Process Improvement of Biogas Production from Anaerobic Co-digestion of Cow Dung and Corn Husk

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Abstract

A study of organic loading rate (OLR) and effect of NaOH concentration on the pretreatment stage of corn husk (CH) was conducted by Face-Centered Central Composite Design (CCF) to improve the biogas production. Three levels of OLR at 25.0 g VS · L⁻¹ · d⁻¹ (OLR25), 35.0 g VS · L⁻¹ · d⁻¹ (OLR35), and 45.0 g VS · L⁻¹ · d⁻¹ (OLR45) were performed with NaOH pretreatment concentration of CH at 25.0 % (N25), 35.0 % (N35), and 45.0 % (N45) (w/w). The optimum production of biogas at 67.6 mL · min⁻¹ with methane concentration of 63.4 % has been obtained at the application of OLR at 43.6 g VS · L⁻¹ · d⁻¹ and NaOH concentration at 33 % (w/w).

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Peer-review under responsibility of the Scientific Committee of HK-ICONS 2014

Keywords: biogas production; codigestion; improvement.
1. Introduction

Biogas could be defined as an end product of anaerobic digestion where a biochemical process is held during which complex organic matter is decomposed in the absence of oxygen, by various types of anaerobic microorganisms\(^1\). Biogas consists mainly of methane (CH\(_4\)), carbon dioxide (CO\(_2\)), and several other traces of gas and impurities. The gas components of biogas are specific to the plant and substrates type, and the composition of the biogas can only be partially controlled. However, the anaerobic digestion of biomass improves with longer time of exposure, where toward the end of the residence time, the concentration of methane increases disproportionately\(^2\).

1.1. Co-digestion of substrates and organic loading rate

Mixing organic matters by two or more substrates, which commonly known as co-digestion, may give a synergistic effect that can result in the higher production rate of the biogas\(^2\). This action improves the biogas production rate and methane concentration in the fermentation process and relies heavily on the organic loading rate (OLR) of co-substrates engage to the process. The most common applications of co-digestion can be found in agricultural biogas plants by using a basic substrate such as animal manure and by adding minor amounts of additional substrates\(^3\).

In anaerobic digestion process, OLR affects the stability of the fermentation process and the rate of gas production by providing digestible substrates for the growth of microorganisms\(^4\). It is an important factor that indicates on how much organic dry matter can be fed into the digester per volume and time unit, and has been defined as the amount of time that the biomass is retained within the digester\(^5\). With a precautious handling, an increase in the OLR might improve the biogas production by avoiding an outrageous OLR feeding which might impair the fermentation process and cause a wash-out of microorganisms.

1.2. Agricultural lignocellulosic biomass residues

Agricultural sector provides abundant biomass residues, and it has variously been estimated that these wastes can account for over 30 % of worldwide agricultural productivity\(^6\). Corn residues which are produced from this sector, which cover the husk, stover, and cob, are lignocellulosic biomass which has been commonly used as one of the substrates for biogas production worldwide. With the high content of cellulose and hemicelluloses, corn residues are considered to be a good substrate for the biogas production.

Lignocelluloses are basically composed of carbohydrates (cellulose and hemicelluloses), lignin, and extraneous materials\(^7\). However, the compact crystalline structure where lignin physically protects the cellulose and hemicelluloses parts makes these materials more resistant to anaerobic digestion\(^8\). In the anaerobic digestion process, if a substrate is well enclosed in lignin structures, the type of disintegration of the substrate becomes important. The structure should be disrupted or defibrated rather than cut, because they are refractory to decomposition under anaerobic conditions\(^9\). Without prior treatments, a slow hydrolysis might occur and biogas production could become low with a long retention time required to produce sufficient amount of biogas\(^9\).
1.3. Sodium hydroxide (NaOH) pretreatment

The pretreatment methods of lignocellulosic-containing substrates involve the use of sodium hydroxide (NaOH). It is the most popular alkali used in alkaline pretreatment, and has been extensively studied to improve biogas yield from lignocellulosic biomass. A study by Chandra et al. with the pretreatment of wheat straw with NaOH showed that NaOH pretreated substrate produced 87.5% higher biogas production and 111.6% higher methane production compared to the untreated wheat straw substrate. Another study conducted by Taherdanak and Zilouei in the utilization of NaOH for wheat pretreatment showed that the best improvement in the yield of methane production was achieved by pretreatment at 75 °C for 60 min, giving a methane yield of 404 mL · g⁻¹ VS. Zheng et al. provided a conclusive statement that NaOH is the most cost-effective and widely used alkali for lignocellulosic biomass pretreatment. However, its utilization should be handled with care, as it might cause Na⁺ ion inhibition of the fermentation process, especially methanogenesis.

