



Application of a mixture design to identify the effects of substrates ratios and interactions on anaerobic co-digestion of municipal sludge, grease trap waste, and meat processing waste



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ABSTRACT

Anaerobic mono- and co-digestion of two municipal sludge wastes (A and C), grease trap waste (B), and meat processing waste (D) were investigated under mesophilic temperature conditions by biochemical methane potential (BMP) assays and kinetic modeling. Wastes ratios in the mixtures were systematically selected based on Simplex Lattice mixture design, and statistical analyses were performed to elucidate possible synergetic and antagonistic effects of wastes interactions on the kinetics and ultimate methane potentials of wastes co-digestion. The mixture of 1/8A + 1/8B + 1/8C + 5/8D (VS basis) showed the highest COD and VS removals of 35.0% and 33.8%, respectively. Substrates B and D with 980 and 641 mL/g-VS methane yields, respectively, had the highest BMP. However, with reaction rate constants of 0.047 and 0.070 d⁻¹, their methane production was very slow. It was observed that diluting these organic-rich but complex substrates with readily soluble wastes (A and C) enhanced their biogas production rate markedly. Statistical analysis showed that the interactions among the substrates in co-digestion did not have a significant impact on the ultimate cumulative methane yields. Nevertheless, these interactions proved to have synergetic and antagonistic effects on the reaction rates, leading to accelerated or hindered methane production rates. As a result, while the methane yield of wastes co-digestion could be predicted by proportional summation of methane yields obtained in mono-digestions of these waste fractions, such linear regressions were unable to provide a good estimation of the rate constants. Quadratic equations, however, were found to estimate the rate constants of the co-digestion process with good accuracy.

1. Introduction

With increasing trends of annual waste disposal, greenhouse gas emission, and energy costs, replacing the conventional waste management strategies with more efficient, environmental-friendly, and sustainable technologies has become a necessity. Anaerobic digestion of organic wastes is proved to be an effective method for stabilizing such substrates and reducing the risk of environmental pollution. In addition, this method has emerged as a viable technique for producing biogas as a source of renewable energy in today's energy-hungry world [1,2].

Anaerobic digestion of single waste streams usually entails undesirable constraints such as long retention times, low conversion efficiency, and sensitivity to waste load and toxic materials [3–5]. On the other hand, anaerobic co-digestion of two or more waste streams, is a promising approach to overcome these problems and enhance the

biotransformation efficiency of the process. Improved nutrient balance and bacterial diversity, dilution of toxic compounds, supplying buffering capacity, and establishing required moisture content are among the merits of anaerobic co-digestion. Furthermore, co-digestion is advantageous if the amount of waste generated at one particular site is not large enough to justify the investment for an on-site anaerobic bioreactor [4,6–8].

Fat, oil, and grease (FOG) are attractive substrates for anaerobic digestion. FOG is usually referred to the lipid-rich materials derived from animals and plants by-products, which are usually generated in restaurants and food processing plants [9–12]. These wastes are high in chemical oxygen demand and volatile solids, and have high ultimate digestibility and biogas production yield. However, due to issues like rapid acidification and process inhibition, sludge floatation, and clogging of pipes and collectors, mono-digestion of these wastes is usually problematic [13,14]. To address these issues, researchers have studied

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the feasibility of diluting FOG with waste activated sludge and performing co-digestion instead of mono-digestion. Many studies have reported on benefits of co-digestion of these types of wastes [9–11,13–15]. Brown and Li [16] reported increased methane yield for co-digestion of food and yard wastes. Grosser et al. [13] also reported the complementary properties of sewage sludge, organic fraction of municipal waste and grease trap sludge in anaerobic co-digestion process, which led to significantly higher methane yield and removal of volatile solids compared to mono-digestion of sewage sludge. In another study, Grosser and Neczaj [14] observed enhanced performance of anaerobic digestion of sewage sludge when mixed with fatty rich materials in semi-continuous bioreactors. They reported higher process efficiency and shorter hydraulic retention times in the case of co-digestion.

However, when conducting co-digestion experiments for verifying the feasibility of mixing different waste streams for biogas production and waste stabilization, it is important to not overestimate the synergistic effects of wastes interactions. For example, it could be that the increased biogas production is not a result of synergistic effect among the wastes but simply due to introducing higher amount of organic matter to the bioreactor. Therefore, for proper investigation of synergistic and/or antagonistic interactions of different wastes, controlled co-digestion experiments should be carried out in a systematic manner. The present work investigates anaerobic co-digestion of four different wastes in seven sets of mixtures: three 2-substrate, three 3-substrate and one 4-substrate mixtures. Selection of the wastes was based on the requirements of a wastewater treatment plant (WWTP), whose authorities are interested in co-digestion of in-house sewage sludge and the wastes received from local food industries. The aim of the study was to evaluate the effects of mixing different combinations of these four waste fractions on the biogas yield and biogas production kinetics. For this purpose, experiments were conducted by following design of experiment (DOE) and statistical analysis was performed on the collected data.

