Review Paper

Biogas Production from Co-digestion of Substrates: A Review

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Abstract

Sustainable development is the projected demand of all nations at present. Only 20 percent of world's primary energy requirement is met by renewable sources like solar and wind energy, hydropower, biomass, municipal and agri-wastes. Especially energy recovery from municipal and agri – wastes have gained importance due to two – fold reason: i. waste volume reduction, ii. energy recovery. The present review article focuses onto detailed aspects of some exhaustive research work in the field of energy generation by co-anaerobic digestion of several potential organic sources with cattle manure. Co-digestion of substrates have been preferred over mono-digestion due to several benefits associated with it. Carbon to nitrogen (C/N) ratio has been identified as the key parameter for improving the digestion of substrates. The average C/N ratio of 20 – 30 has been stated as optimum for maximum yield of biogas and corresponding methane in it by almost all workers referenced below. Mostly, specific methane production, ultimate methane production, methane production rate has been determined for evaluating the co-digestion process. Improvement in C/N ratio, higher bio-degradability, effective volatile solids (VS) removal, eco-friendly sludge production has been regarded as merits of co-digestion process.

Keywords: Sustainable development, Energy recovery, Renewable sources, Co-digestion, C/N ratio, Biogas, Methane, Biodegradability, Volatile solids, Sludge.

Introduction

Global prosperity and human development has been driven by energy, which has been identified as an essential requirement of mankind. Globally energy is a key factor for running household, commercial and industrial activities. Dependency conventional sources like coal, petroleum etc as primary source of energy, led to the ecological imbalance, climatic alterations, health hazards and degradation of natural resources¹. More than 80% of global energy consumption emerges from the combustion of conventional sources². Rapid increase in population and their substantial burning of fossil fuels have contributed to increase in global warming due to green house gas (GHG) emissions³. Hence, renewable sources of energy can be a key option as a potential substitute over fossil fuels. Energy recovery from biomass at a large scale without affecting environment and human activity has been encouraged⁴.

One such well known and widely used method for bioconversion of wastes into fuel is anaerobic digestion (AD), which is regarded as the simplest technique due to its very limited environmental impact $^{5-8}$ and high energy recovery potential 5,6,9 . Though extensive research has been conducted on bio-gasification of wastes by AD process, further research is currently in progress in order to enhance the biogas generation, achieve faster degradation rates and decrease the amount of ultimate leftovers to be disposed 6,7,10 . Ideally biogas is collectively CH_4 and CO_2 in the ratio of 3:1, with certain

contaminant remains of SO₂, NO₂ etc. CH₄ is a potent GHG among other gaseous fractions in biogas and henceforth is a notable contributor of global warming. So, bio-gasification of organic content serves both the purposes: i. waste volume reduction and waste treatment, ii. biogas generation as alternative and renewable energy source. The remains of waste treatment after AD process acts as superior manure that can be easily utilized in agricultural lands as a substitute for fertilizer. Thus, AD process is a promising method of volatile solid conversion to gaseous fuel and manure as degraded by-product thereby solving ecological and agrochemical issues.

AD process is dependent on specific microbial consortium for degradation of biomass through four main stages namely hydrolysis, acidogenesis, acetogenesis and methanogenesis. Researchers have reported that methanogenesis is the rate limiting step for easily biodegradable substances whereas hydrolysis is the rate limiting step for complex organic wastes due to formation of toxic by-products like complex heterocyclic compounds and un-desirable volatile fatty acids. Biogasification process is highly dependent on environmental conditions¹¹ like pH, temperature, C/N ratio, C/P ratio, size of particle, inhibitors and kind of substrate being utilized to recover energy from it. Optimum combination of operating process parameters are needed to improve the inconveniences and improve process efficiency. The substrates belong to different classes and have been categorized into further

subclasses on the basis of their proximate and ultimate properties and some of them have been summarized in Table-1 and 2 respectively.