2. Methods

2.1. Materials

Cow dung (CD) was taken from a local ranch at Gombak, Selangor State of Malaysia. The dung was collected and stored in a perforated container at room temperature for 7 d before use. Inoculums were taken from the slurry of operating biogas digesters in the anaerobic codigestion of cow dung, palm oil leaves, corn husk, and grass-cuttings.

Corn husk (CH) was taken from a local farming area at Salak Tinggi, Selangor State of Malaysia. The husk was cut in size of 5 cm to 10 cm and sun dried prior to drying process at 60 °C for 8 h. The dried husk was then grinded into a granular form and screened with a sieve size of 1.0 mm. The husk was then stored in a closed container at room temperature prior usage.

2.2. Fermentation setup

Cow dung was used as an initial basic substrate, complemented with CH as co-substrate assigned as a measured feeding input for the digesters. A total of six 9 d were used for the observation of the anaerobic fermentation process with two independent variables in the fermentation process. The first factor was the OLR of CH at 25.0 g VS · L⁻¹ · day⁻¹ (OLR₂₅), 35.0 g VS · L⁻¹ · d⁻¹ (OLR₃₅), and 45.0 g VS · L⁻¹ · d⁻¹ (OLR₄₅) per digester. The second factor was the concentration of NaOH for the pretreatment of CH at 25.0 % (N₂₅), 35.0% (N₃₅), and 45.0 % (N₄₅) of NaOH (w/w).

The fermentation was conducted in continuous and mesophilic condition, by initially mixing 30 kg of CD and 15 L of inoculums. Tap water was then added to the mixture, stirred for homogenous final mixture until working volume of the digester reached 90 L. Incubation of CD to the inoculums took 7 d prior to the feeding process of co-substrate.

2.3. Corn husk pretreatment

The pretreatment was conducted by soaking CH into the solutions of NaOH. The NaOH solution was prepared by dissolving the designated amount of NaOH with distilled water, thus providing nine level concentrations of solution (%) as shown in Table 1.

| Table 1. Sodium hydroxide solution preparation in terms of volume percentage |
|-----------------------------|-----------------------------|-----------------------------|-----------------------------|
| Factor 1: OLR | Factor 2: NaOH | NaOH (g) | NaOH solution (% w/v) |
|----------------|----------------|----------------|-----------------------------|
| OLR₂₅ | N₂₅ | 21.02 | 0.93 |
| OLR₂₅ | N₃₅ | 29.43 | 1.31 |
| OLR₂₅ | N₄₅ | 42.04 | 1.87 |
| OLR₃₅ | N₂₅ | 29.43 | 1.31 |
| OLR₃₅ | N₃₅ | 41.20 | 1.83 |

Continued on next page
The designated concentration of CH was added and stirred into the NaOH solution until it reached homogenous state to the designated level. The feeding mixtures were stored in closed vessels at room temperature for 5 d.

2.4. Fabrication of digesters

Three identical cylindrical digesters were made of polyethylene with total capacity of each at 160 L. The temperature of the fermentation rely on the ambient temperature and the mixing process is passive stirring, which occurred by the insertion of new feeding and the subsequent thermal convection streams as well as by the up-flow of gas bubbles. Each digester was connected to a water displacement unit, manometer, and thermometer. The schematic arrangement for a unit of digester is provided from Figure 1.

The three digesters were utilized and differed based on the OLR factors. The feeding strategy was conducted in a continuous manner by flowing in 3 L CH co-substrate, making the hydraulic retention time (HRT) set at 30 d. The feeding process was repeated for every 24 h.

2.5. Analytical procedures

Biogas production rate, methane concentration, and slurry samples were taken every 24 h prior the regular feedings. The gas production rate was measured by water displacement method described by Parajuli. The biogas concentration were measured by gas analyzer GA-m Binder Combimass® by taking 3 L of gas sampling volume directly from the digesters. Slurry samples were taken at 600 mL from the effluent of the fermentation and stored in an air-tight container with temperature set to 4 °C prior the analysis.

Total solids (TS) and Volatile solids (VS) of substrates and slurry samples were measured according to the standard methods provided by US Environmental Protection Agency (EPA). Samples of CD and CH, each in
triplicate, were analyzed to approximate the content of organic fraction present prior the start of the fermentation process. The samples of CD were taken before the incubation of the basic substrate with inoculums at an average VS value observed at 300.5 g VS · kg⁻¹ CD, and organic dry matter content as much as 85.62 % of total dry matter. The samples of CH were taken from the dried and granulated CH with an average value of VS at 0.892 g VS · g⁻¹ dried-granulated CH.

