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Optimization of bio gas production from slaughterhouse wastes

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This study focuses on the production of biogas from slaughter house waste to generate optimal methane yield which has high calorific value under optimum temperature, pH and substrate proportion (blood, manure and undigested food material). In this study, two different kinds of experiment were done. The first experiment was carried out to determine the pH, total solid, volatile solid and ash content of each type of waste fermented independently at optimal temperature (35°C) and pH 7. The second experiment was done by mixing three types of wastes based on crossed D-optimal design expert software, a well-accepted statistical technique used to design and optimize the experimental process. It involves choosing the optimal experimental design and estimating the effect of the several variables which have 42 runs at different proportions of the substrates. After the experiment, the following parameters: methane content and volume of biogas, pH, temperature, total solid, volatile solids, and ash content of each type of waste were determined. From individual type of 0.351 sample waste (blood, manure and undigested content), maximum biogas production and methane composition obtained were 01, 71, 41 and 0%, 66% and 54%, respectively. From the 42 runs experimental result, the best optimal methane composition and biogas produced were 79.26% and 0.381 l/g, respectively. Optimal conditions included 20% blood, 20% manure, and 60% undigested content substrate at pH 7.88 and t 32.49°C temperature.

Key words: Digester, biogas, methane, anaerobic digestion, abattoirs.

INTRODUCTION

Nowadays, the discharge of wastes and demand of energy is increasing from time to time because of fast growing population, urbanization and industrialization. In addition, growing scarcity of petroleum and coal,

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deforestation of forests for fuel and problems related to emission of greenhouse gases such as CO_2, methane, and hydrogen sulfide from landfill sites are the challenges facing developing countries (Ali et al., 2014). To overcome the energy crisis, environmental pollution and global warming, renewable energy sources such as generation of energy from wastes (biogas), solar, wind, thermal and hydro powers should be utilized. In Ethiopia, high percentage of total energy consumption depends on biomass which results in deforestation and climate change (Amare et al., 2015).

In Ethiopia, there is no organized strategy for disposal of solid as well as liquid wastes generated in abattoirs. The solid slaughterhouse waste is collected and dumped in landfills or open areas while the liquid waste is sent to municipal sewerage system or water bodies, thus endangering public health as well as terrestrial and aquatic life (Ukpong et al., 2013). Wastewater from slaughterhouses is known to cause an increase in biological oxygen demand (BOD), chemical oxygen demand (COD), total solids, pH, temperature and turbidity, and may even cause deoxygenation of water bodies (Tekenah et al., 2014)

In Ethiopia, enough attention has not yet been given to biogas production from slaughter house waste; consequently, the countries are among methane emitter countries (Global Methane Initiative, 2011). Besides, they are not gaining enough amount of advantage from the resource like bio-slurry which can produce organic fertilizer. As a result, the countries import huge amount of petroleum products from abroad while it is possible to use bio-gas produced from high resource of livestock and slaughter wastes to significantly reduce the consumption of fossil fuel. In addition, the byproduct of biogas production, bio-slurry can be used as input for fish farming in commercial fish ponds.

This project aims at utilizing abattoir wastes to produce bio gas from the abundant resource of slaughter house waste by using anaerobic digestion. This can be used as source of energy for abattoir and for mitigating greenhouse gases by decreasing methane being emitted into the atmosphere from slaughter house wastes and livestock industry.

MATERIALS AND METHODS

Raw materials

The slaughterhouse considered in this study is Addis Ababa Abattoirs Enterprise in Addis Ababa city. Freshly voided blood, stomach content, and manure of slaughterhouse wastes were the major feedstock collected from Addis Ababa slaughter house. The sampling site is the three slaughtering houses for goat, sheep and cattle containing mixed effluent. The collected fresh sample was immediately fed to the digester to avoid being outside the reactor.

Sample preparation was done in two different ways: the first one was done without mixing the waste and without inoculum. The second one involved mixing the three types of wastes at desired proportion with inoculum (Table 4). The sample prepared in the 42 runs was done by mixing three types of wastes in the laboratory. Water was added in both procedures to obtain the desired total solid concentration of 7 to 10% inside the reactor, and the ratio of the waste to water was 1:1. The characteristics of the wastes are shown in Table 1.

Inoculum

The inoculum used in this study was bio slurry of cattle dung which contains active microbes essential for anaerobic digestion process. The percentage of inoculum for anaerobic fermentation of the organic waste is approximately 20% of the volume of the feed sample. The pH, total solid and volatile solid of the inoculums were 7.1, 9% and 80.4%, respectively. The inoculum was collected from household biogas digester in Addis Ababa city and fresh bio slurry was directly used.

Experimental set up

The study was conducted in Environmental Engineering in School of Chemical and Bio Laboratory at Addis Ababa Institution of Technology Laboratory Scale Anaerobic Digester (Figure 1). The experiments were carried on batch laboratory scale reactors at 500 ml; water was filled in the water bath to regulate the temperature of the reactors. It was operated by setting the water bath temperature separately at the desired value within the range of 20 to 45°C. The different designed temperatures are shown in Table 4. The bottles were closed by rubber stoppers equipped with glass tubes for gas removal and the glass was connected to gas line which conveys it to the gas bag. The volume of biogas generated from the reactors was measured by gas syringe and the composition of the gas was evaluated by gas analyzer.

Laboratory analysis

Characterization of the sample was made by taking fresh voided blood, manure and undigested content without mixing them in order to know the TS and VS of each type of waste. This helps to decide the amount of water to be added to the reactor to get optimal value of the substrate total solid concentration inside the reactor, which is from 7 to 10% (Yadav et al., 2014). The waste was characterized using Standard Methods (APHA, 1995): total and volatile solids (TS and VS), total fixed solids (TFS) and pH. DM was measured after drying at 105°C for 24 h, and ash after heating to 525°C for 5 h. VS was determined by subtracting the amount of ash from the amount of dry matter.