Further, better volatile solid removal has been achieved by practicing co-fermentation also known as co-digestion that increases the load of mixed nutrients and accelerates the breakdown of macro-molecules in substrates by bio-stimulation studied during the last 15 - 20 years 12-16. Co-digestion is a simple approach of AD by mixing wastes together in different ratio and proportion keeping the C/N ratio within the desired range of 25 - 30. Particularly mixing organic substrates have resulted in the production a mixture with C/N ratio in the optimal range of $20 - 30^{17}$. Higher yield of biogas by codigestion is associated with the synergistic effect of the microorganisms¹⁸. Hence, co-digestion is preferred over AD of waste alone due to its several advantages 19,20 like: i. dilution of toxic components present in any one of the substrate has high associated toxicity; ii. improves carbon to nitrogen ratio of substrate; iii. sludge of superior quality is produced; iv. pH adjustment; v. adjustment of moisture content^{8,18}. The mentioned benefits have a positive effect on improved stability and performance of the process and leads to higher biogas yield and energy contribution. Optimum operational conditions for co-digestion, in terms of percentages of substrates cannot be defined as a standard but further investigations are needed to reach a benchmark for desired results in terms of biogas yield^{8,21-25}. Therefore, this paper aims to review the detailed aspects of co-digestion of substrates for energy generation.

Substrates for co-digestion: All kinds of biomass composed of macro-nutrients like carbohydrates, proteins, lipids, cellulose and hemicelluloses are suitable for bio-gasification. Theoritical yield of biogas varies largely on the presence of three major macro-nutrients: i. lipids, ii. carbohydrates and iii. proteins. Apart from this categorization ligno-cellulosic wastes have been studied widely for co-digestion process; the major nutrient present in these wastes is cellulose and hemicelluloses. The biodegradability of these nutrients vary largely. Lipids are the highest contributors of biogas with longer hydraulic retention time due to retarded bio-degradability, whereas proteins and carbohydrates exhibit quicker conversion rates with lower biogas yield.

Lipids are generally classified as solids (waxes and greases), liquids (oils) and fats found in dairy wastes, slaughter-house remains, oil-refineries etc²⁶. Lipids posses higher number of C and H atoms in their molecule and this makes them the source of attraction for higher methane potential but at the same time they appear to be inhibitory to methanogenic activity and lead to sludge floatation by getting adsorbed onto biomass²⁷.

Carbohydrates are the main contents of organic wastes from agricultural sector, food processing industries and source-sorted organic fractions of municipal garbage. These wastes are highly prone to formation of organic acids upon fermentation, leading to reactor souring. The volatile fatty acid (VFA) accumulation increases and leads to subsequent drop in pH, if the acidogenic activity in the reactor dominates over the methanogenic activity. Hence, anaerobic digestion of these wastes are highly dependent on the ratio of acidogenic and methanogenic process.

Slaughter houses, meat and fish processing industries, poultry farms etc generate huge pile of discarded organic matter containing several body parts suitable for bio-gasification through anaerobic digestion. These wastes exhibit high biological oxygen demand (BOD), high organic matter content, high nitrogen content, low C/N ratio²⁸⁻³⁰. There has been a serious problem in anaerobic digestion process with these wastes. High ammonia concentration of animal wastes have been considered as a factor of inhibition for methanogenic activity³¹. A significant increase in ammonia concentration occurs due to fermentation of wastes. This problem becomes particularly serious when reactors are fed with wastes rich in protein⁸.