2.6. Design of experiment and statistical analysis

Face-Centered Central Composite Design (CCF) was selected to find the optimal process settings of CH OLR and NaOH pretreatment, estimate their interaction and quadratic effects, as well as predicting the shape of the response surface investigated. A set of ten experimental runs with two centre points was replicated in an experimental augmentation, resulted in a total of twenty experimental runs with four centre points. Three different levels of low (-1), medium (0), and high (+1) were accounted to each independent variable. The design of experiments recipe and analysis were generated by Design Expert Software® (Version 7.0.0, Stat-Ease Inc., Minneapolis, USA). The data were checked through the Lack of Fit test and Analysis of Variance (ANOVA), and compared with the predicted values generated by the software. The flow of the experimental runs is provided from Table 2.

Optimization computation with the software was conducted based on the research responses toward the operating conditions. The optimization was aimed to search the combination of factor levels that simultaneously satisfy the requirements placed on each of the responses and factors.

| Table 2. Flow of experimental runs based on the time of fermentation and digester |
|-----------------------------|-----------------------------|-----------------------------|
| Time of fermentation (d) | Digester identification | Independent variables |
|                            |                            | (In coded factors)          |
| Day 1 of fermentation   | Digester 1                 | OLR₃₅                      |
|                          | Digester 2                 | OLR₃₅                      |
|                          | Digester 3                 | OLR₄₅                      |
| Day 2 of fermentation   | Digester 1                 | OLR₃₅                      |
|                          | Digester 2                 | OLR₃₅                      |
|                          | Digester 3                 | OLR₄₅                      |
| Day 3 of fermentation   | Digester 1                 | OLR₃₅                      |
|                          | Digester 2                 | OLR₃₅                      |
|                          | Digester 3                 | OLR₄₅                      |
| Day 4 of fermentation   | Digester 1                 | OLR₃₅                      |
|                          | Digester 2                 | OLR₃₅                      |
|                          | Digester 3                 | OLR₄₅                      |
| Day 5 of fermentation   | Digester 1                 | OLR₃₅                      |
|                          | Digester 2                 | OLR₃₅                      |
|                          | Digester 3                 | OLR₄₅                      |
| Day 6 of fermentation   | Digester 1                 | OLR₃₅                      |
|                          | Digester 2                 | OLR₃₅                      |
|                          | Digester 3                 | OLR₄₅                      |
3. Results and discussion

3.1. Biogas production rate

The experimental design and the actual response values paired with the biogas production rate are presented from Table 3. The suggested model is quadratic (p-value 0.0186) and the Predicted R-Squared of 0.7160 is in reasonable agreement with the Adjusted R-Squared of 0.8107. Both independent variables of OLR and NaOH concentration affected the production rate significantly.

The 3D contour for the production rate is presented in Figure 2. The maximum rate obtained was 75.8 mL biogas min⁻¹ with 45 g VS · L⁻¹ · d⁻¹ of OLR and 25.0 % application of NaOH pretreatment concentration, whilst the minimum observed at 20.52 mL · min⁻¹ of biogas with 25 g VS · L⁻¹ · d⁻¹ of OLR and 45.0 % of NaOH (w/w). The contour of 70.00 mL · min⁻¹ of production rate on the range of (44.25 to 45.00) g VS · L⁻¹ · d⁻¹ OLR, constrained with (27.0 to 39.0) % NaOH can be seen from Figure 3.

In general, higher value of OLR gave higher production rate of biogas. A production rate above 45.00 mL · min⁻¹ of biogas was observed when the OLR given was above 35 g VS · L⁻¹ · d⁻¹. In the same way, NaOH tend to increase the rate when the application was in the range of (30.0 to 37.5) % of NaOH with constraint of OLR application more than 30.0 g VS · L⁻¹ · d⁻¹. Below than the mentioned OLR value, the production rate would be lower than 34.00 mL biogas · min⁻¹.

