



# **Characterization and Ranking of Various Mix Ratios of Cow, Pig and Sheep Manure**

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## **Authors' contributions**

*This work was carried out in collaboration among all authors. Author SCM designed the study, managed the literature searches, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors DMN and BOO managed the analyses of the study. All authors read and approved the final manuscript.*

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## **ABSTRACT**

This work determined the characteristics of various mix ratios of the cow to pig to sheep manures and ranked them with help of principal component analysis (PCA). Ten mix ratios (by mass) namely 1:1:1, 3:1:1, 1:3:1, 1:1:2, 2:1:1, 1:2:1, 1:1:2, 1:3:3, 3:3:1, 3:1:3 of cow, pig and sheep manures respectively were selected. Laboratory analysis was done to determine the total solid (TS) content, carbon to nitrogen ratio, pH, and volatile solid (VS) content using standard procedures. The results obtained (except that of pure feedstocks) were subjected to principal component analysis to determine the principal component scores for the mix ratios to enable ranking. The total solids content of pure cow, pig, and sheep manure were found to be 19.18%, 23.50%, and 30.35% respectively. Corresponding carbon to nitrogen ratios values were 23.68, 13.27 and 29.00, pH values were 6.50, 7.90 and 7.00 and volatile content were 88.37%, 84.57% and 80.00%. Upon mixing the three manures at various mix ratios total solid content varies from 22.28% to 26.75%. Total solids content, carbon to nitrogen ratio, pH and volatile solids content varies from 22.28% to

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26.75%, 18.76 to 25.05, 7.13 to 7.56 and 85.94% to 82.59% respectively. Using the first principal component scores mix ratio 1:1:3 of cow dung, pig, and sheep manure was the top-ranked followed by 3:1:3 and the third one was 1:1:2 with scores of 2.540, 1.638, and 1.580 respectively. The 4<sup>th</sup> ranked mix ratio was 1:3:3, 5<sup>th</sup> ranked was 1:1:1, 6<sup>th</sup> ranked was 2:1:1, the 7<sup>th</sup> one was 3:1:1, 8<sup>th</sup> one was 1:1:2, then 3:1:1 and lastly 1:3:1 with the scores of 0.191, -0.006, -0.147, -0.259, -1.440, -1.810 and -2.287 respectively. Higher positive scores were associated with a possibility of producing higher biogas yield possibly due to the right combination of the several parameters in the mixture while a lower score might indicate a lower gas yield due to an improper combination of parameters. It was then concluded that principal component analysis is a suitable method for selecting few mix ratios to use in anaerobic digestion among the many. It saves on time and resources due to the reduced number of experiments.

**Keywords:** *Mix ratios; total solids; volatile solids, pH; anaerobic digestion; principal component analysis.*

## 1. INTRODUCTION

Various feedstock for anaerobic digestion includes an organic fraction of municipal solid waste (OFMSW), sewage sludge, grass clippings/garden waste, food remains manure (cattle, pig, poultry), energy crops, algal biomass, harvest remains, and waste from food or beverage processing, dairy, starch industry, sugar industry, pharmaceutical industry, cosmetic industry, biochemical industry, pulp and paper, slaughterhouse/rendering plant [1]. Various anaerobic digestion feedstocks considerably vary in qualitative and quantitative composition, homogeneity, fluid dynamics, and biodegradability as evident in Table 1.

Anaerobic digestion is affected by different factors namely pH, temperature, retention time, particle size, total solid content, volatile solid content, inoculums, dry matter content, and organic loading rate [20]. Retention time is the time needed for the complete depletion of the substrate to be attained. It depends on the feedstock's composition, temperature, pH, and particle size. The higher the total solid content in the feed the higher the retention time, favorable temperature ranges decrease the retention time and the smaller the particles size the shorter the retention time due to high reaction rates [20]. The shorter the retention time the greater the bacteria

washed out in the digester than their replication. Approximate values of retention time of liquid cow manure, liquid pig manure, and liquid chicken manure range from 20-30, 15-25, and 50-80 respectively [21]. This shows that anaerobic digestion of different mix ratios is time-consuming and expensive. Hence, the need of using tools that reduces the number of anaerobic digestion experiments such as principal component analysis (PCA).