Experimental procedure for evaluation of biogas production potential from each type of waste

To know the capacity of gas production from each type of waste, single type of slaughter house waste (manure, blood and undigested content) was mixed with water. 0.51 separate reactor was used and 0.351 substrate was fed to the reactor without inoculums. The experiment was carried out at optimum temperature range of 35.6°C (regulated by water bath) and initial substrate of pH 7.5 (Hafid et al., 2011). The waste pH (at feeding time), methane composition and volume of biogas were measured by gas analyzer and gas syringe, respectively at HRT25, 35, and 45 days; and the substrate was mixed once a day to maintain intimate contact between the microorganisms and the substrate.

Table 1. Characteristics of blood, undigested stomach content and manure before digestion.

Type of Waste	TS(%)	VS/TS (%)	Ash(%)	pH
Blood	11±0.53	89.9±0.9	10.1±0.54	7.8±0.5
Manure	18.95±0.25	81.53±1.2	18.5±0.77	7.5± 0.2
Undigested Stomach Content	15.35±0.6	84.85±1.3	15.15 ± 0.5	7.01± 0.3

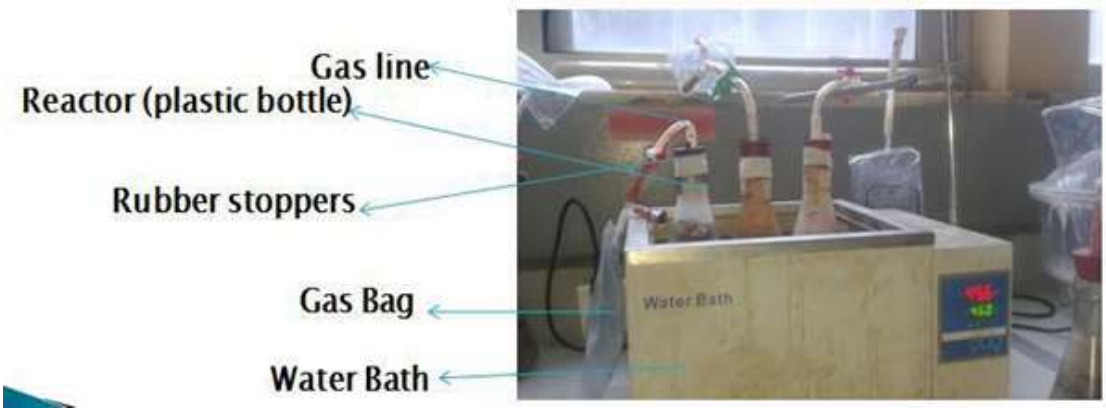

Figure 1. Laboratory scale anaerobic digester set up.

Experimental procedure for evaluation of biogas production potential from the mixture of three types of wastes

The study aimed to investigate the maximum volume of biogas and methane composition by mixing three types of wastes at different initial substrate proportions, temperature and pH adjusted using design expert software (Table 4). The reactors had 0.51 total volume and 0.41 substrate fed to it. Initial pH was adjusted at the feeding time at designed levels of 4, 5.5, 7, 8.5 and 10 by using nitric acid; sodium hydroxide was used to adjust the initial pH of the feedstock (Rodriguez et al., 2016). Volume of biogas production and methane yield was measured by gas syringe and gas analyzer, respectively.

Data analysis

Crossed D-Optimal Designs is a type of design expert software used for the experiment; it has process factors and mixture components. It is a well-accepted statistical technique used to design and optimize the experimental process; it involves choosing the optimal experimental design and estimating the effect of the several variables independently and also their interactions simultaneously (Mark, 2002). This technique was used for the optimization of methane production and volume of gas production by using the Stat-Ease software with Design Expert v.6; the experiment was applied to obtain optimum operating conditions for the factors involved. In this study, two process factors (Initial pH and temperature) and three mixture components (blood ratio, manure ratio and undigested content) were selected to study their effect on biogas production. First, maximum and minimum input value of blood, manure and undigested content mixture were given to the software with blood range (10 to 20%), manure range (10 to 20%) and undigested stomach content range (60 to 80%). Total amount of mixed substrate fed to the reactor that is 0.41 and the maximum and minimum value of the waste were decided based on the article (Medina et al., 2014) and available resource in Addis Ababa Abattoir Enterprise. According to this Crossed D-optimal design software, 42 runs of experiments were conducted.

RESULTS AND DISCUSSION

Characterization of individual raw slaughterhouse waste

Characteristics of blood, manure and undigested content (rumen) used in the study were determined and the observed results are shown in Table 1.

The total solid of blood, manure and undigested content were 11, 18.95 and 15.35%, respectively and the volatile solid of the three wastes accounts for 89.9, 81, 53 and 84.85% of the total solid, respectively. When compared with other studies, the result of this study has slight variation because of several factors which include feeding type of animal, type of on-site sanitation system, way of sampling system and amount of ageing that has taken place (Alvarez and Liden, 2008).

Biogas production potential and methane composition of each type of slaughter house waste

Major findings of the anaerobic digestion of each type of slaughterhouse waste from 0.350 L of substrate at optimal condition of 35°C and pH 7.5 (Hafid et al., 2011)

Table 2. Biogas potential and composition of methane for blood, manure and undigested stomach content.

HRT	Blood		Manure		Undigested Stomach Content	
	Volume of Biogas(lit)	Methane Composition (%)	Volume of Biogas(lit)	Methane Composition (%)	Volume of Biogas(lit)	Methane Composition (%)
25	0	0	2.2	53.4	0.8	34
35	0	0	3.2	66	2	46
45	0	0	1.6	57	1.2	54

are shown in Table 2.

The cumulative biogases produced during the digestion of the feed stocks for 45 days are presented in Table 2. From the digestion of manure, the total production of biogas was 2.51, 3.21 and 1.61 at HRT 25, 35 and 45 days, respectively from 350 ml of the substrate. The maximum methane yield from manure was 66% at HRT 35 days.

From the undigested content, the total biogas volume was 0.81, 21 and 1.21 at HRT 25, 35 and 45 days, respectively from 0.3501 of substrate. The maximum methane yield obtained from undigested content was 54% at HRT 45 days.

From blood, there was no biogas production and methane was generated at equal volume of substrate and HRT like that of manure and undigested content.

