

Article

Co-Digestion of Sewage Sludge and Cattle Slaughterhouse Waste for Wastewater Treatment Plant Energy Self-Sufficiency—A Case Study

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Abstract

The need to increase the energy self-sufficiency of wastewater treatment plants (WWTPs), and ultimately to achieve energy neutrality of these facilities, encourages the search for solutions that intensify biogas production. Co-digestion of sewage sludge (SS) with high-energy potential organic waste is a solution that enables an increase in biogas yield in WWTPs. Such an approach allows not only for more efficient use of existing infrastructure but also for increased energy recovery from waste. This article evaluates the use of cattle slaughterhouse waste (CSW) as a co-substrate in the co-digestion process with sewage sludge. The analysis covered biogas production efficiency, its quality, and the significance of this approach for the operation of WWTPs. The study was conducted in laboratory reactors operating under mesophilic conditions (36–37 °C), with different percentage shares of CSW (2, 8, 12, and 25% of TS). The obtained biogas yield increased from $0.487 \pm 0.024 \text{ m}^3/\text{kg VS}$ for SS to $0.527 \pm 0.026 \text{ m}^3/\text{kg VS}$ at 25% CSW, while for CSW alone this parameter reached $0.625 \pm 0.031 \text{ m}^3/\text{kg VS}$. Methane yield increased from $0.322 \pm 0.016 \text{ Nm}^3 \text{ CH}_4/\text{kg VS}$ (SS) to $0.344 \pm 0.017 \text{ Nm}^3 \text{ CH}_4/\text{kg VS}$ (25% CSW), while for CSW it reached $0.452 \pm 0.023 \text{ Nm}^3 \text{ CH}_4/\text{kg VS}$. In the simulation of the co-digestion process with this waste, an increase in energy production from 3.1% to 51.2% was achieved depending on the percentage share of CSW. The conducted research clearly demonstrated that the addition of CSW improves the energy balance of WWTP, which represents a significant technological benefit.

Keywords: sewage sludge; co-digestion; cattle slaughterhouse waste as co-substrate; biogas yield; biogas composition; energy efficiency



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1. Introduction

In recent years, anaerobic digestion (AD) has been recognized as an effective method for stabilizing sewage sludge (SS) that enables the production of renewable energy. This process is widely used in large wastewater treatment plants (WWTPs) with a flow rate

exceeding 20,000 m³/day. In addition to producing biogas, a versatile energy carrier, the implementation of this technology enables the reduction in greenhouse gas emissions and the achievement of energy self-sufficiency for WWTPs [1,2]. Furthermore, once specific requirements are met, it also enables utilizing digestate in agriculture [3]. Despite its many benefits, the mono-digestion of SS faces numerous technological challenges related to the poor degradation combined with a long retention time. Furthermore, SS in many cases contains toxic substances such as heavy metals, ammonia, and volatile fatty acids, which can inhibit methanogenic bacteria. It is also characterized by an unfavorable C/N ratio [4]. These limitations mean that the current research on the use of SS in AD is focused on process optimization, with the greatest emphasis placed on increasing biogas production and, consequently, improving the profitability of WWTPs with high energy demands [5]. The measures taken in this regard are aimed at employing the processes that improve the properties of SS, i.e., conditioning and disintegration, particularly through the use of thermal methods (e.g., thermal hydrolysis) and physical methods (e.g., acoustic and hydrodynamic cavitation) in both cases. However, the implementation of these strategies entails additional financial costs for WWTPs, increased energy demand, and staff training [5,6].

Another interesting option aimed at intensifying the SS anaerobic bioconversion process is co-digestion with other organic waste, i.e., the so-called co-digestion process [7]. It is worth emphasizing that waste-to-energy processing has been gaining importance in recent years, as this action allows for solving economic and environmental problems [8,9]. Research conducted in recent years indicates that anaerobic co-digestion is one of the main directions for increasing the energy self-sufficiency of wastewater treatment plants while simultaneously improving the management of various organic wastes [10]. As numerous scientific studies show, the implementation of this strategy in WWTPs offers a number of advantages: increased biogas production, improved sludge digestion, and enhanced quality of the reactor feedstock through increased levels of macro- and micronutrients, as well as the biodegradable fraction [11–13]. Equally important is the ability to dilute potentially toxic compounds (e.g., ammonia, pharmaceuticals, heavy metals, or higher fatty acids) [10]. In addition to maximizing methane yield, research is increasingly focused on optimizing substrate selection, mixing ratios, process stability, and full-scale implementation of strategies in line with the principles of the circular economy [14]. Recent reviews emphasize the use of co-substrates (e.g., food waste, manure, degreaser waste, and slaughterhouse by-products) due to their high biodegradability and methane potential [10]. Among the many possibilities, fat-rich waste is an interesting option; due to its high methane production potential, it is an attractive substrate for the AD of SS. Its addition to sludge can significantly increase methane production by as much as 50–300% [11,15,16]. The efficiency of anaerobic co-digestion strongly depends on the substrate properties, and synergistic effects should also be assessed for each substrate–co-substrate mixture [17]. Sources of fat waste include dairies, slaughterhouses, food processing plants, and industrial facilities such as biodiesel production plants. However, their use in municipal WWTPs can pose numerous technological challenges, including inhibition by long-chain fatty acids (LCFAs), the potential for overloading the reactor with organic compounds, and foaming within the reactor [18]. For this reason, their addition should be carefully controlled, and their implementation in operational facilities should be preceded by studies to determine the most favorable proportions [19,20].

This study presented a comprehensive analysis of the potential for using cattle slaughterhouse waste (CSW) as an additive to SS to intensify methane production at the municipal wastewater treatment plant (WWTP) in Dębica (Poland, Subcarpathian Voivodeship). For this purpose, the following were conducted: an analysis of the AD process at the selected WWTP and the energy self-sufficiency of the facility, biogas yield studies for cattle slaughter-

house waste and mixtures of SS with this waste, an assessment of the co-digestion process using this waste group, along with a SWOT analysis. Thus far, such analyses have not been the subject of scientific research. The novelty of this study is the systematic evaluation of different CSW addition ratios (2–25% TS) under mesophilic anaerobic digestion conditions and their combined effects on biogas yield, methane production, gas quality, and the overall energy balance of the WWTP. This research demonstrates the potential of integrating an industrial organic waste stream into existing WWTP infrastructure to simultaneously enhance renewable energy recovery and promote waste valorization. The results provide quantitative evidence that CSW addition can increase methane yield and simulated energy production, supporting a pathway toward more energy-neutral and circular wastewater treatment systems.

2. Materials and Methods

2.1. Characteristics of the Study Site

The study was conducted at the WWTP in Dębica, which has a design flow rate of $Q_{av\ d} = 21,000\ m^3/d$. The plant receives municipal wastewater, industrial wastewater (mainly from meat processing plants, car washes, and small food service establishments), and stormwater, as well as the wastewater delivered by vacuum trucks. The process flow of the treatment plant (Figure 1) consists of the following stages: a mechanical coarse screen, two fine belt-and-hook screens, a two-chamber longitudinal grit chamber, two radial primary settling tanks, two biological reactors with three-phase activated sludge (a modified A2/O system with pre-denitrification and selectors), and two secondary settling tanks. The mechanical treatment stage ensures the removal of screenings, mineral suspended solids, easily settleable organic suspended solids, and fats from the wastewater. The biological treatment stage enables the integrated removal of carbon, nitrogen, and phosphorus compounds. Sludge processing activities include thickening processes (carried out in gravity thickeners for primary sludge and in a belt thickener for excess sludge), anaerobic stabilization (carried out in a closed AD chamber), dewatering (carried out in decanter centrifuges), and hygienization (carried out using quicklime).

The sludge stabilization process is carried out in a single-stage mesophilic AD system. For this purpose, two closed AD chambers with a total capacity of $4400\ m^3$ have been provided, into which, in addition to thickened sludge, fats and other solids floated in the primary clarifiers are directed. Process operating parameters: $36\ ^\circ C$ —temperature; 300 mm H_2O —biogas pressure in the chamber; 94–95%—average moisture content of sludge fed into the digester; 97.2–97.4%—moisture content of digested sludge. According to design specifications, the amount of total solids content in the digester feedstock is 9392 kg TS/day, and the proportion of individual sludge types is as follows: 5273 kg TS/day—primary sludge; 3119 kg TS/day—excess biological sludge; 1000 kg TS/day—chemical sludge. The total solids load in the digested sludge is 6377 kg TS/day. The biogas produced during the stabilization process contains an average of 60% methane, which is captured and used to generate heat and electricity for the needs of the WWTP and the administrative building of Wodociągi Dębickie Ltd. (Dębica, Poland).

2.2. Methodology for Assessing the Present State

The first phase of the work involved an assessment of the efficiency of the SS digestion process and the energy self-sufficiency of the WWTP under analysis. For this purpose, a detailed analysis of the data from a one-year period (2023), which was provided by Wodociągi Dębickie Ltd., was carried out. The scope of the data provided included, among other things, average daily flow rates, the daily volume of sludge produced—i.e., primary thickened sludge (PS) and excess thickened sludge (ES)—total solids (TS) and volatile

solids (VS) content in the aforementioned sludge, daily biogas production, daily energy consumption, and energy production from biogas.