In co-digestion studies where effects of wastes' interactions on digestion process and biogas production are of interest, mixing ratios of the wastes are the experimental parameters and not the total amount of wastes mixture. Choosing the mixing ratios based on the "scattergun" procedure, where a large number of combinations are tried, usually requires large expenditures in terms of experiments' time and cost, and therefore better methods are sought. A full factorial experiment, where the response is dependent on changing the level of one factor at fixed levels of the other factors or changing the levels of two/more factors simultaneously, is not suitable for such system since the responses are affected by the amount of factors used. Such studies should be treated as mixture experiments. Mixture experiments are a special class of response surface experiments in which the product under investigation is made up of several components or ingredients. In a mixture experiment, the independent factors are proportions of different components of a blend and the proportions of the components must sum to 100%. This method is advantageous to (i) determine whether there exist some combination of the mixtures' ingredients leading to desirable product properties, i.e., higher methane yield and production rate, and higher VS and COD removals, and (ii) study the roles of different ingredients to gain a better understanding of the system. Besides, by applying this design, it is possible to mathematically model the system responses. These models are surface models enabling to optimize the proportions of the components for or predict, for any mixture of substrates, a target response variable [17].

To optimize mixing ratios and investigate possible synergy of co-substrates in AD process, in many co-digestion investigations reported in the literature, no technical basis has been followed in designing the experiments and selecting the wastes proportions in the mixtures. For example, in an attempt to maximize methane production from waste activated sludge, Alqaralleh et al. [11] used mixtures of 20%, 40%, 60%, and 80% (VS basis) FOG as co-substrate. In contrast, a threshold

concentration of greasy sludge was reported in another study, beyond which co-digestion with waste activated sludge is inhibited [18]. In other studies, a few mixtures were chosen to evaluate synergy and optimize mixing ratio of sludge samples in co-digestion with other wastes [19,20]. Furthermore, a proper basis is required for mixing different wastes and choosing their ratios in the mixture; in some studies, the proportions of the wastes have been selected based on volume such as by Obulisamy et al. [21], Xie et al. [22], and Park et al. [23]. The problem with using volume as the basis for mixing the substrates is that since the concentration of biodegradable matters varies from one waste to another, it is not clear in such studies whether the concentration or the nature of the biodegradable matter is affecting the methane production. Choosing the substrates ratios based on the total or volatile solid (TS or VS) content can provide a better picture of the synergistic or antagonistic interactions of the wastes.

Experiments in the present study were designed based on Simplex lattice mixture design of first degree (augmented with centre and axial points) and the substrates were mixed based on their content of volatile solids. All other experimental parameters were fixed so that a systematic comparison could be performed. Data obtained from these experiments were then statistically analyzed. The present study provides a comprehensive understanding of the effects of interactions among these different substrates on anaerobic co-digestion and it determines how these interactions alter the methane production in terms of cumulative yield and production rate.

2. Materials and methods

2.1. Substrates and inoculum

Four different wastes (two wastewater sludge samples, grease trap waste, and food processing waste from a local meat processing plant) were collected for the experiments. All the samples were obtained from a local wastewater treatment plant (WWTP) in Melbourne, Australia. The samples were denoted as follow: waste activated sludge from the WWTP (A), grease trap waste (B), wastewater treatment sludge from a second WWTP (C), and meat processing waste (D). The effluent from anaerobic digester of the WWTP, which provided the samples, was used as the inoculum. All the wastes samples were provided by the plant and were stored in closed containers at 4 °C upon arrival at University laboratory. Within two days, all the samples were characterized and batch experiments were then started. Key properties of the substrates and inoculum, determined according to methods described in Section 2.4, are summarized in Table 1.

2.2. Design of experiments and statistical analyses

It is well-established that the OVAT (one-variable-at-a-time) approach is not an efficient method for designing experiments, in

Table 1
Characteristics of substrates and inoculum. Data are averaged values of two replicates.

Parameters	Unit	Substrates				Inoculum
		A	B	C	D	
TS	%	2.97	16.28	1.01	9.26	2.91
VS	%	2.49	13.89	0.66	7.07	2.13
VS/TS	%	83.8	85.3	65.8	76.4	73.3
tCOD	mg/L	49700	245750	9430	188860	40300
sCOD	mg/L	7370	10980	340	4160	3210
sCOD/tCOD	%	14.8	4.5	3.6	2.2	8.0
Total N	mg/L	1470	2790	539	2655	2255
Total NH ₃ -N	mg/L	315	444.6	31.3	36.3	678
Total P	mg/L	1159.2	3134.7	888.3	5089.5	1590.8
Volatile fatty acids	mg/L	1106	2091	–	1029	80
pH	–	7.15	5.23	7.48	5.36	7.28

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