



# Kinetic parameter estimation model for anaerobic co-digestion of waste activated sludge and microalgae



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## HIGHLIGHTS

- Kinetic parameter estimation models were proposed for anaerobic co-digestion.
- The models considered co-substrate ratios and single substrate kinetic parameters.
- The models were used to determine the first-order kinetic and Monod coefficients.
- The models could estimate kinetic parameters for the microalgae-WAS co-digestion.

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## ABSTRACT

Anaerobic co-digestion has a potential to improve biogas production, but limited kinetic information is available for co-digestion. This study introduced regression-based models to estimate the kinetic parameters for the co-digestion of microalgae and Waste Activated Sludge (WAS). The models were developed using the ratios of co-substrates and the kinetic parameters for the single substrate as indicators. The models were applied to the modified first-order kinetics and Monod model to determine the rate of hydrolysis and methanogenesis for the co-digestion. The results showed that the model using a hyperbola function was better for the estimation of the first-order kinetic coefficients, while the model using inverse tangent function closely estimated the Monod kinetic parameters. The models can be used for estimating kinetic parameters for not only microalgae-WAS co-digestion but also other substrates' co-digestion such as microalgae-swine manure and WAS-aquatic plants.

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## 1. Introduction

Anaerobic digestion technology has been used in waste management for several purposes such as waste stabilization, solids reduction, and energy production (Angelidaki et al., 2003; Kythreotou et al., 2014). With the increasing interest in protecting environments and producing renewable energy, this technology becomes more popular due to its ability to produce biogas from waste (Kythreotou et al., 2014). However, anaerobic digestion of some substrates such as waste activated sludge, agricultural waste, and microalgae results in low biogas yield, because the substrate has low organic loadings (low carbon content) and high ammonia concentrations that negatively impact on the activity of methanogens during anaerobic digestion (Mata-Alvarez et al., 2014). Anaerobic co-digestion, which is the simultaneous digestion of two or

more substrates, could be a feasible option not only to overcome this drawback by supplying missing nutrients from co-substrates and diluting the potential toxic substances, but also to stimulate synergistic effects on microorganisms (Mata-Alvarez et al., 2000). Many substrates, including animal waste, sewage sludge, municipal organic solid waste, agricultural waste, fats, oil, grease, and microalgae have been used for co-digestion (Mata-Alvarez et al., 2014). In particular, studies on anaerobic co-digestion using microalgae have been increased for the last decade because microalgae have an ability to treat wastewater with high biomass productivity (Pittman et al., 2011). Due to this ability, microalgae have been used for nutrient recovery in nutrient rich wastewater such as rejecting water from sludge dewatering (Pittman et al., 2011; Olsson et al., 2014). Moreover, wastewater treatment integrated with microalgae cultivation and subsequent production of biogas from the co-digestion using Waste Activated Sludge (WAS) and microalgae can be one of the most promising options for renewable energy production at wastewater treatment plants (Ajeej et al., 2015; Wang et al., 2016).

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Anaerobic co-digestion has the same mechanism as anaerobic digestion that consists of a series of biological conversion processes in which multiple microorganisms break down biodegradable organic substances, and these processes are described by four major steps, including hydrolysis, acidogenesis, acetogenesis, and methanogenesis (Batstone et al., 2002; Gavala et al., 2003; Vavilin et al., 2008). It is generally accepted that hydrolysis and methanogenesis are rate limiting steps in the anaerobic digestion process (Gavala et al., 2003; Ariunbaatar et al., 2014). Due to enzymatic activity by hydrolytic bacteria to break down the large organic matters, hydrolysis is considered to be a slow reaction. On the other hand, methanogenesis is considered as another rate limiting step, because methanogenic bacteria require complex environmental conditions that are hard to maintain in digesters. For example, nitrogen contents between 3.5 and 8.7% in the substrates may result in methanogenesis inhibition (Costa et al., 2012). When the pH drops below 7.0 as a result of fast acidogenesis and acetogenesis steps, the activity of the methanogens is inhibited (Schwede et al., 2013). For the co-digestion of microalgae and WAS, hydrolysis and methanogenesis can be also considered as the rate-limiting steps because microalgae affect these steps (Costa et al., 2012). For instance, a hemicellulose composition of the microalgae cell wall impacts on the hydrolysis of the co-digestion (Northcote et al., 1958; Wang et al., 2013). Also, a high ammonia concentration resulting from degradation of protein content in microalgae negatively affects the methanogenic bacteria activity (Mairet et al., 2011).

The rates of these two steps have been described by different kinetic models, such as the first-order kinetic model, Monod model, and Andrews model (Kythreotou et al., 2014). Among these models, the first-order kinetic model was mostly used to explain the rate of hydrolysis, whereas the Monod model was commonly applied in kinetic modeling of methanogenesis. Vavilin et al. (2008) reviewed existing kinetic models for the hydrolysis of particulate organic materials in anaerobic digestion. For anaerobic digestion of complex organic substrate, they suggested a modified first-order kinetic model taking into consideration of non-biodegradable fraction of the substrate. In addition to improving the rate expression of the kinetic models, the determination of the kinetic parameters is critical for the overall model prediction.