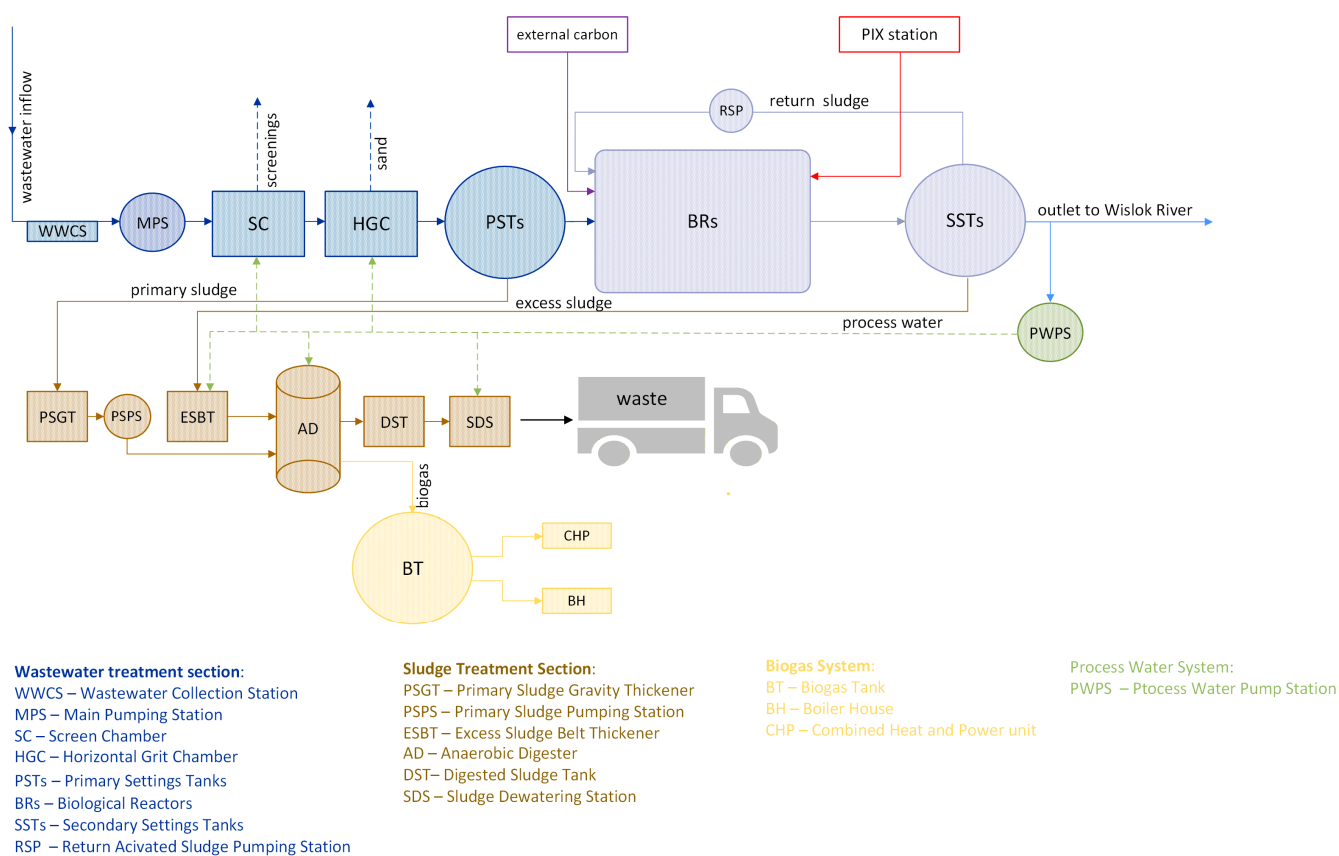


Figure 1. A process flow diagram of the WWTP in Debica (authors' own illustration).

On the basis of the data above, the following analyses were made: the quantitative and qualitative balance of individual sludge streams directed to the digester; biogas yield in the AD process in relation to volume and the unit total solids and volatile solids of the sludge (the parameters were calculated as the ratio of the daily biogas volume and, respectively, the daily sludge volume, the daily TS content, and the daily VS content); actual technological parameters of the digester; digestion efficiency (a parameter calculated as the ratio of the difference between VS before and after the digestion process to the VS before the digestion process), energy consumption (a parameter calculated as the ratio of the treatment plant's daily energy consumption to the daily volume of treated wastewater), and the energy self-sufficiency (a parameter calculated as the ratio of the energy produced from biogas to the total energy consumption) of the analyzed WWTP.

2.3. Methodology for Assessing the Suitability of Cattle Slaughterhouse Waste for the Co-Digestion Process

Cattle slaughterhouse waste (CSW) was used in the co-digestion process with sewage sludge (SS) (a mixture of PS and ES mixed in a 3:1 ratio) (Figure 2a). The factors taken into account included the availability of the raw material on the local market and its significant methane potential. The co-substrate originated from a cattle slaughterhouse and consisted of a mixture of the fraction retained in the flotation tank and, for example, the stomach contents of the slaughtered cattle; consequently, it contained residues of feed, straw, hay, etc. (Figure 2b). The heavy metal content in the waste was as follows: <0.4 mg/kg TS for cadmium; <4.0 mg/kg TS for lead; 10.2 ± 1.6 mg/kg TS for nickel; 106.0 ± 23.0 mg/kg TS for zinc; 12.7 ± 1.7 mg/kg TS for copper; and 0.0258 mg/kg TS for mercury. Furthermore,

calcium and magnesium account for 1% and 0.07% of the TS of this waste, respectively. The average amount of waste generated is approximately 197 Mg/month.



Figure 2. A mixture of primary and excess sludge in a 3:1 ratio (SS) (a) and cattle slaughterhouse waste (CSW) (b) (authors' own photographs).

To assess the suitability of CSW for supporting the AD process at the WWTP in Dębica, physicochemical analyses and methane potential tests were conducted on SS from the mentioned WWTP and CSW. Additionally, to select the most favorable co-substrate dosage, four mixtures were prepared, differing in the percentage of fatty waste. The following co-substrate percentages were used in the tested mixture: 2% (2% CSW), 8% (8% CSW), 12% (12% CSW), and 25% (25% CSW) of TS (relative to the total solids of the sludge). The reference sample was the aforementioned mixture of primary and excess sludge. The adopted doses were selected based on the technical limitations of the existing digesters at the WWTP in Dębica as well as the availability of this co-substrate (the amount of substrate that the producer might supply).

The physicochemical analyses of substrates included the determination of pH, TS, VS, Kjeldahl total nitrogen, sulfur (S), iron (Fe), chlorides (Cl), ether-extractable substances (EXS), and total organic carbon (TOC). Moreover within the AD experiment the stability parameters such as alkalinity, volatile fatty acids (VFAs), and ammonia nitrogen (AN) were controlled. Most of the analyses were performed using the spectrophotometer Hach Lange UV–VIS DR 3900 (Hach, Loveland, CO, USA) according to Hach analytical methods (www.hach.com). Total and volatile solid analyses were performed in accordance with the Standard Methods for the Examination of Water and Wastewater [21]. In turn, the pH values were monitored using the HQ 40D Hach-Lange multimeter (Hach, Loveland, CO, USA). Ether-extractable substances (EXS) were determined by an accredited external laboratory (Polbiotech Laboratory, Poznań, Poland) using its validated analytical procedure based on solvent extraction with petroleum ether. Total organic carbon was analyzed using a TOC analyzer (Shimadzu, TOC-V_{CPN}, Kyoto, Japan)—samples were combusted in an oxygen atmosphere at temperatures > 600 °C and the evolved CO₂ was quantitatively measured by infrared detection.

The characteristics of the mixtures used are presented in Table 1.

Table 1. Physicochemical properties of SS, CSW and their mixtures (average values and standard deviation are shown).

Parameter	Unit	Sample					
		SS	CSW	2% CSW	8% CSW	12% CSW	25% CSW
pH		6.04 ± 0.30 a	6.02 ± 0.30 a	5.85 ± 0.29 a	5.88 ± 0.29 a	5.97 ± 0.30 a	6.01 ± 0.30 a
TS	g/kg	38.0 ± 1.90 a	147.0 ± 7.35 b	40.0 ± 2.00 a	41.0 ± 2.05 a	39.0 ± 1.95 a	41.0 ± 2.05 a
VS	g/kg	30.2 ± 1.51 a	131.4 ± 6.57 b	32.0 ± 1.60 a	32.7 ± 1.64 a	31.2 ± 1.56 a	32.8 ± 1.64 a
VS	%TS	79.4 ± 3.97 a	89.4 ± 4.47 b	80.1 ± 4.01 a	79.9 ± 4.00 a	79.9 ± 4.00 a	80.0 ± 4.00 a
N _{Kj}	g/kg	2.30 ± 0.12 a	6.70 ± 0.34 b	2.50 ± 0.13 a	2.60 ± 0.13 a	2.40 ± 0.12 a	3.00 ± 0.15 c
S	g/kg	0.27 ± 0.01 a	0.82 ± 0.04 b	0.36 ± 0.02 b	0.38 ± 0.02 b	0.35 ± 0.02 b	0.34 ± 0.01 a
Fe	g/kg	0.26 ± 0.01 a	5.70 ± 0.29 b	0.28 ± 0.01 a	0.32 ± 0.02 c	0.27 ± 0.01 a	0.30 ± 0.02 c
Cl	g/kg	nd	1.90 ± 0.10	nd	nd	nd	nd
EXS	g/kg	0.89 ± 0.04 a	60 ± 3.00 b	2.10 ± 0.15 c	5.75 ± 0.75 d	8.12 ± 1.1 e	15.75 ± 1.3 f
TOC	g/kg	12.54 ± 0.63 a	608 ± 30.40 b	23.81 ± 2.1 c	62.1 ± 3.1 d	85.6 ± 3.7 e	170.1 ± 9.8 f
C/N	-	6 a	37 b	6.8 a	8.9 a	10.5 b	17 c

nd—not determined; Note: values followed by the same letter are not statistically different (ANOVA, $p < 0.05$, Tukey test).

In turn, biogas yield tests were conducted in accordance with the VDI 4630 guidelines, which outline the general procedure for various types of tests assessing biogas productivity from different materials. The biogas potential assessment was carried out in 0.5 dm³ glass digesters. An inoculum was placed in each tank, consisting of a properly prepared suspension rich in microorganisms involved in the methanogenic anaerobic digestion process. This sample was taken from an agricultural biogas plant, diluted 1:1 with deoxygenated tap water, and incubated at 38 °C for 4 weeks—during incubation, organic compounds were digested. The average TS value for the inoculate was approximately 2%, while VS was approximately 60% of TS. The proportions between the inoculate and each tested substrate were selected so that the initial digester load was equal to (or very close to) 5 kg VS/m³. The ratio of VS from the substrate to VS from the inoculate was less than 0.50, as specified by the VDI 4630. In this experiment the substrate to inoculum ratio varied between 0.171 and 0.182 depending on the feedstock composition. The laboratory digester was sealed tightly, and its gas space was flushed with nitrogen to remove oxygen. The contents of the fermenter were stirred periodically (at least once a day). The process was conducted at a temperature of 36–37 °C. The biogas produced was collected in so-called eudiometers, and its volume was measured every day of the experiment. The composition of the biogas was determined using a Geotech BIOGAS 5000 analyzer (Coventry, UK). The study was conducted until daily increases in biogas were observed, i.e., for a period of approximately 5 weeks. During laboratory tests, the digestion of the inoculate itself was carried out in parallel, and the obtained volume of biogas (biogas yield from the inoculate was 20–50 cm³/0.5 dm³ of inoculate), after appropriate calculation, was subtracted from the biogas produced from the substrate and inoculate mixture. The results obtained from observing the mono- and co-digestion processes include:

- The amount of methane and biogas obtained per unit mass of substrate, also calculated per 1 kg of total solids and 1 kg of volatile solids;
- The time required to produce 90% of the biogas;
- The content of CH₄, CO₂, O₂, and H₂S in the biogas;
- The VS removal (%);
- The calculated amount of digestate remaining for disposal after the digestion of 1 ton of substrate;
- The calculated mass of substrate required to produce 1 MWh of electricity.

The presented results represent the average of three repetitions.

In order to assess the effect of CSW addition on the co-digestion process, the synergy index (SI) was calculated for each tested mixture. This index was calculated according to the method proposed by Nielfa et al. [21], i.e., as the ratio of the experimental value of methane yield from the co-digestion mixture (BMP_{exp}) to the calculated theoretical value of methane yield for individual mixtures (BMP_{th}), taking into account the VS of each substrate included in the final mixture. The results were interpreted as follows: SI values > 1 indicate that the mixture has a synergistic effect on the final production, SI = 1 indicates that the substrates act independently of the mixture, SI < 1 that the mixture has a competitive effect on the final production.

