Screening of Factors Affecting Pre-treatment by Ammonia-N Removal from Poultry Manure Wastewater by Using Soil Water to Improve Biogas Production

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Abstract. Direct land application of poultry manure wastewater (PMW) is the most common utilization option, but it can result in environmental pollution of waters, odor nuisance and hygienic problems which can support the spread of disease. One of the efficient methods to treat PMW is by anaerobic digestion. However, excess ammonia-N can cause inhibition in the process. Therefore, pre-treatment for anaerobic digestion of PMW need to considered. In this study, the factorial analysis and best pre-treatment condition of PMW were determined for the purpose of improving biogas production. The design of experiment (DOE) in this study was generated by using the Design Expert. Two-level full factorial design (FFD) was used to construct experimental table, analyzed the main factors that affect the process and determined interactions between the factors. Five factors were chosen for factorial screening namely: agitation (0 or 200 rpm), reaction time (2 to 5 hours), type of soil (peat soil (PS) or poultry farm soil (PFS)),soil to water ratio (1:1 or 1:4) and PMW to soil water (SW) ratio (1:4 or 2:3). Based on the result, agitation gave highest contribution at 38.36% followed by PMW to SW ratio at 29.76% contribution. In term of interaction, agitation and reaction gave the highest contribution to pre-treatment process at 3.33% contribution. The best pre-treatment condition suggested by Design Expert software was using PS as source of soil at soil to water ratio of 1:6, and mixed with PMW at 1:4 ratios without agitation for 5 hours reaction time. Application of this best pre-treatment condition to biogas production showed improvement in biogas yield by 82 % where biogas yield was up to 0.0248 L/g COD by using treated PMW compared to only 0.0045 L/g COD biogas yield by using untreated PMW.

Introduction

In Malaysia, from the statistics produced by the Department of Veterinary Services (DVS), from year 2001 to 2011, the output of livestock products from poultry industry has increased 35.6% from 7532.55 ('000) metric tons to 11692.47 ('000) metric tons. Along with the increasing production of poultry, the amount of poultry manure is also rising.

At present, anaerobic digestion is an efficient conversion process for poultry manure producing a collectable biogas mixture while reducing the adverse impact on the environment [5]. In general, poultry manure (PM) contains two forms of nitrogen, which are uric acid and undigested proteins, which represent 70% and 30% of the total nitrogen in PM, respectively [1]. Anaerobic decomposition of uric acid and undigested proteins in PM resulted in the production of high amounts of unionized ammonia and ammonium ions [14, 3]. Excess of ammonia can inhibit the anaerobic microbial consortia necessary for the production of biogas [13]. Air stripping can be used to remove ammonia from materials to be digested [8], or ammonia may be removed from anaerobic digester by addition of phosphorite or, supposedly by either immobilizing methanogens, which increases the buffering capacity of the medium or by exchanging ammonium ions for cations [7]. Based on study by Abouelenien *et al.,* (2010), when poultry manure was anaerobically digested, 82% of the produced ammonia was successfully removed by recycling the biogas. However, methanogenic bacteria became acclimatized after long lag phase and longer time was taken. Thus, it is difficult to control ammonia inhibition in the practical operation. In the light of these concerns, the aim of this study was to study the best conditions for pre-treatment of PMW for ammonia removal by using soil mixed culture.

Soil contains a complex biological community of microscopic organism, including bacteria, protozoa, and fungi, among others. Some of these organisms feed on the organic matter in wastewater [6]. Mixed culture from soil did contain high amount and variety species of bacteria such as Enterobacter Soli [12]. Ammonia-N from waste resulted from denitrification of nitrogen. Denitrification requires the presence of nitrate and organic carbon as an energy source for denitrifying bacteria under anaerobic conditions. This make the used of soil mixed culture in ammonia-N removal in pre-treatment as one of the most suitable option while reducing cost compare to use chemical and additives and to ensure highest productivity in return.

