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Effects of seasonal and local co-feedstocks on the performance of continuous anaerobic digestion of cattle slurry

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ABSTRACT

The study's aim was to assess the robustness of farm-type anaerobic digestion with cattle slurry as the main feedstock under the change of co-feedstock type and quality. Fish biomass, rainbow trout offal, potato, and reindeer offal were investigated as possible co-feedstocks in a 428-day semi-continuous reactor experiment. Using fish biomass or rainbow trout offal as co-feedstock (19 % of VS of the feed) produced an average of 220 and 305 L CH_4/kg VS compared to the 201 L CH_4/kg VS for the manure control. For other co-feedstocks, the differences were not statistically significant. Despite recommendations to acclimate biogas process to new feedstocks with caution, no disturbances in the biogas process were observed, even with sudden changes in co-feedstocks. The use of locally available and seasonal co-feedstocks improved the agronomic quality of the digestates and could be important in securing farms' supply of and self-sufficiency in both energy and fertilizers.

1. Introduction

Global and EU goals for the circular economy encourage the more efficient and sustainable use of different materials through e.g., anaerobic digestion (AD). AD is a widely used technology for processing and stabilizing biomasses and producing renewable energy and recycled nutrients for agriculture. AD is used in both urban and rural contexts; large digesters are often practical in urban areas near municipal and industrial feedstocks and end-use possibilities for biogas. However, in rural areas, AD plants are usually smaller and utilize different agricultural biomasses, e.g., manure and crop biomasses, and provide energy and nutrients for farms or farm cooperatives or larger networks, depending on the location (Ahlberg-Eliasson et al., 2017). Such agricultural AD plants have been reported to also digest co-feedstocks, e.g., different types of food industry waste (Ahlberg-Eliasson et al., 2017; Chodkowska-Miszczuk et al., 2021). With co-feedstocks, it is possible to increase the biogas and nutrient production capacity of farm-scale digesters and the self-sufficiency and economy of a farm (Winquist et al., 2019). However, in sparsely populated remote areas, there may not be a steady flow of certain co-feedstocks, because the availability of certain biomasses can be highly seasonal (Ervasti et al., 2019). This poses a challenge to farm-scale AD plants, because digesters are conventionally designed for steady flows of certain feedstocks, and rapid changes in feedstock compositions could compromise the process stability and the biogas production itself (Ghofrani-Isfahani et al., 2020).

The rapid change of feedstock composition can affect the stability of the digestion process, as the microbial communities lack the time to adapt to changes in the feedstock composition. This could lead to an accumulation of process intermediates such as volatile fatty acids (VFA) (Ghofrani-Isfahani et al., 2020). In addition, increasing the portion of protein-based feedstocks can also elevate the nitrogen load on the digester, which after mineralization into ammonia, can inhibit methanogenesis and cause process disturbances. However, co-feedstocks often have a synergistic effect with the main feedstocks of the AD process, which can lead to increased biogas production (Ebner et al., 2016; Tufaner and Avşar, 2016). The use of co-feedstocks can balance the C/N ratio to avoid ammonia inhibition and elevate concentrations of trace elements, which improves the process. In addition, pH and buffer capacity can be enhanced by the selection of specific co-feedstocks (reviewed in Tufaner and Avşar (2016)).

Facilitating the change of feedstock composition would increase the flexibility of a farm-scale biogas plant and reduce dependence on the availability of certain types of feedstocks. The flexibility of AD plants is usually considered as the ability to produce electrical energy according

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to its demand, which can be achieved by various engineering solutions, as well as by managing the substrate and its feeding (Mauky et al., 2017). Flexibility can also be seen as the ability to tolerate different process temperatures or organic loading (Ahlberg-Eliasson et al., 2021). Previously, a change of feedstocks has been experimentally studied from the process control perspective (Ghofrani-Isfahani et al., 2020) but not focusing on the seasonality and/or availability of the feedstocks. Utilizing locally produced biomasses in a farm-scale biomass plant in remote sparsely populated areas has advantages for not only the AD plant (Wang, 2014) but also the local waste management and environment. In remote areas, the produced biomasses, from e.g., the food industry and slaughterhouses, are conventionally either transported hundreds of kilometers to be utilized in centralized treatment plants or landfilled, with the risk of uncontrolled greenhouse gas emissions and nutrient loading. If suitable centralized treatment is unavailable, processing these materials locally in farm-scale AD plants constitutes a more sustainable and resource-efficient use of these materials. The use of local feedstocks can also promote nutrient recirculation and the minimization of eutrophication if excess nutrients are removed from the lakes through fish removal (Boros, 2022), then further utilized in AD and used as fertilizers in farm's crop production.

This study's aim was to assess the biogas production potential of cattle slurry digestion with respect to the change of co-feedstocks. Moreover, our study evaluated the robustness of the process – whether it could tolerate rapid changes in co-feedstock composition. The experimental setup simulated a farm-scale operation, with the farm's own cattle slurry and excess grass silage as base feedstocks for the AD process. The long-term co-digestion performance of the reactor with different locally and seasonally available feedstocks – potato biomass, fish biomass from fish removal, offal from farmed rainbow trout, and reindeer offal – were studied using laboratory-scale digesters. This approach provides novel insights to the use and rapid changes of locally and seasonally available co-feedstocks with distinctive properties.

2. Materials and methods

2.1. Origin of materials

The main feedstock of the AD process was dairy cattle slurry (CS) collected from the research dairy farm of Natural Resources Institute Finland (Luke) in Jokioinen, Finland. At the time of the experiment, the farm housed 209 dairy cows and heifers in loose housing. The slurry was mixed prior to the pumping and stored in a 1 m^3 plastic container at +2 °C for the duration of the experiment. Grass silage (GS) was a mixture of timothy and meadow fescue from the 2nd harvest (Jokioinen, Finland). Potato (P) biomass consisted of second-class almond potatoes which had been rejected for consumer use due to their size. The potato sample originated from a potato farm in southern Lapland. Fish (F) mass consisted of common roach (*Rutilus rutilus*) and common bream (*Abramis brama*) from fish removal (biomanipulation). The guts from farmed rainbow trout were collected from a freshwater fish farm (Kemijoki, Finland) and referred to as fish offal (FO). Reindeer intestines were obtained from one slaughtered adult animal, referred to as reindeer offal (RO). The feedstocks, excluding the slurry, were stored in a freezer (−20 °C), thawed in weekly portions, and then stored at +4 °C. Co-feedstocks were ground using knife mills (Oviation 3, Moulinex, France; Retsch Grindomix GM300 knife mill, Retsch Gmbh, Germany).

Inocula were taken from a mesophilic wet-type farm-scale biogas reactor (Luke Maaninka, Kuopio, Finland), treating the slurry from 120 dairy cows. The inoculum for batch tests and the continuous experiment was taken at different timepoints. Prior to batch tests, the inoculum was sieved to obtain an even composition and remove coarse particles.

2.2. Batch test

Biochemical methane potential (BMP) and residual methane potential (RMP) were determined using an AMPTS II automated test system (Bioprocess Control Ltd., Sweden). BMPs were determined for both individual feedstocks and feedstock mixtures. In co-digestion tests, the share of feedstocks corresponded to the feed mixtures in continuous experiments. The tested materials were added to 500 mL borosilicate test bottles according to a substrate/inoculum volatile solids (VS) ratio of 0.5. In co-digestion tests, the amount of substrate referred to the total VS of the substrate mix. Sodium bicarbonate was used as a buffer with a dosing of 3 g/L, and distilled water was added to the bottles to reach a uniform liquid amount of 400 g. The headspaces of the bottles were flushed with N_2 to attain anaerobic conditions. The carbon dioxide of biogas was trapped in 3 M sodium hydroxide, after which the CH_4 volumes were measured using a water-displacement-based system. Samples were incubated for 55 days in mesophilic conditions (37 °C), and the contents of the bottles were mechanically mixed for 1 min per hour at 84 rpm. RMP tests were not inoculated; 300 g of digestate were added to 500 mL bottles, and the bottles were flushed with N_2 gas. No buffering agent was used. RMP tests were run for 60 days at 37 °C. All batch tests were performed as triplicates.