The quality and quantity of organic matter available for use in a biogas plant constitutes the basic factor of biogas generation. The volumetric yield of biogas per kilogram (kg) varies from one substrate to another depending on the composition as well as nature of the substrate. In addition, the percentage of methane obtained from the resultant biogas also varies independently according to type of biomass material (Dioha, 2013). Digestion rate of undigested content is low as compared to that of manure because undigested content is lignocellulose carbohydrate in nature which cannot be degraded easily by bacteria and is resistant to hydrolysis; hence, needs long time for digestion.

Biogas produced from blood was zero whereas manure and undigested content have good biogas potential from the same amount of substrate (0.351), because blood has naturally high amount of nitrogen. This means the C:N ratio of blood is three, which is very small. The C:N ratio decreases and inhibits gas production because ammonia accumulates inside the digester, which indirectly affects the composition of methane and retention time (Tesfaye, 2009). Proteinaceous waste products in large quantities in the AD process are not recommended due to the increased risk of inhibition by ammonia (Etelka et al., 2010). In addition to this, this is a well-known source of sulfide formation during anaerobic degradation. The increased concentration of sulfides in the digester leads to higher concentrations of corrosive H_2S in the biogas and can further lead to sulfide inhibition of the methanogens (Chen et al., 2008). However, protein, a rich energy resource, co-digests with materials with high carbon content to achieve a balanced process (WCECS, 2014). From the literature, it is seen that by nature protein has high biogas potential and a composition of 0.53 m^3/kg VS and CH_4:CO_2 is 60:40 (Kovács et al., 2010).

While manure has a good potential of biogas, which is C:N ratio 20:1; the optimal carbon/nitrogen (C/N) ratio for anaerobic bio digestion is between 20:1 and 30:1. Manure without co-digestion with other substrate has large amount of gas production, short hydraulic retention time and methane composition as compared to blood and stomach content (Table 2).

Identification of optimal mix ratio of wastes with optimal process factor (pH and temperature) for high methane production

Runs (42) were generated to determine the maximum methane yield from the mixture of blood, manure and undigested content at different ratio, temperature and pH using Crossed D-Optimal design expert software, for the optimization of biogas production. The volume of substrate solution fed to the reactor was 0.41 which was solid substrate solution (Table 3) and the selected substrate composition was manure (20%), blood (20%) and rumen (60%) (Table 5). The Laboratory result is shown in Table 4 with the experimental values of methane yield and volume of resulting biogas production.

Optimization

The numerical optimization finds a point that maximizes the desirability function. Table 5 presents the specific optimum conditions of methane composition by considering pH and temperature with the three substrate mixtures; the goal is to obtain maximum methane yield and biogas production at optimum conditions. Table 5 shows that the suitable optimum formulation (manure 20%, blood 20% and rumen 60%) was at pH 7.89 and

Table 3. Volume and solid concentration of the selected sample.

Items	Unit	Amount
Volume of Sample solution	ml	400
TS concentration of sample	gm/400ml	30
	%	7.5
Volatile solid per total solid content of sample	gm /ts	25.67
	%	84.8
Selected Sample Composition of substrate (Manure, Blood and Undigested Stomach content)	%	20,20,60

Table 4. Laboratory results of methane yield and amount of biogas Production from mixtures of blood, manure and undigested stomach content.

Run	Block	Component 1 A:Manure %	Component 2 B:Blood %	Component 3 C:Rumen %	Factor 4 D:Temp ^{o}C	Factor 5 E:pH pH	Response 1 Methane composition %	Response 1 Volume of BioGas %
1	Block 1	10	10	80	45	4	0	0
2	Block 1	10	15	75	26.25	8.5	56	7
3	Block 1	20	20	60	32.5	10	63	8.67
4	Block 1	20	10	70	45	4	0	0
5	Block 1	20	10	70	32.5	10	68.1	9
6	Block 1	20	20	60	32.5	10	64	9
7	Block 1	10	20	70	45	10	59	3.6
8	Block 1	20	15	65	20	10	48	4.2
9	Block 1	10	10	80	20	7	57	4.8
10	Block 1	10	10	80	20	10	50	5
11	Block 1	15	15	70	20	7	57	4.8
12	Block 1	10	20	70	45	10	58	3.8
13	Block 1	15	10	75	45	10	59	3.8
14	Block 1	10	20	70	20	4	0	0
15	Block 1	15	10	75	26.25	5.5	50	4
16	Block 1	10	15	75	32.5	4	0	0
17	Block 1	10	20	70	20	10	49	4.5
18	Block 1	10	10	80	32.5	7	67	10
19	Block 1	20	10	70	20	7	67	5
20	Block 1	15	10	75	38.75	5.5	50	3.8
21	Block 1	10	20	70	32.5	7	70	11

Table 4. Contd.

22	**Block 1**	**15**	**15**	**70**	**45**	**10**	**57**	**2.8**
23	Block 1	10	10	80	45	10	56	3
24	Block 1	20	10	70	20	10	50	5
26	Block 1	10	20	70	45	7	60	6.4
27	Block 1	15	15	70	45	10	74.2	12
28	Block 1	20	15	65	20	4	0	0
29	Block 1	20	20	60	32.5	4	0	0
30	Block 1	20	20	60	45	7	58	6
31	Block 1	20	20	60	45	10	50	2.6
32	Block 1	20	20	60	45	7	57	5.88
33	Block 1	20	15	65	45	4	0	0
34	Block 1	10	15	75	32.5	4	0	0
35	Block 1	15	20	65	32.5	7	69	9.8
36	Block 1	15	20	65	20	4	0	0
37	Block 1	20	20	60	45	4	0	0
38	Block 1	20	20	60	20	4	0	0
39	Block 1	20	10	70	45	10	59.4	3.5
40	Block 1	20	20	60	45	4	0	0
41	Block 1	10	10	80	20	4	0	0
42	Block 1	15	10	75	20	10	49	4.5

Table 5. Optimization conditions and desirability of model.

