

# **Temperature effect on performance and methane emissions of highly controlled replicate septic tanks**

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## Abstract

Septic tanks are widely used for decentralized wastewater treatment but remain poorly characterized with respect to greenhouse-gas emissions, particularly under variable temperature regimes. Understanding how temperature influences treatment performance and methane production is essential for improving both emission inventories and environmental sustainability through tailored mitigation strategies. This study used a unique set of twelve full-scale, replicate septic tanks fed with real domestic wastewater to isolate the effect of four controlled temperature conditions (ambient, insulated, 20 °C, 30 °C). Continuous monitoring of headspace gas, dissolved methane and water quality enabled a complete carbon balance across gaseous, liquid, and sludge pathways. Higher temperatures enhanced organic degradation and established stronger anaerobic conditions, increasing methane production from ~42% under insulated conditions to ~54% at 30 °C. Critically, temperature governed methane partitioning: at 30 °C, desorption was favored and ~70% of the methane accumulated in the headspace, whereas at lower temperatures ( $\leq 20$  °C), a greater fraction (often >80% of methane) remained dissolved and was discharged with the effluent. This behavior reveals that dissolved methane can represent a substantial fraction of total methane generated in septic tanks, yet it is typically not quantified separately in standard emission assessments. By jointly accounting for gaseous and dissolved pathways, our results show that total methane release from septic tanks may be higher than estimates based solely on headspace measurements. These findings highlight the importance of explicitly considering dissolved-phase dynamics when refining emission factors and developing mitigation strategies.

## Keywords

Decentralized sanitation; septic tanks; temperature effect; methane partitioning.

## 1. INTRODUCTION

Septic tanks have long been a cornerstone of basic, decentralized wastewater treatment systems, particularly in areas where centralized treatment systems are impractical or unavailable. They are low-cost and are used to manage domestic wastewater for millions of households worldwide. A standard septic tank is a very simple technology, consisting of a tank made of one or more chambers. Wastewater separates into three layers: solids settle at the bottom, clarified water remains in the middle, and floating materials, including fats, form a foam layer at the top. Partial microbial degradation of pollutants in the wastewater and sludge occurs through anaerobic digestion (AD). The AD generates more microbial cells (biomass) and gases, such as methane and carbon dioxide. Septic tank effluent is usually discharged using a network of pipelines into a drainage field, where it undergoes additional treatment through filtration, gas desorption, and microbial degradation in the soil. Despite their prevalence, septic tanks are poorly monitored and regulated and often fail to meet discharge standards (Ahmed et al., 2005; Ravi and Johnson, 2021; Soewondo et al., 2025; Withers et al., 2012), which leads to pollutant hotspots in the surrounding environment (Richards et al., 2016; Scott and Parsons, 2005). Furthermore, it has been suggested that the emissions of methane from septic tanks could constitute a significant portion of global emissions of greenhouse gas from wastewater treatment (Cheng et al., 2022; Manga and Muoghalu, 2024), although direct measurements of septic tank emissions are rare. The juxtaposition of needing affordable decentralised sanitation with the potential for septic tanks to pollute local environments and the global atmosphere makes experimentally quantifying the treatment outcomes and methane emissions an imperative.

The former of these, treatment outcomes, has been addressed, in a large part, by a long but sparse literature on research into septic tanks' performance and their environmental implications, dating back to the 1970s (DeWalle and Schaff, 1980; Lawrence, 1973; Yates,

1985). This has led to new designs, with enhanced treatment, to meet increasingly strict regulatory standards on discharges (Koottatep et al., 2025, 2020; Saeed et al., 2024; Singh et al., 2019; Sorenson et al., 2023). Notwithstanding these innovations, it is widely accepted that failure to regularly remove the sludge from tanks is a major cause of downstream septic tank pollution (Tan et al., 2021; Wardhani et al., 2024). One way of mitigating this is to enhance the biodegradation within the tank using AD processes to reduce the rate of sludge accumulation and hence the frequency with which the tank needs to be emptied and thus reduce homeowners' maintenance costs (Mahon et al., 2022; Pussayanavin et al., 2015). Typically, microbial activity correlates positively with temperature (Viessman Jr. and Hammer, 1999) and so methods of increasing the temperature of the tank have been deployed to increase sludge degradation. Polprasert et al. (2018) demonstrated the efficacy of this approach, using solar thermal energy to increase the tank temperature, with Connelly et al. (2019) correlating the increased degradation with increased abundance in key organisms, such as hydrolysers. However, inevitably, by increasing the AD of organic material the rate of methane production is increased. And, given the potency of methane as a greenhouse gas, this improvement of treatment at the expense of increased methane emissions presents a dichotomy.

In recent years, there have been some efforts to estimate the extent of methane emissions from decentralized sanitation using models. So, for example, in USA, where approximately 25% of the population rely on on-site systems, the methane emissions produced have been estimated to 44% of the total emissions from domestic wastewater treatment systems (EPA, 2024). A similar proportion of the population of continental Europe rely on septic tanks. In Scotland, where more than 200,000 septic tanks, serve approximately 10% of the population, 90% of whom are located in rural areas (Lawson et al., 2024), it has been shown that the carbon footprint of conventional septic tanks can be 7 times higher per capita than for large urban wastewater treatment plants (Gupta et al., 2024). However, the models used in these studies

are based on very sparse empirical data and, as a result, the predictions are subject to significant uncertainties.

It is imperative, therefore, that we measure emissions (Poudel et al., 2023). Historically, emissions from on-site technologies such as septic tanks have been neglected, and there is a shortage of direct field measurements (Burchart-Korol and Zawartka, 2019). Diaz-Valbuena et al. (2011) conducted a study in real septic tanks to assess methane emission rates, considering both direct emissions (measured using a gas flux chamber and directly from the vent pipes) and dissolved methane. Whilst their measurements from dissolved methane accounted for only a small fraction (up to 11%) of the total in water temperatures from 12 to 27 °C, it has been shown that methane can be supersaturated at low temperatures, increasing the environmental impact of technologies performing anaerobic digestion (Gómez-Borraz et al., 2022). In STs where biogas is not recovered, the emission becomes the total amount of methane produced, which includes both the gas measured in the gas phase and the fraction that remains in the liquid, as dissolved methane. Because methane solubility in water increases at lower temperatures, dissolved methane poses a problem when septic tanks operate under low temperatures (<15°C), which is common in much of the global north (Mahon et al., 2022). One of the difficulties in drawing conclusions on the factors affecting emissions in real septic tanks is that each one is different and subject to the vagaries of, amongst other things, local weather and influent composition (Connelly et al., 2019). Field studies of methane emissions have been complemented by laboratory studies, which do provide valuable insights, but fail to capture the complexity of real systems (Dubois et al., 2022; Shaw and Dorea, 2021). The ideal, therefore, is a system of closely monitored, field-based, replicate septic tanks that retain the complexity of real influent but where elements of the operating conditions can be controlled.

