



Comparative Assessment of Biogas Production Potential of the Most Abundant Agro-residues in Turkey

Türkiye'deki Yaygın Tarımsal Atıkların Biyogaz Üretim Potansiyelinin Değerlendirmesi

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Abstract

Recent energy policies promote energy generations from green resources to meet sustainability criteria. Since Turkey is one of the largest agricultural producers globally, it has great biogas production potential. This study aims to evaluate the biomethane yields of the most abundant agro-residues in Turkey and to assess their potentials for contribution to biogas production. Within this scope, sunflower heads, tea residues, cotton stalks, and crop residues; wheat, rye, and triticale straws were collected from different regions of Turkey. Anaerobic batch digesters were conducted to investigate the biomethane production of the selected feedstock and operated for 30 days at 37°C. Each setup was conducted in triplicates and methane productions were monitored online. The main methane production route of the inoculum was determined as acetoclastic methanogenesis while *Cloacimonetes*, *Firmicutes*, and *Bacteroidetes* composed the core bacterial phyla. The greatest methane yield was observed in the digesters operated with the wheat straw followed (164 NmL/gVS_{influent}) by triticale straw and sunflower head. The lowest yields were calculated for the digesters fed with the cotton stalks (71 NmL/gVS_{influent}). To increase the biomethane potential yields in the anaerobic digesters operated with agro-residues and to make the anaerobic digesters more feasible, operational conditions should be optimized and physico-chemical and biological pre-treatment techniques and/or bioaugmentation applications should be integrated into the systems.

Keywords: *Agricultural residues, Anaerobic digestion, Biomethane, Renewable energy, Waste management*

Öz

Günümüzdeki enerji politikaları, sürdürülebilirlik kriterlerinin sağlanması yeşil kaynaklardan enerji üretimini teşvik etmektedir. Dünyadaki en büyük tarım üreticilerinden biri olan Türkiye, büyük bir biyogaz üretim potansiyeline sahiptir. Bu çalışmada, Türkiye'deki yaygın tarımsal atıkların biyometan verimlerinin incelenmesi ve biyogaz üretimine katkı potansiyellerini değerlendirilmesi amaçlanmaktadır. Bu kapsamda, Türkiye'nin farklı bölgelerinden substrat olarak değerlendirilmek üzere tahıl atıklarından buğday, çavdar ve tritikale ile ayçiçeği başları, çay artıkları, pamuk sapları

toplanmıştır. Seçilen atıkların biyometan üretiminin belirlenmesi için anaerobik çürütücüler kurularak, 30 gün süre ile 37 ° C'de kesikli olarak işletilmiştir. Çürütücü deney setleri üç tekrarlı olarak kurulmuş, metan üretimleri eş zamanlı kaydedilmiştir. Aşı çamurundaki metan üretiminin ağırlıkla asetoklastik metanojenik yolağı izlediği belirlenirken, bakteriyel komünite *Cloacimonetes*, *Firmicutes* ve *Bacteroidetes* türlerinden oluşmuştur. En yüksek metan verimi buğday samanı ile işletilen çürütücülerde gözlenirken (164 NmL/gUKM_{giriş}), bunu tritikale samanı ile ayçiçeği başları izlemiştir. En düşük biyometan verimi pamuk sapları ile işletilen çürütücülerde kaydedilmiştir (71 NmL/gUKM_{giriş}). Tarımsal atıklarla işletilen çürütücülerde biyometan potansiyel verimlerini artırmak ve anaerobik çürütücüleri daha uygulanabilir hale getirmek için, işletme koşulları optimize edilmeli ve fiziko-kimyasal ve biyolojik ön arıtma teknikleri ve / veya biyoaugmentasyon uygulamaları sistemlere entegre edilmelidir.

Anahtar Kelimeler: Anaerobik çürütme, Atık yönetimi, Biyometan, Tarımsal atık, Yenilenebilir enerji

1. Introduction

Renewable energy alternatives have come into prominence concerning to maintain energy security and balance energy costs as well as pressures on greenhouse gas emissions. Thus, serious actions and plans are taken into account globally at the international and national levels [1]. The European Union (EU) set stringent targets for a sustainable society that intends a 40% reduction in greenhouse gas emissions. Moreover, they targeted to enhance the installed energy generation capacity by 27% from renewable sources until 2030 [2]. In order to diversify energy sources, the Turkish government released Turkey's Renewable Energy Action Plan and targets to maximize the share of energy production from green sources to 20% by 2023 [3]. These policies have promoted bioenergy generation from green resources.