The decisions on the ratio between feedstocks have been associated with optimization of carbon to nitrogen ratio and also the right combination of the several other parameters in the mixture such as macro and micronutrients, pH and alkalinity, inhibitors and toxic compounds, biodegradable organic and dry matter [22]. Hence, the need for mix ratios analysis to proof this philosophy by checking if biophysical characteristics fall within the recommended range or not.

PCA is a multivariate technique that analyzes data described by several inter-correlated dependent variables to extract the important information to represent it as a set of new orthogonal variables called principal components. It is mostly used as a tool in exploratory data analysis and for developing predictive models [23].

**Table 1. Feedstock characteristics**

Animal manure	pH	TS (%)	VS (%)	C/N ratio	Reference
Cattle manure	7.1-8.6	14.5-22.7	11.9-72.0	14.59-18.9	[2-6]
Pig manure	6.4-7.5	8.2-36.7	6.2-82.8	5.7-13.5	[5,7-12]
Chicken manure	6.9-7.4	20.0-92.6	18.3-84.1	7.5-9.75	[5,2,13-16]
Sheep manure	7.16-8.1	22.3-40.0	18.7-72.7	11.3-14.7	[2,3,17]
Goat manure	7.9	33.7-55.5	27.7-89.4	18.0	[3,17,18]
Donkey manure	6.8	19.8	14.4	-	[19]

According to Reiris and Brooks [24] applications of PCA includes dimensionality reduction, principal component regression, ranking, and total least squares regression. It has been used for the description and visualization of the parameters under study [25], to distinguish instability periods and also different process performance or waste compositions [26], to monitor industrial processes [27], and in wastewater treatment processes [28].

PCA has been used in anaerobic digestion of various organic matter. For example, Nikiema et al. [29] used the tool to reduce geometric space and visualize data by the use of a linear combination of variables that maximizes variance. Leite et al. [30] used it to compare single and two-stage anaerobic digestion (AD) process performances when treating waste-activated sludge (WAS) to increase their monitoring and control. From the literature, PCA has not been used to rank various mix ratios of anaerobic digestion feedstocks. Hence, the research utilizes the tool to rank various mix ratios of the cow to pig to sheep means to select a few among the many for anaerobic digestion tests to save on time and resources.

## 2. MATERIALS AND METHODS

### 2.1 Selection of the Mix Ratios

Cow, pig, and sheep manures were chosen to be used in the research as the feedstocks for digestion due to their abundance availability at Tatton Farm, Egerton University. The manures mixed at varying ratios based on the recommendations by Levi and Dorothy [31] that ratio of 1:1 of pig manure to cow dung and by Ngunjiri et al. [32] that ratio of 7:3 (approximately 2:1) sheep manure to cow manure that gave the highest biogas yield. Ten mix ratios (by mass) namely 1:1:1, 3:1:1, 1:3:1, 1:1:2, 2:1:1, 1:2:1, 1:1:2, 1:3:3, 3:3:1, 3:1:3 of cow, pig and sheep manures respectively were selected to cover the range suggested by the two researchers with the three manures.

### 2.2 Characterization of the Mix Ratios

Laboratory analysis was done to determine the biophysical characteristics of the ten mix ratios of cow dung, pig manure, and sheep manures. The total solid (TS), pH, and volatile solid (VS) were determined according to APHA standard [33]. Total nitrogen was estimated by Kjeldahl method [34] and the carbon content was determined by

Walkey black method. The carbon to nitrogen ratio was determined by dividing the results of carbon content by that of nitrogen. Results are in Table 2.

