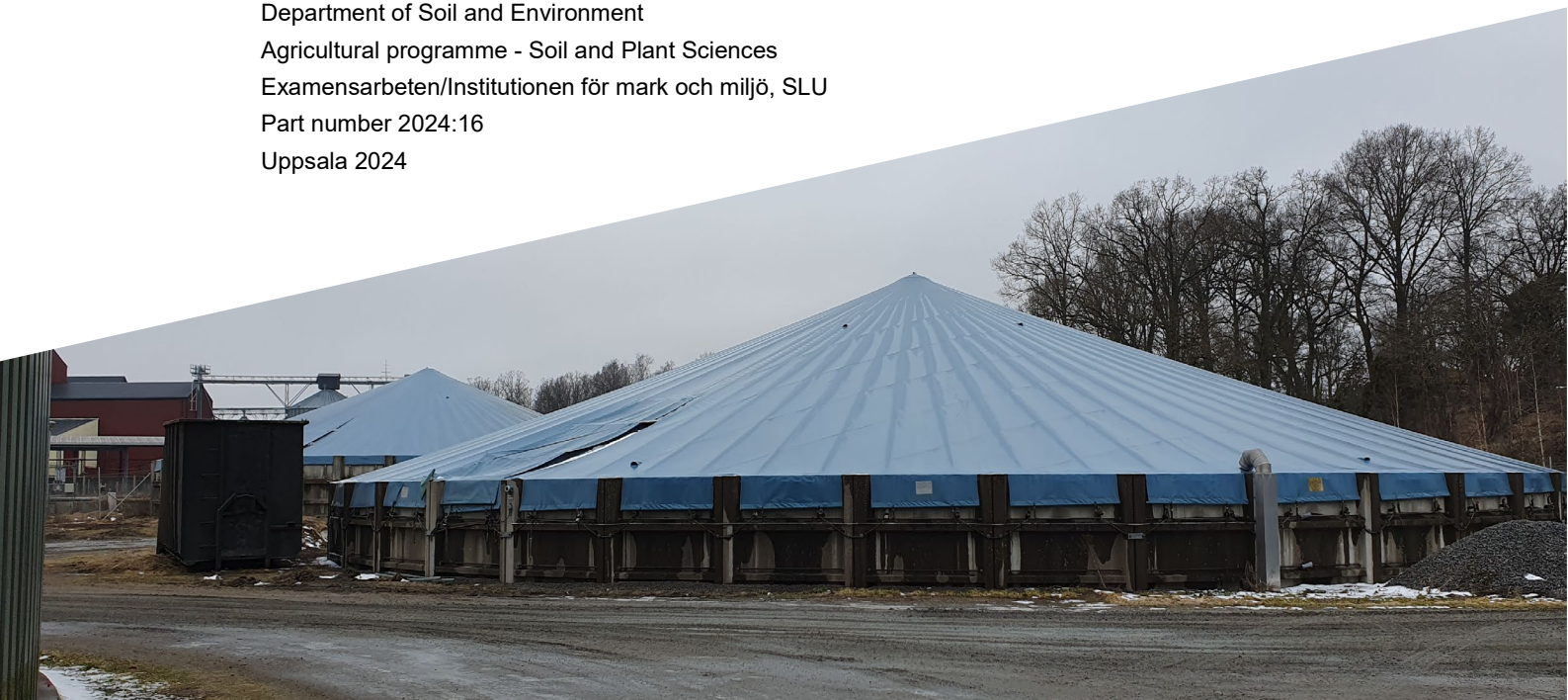




The Effect of Ammonium Sulphate on Methane emissions when added to organic fertilizers

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Effekten av ammoniumsulfat på metanemission när det tillsätts till organiska gödselmedel

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Keywords: Ammonium sulphate, methane, digestate, cattle manure, hydrogen sulphide

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Abstract

Methane (CH₄) is the most common greenhouse gas being emitted from Sweden's agriculture, where approximately 4.5% of Sweden's total emission in 2022 originated from livestock manure storage facilities, highlighting the importance of optimizing manure degradation processes. This study was part of a project led by Research Institutes of Sweden, which aimed to investigate if CH₄ emissions could be reduced by adding ammonium sulphate ((NH₄)₂SO₄) to organic fertilizers. The quantitative study was conducted under mesophilic conditions (37°C) for 29 days, focusing on the effects added (NH₄)₂SO₄ had on samples of two types of digestates and one type of cattle manure. In addition to CH₄ emission, hydrogen sulphide (H₂S) emissions – known for its toxicity at low concentrations - were measured. The study also examined changes in volatile solids (VS), pH levels, and volatile fatty acid (VFA) composition before and after the experiment.

The results were conclusive, CH₄ emissions were reduced when (NH₄)₂SO₄ was added to the respective fertilizer. However, H₂S emission increased significantly in all samples containing (NH₄)₂SO₄. This suggests that adding (NH₄)₂SO₄ to organic fertilizers could potentially be a way of reducing CH₄ emission during the storage of digestates or animal manures. However, further research is needed to determine its applicability under farm conditions. Additionally, it is crucial to assess the risks posed by H₂S exposure to workers handling such materials under realistic working conditions.