The energy balance of the wastewater treatment plant was developed based on operational data including monthly electricity consumption, the daily volume of treated wastewater, biogas production, and the amount of electricity generated in the CHP system. Electricity consumption was presented on a monthly basis, and a specific energy consumption index was determined to assess energy efficiency, relating monthly energy consumption to the volume of treated wastewater (kWh/m³). Similarly, specific electricity production from biogas was determined relative to unit of biogas (kWh/Nm³), SS volume (kWh/m³ of SS), total solids (kWh/kg TSS), and volatile solids of the sludge (kWh/kg VS). Based on the results of laboratory tests of the biogas yield of CSW and its mixtures with SS, the specific biogas yield was determined for the analyzed co-substrate substrates (2, 8, 12, and 25% CSW). The obtained values were used to develop a computational model that estimated the potential biogas production under the operating conditions of the wastewater treatment plant for each co-substrate dosage, taking into account the actual amount of SS directed to the anaerobic digester. Then, based on the indicators of electricity production from biogas, the potential electricity production was calculated and compared with the actual energy consumption of the facility. The obtained results formed the basis for a simulation assessment of the impact of using CSW on the energy self-sufficiency of the Dębica WWTP.

2.4. Statistical Analysis

The statistical analysis was performed using analysis of variance (ANOVA) (Shapiro–Wilk’s, Levene’s and Tukey’s tests were applied) and the StatsoftStatistica software (v13). The differences were assumed to be statistically significant at $p < 0.05$.

3. Results

3.1. Assessment of the Anaerobic Digestion Process Efficiency at the Selected Wastewater Treatment Plant and the Energy Self-Sufficiency of the Facility (Current Status)

The amount of SS generated in the analyzed year, including PS and ES, was $65.0 \pm 10.6 \text{ m}^3/\text{d}$ and $22.7 \pm 4.2 \text{ m}^3/\text{d}$, respectively (Figure 3a). The total volume of generated sludge ranged from 57.0 to $127.0 \text{ m}^3/\text{d}$ (average value: $87.7 \text{ m}^3/\text{d}$). During this period, the average daily flow to the treatment plant ranged from 8249.0 to $21,426.0 \text{ m}^3/\text{d}$. The TS content in the PS and ES was 3.5% and 6.0%, respectively. In turn, the VS content reached 85% and 75% in PS and ES, respectively. On the basis of the above, the mass of sludge directed to the digester was determined (Table 2, Figure 3b,c). During the analyzed period, the ratio of PS to ES directed to the AD process ranged from 1.2:1 to 7.3:1, with an average of 3.0:1. Changes in the percentage share of individual sludge types directed to the digester during the analyzed period are presented in Figure 3d.

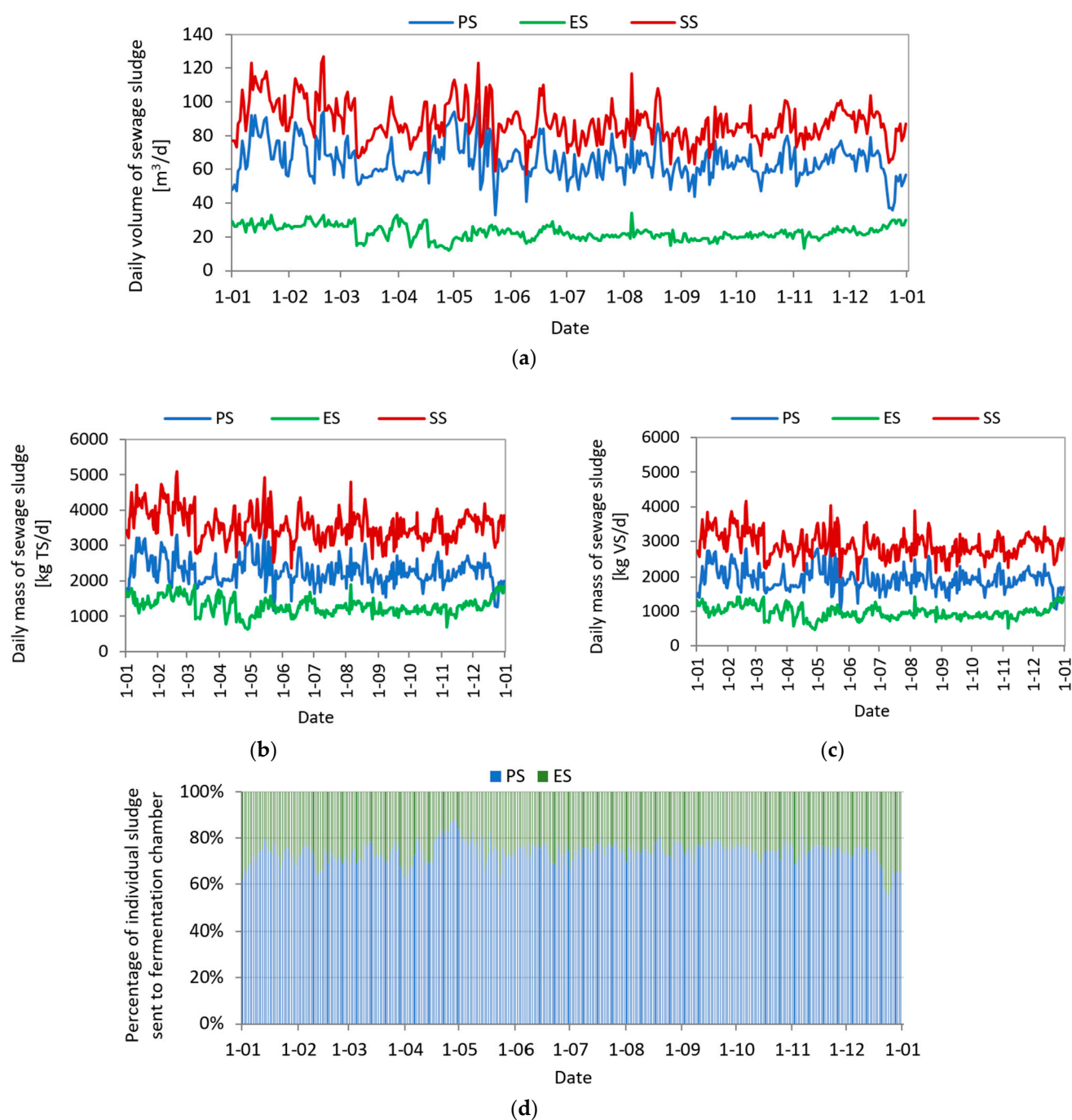


Figure 3. Changes in the daily values of volume (a), TS (b), and VS (c) of the PS and ES directed to the digester, and the percentage of each type of sludge directed to the digester (d).

Table 2. The mass of SS directed to the digester.

Value	Sludge Mass [kg TS/d]			Organic Sludge Mass [kg VS/d]		
	PS	ES	SS (PS + ES)	PS	ES	SS (PS + ES)
Minimum	1155.0	624.0	2363.0	981.8	468.0	1915.8
Maximum	3465.0	1904.0	5105.0	2945.3	1428.0	4157.8
Mean	2275.9	1293.6	3569.5	1934.5	970.2	2904.7
Std. deviation	371.8	244.0	437.4	316.0	183.0	359.5
15th percentile	1960.0	1060.0	3141.8	1666.0	795.0	2564.2
85th percentile	2639.0	1586.0	4019.8	2243.2	1189.5	3277.3

The data analysis made it possible to determine the actual operating parameters of the digester as recorded during operation in the study period (Table 3). The daily dose of fresh sludge (a mixture of PS and ES) introduced into the digester ranged up to approximately 5.8% of the chamber's active volume. The hydraulic retention time (HRT) during the analyzed period ranged from 17.2 to 38.3 days. This is a typical duration for classical anaerobic digestion of SS [22]. The digester operated at a very low organic loading rate, i.e., in the range of 0.877 to 1.903 kg TS/(m³·d), which is lower than the recommended value for this type of installation. In practice, it is recommended to operate digesters for mesophilic anaerobic digestion of a mixture of primary and excess sludge at a loading rate of up to 2.25 kg TS/(m³·d). Conversely, when the HRT is reduced to 20 days, it is recommended to maintain the digester loading at 4.8 kg BOD/(m³·d) [22]. It is worth noting that the designed loading rate of the digester at the Dębica WWTP is 2.13 kg TS/(m³·d). The VS content in the digested sludge during the analyzed period averaged 2.2%. The low loading rate of the anaerobic digestion chamber resulted in low digestion efficiency—a maximum of 55.1% (average 45.2%). Changes in this parameter during the analyzed period are presented in Figure 4.

Table 3. The process/actual parameters of the digester.

Value	Daily Sludge Dose (PS + ES Mixture) [%/d]	HRT [d]	Loading of the Digester A [kg VS/(m ³ ·d)]	Digestion Efficiency [%]
Minimum	2.6	17.2	0.877	18.3
Maximum	5.8	38.3	1.903	55.1
Mean	4.0	25.4	1.329	45.2
Std. deviation	0.5	3.3	0.165	5.6
15th percentile	3.5	21.8	1.174	42.5
85th percentile	4.6	28.4	1.500	48.9

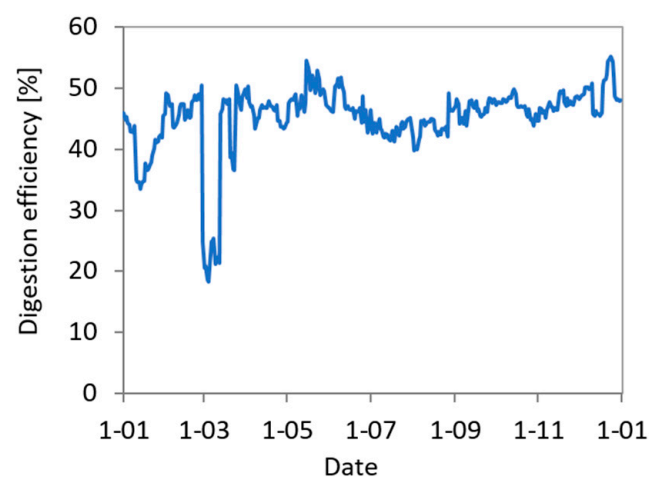


Figure 4. Digestion efficiency during the analyzed period.

Biogas production in the year under review ranged from 104.0 to 2742.0 Nm³/d (Figure 5), with an average of 1244.9 ± 350.9 Nm³/d. The available data show that the majority of the biogas produced was directed to the cogeneration unit (ranging from 65.2% to 87.7%, with an average of 79%). The WWTP also used the produced biogas to power the boiler room (ranging from 6.2% to 34.8%, with an average of 19.8%). A small percentage, most often excess biogas, was directed for flaring (max. 7.2%, average 1.2%), particularly during the summer. This distribution confirms the effective utilization of the biogas produced at the Dębica WWTP.

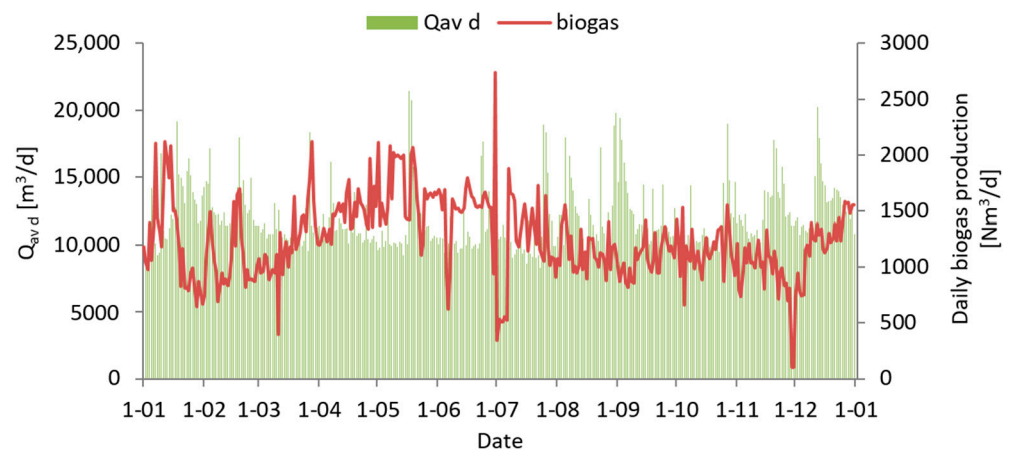


Figure 5. Variations in biogas production at the analyzed wastewater treatment plant.