2.3. Setup of continuous experiments

Continuous reactor experiments were run in two identical stainless-steel laboratory-scale reactors (Metener Ltd., Finland), referred to as R1 and R2, with a working volume of 10 L each. The reactors were operated for 428 days under mesophilic conditions (37 °C). Feeding was carried out manually on working days, and the digestate overflowed by gravity via a U-tube. The reactors were mixed (32 rpm) semi-continuously with a mixing cycle of 5 s on and 60 s off. Biogas volume was measured with a reactor-specific drum-type gas meter (Dr.-Ing. Ritter Apparatebau GmbH & Co. KG), after which the gas was collected into aluminum gas bags (Tesseraux Spezialverpackungen GmbH). Cumulative biogas volumes and compositions were analyzed on working days.

2.4. Experimental procedure of continuous experiments

Two parallel semi-continuously stirred tank rectors (SCSTR) simulated the operation of a farm-scale biogas plant using local feedstocks. The experiment was started with control periods using typical farm materials, cattle slurry, and grass silage as feedstocks. The use of co-feedstocks with different characteristics was tested sequentially one at a time. The experiment consisted of a total of 6 feeding periods (Table 1). Shifts between co-feedstocks were conducted with full organic loads of both main- and co-feedstocks without the acclimatization of microbiota.

At the start of the experiment, both reactors R1 and R2 were filled up to the operational volume with the inoculum. The inoculum had a TS of 5.7 % and VS of 4.1 %. The first control feeding period, period 1 (P1), was run for 55 days (days 1–55) as a mono-digestion of cattle slurry followed by a second control feeding period, period (P2). P2 was run for 61 days (days 56–116), using the usual farm-scale feedstocks, cattle slurry, and grass silage. Test periods with co-feedstocks (P3–P6) were run for at least 3 hydraulic retention times (HRTs) each, apart from the last P6, in which the availability of co-feedstock restricted the length of the test period to 2 HRTs. The HRT was a constant 23 days throughout the experiment. Distilled water was used in the required amounts to equalize the HRTs of all periods.

The organic loading rate (OLR) varied from 1.6 $kgVS/m^3d$ in the first control period to 2.2 $kgVS/m^3d$ in the second control period and reached 2.8 $kgVS/m^3d$ in the co-feedstock test periods (Table 1). The amount of cattle slurry feed was kept constant throughout the experiment, as well as the amount of grass silage when it was used. The VS proportion of co-feedstock was 19 % of the total amount of VS addition (Table 1).

Digestate sampling was performed once a week from the parallel reactors, prior to daily feeding. RMP tests were done from the digestate collected after periods 2–6. In addition, a more detailed characterization of digestates, including organic composition, nutrient value, and trace

Table 1

Feeding periods, period durations, organic loading rates (OLRs), hydraulic retention times (HRTs), and proportions of the total amount of volatile solids (VS) and fresh matter (FM) fed to the reactors.

Period	Feedstocks	Operating days	OLR (kg VS/m^3d)	HRT (d)	Composition of feed (VS basis)	Composition of feed (FM basis)[a]
1	Cattle slurry (CS)	1–56	1.6	23	CS 100 %	CS 88 %
2	CS + Grass silage (GS)	57–117	2.2	23	CS 71 %; GS 29 %	CS 88 %; GS 4.6 %
3	CS + GS + Potato (P)	118–200	2.8	23	CS 57 %; GS 24 %; P 19 %	CS 88 %; GS 4.6 %; P 5.0 %
4	CS + GS + Fish (F)	201–285	2.8	23	CS 57 %; GS 24 %; F 19 %	CS 88 %; GS 4.6 %; F 6.1 %
5	CS + GS + Fish offal (FO)	286–385	2.8	23	CS 57 %; GS 24 %; FO 19 %	CS 88 %; GS 4.6 %; FO 2.4 %
6	CS + GS + Reindeer offal (RO)	386–428	2.8	23	CS 57 %; GS 24 %; RO 19 %	CS 88 %; GS 4.6 %; RO 8.0 %

[a] Water was added to the feedstock mixture in periods 1–5 to equalize HRTs.

metals was done after each period.

2.5. Chemical analyses

Fresh feedstock and digestate samples were analyzed for total solids (TS) and VS using the standard SFS 3008 method (Finnish Standards Association, 1990). The pH was measured with a VWR pH 110 pH-analyzer (VWR International). Total Kjeldahl nitrogen (TKN) was determined according to a standard method (AOAC, 1990) as in Ervasti et al. (2019), and ammonium nitrogen (NH_4-N) according to McCullough (1967). For the C/N ratio, total C and N were analyzed using Duma's method according to the manufacturer's instructions with a Leco CN-2000 Elemental Analyzer (Leco Corp., USA). Soluble chemical oxygen demand (sCOD) was analyzed according to SFS 5504 (Finnish Standards Association, 2002). VFAs, i.e., acetic, propionic, isobutyric, n-butyric, isovaleric, valeric, and caproic acids, were analyzed as in Tampio et al. (2014) using an HP 6890 gas chromatograph (Hewlett-Packard, Little Falls, USA) with a 10 m × 0.53 mm × 1 μm HP-FFAP capillary column (Agilent Technologies, USA) and a flame ionization detector. From freeze-dried samples, the total sugars were determined by colorimetric method (VALORGAS, 2011), crude fat with a Soxcap-Soxtec-Analyser (AOAC, 1990; Foss Tecator Application Note AN 390), and the protein content was calculated by multiplying the organic nitrogen (TKN – NH_4-N) by 6.25. Total phosphorus (Ptot), total potassium (Ktot), and other trace elements (Ca, Cu, Fe, Mg, Mn, Na, Zn) were analyzed with an ICP-OES according to the manufacturer's instructions after HNO_3 digestion (Huang and Schulte, 1985). Soluble phosphorus was determined after filtration with a Skalar Scan+ analyzer. Gas composition (methane (CH_4) and carbon dioxide (CO_2)) was analyzed with a gas chromatograph (Perkin Elmer Arnel Clarus 500 (Pakarinen et al., 2008) on days 1–67. On days 68–428, gas composition (CH_4, CO_2, and hydrogen sulfide (H_2S)) was measured with a portable Combimass GA-m gas analyzer (Binder Engineering GmbH, Germany).

2.6. Calculations

TS contents with the highest total VFA contents (GS and RO) were compensated due to the compounds volatilized during the analyses according to Porter & Murray (Porter and Murray, 2001) (Eq. (1)).

$$\text{Compensated total solids (g/kg)} = 19.96 + 0.9793^*(1.011^*\text{TS}_{100} + 1.24), \quad (1)$$

where TS_{100} is the oven total solids (TS_{100} = oven at 100 °C).

The productions of CH_4 in batch tests (BMP and RMP) were calculated as L CH_4/kg VS, and the results are given as averages of three parallel bottles with their standard deviations. The estimated CH_4 production in co-digestion in batch tests was calculated as a weighted sum of the BMPs of individual feedstocks. Theoretical biochemical methane potential (BMP_{th}) was calculated with the method derived from Nielfa et al. (2015) and Yan et al. (2021). The method is based on the specific theoretical stoichiometric CH_4 production capacities of different organic fractions, which for carbohydrates are 415, for proteins 496, for lipids 1014, and for VFA 373 L CH_4/kg VS. In the present study, the analyzed total sugars concentration was used to describe carbohydrates.

