



## Research Article

# Biodegradation of high cellulose-lignin content agricultural wastes in bioreactors

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## ABSTRACT

The bioreactor landfill is a solid waste disposal method that provides rapid degradation of solid waste and acquisition of methane. Bioreactors in which leachate circulation is carried out are generally operated anaerobically. The biodegradation of wastes with high lignin and cellulose content is very difficult. Especially under anaerobic conditions (moreover, if there is a lack of moisture), such wastes almost never decompose. In this study, the degradation of waste sunflower stalks that are difficult to biodegrade and have a high lignin-cellulose content and the production of methane gas in semi-aerobic bioreactors have been investigated. Sunflower stalks were loaded into the bioreactors in different proportions and mixed with the organic fraction of municipal solid waste (OFSWM). The bioreactors have been operated under different operating conditions. The contents of cellulose, hemicellulose, lignin, and initial and final organic matter in the wastes loaded into the bioreactors were examined. Parameters such as pH, COD, BOD5, TKN, NH<sub>4</sub>-N in leachate were analysed and the amounts of total and methane gas were measured. Initially, all bioreactors have been operated anaerobically. In the decomposition of the sunflower stalk, while 43% of the organic matter removal was achieved in the anaerobic bioreactor, 60% of the organic matter removal was realized in the semi-aerobic/anaerobic bioreactor. The other agricultural wastes were then subjected to decomposition under semi-aerobic/anaerobic operating conditions. As a result of the study, it can be said that semi-aerobic pretreatment accelerates the decomposition of agricultural waste with a high lignin and cellulose content, decreases the COD values of leachate, and increases the amount of methane.

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## INTRODUCTION

Agricultural waste, known as biomass, has significant potential to produce sustainable energy from renewable fuels [1]. Lignocellulosic materials, such as agricultural waste, are widely accepted as the most suitable raw materials

for the production of biomethane due to their relatively low costs, high availability, and lack of direct competition with the production of food and feed [2]. Lignocellulosic biomass is composed of cellulose, hemicellulose, lignin, oil, starch and proteins [3].

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Sunflower stalks, which are lignocellulosic biomass, produce 78 to 182 million tons globally and are typically disposed of as garbage or burned in fields, resulting environmental pollution [4]. Sometimes, sunflower remains (stems, leaves, heads) are left in the fields after the seed harvest [5]. In contrast, the high cellulose and hemicellulose content of the sunflower stalk makes it a potential raw material for the production of biomethane [6]. However, in the anaerobic decomposition of sunflower stalk, which contains high-crystalline cellulose, a lignin of a natural structure and hemicellulose, hydrolysis is rate-limiting and adversely affects methane production [7].

Pruning waste contains organic compounds such as lignin, which prevents and slows the aerobic degradation of cellulose, which is resistant to biodegradation, or hemicellulose, which is slow/moderately degradable [8].

Hazelnut husks are green plant structures that surround and protect the hazelnut during the growth process. At harvest time, this plant structure is collected with the hazelnut, separated from the hazelnut in the factory, and converted to waste. Turkey, which hosts 73% of the world's hazelnut production, is the world's largest hazelnut producer (400.000–450.000 tons/year). Each year after the hazelnut harvest, the hazelnut waste is burnt or left in the field [9]. Anaerobic decomposition of organic substances containing lignocellulosic matter, such as hazelnut husks, takes a long time and produces inefficient biogas [10].

In recent years, it has been demonstrated that bioreactor landfill technology supports the decomposition and stabilization of biodegradable organic waste by adding water, recirculating leachate, or injecting air [11].

Bioreactor systems are landfill systems that use advanced controlled microbial processes to convert and balance organic waste compounds that can be easily or partially decomposed in as little as 5-8 years [12]. The bioreactor areas may operate under various operating conditions. 4 types of bioreactor systems have been developed in line with the studies carried out so far. These are; anaerobic, aerobic, facultative, and hybrid bioreactors [13].

In this study, the effect of adding an organic fraction of solid waste to the biodegradation of sunflower stalk was examined. The other objective is mixing these wastes at different rates within themselves by applying different operating conditions in bioreactors and observing methane production.

## MATERIALS AND METHOD

### Laboratory-Scale Simulated Bioreactor

Two stainless steel cylindrical bioreactors with a height of 30 cm, a diameter of 10 cm and a reactor volume of 2.4 L were used in experiments (Figure 1). The anaerobic reactor has a part that allows air to be supplied with the help of a diffuser from the lower part to provide aerobic conditions. It was operated under mesophilic conditions (33-35°C). According to the meteorological data in the region, enough water was added the reactor in rainy days.

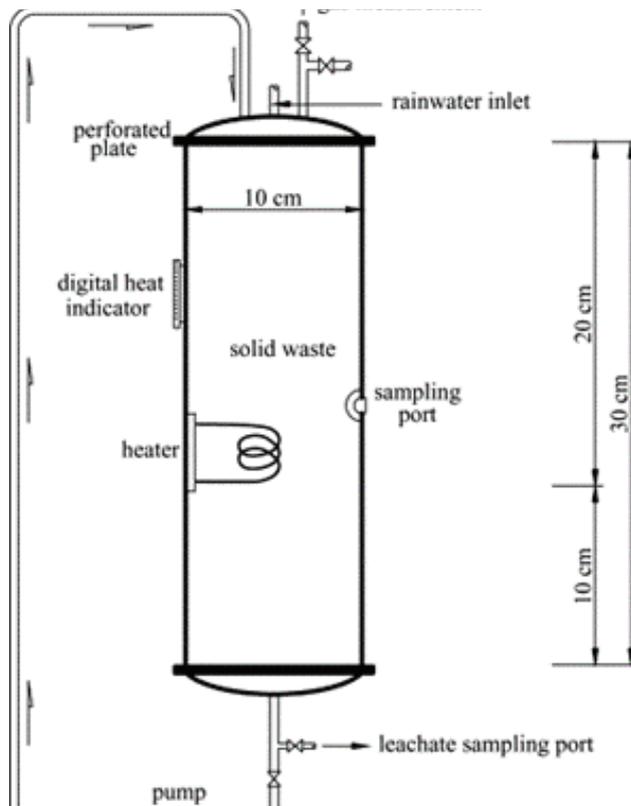


Figure 1. Laboratory-Scale Simulated Bioreactor.