Keywords: Ammonium sulphate, methane, digestate, cattle manure, hydrogen sulphide

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Abbreviations

$(\text{NH}_4)_2\text{SO}_4$	Ammonium sulphate
CH_3COO^-	Acetate
CH_4	Methane
CO_2	Carbon dioxide
FeCl_3	Ferric chloride
H_2S	Hydrogen sulphide
H_2	Hydrogen gas
LCFA	Long chain fatty acid
NH_3	Ammonia
NH_4^+	Ammonium
$\text{NH}_4\text{-N}$	Ammonium-nitrogen
O_2	Oxygen
N_2O	Nitrous oxide
RISE	Research Institutes of Sweden
S^{2-}	Sulphide
SO_4^{2-}	Sulphate
SRB	Sulphur-reducing bacteria
TA	Total alkalinity
TS	Total solids
VFA	Volatile fatty acids
VS	Volatile solids

1. Introduction

In Sweden's agriculture, methane (CH₄) and nitrous oxide (N₂O) are the main greenhouse gases being emitted, with CH₄ being the predominant gas (The Swedish Environmental Protection Agency 2024). CH₄ has a global warming potential 28 times greater than carbon dioxide (CO₂) over a 100-year period (Baldé et al. 2016), meaning that by reducing its emission, the near-term global warming could be mitigated as well (IPCC 2023). In 2022, about 4.5% of the total agricultural CH₄ emission in Sweden came from livestock manure storage facilities, stored primarily for its use as an organic fertilizer (The Swedish Environmental Protection Agency 2024). Globally, about 4.7% of the annual anthropogenic CH₄ emission come from manure degradation (Vechi et al. 2023).

Among organic fertilizers, livestock manure is the most commonly used in Sweden, followed by digestates from biogas plants (Swedish board of Agriculture 2023). A biogas plant is a facility where organic matter – such as food waste and waste from slaughterhouses (Energigas Sverige 2023) - is subjected to anaerobic conditions to produce biogas (CH₄) through microbial decomposition. The primary goal of the facility is to produce CH₄, which can be used for various purposes, such as vehicle fuel, heating, or electricity. In addition to the produced biogas, a digestate is formed that can be used as an organic fertilizer. The gases produced are mainly CH₄ (55-80%) and CO₂ (20-45%) but also include nitrogen (N) (0-1%), hydrogen sulphide (H₂S) (50-2000 ppm), oxygen (O₂) and hydrogen gas (H₂) (Christensson et al. 2009). The exact proportion of each gas formed depends on the organic matter added to the facility and the activity of the methanogens, i.e. the anaerobic microorganisms that produces CH₄ as a byproduct of their metabolism (Jarvis et al. 2009). For a high CH₄ content, a substrate rich in fat and protein is needed, while a substrate rich in carbohydrates will produce a lower CH₄ content (Christensson et al. 2009). The activity of the methanogens continues even after the digestate leaves the biogas facility. In one study, stored digestates from biogas plants emitted around 12% of the total annual quantity of the CH₄ produced in the digester (Baldé et al. 2016). Therefore, reducing CH₄ emissions from stored organic fertilizers, such as livestock manure and digestates, is of increasing interest.

Ammonium sulphate $(\text{NH}_4)_2\text{SO}_4$ is a commonly used sulphur fertilizer in agriculture, produced primarily as a commercial fertilizer. It can also be produced in agro bio-refineries, where biomass is processed to obtain bioenergy and other products, similar to a biogas plant (Szymańska 2019). Studies have shown that $(\text{NH}_4)_2\text{SO}_4$ reduces the CH_4 emission when applied to rice (*Oryza*) fields, which have significant CH_4 emissions annually. This is due to its effect on the microbial activity of methanogens and sulphate-reducing bacteria (SRB). With an increase in sulphur containing compounds, methanogens are outcompeted by SRB in the anaerobic digestion, since SRB thrives in sulphur rich environments (Lindau et al. 1993; Denier van der Gon et al. 2001). In one study, CH_4 emission were reduced by about 60-70% (Denier van der Gon et al. 2001). For this reason, this study aims to determine if $(\text{NH}_4)_2\text{SO}_4$ can have a similar effect on CH_4 emissions when added to organic fertilizers.

1.1 The anaerobic digestion

The anaerobic digestion in a biogas plant is similar to the degradation of livestock manure in a storage facility, so to understand how $(\text{NH}_4)_2\text{SO}_4$ can affect the organic fertilizers used in this study, it is of relevance to have insight in how the digestion in a biogas plant occurs. In a biogas plant, the digestion is divided into four different process steps: hydrolysis, acidogenesis, acetogenesis and methanogenesis. In each step of the process different microorganisms are active, specialized for that specific step (Christensson et al. 2009). The steps can take place simultaneously.

In hydrolysis, the initial larger molecules such as sugars, fats and proteins in the organic matter are degraded to smaller molecules as simple sugars, fatty acids, and amino acids when reacting with water added to the solution (Anukam et al. 2019). Some of the commodities produced in this step can be directly used by methanogens, for example acetate (CH_3COO^-) and H_2 , while other needs to be decomposed in further digestion (Ibid.).

