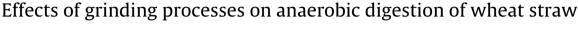
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# Industrial Crops and Products

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# ABSTRACT

Lignocellulosic biomass represents an important part of the agricultural wastes that can be transformed into a renewable energy. The biodegradability and biodegradation kinetics of ultra-finely ground wheat straw was studied under mesophilic anaerobic conditions. Biological methane potential (BMP) tests and batch reactors were performed on samples resulting from successive grinding processes (759–48  $\mu$ m). The main conclusion is that micronization do not improve methane yield but has a positive effect on the biodegradation kinetics. In addition, the results presented here showed for the first time that no significant increase of kinetics was observed below a size threshold value around 200  $\mu$ m. This can be explained by the modifications in the lignocellulosic network from 759 to 200  $\mu$ m but not below 200  $\mu$ m. Therefore, micronization, increasing significantly the kinetics of anaerobic digestion, can decrease the retention time or the size of the digesters to treat the same quantity of waste.

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# 1. Introduction

Because of earth global warming and the reduction of fossil fuels resources, the use of renewable energy sources is becoming increasingly necessary. The conversion of biomass into energy can be achieved in many of ways, for example by producing fuel thanks to various technologies. This study explores the potential of anaerobic digestion (AD) of lignocellulosic biomass. The AD can process biomass and produces a gas rich in methane and digestates rich in mineral elements (Mata Alvarez et al., 2000). Nowadays, processing lignocellulosic biomass thanks to AD has become of much interest. Researchers explore the possibility of transforming some non-food competitive agricultural crops in energy.

However, the hydrolysis step of complex polymeric substances constitutes the rate limiting step (Vavilin et al., 1996), and therefore, it has engaged the most attention from researchers. In order to increase the hydrolysis rate and optimize the biological trans-

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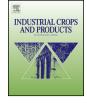
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formations, physic and/or chemical pretreatments need to be implemented, such as mechanical (grinding), physical (thermal) (Maroušek, 2013), chemical (acid or alkali) (Nieves et al., 2011), or a combination of them (Barakat et al., 2013). These pretreatments aimed at increasing the substrate surface area, decreasing crystallinity and/or disrupting lignin-hemicelluloses cross-linked network. Therefore, biodegradable compounds become accessible to enzymes and microorganisms. Mechanical fractionation is a necessary step in lignocellulosic bioconversion to (i) decrease particle size and increase total accessible specific surface area (ii) increase pore size of particles and the number of contact points for interparticle bonding in the compaction process and eventually (iii) decrease cellulose crystallinity. All these parameters improve the digestibility and the conversion of saccharides during enzymatic hydrolysis (Khullar et al., 2013; Silva Ghizzi Damasceno et al., 2012; Wang et al., 2013) and bioconversion (Barakat et al., 2013; Galbe and Zacchi, 2012; Lindmark et al., 2012; Palmowski and Müller, 2000a; Taherzadeh and Karimi, 2008). Mechanical fractionation can represent an interesting and efficient pretreatment or raw lignocelllulosic matter for bioconversions without water addition and effluents production (Barakat et al., 2013).

Some authors investigated the effect of reducing particle size and solubilizing food wastes. Izumi et al. tested substrates bead milled (food wastes) in batch process experiments at mesophilic temperature (37 °C) for 16 days (Izumi et al., 2010). The reduction in diameter from 0.843 to 0.391 mm involved 40% of total





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*Abbreviations:* AD, anaerobic digestion; d<sub>med</sub>, median diameter; COD, chemical oxygen demand; VFA, volatile fatty acids.

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chemical oxygen demand (COD) solubilization. In batch experiments, particle size reduction from 0.888 to 0.718 mm improved the methane production by 28%. However, excessive particle reduction resulted in volatile fatty acids (VFA) accumulation, which composition depended of particle size reduction. Acetic acid was accumulating when particle size was reduced, while the concentration in butyric acid was decreasing for particle size inferior to 0.5 mm. No significant effect was observed on propionic acid. The maximum substrate utilization constant was inversely proportional to particle size (reduction from 2.14 to 1.02 mm) in anaerobic thermophilic digestion in food wastes (Kim et al., 2000). Sharma et al. reported the effects of particle size on agricultural and forest residues on biogas generation through anaerobic digestion in batch digesters at 37 °C (Sharma et al., 1988). Out of five particle sizes (0.088, 0.40, 1.0, 6.0, and 30.0 mm), the maximum quantity of biogas was produced with the smallest particles. Palmowski et al. (2000) investigated the effect of comminution (to millimeters or hundreds of micrometers) of several organic solids on the particles biodegradation (Palmowski and Müller, 2000b). The gas production was increased up to 20% during AD in batch digesters, depending on the state of the ground sample and on the characteristics of the substrate. The maximum gas production increase was observed in the case of high fiber content substrates. The generation of active surface area and the reduction of material structure implied that cell compounds from areas, previously difficult to reach for microorganisms and enzymes, were released and therefore became accessible. No improvement for more biodegradable substrates such as a mixture of apples, carrots and potatoes, meat was observed. Considering small particles (specific surface greater than  $20 \text{ m}^2/\text{kg}$ ), a small effect was observed on biogas production, but a significant effect was observed in the case of big particles (specific surface ranged from 3 to  $20 \text{ m}^2/\text{kg}$ ). An increase in gas production rate led to a decrease in digestion time, which allowed the size of the reactor to be reduced without any losses in gas production. Mshandete et al. (2006) studied the degradation and biogas production potential of sisal fiber with fiber sizes ranging from 2 to 100 mm, at 33 °C (Mshandete et al., 2006). The total fiber degradation increased from 31% to 70% for the 2 mm fibers compared to untreated sisal fibers. Moreover, the methane yield was inversely proportional to particle size with an increase of 23% when the fibers were cut at 2 mm size (0.22 m<sup>3</sup>CH<sub>4</sub>/kg organic matter for  $2\,\text{mm}$ , compared to  $0.18\,\text{m}^3\,\text{CH}_4/\text{kg}$  organic matter for untreated fibers). Hu et al. presented the influence of cellulose particle size  $(50-100 \,\mu\text{m})$  on anaerobic degradation by rumen microbes. Particle size reduction resulted in a decrease of methane production and an increase of soluble products (particularly VFA) (Hu et al., 2005). The majors VFA were acetate and propionate. The maximum substrate utilization rate coefficient doubled with a decrease in the average particle size from 2.14 to 1.02 mm. The influence of grinding pretreatment of cardboard materials was studied on various aspects of the AD (biodegradation rate and methane production) (Pommier et al., 2010). The authors observed no significant differences, neither on the biogas production quantity, nor on the biogas production rate.