Materials and Method

The materials and method consist of two parts. The first part consists of the pre-treatment while the second one is the anaerobic digestion, which to validate the efficiency of the first part.

Pre-treatment for PMW

Poultry manure wastewater preparation. Fresh PM was collected from a moderate size poultry farm located at Kuantan, Pahang, Malaysia and was stored at 4°C. In order to maintain the moisture consistency, the PM was mixed with distilled water at 1:1 feed ratio, thoroughly for 5-10 minutes. It is reported that nutrients present in the manure can be sufficient for anaerobic microbial growth if sufficient amount of water is present [5].

Soil water preparation. The first type of soil was collected at one specific site of UMP, Pahang (Malaysia), namely peat soil (PS). The other one was collected near the poultry farm area, namely poultry farm soil (PFS). The soils were kept frozen just prior to use. Upon pre-treatment, the soil was then mixed thoroughly for 5-10 minutes with distilled water to produce soil water (SW). The soil to water ratio was determined as in Table 1.

Experimental set up and statistical analysis. The design of experiment (DOE) in this study is generated by using the Design Expert Version 7.1.6 software (Stat-Ease Inc., Minneapolis, USA). Factorial design allowed determination whether interactions between the factors occurred and also to obtain quantitative cause-effects relationships [15]. Responses were analyzed using analysis of variance (ANOVA) combined with the F-test to evaluate significant factors at the level of 5% ($p<0.05$). The factors chosen were listed as in Table 1 below.

Table 1: Independent variable use in factorial design

Independent Variable	Model symbol	Low actual	High actual
Agitation (rpm)	A	0	200
Reaction Time (hour)	B	2	5
Type of soil	C	PS	PFS
Soil : water	D	1:6	1:1
PMW : soil water	E	1:4	2:3

condition. The flask were filled with PMW first, and then reaction time started as soon as the SW was added. Mixing took place by using the New Brunswick Scientifics Shaker for agitation purpose. Ammonia-N and Chemical Oxygen Demand (COD) concentration were determined by using HACH Spectrophotometer DR/2400 @ DR/2800 following Method 8155 and Method 8000, respectively with suitable dilution factor.

Anaerobic digestion for PMW

Soil mixed culture preparation. Soil used in soil mixed culture preparation was different from the soil used for pre-treatment part. The soil used in soil mixed culture was poultry soil collected besides the poultry barn. Treated PMW was acclimatized with soil mixed culture anaerobically at 1:4 ratios, producing seeding to be use in anaerobic digestion process. The acclimatization hydraulic retention time (HRT) was 30 days, running at ambient temperature.

Experimental Set up. Anaerobic digestions were carried out at lab scale by two set of experiment. One flask was feed with untreated PMW while the other was feed with treated PMW. After that, seeding was added to each flask, and reaction time started. The seeding to feed ratio was kept constant for both flask at 1:4. The flasks were then covered with silicone tube with gas line to the biogas collector to ensure anaerobic condition within flask. The biogas was collected by using water displacement method. No agitation applied during the whole digestion process and was run at room temperature. Reading of biogas volume was taken daily until gas productions eventually stop. Biogas yield can be evaluated as summarize in Eq.2 below.

Biogas Yield (Y) = (biogas production volume/ initial concentration of COD) L/g COD (2)

Result and Discussion

Main Effect Analysis. The initial ammonia-N concentration of PMW used in this study varied between 600-2100 mg/L, oppose with Liu *et al.*, (2012) at range 400-3000 mg/L. Liu and Sung (2002), stated that ammonia concentration below 200 mg/L are beneficial to anaerobic process. However, ammonia inhibition can start at ammonia content up to 1000 mg/L [10].