### Anaerobic and Semi-aerobic/ Anaerobic Bioreactor Operating Conditions

Leachate recirculation in bioreactors under anaerobic and semi-aerobic/anaerobic operating conditions was achieved with peristaltic pumps. Leachate recirculation between 10% and 15% (300 ml) of reactor volume has been shown to be appropriate in several studies in terms of methane yield [14]. The peristaltic pumps used in leachate recirculation were set to operate for 15 minutes every 8 hours with a timer. For the measurement of total gas and methane gas, specially manufactured scaled glass materials were installed in each reactor.

In bioreactors that have been operated under semiaerobic / anaerobic conditions, reactors are aerated for 1 hour in the form of 5 minutes of aeration and 3 minutes of rest with the help of a diffuser 5 days a week to ensure that aerobic conditions have been realised. After the aerobic operation process was completed, the system have been operated anaerobically for methane production.

### Loading of Agricultural Solid Waste and Organic Fraction of Municipal Solid Waste Mixtures into Bioreactors

The organic fraction of municipal solid waste (OFMSW) used in bioreactors was obtained from kitchen waste in the central cafeteria of Pamukkale University, and the sunflower stalk (SS) was obtained from the sunflower pruning waste from a sunflower field in the Tavas district of

**Table 1.** Amount of lignin, cellulose, and hemicellulose (%) of the sunflower stalk

	Hemicellulose (%)	Lignin (%)	Cellulose (%)
Sunflower Stalk	43.67±0,8	17.23±0,2	24.46±0,3

**Table 2.** Water content and organic matter analysis in waste mixtures used in bioreactors

	Water Content (%)	Organic Matter (%)
RA1	71±0,3	82±3,3
RA2	76±5,2	89±4,4
RSA1	26±2,3	88±1,2
RSA2	12±0,4	85±0,3

Denizli province. The first anaerobic bioreactor (RA1) was filled with 20% OFMSW + 80% SS, and the second reactor (RA2) was filled with 50% OFMSW + 50% SS, and waste was loaded into both reactors and the reactors have been operated for 90 days.

For semi-aerobic / anaerobic treatment, the mixture waste rates were determined as 20% OFMSW + 80% SS (RSA1) and the second reactor was 100% SS (RSA2). Reactors have been operated for 90 days. Sewage sludge supplied from Denizli Wastewater Treatment Plant Anaerobic Sludge Tank was used as inoculum for bioreactors. Table 1 shows the amount of lignin, cellulose, hemicellulose (%) of sunflower stalk used in reactors. Table 2 shows the results of the water content and organic matter analysis of the prepared waste mixtures before loading into the reactors.

### Analytical Methods

The total COD was measured colorimetrically by the closed reflux method in bioreactor leachate samples [15]. Leachate samples' pH were measured using the model pH meter HANNA HI 221. Volatile fatty acid concentration was measured by the titrimetric method determined by Anderson and Yang [16]. BOD<sub>5</sub> was measured using the WTW Oxi Top IS system. Ammonium nitrogen and Total Kjeldahl Nitrogen analyses were performed with standard method. Total gas generated in the bioreactors was measured by passing through a solution containing 10% NaCl and 2% H<sub>2</sub>SO<sub>4</sub> using the liquid displacement method [17]. The methane gas generated in the bioreactors was measured by passing through a solution containing 3% NaOH using the liquid displacement method [18]. The analyses of water content and organic matter in solid waste were carried out before and after loading in bioreactors according to the standard method [15]. The contents of hemicellulose, lignin, and cellulose were determined using the processes specified by [19].

## RESULTS AND DISCUSSION

### Sunflower Stalk Biodegradation in Anaerobic Bioreactors

#### COD, pH, VFA Variations in Leachate Produced from Anaerobic Bioreactors

Figure 2 (a) shows change in COD concentrations. In leachate samples from anaerobic bioreactors, the initial COD concentrations were, respectively; 22026 mg/l in RA1 and 14250 mg/l in RA2. The COD removal yields were calculated as 90% in RA1 and 97% in RA2. Dumlu [20] reported that the digestion of agricultural waste consisting of roots, stems, leaves, and fruits resulting from the production of tomatoes, peppers, cucumbers, eggplants, and zucchini was achieved as a result of the 60.49% COD removal yield of 100 days of anaerobic digestion. The cellulose content of the agricultural waste used in that study was 26.40% and the lignin content 12.28%. It can be said that the sunflower stalk used in our study has a lower cellulose content than in this study, and higher COD removal yields are achieved as a result of mixing the sunflower stalk with OFMSW.

Acid accumulation occurred in bioreactors in the first stages of anaerobic treatment and a decrease in pH values. This situation was observed in Figure 2 (b). In order to provide alkalinity to the environment and to increase the pH values, 0.6% NaHCO<sub>3</sub> solution was added until the desired pH values were obtained at regular intervals from the 21<sup>st</sup> to the 51<sup>st</sup> day of the enterprise. pH values were measured between 4.96-7.43 and 4.98-7.48, respectively, in RA1 and Rector2.

It has been reported that VFA occur during the acidogenesis stage in anaerobic breakdown and often cause a decrease in pH, while low pH affects the activity of methanogens and thus affects a rapid decrease in methane production [21]. The VFA concentrations measured on day 38 were 6335 mg/l and 3290 mg/l in RA1 and RA2, respectively (Figure 2 (c)). VFA concentrations decreased in a similar way to COD concentrations [22]. During the operation of the reactors, RA2 had higher pH values than RA1. As a result, lower VFA concentrations were measured in RA2. VFA is inversely proportional to pH, meaning that the higher the occurrence of VFA, the lower its pH [23].