In the SCSTR experiment, the OLR (kg VS/m^3d) is given as the daily average of substrates fed to the reactor over a one-week period, because the feeding was accomplished only for 5 days a week, excluding public holidays. Volumetric CH_4 production was calculated as liters of CH_4 per liter of digester working volume per day (L CH_4/L d), and specific methane production (SMP) was calculated as liters of CH_4 per g of feedstock VS added. For biogas, calculations were done correspondingly. CH_4 and biogas production results, both volumetric and specific, are presented as averages over a one-week (7 days) period, which includes both feeding and non-feeding days. All gas volumes were converted to STP conditions (101.3 kPa, 273.15 K) according to the ideal gas law.

2.7. Statistical analyses

The specific methane production (L/kg VS) and the pH levels were not normally distributed, and the variances differed substantially between different feeding periods. Thus, basic parametric methods such as *t*-test or ANOVA were not applicable. Comparisons between the average values were therefore carried out using non-parametric methods. The H_2S and NH_4-N levels showed a clear trend line, so comparing averages would be meaningless, since it is not obvious from the graphs that an equilibrium state was achieved for any period. What comes to other variables, they were measured less frequently resulting in insufficient number of measurements to obtain the necessary statistical power to detect meaningful differences.

To determine whether the methane production and the pH levels differed between the feedstocks, the sample distributions and their differences were simulated by a non-parametric bootstrap (Efron and Tibshirani, 1986). A two-tailed alternative hypothesis was adopted. For each comparison, 100,000 bootstrap samples were simulated.

Given the large number of comparisons made (60 in total), there is a need to adjust the p-values to control the false discoveries. In addition to the trend lines detected in other variables, this is an important reason why it is not possible to carry out statistical test for every variable while preserving sufficient statistical power for the primary variables of interest. We take a conservative approach here, using Bonferroni-corrected p-values for statistical inference (Bonferroni, 1936). For transparency and meta-analysis purposes, we also report the unadjusted p-values. All calculations were performed with the statistical software R (R Core Team, 2022).

3. Results and discussion

3.1. Feedstock characteristics

Cattle slurry, a common feedstock for farm-scale biogas production, had lower TS and VS content (TS 5.1 %, VS 4.2 %) than the studied co-feedstocks (TS 16.9–51.2 %) (Table 2). Higher VS contents are generally preferred when aiming to increase energy production with co-feedstocks. Cattle slurry characteristics corresponded with previously reported results (TS of 6.2 % reported for cattle slurry samples in Solli et al. (2014); an average C/N ratio of 10.6 was reported in Ahlberg-Eliasson et al. (2017)). The protein content of the CS (154 g/kgTS)

Table 2
Characteristics of the studied feedstocks.

	Cattle slurry (CS)	Grass silage (GS)	Potato (P)	Fish (F)	Fish offal (FO)	Reindeer offal (RO)
TS (%)[a]	5.1 ± 0.2[b]	34.0 ± 1.3[c]	25.7	25.6	51.2	16.9
VS (%)[a]	4.2 ± 0.1[b]	31.1 ± 1.1[c]	24.7	20.0	50.3	15.3
VS/TS (%)	82.3 ± 1.0[b]	91.4 ± 0.3[c]	96.0	78.1	98.3	90.5
Compensated TS (%)		35.8 ± 1.3[c]				18.8
Compensated VS (%)		32.8 ± 1.1[c]				17.2
NH4-N (g/kg)	1.1 ± 0.01[c]	0.3	0.2	0.1	0.6	1.2
TKN (g/kg)	2.3 ± 0.01[c]	7.3	3.8	29.0	13.6	16.5
N (g/kg)	1.6 ± 0.2[c]	8.3	4.0	28.4	18.9	17.2
C (g/kg)	27.9 ± 2.0[c]	170.0	109.5	105.7	359.2	96.7
C/N	17.0 ± 0.4[c]	20.4	27.7	3.7	19.0	5.6
Ptot (g/kg)	0.5 ± 0.1[c]	1.0	0.4	11.6	2.0	2.2
Ktot (g/kg)	2.6 ± 0.3[c]	10.9	4.9	2.4	1.9	2.2
VFAtot (g/L)	4.1 ± 1.8[c]	9.6	0.2	0.2	1.2	7.6
sCOD (g/L)	9.7 ± 2.5[c]	122.0	21.7	74.3	228.6	63.1
Total sugars (g/kg)	9.5	65.3	176.0	2.3	5.8	7.5
Crude fat (g/kg)	3.7	21.1	0.1	18.8	353.1	30.9
Protein (g/kg)	7.9 ± 0.1[c]	44.1	22.6	180.3	81.1	95.8
Sulfur S (g/kg)	0.3 ± 0.1[c]	0.7	0.4	2.0	ND	1.2
C/S	86 ± 10[c]	228	269	52	ND	81

[a] Standard method SFS 3008 analysis results.
[b] Average value of three samplings during the experiment.
[c] Average value of two samplings during the experiment. ND, not determined.

reflected previous results by Ebner et al. (2016) (140 g/kgTS), but e.g., Solli et al. (2014) reported much higher protein content for CS (370 g/kgTS). Differences can be due to the analytical methods, as well as cow diets or manure management practices.

Co-feedstocks had distinctive properties and different shares of organic fractions due to their different origins. Two of the co-feedstocks were plant-based (GS and P), and three were animal by-products (F, FO, and RO). Highest total sugar proportions were analyzed from plant biomasses: P (176 g/kg), followed by GS (65.3 g/kg). The GS characteristics corresponded well with previously reported results by Wall et al. (2014) (TS 29 %, C/N ratio 26, protein concentration 46 g/kg). The potato had a higher TS content of 25.7 % than previously reported by Yan et al. (2021) (18.5 %). While the C/N ratio (26–27) and TKN (3–4 g/kg) concentrations resembled those found in the present study, there were significant differences in protein and fat content. Yan et al. (2021) reported protein and fat concentrations for potato to be 1.5 g/kgTS and 90.3 g/kgTS, while these fractions had concentrations of 88 g/kgTS and 0.4 g/kgTS in the present study.

The studied animal by-products were rich in fat and/or protein. Especially F was rich in protein (180.3 g/kg), while RO had a significant share of both fat, 30.9 g/kg, and proteins, 95.8 g/kg. FO differed from the other co-feedstocks in having an extremely high crude fat concentration, 353.1 g/kg. Animal by-products can have varying characteristics, depending on the origin and processing of the material. In the present study, the TS concentrations for the F and FO samples were 25.6 % and 51.2 %, while e.g., Vivekanand et al. (2018) reported TS of 32 % for fish ensilage. Solli et al. (2014) reported TS of 35.1 %, NH_4-N of 0.4 g/L, a fat concentration of 194 g/kg, and a protein concentration of 142 g/kg for ensiled fish waste consisting mainly of dead salmon from fish farms. These values were in the range of results from F and FO samples in the present study (Table 2). Previous information about reindeer slaughtering wastes was unavailable, but the characteristics of the RO sample (TS 16.9 %, TKN 16.5 g/kg, C/N 5.6) corresponded to results from slaughterhouse waste consisting of mainly lamb and goat stomachs (TS 28.8 %, N 29.1 g/kg, C/N 6.4, (Moukazis et al., 2018)). The same study also reported characteristics for bladders and intestines, but these had lower N content (6.8 g/kg) and a much higher C/N ratio (36) than the present study's RO sample.