Data analysis showed that from 1 m³ of SS fed into the digester, it is possible to obtain between 1.1 and 31.9 Nm³ of biogas per m³ of sludge, with an average of 14.3 ± 4.0 Nm³ of biogas per m³ of sludge. At another WWTP in Poland—the Zamość WWTP—a comparable biogas yield of 14.8 ± 1.2 Nm³/m³ of sludge was recorded in a conventional mono-digestion process [23]. A more reliable indicator is the ratio of biogas production to each unit of total solids content (m³ of biogas/kg TS) and volatile solids content (m³ of biogas/kg VS). Table 4 summarizes the basic descriptive statistics for the aforementioned indicators. It can be noted that the average biogas yield in the analyzed period was 0.352 Nm³/kg TS and 0.432 Nm³/kg VS.

Table 4. A summary of the indicators for biogas yield at the Dębica WWTP.

Value	Biogas Production [m ³ /d]	Biogas Yield		
		[Nm ³ /m ³ Sludge]	[Nm ³ /kg TS]	[Nm ³ /kg VS]
Minimum	104.0	1.1	0.027	0.034
Maximum	2742.0	31.9	0.747	0.921
Mean	1244.9	14.3	0.352	0.432
Std. deviation	350.9	4.0	0.099	0.122
15th percentile	912.8	10.2	0.251	0.309
85th percentile	1589.8	18.2	0.451	0.554

Energy consumption at the considered WWTP ranged from 152,707 to 172,767 kWh/month during the year under review (average: 163,054.58 kWh/month). It is worth noting that the largest consumers of electricity during this period were the facilities related to biological wastewater treatment processes. Annual electricity consumption for the operation of biological reactors (blowers, mixing pumps, internal and external recirculation pumps, measuring probes, etc.) amounted to 1211,456.90 kWh/year, with blower energy consumption alone accounting for 424,895 kWh/year. These figures represent nearly 62% and 22% of annual energy consumption, respectively. By comparison, the grit chambers consumed approx. 31,000 kWh/year, which accounts for just over 1.5% of annual energy consumption. The estimated electricity consumption rate per unit volume of treated wastewater ranged from 0.260 to 0.655 kWh/m³ of wastewater (Figure 6a). Electricity production per unit volume of biogas and sludge ranged from 0.960 to 18.611 kWh/Nm³ of biogas (average value: 2.139 kWh/Nm³ of biogas) and from 16.441 to 58 kWh/Nm³ of sludge (average value: 27.932 kWh/Nm³ of sludge) (Figure 6b,c). In contrast, the energy production values relative to the TS and VS content of the sludge ranged from 0.409 to 1.376 kWh/kg TS of

sludge (average value: 0.686 kWh/kg TS of sludge) and from 0.502 to 1.728 kWh/kg VS of sludge (average value: 0.843 kWh/kg VS of sludge) (Figure 6d,e).

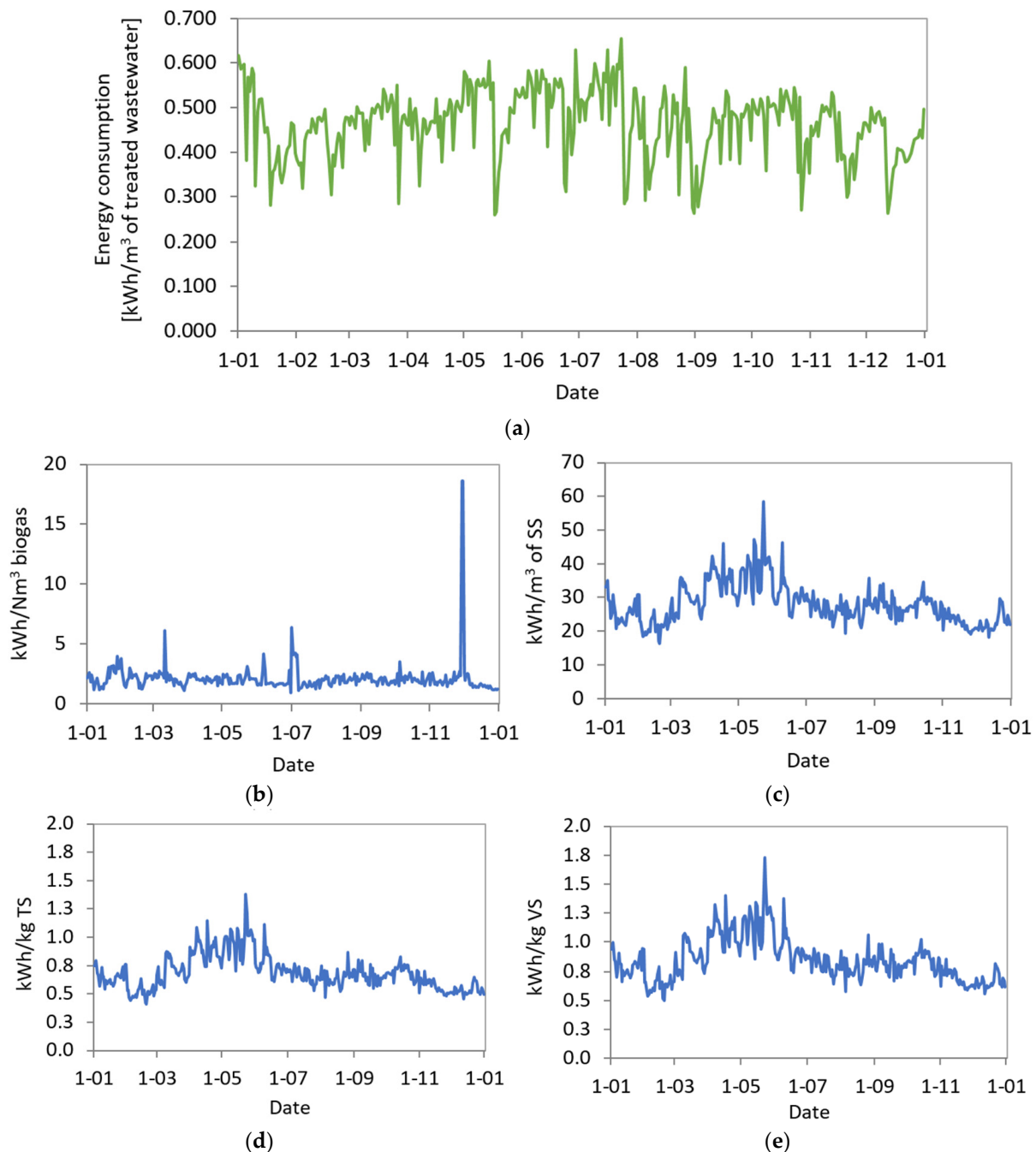


Figure 6. Changes in the energy consumption index relative to the volume of treated wastewater (a), as well as electricity production per unit of biogas (b), SS volume (c), total solids (d), and volatile solids of the sludge (e).

The energy self-sufficiency of the Dębica WWTP in 2023 on a monthly basis, estimated as the ratio of electricity generated on-site to total energy consumption at the facility, ranged from 35.42% to 61.89% (average value: 44.93%), and changes in this parameter are presented in Figure 7. The lowest values of the energy self-sufficiency index occurred mainly in the fall and winter months, indicating that the treatment plant was more dependent on energy drawn from the power grid during these periods, and thus highlighting the need to increase biogas production.

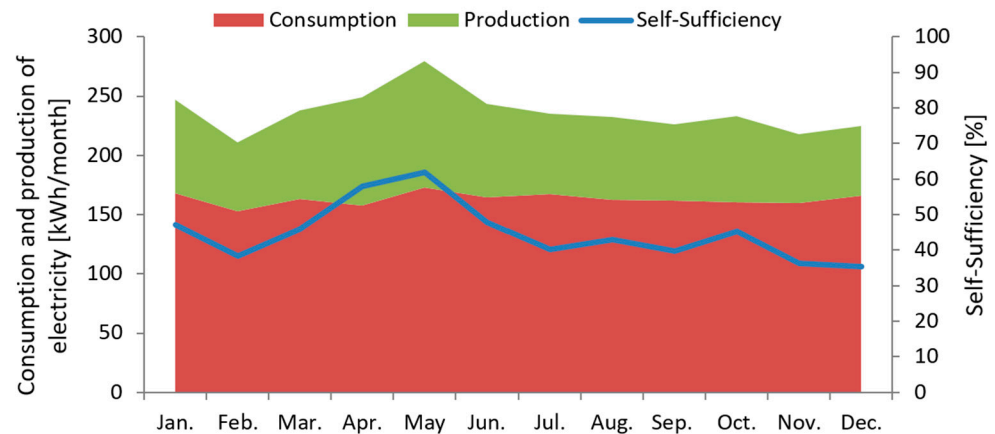


Figure 7. Production and consumption of electricity including self-sufficiency.

3.2. Analysis of the Methane Potential of Sewage Sludge, Cattle Slaughterhouse Sludge, and Their Mixtures

Selected physicochemical parameters of the tested SS, CSW, and their mixtures are summarized in Table 1. The VS value in the tested sludge—a parameter that significantly influences the amount of biogas produced during anaerobic sludge stabilization—is 30.2 ± 1.51 g/kg, which constitutes $79.4 \pm 3.97\%$ of TS. The CSW, in relation to SS, is characterized primarily by higher VS and TOC values. The TOC values in the tested CSW are nearly 50 times higher than in the tested SS. Furthermore, CSW contained significant amounts of ether-extractable substances (60 ± 3.00 g/kg TS). Nevertheless, these amounts were significantly lower than those reported in the literature. For example, Renggaman et al. [24] tested the suitability of cattle slaughterhouse waste with a fat content of $57.4 \pm 0.3\%$.

The addition of CSW to the SS resulted in an improvement in the quality of the reactor feed with respect to TS and VS. The values of these indicators in the individual mixtures are higher than those determined for SS (approx. 6%) and, at the same time, significantly lower compared to the parameters determined in the CSW (by approx. 350% for TS and approx. 410% for VS, respectively). In the case of pH, the values of this parameter in the mixtures with the addition of CSW are slightly lower. However, it should be noted that an analysis of the quality results of the produced mixtures does not indicate significant differences between the pH, TS, and VS of the individual samples.

The results regarding the sulfur content in the tested mixtures show greater variation. The sulfur content in the 2% CSW, 8% CSW, and 12% CSW mixtures was approximately 30–40% higher than in the SS. The observed increases were significantly higher as compared to SS. The obtained values, despite the absence of an upward trend for the 25% CSW mixture, suggest that the use of the CSW substrate in the co-digestion process will increase the sulfur content in the mixture.