Anaerobic digester technology can fulfill these targets. It not only generates green energy (e.g. electricity, heat, and fuel), but it also contributes to waste management and the reduction of greenhouse gas emissions [4,5]. The developments on anaerobic digesters enable us to get closer to net-zero emissions in power generation [6]. Therefore, governments incentivize the spread of anaerobic digesters due to economic and environmental profits. It is projected that biogas plants will reach the capacity of almost 20x10⁹ m³ and meet almost 3% of the EU's current gas consumption by 2030 [4].

The flexibility of anaerobic systems enables operation with a wide variety of feedstock including energy crops, manure, municipal solid waste, etc., and so widens their implementations

from farm to large scale [7-9]. Since food security comes first in crop production, the selection of feedstock is critical for anaerobic digesters [5]. Due to the upward trend of bioeconomy, the valorization of wastes has become the center of interest [6]. Agricultural residues such as crop leftovers are one of the most favorable feedstocks for anaerobic digesters because of their high abundance and low nutritional values. Two types of agro-residues are produced in the agricultural sector; while primary residues are generated in the field during harvesting include straws, stalks, etc., secondary residues are generated during processing include husk, bagasse, etc. [10]. They are generally spread in the fields contributing carbon content of the soil and/or used as bedding for livestock in the farms [2].

Agriculture has crucial importance in Turkey in which the country is accounted for as the seventh-largest global producer [11]. According to TUIK, total productions of the selected feedstocks are as follows; wheat (durum and regular in total): 19 million ton, cotton: 3 million tone, sunflower 2.1 million ton, tea: 1.4 million ton, rye: 310,000 ton and triticale 274,136 ton in 2019 and the crop yields in the corresponding fields can be ordered as cotton (unginned: 460 kg/daa) > sunflower (for oil: 289 kg/daa) > rye (277 kg/daa) > wheat (276 kg/daa) > cotton(ginned:170 kg/daa) [12].

Due to the agro-industrial activities, a large amount of residues is generated in the fields, and the country holds great potential for biomass-based energy production which has not taken into account for long periods. However, it is projected that the country's energy demand will be increased in the future and the government

targets to fulfill this need on time with affordable tariffs [13].

Biomethane potential test (BMP) is a common method to evaluate the amount of methane generated from various feedstock which is an important issue to determine the suitable substrates for biogas plant operation [14,15]. Besides showing a methane potential, BMP also delivers the methane production over time and the data is generally given as a specific methane production (SMP) curve [14].

Inoculum selection is one of the key factors in anaerobic processes and seed sludge should include the necessary microorganisms to perform all steps of anaerobic degradation, namely, hydrolysis, acidogenesis, acetogenesis, and methanogenesis [9]. Next-generation sequencing methods are commonly used molecular techniques to reveal the microbial composition of the biogas plants [16–18] as well as the seed sludge before the start-up of anaerobic digesters [9,19]. Rapid developments in the next-generation sequencing platforms enable greater sequencing depth and high-resolution analyses [20] which provide deeper information about the community structure.

The specific aim of this paper was to examine the biomethane potential of the most common agricultural residues in Turkey. For this purpose, six different agro-residues that are spread in the

fields with large availability were selected and tested in the batch anaerobic digesters and biomethane yields were evaluated.

2. Material and Method

2.1. Inoculum and Agro-residues

The inoculum (active seed sludge) was collected from a large-scale anaerobic digester operated at 37° C, in Antalya, Turkey. Within all agro-residues in Turkey, wheat, rye, and triticale straws, cotton stalk, sunflower head, and tea residues were selected as feedstock in this study. Whereas wheat, rye, and triticale straws were collected in Bursa, tea residues were collected from Rize, sunflower heads were provided from Edirne, and cotton stalks were received from Aydin. Physicochemical characterizations of the substrates and anaerobic inoculum were determined by total solids (TS), volatile solids (VS), sCOD, and alkalinity parameters according to standard methods [21]. pH levels were measured by a HANNA HI 221 Microprocessor pH meter (Germany). The physicochemical characterization of the inoculum and feedstock were presented in Table 1. Bacterial and methanogenic archaeal community patterns of the inoculum were determined by next-generation sequencing-based metagenomics by the Ion Torrent PGM® platform as described in Ince et al. [22].