Manure	Blood	Rumen	Temp.	pH	Methane Composition	Volume of Biogas	Desirability	
20	20	60	32.49	7.88	79.26	9.792	0.914	**Selected**
20	20	60	32.40	7.88	79.25	9.791	0.914	
20	20	60	32.62	7.87	79.23	9.791	0.914	
18.67	20	61.33	30.92	7.58	76.30	9.65	0.908	
17.22	20	62.78	31.14	7.56	74.20	9.60	0.908	

temperature of 32.31°C giving the highest value of 0.914 for methane composition and volume of biogas per feeding selected.

Optimum temperature

This study was conducted at room, mesophlic and thermophlic temperature ranges; the minimal and maximal temperature value was from 20 to 45°C based on the Crossed D-Optimal Design Expert

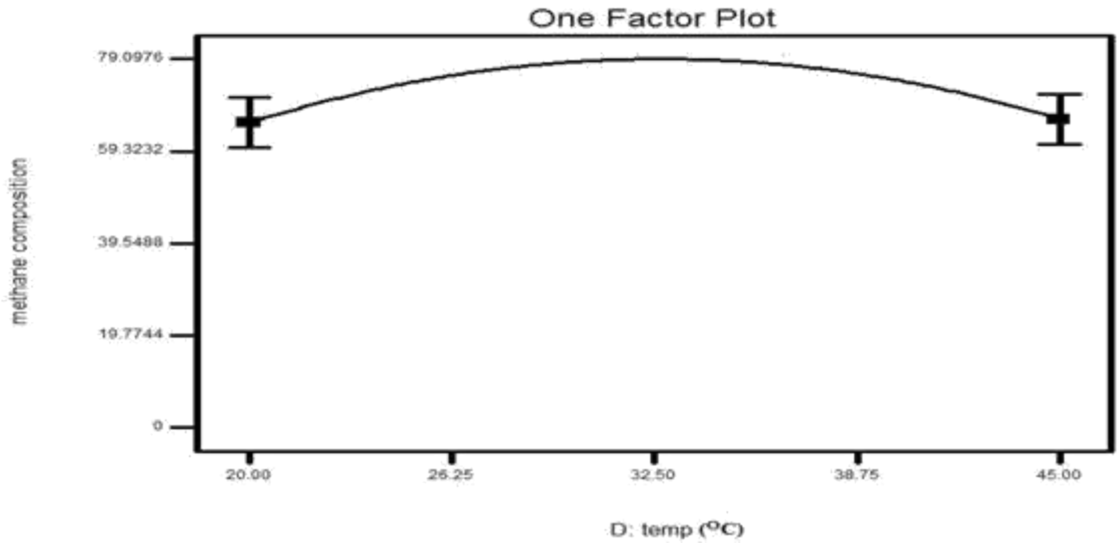

Figure 2. Methane yield versus temperature at selected pH and component of mixture.

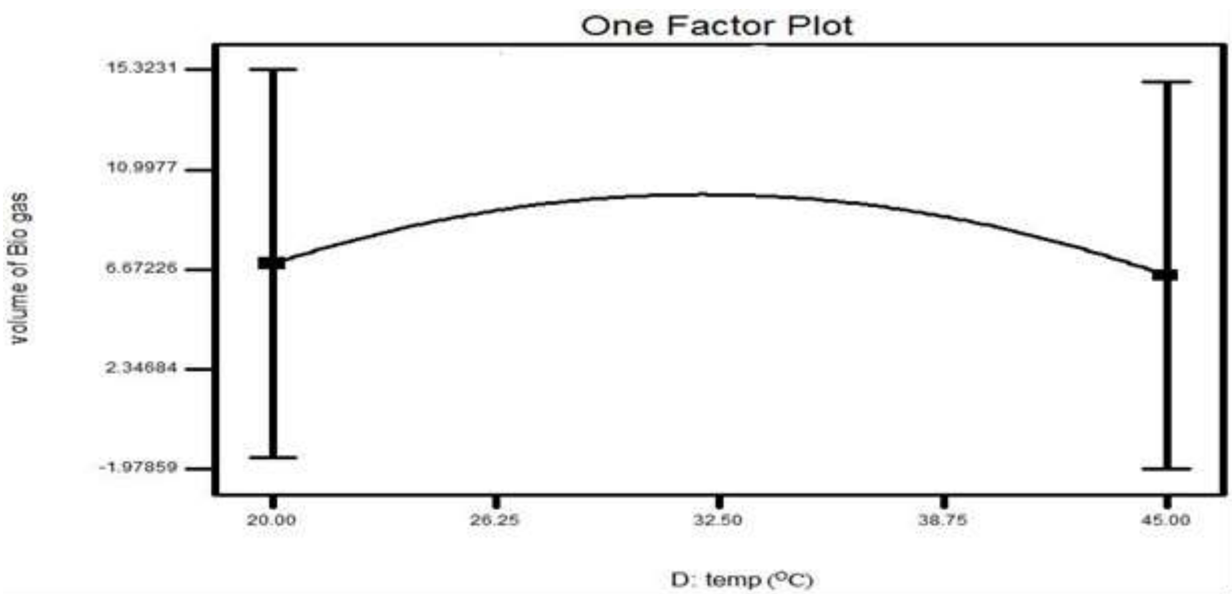

Figure 3. Volume of Bio gas versus temperature at selected pH and component of mixture.

software which was designed at 20, 26.25, 32.5, 38.5 and 45°C. Methane yield at this temperature point was 65.56, 76.1, 79.2, 76.10 and 66.41%, respectively (Figure 4). Volume of biogas and methane composition at controlled pH and optimal mixture of substrate are as shown in Figures 2 and 3.

In this study, the optimal methane composition was found at 32.5°C temperature and methane yield of 79.25%; in other words, this temperature range is that of the temperature range for mesophlic which is the recommended range in literatures. Tchobanoglous et al. (2003) pointed out that reactor temperatures between 25 and 35°C are generally the preferred optimal to support biological-reaction rates and provide a more stable treatment.

When the temperature was set at room temperature (20°C), the volume of biogas and methane composition decreased as shown in Figure 2. Tchobanoglous et al.

