The Effect of Adding Corn Silage at Different Ratios to Orange and Tangerine Wastes on Biogas Production Efficiency

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ABSTRACT

In this study, biogas production efficiencies of mixtures obtained by adding corn silage (CS) to citrus industrial wastes at different ratios were determined. Orange (OJPW) and tangerine processing juice wastes (TJPW) (crusts and shells) were selected as materials in the study. 25%, 50%, 75% of CS was added to these selected wastes. Changes in the obtained mixture chemical properties (dry matter, dry organic matter, crude ash, crude protein, crude oil, Acid Detergent Fiber (ADF), Neutral Detergent Fiber (NDF)), biogas production and methane content in the biogas were investigated. The results of the study showed that the highest crude protein content was found in 100% TJPW (10%), raw fat percentage in 100% TJPW (5.14%), dry matter content in 100% CS (93.56%), ADF in 100% CS (22.74%) and the NDF in a mixture of 25% OJPW + 75% CS (45.08%).

The highest methane production was determined for a mixture of 100% TJPW and 50% TJPW + 50% OJPW (0.46 m^3 kg^{-1} ODM). Also the highest biogas production was determined in a mixture of 50% OJPW + 50% TJPW (0.90 m^3 kg^{-1} ODM). The mixing of CS in TJPW and OJPW reduced significantly the production of methane and biogas in the mixture. As a result of the statistical analysis, significant differences ($P \leq 0.05$) were found in both methane and biogas production of agricultural wastes.

Keywords: Orange wastes; Mandarin wastes; Corn silage; Biogas; HBT

1. Introduction

Today the gradual decrease of fossil fuel resources has increased the need for renewable energy sources (Üçok et al 2016). In recent years, due to its contribution to the sustainable development of countries, there is increasing interest in the energy obtained from biomass, especially biogas (Mansourpoor & Shariati 2012; Üçok et al 2016;

Mojtaba & Ahmad 2012). Wastes emerged from agriculture, and related industry is major sources of biogas production. Application of biogas technology provides not only the disposal of waste but also energy production (Deublein & Steinhauser 2008).

Citrus fruits are among the most grown and consumed fruit group in the world. In this regards, China ranks first with about 32 million tons,

followed by Brazil with 20 million tons and the USA with 10 million tons. Turkey is ranked the eighth with 3.7 million tons (FAO 2015). Approximately 85% of citrus production in Turkey is produced in the Mediterranean region (TUİK 2015). Turkey has an important position in the production of citrus fruits especially regarding exports to the Middle East (Iraq, Saudi Arabia, and Iran), Russia, Ukraine, Romania, Poland, and Bulgaria. However, there are significant problems in the export of products grown in recent years due to political and geopolitical problems among these countries, to which exports have been made. Therefore, most of these products can not be consumed due to excess supply in the domestic market (Çallı 2012).

Processing the citrus fruits into juice, a large part of the separated pulp consisting of crust which is rich in cellulose and pectin can be utilized in bioprocesses with the aid of preprocessing. This may create an opportunity for the fruit juice industry in Turkey for the valorization of wastes (crusts and pulps) existed from the processing of orange and tangerine regarding the production of biogas.

The efficiency of biogas production from fruit processing wastes can be increased by co-fermentation with wastes which have higher biogas potential (Elaiyaraju & Partha 2012). This may encourage the establishment of small-scale biogas plants in the region, leading to the prevention of environmental pollution and the utilization of significant energy resources. In Europe, there are many central biogas plants that are operated successfully, producing biogas from thousands of farm-type and organic household and industrial wastes producing biogas from animal wastes and energy plants (Çallı 2012). Although numerous studies on the usage of fruit pulps and wastes for biogas production have been carried out (Aslanzadeh & Özmen 2009; Elaiyaraju & Partha 2012; Nguyen 2012; Wikandari et al 2014a; Wikandari et al 2014b), there are limited studies on co-fermentation of two plant-based materials to increase biogas production efficiency concerning fruit pulps and wastes.