Table 1. Physico-chemical properties of the agro-residues & inoculum

Inoculum / Agro-residues	pH	VS/TS	Total Solids (TS) (%)	Volatile Solids (VS) (%)	sCOD (mg/L)
Anaerobic Inoculum	7.52	63	3.5	2.2	7400
Tea residues	6.20	90	52.0	47.6	6800
Cotton stalk	6.90	86	79.0	68.0	4400
Rye straw	6.75	89	92.8	82.4	5800
Triticale straw	6.58	93	81.1	75.3	6310
Wheat straw	6.47	94	82.0	77.4	6890
Sunflower head	7.20	83	89.1	74.0	5180

2.2. Biomethane Potential Test

Biomethane production potentials of the selected agro-residuals were determined using the Automatic Methane Potential Test System (AMPTS) II (Bioprocess Control, Sweden) with 24 reactors in two AMPTS runs according to the

protocol described by Ozbayram et al. [23]. The experiments were conducted in 500 mL glass reactors with a working volume of 400 mL. The inoculum/substrate ratio was 2 based on volatile solids, the substrates were milled and their sizes were reduced before the experiments. Triplicate reactors were set up for each feedstock and

operated in batch mode for 30 days under mesophilic conditions (37°C). The reactors were flushed with N₂ gas for the provision of anaerobic conditions. Online methane production was recorded by the counting unit of AMPTS. Furthermore, the blank reactors were set up to subtract the background methane production from the inoculum.

3. Results

In this study, the biomethane potentials of the most abundant agro-residues in Turkey were assessed. For this purpose, before the digester experiments, the inoculum's microbial community structure was examined. The bacterial community pattern of the inoculum was presented in Figure 1. *Cloacimonetes*, was the most abundant phylum representing 25% of the total reads of the bacterial community. It was followed by *Bacteroidetes*, *Firmicutes*, and *Proteobacteria* species. Only less than 1% of the total reads could not be assigned to any known bacterial taxa.

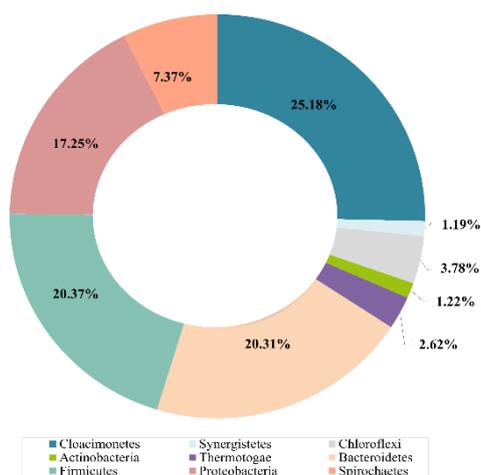


Figure 1. Bacterial community patterns of the anaerobic inoculum

The methanogenic community structure of the anaerobic inoculum was given in Figure 2. More than half of the total reads were affiliated to *Methanosaetaceae* species followed by *Methanobacteriaceae* and *Methanospirillaceae*.

Together, they represented more than 95% of the total sequences. The portion of the total reads which could not be assigned to any taxa was negligible (<0.5%).