In acidogenesis, the smaller molecules formed in hydrolysis are used as substrates for fermenting microorganisms which produce organic acids, alcohols, ammonia (NH_3), CO_2 and H_2 (Jarvis et al. 2009). During both hydrolysis and acidogenesis different fatty acids are produced which is then degraded in acetogenesis and methanogenesis. A distinction is made between volatile fatty acids (VFA or FOS) and long chain fatty acid (LCFA), VFA being short-chain fatty acids (two to six carbon atoms) and LCFA being fatty acids with aliphatic tails (thirteen to sixteen carbon atoms). Both VFA and LCFA are frequently analysed in a biogas plant. This since if the number of fatty acids accumulates, and there for the decomposition of them is lacking, it can be a result of the methanogens being inhibited which then

results in a low production of CH₄. For the most favourable production of CH₄ the pH in the biogas facility needs to be between 7.0 and 8.5, and usually an increase in VFAs result in a lower pH (Idib.). For this reason, regular tests are run on the solutions alkalinity (i.e. the solutions buffering capacity), which is the measurement of the solutions ability to withstand a decrease in pH caused by added acids, like VFA (Jantsch et al. 2003). One common measurement in a biogas plant is the quota of FOS/TAC, also known as VFA/TA, where TA or TAC is the total alkalinity in the sample. If FOS/TAC <0.3 the anaerobic digestion is stable. Between 0.3-0.5 the system is experiencing some instability. If FOS/TAC is >0.5 the system is experiencing clear instability (Jarvis et al. 2009). An important premise for the value of FOS/TAC is that substrate is supplied regularly in the biogas plant, as the substrates effects the activity of the microorganisms and therefore also the stability of the anaerobic digestion.

Acetogenesis and methanogenesis are in close relation to one another, mostly due to the concentration of H₂ accumulated in the previous step, acidogenesis. The metabolism of acetogenic bacteria is inhibited by a high concentration of H₂ while methanogens can use it to produce CH₄ (Anukam et al. 2019). The syntrophic interaction between these different microorganisms is very important in a biogas plant. The methanogens frequently remove H₂ and maintains a low concentration, which enable acetogenic bacteria to decompose fatty acids, alcohols and VFAs to produce CH₃COO⁻, acetic acid (CH₃COOH), CO₂ and H₂ (Idib.). In the last step of the anaerobic digestion, methanogenesis, the produced CH₃COO⁻, and H₂ is then converted by methanogens into CO₂ and CH₄ (Idib.).

1.2 Factors affecting the production of CH₄

There are several factors affecting methanogens activity and therefore CH₄ production and emission. For instance, pH and alkalinity (as mentioned earlier), temperature, and volatile solids (VS) (Jarvis et al. 2009; Vechi et al. 2023). Usually in a biogas plant the temperature is either mesophilic, around 37°C, or thermophilic, around 55°C (Christensson et al. 2009). Thermophilic conditions lead to a faster degradation than mesophilic (Jarvis et al. 2009). However, the microorganisms subjected to thermophilic conditions are more sensitive for a drop in temperature than mesophilic since usually few mesophilic species, i.e. microorganisms active at 37°C, is present in a thermophilic biogas plant, as these have perished at the high temperatures (Ibid.). This means that in the event of a temperature drop in a thermophilic environment, it may take time before CH₄ production goes up again (Ibid.). More microorganisms are usually active in mesophilic conditions, meaning mesophilic temperatures enable a greater diversity of microorganisms and a more stable and slow decomposition of substrates manure (Ibid.).

The VS is the amount of organic matter in a substance which is combustible at 500°C, i.e. VS represents the organic compounds that is emitted as gas when the sample is heated to 500°C. In a biogas plant, VS is measured after calculating the amount of dry matter content, i.e. the measurement of how much total solids (TS) or liquid a material contains after the water content evaporated at 105 °C (Swedish Waste Management 2009). Since a digested residue is more decomposed than an undigested manure, the VS content is usually lower for a digestate than for the original substrate. A material with a high VS content usually has a higher gas exchange per unit of volume than a material with a low VS content, due to more available degradable material (Ibid.). As mentioned earlier, the composition of said material also affects the methanogens activity and rate of degradation since different substrates differ in their chemical energy (Vechi et al. 2023).

1.3 Sulphate-reducing bacteria

In addition to methanogens there are other microorganisms in a biogas plant that can compete for the substrates produced, for example sulphate-reducing or nitrate-reducing organisms (Jarvis et al. 2009). Usually, methanogens are the dominant organisms but in case of an increase concentration of sulphur compounds SRB can increase its growth at the expense of methanogens (Ibid.). This since SRB has a higher affinity and faster growth rate at low substrate concentrations for both H₂ (Kristjansson et al. 1982) and CH₃COO⁻ (Schönheit et al. 1982) than methanogens. This enables SRB to be in a syntrophic relationship with acetogens instead of methanogens and out-compete them for the available substrates in the acetogenesis process step (Jarvis et al. 2009; Lindau et al. 1993; Denier van der Gon et al. 2001).

In the chemical reaction, SRB reduce sulphate (SO₄²⁻) to sulphide (S²⁻) while producing H₂S (Chen et al. 2007), a highly poisonous gas, even in low concentrations. With a concentration of 1000 ppm, it is instantly deadly for humans (U.S. Department of Labor. n.d.). Fatalities and injuries have occurred at livestock manure storage facilities where workers have been subjected to low concentrations of H₂S (Beaver et al. 2007; Park et al. 2016). More so, methanogens can also be inhibited by a high concentration of H₂S, because of the gas toxicity (Zhou et al. 2016). In a biogas plant or a livestock manure storage facility, it is important to make sure that the concentration of H₂S is not too high and that SRB is not the predominant microorganism. One way of doing so is to add ferric chloride (FeCl₃) to the digestate, since FeCl₃ makes existing sulphur-containing compounds inaccessible for SRB in the substance when iron precipitates sulphide (Edström et al. 2013; Zhou et al. 2016). That being said, H₂S does not only form due to the activity of SRB but is also produced in other steps in the anaerobic digestion. For

an example, it is produced during the breakdown of sulphur-containing amino acids such as cysteine and methionine and can be emitted in large quantities when protein-rich waste is degraded, as undigested livestock manure (Jarvis et al. 2009).