The aim of this study was to understand the trade-offs between enhanced treatment and methane emissions using a unique system of twelve identical real-scale septic tanks fed from

the same waste stream. The tanks were operated as groups of three replicates under four different temperature regimes, with the intention of varying the biological degradation rates, and were instrumented to accurately and continuously measure emissions along with a range of physical and chemical characteristics. The use of triplicate tanks and a common waste stream give confidence that the conclusions we draw on treatment outcomes and emissions are a result of controlling temperature and not an artifact of tank-specific complex biochemistry.

## 2. MATERIALS AND METHODS

### 2.1 System set-up and operation

The study was conducted in Gauldry, a small rural community in Fife, Scotland, with approximately 600 people. It has a temperate marine climate and is located at sea level. The average yearly temperature is 10 °C, with average-daily temperatures ranging from 14 to 19 °C during summer (June to August) and typically 5 to 10 °C during winter (November to April). There, a unique system consisting of 12, 1-m<sup>3</sup> septic tanks (STs) was constructed and operated for eight months, from April to November 2023 (Figure 1). The effective volume of each ST was 900 L (0.75 m height x 1 m wide x 1.2 m long), and it consisted of a one-chamber reactor with a water seal for the inlet and outlet to prevent the gases from escaping the STs' headspace. Each ST included four sample ports on the side for water and sludge sampling at different height levels (0.5 m, 0.25 m, 0.5 m, and 0.70 m from the bottom of the ST). The STs were inoculated with 80-90 L of primary sludge from the local wastewater treatment plant.

The STs were fed with real domestic wastewater collected after a coarse screen and a grit channel. It consisted primarily of domestic raw sewage augmented by some rainwater runoff, as there is no industrial activity in the village. A macerating pump fed the raw wastewater to a manifold, which distributed the influent into 12 independent 100-L conical tanks. Using

electrically controlled valves, 100 L of wastewater were gradually released into each ST, twice a day, simulating a 4.5-day hydraulic retention time (HRT). Each 'feeding period' lasted approximately 50 minutes by doing 24 cycles as follows: 10 seconds on (valve open)/120 seconds off (valve closed), with an approximate inlet water flow of 0.42 L/s.

Three conditions were tested using real triplicates, including a set of insulated STs (INS) and two heated sets using thermal jackets (Total Thermal Services, UK) to hold the water temperature close to 20°C (20C) and 30°C (30C). A triplicate for the control (CON) was operated under ambient conditions. The heated tanks were regulated by a 0.5-m thermocouple attached to the top of each ST, measuring the water temperature at a height of 0.5 m.



**Figure 1.** Septic tank system location in Scotland (Gauldry) and experimental set-up.

## 2.2 System monitoring, sampling and analytical methods

Every ST unit featured real-time monitoring of pH, water temperature, and headspace methane concentration. The monitoring system was 5G-enabled, allowing valves and sensors to be controlled remotely from a computer, and ensuring that all data were continuously uploaded to

the cloud. A control box was integrated per ST triplicate to record the data remotely every 5 minutes for pH and temperature, and every 4 hours for the headspace methane concentration. Each control box consisted of a plastic container with a microcontroller, the methane monitoring unit (gas pump, return valve, methane sensor, and filters), pH probe inputs, and thermocouple inputs. For the pH measurement, a pH probe was attached to the top of each ST and submerged approximately 0.3 m below the water level; whereas for the temperature, two thermocouples measuring 0.5 and 1 m in length were attached to the top of each ST unit. An additional thermocouple was set outside one of the STs to record the ambient temperature. A Gascard NG infrared methane gas sensor (Edinburgh Instruments Ltd.) was installed to measure the headspace methane concentration on each ST. A water trap and silica beads filter were installed before the sensor inlet to avoid moisture (>95% relative humidity) from entering the methane sensor. A headspace gas sample from each ST was taken using a gas pump with a flow rate of 0.6 L/h for 5 min and returned to the same ST through a one-way valve. After that, a clean air stream (with the same flow rate) was passed through the filter and sensor to clean the line. The procedure was repeated until all the STs' headspaces were measured. Additionally, a Geotech Biogas 5000 portable analyzer (Cadmus, UK) was used monthly to measure the concentration of methane, carbon dioxide, oxygen, and hydrogen sulfide from the STs' headspace using the same ports for the on-line methane sensor monitoring.

Once a week, one liter of raw sewage (influent) and effluent samples from each ST were taken in triplicate and stored at 4°C until their analysis (no later than 24 h) to monitor water quality parameters according to the Standard Methods (APHA, 2017). The analyses performed included weekly tests for total and soluble chemical oxygen demand (tCOD, sCOD), total suspended solids (TSS), total organic carbon (TON), and total nitrogen (TN); bi-weekly tests for nitrate, nitrite, ammonia, sulfate and total phosphorous (total P); and monthly samples for biochemical oxygen demand (BOD<sub>5</sub>) and alkalinity. Additionally, pathogen removal was



evaluated through the presence or absence of fecal coliform after incubation using the membrane method, where raw wastewater was compared to treated effluent for each condition. The removal efficiency corresponds to the percentage of reduction of each parameter after the treatment in the ST.

Monthly 0.5-L samples were taken from the sample ports (0.1, 0.25, 0.5, and 0.70 m) to monitor the stratification of dissolved oxygen (DO) and oxidation-reduction potential (ORP) using a Go-direct optical dissolved oxygen (Vernier, USA) and ORP probes (Hanna Instruments, USA), respectively. Finally, the sludge depth was monitored at the beginning and end of the experiment using a 5-ft sludge judge (Cole-Parmer, USA).