In this study, it was aimed to determine the biogas production efficiencies of the mixtures obtained by adding corn silage to citrus industrial wastes at different rates. For this aim, corn silage has been added to the orange and mandarin wastes produced during fruit juice industry production activities at different rates. The chemical properties of these obtained mixtures, biogas productions and methane rates in the biogas have been determined.

2. Material and Methods

2.1. Material preparation

In this study, orange juice processing waste (OJPW) and tangerine juice processing waste (TJPW) in the form of pulp were selected as materials, and corn silage (CS) with a high biogas potential was selected as co-fermentation material. The OJPW and TJPW were received from the enterprises producing fruit juice in Çukurova Region, and the CS was obtained from Animal Feeding Center at Çukurova University. The samples were dried at room temperature for three weeks at Biogas laboratory, Department of Biosystem Engineering in Kahramanmaraş Sütçü İmam University. The moisture contents of the samples dried at room temperature were determined as 82.89%, 82.66%, and 64.65% (wet basis, w.b.) for TJPW, OJPW, and CS, respectively, before grinding. The dried samples were ground in a grinder to a grain size of 1 mm (based on the VDI 4630 2006 standard). The ground samples were weighed with a precision of 0.2 g $\pm$ 0.1 µg to fit the established experimental conditions.

In this study, the ground materials were taken in quantities appropriate to the standard conditions (0.2 g) and the samples in Table 1 were prepared. Samples were placed in 100 mL glass syringes as three replicates. Dry matter (drying in the oven), crude oil (according to the Soxhlet method), crude protein (according to the Kjeldahl method), crude ash analyses (in the ash furnace) were analyzed based on AOAC (1990), and Acid Detergent Fiber (ADF) and Neutral Detergent Fiber (NDF) analyses were conducted according to Van Soest et al (1991).

Table 1- Biogas production materials

Material	Number of samples
100% OJPW	3
100% TJPW	3
100% CS	3
75% OJPW+25% CS	3
50% OJPW+50% CS	3
25% OJPW+75% CS	3
75% TJPW+25% CS	3
50% TJPW+50% CS	3
25% TJPW+75% CS	3
50% OJPW+50% TJPW	3

2.2. Biogas and methane measurements

Methane rates in biogas and biogas production were determined by the HBT method (Heffrich & Oechsner 2003). In the study, the prepared samples were placed into 100 mL glass syringes located in the incubator. In the same way, three inoculum syringes, which were prepared using burette, each receiving 30 mL of inoculum for control group samples, were also placed in the sections in the incubator. The syringe piston was removed before the weighed samples were placed in the syringe, and plastic clips were attached to the silicone hoses in the tip of the injectors and used for gas transfer. Vaseline was applied to the pistons of the injectors, leaving a three-finger space at the top and bottom, to prevent gas escape during analysis. Afterwards, the glass syringes were made ready to use by attaching its pistons and closing the clips. The syringes were placed horizontally into the incubator at 37 °C after the inoculum was placed. The methane measurement system used to determine the methane content prior to operations in the incubator was calibrated with a calibration tube (60.5% CH$_4$) (S-AGM plus 1010 sensor). The purpose of calibration is to verify that the measured gas is at standard conditions (0 °C and 1013 hPa). Experiments lasted for 35 days. Measurements were made every 6 hours for the first 6 days, 8 and 12 hours later on the subsequent days, and the methane efficiency in each sample was determined. While Equation (1) was used to calculate the normal volume of the produced gas in the glass syringes prepared for each sample of the materials studied, Equation (2) was used to

determine the methane content of the formed biogas, and Equation (3) was used to calculate cumulative methane over time (VDI 4630 2006).

$$V_0^{tr}=V \left(\frac{(P-P_w)(T_0)}{(P_0)(T)} \right) \tag{1}$$

Where; V_0^{tr}, Volume of gas under normal conditions (mLN); V, Volume of gas read (mL); P, Air pressure at the time of reading (hPa); P_w, The steam temperature of the water in the outside (hPa); T_0, Normal temperature (273 K); P_0, Normal pressure (1013 hPa); T, Temperature of the gas which has undergone fermentation in the outside (300 K)

$$C_{CH_4}^{tr} = C_{CH_4}^{f} \left(\frac{P}{(P-P_w)} \right) \tag{2}$$

Where; $C_{CH_4}^{tr}$, The volumetric methane content in dry biogas (%); $C_{CH_4}^{f}$, Volumetric methane content in moist biogas (%); P, Gas pressure during reading (hPa); P_w, The steam temperature of the water in the outside (hPa).