As mentioned, the concentration of S^{2-} in the biogas plant is determined by the competition and interaction between SRB and other microorganisms in the facility (Chen et al. 2007). SRB cannot degrade bigger molecules available in the early stages of anaerobic digestion but needs the initial breakdown to be done by other organisms (Ibid.). For this reason, the competition for substrates is mainly between methanogens and SRB in acetogenesis. Other factors affecting the competition, apart from a high concentration in SO_4^{2-} , is the temperature. At mesophilic conditions SRB are dominant, while at thermophilic conditions methanogens are dominant (Chen et al. 2007).

1.4 Study aim

The aim of this study is to examine the effect $(NH_4)_2SO_4$ has on CH_4 and H_2S emissions when added to two different kinds of biogas digestates and one cattle manure. This as part of a project led by Research Institutes of Sweden (RISE) together with Swedish University of Agricultural Sciences (SLU) and the companies EasyMining and More Biogas. The prospect with the project is to receive a fertilizer with a high plant nutritional value while at the same reduce its CH_4 emission during storage. Due to time constraints while writing a bachelor's thesis, this essay's focus will be on CH_4 emissions while the plant nutritional value of the organic fertilizer before and after $(NH_4)_2SO_4$ is added will only be looked at briefly.

When $(NH_4)_2SO_4$ is added, the respective fertilizer is suspected to (1) have a reduced amount of CH_4 emission due to the available amount of sulphur, i.e. SRB will effectively inhibit methanogens and (2) the emissions of H_2S will increase because of said activity of SRB. How substantial the emission is will give an indication of possible risks and/or benefits with adding $(NH_4)_2SO_4$ to biogas digestates or/and manures.

2. Materials and method

The quantitative study, including preparatory work, took place at Research Institute of Sweden (RISE) laboratory in Uppsala, Sweden from week 12 to week 18. The actual test of the respective substance took place for 29 days, from 2024-04-03 to 2024-05-02.

Before the actual test begun, a test trial was performed during 5 days in room temperature too see the initial microbial activity in the different organic fertilizers. The test consisted of three bottles with the respective fertilizer in them and no substance added. During this test trial, very little pressure was formed in the bottles. Because of this, the decision was made to subject the bottles to mesophilic conditions (37°C), to receive a result during the prevailing time constraints when writing a bachelor thesis. Meaning, this study is a pilot study of the processes, to see relative differences between treated digestates and cattle manure during controlled temperature conditions.

2.1 Materials

The $(\text{NH}_4)_2\text{SO}_4$, with a concentration of 33% and a pH of 5.28, were supplied from EasyMining in Uppsala, Sweden. It was produced as a part of EasyMining's Aqua2N process, a process in which nitrogen (N) is extracted and separated from wastewater with a precipitation chemical. The equipment for the experiment were supplied by RISE. AgriLab, a commercial laboratory in Uppsala, Sweden, performed selected measurements of macro- and micronutrients, contents of total N and carbon (C) and ammonium-nitrogen ($\text{NH}_4\text{-N}$) analysis.

The cattle manure and one of the biogases digestates, produced from mainly cattle and swine manure in mesophilic conditions, were supplied by Lövsta agricultural research station in Uppsala, Sweden. The cattle manure was undigested and had therefore larger pieces of organic material in it. The other biogas digestate, produced from mainly poultry, cattle and swine manure in thermophilic conditions, were supplied by More Biogas in Läckeby, Sweden. More biogas digestate had ferric chloride added to it. To reduce the risk of microbial activity and emission of gas before the start of the project the samples were kept in a fridge at ~8°C.

2.2 Dry matter content and volatile solids

The dry matter content (%), or TS, were measured by adding about 50 grams of the respective slurry, i.e. cattle manure or digestate, in 9 different aluminium containers, three for each slurry, and then them being subjected to air-drying at 103°C for 8 hours. The mass remaining after the heat-treatment were divided with the start weight of each sample, multiplied with 100 to receive the mass percent of the TS content.

The VS content (%) was measured by an additional heat-treatment of the TS samples, where the samples were subjected to air-drying at 500°C for 8 hours. The mass of the ash remaining were divided with the previously measured TS mass and multiplied with 100 to receive the glow residue, which is the remaining material after heat-treatment consisting of inert materials and ash. VS, which is stated in weight percent of TS, was then calculated by subtracting the glow residue from 100.

2.3 pH and nutrient content

To see if there were any direct change in pH after $(\text{NH}_4)_2\text{SO}_4$ were added, pH were measured with Hanna edge HI2002-01 dedicated pH/ORP at the start of the study before and after $(\text{NH}_4)_2\text{SO}_4$ were added. pH was also measured when the experiment was ended, but not any time during the study after the glass bottles were sealed.

The content of macronutrients (phosphorus, calcium, potassium, magnesium, sodium, and sulphur) and micronutrients (iron, copper, manganese, and zinc) of the three slurry's were determined by AgriLab before and after the study was ended with ICP-OES. Contents of total C and N were estimated with a CN auto-analyser (Carol Erba NA2000, Milan).

2.4 FOS/TAC and the composition of VFA

FOS/TAC of the respective sample were measured with Hach titrator AT1000 before and after the study was ended. Each sample were measured individually with two different replicates. All samples were at room temperature. Approximately 5 g of a sample were filtered in a container, to remove bigger sized organic matter such as straws, and then 50 ml of deionized water was added to the same container. The container was then placed in the Hach titrator. If the test result varied greatly between the two samples further tests were run on new samples until the result did not vary too much.