The kinetic parameters are usually obtained from kinetic studies using an experimental approach (Lübken et al., 2015). This approach provides accurate kinetic information under specific conditions, but it requires time, energy, labor, and cost to obtain the results. There are many kinetic studies for anaerobic digestion, especially anaerobic digestion of sludge from wastewater treatment plant which has been well documented by Gavala et al. (2003). Based on the previous kinetic studies, it is found that majority of the studies focused on single substrates and limited studies dealt with determining the kinetic parameters for co-digestion. Costa et al. (2012) investigated methane production potential of anaerobic co-digestion of *Ulva* sp. and WAS in batch mode at mesophilic conditions. The parameters of the first-order kinetic model for different ratios of co-substrates were determined in the study (Costa et al., 2012). Neumann et al. (2015) studied anaerobic co-digestion of lipid-spent *Botryococcus braunii* with WAS and glycerol. They also determined the kinetic parameters for the first-order kinetic model under different ratios of the co-substrates. Zhen et al. (2015) evaluated the technical feasibility of anaerobic co-digestion of mixed microalgae and food waste in batch tests and explained the kinetics of methane production using the first order kinetics. The results from these prior studies showed that kinetic parameter values were different between single and multiple substrates. Depending on a ratio of co-substrates on a volatile solid basis (or percentage), the kinetic parameters for the co-digestion can be quite different. In addition, the kinetic

information for co-digestion of WAS and microalgae was very limited. Extensive experiments therefore need to be conducted in order to obtain kinetic parameters under different ratios of co-substrates.

This study aims at providing an alternative approach for estimating the kinetic parameters for co-digestion of microalgae and WAS under different ratios of co-substrates with limited kinetic experiments. The proposed kinetic parameter estimation models considered key factors which are ratios of co-substrates and the kinetic parameters for the single substrate. Among the existing kinetic models, the most applicable ones were selected – the modified first-order kinetic model for hydrolysis and the Monod model for methanogenesis (McCarty and Mosey, 1991; Vavilin et al., 2008). To demonstrate the applicability of the parameter estimation models, the models were applied to the published data from literature.

## 2. Methodology

### 2.1. Experimental method

#### 2.1.1. Microalgae cultivation

Indigenous *Chlorella* sp. was cultivated in 2L batch glass photobioreactors in two times diluted real centrate. The enrichment and identification of the algal species was done as described in Halfhide et al. (2015). The centrate was collected from the Northeast Water Reclamation Facility, NWRF (located in Clearwater, FL), which contains  $397 \pm 145$  mg  $\text{NH}_4\text{-N/L}$  and  $238 \pm 59$  mg TP/L. In order to remove particles, the centrate was filtered through glass fiber filters (Fisher Scientific, USA) with pore size of  $0.45 \mu\text{m}$ . The detailed characteristics and preparation of the centrate were described in Lee and Zhang (2016). The reactors were maintained at  $22 \pm 1$  °C in a temperature-controlled room. The cultures were kept suspended by aeration (0.03%  $\text{CO}_2$ ). A 24 h continuous light (about 9000 lx) was provided by 13 W fluorescent lamps.

#### 2.1.2. AD reactor set-up

Batch-type anaerobic digestion experiments were performed in duplicates of 100 mL glass serum bottles with a working volume of 40 mL for 20 days. The reactors were maintained at 35 °C and manually mixed twice each day. Anaerobic digested sludge and WAS were collected from NWRF. The anaerobic digested sludge was used as inoculum for the tests. The waste activated sludge was prepared by gravity setting or centrifugation, while the microalgae were harvested by centrifugation (3000 rpm, 15 min), in order to reach targeted Volatile Solids (VS) concentrations (5%). The characteristics of WAS, microalgae, and inoculum are shown in Table 1. To evaluate the effect of varying microalgae and WAS ratios on digestion performance, microalgae and WAS were added to the reactors to achieve the following mass (VS) composition: 100% WAS, 5% microalgae with 95% WAS, 10% microalgae with 90% WAS, 25% microalgae with 75% WAS, 40% microalgae with 60% WAS, 50% microalgae with 50% WAS, 75% microalgae with 25% WAS and 100% microalgae. A Substrate to Inoculum ratio (S/I) of 1 g VS/g VS was used for all experiments. Each bottle was purged with  $\text{N}_2$  gas before sealing to remove oxygen.

**Table 1**  
Characteristics of waste activated sludge, microalgae, and inoculum.

Parameters	Microalgae	Waste activated sludge	Anaerobic inoculum
TS (g/L)	$76.5 \pm 3$	$21.1 \pm 1.2$	$26.7 \pm 4.5$
VS (g/L)	$48.7 \pm 1.8$	$15.2 \pm 0.8$	$18.8 \pm 3$
COD (g/L)	$73.8 \pm 0.2$	$20.9 \pm 0.6$	$11.4 \pm 0.9$
TN (mg/L)	$1120 \pm 57$	$1590 \pm 74$	$739 \pm 20$
TP (mg/L)	$136 \pm 13$	$272 \pm 19$	$562 \pm 18$

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