$$M_{CH_4}(t) = M_{CH_4}(0)+\int_{t_1}^{t_2} M_{CH_4}(t)\, dt \tag{3}$$

Where; $M_{CH_4}(t)$, Cumulative methane production (Nm^3 CH$_4$ kg^{-1} ODM^{-1} min^{-1}); $M_{CH_4}(0)$, Methane production when t = 0 (Nm^3 CH$_4$ kg^{-1} ODM^{-1} min^{-1}); t_2-t_1, Time between two measurements (min).

2.3. Preparation of ınoculum

Sludge, which is a mixture of liquid + solid phase, was received from Gaziantep Water and Sewerage Administration (GWSA) central wastewater treatment plant. It was filtered through four layers of cheesecloth and was mixed with 1:2 buffer solution to prepare the inoculum. Buffer solution consisted of 500 mL of distilled purified water, 0.1 mL of solution A, 200 mL of solution B, 200 mL of solution C, 1 mL of resazurin (0.1%, w v^{-1}) solution C, and 40 mL of solution E. Solution A was prepared by dissolving 13.2 g of CuCl$_2$2H$_2$O, 10.0 g of MnCl$_2$4H$_2$O, 1.0 g of CoCl$_2$6H$_2$O, 8.0 g of FeCl$_2$6H$_2$O in distilled water, completing the volume to 100 mL. Solution B was prepared by dissolving 35 g of NaHCO$_3$ and 4 g of NH$_4$HCO$_3$ in distilled

water, completing the volume to 100 mL. Solution C was prepared by dissolving 5.7 g of Na_2HPO_4, 6.2 g of KH_2PO_4, 0.6 g of $MgSO_4 7H_2O$ in distilled water, completing the volume to 1000 mL. Solution D was prepared by dissolving 0.5 g of resazurin in distilled water, completing the volume to 100 mL. Solution E consisted of 95 mL of distilled water, four mL of N-NaOH and 625 mg of $Na_2S_9H_2O$.

2.4. Evaluation of data

The mean and standard deviation values, statistical analyzes and variance analyzes of the measurements made in three replicates were determined, and the obtained values were interpreted by transferring them into the figures and tables.

3. Results and Discussion

3.1. Chemical properties of materials

The chemical properties of the mixes obtained as a result of the analysis are given in Table 2. As can be seen from the Table 2, in the chemical analyses conduced in the samples, the highest crude protein ratio was found for the 100% TJPW (10%); the highest crude fat was determined for the 100% TJPW (5.14%); the highest dry matter content was found for 100% CS (93.56%); the highest ADF ratio

was determined for 100% corn silage (22.74%); and the highest NDF rate was determined to be in the mixture of 25% OJPW+75% CS (45.08%).

3.2. Biogas and methane production values of materials

The cumulative specific methane production values of the inoculum and mixes based on time are shown in Figure 1. According to results, the maximum methane production in the studied materials ranged from between 30 and 35 days (Figure 1). In the study, the highest methane production (0.46 m^3 kg^{-1} ODM) was determined for 100% TJPW, and the mixture of 50% TJPW + 50% OJPW, and the highest biogas production (0.90 m^3 kg^{-1} ODM) was found for the mixture of 50% OJPW + 50% TJPW. The cumulative specific methane and biogas productions and methane rates in the biogas are shown in Table 3. Methane and biogas production in mixtures containing CS at different ratios of OJPW and TJPW varied between 0.31-0.46 m^3 kg^{-1} ODM and 0.62-0.90 m^3 kg^{-1} ODM, respectively (Table 3). The change in the cumulative specific methane and biogas production values are given in Figure 2. The results of variance analysis of methane production, biogas production, and biogas methane rates are given in Table 4.