To measure the composition of VFAs of respective slurry, samples were gathered in 15 ml centrifuge test tubes before and after the study was terminated. The samples taken before the test was ended were placed in a freezer, to prevent unwanted activity in the sample that could affect the result, until all samples were gathered. In total 10 test tubes were collected, one for each sample before the study began, one for each control sample and one for each test sample.

All the samples were centrifuged for 10 minutes in 1500 rpm. After the centrifugation, 1 ml of the fluid at the top of respective sample were collected using Finnpiquette F2 and placed in a 1.5 ml centrifuge test tube. There were two test tube for each sample, in total 20 samples. To the test tubes, 0.1 ml 5 M sulfuric acid (H_2SO_4) were added using Finnpiquette F3 in respective sample. The test tubes were then centrifuged once again for 15 minutes at 4000 rpm. After the centrifugation, approximately 1 ml in total were gathered from each sample into an oral syringe and then filtered through a syringe filter (0.22 μm) to a glass test tube. In total 10 samples (one for each sample) were gathered. The VFAs in each sample were then measured by staff at RISE with liquid chromatography, using HPLC at SLU BioCenter in Uppsala.

2.5 Experimental set-up and measurements

$\text{NH}_4\text{-N}$ analysis was performed by staff at RISE through steam distillation and titration to receive the initial amount of available ammonium (NH_4) in the respective samples. The results of the distillation were also confirmed with results from AgriLab, who determined the $\text{NH}_4\text{-N}$ content by flow injection analysis. This information was then used to calculate the amount of $(\text{NH}_4)_2\text{SO}_4$ (ml) that needed to be added to double the amount of NH_4 in the respective samples. To calculate this the molar mass and concentration of the $(\text{NH}_4)_2\text{SO}_4$ were used as a starting point.

Each material had three replicates for the test samples and three for the control samples. The control samples had tap water added to it instead of $(\text{NH}_4)_2\text{SO}_4$ so all samples would contain the same volume. In addition to this, Lövsta biogas digestate was also tested with a quadrupled amount of $(\text{NH}_4)_2\text{SO}_4$ with three replicates. In total 21 glass bottles were used for the experimental set-up.

To calculate the total volume of the bottles, 1000 ml glass bottles were weighed individually and then filled with tap water to the neck edge of the bottle, to see how much the bottles were able to contain. The glass bottles were then filled with approximately 500 g, which corresponded to about 500 ml, well stirred material

each and sealed with a silicone stopper, so no gas was able to leave the bottles. When the bottle was sealed, $(\text{NH}_4)_2\text{SO}_4$ was added by a syringe through the silicone stopper in the test samples. Tap water was added to the control samples with the same method.

The biogas digestates had more NH_4 from the starting point than the cattle manure, which meant that more $(\text{NH}_4)_2\text{SO}_4$ was added to these bottles. 19 ml was added to the biogas digestates and 10 ml to the cattle manure (Table 1). All the samples were then placed in a heating cabinet at 37°C , to mimic a mesophilic temperature.

Table 1. Starting amount of $\text{NH}_4\text{-N}$ in the samples, needed amount of $\text{NH}_4\text{-N}$ and volume of $(\text{NH}_4)_2\text{SO}_2$ added to achieve double and quadrupled amount of $\text{NH}_4\text{-N}$, respectively, in 500 ml of fertilizer

	$\text{NH}_4\text{-N}$ starting point (mg/l)	Needed addition of NH_4 (mg/l)	Added $(\text{NH}_4)_2\text{SO}_2$ (ml)
A: More Biogas biogas digestate, double	1.35	1.35	19.0
B: Lövsta biogas digestate, double	1.30	1.30	19.0
C: Lövsta cattle manure, double	0.72	0.72	10.0
D: Lövsta biogas digestate quadrupled	1.30	2.60	38.0

Approximately every two to four days CH_4 and H_2S emissions were measured. This to ensure that enough gas had accumulated in the bottles, since this is not always the case at close intervals of testing. The microbial activity was higher in the beginning of the study, when abundant substrate was available, which meant that the sampling frequency was higher then. All the measurements were taken in a fume hood to reduce the risk of being exposed to dangerous gasses, such as H_2S .

Before gas sampling, the samples were taken out of the heating cabinet and placed at room temperature ($\sim 21^\circ\text{C}$) to slow down the activity of the microbes in the samples. After waiting for about 30 minutes after taking out the samples the bottles were shaken, to make gas accumulated at the bottom of the bottle rise above the different slurries, and then temperature and pressure was checked. The gas temperature was measured with Fisherbrand Traceable Circle Laser through the

glass of the bottle. Pressure was measured with Greisinger GMH-3181-12, with a needle through the silicone stopper.

CH₄ emission was measured by the removal of 1 ml of gas from respective sample by a syringe through the silicone stopper. Before the sample was taken gas was drawn up and down the syringe three times to mix the gasses in the sample. The samples were then measured by staff at RISE with gas chromatography using Perkin Elmer Arnel, Clarus 500, at SLU BioCenter in Uppsala. At the end of the study, a one-way Anova analysis was made of the results of the respective slurry's CH₄ emission by staff at SLU, to investigate if differences between the CH₄ mean values were significant. The significance limit set was $p < 0.05$. Tukey-Kramers test was used for pairwise comparisons of the different treatments.