3.2. Measured and theoretical methane production

3.2.1. BMPs of individual feedstocks

The cumulative methane yields of the feedstocks were determined in a batch test, using both mono-digestion and co-digestion. Because the BMP test was carried out after the SCSTR experiment, the VS analyses were repeated along with the analysis of the inoculum. These VS results (CS 4.5 %, GS 33.6 %, P 24.4 %, F 20.7 %, FO 63.5 %, RO 17.3 %, and inoculum 2.8 %; compensated VS was used for GS and RO) were used to set up the experiment. Methane potentials were also evaluated theoretically based on the content of organic components, lipids, proteins, total sugars, and VFA. The theoretical approach enables quick estimation of biodegradability of different substrates, compared to the rather long-lasting (1–2 months) experimental procedure. Theoretical methane potential (BMP_{th}) for both under- and overestimated CH_4 production was compared to the yields measured experimentally (Table 3).

The tested co-feedstocks reached high BMPs, from 347 to as high as 971 L/kg VS (Fig. 1a). Plant biomasses (GS and P) had CH_4 productions of around 350 L/kg VS (GS 356 L/kg VS; P 347 L/kg VS), whereas animal-based side streams had even higher CH_4 production potentials: 447 L/kg VS for F; 410 L/kg VS for RO; and as high as 971 L/kg VS for FO. The observed elevated CH_4 potentials support the aim of increasing the CH_4 production of the CS-based process through distinctive co-feedstocks. The obtained BMP of the main feedstock CS (153 L/kg VS) was low compared to the co-feedstocks, but it was also lower than usually reported (230 L/kg VS in Scarlat et al. (2018)). This is related to the low VS/TS ratio (82 %) obtained in the sample, indicating the lower biodegradability of the material. Furthermore, the BMP_{th} of CS (280 L/kg VS) was considerably higher than the measured yield. Both the collection practices and storage time (at the farm and in the laboratory during the SCSTR experiment and before the BMP test) could have affected the characteristics of the CS sample and led to the aerobic degradation of organic matter and the moderate methane production in the BMP tests.

The high experimental BMP obtained with the FO sample (971 L/kg VS) reflected its composition well (Table 2), especially its high organic matter and lipid content. In addition, the production rate of CH_4 was low (Fig. 1a), which apparently originated from a high share of lipids, which are known to be slowly degradable (Vidal et al., 2000; Yoon et al., 2014), but with a sufficient substrate to inoculum VS ratio (0.5), no lag time at the beginning of batch test was observed. The BMP_{th} of the FO was also high (782 L/kg VS) but lower than the experimental value. The

Table 3
Theoretical and experimental CH_4 yields (BMP 55 d) of the individual feedstocks, estimated and experimental CH_4 yields (55 d) of the feedstock mixtures, and the differences between these values.

	Theoretical CH_4 potential[a] (L/kg VS)	Experimental BMP (L/kg VS)	CH_4 yield obtained experimentally in relation to theoretical potential (%)	Estimated CH_4 yield based on the BMPs of the individual feedstocks[b] (L/kg VS)	CH_4 yield obtained experimentally in relation to estimated yield (%)
Cattle slurry (CS)	280	153 ± 11	55		
Grass silage (GS)	226	356 ± 13	157		
Potato (P)	344	347 ± 65	101		
Fish (F)	538	447 ± 35	83		
Fish offal (FO)	782	971 ± 34	124		
Reindeer offal (RO)	536	410 ± 25	77		
CS + GS		207 ± 20		212	97
CS + GS + P		233 ± 9		237	98
CS + GS + F		247 ± 25		256	96
CS + GS + FO		412 ± 29		383	108
CS + GS + RO		248 ± 23		253	98

[a] Based on the organic fraction composition.
[b] Based on the BMPs of the individual feedstocks; weighted sum considering the shares of feedstocks in the mixture.

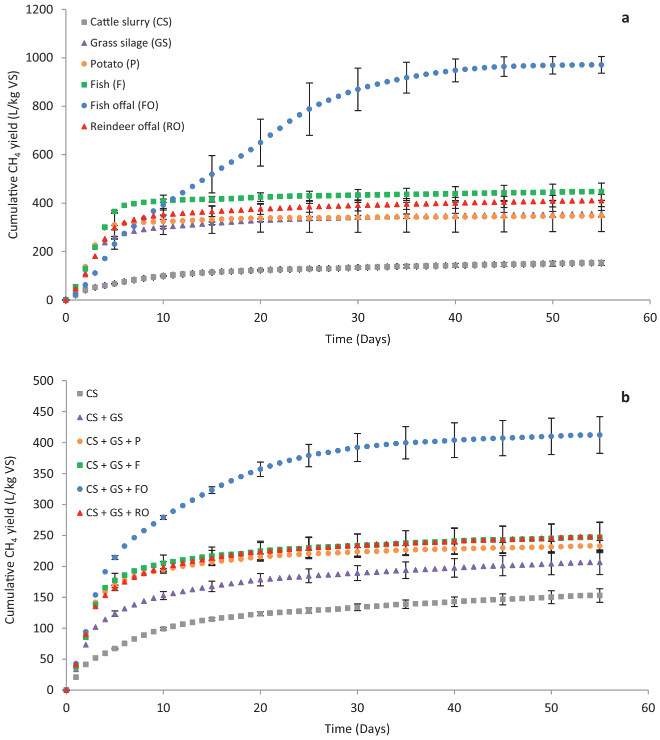

Fig. 1. Cumulative CH_4 yields of mono-digestion of individual feedstocks (a) and co-digestion of cattle slurry (CS) and co-feedstocks (b). Note the different ranges of the y-axes. Standard deviations are plotted in 5-day intervals.

experimental methane production achieved was also very close to the maximum methane production potential of lipids (1014 L/kg VS) which indicates that the FO sample had high biodegradability, or degradation of the inoculum may also have occurred (Yoon et al., 2014).

The measured BMP of GS (356 L/kg VS) was comparable with results from previous studies (337–354 L/kg VS, reviewed in Zhang et al. (2021)) but BMP_{th} (226 L/kg VS) underestimated the CH_4 potential. This may be derived from the calculation method used, which excludes the fiber components, cellulose and hemicellulose, which are shown to be partially degradable in anaerobic digestion (Naroznova et al., 2016). For the other studied co-feedstocks (P, F, and RO), the theoretical approximation of BMP showed equal or slightly higher CH_4 potentials than yields. The calculation method did not account for the biodegradability of the feedstocks, and combining the biodegradability data with the calculations has been shown to specify the estimate of BMP_{th} (Labatut et al., 2011).

Both P and F showed high CH_4 production rates; 90 % of cumulative CH_4 production was achieved in 6 and 8 days respectively. The organic matter of P consisted mainly of sugars, while the share of fat was small, whereas F had a high proportion of proteins. In addition, cumulative CH_4 productions of P (347 L/kg VS) and F (447 L/kg VS) were in line with earlier studies (Allen et al., 2016; Fonseca et al., 2020), even though the inoculum from the farm-scale plant was not adapted to distinctive feedstocks. With fish biomass, fish species and applied processing procedures (cutting/fileting) strongly affect to biomass characteristics, leading to CH_4 potentials ranging from 188 to 691 L/kg VS (Fonseca et al., 2020; Vivekanand et al., 2018).

3.2.2. *Batch co-digestion*

Co-digestion batch tests were executed with feed mixtures corresponding to the SCSTR experiment. The cumulative methane yields in the co-digestion of CS and studied co-feedstocks varied from 207 to 412 L/kg VS. The highest yield of 412 L/kg VS was achieved with the co-digestion of CS, GS, and FO (Fig. 1b). Compared to the methane production potential of CS alone, the co-digestion increased the methane yields per VS by 35 %, 52 %, 61 %, 170 %, and 62 % using co-feedstocks, GS, GS + P, GS + F, GS + RO and GS + FO respectively. Compared to the co-digestion of CS and GS, the additional feedstock increased the VS-based methane yields by 13 %, 19 %, 100 %, and 20 % with P, F, FO, and RO respectively.