#### Cumulative Gas Generated in Anaerobic Bioreactors

During the decomposition of wastes under anaerobic conditions, organic matter is converted into biogas through microbial activities [24]. The cumulative total gas was measured as 12.42 L in RA1 and 18.88 L in RA2 over a 90 days operating period respectively (Figure 3). The high ratio of sunflower stalks, which had a high content of cellulose and

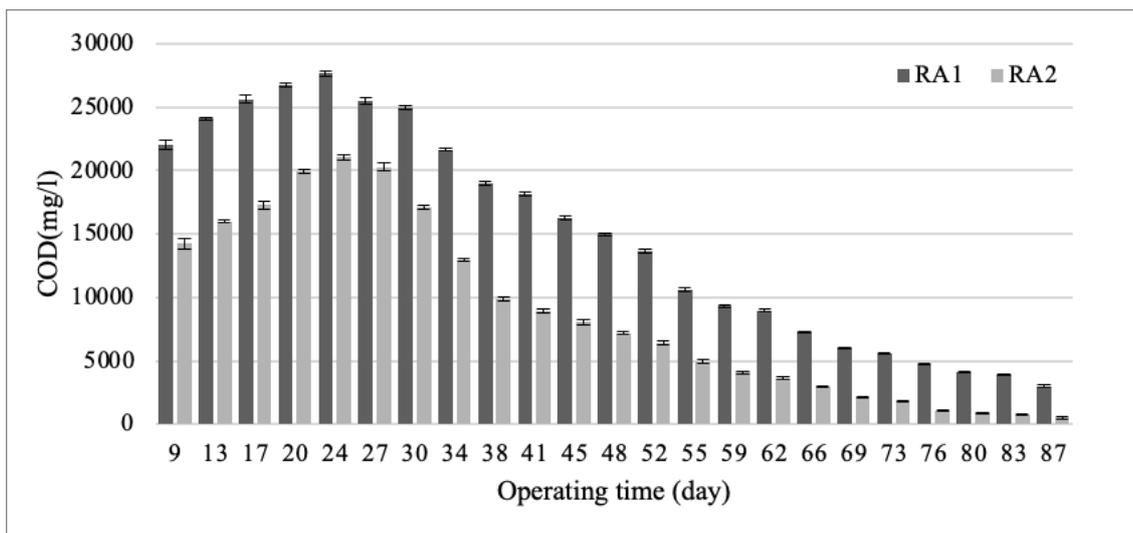
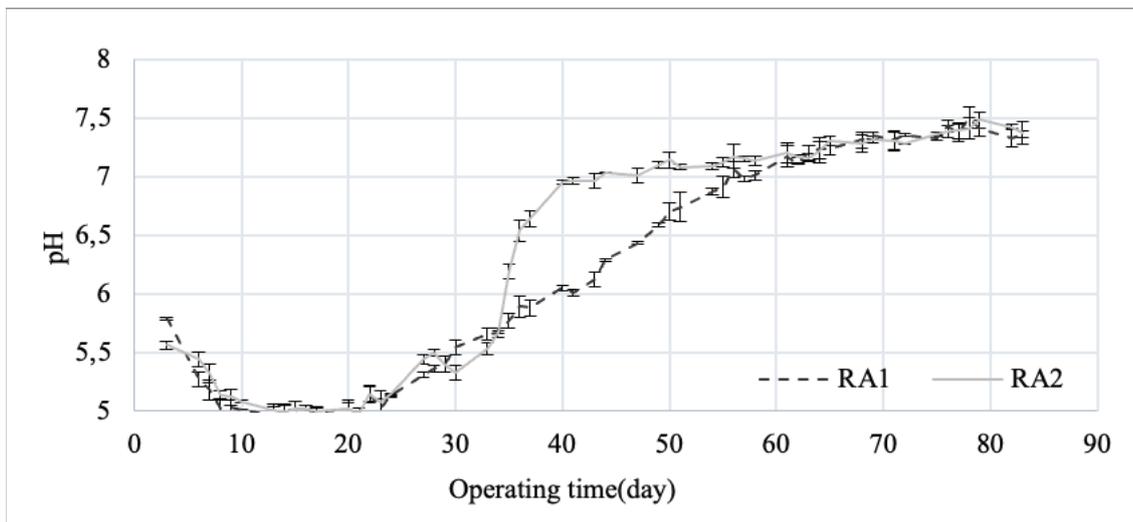
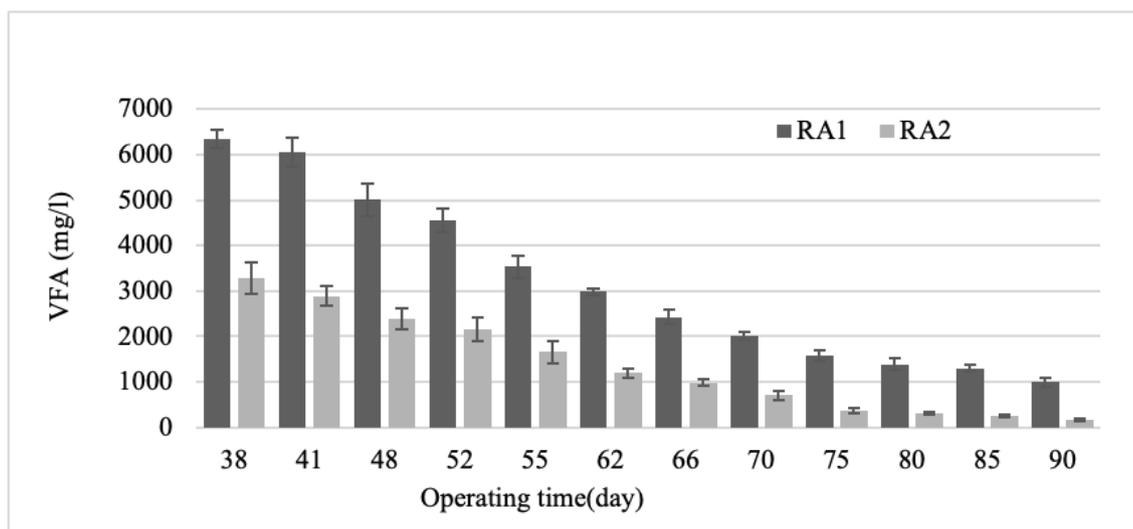


Figure 2. (a) Change in COD concentrations.