H₂S emission was measured using Geotechs' Biogas5000. Gas was collected from the samples in a gas bag that was connected to the glass bottle with a needle through the silicone stopper. To ensure no gas leakage a peang tong was used to seal the tube connected between the needle and the gas bag before and after the sample was gathered. To obtain enough gas to perform the measurements on, gas from the three test samples were collected in the same bag and likewise gas from the three control samples were collected in another bag for each slurry sample. The gas bag was then connected to Biogas5000 and after one minute the data was collected. For these measurements no statistical evaluation was possible, since Biogas5000 only provides one result when measuring, meaning there is no mean values to perform a significance analysis on.

If the bottles still had some pressure in them after the samples had been taken, they were emptied with a needle through the silicone stopper, so that the pressure was restored to the original pressure that was in the bottles when they were sealed (0 mbar).

2.6 Calculations for methane emissions

To calculate the respective bottles headspace (Nml, N standing for normal condition with the room temperature being ~21°C and the pressure 1 mbar), meaning the total gas volume the bottle was able to contain, the volume of the added material (ml), i.e. the slurry and (NH₄)₂SO₄ or water, was subtracted from the measured volume (ml) of the specific bottle when containing only tap water.

The total gas volume (Nml) was calculated using the atmospheric pressure (atm, measured in mbar), the prevailing pressure (ρ_{bottle}) and temperature (T_{bottle}) of the

respective bottle at the time of the sampling, multiplied by the headspace of the bottle (Equation 1).

$$\left(\left(\frac{atm + \rho_{bottle} \times \left(\frac{273 K}{T_{bottle}} \right)}{atm} \right) - 1 \right) \times headspace_{bottle} \quad (1)$$

To receive the total CH₄ volume (Nml) in the respective sample the total gas volume was multiplied with the measured CH₄ content (%) at the time of the sampling.

The accumulated CH₄ production (Nml) was then calculated by summing the respective samples measured values of total CH₄ volume at standard conditions.

Lastly, the specific CH₄ production was calculated by dividing the accumulated CH₄ production with the mass of the substrate (gVS). The mass of the substrate was calculated by multiplying the volume of substrate added (ml) with its VS (%), and then dividing the result by 100.

3. Results

3.1 Volatile solids, pH and FOS/TAC

At the beginning of the project, all samples had a high content of VS which then decreased during the study (Table 2). The cattle manure had the highest amount with 82.4%, Lövsta digestate thereafter with 75.8% and More Biogas digestate lastly with 72.2%. The reduction of VS was modest for most of the samples, except for the cattle manure sample with added $(\text{NH}_4)_2\text{SO}_4$ which decreased from 82.4% to 75.5%.

Table 2. Respective samples dry matter content (TS%), glow residue (%) and volatile solids (VS%) mean values of the three replicates at the beginning and end (Day 29) of the study. “” stands for the sampling at Day 29. “AS” stands for ammonium sulphate*

Sample	Mean TS (%)	Mean glow residue (%)	Mean VS (% of TS)	Standard deviation of VS (±)
More Biogas	5.7	27.8	72.2	0.24
*More Biogas control	5.0	31.0	69.0	0.14
*More Biogas AS	6.5	30.9	69.1	0.21
Lövsta digestate	6.4	24.2	75.8	0.12
*Lövsta digestate control	5.9	26.1	73.9	0.28
*Lövsta digestate AS	7.2	27.0	73.0	0.16
*Lövsta digestate quadrupled amount AS	8.3	24.0	76.0	0.15
Cattle manure	17.6	17.6	82.4	0.10
*Cattle manure control	5.3	20.7	79.3	0.21
*Cattle manure AS	5.9	24.5	75.5	0.27

The cattle manure also had the biggest change in pH during the study (Table 3). At the beginning, pH was measured to 7.5 in the manure and at the end of the project, pH was measured to 6.8 for the sample with added $(\text{NH}_4)_2\text{SO}_4$ and 7.1 for the control sample. Lövsta digestate and More Biogas digestate did not have as large of a change in pH, all but one samples had a small decrease in pH. The Lövsta sample with a quadrupled amount of $(\text{NH}_4)_2\text{SO}_4$ had a higher pH at the end of the study.

Table 3. pH and mean of FOS/TAC at the beginning and end (Day 29) of the study. “*” stands for the sampling at Day 29. “AS” stands for ammonium sulphate

Sample	pH	Mean FOS/TAC
Lövsta digestate	7.8	0.15
*Lövsta digestate control	7.5	0.17
*Lövsta digestate AS	7.6	0.19
*Lövsta digestate quadrupled amount AS	7.9	0.18
More Biogas	7.9	0.23
*More Biogas control	7.5	0.22
*More Biogas AS	7.7	0.26
Cattle manure	7.5	0.87
*Cattle manure control	7.1	0.88
*Cattle manure AS	6.8	1.35

The value of the measured FOS/TAC for the respective digestate was under 0.3, which indicated stable conditions, both at the beginning and end of the study. The cattle manure sample had a measured value of FOS/TAC >0.5, showing a clear instability in the system. With added $(\text{NH}_4)_2\text{SO}_4$, it had an increase from 0.87 to 1.35, which indicated that volatile fatty acids (VFAs) had accumulated. It also had an increased accumulation of VFAs, especially for the sample with added $(\text{NH}_4)_2\text{SO}_4$, which had a significant accumulation of CH_3COO^- and went from 1.8 g/L to 7.6 g/L (Table 4). More Biogas digestate had an increase in CH_3COO^- in the sample with added $(\text{NH}_4)_2\text{SO}_4$ and a decrease in the control sample. Lövsta digestate had no results during the VFA measurement.