Table 3. Experimental design using CCF on two factors and its actual response of biogas production rate

| Run | Factor 1: OLR (g TS/ 3 L feeding · day⁻¹) | Factor 2: NaOH pretreatment (% w/w) | Response 1: Production rate (mL · min⁻¹) |
|-----|------------------------------------------|-------------------------------------|----------------------------------------|
| 1   | 45.00 (151.35)                           | 25.00 (37.84)                       | 58.43                                  |
| 2   | 35.00 (117.71)                           | 25.00 (29.43)                       | 40.35                                  |
| 3   | 25.00 (84.08)                            | 25.00 (21.02)                       | 30.33                                  |
| 4   | 45.00 (151.35)                           | 25.00 (37.84)                       | 75.81                                  |
| 5   | 35.00 (117.71)                           | 25.00 (29.43)                       | 42.86                                  |
| 6   | 25.00 (84.08)                            | 25.00 (21.02)                       | 26.92                                  |
| 7   | 45.00 (151.35)                           | 35.00 (52.97)                       | 66.12                                  |
| 8   | 35.00 (117.71)                           | 35.00 (41.20)                       | 35.86*                                 |
| 9   | 35.00 (117.71)                           | 35.00 (41.20)                       | 35.44*                                 |
| 10  | 25.00 (84.08)                            | 35.00 (29.43)                       | 26.53                                  |
| 11  | 45.00 (151.35)                           | 35.00 (52.97)                       | 75.56                                  |
| 12  | 35.00 (117.71)                           | 35.00 (41.20)                       | 50.63*                                 |
| 13  | 35.00 (117.71)                           | 35.00 (41.20)                       | 50.30*                                 |
| 14  | 25.00 (84.08)                            | 35.00 (29.43)                       | 48.67                                  |
| 15  | 45.00 (151.35)                           | 45.00 (68.11)                       | 69.23                                  |
| 16  | 35.00 (117.71)                           | 45.00 (52.97)                       | 34.23                                  |
| 17  | 25.00 (84.08)                            | 45.00 (37.84)                       | 20.83                                  |
| 18  | 45.00 (151.35)                           | 45.00 (68.11)                       | 55.79                                  |
| 19  | 35.00 (117.71)                           | 45.00 (52.97)                       | 28.39                                  |
| 20  | 25.00 (84.08)                            | 45.00 (37.84)                       | 20.52                                  |

*) Data center points
The maximum rate of biogas production at 75.8 mL · min⁻¹ by the application of 45 g VS · L⁻¹ · d⁻¹ (which is equal to 0.135 kg VS · d⁻¹), with the assumption of constant production rate, is equivalent as 808.6 L biogas kg⁻¹ VS 90 L⁻¹ working volume. This amount could be converted to 8.9 L biogas kg⁻¹ VS · L⁻¹ working volume. As a comparison, Zhu et al. [15] studied the effect of NaOH pretreatment on corn stover and obtained a maximum accumulative yield of 372.4 L kg⁻¹ VS by 5.0 % NaOH in 40 d of batch process at 2.0 L fermentation. This value was equal to 4.66 L biogas kg⁻¹ VS · d⁻¹ · L⁻¹ of fermentation.

Based on these responses, the increase of OLR above 45.0 g VS · d⁻¹ · L⁻¹ is expected to increase the rate, with the execution still within the acceptable corridor of NaOH pretreatment concentration. At the opposite side, the lower region of 34.00 ml biogas · min⁻¹ with OLR below 32.5 g VS · d⁻¹ · L⁻¹ and NaOH below 30.0 % could be taken as a hint that low OLR particularly and low concentration of NaOH is susceptible to low production of biogas.

3.2. Methane concentration

The experimental design and the actual response values of methane concentration is presented in Table 4. Both OLR and NaOH pretreatment concentration are significantly affecting the response. The model for the evaluation followed a quadratic model, with an F-value of 33.00 implying that the model is significant. The Predicted R-Squared of 0.8348 is in reasonable agreement with the Adjusted R-Squared of 0.8938.

The 3D response surface contour for methane concentration is presented in Figure 4. The maximum methane concentration was 68.8 % by volume, obtained at 45.0 g VS · d⁻¹ · L⁻¹ OLR and 45.0 % NaOH pretreatment concentration. The minimum content observed was at 57.1 % of methane at 45.0 g VS · d⁻¹ · L⁻¹ OLR and 25.0 % application of NaOH (w/w).
Table 4. Experimental design by CCF of two factors and the actual response of methane content.