### 2.3 Carbon mass balance using COD and dissolved methane measurements

Lobato et al. (2012) proposed a model for a carbon mass balance considering COD for wastewater treatment in anaerobic reactors that, for the first time, integrated both the methane lost with the effluent (dissolved methane) and the portion used in sulfate reduction. Following their model, we incorporated into our results the estimation of the COD fraction used in sulfate reduction, considering the influent sulfate concentration, biomass production (sludge retained in the reactor), and the dissolved methane. So then, the complete COD mass balance included the fractions (%) of soluble COD lost in the effluent ( $COD_{not\_converted}$ ), for biomass production ( $COD_{sludge}$ ), for methane production, both in the biogas ( $COD_{CH_4\_biogas}$ ) and dissolved ( $COD_{CH_4\_dissolved}$ ), and used for sulfate reduction ( $COD_{SO_4}$ ). Thus,

$$COD_{influent} = COD_{not\_converted} + COD_{sludge} + COD_{CH_4\_biogas} + COD_{CH_4\_dissolved} + COD_{SO_4} ,$$

(1)

where  $COD_{influent}$  represented the total amount of COD fed to each ST. The  $COD_{not\_converted}$  fraction refers to the portion of influent COD that was not removed or transformed within the

203 system and therefore remained in the effluent samples. Additional grab liquid samples were  
204 taken by each set of STs in triplicate, to corroborate the above dissolved methane estimation  
205 following the methodology proposed by Souza et al. (2011). The gas samples were analyzed in  
206 an FID gas chromatograph (Agilent Technologies 7890B) using a HayeSep Q column.

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## 3. RESULTS AND DISCUSSION

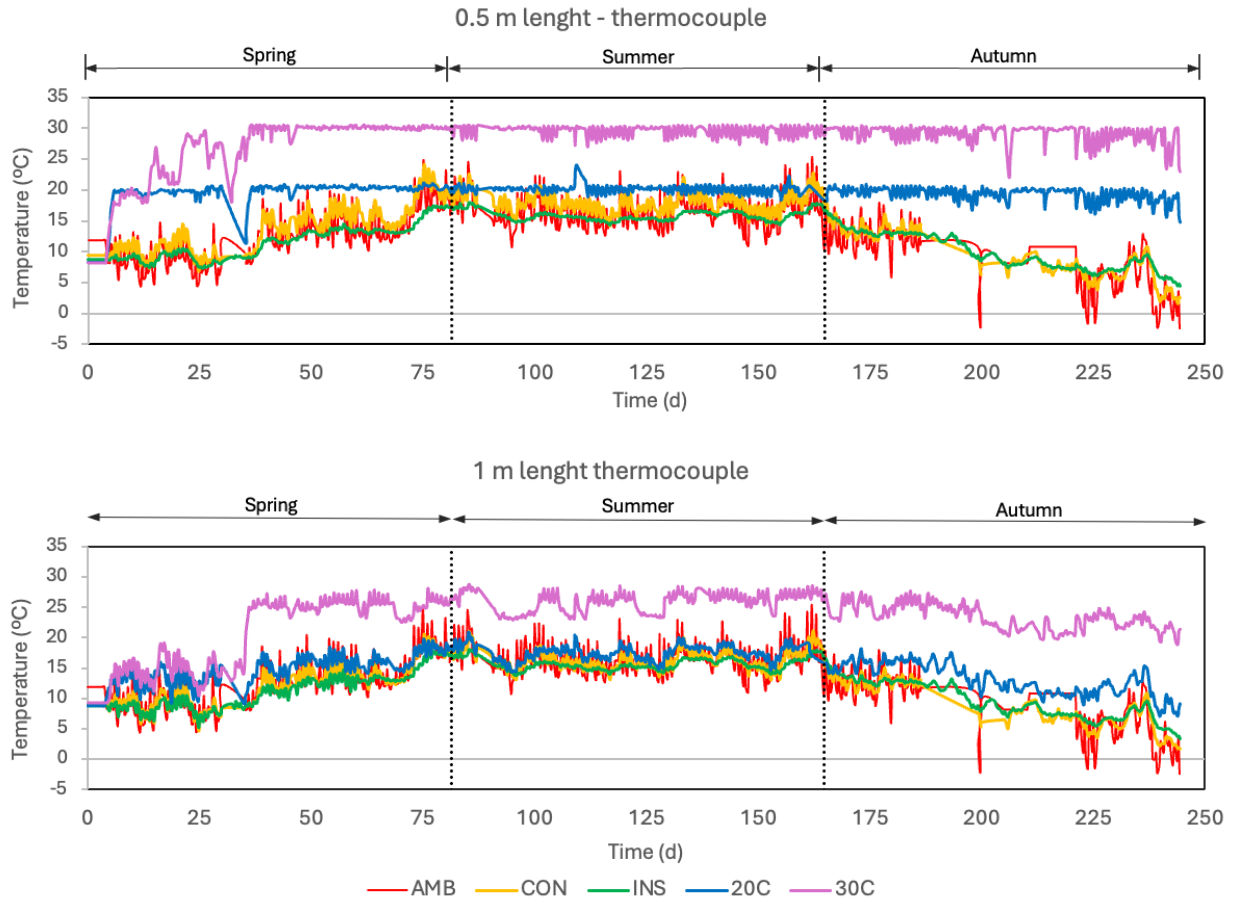
### 3.1 Temperature and pH monitoring

The STs were operated for 244 days. During this time, the ambient temperature was recorded and compared to the bulk-water temperature of the control (CON), insulated (INS), and heated at 20 (20C) and 30 °C (30C) STs. As mentioned above, two thermocouples were installed to measure the water temperature, where the 0.5 m long one was used to control the heated STs, and the 1m long one measured the temperature close to the bottom of the ST. Figure 2 shows the average temperature profiles for each triplicate at the two locations of the thermocouples (0.5 and 1 m depth). For the 0.5 m thermocouples, the CON STs presented a slightly higher temperature compared to the INS STs during spring and summer. In contrast, the insulation in the INS STs provided a thermal buffering effect, reducing the impact of low ambient temperature in the colder months (after day 200).

The 20C was the most stable condition in terms of temperature, with variations within 5 degrees from the desired temperature due to the incoming lower-temperature raw wastewater during the feeding times. Meanwhile, for the 30C STs, it was necessary to add a supplementary heating element beneath them at the beginning of the experimental work to reach the desired temperature on day 32. From day 37 onwards, the temperature remained stable in the 30C set.

Since the mixing effect is extremely low in septic tanks (Mahon et al., 2022), the temperature recorded at the bottom of the STs followed very closely the ambient temperature, except for the 30C that had the extra heating element integrated. Similar to the INS STs, the insulation jacket provided a thermal buffering effect to the heated STs.

According to Shaw and Dorea (2021), a large number of studies on septic tanks indicate that the operating temperature of these systems is within 10-40 °C, with a median of 24 °C.