### 2.3 Principle Component Analysis (PCA)

A MATLAB R2013a (8.1.0.604) mathematical software was used to determine the principal component loadings(coefficients), respective latent, scores and scree plot of the input variables.

#### 2.3.1 Principal component analysis Matlab script

##### a) Principle component coefficients, scores and latents

```
%A represent measurements taken of the
different mix ratio
>> A= [TS C/N Ph VS];
% determination of the size of the data
>> [n m];
% calculation of mean
>> A mean = mean (A);
% calculation of standard deviation
>> A std = std (A)
% standardization which means subtracting the
sample mean from each observation then
dividing by standard deviation
>> B =Z score (A);
% determining the coefficients, respective latent
and scores
% `Coffecient` principal component vectors
% `latent` eigenvalues of covariance matrix of A
arranged in order
% `score` projection of the original data onto the
principal component axis
>> [COEFF SCORE LATENT] = princomp (B)
```

##### b) Scree plotting script

```
A= [TS C/N Ph VS];
Amean=mean(A);
Astd=std(A);
B=zscore(A);
[Coefs, Score, latent] = pca(B);
explained=cumsum(latent) ./sum(latent);
scree(explained);
xlabel ('principal component');
ylabel ('Variance Explained (%)')
The scores obtained were used to rank the
different mix ratios. The results are presented in
Tables 3 and 4. Scree plot help to select the
principal component to be used (Fig. 1).
```

### 3. RESULTS AND DISCUSSION

#### 3.1 Characterization of the Mix Ratios

Characteristics of various mix ratios of cow dung, pig manure, and sheep manure are summarized in Table 2.

From Table 2, total solids content obtained for the pure cow, pig, and sheep manures were 19.18%, 23.50%, and 30.35% respectively. Upon mixing the three feedstocks at various mix ratios total solid content varies from 22.28% to 26.75%. The recommended value for biogas digester slurry is between 8% to 12% [35]. Thus, the manures were diluted with water to the required consistency before being introduced into the digester.

The C/N ratio of pure cow, pig, and sheep manure obtained was 23.68%, 13.27%, and 29.00% respectively and their corresponding pH values were 6.50, 7.90, and 7.00. Mixing the three manures at various mix ratios causes the carbon to nitrogen ratio to range from 18.76% to 25.05%. Corresponding pH values also vary from 7.13 to 7.56.

The volatile content obtained were 88.37%, 84.57%, and 80.00% of pure cow, pig, and sheep manure respectively. It's evident that mixing the three feedstocks at varied mix ratios also results in varied VS content. The values obtained ranged from 85.94% to 82.59%.

The decisions on the ratio between feedstocks have been associated with optimization of carbon to nitrogen ratio and also the right combination of the several other parameters in the mixture such as macro and micronutrients, pH and alkalinity,

inhibitors and toxic compounds, biodegradable organic and dry matter [36]. This agrees with the results presented in Table 2. Mixing cow, pig, and sheep manure at varying mix ratios enable their C/N ratios to fall within the recommended range which is between 20:1 and 30:1 [37] except that of mix ratio 1:3:1 (C/N ratio 18.76). Their pH also varied within a tolerable range for methane formation which is from 5.5 to 8.5 [38].

#### 3.2 Principal Component Analysis

The total solids, carbon/nitrogen ratios, pH, and volatile solids presented in Table 2 (except that of pure feedstocks) were subjected to principal component analysis to determine the principal component scores for the mix ratios to enable ranking.

Table 3 shows the obtained principal component coefficients for the four principal components.

Table 4 shows the scores for the various mix ratios of cow manure pig manure and sheep manure

Scree plotting Fig. 1 helps to select the component to be used and the ones to be left out. The first two components were obtained to account for 99% of the variance.

Fig. 1 shows that PC<sub>1</sub> alone accounts for 91% and was obtained to be suitable for analysis leaving out the 2<sup>nd</sup>, 3<sup>rd</sup>, and 4<sup>th</sup> principal components. This agrees with what Reris and Brooks [24] explained about the first PC as the direction along which the measurements are most varied in the variables that are most heavily loaded.