The possible synergistic effects of co-digestion in the batch test were evaluated by calculating estimated CH_4 yields. The estimate was based on the CH_4 yields of individual feedstocks in mono-digestion, according to Vivekanand et al. (2018). In co-digestion BMP tests, CH_4 production was 1.9–3.6 % lower in most of the feedstock mixtures compared to the estimated production that was calculated based on the BMPs of individual feedstocks. Only in the co-digestion mixture containing fish offal (FO), the experimental CH_4 production was 7.8 % higher than the estimated production. Overall, the difference between measured co-digestion BMP and the estimated one (−4–8 %) was similar, which has previously been reported for different feedstock mixes (−5–20 %) in a study by Ebner et al. (2016).

The co-digestion of different feedstocks is often justified by the more balanced supply of nutrients and trace element, and a more favorable C/N ratio and dilution of possibly inhibiting substances (ammonia, long-chain fatty acids), but antagonistic effects can also be observed (Ebner et al., 2016; Vivekanand et al., 2018). Negative effects can occur, e.g., due to disturbances and/or inhibition in the batch digestion. In the present study, the experimental co-digestion BMP was at most 3.6 % lower than the estimated co-digestion, which does not yet indicate antagonistic effects. Another explanation for the lower experimental BMPs of co-digestions is the low CH_4 production capacity of the CS sample, which probably affected all the co-digestion tests. As a method for studying synergistic effects in co-digestion, BMP has also been criticized because of the high proportion of inoculum it contains. Synergistic effects may appear inter alia due to the better nutrient and trace element balance (Nielfa et al., 2015), but a healthy inoculum effectively prevents nutrient and trace element depletion (Koch et al., 2020).

3.3. *Reactor experiments*

A semi-continuous laboratory experiment was run for 428 days, during which two control periods and four co-feedstock periods were executed. The digestion process was monitored for biogas and CH_4 production and for possible inhibition or disturbance during the rapid changes of the co-feedstocks. The monitored process parameters consisted of specific CH_4 and biogas production, volumetric CH_4 and biogas production, pH, NH_4-N content, and CH_4 and H_2S concentration in the gas (summarized in Figs. 2 and 3 and Table 4).

3.3.1. *CH_4 production*

Both reactors showed stable performance over the co-feedstock changes without disturbances either in CH_4 or biogas production (Fig. 2). Volumetric CH_4 production began to increase after the P1 control period, where it was 301 L CH_4/m^3d, and the highest volumetric CH_4 production was observed in P5, 844 L CH_4/m^3d. All co-feedstock periods, including P2 with only GS as co-feedstock, had higher volumetric CH_4 productions than the P1 control. Like volumetric productivities, the highest SMP was obtained in P5 (305 L CH_4/kg VS). Otherwise, the SMP remained rather steady over the experiment. The difference between average SMP during P1 (with CS as the only feedstock, 201 L CH_4/kg VS) and P2 (GS as co-feedstock, 204 L CH_4/kg VS) was not statistically significant (see supplementary material). Adding P (P3) or RO (P6) to the co-feedstock mixture with GS did not result in statistically significant changes in SMP either, with average CH_4 productions of 216 L CH_4/kg VS and 195 L CH_4/kg VS respectively. On the other hand, co-feedstocks fish (F) during P4 and fish offal (FO) during P5 resulted in statistically significantly higher SMPs (adj. $p < 0.05$), 220 L CH_4/kg VS and 305 L CH_4/kg VS respectively (Table 4). Despite the statistical insignificance of the changes in SMPs, the use of all the tested co-feedstocks (GS, P, F, FO, and RO) improved the total CH_4 production and was thus positive from the farm's energy production perspective. Specific biogas production (L biogas/kg VS) had nearly similar trend line to SMP, and periods P3–P5 showed statistically significant increase in specific biogas production compared to P1. Volumetric biogas production ranged from 500 L biogas/kg VS in control (P1) to over 1400 L biogas/kg VS in P5 (Fig. 2).

The experiment was started with control period P1, where CH_4 production was moderate. Adding GS to the feedstock mixture (P2) from day 57 onwards increased the OLR from 1.6 to 2.2 kg VS/m^3d and enhanced the volumetric CH_4 production from 301 to 447 L CH_4/m^3d, while SMP remained around 200 L CH_4/kg VS (Fig. 2). The SMP in P1 (201 L/kg VS) exceeded the BMP of the CS (153 L/kg VS), which was probably due to a deficient BMP rather than exceptionally high productivity in the SCSTR experiment. As stated earlier, the BMP test was conducted after the SCSTR experiment, and the degradability of CS may have decreased by the time of the batch test. It is known that BMP tests consistently overestimate the CH_4 production compared to SCSTR operation (Holliger et al., 2017). In P2, the SMP was 99 % from the BMP of the CS + GS mixture, which is a high recovery rate for biomethane potential in SCSTR. This may partly be due to the high productivity of recently collected CS, as well as an adapted microbial community and the constant properties of GS throughout the studies, as this material was stored in a frozen state.

Supplementing the CS + GS feedstock mixture with additional feedstocks in P3–P6 increased the OLR to 2.8 kg VS/m^3d. The first additional co-feedstock was P during test period P3, where volumetric CH_4 production increased by up to 585 L CH_4/m^3d, whereas the SMP increased slightly to 216 L/kg VS, but not statistically significantly (Fig. 2, Table 4). Similar type of results was found by González et al. (2021) where potato peels were co-digested with sheep manure, increasing SMP from 196 L/kg VS to 214 L/kg VS. The degradation of carbohydrate-rich P was shown to be a rapidly degrading substrate in the BMP test (Fig. 1), and it can therefore be concluded that the majority

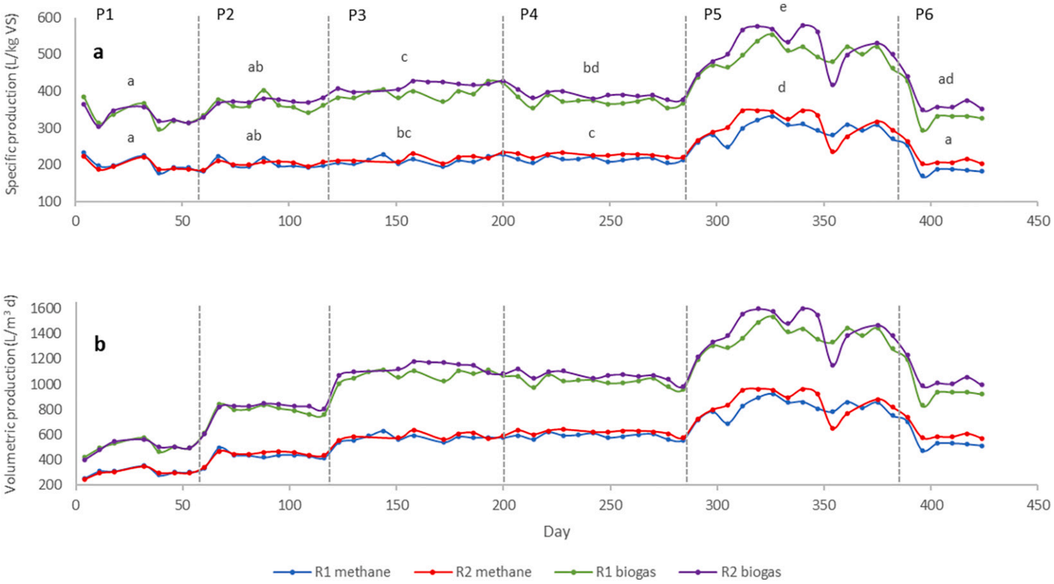

Fig. 2. Specific methane and biogas production (a) and volumetric methane and biogas production (b) for both reactors during test periods 1–6 (P1–P6). Different letters (a–e) in figure a indicate statistically significant differences ($p < 0.05$) between the period averages.