**Figure 2.** Average temperatures recorded at the 0.5 and 1 m length thermocouples in the control (CON), insulated (INS), and 20 (20C) and 30 °C (30C) heated septic tanks, compared to ambient temperature (AMB).

The pH in the STs was very stable during the experimentation. The average values recorded were  $7.1 \pm 0.5$ ,  $7.0 \pm 0.4$ ,  $6.7 \pm 0.2$  and  $6.6 \pm 0.3$  for the CON, INS, 20C and 30C STs, respectively. Some algal growth was observed in the control STs since the tank (white colour) was not covered by any jacket and received direct sunlight. That could have influenced the slightly higher pH values, while the lower pH in the heated STs is an indication of the enhanced microbial activity, which means a higher concentration of volatile fatty acids and carbonic acid (from the produced  $\text{CO}_2$ ) in the bulk liquid (Wang et al., 2019).

### 3.2 Water quality analysis

The influent can be considered a low-medium strength domestic wastewater. The seasonal variation of the raw wastewater used is presented in Table 1. High sCOD and tCOD concentrations were found during summer and declined in autumn. This trend may be attributed to increased biological activity in warmer months and dilution effects from rainfall in autumn. For example, BOD, tCOD and SST values ranged from 25 to 262 mg/L, 71 to 855 mg<sub>tCOD</sub>/L and 26 to 442 mg<sub>SST</sub>/L, with the lowest values occurring in autumn after rain events and the highest in summer. It is noteworthy that, because the raw sewage was drawn from an open channel leading into an open primary sedimentation tank, ambient conditions influenced the wastewater strength, including dilution by rainwater during autumn. In general, all the parameters are in accordance with previous reports for this type of wastewater (domestic with rainfall influence) in the UK (Martin Garcia et al., 2013; Trego et al., 2021).

Table 1. Seasonal variation of domestic wastewater quality parameters (Mean values  $\pm$  standard deviation).

Parameter	Unit	Season		
		Spring	Summer	Autumn
BOD	mg/L	199 $\pm$ 80	191 $\pm$ 101	117 $\pm$ 11
tCOD	mg <sub>tCOD</sub> /L	474 $\pm$ 195	549 $\pm$ 241	203 $\pm$ 140
sCOD	mg <sub>sCOD</sub> /L	143 $\pm$ 51	205 $\pm$ 93	89 $\pm$ 56
SST	mg/L	198 $\pm$ 91	217 $\pm$ 117	88 $\pm$ 65
Total N	mg/L	37 $\pm$ 21	55 $\pm$ 25	18 $\pm$ 14
NO <sub>2</sub> <sup>-</sup>	mg/L	0.5 $\pm$ 0.4	0.0 $\pm$ 0.0	0.3 $\pm$ 0.2

NO <sub>3</sub> <sup>-</sup>	mg/L	0.8 ± 0.5	0.4 ± 0.0	1.4 ± 1.0
NH <sub>4</sub> <sup>+</sup>	mg/L	31 ± 19	42 ± 19	19 ± 9
Total P	mg/L	4.7 ± 1.9	6.1 ± 2.5	2.5 ± 2.1
SO <sub>4</sub> <sup>-2</sup>	mg/L	45 ± 10	49 ± 4	28 ± 17
Conductivity	mg/L	0.7 ± 0.0	0.8 ± 0.2	0.4 ± 0.3
Alkalinity	mg/L	286 ± 0	335 ± 124	136 ± 117

256

257 The effectiveness of septic tanks in waste removal varies significantly based on several factors,  
258 including the type of wastewater (blackwater alone or combined), the number of chambers, and  
259 the frequency of sludge removal, among others. Therefore, this experiment can be considered a  
260 typical example of a one-chamber septic tank treating real domestic wastewater under good  
261 maintenance practices, operating under different temperature regimes.

262 According to Dasgupta and Agarwal (2021) BOD removal in a conventional ST is expected to  
263 be between 30 and 50%. Our findings showed BOD removals above 55% for all the treatments,  
264 with averages of 55 ± 12, 55 ± 13, 62 ± 12 and 73 ± 13 % for the CON, INS, 20C, and 30C  
265 conditions, respectively. These values corresponded to average BOD concentrations in the  
266 effluent ranging from 53 to 90 mg/L, with absolute concentrations varying between 28 and 157  
267 mg/L (from ST 20C and INS). Figure 3 shows the seasonal variation for tCOD, sCOD and TSS  
268 removal efficiency for the control (CON), insulated (INS), heated at 20 (20C) and 30°C (30C)  
269 triplicates. A positive correlation was observed between operational temperature and RE across  
270 all parameters. The 30C condition consistently demonstrated the highest RE. However, all the  
271 conditions exhibited their highest RE for the three parameters during the spring season. This  
272 could be related to the start-up period, during which a higher amount of solids and organic  
273 matter retention may have occurred in the system. Both tCOD and TSS removal reached a

semi-steady state from the beginning of the operation, despite variations in the influent strength, demonstrating the robustness of the technology. The average tCOD removal in the heated STs was above 65%, with effluent tCOD concentrations ranging from 78 to 461 mg/L. In comparison, the CON and INS STs achieved a tCOD removal efficiency of 56-60%, with the lowest values being 107 and 102 mg<sub>tCOD</sub>/L, respectively. The TSS removal was above 80% and up to  $92 \pm 3\%$  in the 30C STs, whereas the CON and INS STs showed peaks in the effluent concentration following the trend from the inlet wastewater (data not shown).