Table 4. The composition of volatile fatty acids in respective sample at the beginning and end (Day 29) of the study. “*” stands for the sampling at Day 29. “AS” stands for ammonium sulphate

Sample	acetate g/L	prop g/L	i-buty g/L	buty g/L	i-val g/L	valerat g/L	Total g/L
Lövsta digestate							0
*Lövsta digestate control							0
*Lövsta digestate AS							0
*Lövsta digestate quadrupled amount AS							0
More Biogas	0.4	0.1		0	0		0.5
*More Biogas control	0.2						0.2
*More Biogas AS	0.7						0.7
Cattle manure	1.8	0.5	0	0.2	0.1	0.1	2.7
*Cattle manure control	2.5	2.5	0.2	0.8	0.3	0.2	6.5
*Cattle manure AS	7.6	2.1	0.3	1.2	0.4	0.2	11.6

3.2 Nutrient content

Since More Biogas digestate had ferric chloride added to it, the result for the nutrient content showed a considerable amount of iron in this digestate, 10 841 mg/kgTS compared to the Lövsta digestate and cattle manure, which had 1608 mg/kgTS and 778 mg/kgTS respectively (Table 5). The More Biogas digestate also had a higher amount of available NH₄-N than the other samples. Compared to the two digestates, the cattle manure had a higher amount of Total-carbon, being 29 kg/ton compared to Lövsta digestate and More Biogas which had 25.8 kg/ton and 22.2 kg/ton respectively. Regarding sulphur content, More Biogas had the highest amount with 0.52 kg/ton and thereafter Lövsta digestate with 0.41 kg/ton. The cattle manure had the lowest amount of sulphur with 0.39 kg/ton.

Table 5. Results of nutrient content of respective fertilizer from AgriLab, Uppsala, at the beginning of the study

Sample	Cattle manure	Lövsta digestate	More Biogas digestate
Dry matter, TS (%)	6.6	6.3	5.6
Tot-nitrogen (kg/ton)	3.2	4.6	4.8
Organic nitrogen (kg/ton)	1.8	2	2
Ammonium nitrogen (NH ₄ -N) (kg/ton)	1.4	2.6	2.8
Tot-carbon (kg/ton)	29	25.8	22.2
Tot-C/Tot-N	9.2	5.6	4.6
Total phosphorus (kg/ton)	0.5	0.87	0.73
Total potassium (kg/ton)	2.4	3.4	3.4
Total magnesium (kg/ton)	0.52	0.67	0.5
Total calcium (kg/ton)	1.3	1.4	1.4
Total sodium (kg/ton)	0.49	0.42	0.74
Total sulphur (kg/ton)	0.39	0.41	0.52
Total Copper (mg/(kgTS))	309	194	204
Total Iron (mg/(kgTS))	778	1608	10814
Total Manganese (mg/(kgTS))	208	318	512
Total Zinc (mg/(kgTS))	191	308	407

Unfortunately, the results of the nutritional composition the samples had at day 29, after (NH₄)₂SO₄ was added, was not received before this essay was submitted. However, NH₄-N analysis performed by staff at RISE showed an increase in NH₄-N in all samples, where the increase corresponds almost to the added amount of (NH₄)₂SO₄ added (Table 6). The control samples also had a minor increase of NH₄-N.

Table 6. The amount of NH₄-N in respective sample at the beginning and end (Day 29) of the study. “*” stands for the sampling at Day 29. “AS” stands for ammonium sulphate

Sample	NH ₄ -N (kg/ton)
Cattle manure	1,4
*Cattle manure control	1,96
*Cattle manure AS	3,57
Lövsta digestate	2,6
*Lövsta digestate control	2,88
*Lövsta digestate AS	6,22
*Lövsta digestate quadrupled amount AS	8,53
More biogas	2,8
*More biogas control	3,19
*More biogas AS	6,24

3.3 The methane and hydrogen sulphide emissions

3.3.1 More Biogas

During the 29 days of the study, the More Biogas sample with added (NH₄)₂SO₄ specific CH₄ production went from 6.6 Nml/gVS to 91.7 Nml/gVS, while the control sample went from 7.4 Nml/gVS to 345.4 Nml/gVS (Figure 1). This means that the CH₄ production in the control sample was almost four times bigger (statistically significant) than in the sample with added (NH₄)₂SO₄. Compared to the control sample, the test sample CH₄ production was 73.5% less at the end of the study.