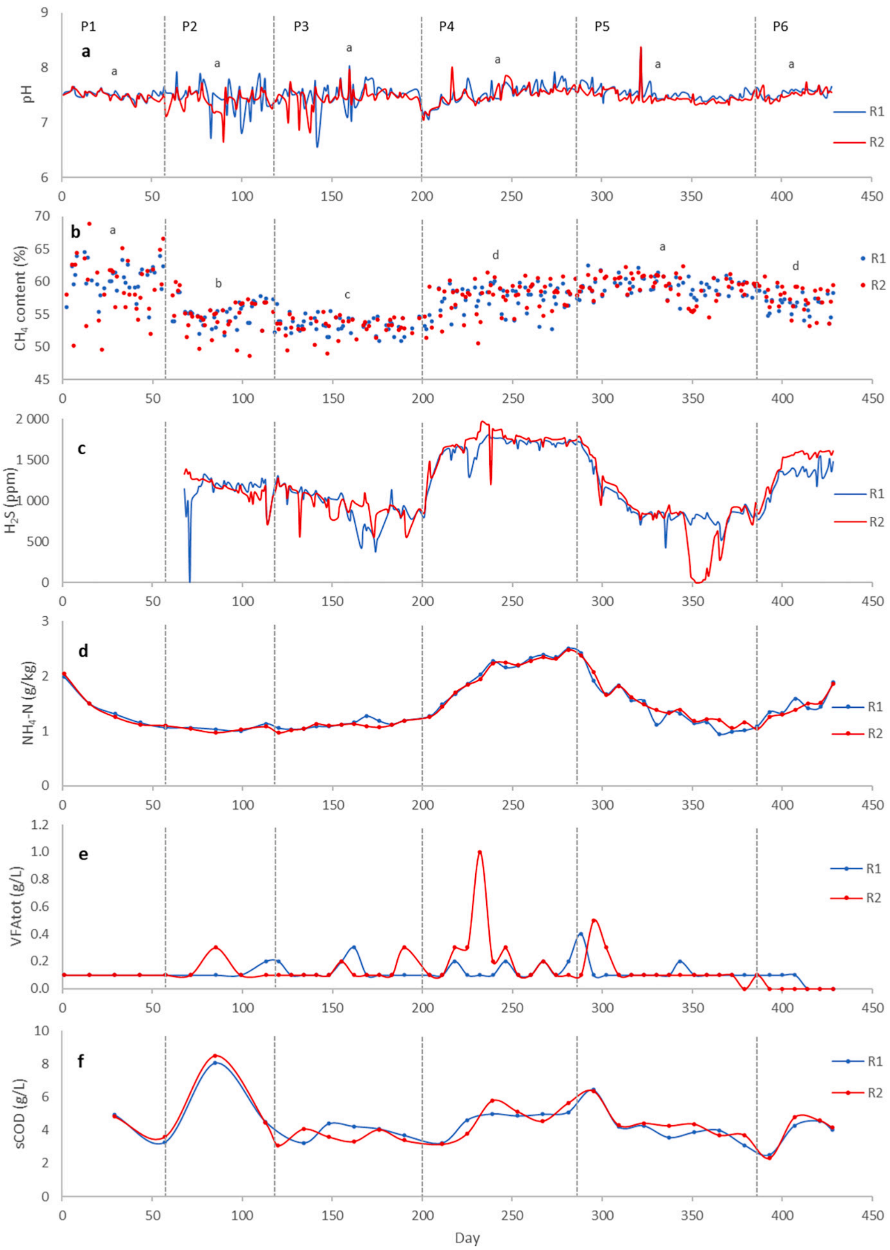

Fig. 3. pH (a), CH_4 (b), H_2S (c), NH_4-N (d), VFAtot (e), and sCOD (f) values during periods 1–6 in parallel reactors R1 and R2. Different letters (a–d) in figures a and b indicate statistically significant differences ($p < 0.05$) between the period averages.

of its CH_4 production potential was achieved, despite the rather short HRT of 23 days. Plant-based biomass such as potato was shown to be practical co-feedstock for a farm-scale biogas plant, as it increased the volumetric CH_4 production and further enhanced energy production.

In P4, the shift of additional co-feedstock type to F showed enhancement in SMP (220 L/kg VS). The reactors also showed robustness in operating efficiently with co-feedstock with distinctive chemical properties, because CH_4 production showed no decrease. The beginning of the P4 test period especially demonstrated that the CS-based digestion process could be flexible, because the CH_4 production stabilized quickly to a new higher level after co-feedstock change.

Although P5 showed the highest SMP (305 L CH_4/kg VS), the feedstock mixture reached only 74 % of the corresponding BMP value (412 L CH_4/kg VS). Both the sole FO and the CS + GS + FO mixture had the highest BMPs among the tested feedstocks and feedstock mixtures, but the FO's BMP was achieved slowly (Figs. 1, 2). These may result from the slow degradation of lipid-rich material. Nevertheless, the capacity for increasing CH_4 and energy production of farm-scale operation was obvious and in line with previous studies (Fjørtoft et al., 2014).

In the last feeding period, P6, SMP was decreased and stabilized to the initial level with P1–P2, and volumetric CH_4 decreased to 551 L CH_4/m^3d. Based on the BMP results, RO could be seen as a promising co-feedstock with respect to CH_4 production, because the BMP of P6 feedstock mixture was equal to the P4 feedstock mixture, for example. Despite the high production in the batch test, only 79 % of the CH_4 potential was achieved in the SCSTR experiments. The degradation rate observed in the batch test was at a corresponding level to the P4 feedstock mixture and did not explain the modest CH_4 production observed in SMP. In both P5 and P6, the possibility of challenges to the microbiota to acclimatize to a very different type of co-feedstock cannot be ruled out. The SMP in P6 (195 L/kg VS) was also lower than has previously been reported for co-digestions with manure and slaughterhouse waste e.g., 260–270 L/kg VS in Luste et al. (2012).

Both the co-feedstock type and its proportion in the feedstock

Table 4
The average values and 95 % confidence intervals for methane production (L CH_4/kg VS) and pH of the parallel SCSTR reactors in periods P1–P6. Feedstocks with average values that have no common superscript letters a-d differ in a statistically significant way at the $p < 0.05$ after controlling for family-wise error rate.

Variable	Period	Feedstock	Average	SD	95 % C.I.
CH_4 (m^3/kg VS)	P1	CS	201^{ab}	17	(192, 210)
	P2	CS + GS	204^{ab}	9	(200, 208)
	P3	CS + GS + P	216^{bc}	11	(211, 220)
	P4	CS + GS + F	220^{c}	8	(217, 223)
	P5	CS + GS + FO	305^{d}	30	(293, 316)
	P6	CS + GS + RO	195^{a}	14	(187, 203)
pH	P1	CS	7.51^{a}	0.07	(7.49, 7.52)
	P2	CS + GS	7.44^{a}	0.22	(7.39, 7.49)
	P3	CS + GS + P	7.49^{a}	0.20	(7.45, 7.53)
	P4	CS + GS + F	7.52^{a}	0.17	(7.49, 7.56)
	P5	CS + GS + FO	7.50^{a}	0.15	(7.48, 7.53)
	P6	CS + GS + RO	7.54^{a}	0.08	(7.52, 7.56)

mixture affect the reactor performance. In the present study, the shares of co-feedstocks were moderate, because the experiment simulated farm-scale operation and the limited local availability of the side streams, which reflected Finnish cattle farm sizes and the remote conditions in Finnish Lapland. Previously, higher co-feedstock proportions of up to 30–40 % of the feedstock mixture's VS, have been reported to be used with cattle manure to increase CH_4 production without process disturbances such as VFA accumulation (Fjørtoft et al., 2014; Lehtomäki et al., 2007). On the other hand, moderate co-feedstock proportions have also been shown to disrupt the AD process. For example Callaghan et al. (1998), conducted their study with cattle slurry and rainbow trout offal, where a fish offal proportion of 6 % (w/w) disrupted the biogas process performance. However, the OLR was significantly higher (5–6.5 kg VS/m^3d) than in the present study.