(b) pH change of bioreactor leachate.



(c) Change in VFA concentrations.

lignin in RA1 was caused to reducing of the total amount of gas. Especially in anaerobic conditions, the difficult decomposition of wastes with a high lignin and cellulose content is the most important factor in this process. Since the decomposition was slow, the total gas formation was also slow.

**Cumulative Methane Gas Production in Anaerobic Bioreactors**

The methane gas measured at the beginning of period is thought to be produced by hydrogen-consuming methane

bacteria. The cumulative quantities of methane calculated during the 90-day operating period were measured as 3.90 L (Figure 4 (a)) in RA1 and 8.13 L (Figure 4 (b)) in RA2, respectively. The percentage of methane in the reactors is 34% in RA1 and 44% in RA2 during the operating period. In the study, in which different MBT (mechanically biologically treated) wastes containing cellulose were used, approximately half of our reactors were used, it was observed that the methane content varied between 58% and 62% [25]. In another study, Zhurka et al. [5] found a methane potential

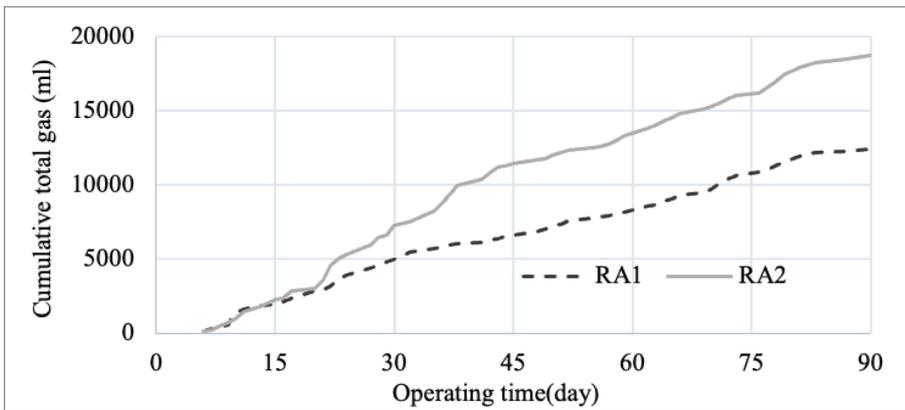


Figure 3. Cumulative total gas generated in bioreactors.

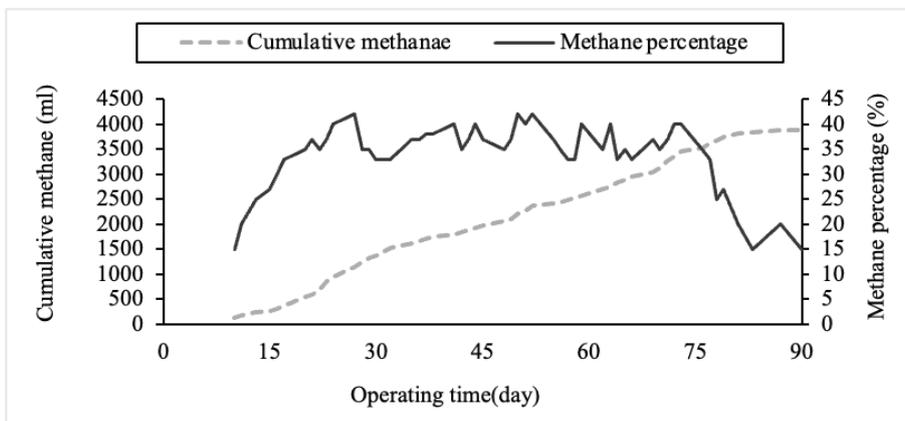
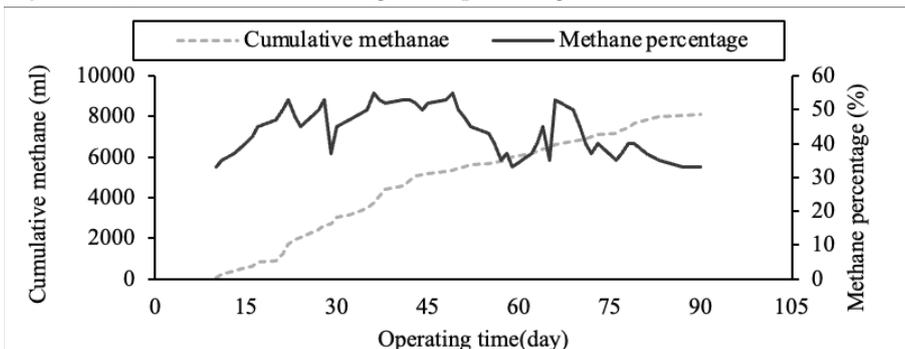


Figure 4. (a) Cumulative methane gas and percentage of methane formed in RA1.



(b) Cumulative methane gas and percentage of methane formed in RA2.

of 132 mL CH<sub>4</sub> g<sup>-1</sup> raw vs. without pretreatment and 144 mL CH<sub>4</sub> g<sup>-1</sup> raw vs. from sunflower stalks using identical alkaline pretreatment.

The highest percentage of methane was 42% in RA1 and 55% in RA2. Lignocellulosic materials are limited by their slow degradation and therefore low methane yield [26]. In parallel with the fact that the amount of sunflower stalk (with a 24.46% cellulose content) is higher in RA1 than in RA2, the amount of methane formed is less. The excess food waste in RA2 has a higher methane potential than the lignocellulosic biomass [27]. In addition, an increase in methane gas amounts was observed in parallel with COD removal yields.