| Run | Factor 1: OLR (g TS/3 L feeding · d⁻¹) | Factor 2: NaOH pretreatment (% w/w) | Response 2: CH₄ content (% v/v) |
|-----|-------------------------------------|------------------------------------|-------------------------------|
| 1   | 45.00 (151.35)                      | 25.00 (37.84)                      | 57.1                          |
| 2   | 35.00 (117.71)                      | 25.00 (29.43)                      | 58.2                          |
| 3   | 25.00 (84.08)                       | 25.00 (21.02)                      | 60.0                          |
| 4   | 45.00 (151.35)                      | 25.00 (37.84)                      | 59.8                          |
| 5   | 35.00 (117.71)                      | 25.00 (29.43)                      | 58.9                          |
| 6   | 25.00 (84.08)                       | 25.00 (21.02)                      | 59.2                          |
| 7   | 45.00 (151.35)                      | 35.00 (52.97)                      | 64.2                          |
| 8   | 35.00 (117.71)                      | 35.00 (41.20)                      | 61.7*                         |
| 9   | 35.00 (117.71)                      | 35.00 (41.20)                      | 61.7*                         |
| 10  | 25.00 (84.08)                       | 35.00 (29.43)                      | 61.7                          |
| 11  | 45.00 (151.35)                      | 35.00 (52.97)                      | 66.7                          |
| 12  | 35.00 (117.71)                      | 35.00 (41.20)                      | 63.5*                         |
| 13  | 35.00 (117.71)                      | 35.00 (41.20)                      | 63.4*                         |
| 14  | 25.00 (84.08)                       | 35.00 (29.43)                      | 63.6                          |
| 15  | 45.00 (151.35)                      | 45.00 (68.11)                      | 68.8                          |
| 16  | 35.00 (117.71)                      | 45.00 (52.97)                      | 66.7                          |
| 17  | 25.00 (84.08)                       | 45.00 (37.84)                      | 64.9                          |
| 18  | 45.00 (151.35)                      | 45.00 (68.11)                      | 68.7                          |
| 19  | 35.00 (117.71)                      | 45.00 (52.97)                      | 66.8                          |
| 20  | 25.00 (84.08)                       | 45.00 (37.84)                      | 62.9                          |

*) Data center points

The increase in OLR and NaOH concentration led to higher methane concentration. The synergistic factors might be increasing of OLR by supplying the substrates for methanogenesis process. However, the high concentration of NaOH might contribute to the provision of alkaline environment, resulting in a suitable living environment for methanogens. This is because methanogens is severely influenced by operating conditions, including feeding rate and pH value\(^1\). A study by Zhu et al.\(^15\) from a biogas production of corn stover, showed that the methane concentration was ranged between 50 % to 60 %. Meanwhile, another study from Zhou et al.\(^16\) with pretreated corn stover with cow dung showed methane content at a higher rate of 75 % to 80 % by volume.
3.3. Optimization of biogas production

The parameters involved in the computation of the process optimization were the independent variables of OLR and NaOH pretreatment concentration, biogas yield response, and methane yield response. There was no extrapolation of the parameters limit attempted to be incorporated in the optimization process, as the response surface model beyond the response and factors is unknown. The response setting of biogas yield was set to 62.5 mL · min⁻¹ as the lower limit. The complete constraints on the parameters of the optimization are provided on Table 5.

Table 5. Summary of criteria on the optimization process with response on the lower limit of biogas yield at 62.5 mL · min⁻¹

| Parameter constraints     | Goal    | Lower limit | Upper limit | Lower weight | Upper weight | Importance |
|---------------------------|---------|-------------|-------------|--------------|--------------|------------|
| OLR (g VS · L · d⁻¹)      | Minimize| 25.0        | 45.0        | 1            | 1            | 3         |
| NaOH (% w/w)              | Minimize| 25.0        | 45.0        | 1            | 1            | 3         |
| Production rate (mL · min⁻¹) | Maximize| 62.5        | 75.81       | 1            | 1            | 3         |
| CH₄ (% v/v)               | Maximize| 57.1        | 68.8        | 1            | 1            | 3         |

The solution provided from the computation of the optimum biogas production rate was at 67.56 mL · min⁻¹ of biogas with methane concentration at 63.4 % of CH₄ by volume. The requirement for the production rate and concentration was to set the OLR at 43.62 g VS · d⁻¹ · L⁻¹ and paired with the concentration of NaOH at 33.14 % (w/w) in the pretreatment of CH.
4. Conclusion

Organic loading rate may affect biogas production rate and methane concentration in a positive manner, where an increasing of OLR provides a better production either concentration or rate of biogas. However, there is a synergistic effect between OLR with the function of NaOH to degrade lignin in the content of co-substrate, can reduce the residence time and improve the quality of the biogas. The result of the process optimization shows that the increased production of biogas yield at 67.6 mL · min⁻¹ with 63.4 % methane concentration require 43.6 g VS · L · d⁻¹ of OLR and the 33.1 % of NaOH (w/w). The requirement for the production rate and concentration was to set the OLR and NaOH at the minimum utilization. However, the given solution needs to be evaluated thoroughly to meet the best option, in terms of cost, efficiency, and practicality.

Acknowledgements

The authors would like to express their gratitude to Ministry of Education (MOE) of Malaysia for supporting this project under KTP Fund (No. KTP12-001-0001).

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