Cellulosic wastes are profoundly found in the environment. Very low or negative cost is associated with these wastes. Agricultural fields, paper and cardboard factories and textile mills provide enormous contribution to potential source of raw material for bio-gasification⁸. They form a part of municipal solid waste and are not always source-sorted, can be directly added for anaerobic treatment. Cellulosic wastes have very high C/N ratio in the range of $173 - 1000^{32}$, where as the optimum C/N ratio recommended for anaerobic digestion process is 20-30²³. Hence, C/N ratio adjustments are essential for a productive operation of anaerobic treatment. Protein rich wastes have been regarded as good buffering agents and at the same time they provide wide variety of essential nutrients but they are characterized by low C/N ratio. Hence, wastes with high carbon content has been co-digested with all substrates with low C/N ratio, thereby decreasing the chances of ammonia inhibition ^{33,34}. Agricultural feed-stocks are ligno-cellulosic wastes; they posses high C/N ratio³². Researchers attempted to co-digest these wastes with cattle manure in order to improve the C/N ratio and also to increase the volatile solid destructions resulting higher biogas yield³⁵. Li et al³⁶ studied the co-digestion of corn stalks and cattle manure in the ratio of VS_{manure} / $VS_{corn-stalks}$ 1:1,1:2,1:3 and 1:4 to determine the optimal C/N ratio for biogas production. The optimum C/N ratio for biogas production was achieved at the condition of VS_{manure} / VS_{corn-stalks} of 1:3.

Apart from the specific potential sources of bio-gasification discussed above, food waste has been regarded as a desirable material for co-digestion with dairy manure because of their biodegradability³⁷⁻³⁹. Studies on biogas production potential of unscreened dairy manure and mixtures of unscreened dairy manure and food wastes have been conducted in batch digesters at 35°C; showed that the methane yield of the co-digested process and unscreened manure after 30 days of digestion were 311 LKg⁻¹VS and 241 LKg⁻¹VS respectively⁴⁰.

Co-digestion of food wastes with manure: Many workers have studied the co-digestion of food wastes with cattle and swine manure. Food sector is an enormous generator of potential rejects for energy recovery. Food wastes consist of fruit and vegetable waste, kitchen waste, fish waste, meat and poultry waste etc. Zhang et al⁴¹ studied the anaerobic co-digestion of food waste and cattle manure in order to determine the key factors for biogas and methane yield. The workers have been successful in determining the key parameters for both batch and semi-continuous tests that revealed that total methane yield can be enhanced by co-digestion of food wastes and cattle manure in the ratio of 2 and optimum C/N ratio of 15.8. In batch tests, the methane yield has been observed to be enhanced by 41.1% corresponding to the methane yield of 388 mLg⁻¹VS and in semi-continuous mode, total methane yield at organic loading rate of 10gVS_{food-waste} L⁻¹d⁻¹ increased by 55.2% corresponding to methane yield of 317 mLg⁻¹VS. Sagagi et al⁴² utilized fruit and vegetable wastes like pineapple, orange, pumpkin, spinach and cattle manure individually to study their biogas production potential and found that highest weekly production rate was in the order of cow dung (1554 cm³ biogas) > pineapple waste (965 cm³ biogas) > orange waste (612 cm³ biogas) > pumpkin waste (373 cm³ biogas) > spinach waste (269 cm³ biogas) respectively. Aragaw et al¹ reported that organic kitchen wastes co-digested with cattle manure improved the biogas production potential as compared to cattle manure alone. Highest daily average and cumulative biogas yield has been obtained from the co-digestion of rumen fluid inoculated cattle manure and organic kitchen wastes mixed in the ratio of 1:3¹. The 1:3, 1:1 and 3:1 mixed ratios of cattle manure and organic kitchen wastes have been observed to produce an enhanced biogas yield from 24.12 to 47.13% and at the same time cumulative biogas yield has been also enhanced by 1.01 - 1.84 times¹. Similarly a study by Earnest and Singh⁴³ reported maximum biogas yield of 245 ml obtained from co-digestion of fruit and vegetable wastes with cow dung in the ratio of 1:1, followed by 230 ml of biogas yield from co-digestion in the ratio of 1:2. Zhang et al⁴¹ studied anaerobic co-digestion of food waste and cattle manure, to overcome the deficiencies of mono-digestion with cattle manure. The study has been carried out to identify the key parameters that identify the biogas and methane yield. Codigestion has been carried out in both batch and semicontinuous mode and found that total methane yield was enhanced by 41.1 % in batch tests with an optimum food waste (FM) to cattle manure (CM) ratio of 2. In semi-continuous mode, total methane yield at an organic loading rate of 10g VSFM/L/day was enhanced by 55.2 %. The workers also reported that addition of cattle manure increased the buffering capacity that facilitated high organic loading without pH control. Similar study by Voegeli et al⁴⁴ reported average daily biogas yield of 290 L/day and 130 L/day with daily feeding of 2Kg wet weight of food waste and market waste respectively. Food waste and market waste exhibited average VS destruction of 92.2% and 85.3% respectively. Garcia-Pena et al⁴⁵ studied anaerobic digestion of fruit and vegetable wastes from the central food distribution market in Mexico City. Methane yield, volatile solids removal and biogas production has been observed to be 50%, 80% and 0.42 m³biogas Kg⁻¹VS. Molinuevo et al⁴⁶ reported increase in methane yield from 111 to 244 mL CH₄/gVS and volatile solid destruction from 50% to 86% with co-digestion of swine manure and vegetable processing waste. The workers also repeated the same experiment for co-digestion of poultry litter and vegetable processing wastes and the biogas yield has been reported to enhancement from 158 to 223 mL CH₄/g VS and volatile solid destructions from 70% to 92%. Effect of co-digestion of vegetable waste and cow dung in ratios of 1:0 to 1:1.5 has been studied by Islam et al⁴⁷ using 4L capacity laboratory scale digesters. The digesters were essentially operated at batch mode at ambient temperature for a hydraulic retention time of 15 days with a total solid content of 8%. The maximum volume of biogas has been produced from co-digestion in the ratio of 1:1.