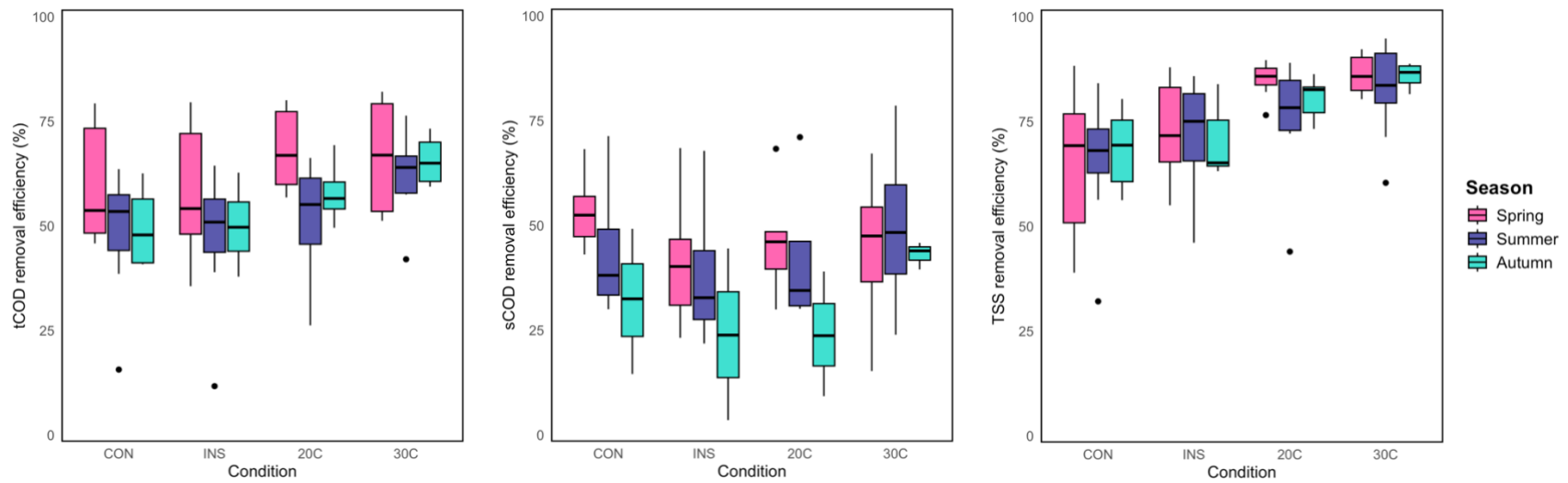
In the case of sCOD, the highest removal efficiency was observed in the 30C STs, followed by the CON condition. This may be due to the algal biofilm growth seen on the walls of the STs, which was promoted by the absence of covering (jacket) in this condition, potentially facilitating aerobic processes within those STs. Similar results were found by Nasr and Mikhaeil (2013) using domestic wastewater in conventional (one chamber) septic tanks under a similar HRT (72 h), at an average temperature of  $27.8 \pm 4$  °C. They reported average removal efficiencies of 65% both for tCOD and TSS, with average influent concentrations of 960 and 295 mg/L, respectively. In this study, the highest tCOD and SST influent concentrations were observed during the summer, at 549 and 217 mg/L, respectively. The removal efficiencies were 64% for tCOD and 85% for SST. Overall, the heated septic tanks (20C and 30C) removed higher concentrations of organic matter and suspended solids.

The seasonal effect was evident in the non-heated STs (CON and INS), especially when colder ambient temperatures were recorded. Once the ambient temperatures dropped below 10 °C, a decrease in both COD and solids removal was observed. Lettinga et al. (2001) described the effects of psychrophilic temperatures in wastewater anaerobic processes: firstly, solids separation due to sedimentation becomes slower under low temperatures when the liquid viscosity increases; then, since the majority of known methanogenic microorganisms are mesophilic and thermophilic, with optimal temperatures of 37 and 55 °C, respectively, it is

expected to have reduced microbial activity in psychrophilic conditions. Previous studies have even reported that anaerobic activity decreases at least one-tenfold when the water-bulk temperature drops in the range of 5 to 35 °C (Viessman Jr. and Hammer, 1999). Still, Halalsheh et al. (2005) reported no significant effect of temperature, when it was between 18 and 25 °C for tCOD and sCOD for UASB reactors fed with high-strength wastewater (tCOD ~ 1500 mg/L).

Nitrogen removal is a crucial step in wastewater treatment, and biologically, it is achieved through a combination of aerobic and anaerobic processes. Therefore, it is natural to have very low nitrogen removal rates after wastewater receives treatment in a septic tank (anaerobic processes only). The main nitrogen compound in domestic wastewater is ammonia ( $\text{NH}_3$ ), which can also be present as ammonium ion ( $\text{NH}_4^+$ ) depending on the pH. In this study, total nitrogen, ammonia, nitrates, and nitrites were measured both in the treated and untreated wastewater. Results shown in Figure 4 indicate that ammonia represents the major nitrogen form in all the samples. It ranged from 25 to 32 mg/L, representing around 86% in the influent and 83 to 94% in the effluent samples, and in accordance with typical values for domestic wastewater (Körner et al., 2001). Total nitrogen removal rates were below 15%, except for the CON STs where the overall nitrogen removal was around 20%, similar to the 22% ammonia elimination. This result can be explained by the fact that the algae biofilm growth in the CON STs' wall could act as microzones where aerobic activity occurred. Overall, our results confirm that nitrification processes and ammonia removal are improbable in STs, as are anammox reactions, due to the slow growth of these microorganisms, which hampers their development in conventional STs (Shaw and Dorea, 2021).





321  
 322 Figure 3. Total and soluble COD (tCOD, sCOD) and total suspended solids (TSS) removal efficiency (%) in the control (CON),  
 323 insulated (INS), and 20 (20C) and 30 °C (30C) heated septic tanks.

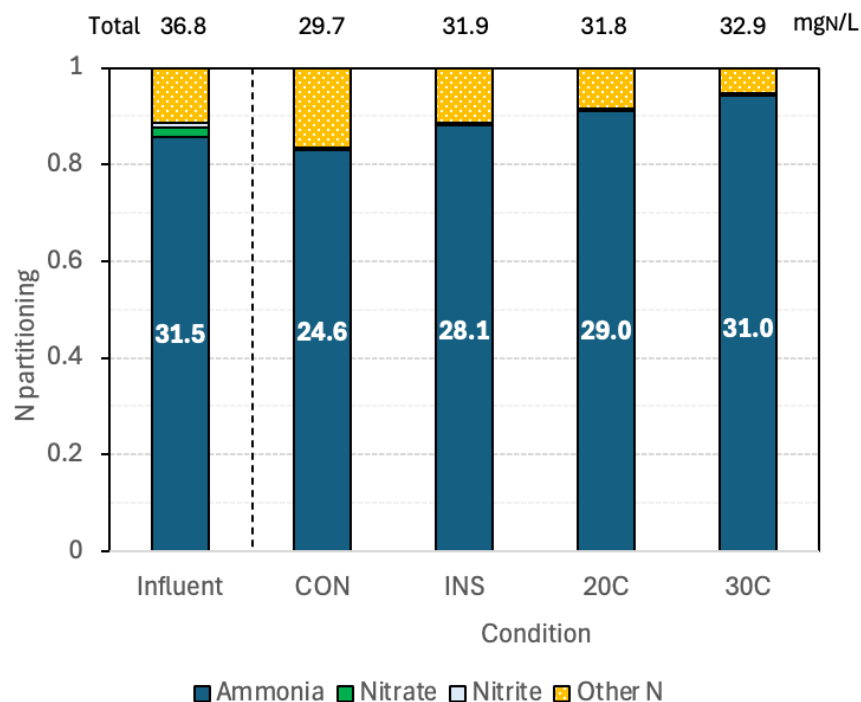


Figure 4. Average nitrogen partitioning in the influent wastewater, and the treated effluent of the control (CON), insulated (INS), and 20 (20C) and 30 °C (30C) heated septic tanks.