Table 2- Chemical properties of mixes

Material	Crude protein content (%)	Crude oil content (%)	Dry matter content (%)	Organic matter content (%)	ADF (%)	NDF (%)
100% OJPW	7.78±0.09[de]	2.30±0.06[d]	88.67±0.02[bc]	95.56±0.15[ab]	14.76±0.08[c]	20.12±0.11[f]
100% TJPW	10.00±0.13[a]	5.14±0.11[a]	88.65±0.19[c]	95.40±0.18[ab]	20.21±0.03[ab]	26.49±0.04[e]
100% CS	7.55±0.06[e]	2.43±0.14[d]	93.56±0.21[a]	95.02±0.26[ab]	22.74±0.18[a]	43.04±0.13[ab]
75% OJPW+25% CS	7.56±0.04[e]	2.38±0.09[d]	90.88±0.05[abc]	96.11±0.17[ab]	19.53±0.13[ab]	34.68±0.27[d]
50% OJPW+50% CS	7.59±0.09[e]	2.56±0.13[d]	91.62±0.46[ab]	93.71±0.39[b]	21.36±0.25[ab]	38.59±0.02[bc]
25% OJPW+75% CS	7.46±0.02[e]	2.50±0.07[d]	92.16±0.31[ab]	97.56±0.41[a]	21.11±0.04[ab]	45.08±0.01[a]
75% TJPW+25% CS	9.27±0.11[b]	4.68±0.09[ab]	89.59±0.15[bc]	95.25±0.16[ab]	19.21±0.08[ab]	37.12±0.02[bc]
50% TJPW+50% CS	8.72±0.04[c]	4.07±0.05[bc]	90.49±0.27[ab]	95.86±0.04[ab]	21.09±0.32[ab]	39.37±0.34[bc]
25% TJPW+75% CS	8.06±0.15[d]	3.40±0.08[c]	92.05±0.34[ab]	96.75±0.047[ab]	22.68±0.38[a]	44.54±0.16[a]
50% OJPW+50% TJPW	8.81±0.07[c]	3.85±0.15[c]	88.70±0.27[abc]	94.85±0.33[ab]	17.65±0.07[bc]	23.44±0.08[ef]

P≤0.05 a, b, c, d, e, the differences between the mean scores of cumulative specific methane and biogas production, represented by different letters in the same column, are significant; P≤0.1 a, b, the differences in the mean scores of the ratio of methane in the biogas indicated by different letters in the same column are significant
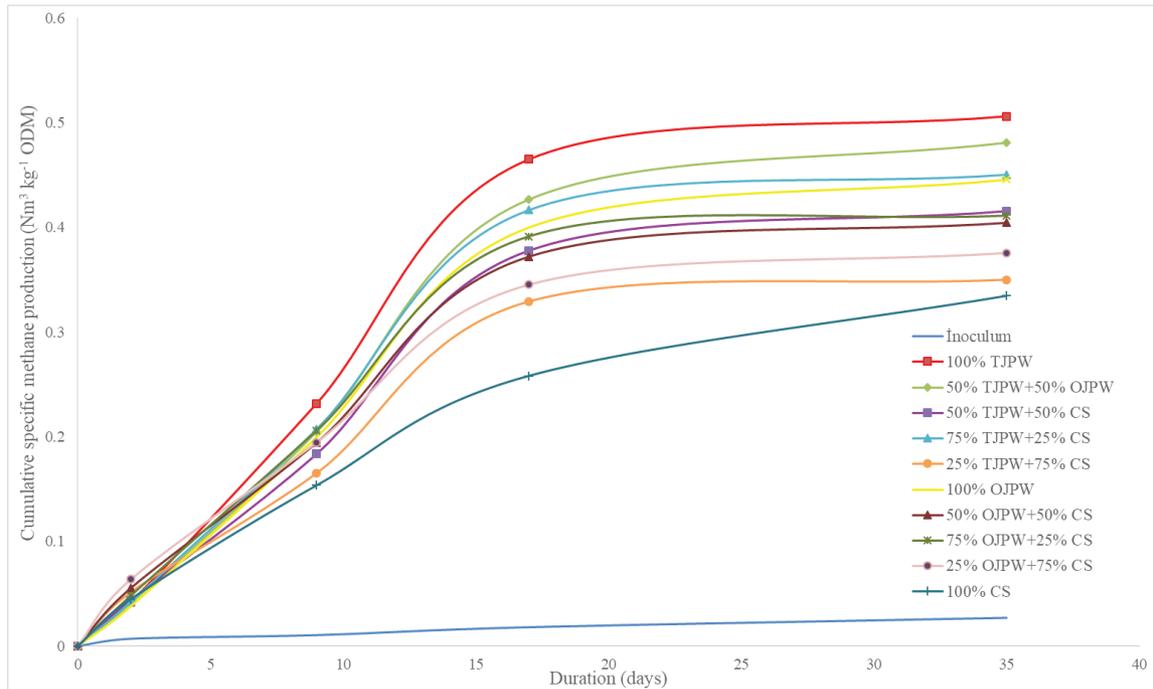
Figure 1- Change in average cumulative methane productions of materials based on time