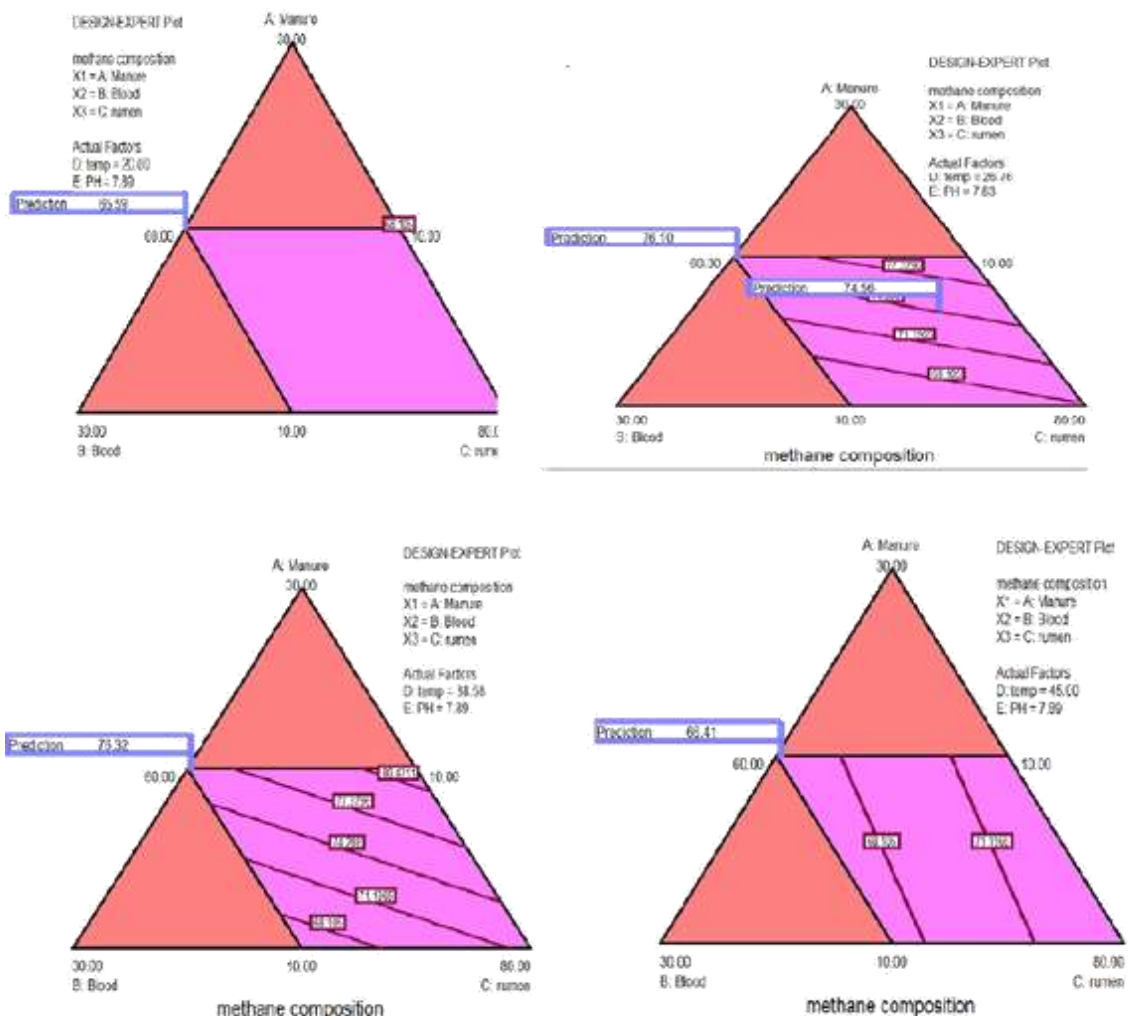

Figure 4. Methane Composition at different temperature and at selected pH and at different Substrate composition.

(2003) reported that temperature ranges approaching 20°C are not suitable for anaerobic digestion and it takes long HRT for digestion and low gas production because the degradation of long-chain fatty-acids is often limiting. If long-chain fatty-acids accumulate, foaming may occur in the reactor and so inhibit the process (Appels et al., 2008).

Temperature within the range of 26 to 38°C has better methane composition in the biogas as compared to temperature lower than 26°C and temperature greater than 38°C (Figure 2). When temperature increased to 45°C, bio gas was produced in a short period of time but the amount of biogas product and methane composition was not as much as that of 26 to 38°C, because the rate of decomposition and gas production is sensitive to temperature. In general, the process becomes more rapid at high temperatures; whereas the digestion process becomes increasingly unstable with rising temperature and requires higher rates of heat inputs; it produces poorer-quality supernatant containing larger quantities of dissolved solids (Tchobanoglous et al., 1991).

When the temperature was reduced to 20°C or increased to 45°C, the efficiency of the treatment decreased markedly because fatty acid formation and NH_3 accumulation inside the digester increased consequently inhibiting gas formation (Appels et al., 2008). Hence, temperature affects the success of the digestion process because the activities of the anaerobe causing waste decomposition are temperature dependent.

Optimal initial pH of substrate

The pH considered in the study was initial pH of substrate when fed to the reactor from acidic to basic, from pH 4 to