Kim et al⁴⁸ studied the effect of temperature and hydraulic retention time for the treatment of food waste. Methane content in biogas yield has been observed to be high for thermophilic conditions than the reactor run in mesophilic conditions. Methane yield has been observed to be highest in the reactor operated at 12 days retention time (223 L CH₄Kg⁻¹ COD degraded). Another study reported biogas yield of 0.35±0.02 m³Kg⁻¹VS resulting from co-digestion of 67% fruit and vegetable waste, 17% solid slaughter house waste and 17% manure in mesophilic digester with a retention time of 30 days and organic loading rate of 0.3-1.3 KgVSm⁻³day⁻¹ ⁴⁹. Kumar et al⁵⁰ determined the effect of 2.5 and 5.0 ppm concentrations of three heavy metals (Cd, Ni and Zn) on bio-gasification of potato waste and cattle manure at 37±1°C. All the three heavy metals have been successful in enhancing the biogas yield from codigestion of the substrate at 2.5 ppm concentration. The biogas yield enhancement has been in the order of Cd > Ni > Zn.

Co-digestion of energy-crops with manure: Although there are few data published on the co-digestion of energy crops with manure, more than 50% of the 3000 biogas plants in Germany has been using energy crops for energy recovery in the form of methane by the end of 2005. These plants have been essentially utilizing maize in co-digestion with different manures and other organic materials⁵¹. The biogas potential of different energy crops have been indicated in Table-3. Lower specific methane yield has been reported in co-digestion of manure with straw compared with digestion of manure alone^{23,24} where as another study reported highest specific methane yield from co-digestion of cow manure and 40% of wheat straw of total solid content⁵². Lehtomaki et al⁵³ investigated effect of crop to manure ratio for methane production by co-digestion of energy crops and crop residues with manure. Sugar beet tops, grass silage and oat straws have been co-digested with cow manure in continuous stirred tank reactors upto 40% volatile solids of crops in the feedstock. Highest specific methane yield of 268 (grass silage), 229 (sugar-beet tops) and 213 (oat straw) L CH₄Kg⁻¹VS_{added} has been obtained with 30% of crop in the feedstock. Volumetric methane yield increased by 65,68 and 16% in reactors fed with

30% VS of sugar beet tops, grass silage and oat straws along with manure compared to manure alone fed in reactors at a similar loading rate. Furthermore, bio-methane potential of co-substrates has been presented in Table-4.