Additionally, total phosphorus and sulfate concentrations were measured as part of the nutrients found in raw domestic wastewater. Both nutrients were within the typical range reported by other studies for typical domestic wastewater, at  $4.5 \pm 0.1 \text{ mg}_{\text{TotalP}}/\text{L}$  overall, and  $41.7$  and  $21.3 \pm 4.5 \text{ mg}_{\text{SO}_4}/\text{L}$  for the raw and treated wastewater, respectively (Sahinkaya et al., 2018). It is known that anaerobic conditions in a septic tank promote the growth of sulfate-reducing bacteria. This set of bacteria

Zuo et al. (2019) demonstrated a strong correlation between  $\text{H}_2\text{S}$  emission peaks and increased water flow, specifically water turbulence, which releases sulfide present in the liquid fraction. This may explain why, in our study, we could not detect  $\text{H}_2\text{S}$  production exceeding 5 ppmv under any of the septic tank conditions tested, despite a decrease in sulfate concentration. Monitoring sulfide and  $\text{H}_2\text{S}$  is crucial yet often overlooked in these systems. For instance,

concentrations as low as 2 mgS/L can compromise the septic tank's integrity by causing corrosion, while H<sub>2</sub>S emissions can present significant health risks.

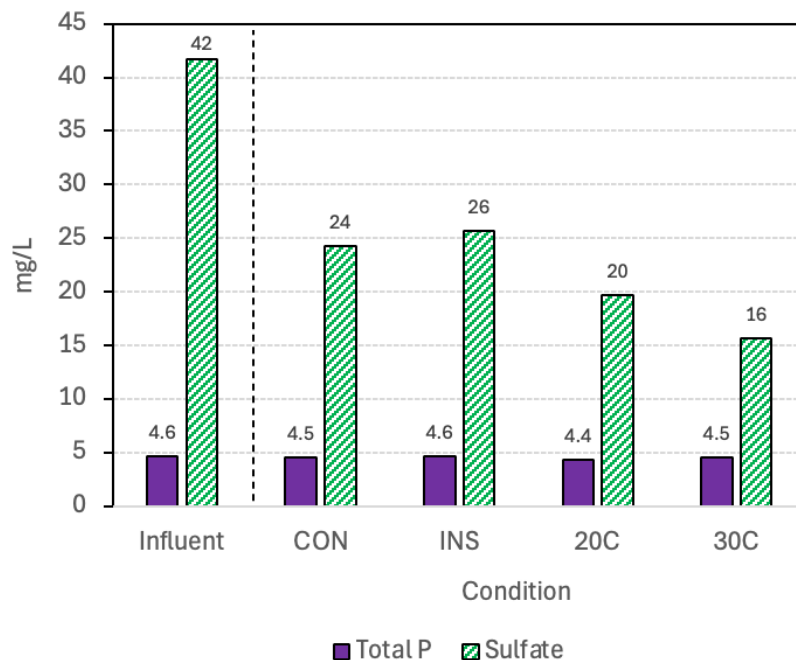


Figure 5. Average total phosphorus (Total P) and sulfate concentration in the influent wastewater, and the treated effluent of the control (CON), insulated (INS), and 20 (20C) and 30 °C (30C) heated septic tanks.

Pathogen content is one of the most important parameters often regulated in direct discharges of treated effluents. In this study, the fecal coliform content was used as an indicator for pathogens. Two sample campaigns were performed at the beginning of the experimental work (May 15<sup>th</sup>), and once the septic tanks demonstrated a semi-steady-state operation (September 11<sup>th</sup>). The results are shown in Table 1. Fecal coliform levels in the raw wastewater ranged from 6 to 12 X 10<sup>7</sup> CFU/100 mL. A one-tenth decrease was observed in the case of CON, INS and 20C STs. In terms of sanitation, heating the water temperature in a septic tank to 30 °C resulted in favorable conditions for a higher fecal coliform removal. However, it is known that other pathogens, such as *Salmonella* spp and helminth eggs, can survive and reproduce at higher temperatures (Scaglia et al., 2014). Therefore, a broader monitoring of pathogens is

recommended depending on the final use of the treated wastewater. Still, fecal coliforms were two orders of magnitude lower compared to Nasr and Mikhaeil (2013), who reported a removal efficiency of 86% at  $27.5 \pm 4.3$  °C.

Table 1. Average values for fecal coliforms (CFU/100 mL) from the raw and treated wastewater at the start and steady state operation.

	May	September
INLET	$6.3 \times 10^7 \pm 5.6 \times 10^6$	$9.0 \times 10^7 \pm 3.4 \times 10^7$
CON	$1.1 \times 10^6 \pm 9.1 \times 10^5$	$2.1 \times 10^6 \pm 1.8 \times 10^6$
INS	$2.9 \times 10^6 \pm 2.2 \times 10^6$	$1.5 \times 10^6 \pm 1.0 \times 10^6$
20C	$1.3 \times 10^6 \pm 7.7 \times 10^5$	$3.1 \times 10^6 \pm 7.8 \times 10^5$
30C	$5.0 \times 10^5 \pm 3.1 \times 10^5$	$2.9 \times 10^5 \pm 1.3 \times 10^5$

### 3.3 Anaerobic conditions and methane emissions monitoring

Dissolved oxygen (DO) and oxidation-reduction potential (ORP) were monitored inside the septic tanks to assess the degree of anaerobicity achieved under each operational condition (Table 2).

Table 2. Dissolved oxygen (DO) and Oxidation-Reduction potential (ORP) monitoring inside the control (CON), insulated (INS), and 20°C (20) and 30°C (30C) heated septic tanks.