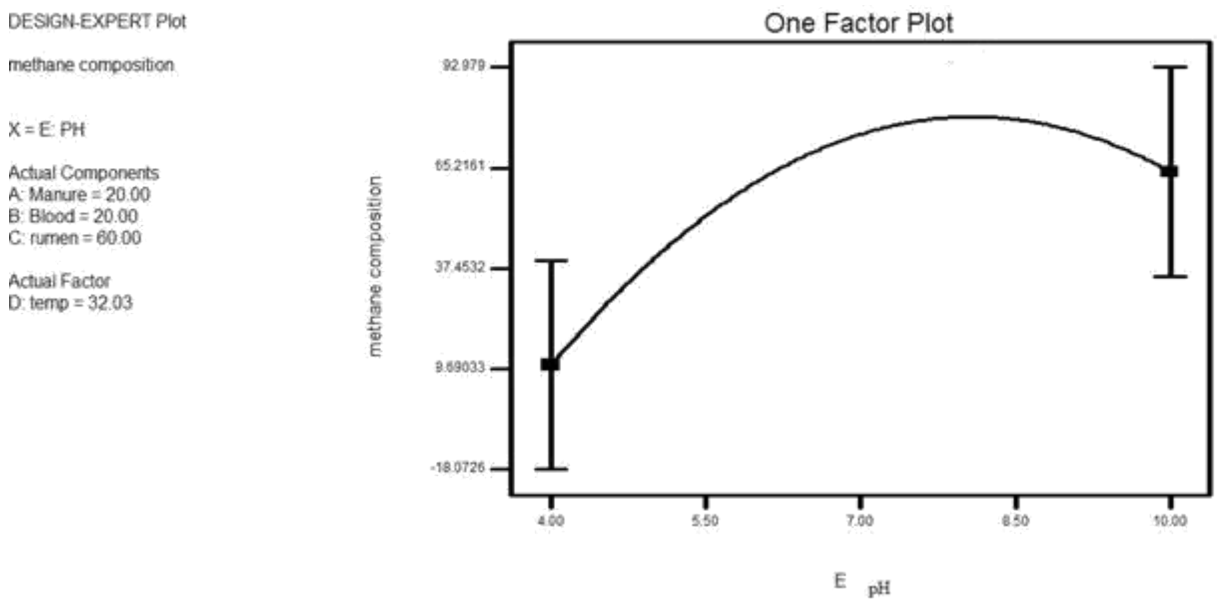

Figure 5. Methane yield versus pH at selected temperature and component of mixture.

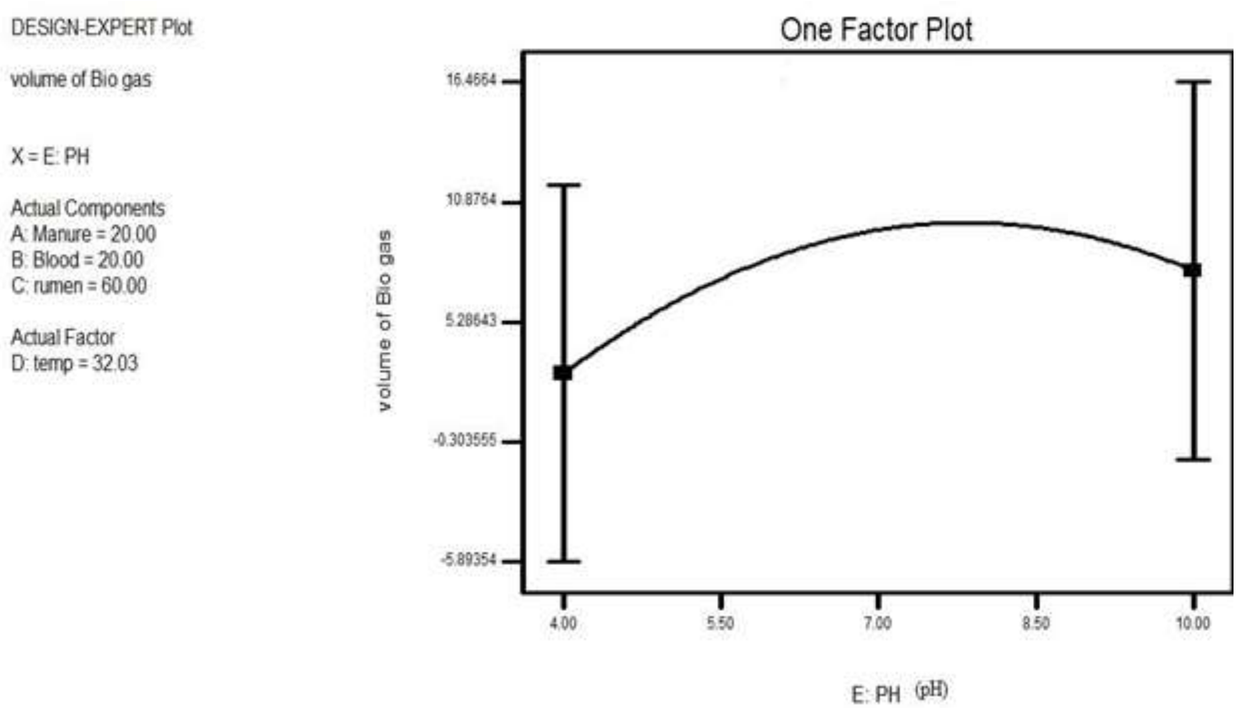

Figure 6. Volume of biogas versus pH at selected constant Temperature and mixture of component.

10. The output value of the pH design of the Crossed D-Optimal Design expert software for experiment gave five pH levels: 4, 5.5, 7, 8.5 and 10; and methane yield at these pH levels was 10, 52.2, 74.6, 78.44 and 64%, respectively (Figure 7). Volume of biogas and methane composition at controlled temperature and optimal mixture of substrate is explained in Figures 5 and 6.