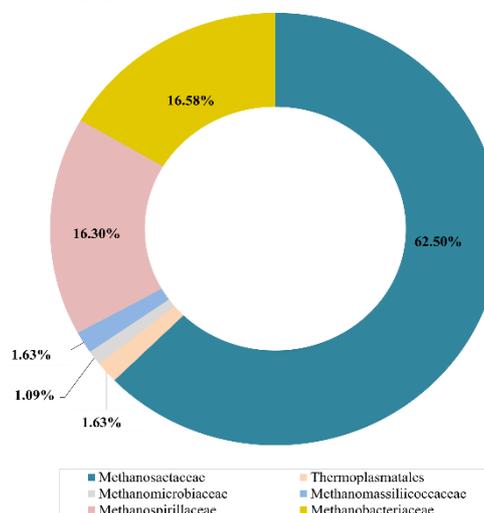


Figure 2. Methanogenic Archaeal community patterns of the anaerobic inoculum

The biomethane potentials of the selected agro-residues were depicted in Figure 3. Biomethane productions were almost completed in two weeks in the digesters operated with triticale straw and sunflower heads and reached the plateau. For the rest of the substrates, the observation of the constant methane values took a week longer. Besides, the biomethane production did not change in the last week of the experiment and the experiment was ended on the 30th day of the operation period. A sharp increase in the biomethane production was observed in the first week of the operation period for the crop residues triticale and wheat straw as well as the sunflower heads. While the methane production continued in the digesters operated with the wheat straw, it almost stopped in the digesters with triticale straw and sunflower head. During the first 10 days of the operation, methane yields were calculated almost the same for tea residues and cotton stalks, higher methane yields were observed in the digesters operated with tea waste.

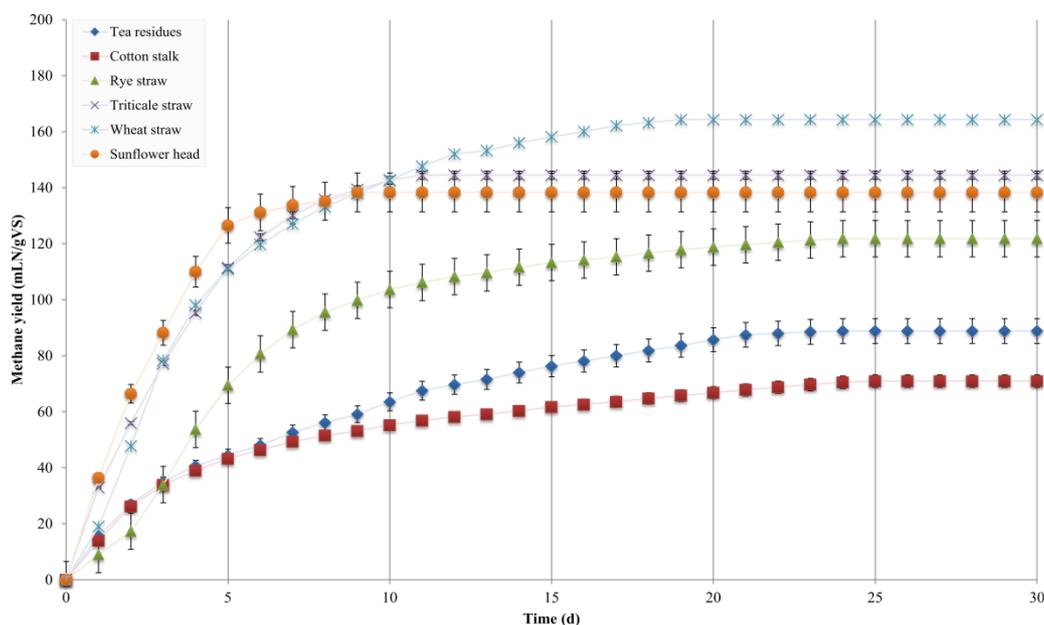


Figure 3. Biomethane potential of the selected agro-residues

The biomethane production of the rye straw was quite similar to the cotton stalk during the first three days of the operation period. However, the difference was started to increase after that day and the methane yield was almost doubled at the end of the operation. The highest amount of methane was measured in the reactors operated with the wheat straw (164 NmL/gVS_{influent}), followed by triticale straw and sunflower head. The cumulative biomethane yield in the reactors fed with rye straw was calculated as 122 NmL/gVS_{influent}. The lowest yields were calculated for the digesters operated with cotton stalks (71 NmL/gVS_{influent}).