Table 2: Analysis of Variance

	Mean Squares	F Value	p-value Prob>F
Agitation	2126.34	75.14	< 0.0001
Reaction Time	120.47	4.26	0.0547
Type of soil	19.96	0.71	0.4127
Soil : water	267.90	9.47	0.0068
PMW : soil water	1649.53	58.29	< 0.0001

A total of 32 runs were made in response to R1, with R squares (R^2) of 0.9132. Factor that having p-value less 0.05 were considered as potentially significant. From Table 2, three factors were considered significant were the agitation, soil to water ratio and also PMW to soil ratio. Reaction time was slightly significant with P value 0.0547 and type of soil is not a significant factor. The significant of factors were supported by the percent contribution by each model as in Table 3 below.

Term	% Contribution
A - Agitation	38.36
B - Reaction Time	2.17
C - Type of soil	0.36
D - Soil : water	4.83
E - CM : Soil water	29.76
AB	3.33
CD	1.22
CE	0.58
AC	0.33
AE	0.22

Table 3: The percentage contribution of each main factor and their interaction

Based on Table 3, agitation at 0 rpm has the highest contribution at 38.36%. This means that mixing or the ability of the soil to treat wastes as in the amount of accessible soil particle surface area as stated by Hygnstrom *et al.*, (2011) was not much necessary to ensure the efficiency of the pre-treatment. Meanwhile, the best PMW to soil ratio or so as considered as food-to-microorganism (F/M) was at 1:4 ratio with 29.76% contribution. This supported by the fact that a high F/M ratio provides a high driving force for metabolic activity and microbial growth and high overall rates of waste converse to biogas [11].

The best soil to water ratio was 1: 1 and the reaction time was 5 hours with 4.83% and 2.17% contribution, respectively. This mean, for efficient ammonia-N removal, more than 2 hours reaction time needed and 5 hours reaction time was preferable. The effect of type of soil was not significant with only 0.36% contributions and can be eliminated in further optimization works. Even the soil type was not significant, but sand soil give higher ammonia-N removal, due the nature of the particular substances in the wastewater [4].

The best pre-treatment condition suggested by Design Expert software was using PS as source of soil at soil to water ratio of 1:6, and mixed with PMW at 1:4 ratios without agitation for 5 hours reaction time. This count for 1.17% of error from expected result. The coded mathematical model for 2^5 factorial designs can be given as Eq. 3 below:

$$R1 = 59.12 - 8.15\,A - 1.94\,B - 0.79\,C + 2.89\,D - 7.18\,E - 2.40\,AB - 0.75\,AC - 0.62\,AE + 0.007188\,BC + 0.29\,BD + 1.46\,CD + 1.00\,CE - 3.45\,ACE - 2.36\,BCD \quad (3)$$

Interactions between Factors. There were two interaction discovered in this study with high effect among other interactions. The first one with 3.33% contribution was the interaction between factor agitation (A) and reaction time (B). Based on Fig. 1 and Table 3, at agitation 0 and 200 rpm, reaction hour of 5 hours was much preferable compare to the other reaction time of 2 hours. In this study, the ammonia-N removal runs at aerobic conditions and according to Yetilmezsoy and Sapci-Zengin (2009), air flows play an important role in ammonia-N volatization. As air introduced, it begins to agitate the solution, creating a removal pathway for dissolved free ammonia to volatize and leave the solution. By referring to Table 3 and Fig.2, the other interaction was in between factor type of soil (C) and soil to water ratio (D), with 1.22 % contribution. The fact that the soil contains a complex microscopic organisms and some of these organism feed on the organic matter in the poultry manure wastewater [6], and in order for reaction to take place, the amount of soil particle was consider as one of the main contribution, where soil to water ratio took place.