Co-digestion of miscellaneous solid organic substrates: Concept of co-digestion with centralized facility has been employed at different corners of the world to make digestion cost-effective because the volume of organic waste wastes generated at a particular site may not be feasible for a treatment plant. Callaghan et al⁵⁴ studied co-digestions of cattle manure slurry with chicken manure at 7.5% and 15% total solid content, fish offal, fruit and vegetable wastes, brewery sludge and dissolved air floatation sludge. Fish offal and brewery sludge as co-digestate with cattle manure slurry produced an increase in methane yield as compared to that of control group of cattle manure slurry alone. Chicken manure at 15% total solids and fruit and vegetable wastes depressed the methane yield. Free ammonia inhibition has been considered as the hindering factor for methane yield from chicken manure. Fish offal has been observed as the highest contributor of specific methane yield followed by brewery sludge and others. Between dissolved air floatation sludge and brewery sludge the former resulted in higher cumulative methane yield compared to control group of cattle manure slurry alone. Ponsa et al⁵⁵ studied the co-digestion of organic fractions of municipal sludge with miscellaneous organic co-substrates like vegetable oil, animal fat, cellulose and protein. Vegetable oil has been concluded as most suitable substrate for co-digestion depending upon the determined parameters like ultimate methane production, maximum methane production rate, specific maximum methane production rate and volatile solids reduction. Other co-substrates (animal fat, cellulose and protein) used in this study, improved few anaerobic digestion aspects like ultimate methane production but initial production rates were poor than that of control group and rates of biodegradability also has not been observed to improve. Kuglarz and Mrowiec⁵⁶, investigated the co-digestion of kitchen remnants and sewage sludge. Higher degree of volatile solid destruction has been observed between 45.7 – 61.7 % for co-digestion of substrates. No significant deterioration of dewatering properties of the digestate has been observed. Gomez et al⁵⁷ studied combined biomethanation potential of primary sludge and vegetable fraction of municipal garbage under mesophilic conditions and compared anaerobic digestion of primary sludge alone. The digestion of co-substrates reported biogas yield of 0.60 – 0.80 Lg⁻¹VS whereas the control group of primary sludge alone yielded 0.4 – 0.6 Lg⁻¹VS respectively. Codigestions of fish wastes, abattoir waste water and waste activated sludge with vegetable fractions have been studied by Bouallagui et al⁵⁸. Anaerobic sequencing batch reactors have been run for 10 days under mesophilic conditions at an organic loading rate of 2.46 – 2.51 gVSL⁻¹day⁻¹ of which vegetable fraction comprised of 90% share in the mixture feedstream. C/N ratio of 22 – 25 has been found to be optimal for the treatment process. Zhang et al³² investigated co-digestion of bio-solids and organic fraction of municipal sewage and compared with anaerobic digestion of bio-solids alone. The C/N ratio has been observed to increase from 8.10 to 17.68 and reductions in total and volatile solids have been reported to be 30% and 65% respectively. Yen et al⁵⁹ performed exhaustive investigations on co-digestion of algal sludge and paper waste as unbalanced nutrients (C/N ratio) of algal sludge supposed to be a limitation factor to anaerobic digestion process. Addition of 50% volatile solids of waste paper to algal sludge feedstock, has been fruitful in terms of increased methane rate of 1170 ± 75 ml/l day as compared to 573 \pm 28 ml/l day obtained from algal sludge alone, operated at a hydraulic retention time of 10 days and organic loading rate of 4gVS/l day. Maximum methane production rate of 1607 ± 17 ml/l day has been observed with addition of 60% volatile solids to algal sludge operated at 5gVS/I day. The optimized C/N range in the co-digestion process has been 20 - 25 and increase in cellulase activity, led to efficient degradation of algal sludge. Sosnowski et al¹⁶ investigated co-fermentation of sewage sludge with organic fractions of municipal solid waste at both thermophilic and mesophilic conditions. Individually five experiments have been performed namely: i. thermophilic batch-digestion of primary sludge and thickened excess activated sludge, ii. cofermentation of sewage sludge and organic fractions of municipal solid wastes in the ratio of 3:1, iii. digestion of organic fraction of municipal solid waste alone at quasi-steady state continuous mode, iv. digestion of sewage sludge (primary sludge and thickened activated sludge at 1:1) alone at quasisteady state, v. co-fermentation of sewage sludge and organic fraction of municipal solid waste in the ratio of 3:1 at quasisteady state continuous mode. Methane concentration in biogas has been observed to be above 60% in all cases. Biogas productivity varied between 0.4 and 0.6 dm³g⁻¹VSS_{add} depending on the substrate added to the digester. Improvement in C/N ratio has been observed from 9:1 to 14:1. Cumulative biogas production increased with increasing proportions of municipal solid waste in the co-fermenting substrate, but biogas production has been achieved more at lower organic load than higher. Biological efficiency of methane production has been 82% and 62.7% for experiment 4 and 5 whereas lower efficiency of 49.3% has been observed with experiment 2. Banadda et al⁶⁰ performed co-digestion experiments of primary sludge obtained from a sewage treatment plant (STP) of Bugolobi with cow manure and brewery sludge respectively. Results showed that co-digestion of Bugolobi STP sludge with brewery sludge increased the biogas production rate by a factor of 3. Maximum biogas yield and production rate has been obtained with codigestion of Bugolobi STP sludge and brewery sludge in the ratio of 1:1, but co-digestion of a mixture (50% STP sludge, 25% brewery sludge and 25% cow manure) has been selected as optimal mixture for practical applications.