**Table 2. Characteristics of different mix ratios of un-digested manure**

Mix ratio	Total solid content (%)	Carbon/Nitrogen ratio	Ph	Volatile solid content (%)
1:0:0	19.18	23.68	6.50	88.37
0:1:0	23.50	13.27	7.90	84.57
0:0:1	30.35	29.00	7.00	80.00
1:1:1	24.34	22.42	7.13	84.31
3:1:1	22.28	23.45	7.57	85.94
1:3:1	24.01	18.76	7.19	82.59
1:1:3	26.75	25.05	7.26	84.42
2:1:1	23.05	23.07	7.40	85.33
1:2:1	24.13	20.13	7.26	83.24
1:1:2	25.85	24.07	7.30	84.38
1:3:3	25.82	21.68	7.30	83.15
3:3:1	22.63	20.54	7.29	84.24
3:1:3	24.58	25.04	7.30	85.55

**Table 3. Principal components'(PC) coefficients**

Characteristic	PC <sub>1</sub>	PC <sub>2</sub>	PC <sub>3</sub>	PC <sub>4</sub>
TS	0.430	-0.597	-0.558	-0.384
C/N	0.593	0.291	-0.322	0.679
Ph	0.613	-0.210	0.757	-0.087
VS	0.297	0.717	-0.113	-0.620

**Table 4. Principal components'(PC)scores**

Mix ratio	PC <sub>1</sub> scores	PC <sub>2</sub> scores	PC <sub>3</sub> scores	PC <sub>4</sub> scores
1:1:1	-0.006	-0.001	-0.002	0.004
3:1:1	-0.259	2.182	-0.010	-0.001
1:3:1	-2.287	-1.260	-0.003	-0.001
1:1:3	2.539	-0.919	-0.002	-0.003
2:1:1	-0.147	1.361	0.013	0.000
1:2:1	-1.440	-0.781	-0.011	-0.000
1:1:2	1.580	-0.572	-0.014	0.001
1:3:3	0.191	-1.566	0.018	-0.0001
3:3:1	-1.810	0.653	0.005	-0.001
3:1:3	1.638	0.902	0.007	0.000



**Fig. 1. Scree plot**

Hence, Table 4 shows that the mix ratio 1:1:3 of cow manure, pig, and sheep manure was the top-ranked followed by 3:1:3 and the third one was 1:1:2 with scores of 2.540, 1.638, and 1.580 respectively. The 4<sup>th</sup> ranked mix ratio was 1:3:3, 5<sup>th</sup> ranked was 1:1:1, 6<sup>th</sup> ranked was 2:1:1, the 7<sup>th</sup> one was 3:1:1, 8<sup>th</sup> one was 1:1:2, then 3:1:1 and lastly 1:3:1 with the scores of 0.191, -0.006, -0.147, -0.259, -1.440, -1.810 and -2.287 respectively.

A higher positive score indicates a higher gas yield possibly due to the right combination of the several other parameters in the mixture such as

macro and micronutrients, pH and alkalinity, inhibitors and toxic compounds, biodegradable organic and dry matter while a lower score might indicate a lower gas yield.

#### 4. CONCLUSION

Upon mixing cow dung, pig manure and sheep manure at various mix ratios, the total solids content, carbon to nitrogen ratio, pH, and volatile solids content varies from 22.28% to 26.75%, 18.76% to 25.05%, 7.13 to 7.567, and 85.94% to 82.59% respectively. Principal component analysis is a suitable method for selecting few

mix ratios to use in anaerobic digestion among the many. Mix ratio 1:1:3, 3:1:3 and 1:1:2 with scores of 2.540, 1.638, and 1.580 respectively were chosen. This ensures reduction in the number of experiments to run and hence saves on time and reduces the resources used.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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