	DO range (mg/L)	ORP range (mV)
CON	1.12 – 3.01	-135 to -211
INS	1.20 – 3.04	-130 to -216
20C	0.88 – 2.10	-183 to -225
30C	0.43 – 2.08	-206 to -261

Results indicate that 30C STs exhibited the most strongly reducing environment, characterized by the lowest DO (0.43 to 2.08 mg/L) and the most negative ORP values (−206 to −261 mV). According to Huynh et al. (2021), lower ORP correlates strongly with higher methane emissions ( $R = -0.67$ ;  $p = 0.034$ ). Particularly, STs with long empty intervals were found under complete anaerobic conditions, demonstrated by ORP values ranging from -230 to -489, with 3.9 to 19 years of storage (Huynh et al., 2021). Another study found ORP values ranged from -150 to -210 mV in conventional STs in the USA (Diaz-Valbuena et al., 2011). Our results overlap with these ranges, with the 30C condition being the most effective at fostering anaerobic conditions. The 20C condition also enhanced anaerobic conditions compared to the CON and INS STs, suggesting a moderate increase in methanogenic potential.

The progressive decrease in ORP with increasing temperature supports the notion that temperature primarily affects methane production indirectly by accelerating organic degradation and depleting residual oxygen. Once anaerobic conditions are firmly established ( $ORP < -150$  mV), however, several authors have reported that further increases in temperature or reductions in DO have limited impact on methane yield (Huynh et al., 2021; Moonkawin et al., 2023). For example, comparative field observations between winter and summer (with ~ 9 °C temperature difference) in STs showed negligible differences in methane emission rates, suggesting that temperature alone (within sub-optimal ranges) may not strongly influence methane emissions if anaerobic conditions are already established. However, studies with more extreme temperature differences, such as mesophilic anaerobic digesters operated at 25 °C and up to 34 °C demonstrate that while biogas yield increases with temperature, yields at 31 °C are still close to those at 34 °C (~90%), whereas those at 25 °C are significantly lower (~70%) (Babaei and Shayegan, 2019). Similar behavior was observed here: after the start-up phase, methane generation in CON and INS tanks stabilized across summer and autumn, while greater variability was found in the temperature-regulated reactors (20C and 30C), likely due to

transient cooling from the daily cold-water inputs. Nonetheless, the 30C STs consistently produced biogas with methane concentrations reaching up to 23% v/v in the headspace, confirming active methanogenesis under enhanced thermal conditions.

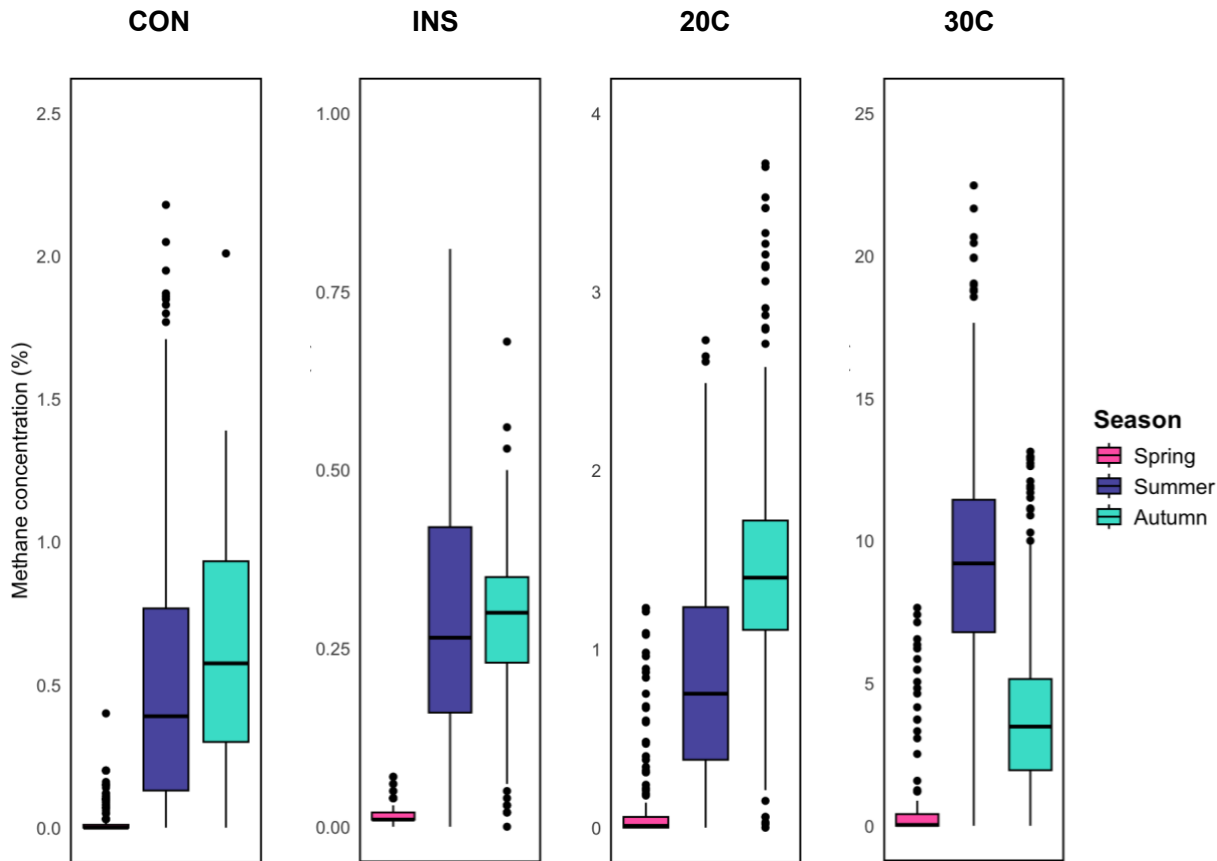


Figure 6. Seasonal variation of the methane concentration in the headspace of the septic tanks.

Beyond headspace accumulation, a substantial portion of methane remained dissolved in the liquid phase, indicating that these systems act as dual emitters. This fraction, shown in Figure 7 as “CH<sub>4</sub> lost in the effluent,” represents the portion of COD converted to methane that escapes collection and contributes to indirect emissions. Several studies have found that most methane emissions from anaerobic wastewater treatment systems result from the release of untreated dissolved methane into the environment (Lobato et al., 2012; Noyola et al., 2006). The coexistence of high headspace concentrations and elevated dissolved fractions suggests incomplete degassing and possible supersaturation of the effluent, particularly under fluctuating

loading and limited mixing. In the last decades, the supersaturation term has been used to refer to a higher amount of dissolved methane than the one theoretically calculated using the equilibrium constant (Henry Law), temperature, and gas phase partial pressure data (Cookney et al., 2016; Noyola et al., 1988; Souza et al., 2011; Zhang et al., 2022). This dual pathway agrees with previous findings in anaerobic domestic wastewater systems, where dissolved methane can represent between 20% and 60% of total emissions depending on temperature, hydraulic retention and turbulence (Crone et al., 2016; Lobato et al., 2012; Souza et al., 2011).