Table 3- Methane and biogas productions and methane rates in biogas of materials

Material	Cumulative specific methane production (m³ kg⁻¹ ODM)	Cumulative specific biogas production (m³ kg⁻¹ ODM)	Methane rate in biogas (%)
100% OJPW	0.41±0.006[ab]	0.85±0.012[ab]	48.42[b]
100% TJPW	0.46±0.011[a]	0.86±0.023[ab]	53.69[a]
100% CS	0.31±0.017[c]	0.62±0.030[e]	50.00[ab]
75% OJPW+ 25% CS	0.38±0.004[bc]	0.76±0.003[bcd]	49.81[ab]
50% OJPW+ 50% CS	0.37±0.013[bc]	0.73±0.019[cde]	50.63[ab]
25% OJPW+ 75% CS	0.34±0.006[c]	0.70±0.007[cde]	48.83[ab]
75% TJPW+ 25% CS	0.42±0.012[ab]	0.82±0.014[abc]	51.16[ab]
50% TJPW+ 50% CS	0.38±0.020[bc]	0.77±0.019[abcd]	49.73[ab]
25% TJPW+ 75% CS	0.31±0.028[c]	0.63±0.044[de]	49.60[ab]
50% OJPW+ 50% TJPW	0.46±0.009[a]	0.90±0.019[a]	50.79[ab]

P≤0.05 a, b, c, d, e, the differences between the mean scores of cumulative specific methane and biogas production, represented by different letters in the same column, are significant; P≤0.1 a, b, the differences in the mean scores of the ratio of methane in the biogas indicated by different letters in the same column are significant.