The total energy potentials of the selected feedstock produced in Turkey in 2019 were calculated and presented in Table 2. According to the data obtained from TUIK [12], wheat was the major crop produced in the country holding the potential to produce nearly 26.5 billion kWh energy. The highest production was in Konya. On the other hand, during cotton production, a great amount of residue, stalk, is produced. In 2019, it was calculated that more than 21 million tons of cotton stalk was generated in the agro-industry which means almost 10.5 billion kWh energy. On the other hand, during tea harvesting, the residue is quite low since the small parts can also have a market value as a low-quality product. Rize was the main producer of

tea. Sunflower was planted both for oil and snack production and the greatest amount was harvested in Tekirdağ. In total, nearly 2.5 billion kWh of energy can be produced from sunflower heads. The highest amount of rye plantation was in Niğde. Triticale is a hybrid of rye and wheat and is mostly used as animal feed. The highest amount of triticale was planted in Çorum in 2019 and 0.33 billion kWh energy can be produced from the straws.

The information given in Table 2 highlighted that the values for the energy potential per unit harvest area of wheat straw and rye straw were greater in the respective city with the highest production (4238 kWh/ha and 3115 kWh/ha, respectively) rather than the values calculated for the whole country (3884 kWh/ha and 3060 kWh/ha). For the rest of the substrates, the yields were less than the overall values.

The VS concentrations in the reactors were in the range of 1.1-1.4% on the first day of the operation period. After 30 days of operation, VS varied between 0.99% and 0.86%. The VS reduction in the reactors were showing the same pattern with biomethane production. Whereas the highest VS reduction was observed in the reactors operated with wheat straw (28.7%), the lowest values were determined in the reactors fed by cotton stalk (4.8%).

Table 2. Total energy potential of the selected agro-residues in Turkey

Type of Crop	Wheat	Triticale	Rye	Sunflower	Cotton	Tea
Production in 2019 (ton)	19,000,000	274,136	310,000	2,100,000	3,014,000	1,407,448
Residue type	Straw	Straw	Straw	Head	Stalk	Residue
Residue to Crop Ratio [13,24]	1.1	1.1	1.1	1,17	7.18	0.22
Available Residue (ton)	20,900,000	301,550	341,000	2,457,000	21,640,520	309,639
Methane Production (m ³)	2,652,962,400	32,924,737	34,280,048	250,908,840	1,044,804,306	12,869,526
Total energy potential (kWh)	26,531,746,370	329,273,707	342,827,904	2,509,289,127	10,448,878,899	128,705,553
Harvest area (ha)	6831853.5	64092.5	3428004.8	751693.1	955613.8	78569.3
Energy potential per unit harvest area (kWh/ha)	3884	5137	3060	3338	10934	1638
Harvesting period	May - July	May - July	May - July	August-September	September-December	April-October
City with the highest production	Konya	Çorum	Niğde	Tekirdağ	Şanlıurfa	Rize
Energy potential of the city with the highest production per unit harvest area (kWh/ha)	4238	4078	3115	2996	9250	1569

4. Discussion and Conclusion

There is an increasing trend in energy consumption in Turkey and energy demand is escalated rapidly. To meet this demand, renewable energy targets are stated in the countries energy policies. Biomass energy is also addressed as one of the green energy sources and it is projected to have more biogas plants shortly. In this study, the biomethane potentials of the most abundant agro-residues were evaluated.

Anaerobic digestion is a sequential process in which diverse microbial communities including hydrolytic, acidogenic, and acetogenic bacteria and methanogenic archaea interact mutually [9]. The lignocellulosic structures of agro-residues limit the hydrolysis rates which then affect the

methane yields [9,25,26]. Thus, appropriate inoculum should be used during the start-up period which includes necessary microbial communities having an ability to degrade lignocellulosic feedstock. The bacterial community structure of the inoculum seems to be consistent with other research that found a similar bacterial profile in the anaerobic systems [22,27] and provides an ability to degrade a wide variety of carbohydrates under anaerobic conditions. The bacterial community of inoculum was dominated by *Cloacimonetes*, *Bacteroidetes*, *Firmicutes*, and *Proteobacteria* species. The members of *Cloacimonetes* are speculated to having a role in hydrolysis and/or fermentation of cellulosic feedstock [9]. On the other hand, the phylum *Firmicutes* include key members of acetogenic and syntrophic bacteria