3.3.2. *Process stability*

Process stability and the possible accumulation of process intermediates or inhibiting components were monitored by measuring pH and analyzing the concentrations of VFA, sCOD, and NH_4-N (Fig. 3). Despite the rapid co-feedstock changes, no signs of process disturbances were detected. Furthermore, the CH_4 content in biogas (%) did not indicate process disturbances. CH_4 contents had some variation over the experiment (Fig. 3b) but fluctuations occurred most likely due the changing characteristics of the co-feedstocks.

In general, no critical pH drops were observed, and there were no signs of acidification. The variation in pH was largest in P2 and P3, where GS and GS + P were used as co-feedstocks (Fig. 3a). However, there were no statistically significant differences between the periods after controlling for the family-wise error rate. For example, in P2, the pH level ranged from 6.72 to 7.93 in R1 and from 6.65 to 7.72 in R2. The varying pH was probably due to the rapid formation of organic acids in both P and GS samples during their storage. Shifting from P3 to P4 induced a temporary decrease of reactors' pH levels. The pH dropped to 7.06 and 7.05 in R1 and R2 respectively but stabilized to the optimal level within one HRT. If the digestate has a high buffering capacity, changes in pH are not easy to detect even if process intermediates such as VFAs are accumulating, as was shown in the study by Liu et al. (2017), where pH values remained stable during the introduction of a different substrate mix.

Total VFA concentrations remained at a moderate level, 0–1 g/L, throughout the experiment in both reactors. Only three measured values exceeded 0.3 g/L (Fig. 3e). High VFA levels could indicate possible process disturbances, which occurred due to the accumulation of the process intermediates. In the previous studies, much stronger VFA accumulation has occurred during the introduction of new feedstock types, resulting in VFA concentrations of up to 1.6–4.6 g/L (Liu et al., 2018). In the present study, a relatively low share (19 % on VS basis) of the total feedstock mixture was changed between co-feedstock periods, which probably explain the process's tolerability and adaptation capability. Concentrations of sCOD remained at a moderate level, exceeding 6.5 g/kg at only one analysis point during P2, as in both reactors R1 and R2 (Fig. 3f). VFA and sCOD analyses supported the conclusions from CH_4 productions that the process was stable throughout the experiment despite the co-feedstock changes.

The biogas process may also be inhibited by high levels of free ammonia originating in an increased NH_4-N concentration and a high pH level, but no signs of NH_4-N-induced inhibition were detected. At the beginning of the experiment, NH_4-N levels were roughly 2 g/kg but dropped to approximately 1 g/kg during P1 (Fig. 3d). The concentration was stable for P2 and P3 but rose steadily to 2.5 g/kg during P4 with F as co-feedstock. The trend was reversed in the fifth period with FO; NH_4-N decreased to close to 1 g/kg by the end of the period. An increasing trend can also be observed for the final period with RO (P6). However, the trendlines did not flatten out, so it is possible that for longer runs with FO or RO, even higher NH_4-N concentrations would have been reached. Previously, the biogas process has been reported to tolerate relatively high levels (3.2–4.4 g NH_4-N/L) of ammonium-nitrogen without process disturbances (Ahlberg-Eliasson et al., 2017).

H_2S concentrations in the produced biogas were around 1200 ppm in period 2 and decreased to around 1000 ppm when shifting to P3 (Fig. 3c). In P4, with fish (F) as a co-feedstock, H_2S concentrations again rose and stabilized at a higher level, 1700–1750 ppm. Over the P5, H_2S concentrations declined and stabilized to around 800 ppm in R1, and in R2, H_2S levels were even observed to drop to close to 0 ppm. In the last period (P6), H_2S concentrations again increased to an average of 1400 ppm in R1 and 1600 ppm in R2. The H_2S concentration measurement began on day 68, during control period 2, and no data are available for P1. H_2S concentrations in the produced biogas reflected the changes in co-feedstocks characteristics, i.e., concentrations of S. According to Peu et al. (2012), feedstocks with a carbon/sulfur (C/S) ratio under 40 have a risk of high levels of H_2S in the biogas. In the present study, all the tested feedstocks had C/S ratios above 50 (Table 2; FO was not analyzed). In addition, the periods in which feedstocks had the lowest C/S ratios were those with the highest H_2S concentrations in the gas: P4 with F as a co-feedstock and P6 with RO as a co-feedstock. In anaerobic digesters, low H_2S concentrations are preferred to prevent corrosion of biogas engines and the cost of H_2S removal.

3.4. *Digestate quality and usability*

3.4.1. *Residual methane potential*

The RMP of the digestates from the SCSTRs were determined at the end of each feedstock period, except after P1. RMP testing enables the measurement of digestate biodegradability, i.e., how much methane potential remains in the digestate after the reactor. The RMP of the digestates after each feedstock period varied from 71 to 98 L CH_4/kg VS (Table 5 and supplementary material), indicating that the materials were not fully degraded during the SCSTR runs. Digestates from P5 (CS + GS + FO) had the highest RMP, 91.1 L CH_4/kg VS for R1 digestate and 97.8 L CH_4/kg VS for R2 digestate. Overall, the RMP was higher after co-feedstock periods P3–P5, reflecting the higher CH_4 productions in the SCSTR experiments (Table 5). After P2 (CS + GS control) and P6 (CS + GS + RO), the RMP was lowest, 72.7–75.5 L CH_4/kg VS and 71.4–73.3 L CH_4/kg VS respectively. Previously, the addition of co-feedstock to the manure-grass-based process has been found to increase RMP from 71 to 134 L CH_4/kg VS (Liu et al., 2018) and to follow the methane production trend in the continuous reactor. Varying RMP values have been reported for different manure-based digestates, depending on the initial feedstocks and operating conditions (HRT, temperature), e.g., 15–103 L CH_4/kg VS (Rico et al., 2011), 40–98 L CH_4/kg VS (Ahlberg-Eliasson et al., 2021), and 1–197 L CH_4/kg VS (Lehtomäki et al., 2007).

The RMP is an essential parameter for assessing the degradability of the feedstocks and to estimate the risks of greenhouse gas and ammonia

Table 5
Characteristics of digestates at the end of the feedstock period presented as averages of samples from R1 and R2. The feedstocks are cattle slurry (CS), grass silage (GS), potato (P), fish (F), fish offal (FO), and reindeer offal (RO).