The mixing of CS with OJPW and TJPW significantly reduced the production of methane and biogas in the mixture. In the other word, the increase in the ratio of CS to OJPW and TJPW negatively affected the methane and biogas production of the mixture. At the other side, maximum cumulative specific methane production (0.46 m³ kg⁻¹ ODM, Table 3) existed for %100 TJPW due to its crude
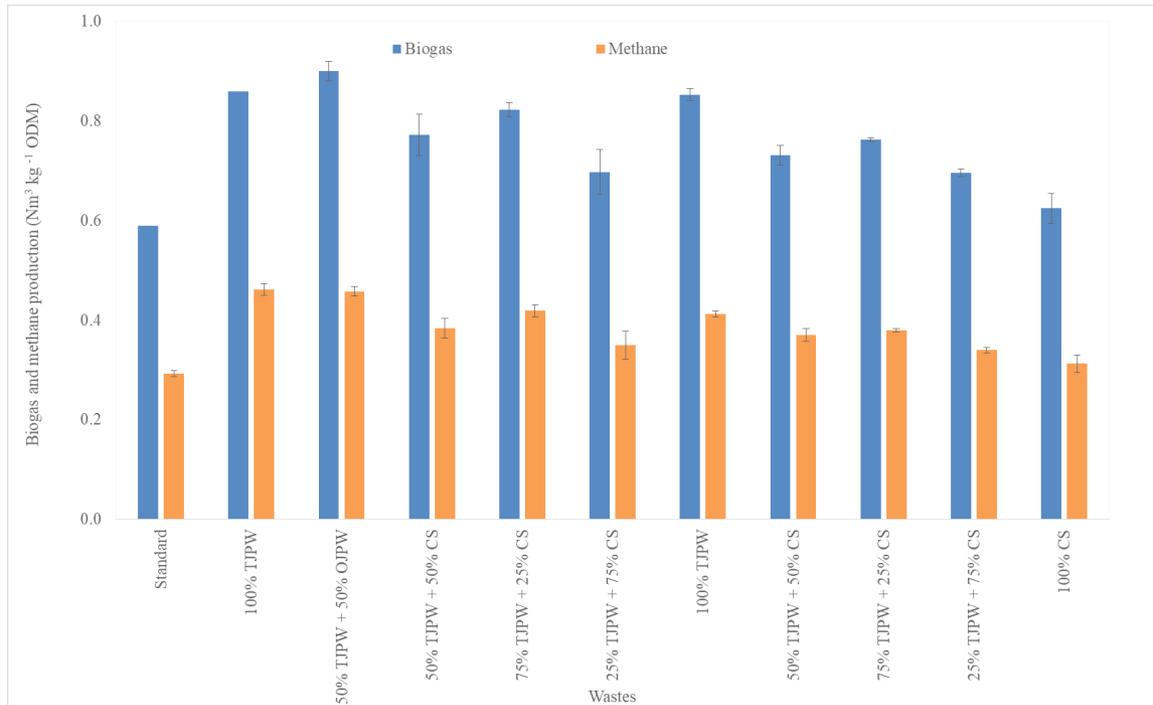
Figure 2- Changes in cumulative specific methane and biogas productions of materials

Table 4- Analysis of variance table of methane, biogas productions and methane ratios of materials

	Source of variance	SD	SS	MS	F value	SEM	P value
Methane	Between groups	9	0.75	0.08	13.236	0.0250	0.000***
	Within groups	20	0.13	0.01		0.0250	
	Total	29	0.88			0.0250	
Biogas	Between groups	9	0.237	0.26	12.653	0.037	0.000***
	Within groups	20	0.042	0.02		0.037	
	Total	29	0.279			0.037	
Methane ratio (%)	Between groups	9	58.174	6.464	2.004	1.466	0.094*
	Within groups	20	64.521	3.226		1.466	
	Total	29	122.696			1.466	

***, the differences between the mean scores of methane and biogas are significant (P≤0.05); *, the differences between the mean scores of methane ratios are important (P≤0.1); *SD*, Standard Deviation; *SS*, Some of Squares; *MS*, Mean Square; *SEM*, Standard Error of the Mean

protein content (10.0%) and crude oil content (5.14%). This findings is supported by the statement of "the amount of methane production increases with the increase in protein and fat by Avcıoğlu (2011).

The cell contents are fast, NDF-ADF (hemicellulose) is slow and ADF (cellulose and lignin) is digested more slowly in the inoculum (Van Soest 1994). Therefore, the digestion of CS occurred later due to high rates of ADF and NDF (Figure 1). In the statistical analysis, both methane and biogas productions of mixes differed significantly (P≤0.05). Methane ratios in the biogas also differed significantly (P≤0.1) (Table 4).