**Variations in BOD<sub>5</sub> Concentrations in Leachate Produced from Anaerobic Bioreactors**

The BOD<sub>5</sub> measurements at different 5 days during the operation period of the bioreactors are shown in Table 3. The BOD<sub>5</sub>/COD ratio is indicative of stabilisation in reactors, and the BOD<sub>5</sub>/COD ratio is initially 0.16 in RA1 and 0.18 in RA2. The BOD<sub>5</sub>/COD ratio between 0.02 and 0.13 means a low biodegradability and indicates an excess amount of organic compounds that are difficult to decompose [28]. On the 34<sup>th</sup>, 45<sup>th</sup> and 59<sup>th</sup> days of the operation, an increase in the BOD<sub>5</sub>/COD ratio was observed as the organic materials in the reactors were broken down and

passed into the leachate. On day 73, as a result of the consumption of organic substances by methane bacteria, a decrease in the BOD<sub>5</sub> / COD rates was observed.

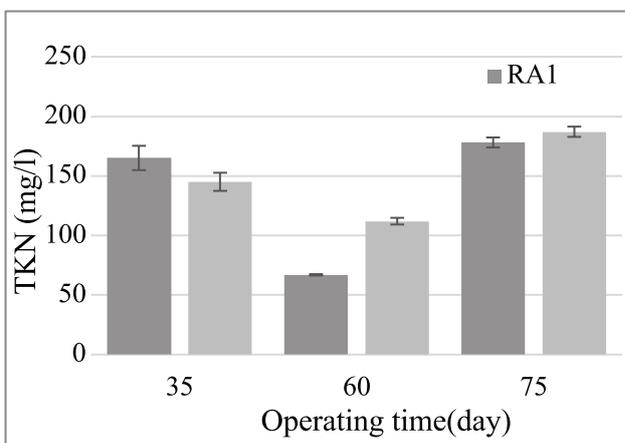
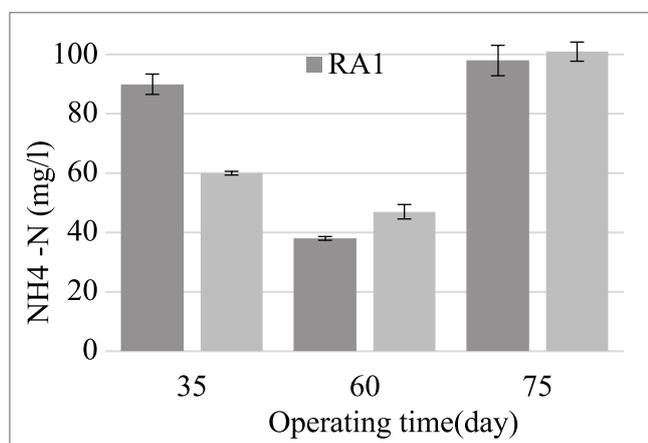
As a result of the loading of sunflower stalk wastes with high content of lignin into the reactors, low BOD<sub>5</sub>/COD ratios are observed. When BOD<sub>5</sub> removal yields are calculated, it is 76% in RA1 and 92% in RA2.

**Variation of NH<sub>4</sub>-N and TKN in Leachate Produced from Anaerobic Bioreactors**

When proteins break down as a result of the breakdown of organic matter, ammonium ions are released. These ammonium ions are potent inhibitors of methanogenic bacteria [29]. During the operation of the bioreactors, NH<sub>4</sub>-N (Figure 5(a)) and TKN (Figure 5(b)) concentrations were measured on 3 separate days. Low concentrations of NH<sub>4</sub>-N and TKN were measured on day due to the use of anaerobic microorganisms in the system for their N and amino acid needs, as organic materials decompose and pass into leachate. On day 75, an increase in NH<sub>4</sub>-N and TKN concentrations was observed again. This increase in concentrations indicates that the degradation of the sunflower stalk with a high lignin and cellulose content is still ongoing. Furthermore, continuous leachate recirculation in all reactors is also effective in this increase.

**Table 3.** Change in the BOD5 concentrations of bioreactor leachate

	RA1				RA2					
	20 <sup>th</sup> day	34 <sup>th</sup> day	45 <sup>th</sup> day	59 <sup>th</sup> day	73 <sup>rd</sup> day	20 <sup>th</sup> day	34 <sup>th</sup> day	45 <sup>th</sup> day	59 <sup>th</sup> day	73 <sup>rd</sup> day
BOD <sub>5</sub> (mg/l)	4275±469	5200±742	5376±223	3354±134	1224±202	4978±323	4538±142	2978±169	1345±183	357±26
COD (mg/l)	26714±142	21669±164	16291±55	9315±90	5565±70	19912±159	12965±127	8050±164	4075±117	1785±36
BOD <sub>5</sub> / COD	0.16	0.24	0.33	0.36	0.22	0.25	0.35	0.37	0.33	0.2



**Figure 5.** (a) Variation of NH<sub>4</sub>-N concentration

(b) Variation of TKN concentration

## Sunflower Stalk Biodegradation in Semi-aerobic/ Anaerobic Bioreactors

### COD, pH, Variations in Leachate Produced from Semi-aerobic/Anaerobic Bioreactors

In leachate samples of semi-aerobic/anaerobic bioreactors, initial COD concentrations were 10181 mg/l and 8777 mg/l in RSA1 and RSA2, respectively (Figure 6 (a)). Toptas and Yay [30], in the intermittent air, increased COD concentrations in the first 38 days and the highest value was measured at 29800 mg/l. There has been a rapid decline since day 38. The COD concentration measured at the end of operation in the intermittent reactor operated for 245 days is 396 mg/l. It is seen that the oxygen supplied to the system from here has a faster decomposition effect. Similar to this study, the COD concentration in RSA1 was 651 mg/l and the COD concentration in RSA2 was 1326 mg/l as a result of the 90 days operating period. The COD removal efficiency is 97% in RSA1 and 94% in RSA2.

The optimum pH range for methane bacteria in mesophilic systems is between 6.5-8.0. When the pH increases below 6.0 and above 8.3, the process is severely restricted [31]. Figure 6 (b) shows an increase in pH values is observed

from the 10th day. This increase in pH values shows that the hydrolysis and acidification phases occur faster with the aeration effect in the reactors. Cossu et al. [32] showed that pre-aeration had a beneficial effect on the increase in pH in the subsequent anaerobic phase. The highest pH values in the reactors were measured on day 42, which were 7.77 in RSA1 and 8.47 in RSA2. During the 90 days operating period, the pH values in the reactors were observed to be between 5.57 and 7.77 in RSA1 and 5.64 to 8.47 in RSA2, respectively.