From pH 7.5 to 8.5, best gas productions and methane

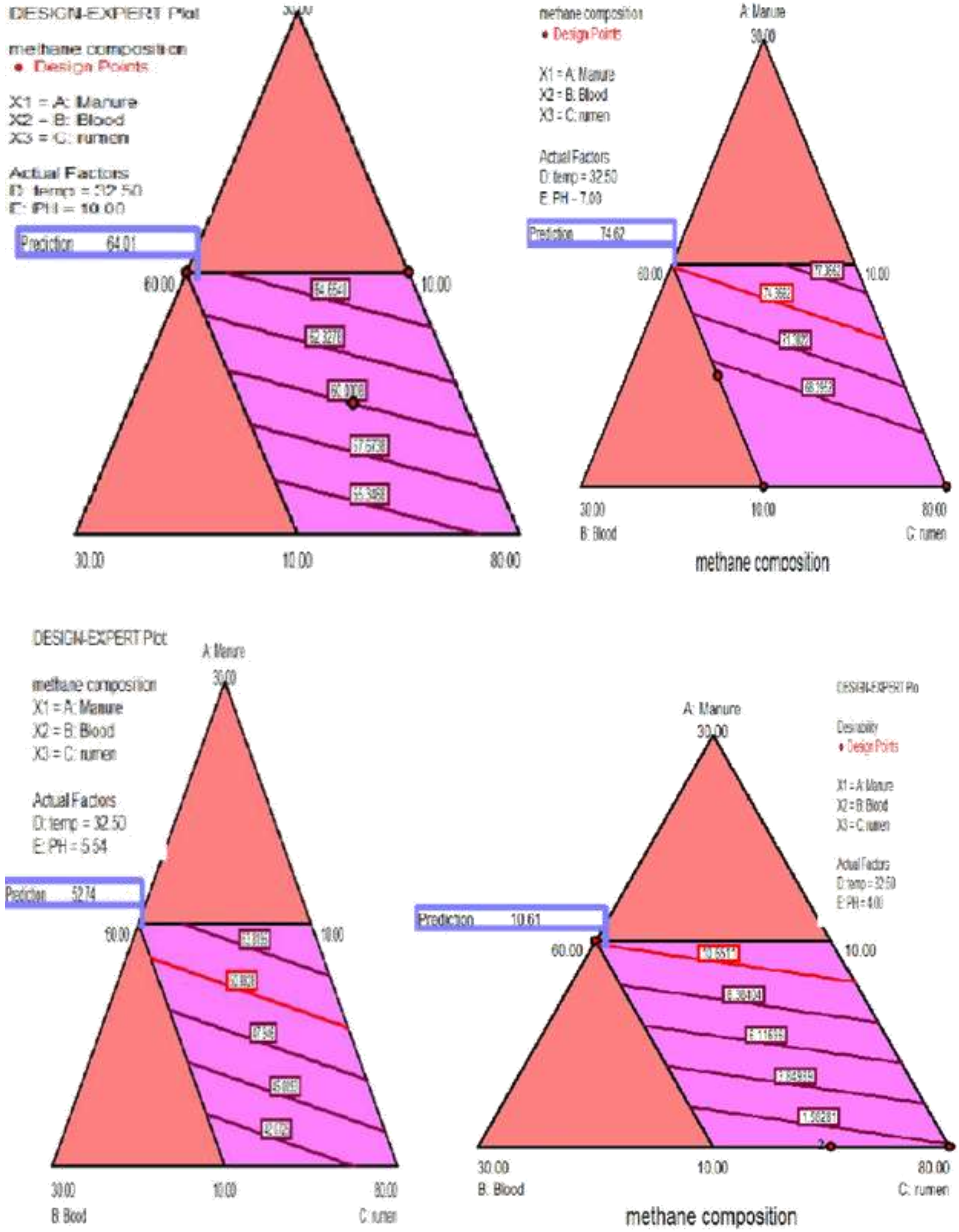

Figure 7. Desirability and methane composition at selected temperature and different pH.

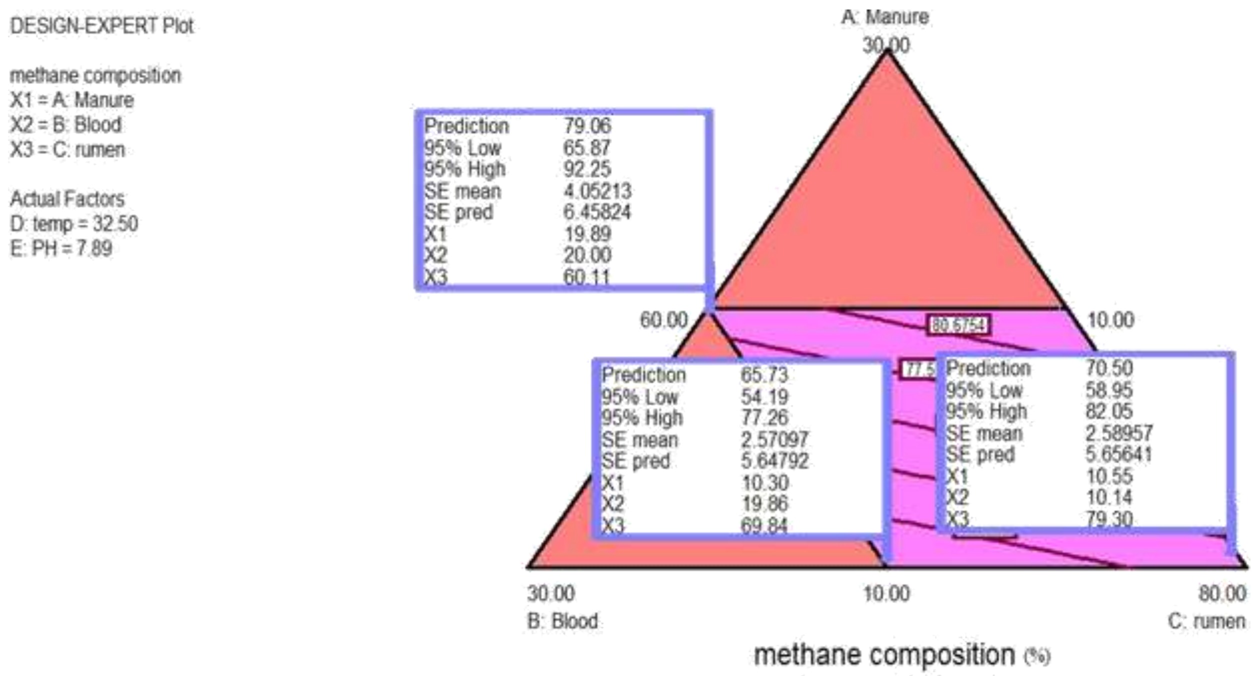

Figure 8. Methane yield at different substrate composition.

yield were obtained; in this study, the optimal pH selected was 7.88 as shown in Table 5, with best desirability value. In another study, pH 7.5 to 8 also showed excellent performance because 85% of the total chemical oxygen demand was removed in anaerobic digester and a short hydraulic retention time (Appels et al., 2008).

