR and TPAR were weighed on scales prior to the addition of inoculum and their weights noted. The SPR and TPAR were inoculated with 4.0 kg of granular sludge (after breaking up the granules in glass reactors by continuous mixing for several days) and made up to 10 kg by the addition of treated effluent from a pilot plant in the laboratory. In both the reactors, the initial total suspended solids (TSS) and volatile suspended solids (VSS) were, respectively, 35 g/L and 30 g/L. During reactor operation, the total weight of the reactor was measured and sludge withdrawal adjusted accordingly to maintain the reactor sludge weight constant at 10 kg. This procedure was followed in order to minimize the error in calculations of solids destruction.

The TPMR was inoculated with 2 kg of the broken granular sludge (the same as that used for SPR and TPAR), and made up to 6 L with treated effluent from the pilot plant. The initial TSS and VSS were 19 g/L and 16 g/L respectively. The reactor was run in a semi-continuous mode with continuous mixing ensured by magnetic stirring. The gas produced was measured and acquired in the same way as in the SPR and TPAR.

### 2.3. Sampling and analysis

The parameters pH, solids, COD and alkalinity were measured in accordance with Standard Methods [19]. The soluble, cellulose, hemicellulose and lignin-like fractions in the feed substrates were analysed according to Van Soest procedure [20] using the Fiberbag system (Gerhardt, Germany). This procedure is based on sequential extraction with neutral and acid detergents, followed by strong acid extraction. The soluble content was obtained by extraction with a neutral detergent solution; the hemicellulose-like compounds were extracted by an acid detergent solution; and the cellulose-like compounds were extracted with sulphuric acid. The remaining fraction corresponds to the lignin content. Volatile fatty acid (VFA) concentration was measured using a gas chromatograph (GC-8000, Fisons Instruments) and biogas composition using a gas chromatograph (Shimadzu GC-8A) as per the procedures described in Ganesh et al., 2013 [21]. Drain samples were analysed for pH, total alkalinity, soluble COD, VFA and total and volatile solids [21].

**Table 1**  
Composition of fruit and vegetable waste (FVW) and cow manure with straw (CM).

Parameters	FVW	CM
TS (%)	12.7	26.5
VS (%)	11.9	22.9
VS/TS (%)	94.1	86.5
Soluble fraction (%)	75.9	19.8
Cellulose fraction (%)	3.9	27.5
Hemicellulose fraction (%)	19.2	49.0
Lignin (%)	1.0	3.7

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