Conclusion

Biogas yield and corresponding methane content in it has been subsequently enhanced by co-digestion of wastes with cow Vol. **5(1)**, 49-57, January (**2016**)

Int. Res. J. Environment Sci.

manure. Specific bio-methane production, ultimate bio-methane production, methane production rate has been regarded as the key parameters for judging the success of co-digestion. The average optimum C/N ratio for co-digestion has been in the range of 20-30. In every possible case, the workers reported that co-digestion of wastes increased the load of nutrients that

led to higher bio-degradability, eliminated cases of ammonia inhibition, increased buffering capacity (caused by ammonia and volatile fatty acids) and finally led to formation of ecofriendly sludge. Hence, co-digestion appears to economic, potential and viable option for generation of alternative renewable source of energy to substitute the fossil fuels^{61,62}.

Table-1
Proximate properties of substrates⁶¹

Biomass	Proximate composition				
	Volatile fraction in dry matter %	Fixed carbon (%)	Ash (%)	Heating value, MJ/Kg dry wt.	
Maize stover	75.2,93.2,89.7	19.3	7.5,6.9,10.3	16.2,16.5	
Sugarcane Bagasse	70.9	7.0	16.0,14.7,22.1	10.0,14.3	
Sugarcane leaves	77.4	14.9	7.7	17.4	
Wheat straw	79.6,91.3,80.6,(88.9)	16.8,11.7	8.3,5.3,10.5,7.7,4.1,(11.1)	16.8,18.4,17.0,18.9	
Rice straw	69.3,70-95,72.7, (84.0)	17.3,11.8	16.2,14.7,15.5,(16.0)	15.3,14.5	
Rice husk	59.5,(75.7)		21.8,17.1,22.5,22.2,(24.3)	12.3, 16.5	

Table-2 Ultimate properties of substrates⁶¹

Biomass	Elemental Composition					
	Carbon %	Hydrogen %	Oxygen %	Nitrogen %	Sulphur %	
Maize straw	45.6	5.4	43.4	0.3	0.04	
Maize stover	43.7,35.2	5.6	43.3	0.6	0.01	
Sugarcane leaves	39.7	5.55	46.8	0.2	-	
Wheat straw	46.7,45.5,(45.8)	6.3,5.1,(6)	41.2,34.1	0.4,(0.42)	0.1	
Rice straw	41.8,36,(41)	4.6,5.3,(5.4)	36.6,43.1	0.7,0.35- 0.70,(0.74)	0.08	
Rice husk	37.9,44.6,38.2,(36.3)	4.82,5.6,5.6,(4.7)	34.9,49.3,33.7	0.43,(0.60)	0.17	

Int. Res. J. Environment Sci.