When pH was up to 8.5 to 10, the amount of biogas decreased from the peak value but not like when the pH approached 4, because to adjust the substrate pH from 8.5 to 10, NaOH was used. Liew et al. (2011) carried out simultaneous solid-state pretreatment using NaOH on fallen leaves; the methane yield increased by 20% during batch tests. It is demonstrated that alkali pretreatment can increase gas yield from lingo-cellulose rich substrates. Carlsson et al. (2012) reported that lime or NaOH was used for treatment of lingo-cellulosic feedstock materials which are resistant to hydrolysis due to their structure and composition. So in this work, our feedstock (slaughter house waste) had undigested content which was lignocellulose material and batch type fermentation was the reason for obtaining good methane composition.

When initial pH was set near 7, there was good biogas production and methane composition with good desirability value (Figure 7). The bacteria involved in anaerobic digestion have a pH range close to 7 for optimal activity (HilkiahIgoni et al., 2008).

When initial pH value was below or equal to 4, there was no biogas production as shown in Figures 5 and 6. HilkiahIgoni et al. (2008) said that the hydrogen-ion concentration of the culture medium has a direct influence on microbial growth because the digestion is inhibited by excessive acidity. In literature, a decrease of pH to 6 and below caused a strong reduction of COD removal and biogas production, with a simultaneous accumulation of volatile fatty acids and ammonia (Appels et al., 2008).

Optimal substrate mixture component

Based on the 42 runs and different experimental results (Figure 8) using the crossed D-Optimal Design Expert software, 20% blood, 20% manure and 60% undigested content were selected as the optimal substrate composition (Table 5); the counter plot in Figure 8 reported the methane yield and amount of biogas production at optimal temperature and pH at different substrate proportion.

In Figures 8 and 9, when approximate value of the first proportion was set at 19.89% manure, 20% blood and 60.11% undigested content volume, the volume of biogas production and methane yield were 9.91 L and 79.25%, respectively. From the second proportion (10.3% manure, 19.86% blood and 69.84%) biogas production and methane yield were 9.3 L and 65.7%, respectively. And from the third proportion (10.55% manure, 10.14% blood, and 79.3% undigested content), volume of biogas

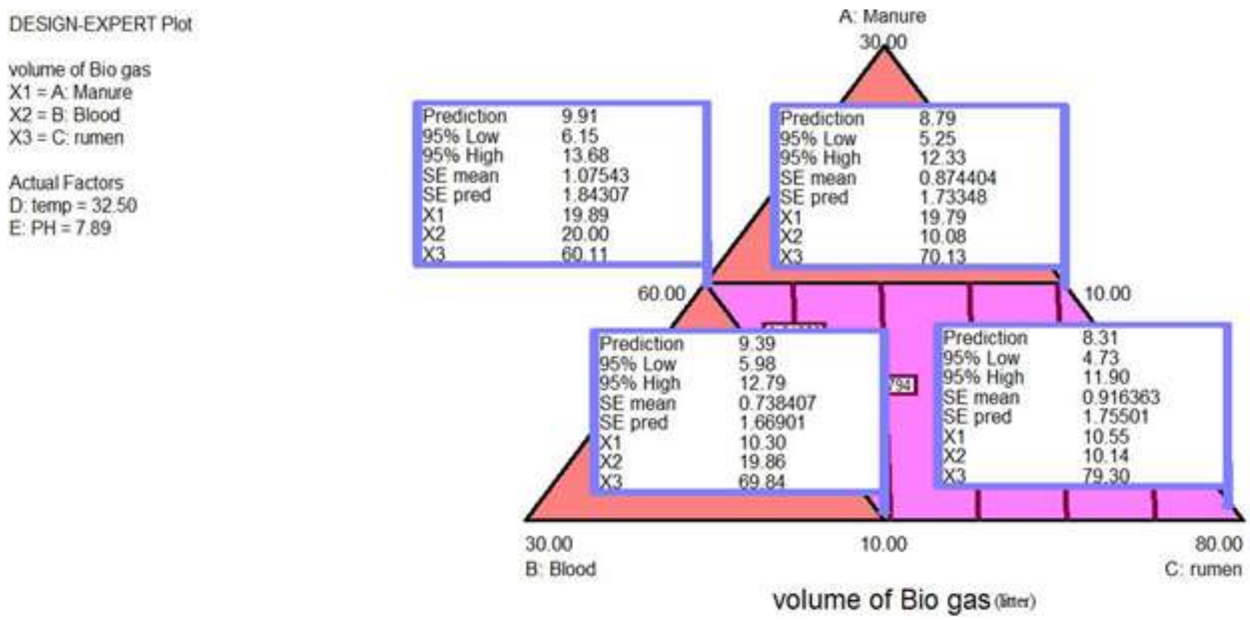

Figure 9. Amount of biogas produced at different substrate.

production and methane yield were 8.3 L and 70.5%, respectively.

The first proportion had better methane yield and volume of biogas than the second and third proportions. The composition of methane yield reduced with decreasing amount of manure (from 20 to 10 %). Mixture having more blood than manure made the methane yield to decrease by 65.5%. This implies too much blood as substrate means low total solid content and mainly protein compounds have low biodegradability. This means increasing biodegradability will give higher methane yield by co-digesting it with other easily biodegradable substrates like carbohydrates.

The third was better than the second as its methane yield increased because it has high amount of undigested content from 70 to 80% which is a lingo-cellulose and a very common carbohydrate plant. The methane yield increased from 65.5 to 70.5%, because cellulose compound has the capacity to minimize the negative impact of protein substrate (blood) inside the digester (Figure 9).

Conclusion

Biogas production from slaughter house waste blood alone is so difficult; it must be mixed with another organic waste. The best and straightforward organic waste in a slaughterhouse is manure and undigested content from the stomach of dead animals and has higher amount of anaerobic bacteria; hence, blood mixed with manure and undigested content at optimal proportion can produce attractive amount of biogas, but the amount of methane yield decreases when concentration of blood is higher than that of manure.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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