The differences in the methane and biogas interaction of the materials that were examined in the study were also evaluated. There was no interaction for OJPW, TJPW, and CS. Mixtures formed by mixing these samples at certain ratios will also be subjected to co-fermentation, so their interactions will be different. For example, the specific methane production values were 0.38 m^3 kg^{-1} ODM for the mixture of 75% OJPW + 25% CS, 0.41 m^3 kg^{-1} ODM for 100% OJPW and 0.31 m^3 kg^{-1} ODM for 100% CS (Table 3). The methane interaction was 75% x 0.41 + 25% x 0.31 = 0.385 m^3 kg^{-1} ODM for the mixture of 75% OJPW + 25% CS (Table 5). The average specific methane production values were 0.38 m^3 kg^{-1} ODM for the mixture of 75% OJPW+25% CS and 0.385 m^3 kg^{-1} ODM as a result of its interaction. The value of difference was calculated to be -0.005 m^3 kg^{-1} ODM, and the value of decrease was calculated to be -1.30% (Table 5). The values in other materials (50% OJPW+50% CS, 25% OJPW+75% CS, 50% TJPW+50% CS, 25% TJPW+75% CS, 50% OJPW+50% TJPW) in the study were calculated in the same evaluation method which was applied to the other mixtures (50% OJPW+50% CS, 25% OJPW+75% CS, 50% TJPW+50% CS, 25% TJPW+75% CS, 50% OJPW+50% TJPW). The increase in the production of methane and biogas as a result of co-fermentation interactions resulted in a maximum of 5.41% in the mixture of 50% OJPW+50% TJPW (Table 5).

Table 5- Interaction differences of methane and biogas

Waste materials	Methane interaction difference (%)	Biogas interaction difference (%)
100% OJPW	0.00	0.00
100% TJPW	0.00	0.00
100% CS	0.00	0.00
75% OJPW+25% CS	-1.30	-4.10
50% OJPW+50% CS	2.87	-0.56
25% OJPW+75% CS	1.44	-12.14
75% TJPW+25% CS	-0.87	2,87
50% TJPW+50% CS	-0.33	4.32
25% TJPW+75% CS	-10.79	-6.74
50% OJPW+50% TJPW	5.41	5.27

The methane values of the OJPW and TJPW were reported as 0.45 Nm^3 kg^{-1} ODM and 0.48 Nm^3 kg^{-1} ODM (Gunaseelan 2004), biogas and methane production from CS, 0.70 Nm^3 kg^{-1} ODM and 0.34 Nm^3 kg^{-1} ODM, respectively (Amon et al 2007). The findings in this study were close to the limit values reported by Gunaseelan (2004) and Amon et al (2007). It is thought that this difference, which was observed at a small level in the study, may be due to the chemical structure of the materials (Avcıoğlu 2011).

Results showed that the wastes examined, the highest raw protein and crude oil ratio was in 100% TJPW, the highest dry matter ratio was in 100% CS, the highest organic dry matter ratio was in the mixture of 25% OJPW+75% CS, the highest ADF ratio was in 100% CS, and the highest NDF ratio was in the mixture of 25% OJPW+75% CS.

5. Conclusions

The highest cumulative specific methane and biogas production values were found in 100% TJPW, 100% OJPW and the mixture of 50% OJPW+50% TJPW, while the lowest such values were found in mixtures of OJPW and TJPW containing 75% CS, and 100% CS. A significant difference (P≤0.05) was found between the methane and biogas productions of the agricultural wastes examined in the statistical analysis. The highest increase in the production of methane and biogas as a result of co-fermentation interaction occurred in the mixture of 50% OJPW+50% TJPW. The co-fermentation of the wastes resulted in differences in the proportions of the cumulative methane, biogas, and methane in biogas. CS mixed at different ratios to OJPW and TJPW reduced methane and biogas production of the mixture.

Recommendations

The biogas production potential of OJPW, TJPW, and CS, and their co-fermentations is high. For this reason, it can be said that they are important materials for biogas plants. New wastes to be generated in case of using the mentioned waste for production of biogas can be used for organic fertilizer in agriculture. Greenhouse gases (methane and carbon dioxide)

that will be released into the atmosphere due to the storage of wastes on uncontrolled conditions will be prevented with the launch of the biogas process. As a result of the elimination of wastes, the odor intensity in the air will decrease, and the environmental problems will be reduced. The level of methane ratios in the cumulated methane, biogas, and biogas in methane that are formed as a result of appropriate co-fermentation interactions can be increased. By providing co-fermentation at an appropriate level, energy production in biogas plants is going to be an able to increase.

Acknowledgements

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