that can use volatile fatty acids and produce acetic acid for further processes, as well as some members can perform hydrolysis. Some species of *Bacteroidetes* can ferment sugars to volatile fatty acids such as acetate, butyrate, and propionate [28]. Whereas some *Proteobacteria* species can hydrolyze various organic feedstock, it also includes well-known butyrate and propionate consumers [27]. Since *Methanosaetaceae* can directly convert acetate into methane and carbon dioxide [29], the results indicated that acetoclastic methanogenesis was the major path for methane generation consistent with the literature [30]. The inoculum included also hydrogenotrophic methanogens namely, *Methanobacteriaceae* and *Methanospirillaceae*. Overall, the results revealed that inoculum had a diverse microbial community that can degrade lignocellulosic biomass to methane under anaerobic conditions. Thus there would not be any constraints due to the inoculum in the reactor experiments.

Lignin, cellulose, and hemicellulose content of the substrate is quite important on biodegradation which acts similarly to a complex shield protecting the structure of the feedstock from enzymatic attacks and hinders the hydrolysis [25]. Thus the degradation rate is highly dependent on the composition of the substrates. The data obtained in this study were compared with the literature and given in Table 3. The studies in the literature about tea waste mostly focused on the spent tea waste, the waste produced after the beverage production [24,31]. Thus the yields are not comparable with this study. On the other hand, in another study, carried out in Zimbabwe, the biomethane production of tea residues after harvesting was evaluated [32]. According to the results, higher yields were observed compared to this study. This inconsistency may be due to the difference in VS/TS values of the products from different regions. In another research, the impacts of chemical pretreatment methods on biomethane production of cotton stalk were examined and the cumulative methane production of the non-pretreated substrate was around 60 mL CH₄/gVS_{influent} which was slightly less than the values detected in this study [25]. This observed difference in this study could be attributed to the experimental set-up and/or inoculum activity. In another research, the authors evaluated bioethanol and biogas production from lignocellulosic biomass including rye straw.

Interestingly, their methane yields were almost three times higher than that of this study (360 mL/gVS_{influent}) [37]. This discrepancy could be attributed to the operating temperature which was 42°C. For triticale straw, in addition to the differences in TS and VS values, the differences may be attributed to the trace element addition in the experimental setup in that study [33]. On the other hand, the biomethane potential range is quite wide for wheat straw (150-250 mL/gVS_{influent}) [34,35] and our results are in line with the previous studies carried out in Turkey and Germany [23,28]. In another study, the biomethane yield for sunflower head was determined as 211 mL CH₄/gVS_{influent} which was higher than the result obtained from this study [26]. However, this result needs to be interpreted with caution because the authors dried the sunflower heads at 40 °C and milled them finely before digestion. It seems possible that these pre-treatments had a positive effect on the methane yield from this substrate.

Currently, most of the plants in Turkey are operated for sewage sludge, leachate, and industrial wastewater management. Although the number of biogas plants with agricultural feedstock is quite low [36], more than 51 million tons/year of cereal straw is produced in Turkey holding a biogas potential of nearly 276.74 PJ/year.

Table 3. Comparison of the methane yields of the selected feedstock

Feedstock	In this study NmL/gVS _{influent}	Methane yield from literature mL/gVS _{influent}	Reference
Tea residues	89	120-130	[32]
Cotton stalk	71	50-60	[25]
Rye straw	122	360	[37]
Triticale straw	145	245	[33]
Wheat straw	164	150-250	[23,34,35]
Sunflower head	138	211	[26]

Studies showed that Turkey holds a great biomass potential for bioenergy generation through anaerobic digestion. Since anaerobic digestion is a sustainable alternative in energy generation, it also contributes to diversifying energy sources and increasing energy security in the country. Agro-residues can contribute a significant share in the biogas sector and therefore, have a significant role in future green energy production. For the more flexible anaerobic digesters, the amount and availability of the feedstock should be determined in each province and the systems should optimize for those feedstocks. In order to increase the biomethane potential yields and to make the anaerobic digesters more feasible, operational conditions should be optimized and physico-chemical and biological pre-treatment techniques and/or bioaugmentation applications should be integrated into the systems. Ensiling and/or co-digestion strategies can also be taken into account due to different harvesting periods of the feedstocks.

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