Larger differences between the individual mixtures were also observed for iron content. Compared to the iron content in the SS, these values are higher by approximately 5 to 23%, which is also determined by the high iron content in the CSW (5.70 ± 0.29 g/kg TS). It should be noted that iron has a beneficial effect on the anaerobic digestion process.

The use of CSW also contributed to an increase in the Kjeldahl total nitrogen content in the tested mixtures. With regard to the results for SS, these values are higher by approximately 5 to 30%. The significant differences were observed for 25% CSW mixture.

It should be highlighted that in the presence of CSW the improvement of the C/N ratio occurred; a statistical enhancement was found for 12 and 25% CSW. For the highest dose of CSW there was an increase of over 2.8 times as compared to SS. This is a beneficial effect, as it is commonly known that a balanced C/N ratio in a feedstock-supplied digester

is essential for a successful process performance. Previous studies demonstrated that an optimal C/N ratio should be varied between 15 and 45 [25,26].

The course of the methanogenic digestion process for all samples can be described as normal. However, certain differences in the course and efficiency of this process are evident between individual samples, as indicated by the parameters used to assess methane potential (Table 5) and the course of the cumulative curves (Figure 8a–f).

Table 5. The parameters related to the assessment of the methane potential of SS, CSW, and their mixtures (average values and standard deviation are given).

Parameter	Unit	Sample					
		SS	CSW	2% CSW	8% CSW	12% CSW	25% CSW
Biogas yield	Nm ³ /kg	0.015 ± 0.001 a	0.082 ± 0.004 a	0.015 ± 0.001 a	0.016 ± 0.001 a	0.013 ± 0.001 b	0.017 ± 0.001 d
	Nm ³ /kg TS	0.387 ± 0.019 a	0.559 ± 0.028 b	0.373 ± 0.019 a	0.395 ± 0.020 a	0.321 ± 0.016 c	0.422 ± 0.021 a
	Nm ³ /kg VS	0.487 ± 0.024 a	0.625 ± 0.031 b	0.465 ± 0.023 a	0.495 ± 0.025 a	0.402 ± 0.020 c	0.527 ± 0.026 a
Methane yield	Nm ³ CH ₄ /kg	0.010 ± 0.001 a	0.059 ± 0.003 b	0.010 ± 0.001 a	0.010 ± 0.001 a	0.008 ± 0.001 a	0.011 ± 0.001 a
	Nm ³ CH ₄ /kg TS	0.256 ± 0.013 a	0.404 ± 0.020 b	0.254 ± 0.013 a	0.252 ± 0.013 a	0.212 ± 0.011 c	0.275 ± 0.014 a
	Nm ³ CH ₄ /kg VS	0.322 ± 0.016 a	0.452 ± 0.023 b	0.318 ± 0.016 a	0.316 ± 0.016 a	0.265 ± 0.013 c	0.344 ± 0.017 a
CH ₄ content in biogas	% v/v	66 ± 3.30 a	72 ± 3.60 b	68 ± 3.40 a	64 ± 3.20 a	66 ± 3.30 a	65 ± 3.25 a
CO ₂ content in biogas	% v/v	31 ± 1.55 a	26 ± 1.30 b	29 ± 1.45 a	34 ± 1.70 a	32 ± 1.60 a	33 ± 1.65 a
Other gases *	% v/v	2.5 ± 0.13 a	1.7 ± 0.09 b	3.1 ± 0.16 c	2.4 ± 0.12 a	2.1 ± 0.11 d	1.6 ± 0.08 b
H ₂ S content in biogas (average)	ppm	145 ± 7.25 a	5 ± 0.25 b	210 ± 10.50 c	320 ± 16.00 d	285 ± 14.25 d	312 ± 15.60 d
VS removal	%	55 ± 2.75 a	66 ± 3.30 b	50 ± 2.50 c	57 ± 2.85 a	45 ± 2.25 c	60 ± 3.00 a
Time when 90% of biogas was produced	d	16	13	12	13	14	12
Estimated mass of digestate from 1 tonne of sludge/substrate	t	0.98 ± 0.049	0.91 ± 0.046	0.98 ± 0.049	0.98 ± 0.049	0.99 ± 0.050	0.98 ± 0.049
Estimated mass of sludge or substrate needed to produce 1 MWh of electricity	t	25.29 ± 1.26	4.15 ± 0.21	24.18 ± 1.21	23.80 ± 1.19	29.76 ± 1.49	21.81 ± 1.09

* “other gases” include O₂, water (water vapor 0–3.5%), NH₃, H₂S, H₂, N₂ and other volatile compounds; Note: values followed by the same letter are not statistically different (ANOVA, $p < 0.05$, Tukey test).

In the case of SS alone and mixtures of SS with CSW, due to the low TS content in these samples, the biogas production yield was not high and ranged at a comparable level of 0.013–0.017 Nm³/kg, with the highest values obtained for the 25% CSW mixture. Significantly higher values were obtained for the CSW substrate; in this case, biogas production was more than five times higher than in the other cases considered. Of course, a rational approach is to compare the results in terms of TS or VS (Table 5). With regard to VS, the highest biogas production was obtained in the case of mono-digestion of the CSW substrate. In turn, when analyzing two-component mixtures, the most favorable results were recorded for the 25% CSW mixture. In this case, compared to the control sample, where only SS was fermented, an increase of over 8% was achieved. However, the observed differences were not of statistical significance. It should be noted that for the 2% CSW and 12% CSW mixtures, a decrease in biogas production of 4.5% and 17.5%, respectively, was observed. With regard to the implementation of this strategy on a technical scale, methane production is key. Compared to the reference sample (SS mono-digestion), the most favorable results were again obtained for the AD of CSW, with methane production averaging 0.452 ± 0.023 Nm³ CH₄/kg VS (Table 5). For comparison, the AD of high-fat CSW yielded 0.5822 ± 0.033 Nm³ CH₄/kg VS [24]. In two-component mixtures, the highest methane production was again observed in the presence of the highest co-substrate dose (25% CSW). Compared to the control, an improvement of 7% was achieved; in this case, methane production amounted to 0.344 ± 0.017 Nm³ CH₄/kg VS. Similarly to biogas production, this increase was not of statistical significance.

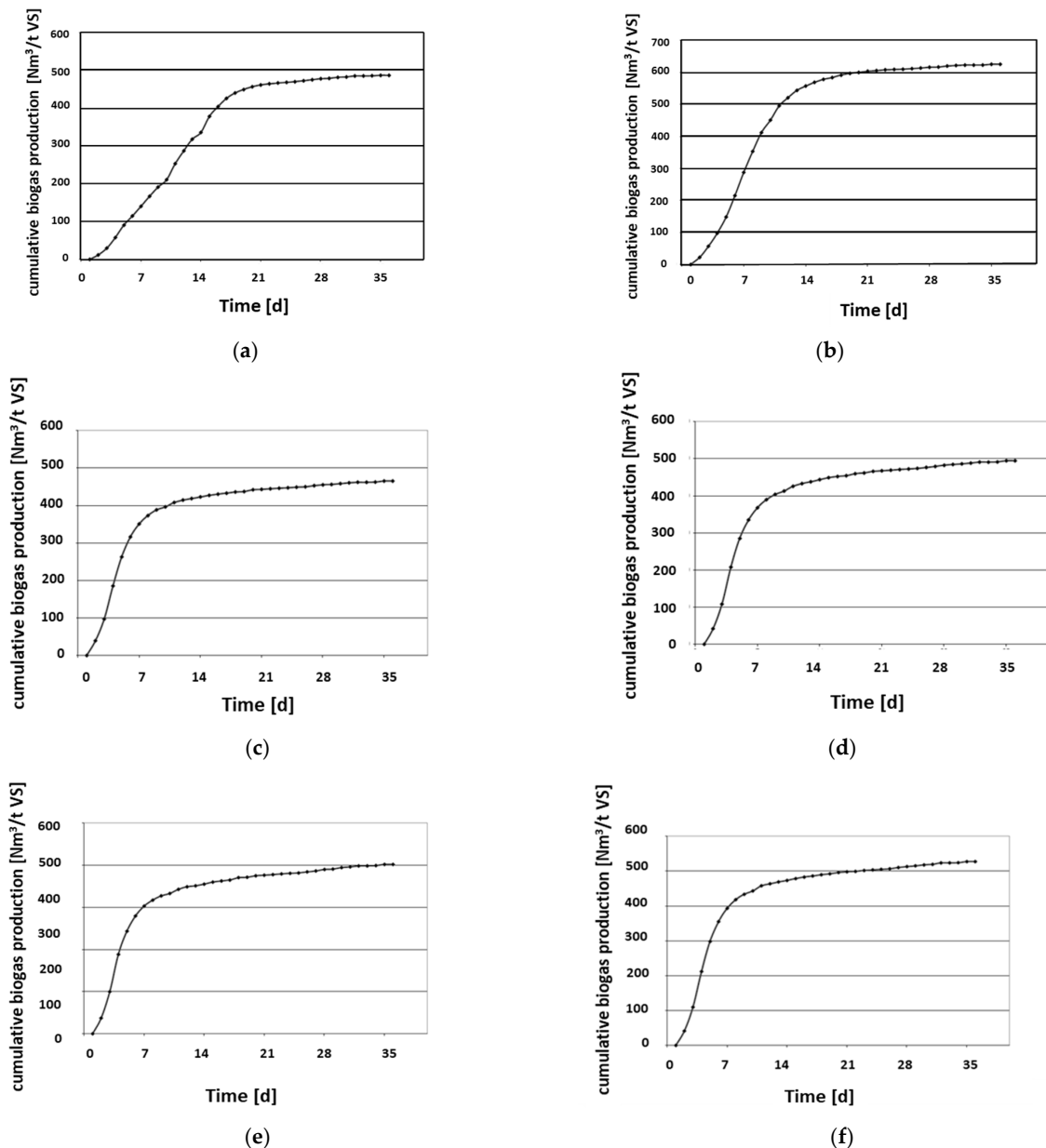


Figure 8. Cumulative biogas production curves for SS (a), CSW (b), the 2% CSW mixture (c), the 8% CSW mixture (d), the 12% CSW mixture (e), and the 25% CSW mixture (f).

The calculated synergy index values (Table 6) were below unity for all tested mixtures, indicating that the experimental methane yields did not exceed the theoretical values predicted from the weighted contribution of individual substrates. Therefore, no synergistic effect was observed under the investigated conditions.

The absence of synergy does not diminish the practical value of co-digestion. CSW exhibited a considerably higher methane potential than sewage sludge, and in the case of the 25% CSW sample resulted in increased methane production and improved energy recovery compared to SS mono-digestion. Consequently, the benefits of co-digestion in this study arise primarily from the utilization of a high-energy co-substrate rather than from positive biological interactions between substrates. From the perspective of wastewater treatment plant operation, such an increase in energy generation remains a significant technological advantage and supports the implementation of co-digestion strategies. As already explained, the absence of synergy does not diminish the practical value of co-

digestion. The benefits of co-fermentation may result from the use of a high-energy co-substrate rather than from positive biological interactions between the substrates.

Table 6. Results of synergy effects produced by co-digestion.