### Cumulative Gas Generated in Semi-aerobic/ Anaerobic Bioreactors

The cumulative gas quantities during the 90 days operating period were measured as 26.64 L in RSA1 and 39.6 L in RSA2 (Figure 7). Due to intermittent air supplied in the first 41 days of operation of the reactors, pH values were measured higher than those of the anaerobic system. When the amount of gas generated in reactors is compared, the total amount of gas in RSA2 is considerably higher than in RSA1. Although there are 100% sunflower stalks in RSA2, the amount of gas formed in parallel with the pH values is higher in RSA2 because the pH values are higher than in RSA1.

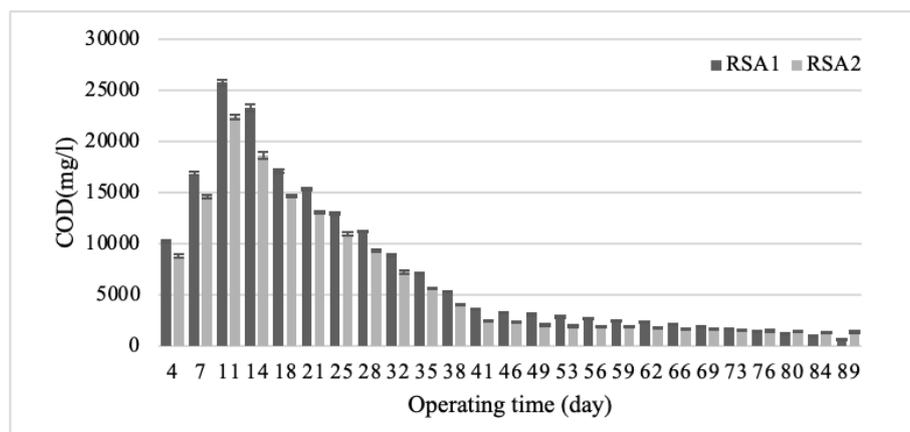
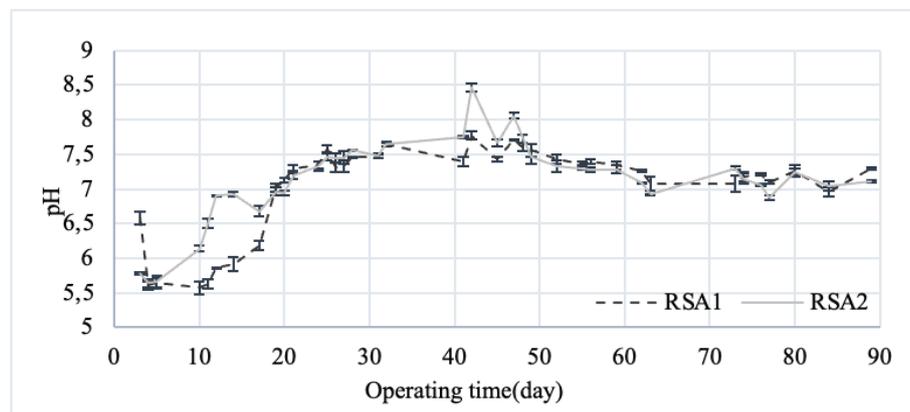


Figure 6. (a) Change in COD concentrations.



(b) Changes in pH of the bioreactor leachate.

**Cumulative Methane Gas Production in Semi-aerobic / Anaerobic Bioreactors**

Traditionally, exposure to oxygen or air is avoided in anaerobic digestion systems to minimise its negative impact on anaerobic microorganisms. However, it has recently been found that methanogenesis also occurs outside of anaerobic environments such as oxygenated freshwater and soil [33]. Nguyen and Khanal [34] reported that exposure

to small amounts of oxygen / air (micro-aeration) benefits anaerobic digestion by increasing hydrolysis, improving methane yield, stabilizing the process, and removing hydrogen sulphide, among others. It has been observed that a high rate of methane gas is formed in reactors compared to the anaerobic system due to the air supplied to the reactors (Figure 8 (a) and (b)). When the aeration process in the reactor was completed and the anaerobic operation was

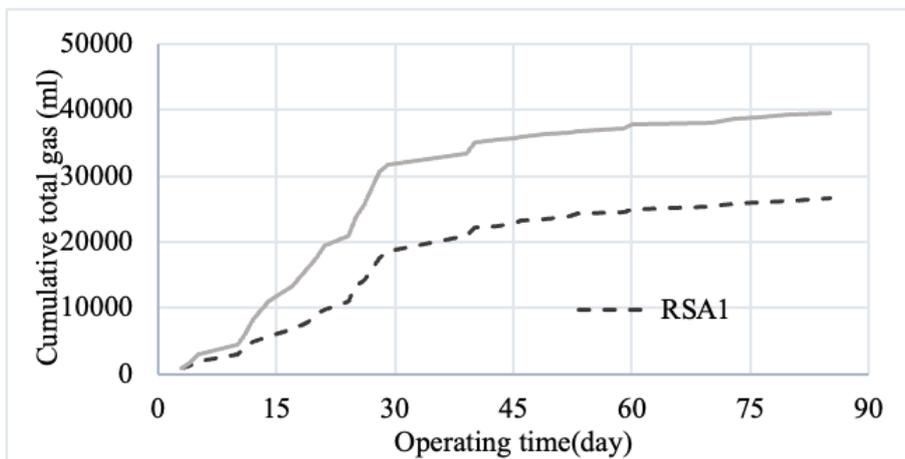


Figure 7. Cumulative total amount of gas generated in bioreactors.