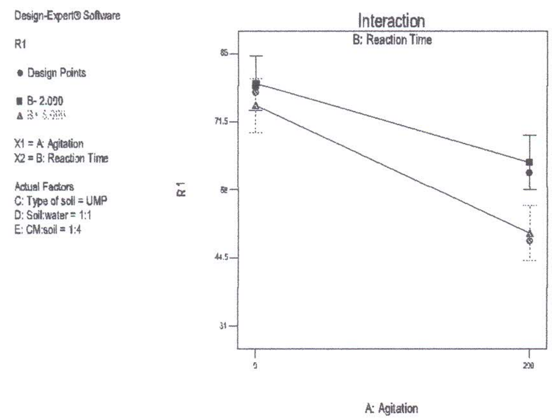

Fig. 1: The interaction graph between factors agitation (A) and reaction time (B)

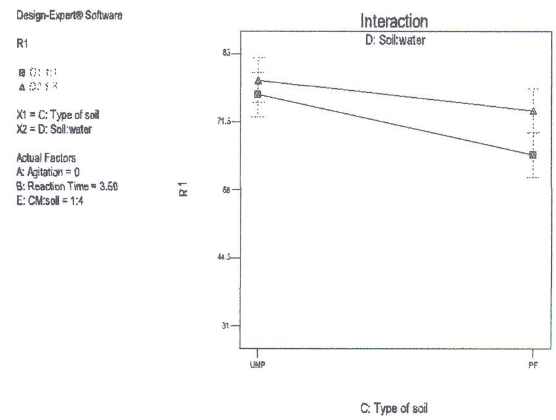

Fig. 2: The interaction graph between factors type of soil (C) and soil to water ratio (D)

Anaerobic digestion analysis. In untreated PMW, the initial COD concentration was 180,000 ± 14,200 mg/L while the treated PMW was 15,200 ± 3400 mg/L. By observing Fig. 3, the biogas yield for treated PMW was noticeable much higher than the untreated PMW. The total gas yield for treated PMW was 0.0248 L/g COD, which was 82.85% higher than that of the untreated PMW with total accumulation of 0.0045 mL/g COD. Regarding to the production of biogas conducted by Abouelenien (2009), fermentation of dry PM for production of methane was studied under laboratory conditions. Biogas was successfully produced after an acclimation period about 254 d. However, compare to this study, by using treated PMW with soil mixed culture, the biogas was successfully produced after an acclimation period of 30 d only. This shows that high ammonia content had to be resolve first before biogas production started. This to ensure that ammonia inhibition could be avoided and response to higher yield of biogas.

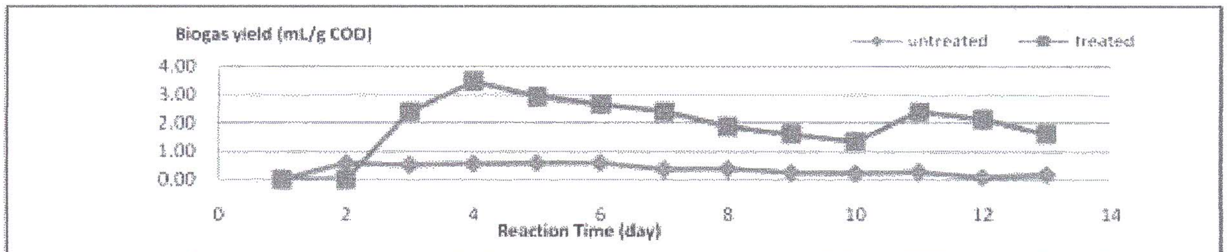

Fig. 3: Biogas yield graph

Summary

The objective of the study was achieved. Agitation gave highest contribution at 38.36% followed by PMW to SW ratio at 29.76% contribution. In term of interaction, agitation and reaction gave the highest contribution to pre-treatment process at 3.33% contribution. The best pre-treatment condition suggested by Design Expert software was using PS as source of soil at soil to water ratio of 1:6, and mixed with PMW at 1:4 ratios without agitation for 5 hours reaction time. Application of this best pre-treatment condition to biogas production showed improvement in biogas yield by 82 % where biogas yield was up to 0.0248 L/g COD by using treated PMW compared to only 0.0045 L/g COD biogas yield by using untreated PMW.

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