Substrate or Tested Mixture	BMP _{exp} Nm ³ CH ₄ /kg VS	BMP _{th} Nm ³ CH ₄ /kg VS	SI
SS	0.322 ± 0.016	0.322	-
CSW	0.452 ± 0.023	0.452	-
2% CSW	0.318 ± 0.016	0.325	0.98
8% CSW	0.316 ± 0.016	0.334	0.95
12% CSW	0.265 ± 0.013	0.339	0.78
25% CSW	0.344 ± 0.017	0.358	0.96

When analyzing the composition of the biogas, the highest methane concentration occurred in the biogas obtained from the AD of the CSW substrate (72%), while in the biogas from the AD of mixtures, these values ranged from 64% to 68%. The CH₄ content in the biogas from CSW was approximately 9% higher compared to SS. This is a direct result of the waste characteristics. Previous studies of other researchers have shown that the AD of waste with a high fat and protein content yields biogas with the highest methane content [27,28]. No upward trend was observed in the CH₄ content of the biogas as the proportion of CSW substrate in the mixture increased. Only in the case of the 2% CSW mixture was a higher CH₄ content relative to SS obtained, along with the lowest H₂S content in the biogas (210 ± 10.5 ppm). In the remaining mixtures, the average H₂S concentration was 320 ± 16.0 ppm, 285 ± 14.3 ppm, and 312 ± 15.6 ppm for 8% CSW, 12% CSW, and 25% CSW, respectively. However, it should be emphasized that the first portions of the biogas produced contained higher H₂S concentrations: 330 ppm, 420 ppm, 450 ppm, and 420 ppm, respectively, for the 2% CSW, 8% CSW, 12% CSW, and 25% CSW mixtures.

If this technology is applied in a municipal WWTP, an efficient biogas desulfurization system must be provided. The biogas obtained from the CSW substrate had the lowest H₂S content (approx. 5 ± 0.25 ppm) despite its elevated sulfur content. However, it should be noted that this waste contained significant amounts of iron (5.7 ± 0.29 g/kg), which likely effectively binds sulfur to iron sulfide—eliminating hydrogen sulfide from the biogas [29]. Simultaneously, CSW is characterized by a high total nitrogen content (6.70 ± 0.34 g/kg), and given its average VS removal of 66%, it can cause problems with excessively high concentrations of ammonium nitrogen, especially at high application rates. Excessively high concentrations of N-NH₄⁺ can inhibit the AD process. In the tested mixtures, this parameter ranged from 2.5 ± 0.13 g/kg to 3.0 ± 0.15 g/kg; thus, given the achieved VS removal there should be no issues with excessively high concentrations of ammonium nitrogen that could inhibit the AD process [30].

Differences were observed in the time at which the maximum daily biogas yield and 90% of the biogas were achieved (Figure 9a–f), as well as in the removal of volatile solids contained in the SS, CSW, and their mixture (Table 5). In the case of SS, the maximum biogas yield was recorded on day 10 (over 1.3 Nm³/t), and 90% of the biogas volume was obtained by day 16 of the study. In this case, a VS removal of 55 ± 2.75% was achieved. The AD process of the CSW substrate was more efficient—the maximum daily biogas yield was recorded on day 6 (approx. 9.5 Nm³/t), 90% of the biogas volume was obtained after 13 days, and by the end of the study, the VS removal had reached 66 ± 3.30%. The addition of CSW substrate to SS shortened the time during which the highest daily biogas yield occurred and during which 90% of the biogas volume was

obtained. In the first case, this occurred on the third day of the test, with values ranging from approx. $2.75 \text{ Nm}^3/\text{t}$ (12% CSW) to approx. $3.4 \text{ Nm}^3/\text{t}$ (25% CSW). In the second case, this occurred after 12 days for the 2% CSW and 25% CSW mixtures, after 13 days for the 8% CSW mixture, and after 14 days for the 12% CSW mixture. The highest degree of AD, recorded approximately 35 days later, was observed for the 8% CSW and 25% CSW mixtures, with an increase of approximately 4% and 9%, respectively, compared to the reference sample. In the remaining mixtures, this parameter reached lower values, ranging from $50 \pm 2.50\%$ and $45 \pm 2.25\%$ for the 2% CSW and 12% CSW mixtures, respectively, which means that at least half of the organic matter contained in the mixture did not decompose easily under anaerobic conditions and remained in the so-called post-digestion mass leaving the digester. The observed lack of an upward trend in biogas production between the 8% CSW and 12% CSW mixtures is most likely due to the heterogeneity of the substrate used for the anaerobic co-digestion experiments. The CSW used in the 12% CSW mixture likely contained a larger amount of lignocellulosic fraction than fats and proteins. It is well known that anaerobic conversion of lignocellulosic biomass results in lower biogas production and also requires a longer retention time.

Another factor that was analyzed in this study is process stability based on total alkalinity, VFA and AN contents, pH values as well as VFA/TA ratio (Table 7). It should be noticed that all analyzed parameters reached the values indicating a stable AD performance. Moreover, in all cases the VFA/ALK ratio was at a low level below 0.35 [31,32]. Importantly, regardless of the adopted co-substrate dose, no deterioration of those parameters was observed. Another positive effect achieved with regard to AN in digestate was the statistically significant reduced concentrations as compared to SS mono-digestion that were found. Drops from 27 to 40% were observed.

Table 7. Stability parameters in batch AD experiments (average values and standard deviation are given).

Parameter	Unit	SS	CSW	2% CSW	8% CSW	12% CSW	25% CSW
TA in feedstock	mg/L	731 $\pm$ 21.1 a	251 $\pm$ 14.2 b	768 $\pm$ 25.7 a	771 $\pm$ 31.30 a	768 $\pm$ 29.8 a	775 $\pm$ 33.2 a
TA in digestate	mg/L	4900 $\pm$ 147 a	3870 $\pm$ 128.1 b	5100 $\pm$ 89.2 a	5250 $\pm$ 55.7 a	5190 $\pm$ 53.1 a	5310 $\pm$ 57.1 a
VFA in feedstock	mg/L	545 $\pm$ 11.2 a	347 $\pm$ 28.1 b	560 $\pm$ 14.1 a	555 $\pm$ 49.7 a	567 $\pm$ 44.5 a	570 $\pm$ 54.5 a
VFA in digestate	mg/L	300 $\pm$ 15.3 a	210 $\pm$ 25.1 b	265 $\pm$ 37.2 a	275 $\pm$ 44.7 a	310 $\pm$ 10.7 a	307 $\pm$ 17.1 a
VFA/TA	-	0.061	0.054	0.048	0.049	0.060	0.058
AN in feedstock	mg/L	120 $\pm$ 10.9 a	89 $\pm$ 8.7 b	117 $\pm$ 1.1 a	134 $\pm$ 14.1 a	129 $\pm$ 12.5 a	130 $\pm$ 7.8 a
AN in digestate	mg/L	74 $\pm$ 7.1 a	59 $\pm$ 6.1 b	54 $\pm$ 6.8 b	47 $\pm$ 5.7 b	44 $\pm$ 4.8 b	49 $\pm$ 6.2 b
pH in feedstock	-	7.44 $\pm$ 0.37 a	7.62 $\pm$ 0.38 a	7.43 $\pm$ 0.37 a	7.44 $\pm$ 0.37 a	7.56 $\pm$ 0.38 a	7.51 $\pm$ 0.38 a
pH in digestate	-	7.67 $\pm$ 0.38 a	7.67 $\pm$ 0.38 a	7.60 $\pm$ 0.38 a	7.58 $\pm$ 0.38 a	7.56 $\pm$ 0.38 a	7.58 $\pm$ 0.38 a

Note: values followed by the same letter are not statistically different (ANOVA, $p < 0.05$, Tukey test).

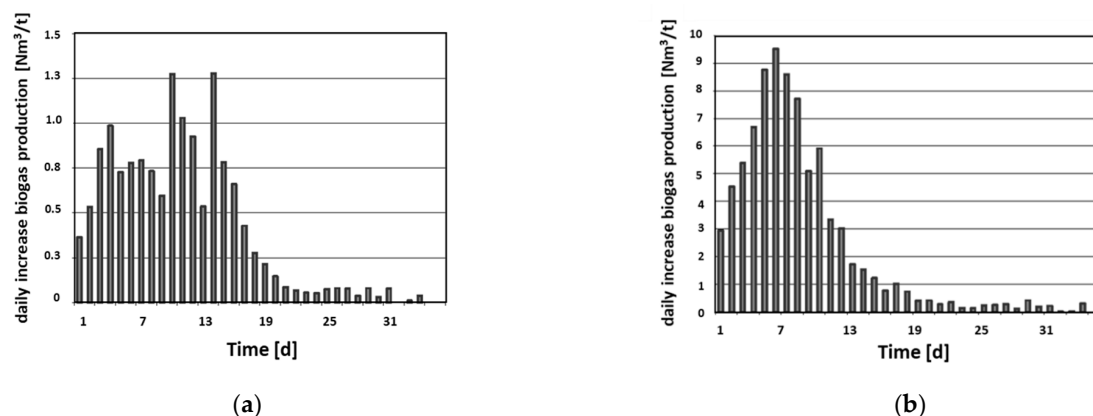


Figure 9. Cont.

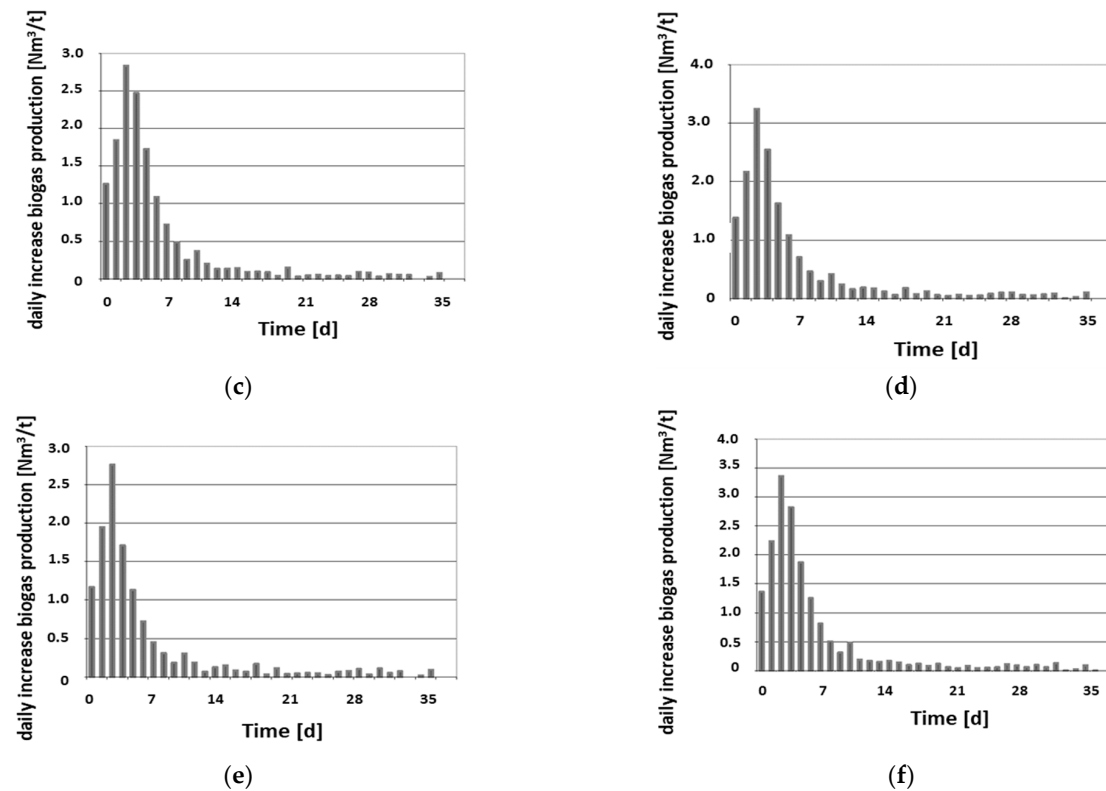


Figure 9. Daily biogas production for SS (a), CSW (b), the 2% CSW mixture (c), the 8% CSW mixture (d), the 12% CSW mixture (e), and the 25% CSW mixture (f).