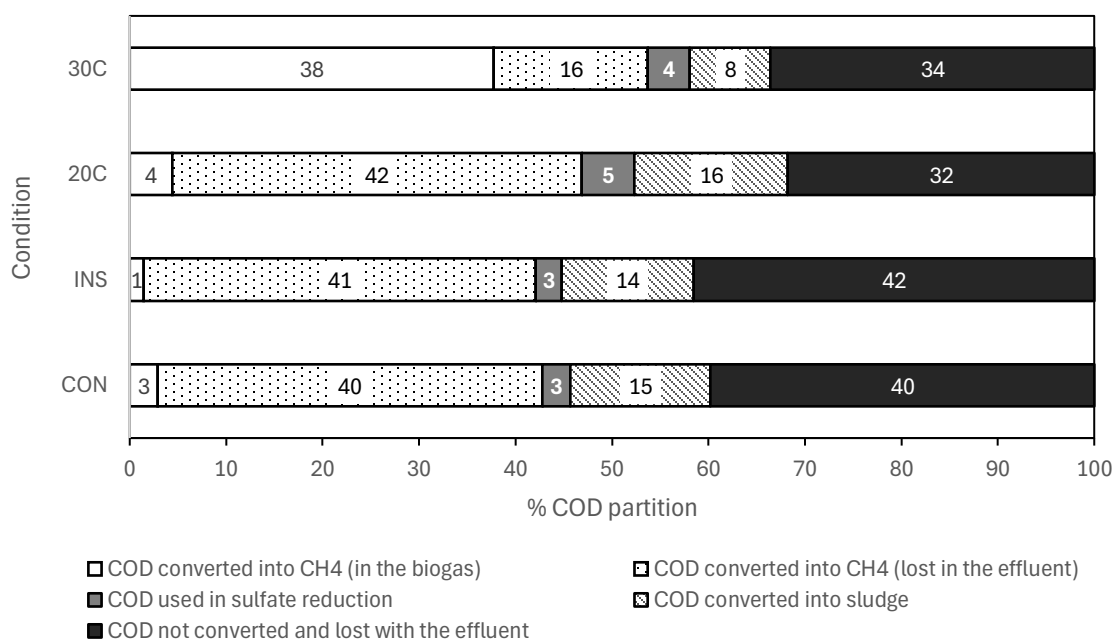


Figure 7. Average COD partitioning in septic tanks.

In our study, the persistence of high dissolved methane concentrations, even under conditions where the headspace contained elevated methane levels, indicates a rapid but incomplete gas-liquid equilibration periodically disrupted by feeding events (turbulence) and therefore transient pressure variations. As expected, since methane solubility is temperature-dependent, this also highly influenced methane partitioning in STs. It is clear that higher temperature (30 °C) enhanced desorption from the liquid phase, leading to greater accumulation of methane in the headspace. Nevertheless, a considerable fraction of the total methane produced, corresponding

to approximately 30% of the total COD transformed into methane, remained in the liquid phase and was subsequently discharged with the effluent. Under cooler conditions (CON and INS), this fraction increased markedly (>90%), highlighting that temperature not only governs methanogenic activity but also the balance between gaseous release and dissolved retention. These results confirm that even in well-developed anaerobic environments, the dissolved phase represents a substantial portion of methane-derived COD. This pathway is rarely measured despite having clear implications for both energy recovery and greenhouse-gas accounting (Crone et al., 2016; Gómez-Borraz et al., 2025, 2017; Huete et al., 2018). This observation challenges conventional assumptions used in global inventories and current emission models. While the IPCC methodology provides a simplified framework for estimating total methane generation from septic tanks, it does not explicitly distinguish between methane released as biogas and methane remaining dissolved in the effluent. Our findings show that, under certain operating conditions, particularly at lower temperatures, a considerable amount of the methane produced can remain in the liquid phase, underscoring the importance of accounting for this route when assessing total methane releases. The clear coupling observed between redox potential, temperature and methane partitioning underscores that STs are dynamic emitters whose gaseous and dissolved fluxes respond to subtle operational and environmental changes. These insights emphasize the need for monitoring approaches that encompass both phases and for inventory frameworks that can better reflect the multiple routes through which methane is released from on-site systems. Recognizing and quantifying both pathways is essential for improving greenhouse-gas assessments of decentralized sanitation systems and for guiding the development of future emission factors and mitigation strategies.



## CONCLUSIONS

This study demonstrates that temperature is a primary driver of both treatment performance and methane dynamics in full-scale septic tanks treating real domestic wastewater. Higher water temperature enhanced organic degradation and strengthened anaerobic conditions, increasing methane generation: the fraction of influent COD converted to methane increased from approximately 45% under insulated conditions to up to 54% at 30 °C. Crucially, temperature also strongly influenced methane partitioning between dissolved and gaseous phases. Under cooler conditions ( $\leq 20$  °C), methane was predominantly retained in the liquid phase, with up to 98% of produced methane remaining dissolved and discharged with the effluent. In contrast, at 30 °C, enhanced desorption favored methane transfer to the gas phase, reducing the dissolved fraction to around 30% and resulting in headspace accumulation. This highlights an emission pathway that is rarely quantified in routine monitoring.

The coexistence of high dissolved and gaseous methane fluxes indicates that septic tanks behave as dynamic dual emitters whose emissions cannot be reliably assessed from headspace measurements alone. These findings underscore the importance of considering dissolved-phase methane in greenhouse gas assessments of decentralized sanitation systems and in future refinements of emission factors.

From a management perspective, strategies aimed at improving treatment performance by increasing temperature must be evaluated alongside their potential to elevate methane emissions. Integrating dissolved-methane monitoring, developing mitigation approaches (e.g., degassing or oxidation stages), and improving inventory methodologies will be essential for accurately capturing the climate impact of septic tanks and informing sustainable sanitation practices.

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## CRedit author contribution

**Tania L. Gómez-Borraz:** Data curation, formal analysis, investigation, methodology, supervision, writing-original draft, review and editing. **Calum Cuthill:** Data curation, investigation, methodology. **Tymon Herzyk:** Investigation, methodology. **Stephanie Connelly:** Conceptualization, funding acquisition. **William T. Sloan:** Conceptualization, funding acquisition, project administration, resources, supervision, writing-review and editing.

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