Period	1	2	3	4	5	6
Feed	CS	CS + GS	CS + GS + P	CS + GS + F	CS + GS + FO	CS + GS + RO
TS (%)	3.8	4.3	4.9	5.2	4.9	5.0
TKN (g/kg FM)	2.2	2.4	2.7	3.8	3.0	3.8
NH4-N (g/kg FM)	1.1	1.1	1.2	2.5	1.1	1.9
NH4-N/TKN (%)	49.0	46.0	44.6	65.9	35.6	49.9
C/TKN	14.8	13.1	12.6	9.2	13.3	10.7
TKN/Ptot	6.1	5.1	5.0	3.2	4.9	5.7
Ptot (g/kg FM)	0.4	0.5	0.5	1.2	0.6	0.7
$P_{soluble}$ (mg/L)	6.4	5.8	11.5	16.4	15.2	14.9
Ktot (g/kg FM)	2.5	2.9	2.9	3.6	3.1	3.2
Ca (g/kg FM)	0.7	0.8	0.9	1.9	1.1	1.1
Cu (g/kg FM)	3.0	2.7	2.9	3.3	3.1	3.3
Fe (g/kg FM)	62.0	72.1	99.4	112.0	109.2	96.6
Mg (g/kg FM)	0.4	0.5	0.6	0.6	0.6	0.6
Mn (g/kg FM)	10.6	12.7	14.9	17.0	15.0	23.9
Na (g/kg FM)	0.3	0.3	0.3	0.4	0.3	0.4
Zn (g/kg FM)	13.2	13.8	15.5	19.9	20.6	16.9
RMP (L/kg VS)	ND	74.1 ± 6.1	85.5 ± 8.7	82.3 ± 8.4	94.5 ± 8.7	72.4 ± 6.3

ND, not determined.

emissions during the digestate storage. The RMP of digestates is highly dependent on the organic matter degradation within the initial biogas reactor. Reduced methane production during the reactor phase is reflected in the RMP result (Ahlberg-Eliasson et al., 2021). In the present study, no process disturbances were detected during SCSTR experiments, and the cumulative RMPs were quite similar with all digestates. This was due to the similar HRT during all periods. The reactor's decreased HRT is known to increase RMP (Rico et al., 2011), as more organic matter is left undegraded. In the present study, the HRT in the reactors was 23 days throughout experiment. This is a fairly low HRT, at least if compared to commonly used HRTs in farm-scale anaerobic digesters (on average, 30 d in Swedish farm-scale plants (Ahlberg-Eliasson et al., 2017). The decrease in RMP value and the risks for subsequent emissions in the storage phase could be minimized by optimizing the HRT.

3.4.2. Agronomic quality of the digestates

The quality and agricultural usability of the digestate was determined after each feeding period. The usability of digestates in agriculture as fertilizers depends on the nutrient content and ratios. In addition, TS affects the use of different spreading equipment, for example. In this study, all digestates were well-suited for slurry spreaders, as the TS of the digestates varied from 3.8 % to 5.2 %, depending on the co-feedstocks used (Table 5), where the increase in TS was due to the higher TS of the feedstock mixture. Using fish (F) as a co-feedstock resulted in a digestate with the highest agronomic quality, with concentrations of 3.8 gTKN/kg, 2.5 gNH_4-N/kg, 1.2 gPtot/kg, and 3.6 gKtot/kg. The share of NH_4-N of the TKN was highest with the co-feedstock F (66 %), which indicates higher usability of the digestate nitrogen for plants. Overall, the nutrient and trace element (Ca, Cu, Fe, Mg, Mn, Na, Zn) content of digestates from periods P2–P6 was increased by the introduction of co-feedstocks, which had a higher nutrient content of the feedstock than the cow slurry. The C/TKN ratio was therefore decreased (from 14.8 to 9.2–13.3) after the introduction of co-feedstocks as the TKN content increased, and the TKN/Ptot ratio decreased due to the Ptot in co-feedstocks compared to the CS feedstock alone. However, $P_{soluble}$ concentrations in digestates increased from 6.4 and 6.8 mg/L in the CS and CS + GS digestate, and to 11.5, 14.9, 15.2, and 16.4 mg/L in co-feedstock mixtures with P, RO, FO, and F respectively.

The value of the digestate as a fertilizer in agriculture is determined by its nutrient content and the ratios of nutrients (e.g., TKN/Ptot ratio), while for soil amendment, the C/TKN ratio is more important. While the concentration of nutrients was increased by the addition of different co-feedstocks to the digestion process, the ratio between TKN and Ptot was decreased compared to the CS digestate. This can be attributed to the higher Ptot content within the co-feedstock. The higher the TKN/Ptot ratio in a digestate, the less additional nitrogen fertilization is needed, although the overall levels are highly dependent on the TKN and Ptot requirements of the fertilized crop. However, with the increased amount of TKN and Ptot in the digestate in the co-feedstock scenarios, the resulting improvement in the nutrient balance and nutrient self-sufficiency on the farm would reduce the need for mineral fertilizers (Tampio et al., 2019). The increased input of trace elements (e.g., Mg, Cu, Fe, and Zn) and different co-feedstocks would not only be beneficial for farming but also for the digester operation by balancing the trace element supply (Molaey et al., 2018).

3.5. Co-feedstocks in farm-scale biogas plants

For a farm-scale biogas plant, cattle slurry is often the main feedstock due to its vast supply. Grass silage is also used as a co-feedstock on a regular or semi-regular basis. However, on its own, slurry does not provide significant methane yields (Tufaner and Avşar, 2016), and to increase the energy potential of farm-scale plants and fulfil the energy requirements of the farm, for example, co-feedstocks may be needed. Animal manure provides a good basis for co-digestion, because it provides a buffer capacity (Acosta et al., 2021) and usually does not contain inhibitory elements (Ebner et al., 2016), thus acting as a diluter for possible inhibitors within the co-feedstocks. However, biogas production in a decentralized farm context struggles with the economic feasibility arising from the poor energy value of manure, reliability, durability, and the high costs of digestion (Wang, 2014). In addition, at least in Finland, manure digestion is not subsidized to the same degree, and corresponding gate fees for manure as those in industrial scale digestion of waste biomasses, for example, are unavailable (Winquist et al., 2019). The use of locally available co-feedstocks, e.g., from food processing and slaughterhouses, could not only improve the energy production potential but also the farm's nutrient balance, as the present study demonstrates. In addition, receiving food industry waste and side streams could improve profitability through gate fees (Chodkowska-Miszczuk et al., 2021). Farm-scale biogas plants do not always operate with full reactor capacity, which would ease the co-feedstock use, because it would not necessarily affect the reactor's loading rate and retention time. The present study showed that it was possible with co-feedstocks to increase the volumetric CH_4 production by 23–89 % (compared to P2 with CS + GS), which enables more efficient use of the available digester volume on farms.

The seasonality and variation of feedstock composition has been raised as the main hurdles for the successful and feasible production of biogas (Wang, 2014). The present study indicates that the change of co-feedstocks had no negative effects on the process stability and methane production if the share of the co-feedstocks was kept low. Co-feedstock use could also be matched with needs of biogas (Mauky et al., 2017). For example, with its higher heat demand, the winter could be an ideal time for utilizing the most energy-producing feedstocks such as fish offal biomass. However, the feedstock's seasonality does not necessarily match the energy requirements. For example, plant biomasses are mainly harvested during the summer and fall, and biomass ensiling and storage options are therefore also required. The use of co-feedstock can therefore require investments from the biogas plant for e.g., storage and feeding systems. The legislation related to the treatment of co-feedstocks should also be considered when using animal by-products such as offal. According to the EU legislation (EU/1069/2009), these materials require hygienization, and for hygienic reasons, these materials should be carefully handled within the farm environment. Naturally, due to e.g., hygienization requirements, the energy demand increases, which should be considered when evaluating the total profitability of the processing. However, e.g., Luste et al. (2012) showed that it was possible to gain positive net energy production when hygienizing animal by-products and using them as co-feedstocks.

4. Conclusions

For a farm-scale digester, the utilization of regionally available, and thus seasonal, co-feedstock in low proportions (19 % of the total VS load) increased CH_4 and energy production compared to sole cattle slurry feeding. Despite recommendations that the biogas process to new feedstocks be acclimated with caution, no disturbances in the biogas process were observed, even with sudden changes in co-feedstocks. This indicates the potential of using a small share of seasonal co-feedstocks with the added value of an increased digestate nutrient content, which would support farms in self-sufficient fertilization. Furthermore, local utilization promotes more sustainable treatment of side streams.

CRediT authorship contribution statement

Satu Ervasti: Conceptualization, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing. **Joel Kostensalo:** Formal analysis, Methodology, Validation, Visualization, Writing – original draft, Writing – review & editing. **Elina Tampio:** Conceptualization, Formal analysis, Supervision, Validation, Writing – original draft, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.biteb.2022.101207.

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