3.3. Evaluation of the Co-Digestion Process Involving Cattle Slaughterhouse Waste

The estimated amount of waste from cattle slaughterhouses directed to the co-digestion process ranges from 47.26 to 102.10 kg TS/day, from 189.04 to 408.40 kg TS/day, from 283.56 to 612.60 kg TS/day, and from 590.75 to 1276.25 kg TS/day, respectively, for 2%, 8%, 12%, and 25% co-substrate content in the digested mass. Converted to volume, these are the following waste quantities: from 0.321 to 0.695 m³/d, from 1.286 to 2.778 m³/d, from 1.929 to 4.167 m³/d, and from 4.019 to 8.682 m³/d. The average values of volatile solids content in the co-digestion mass, in relation to the respective percentage shares, are 63.82 ± 7.82 kg VS/d, 255.29 ± 31.28 kg VS/d, 382.94 ± 46.99 kg VS/d, and 797.78 ± 97.75 kg VS/d. The hypothetical increase in biogas production in the digesters following the application of co-digestion with CSW is shown in Figure 10. In the AD process, this parameter was 1244.9 ± 350.9 Nm³/d. The co-digestion simulation indicates an increase in the amount of biogas produced: 1358.0 ± 166.4 Nm³/d for 2% CSW; 1522.7 ± 186.6 Nm³/d for 8% CSW; 1283.3 ± 157.2 Nm³/d for 12% CSW; and 1882.9 ± 230.7 Nm³/d for 25% CSW (Table 8). A rational approach to the co-digestion of SS with CSW allows for stable daily biogas production in the range of 1200–2200 Nm³/d, depending on the percentage of co-substrate.

The addition of fatty waste to the digesters allows increasing the organic loading rate of the chamber to 2.43 kg TS/(m³·d). With a rational approach, it is possible to increase the substrate loading by several percent, which—as previously noted—is advisable from the perspective of operating the digesters; currently, the fermentation chamber is operating at a lower substrate loading, ranging from 0.877 to 1.903 kg TS/(m³·d), while the design value of this parameter is 2.13 kg TS/(m³·d), and the literature/recommended value is up to approx. 2.25 kg TS/(m³·d). The use of the co-digestion process allows for increased energy production (Figure 11). The simulation shows that adding CSW to the AD mass

results in an increase in this parameter by an average of 9.1%, 22.3%, 3.1%, and 51.2% for 2% CSW, 8% CSW, 12% CSW, and 25% CSW shares of slaughterhouse waste, respectively.

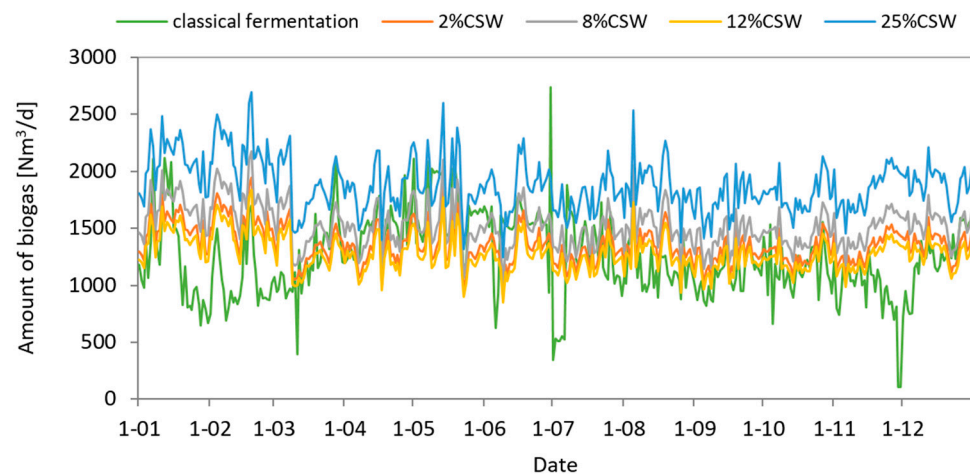


Figure 10. Changes in the amount of biogas produced in the co-digestion process.

Table 8. A summary of basic descriptive statistics for biogas production in the co-digestion process.

Value	2% CSW	8% CSW	12% CSW	25% CSW
Minimum	899.0	1008.1	849.5	1246.5
Maximum	1942.2	2177.8	1835.3	2692.9
Mean	1358.0	1522.7	1283.3	1882.9
Std. deviation	166.4	186.6	157.2	230.7
15th percentile	1195.3	1340.3	1129.5	1657.3
85th percentile	1529.4	1714.8	1445.2	2120.4

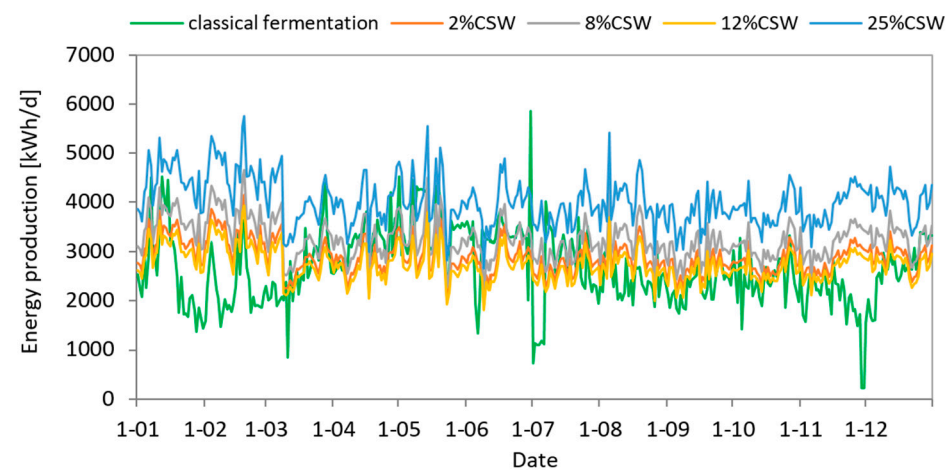


Figure 11. Changes in estimated energy production (taking co-digestion into account).

3.4. Strategic Assessment of the Proposed Co-Digestion Approach

SWOT analyses allow for the translation of the experimental results into a practical framework that can support decision-making by WWTP operators, policymakers, and investors. The use of this kind of fatty waste as a co-substrate in the anaerobic digestion process may bring both technological and energy benefits, but also potential operational risks. Table 9 presents a SWOT analysis, highlighting the strengths and weaknesses of the proposed solution, as well as the resulting opportunities and threats.

Table 9. A SWOT analysis for the simulated co-digestion process.

Strengths	Weaknesses
- the potential to increase biogas and methane production - more efficient use of existing infrastructure for sludge stabilization - improved energy balance of the wastewater treatment plant - a potential source of revenue due to the ability to accept waste	- risk of destabilizing the AD process if the CSW content is too high in LCFAs - potential for scum and foam formation in the digester - changes in the rheological parameters of the sludge - the need for modifications to the process system, including equipment for receiving and storing fats (tanks, heating systems, pumps) - the need to homogenize the sludge and co-substrate before introducing them into the digester - the potential for increased load on the sludge dewatering system
Opportunities	Threats
- increased production of electricity and/or heat - reduced operating costs for the wastewater treatment plant (lower energy purchase costs) - opportunities for collaboration with industry (facilities supplying co-substrates) - improved energy efficiency of the treatment plant (ultimately meeting the requirements of the new Wastewater Directive) - increased chances of acquiring funding for energy and biogas investments as part of the implementation of a circular economy	- variability in the quantity and quality of CSW - risk of AD instability due to uncontrolled co-substrate dosing - operational issues with the process system (pipeline blockages, fat buildup, pump problems) - increased hydrogen sulfide concentration in biogas (potential for system corrosion) - health or odor risks associated with co-substrate storage - changes in regulations regarding the acceptance of external waste

A SWOT analysis enabled a rational assessment of technological, operational, environmental, and economic factors that may be important for implementing the proposed co-digestion strategy on a real scale. A similar approach was previously used to strategically evaluate sewage sludge management technologies [33]. Cengiz et al. [34] indicated that the implementation of co-digestion technology should consider not only methane yield but also technical, environmental, and operational aspects. The analysis clearly indicated that the main advantage of the proposed solution is increased energy recovery resulting from the use of a high-energy co-substrate without the need to build new anaerobic digestion infrastructure. Furthermore, such a strategy enables better organic waste management and supports the concept of a circular economy. The main weaknesses and threats of this solution are related to the variability of CSW composition, co-substrate delivery logistics, and potential process inhibition due to excessive organic loading.

4. Discussion

The quantity and composition of biogas depend on the chemical composition of the feedstock undergoing AD and the selected process parameters, namely retention time, temperature, and the organic loading rate of the reactor [35]. Currently, the most commonly used systems employ a mixture of several substrates. The diversity of substrates contributes to better biogas parameters and increases the security of raw material supply. The feedstock, i.e., the substrate for biogas production, should also be selected with a view to maximizing biogas yields, ensuring the stability of the AD process, and enabling the utilization of the resulting digestate [36,37].

The CSW used in the study exhibited characteristics typical of this type of waste, namely a high content of organic matter expressed as TS, VS, and TOC [27,38,39]. In particular, the presence of readily biodegradable compounds in CSW appears to be especially

beneficial, as they undergo rapid conversion to methane [10]. Additionally, a high iron content in the substrate can have a beneficial effect on the AD process. This macronutrient has the ability to bind hydrogen sulfide, acts as a cofactor for key enzymes, serves as a micronutrient for methanogens, and stabilizes the process [40].