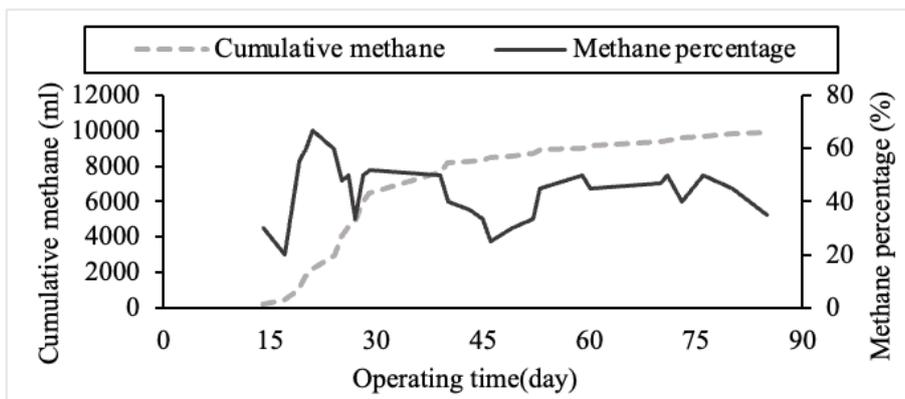
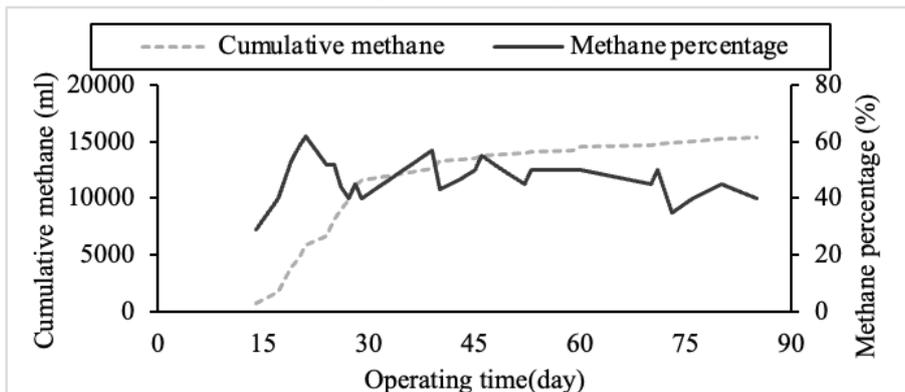


Figure 8. (a) Cumulative methane gas and percentage of methane formed in RSA1.



(b) Cumulative methane gas and percentage of methane formed in RSA2.

continued, less methane gas was formed than the aerobic part. In the aerobic part, methane bacteria consumed the organic substances in the environment and when the anaerobic part was passed, less methane gas was formed than the aerobic part due to the fact that there was less organic matter in the environment. Cumulative quantities of methane in semi-aerobic / anaerobic bioreactors were 9.93 L in RSA1 and 15.36 L in RSA2, respectively. The percentage of methane in the reactors was 44% of the total gas in RSA1 and 47% in RSA2, respectively, during the operating period. The percentage of methane was highest on day 21 at 66.6% in RSA1 and 62% in RSA2.

#### Variations in BOD<sub>5</sub> Concentrations in Leachate Produced from Semi-aerobic / Anaerobic Bioreactors

According to Table 4, it was observed that organic substances digest and pass through leachate through the air distributed to the reactors intermittently before the anaerobic system. On day 38, the BOD<sub>5</sub> / COD ratios increased to 0.4 in RSA1 and 0.35 in RSA2, which is an indication that organic substances are broken down and passed into leachate. BOD<sub>5</sub> concentrations on day 80 were 154 mg/l and 262 mg/l in RSA1 and RSA2, respectively. The BOD<sub>5</sub> / COD ratios are 0.13 in RSA1 and 0.19 in RSA2.

#### Variation of NH<sub>4</sub>-N and TKN in Leachate Produced from Semi-aerobic/Anaerobic Bioreactors

Since the reactors were intermittently aerated during the first 41 days of operation, a decrease in TKN and

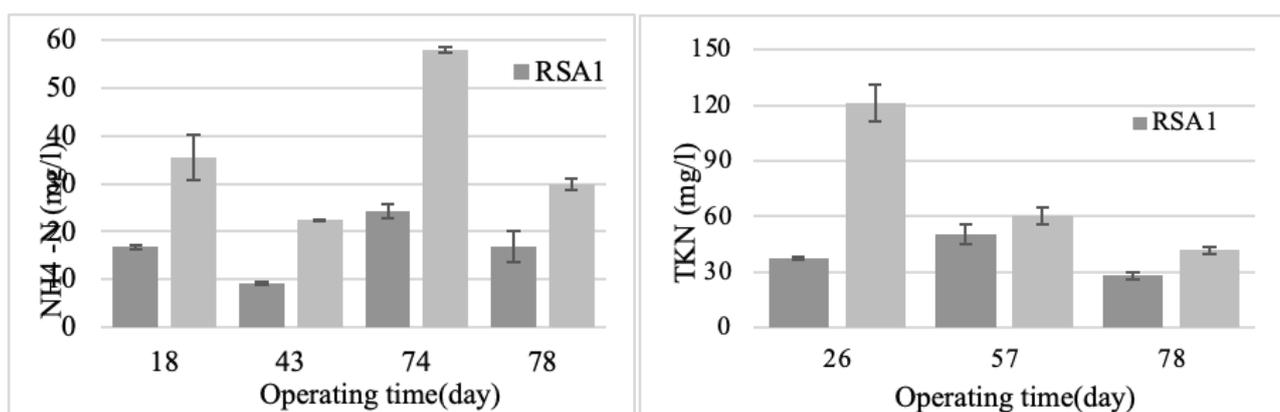
NH<sub>4</sub>-N was observed (Figure 9 (a) and (b)). An increase in NH<sub>4</sub>-N concentrations was observed when the system was operated anaerobically after intermittent aeration. Nag et. al. [35] evaluated the aerobic-anaerobic storage method using intermittent aeration. The mass of ammonium nitrogen decreased sharply from its high values as a result of the stripping of air and the removal of free ammonia. The observed decrease in NH<sub>4</sub><sup>+</sup> concentrations is in line with findings from previous studies showing that hybrid conditions created by cyclic/intermittent aeration are suitable for reducing total nitrogen, especially ammonium concentrations.