These studies demonstrated that the particles size reduction generally allows an increase of the biogas potential and/or the digestion rate. Reducing particle size can also involve VFA accumulation, and therefore, may inhibit methanogens generate a decrease of AD performances. Most of these articles investigated the effect of particle size reduction to some hundreds micrometers. Moreover, the conclusions could be opposite depending on either the size reduction or the type of substrate.

Therefore, the hydrolysis limitation due to substrate specific surface area and accessibility of organic matter to microorganisms is still disputed and needs to be clearly demonstrated. The novelty of our study was the investigation of the effect of lignocellulosic biomass micronization on the AD performances (Silva Ghizzi Damasceno et al., 2012, 2011). Indeed, most of the previous articles dealt with coarse milling, with a minimum particle size around  $100-200 \,\mu$ m. Here, the present work aimed at determining if a significant variation of the median diameter of particles (more than one order of magnitude), obtained by micronization, could increased the maximum biogas production value or/and the kinetics of the AD. This study focuses on one type of mechanical grinding (centrifugal grinding) in order to focus on the influence of the particle size reduction on the AD.

# 2. Material and methods

Wheat straw (*Triticum aestivum* cv. Apache) was grown in Aveyron (FR) and harvested in 2007. The size reduction process was detailed and presented in Silva Ghizzi Damasceno et al. (2011) and constituted 5 steps. Wheat straw was first comminuted using a cutting mill (Retsch SM2000, Haan, Germany) equipped with a 2.0 mm sieve followed by 4 successive steps of centrifugal grinding (Retsch ZM200, Haan, Germany) using 1.0, 0.5, 0.25 and 0.12 mm sieves.

### 2.1. Granulometry and specific surface analysis

Particle size distributions were determined from  $0.02 \,\mu\text{m}$  to 2000  $\mu\text{m}$  using a laser diffraction granulometer Mastersizer 2000 (Malvern Instruments Ltd., United Kingdom) in ethanol suspensions in duplicates at least. The wheat straw samples were characterized as their median diameter ( $d_{med}$ ) obtained from the particle size distributions. For knowing their median diameter and assuming that particles are spherical, the surface area could be estimated.

Surface area = 
$$4 \times \text{Pi} \times (d_{\text{med}}/2)^2$$
 (1)

Bulk density was determined as the tapped volume of 1 g of powder in a graduated cylinder.

#### 2.2. Biochemical composition of the wheat straw

The dry matter and organic matter (DM and OM) were measured according to standard APHA methods (1992) by drying the biomass at  $105 \degree C (24 h)$  followed by incineration at  $550 \degree C (2 h)$ .

The cellulose and hemicellulose contents of each class were determined by gas chromatography (GC) after a two-step sulfuric acid hydrolysis ( $36N H_2SO_4$  for  $30 \min$  at  $25 \circ$ C then  $2N H_2SO_4$  for 2 h at  $100 \circ$ C) and derivatisation as alditol acetates (Blakeney et al., 1983). The alditol acetates obtained were injected on a DB 225 capillary column (J&W Scientific, Folsom, CA), using allose as the internal standard. Cellulose content was calculated as the sum of anhydro-glucose content. Hemicelluloses content was calculated as the sum of anhydro-arabinose, anhydro-xylose, anhydromannose and anhydro-galactose contents.

Klason lignin was determined as the insoluble residue remaining after a two-step sulfuric acid hydrolysis of cell wall polysaccharides (Monties, 1984). Briefly, 200 mg of sample was suspended in 2 mL of 72%  $H_2SO_4$  for 2 h at 20 °C. These acidic suspensions were then diluted to 5% with deionized water and boiled for 3 h then filtered. The remaining residues were dried at 105 °C and then incinerated at 550 °C for 6 h.

The protein content was determined by the KJELDAHL method (total nitrogen  $\times$  6.3).

#### 2.3. Structural analysis

The Fourier transform infra-red (FTIR)-spectroscopy was performed on 2 mg of dry sample carefully mixed with 100 mg Download English Version:

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