The substrate used also exhibited an elevated sulfur content compared to SS. Under anaerobic conditions, sulfur is reduced by sulfate-reducing bacteria (SRB) to hydrogen sulfide, which can result in a deterioration of biogas quality and cause technological problems related to corrosion [41]. Previous studies have shown that sulfur has a toxic effect on AD microorganisms, particularly methanogens; inhibition was observed at sulfur concentrations ranging from 100 to 800 mg/L [42]. Another problematic issue is the elevated nitrogen content in the applied substrate. Moderate amounts of nitrogen in the AD process are beneficial; it is an essential nutrient for protein synthesis by anaerobic microorganisms. However, an excess of nitrogen can lead to ammonia inhibition, which in turn can cause process instability, low biogas production efficiency, and even system failure [43,44]. The applied substrate also contained heavy metals, with elevated levels of Ni, Zn, and Cu observed. However, their presence is essential for proper anaerobic decomposition. A moderate amount of Cu (>300 mg/kg) in the reactor feedstock is essential for maintaining microbial activity, as Cu acts as a cofactor stimulating enzymatic reactions. Ni is considered the most toxic chemical element; however, it is essential for maintaining the activity and metabolism of microorganisms [45]. Zn, on the other hand, improves the efficiency of protein hydrolysis. Additionally, it is considered an essential element for methanogens, as it limits the accumulation of short-chain fatty acids and increases methane yield [46].

The studies conducted thus far have shown that methane production from food waste depends on its physicochemical characteristic, origin, and applied dose. Its methane potential varies widely, ranging from 0.4 to 0.8 m³ CH₄/kg VS, depending on the exact composition—i.e., the proportion of fats, proteins, and carbohydrates [38,47]. In this study, the obtained methane potential of the substrate was relatively low, and established at the level of 0.452 m³ CH₄/kg VS, which resulted from the heterogeneity of the substrate, and in particular the presence of a lignocellulosic fraction, resulting from the presence of feed residues, straw, and hay in the ruminants' stomachs (Figure 2). It should be noted that the AD of the CSW substrate, in terms of biogas yield, showed an efficiency similar to the literature data summarized in Table 10, which presents the results of the methanogenic digestion of selected fatty waste from slaughterhouses.

Table 10. Biogas potential of selected slaughterhouse waste (authors' own elaboration based on Czarkowska and Czarkowski [48]).

Substrate Type	Biogas Yield		Methane Content in Biogas [%]
	[m ³ /t Fresh Matter]	[m ³ /t VS]	
Pig stomach contents	20–60	250–450	60–70
Ruminant stomach contents	20–60	200–400	58–62
Flotation sludge	35–280	900–1200	60–75
Blood	50–70	300–450	50–55

The study evaluated the effectiveness of co-digestion a selected CSW substrate with SS at four different concentrations, where the percentage of the co-substrate in the tested mixture was 2%, 8%, 12%, and 25%, respectively. The obtained results showed that the addition of 25% *v/v* CSW to SS resulted in an increase in biogas and methane production by approximately 8% and 7%, respectively, compared to the control sample. In the remaining cases, comparable results or lower values were obtained compared to the mono-digestion

of SS. This was mainly due to the presence of the lignocellulosic fraction, which is difficult to biodegrade [49,50].

Co-digestion of some substrates can result in synergistic or antagonistic effects. No synergistic effect was obtained in these studies (Table 6). Nielfa et al. [21] conducted research on the co-digestion of biological sludge and organic fraction municipal solid waste (OFMSW). Their BMP test results confirmed synergism between the tested substrates. SI values ranged from 1.065 to 1.102. The effect increased with the addition of OFMSW. The authors indicated that the synergistic effects could be determined by, for example, additional alkalinity, trace element content, or nutrients. Arribas et al. [51] obtained antagonistic effects for mixtures with 5%, 15%, and 25% weight of biological sludge as co-substrate (major substrate—solid waste), and as the authors point out, competitive effects could be caused by pH, ammonia toxicity, or high concentrations of volatile acids. Co-digestion can provide synergistic benefits, but the effectiveness of the process depends on the choice of substrates and the proportions [17].

The studies on the co-digestion of SS with slaughterhouse waste conducted to date have shown that methane production with the addition of this waste varies widely; depending on the type of waste and the process parameters used, values ranging from 0.46 to 0.735 m³ CH₄/kg VS have been reported [11,52]. Table 11 shows the examples of co-digestion of SS with various lipid-rich wastes.

Table 11. Process performance in co-digestion of SS and lipid-rich wastes.

Lipid Waste Type	Co-Substrate Share in the Feedstock <i>v/v</i> on VS Basis	T °C	Methane Content (%)	VS Removal (%)	Biogas/Methane Yields (Y _b /Y _m)	Improvement vs. SS Mono-Digestion	The Most Beneficial Effect	Reference
grease trap sludge	10–30%	35	65–71%	45–58	0.295–0.344 m ³ CH ₄ /kg VS	9–27%	Pilot-scale AcD 30% <i>v/v</i> , Y _m = 0.344 m ³ CH ₄ /kg VS	[53]
slaughterhouse waste	20–60%	37	70.5–78.3	60.6–70.4	0.647–0.736 m ³ /kg VS	49–70%	40% <i>v/v</i> , TS = 4%, Y _b = 0.736 m ³ /kg VS	[54]
greasy sludge from flotation system (meat industry pork processing)	7–87%	36	63–39	17–43	0.158–0.546 m ³ CH ₄ /kg VS	60–210%	52% <i>v/v</i> , Y _m = 0.546 m ³ CH ₄ /kg VS	[55]
trapped grease waste from WWTP	4–37%	35	68–70	44–55	0.278–0.369 m ³ CH ₄ /kg VS	110–150	23% <i>v/v</i> , Y _m = 0.369 m ³ CH ₄ /kg VS	[56]
grease interceptor waste from a foodservice establishment	10–40%	37	68.2–70.1	-	0.502–0.752 m ³ CH ₄ /kg VS	280–420	20% <i>v/v</i> , Y _m = 0.75 m ³ CH ₄ /kg VS	[57]
pig slaughterhouse wastes	30–50%	35	67.4–77.4	57–62.16	0.472–0.609 m ³ CH ₄ /kg VS	96–140	50% <i>v/v</i> , Y _m = 0.609 m ³ CH ₄ /kg VS	[58]
palm oil mill effluent	50%	37			0.151 m ³ CH ₄ /kg COD _{added}	180%		[59]
poultry slaughterhouse wastes	5%TS 7%TS	34	73%	66	5% TS: 0.109–0.631 m ³ /kg VS 0.024–0.462 m ³ CH ₄ /kg VS 7% TS: 0.125–0.443 m ³ /kg VS 0.013–0.323 m ³ CH ₄ /kg VS	-	5% TS Y _m = 0.462 m ³ CH ₄ /kg VS Y _b = 0.631 m ³ /kg VS	[60]
fat, oil and grease	27.3: 72.7%	35		83%		96.8%	244 mL/gCOD added	[61]
flotation sludge	70 and 90 <i>v/v</i> %	37					90% <i>v/v</i> 0.529 m ³ CH ₄ /kg VS removed	[62]

Co-digestion with SS also showed a significant methane yield ranging from 65 to 78%. In contrast, their addition to SS resulted in an increase in methane production of up to 175% compared to the mono-digestion of SS [63]. Compared to the presented data, a relatively small increase in methane production was observed compared to the AD of SS alone. The lower values obtained for the slaughterhouse waste used result, among other things, from the lower fat content in its composition, while a higher proportion of the lignocellulosic fraction stems from the presence of feed residues, straw, and hay in the stomachs. It should be noted that slaughterhouse waste itself constitutes a diverse group, which includes slaughterhouse blood, stomach and intestinal contents, offal, tissues, and other meat and fat waste, and its exact composition.

On a technical scale, the waste from the poultry industry applied to anaerobic digesters at the WWTP in Kozięgłowy (Poznań, Poland) resulted in an increase in the biogas production rate of up to 30% (from 0.38 m³/kg VS to 0.49 m³/kg VS) [64]. Promising results were obtained at the WWTP in Brzeg, where the feasibility of co-digestion SS with waste fats was investigated. After introducing the co-substrate, an increase in biogas production of up to 80% was observed, yielding 107 m³ of biogas from 1 Mg of fats. Studies have shown that a fat content of up to 25% in a single batch of fermentation mass fed into the digester does not disrupt the mesophilic AD, even with varying properties of the waste fat. However, a negative aspect of feeding fat—a substrate with a higher organic content than sludge—is a reduction in VS removal [65]. At the WWTP in Rzeszów (398,000 PE), biogas yield ranged from 0.252 to 0.519 Nm³/kg TS during the co-digestion of SS with waste fats (edible fats, used cooking oils) and distillery stillage [66]. Waste from the poultry industry is also frequently used as a component in co-digestion mixtures. The biogas potential of poultry waste varies widely and can range between 0.57 Nm³/kg TS [67], 0.61 Nm³/kg TS [68], 0.631 Nm³/kg TS [60], and even 1.24 Nm³/kg TS [64]. Significant co-digestion efficiency was also observed when using SS mixed with fat sludge from the preliminary treatment plant of a poultry slaughterhouse (the WWTP in Ilawa). Fat waste from poultry processing was added to the SS mixture in the form of an emulsion with a total solids content of 11.5%. The average daily volume of waste fat ranged from 9.8 to 22.6 m³/d, with an average value of 14.7 ± 3.4 m³/d. From 1 kg of TS of all substrates introduced into the digesters during the period under consideration, 0.220 to 0.451 Nm³ of biogas was obtained, and the average unit biogas yield was 0.321 Nm³/kg TS [69].

An analysis of the topic indicates the possibility of co-digestion SS with slaughterhouse waste, which includes intestines, stomachs, rumen contents, adipose tissue, blood, and fat. The proportion of slaughterhouse waste that can be used in the co-digestion process depends on the dry matter content [70]. As has already been demonstrated, substrates from slaughterhouses and meat processing plants have high energy potential for biogas production. The use of slaughterhouse waste for co-digestion requires proper preparation. Some slaughterhouse waste should be shredded, homogenized, and sometimes even sanitized before use.

5. Conclusions

An analysis of the current situation revealed that the digester at the WWTP in Dębica was operating at a low TS load (resulting in low digestion efficiency and biogas production). The implementation of the co-digestion process of SS with CSW for the analyzed WWTP might lead to an increase in biogas production, and hence an increase in energy production ranging from 3.1% to 51.3% on average, depending on the substrate composition, allowing in this way the energy neutrality of this facility.

Another argument for using CSW as co-substrate to SS is the possibility of energy recovery from waste, the disposal of which is problematic and requires significant financial

investment. On the other hand, challenges may include technical issues related to clogging of the anaerobic digester and process lines, the formation of foam in the reactor, and the leaching of biomass, as well as the need for strict control of the co-digestion process due to the potential for inhibition by LCFAs and ammonia. Another issue that should be considered is increased hydrogen sulfide content in biogas, which may lead to the necessity of using highly effective solutions for biogas purification; further studies in this field should be conducted.

The SWOT analysis confirmed the practical suitability of CSW as a co-substrate for SS digestion. However, implementation of this strategy should be preceded by a technical and economic analysis for specific locations, taking into account local operating conditions and substrate availability.

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Abbreviations

The following abbreviations are used in this manuscript:

AD	anaerobic digestion
BMP _{exp}	experimental value of methane yield from the co-digestion mixture
BMP _{th}	calculated theoretical value of methane yield for individual mixture
CSW	cattle slaughterhouse waste
ES	excess thickened sludge
EXS	ether-extractable substances
HRT	hydraulic retention time
LCFAs	long-chain fatty acids
OFMSW	organic fraction municipal solid waste
PS	primary thickened sludge
SI	synergy index
SS	sewage sludge
TOC	total organic carbon
TS	total solids
WWTP/WWTPs	wastewater treatment plant/plants
VS	volatile solids

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