 ${\bf Table \hbox{-} 3} \\ {\bf Biogas\ potential\ of\ different\ energy\ crops}^{62}$

Crop	Biogas yield (Nm³ton⁻¹VS added)	Methane content (%)	
Sugar beet	730-770	53	
Fodder beet	750-800	53	
Maize	560-650	52	
Corn cob mix	660-680	53	
Wheat	650-700	54	
Triticale	590-620	54	
Sorghum	520-580	55	
Glass	530-600	54	
Red clover	530-620	56	
Sunflower	420-540	55	
Wheat grain	700-750	53	
Rye grain	560-700	53	

VS: volatile solids

Table-4
Bio-methane potential of organic co-substrates

Co-substrates	Ratio	Operational conditions	CH ₄ yield (Nm ³ ton ⁻¹ VS added)	References	
Pig manure : corn stover	75:25 (VS basis)	V=30 L; T= 39 ⁰ C	210	21	
Pig manure : wheat straw	75:25 (VS basis)	V=20L;	240	24	
rig manure . wheat straw	50:50 (VS basis)	$T = 35^{\circ} C$	220		
Dia manusa a notata vuosta	85:15 (VS basis)	V=3.5 L;	210-240	25	
Pig manure : potato waste	80:20 (VS basis)	$T=35\pm1^{0} C$	300-330		
OFMSW : vegetable oil	83:17(dry wt basis)		699±6		
OFMSW: animal fat	83:17(dry wt basis)	V=1L;	508±16	55	
OFMSW : cellulose	83:17(dry wt basis)	$T=37^{0} C$	254±10		
OFMSW : protein	83:17(dry wt basis)		288±7		
Buffalo manure : maize silage	70:30 (VS basis)	V=0.5 L; T=35±1 ⁰ C	358.23±44.15	8	
Cow manure : straw	90:10 (VS basis)		145±9		
	80:20 (VS basis)	V=1.5 L;	159±19	35	
	70:30 (VS basis)	$T=35\pm1^{0} C$	213±17		
	60:40 (VS basis)		188±19		
Cow manure : barley straw	80:20 (Volume basis)	V=100 L; T=35 ⁰ C	160	22	
Cow manure : forage beet silage	83:17 (VS basis)	V= 20 L; T=35 ⁰ C	400	63	
Cow manure : fruit and	80:20 (dry wt basis)	V=18L;	380	28	
vegetable waste	70:30 (dry wt basis)	$T=35\pm0.5^{\circ} C$	340		

Co-substrates	Ratio	Operational conditions	CH ₄ yield (Nm ³ ton ⁻¹ VS added)	References
	60:40 (dry wt basis)		380	
	50:50 (dry wt basis)		450	
	25:75 (VS basis)	V=2L; T=30±8 ⁰ C	107.03	
Organic kitchen waste : cow manure	50:50 (VS basis)		128.14	1
cow manare	75:25 (VS basis)	1-3020 C	146.53	
	25:75 (VS basis)	V=4L; T=35±1 ⁰ C	968±73	
Algal sludge : waste paper	50:50 (VS basis)		1170±75	59
	75:25 (VS basis)		317±114	
Food waste : cow manure	67:33 (VS basis) (Batch)	V=1L; T=35±1 ⁰ C	388	41
rood waste . cow manure	67:33 (VS basis) (Semi-continuous)		317	
Dairy manure : potato waste	75:25 (VS basis)		227.7	
Dairy manure : used oil	75:25 (VS basis)	V=0.25L; T=35±1 ⁰ C	360.6	64
Dairy manure : cheese whey	75:25 (VS basis)		252.4	
Dairy manure : switch grass	75:25 (VS basis)		207.8	

VS: volatile solids; V: volume; T: temperature

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