#### Comparison of Anaerobic and Semi-aerobic/Anaerobic Bioreactors

Although RA1, one of the anaerobic bioreactors, and RSA1, one of the semi-aerobic / anaerobic bioreactors, had the same mixture ratios of wastes, in RA1, the hydrolysis phase, which is one of the advantages of the aerobic process, took place faster and in a shorter time compared to the anaerobic system, so the organic substances contained in the wastes passed into the leachate in a shorter time and higher removal efficiencies were obtained. In RSA2, although 100% sunflower stalks were used, COD decreased at faster rates in short periods in both reactors. This is because the aerobic process is faster than the anaerobic process. The COD concentrations measured at the end of a long operating period indicate that wastes with high lignin and cellulose content are better decomposed under

**Table 4.** Change in the BOD<sub>5</sub> concentrations of bioreactor leachate

	RSA1				RSA2			
	21 <sup>st</sup> day	38 <sup>th</sup> day	56 <sup>th</sup> day	80 <sup>th</sup> day	21 <sup>st</sup> day	38 <sup>th</sup> day	56 <sup>th</sup> day	80 <sup>th</sup> day
BOD <sub>5</sub> (mg/l)	5058±613	2119±257	394±84	154±12	3923±209	1394±111	462±42	262±15
COD (mg/l)	15328 ±108	5299±42	2626±95	1186±19	13078±113	3983±55	1848±51	1377±60
BOD <sub>5</sub> / COD	0.33	0.4	0.15	0.13	0.3	0.35	0.25	0.19



**Figure 9.** (a) Variation of NH<sub>4</sub>-N concentration

(b) Variation of TKN concentration

semi-aerobic / anaerobic operating conditions than anaerobic process.

The pH values in semi-aerobic / anaerobic operating conditions are higher than the pH values in anaerobic operating conditions. The reason for this is that the hydrolysis and acidification phases of the intermittent air supplied to the reactors take place at shorter times and the reactors pass to the methanization phase faster.

The total amount of gas measured in semi-aerobic / anaerobic operating conditions is higher than anaerobic bioreactor. The decomposition of sunflower stalks with a high lignin and cellulose content in semi-aerobic / anaerobic conditions occurs faster than under anaerobic operating conditions, and the pH values are higher due to the intermittent air, which causes more gas to be formed.

In comparison to anaerobic operation, the amount of methane gas formed in semi-aerobic / anaerobic operation conditions was higher than in the anaerobic system. This is since the hydrolysis and acidification phase is completed in shorter periods of time and then the transition to the methanation phase is faster due to the aeration of semi-aerobic / anaerobic bioreactors with intermittent air for 41 days. Ebrahimi et al. found that methane production from untreated stem and capitulum was very low compared to other crude samples and suggested that a pretreatment is required for anaerobic degradation of lignocellulosic biomass [36]. As in this study, aerobic pretreatment confirms the increase in the methane gas amount.

At the end of the experiment, it was observed that the removal of ammonium nitrogen in aerobic-anaerobic reactors was slightly higher than in the aerobic reactor. Studies in the literature show that a semi-aerobic bioreactor landfill can not only accelerate the landfill stabilization process and reduce the concentration of organic matter in the leachate, but also reduce the concentration of ammonia in the leachate [37, 38].

Due to intermittent aeration made due to the loading of sunflower stalk waste with a high lignin content of lignin into the reactors, higher BOD<sub>5</sub> / COD rates are observed according to anaerobic operating conditions with the breakdown of organic materials and their passage into leachate. When the BOD<sub>5</sub> removal yields are calculated, it is 97% in RSA1 and 93% in RSA2.

## CONCLUSION

According to the analysis results, 90% and 97% COD removal, 84% and 94% VFA removal, 76% and 92% BOD<sub>5</sub> removal and 43% and 52% organic matter removal were achieved in RA1 and RA2, respectively. The cumulatively calculated total gas amounts were 12.42 L and 18.88 L in RA1 and RA2, respectively. Cumulative methane amounts were 3.895 L in RA1, 8.13 L in RA2 and methane percentage was measured as 34% in RA1 and 44% in RA1. It was observed that the decomposition rate was slower in the reactor with more sunflower stalks than in the reactor with more domestic waste due to the high lignin and cellulose

content. In the next operating condition, semi-aerobic / anaerobic operating conditions, the reactor with more sunflower stalks was selected and its degradation was observed under semi-aerobic / anaerobic conditions.

Under semi-aerobic / anaerobic operating conditions, sunflower stalks were mixed with organic fraction of municipal solid waste at a ratio of 20% OFMSW + 80% SS in RSA1 and 100% sunflower stalks in RSA2 and their decomposition and methane production in bioreactors were observed for 90 days. The reactors were intermittent aerated for 41 days and anaerobically operated for 49 days for methane production. Under semi-aerobic operation conditions, RSA1 and RSA2 achieved 86% and 89% COD removal, respectively. After 90 days of operation, RSA1 and RSA2, respectively; 97% 94% COD removal, 97% 93% BOD removal and 60% and 58% organic matter removal were achieved. The cumulatively calculated total gas amounts of RSA1 and RSA2 were 26.64 L and 39.6 L, respectively. Cumulative methane amounts in RSA1 and RSA2; 9,929 L and 15,351 L methane gas was measured and the percentage of methane in the reactors was 44% and 47% of the total gas.

Due to the high lignin and cellulose content of agricultural waste, the decomposition process takes place in long stages with anaerobic treatment methods. Therefore, it has been determined that the decomposition of agricultural wastes with a pre-aerobic treatment to accelerate the decomposition process and then their anaerobic operation for methane production contribute to the decomposition process of the wastes.

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## CONFLICT OF INTEREST

The authors declare no potential conflicts of interest regarding the research, authorship and/or publication of this article.

## DATA AVAILABILITY

The data used to support the findings of this study are included within the article.

## AUTHOR'S CONTRIBUTIONS

All authors are contributed equally to bring out this article.

## ETHICS

There are no ethical issues with the publication of this manuscript.

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