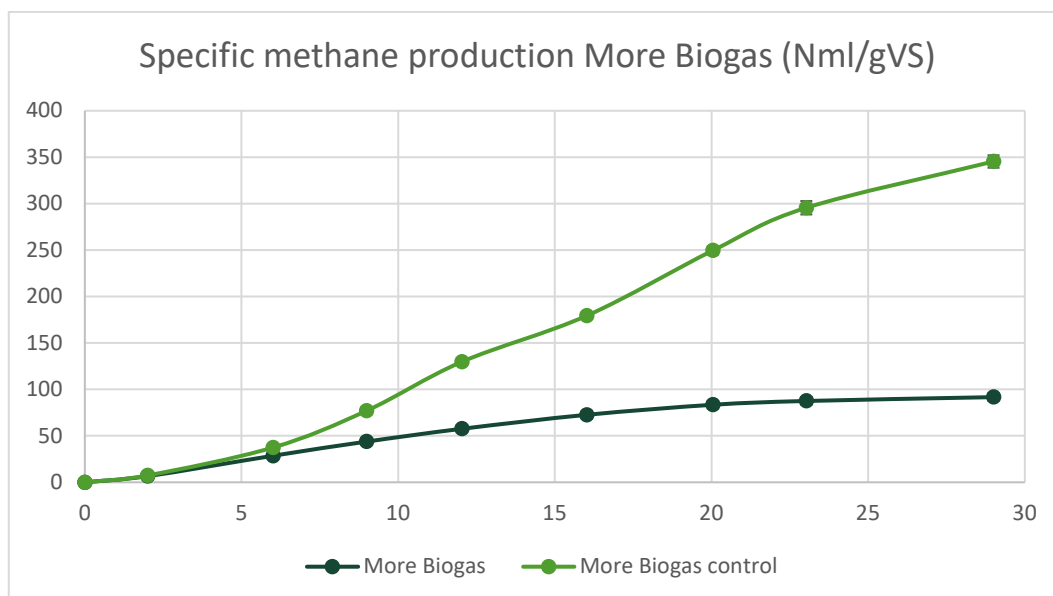


Figure 1. Accumulated values for specific CH_4 production (Nml/gVS, N standing for normal conditions) for More Biogas digestate with and without (control) added ammonium sulphate.

The standard deviation at the beginning of the study for the sample with added $(NH_4)_2SO_4$ was ± 0.2 and at the end ± 4.0 (Table 6). For the control sample it was ± 1.5 at the beginning of the study and ± 6.7 at the end.

Table 7. Accumulated values for specific CH_4 production (ml/gVS) for More Biogas digestate with the standard deviation

Days of the study	2	6	9	12	16	20	23	29
More Biogas AS	6.6 \pm 0.2	28.6 \pm 0.4	43.8 \pm 2.7	57.6 \pm 3.7	72.7 \pm 4.4	83.5 \pm 4.2	87.6 \pm 4.1	91.7 \pm 4.0
More Biogas control	7.4 \pm 1.5	37.4 \pm 2.0	77.0 \pm 3.7	129.7 \pm 1.8	179.4 \pm 3.0	249.6 \pm 4.5	295.6 \pm 7.2	345.4 \pm 6.7

At the same time, the H_2S content increased in the sample with added $(NH_4)_2SO_4$. From a low production of 4 ppm at day 6, the test sample reached the highest value that the Biogas5000 equipment could obtain, namely 5000 ppm, at day 16 (Figure 2) and maintained the production till the end of the project. The control sample kept a low and stable H_2S production during the study, 370 ppm being the highest measured value at day 23. This means that the sample with added $(NH_4)_2SO_4$ almost had 14 times bigger H_2S emission than the control sample.

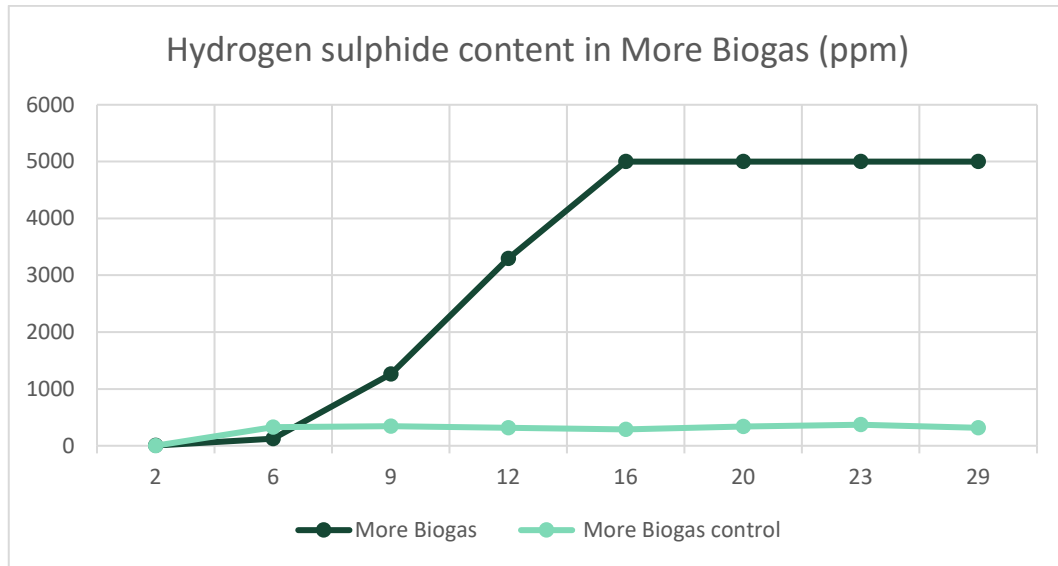


Figure 2. H₂S content for More Biogas digestate with and without (control) addition of ammonium sulphate.

3.3.2 Lövsta digestate

The specific CH₄ production for Lövsta digestate was consistent in its increase. For the control sample, the CH₄ production was significantly higher than for the test samples. It went from a production of 6.3 Nml/gVS at day 2 in the study to a production of 267.8 Nml/gVS at the end (Figure 3), almost a double amount of CH₄ production compared to the test samples. The test sample that had a quadrupled amount of (NH₄)₂SO₄ added to it had the lowest production of CH₄ and went from a production of 4.6 Nml/gVS at day 2 to a production of 114.9 Nml/gVS at day 29. The test sample that had a double amount of (NH₄)₂SO₄ added to it had a very similar production (differences were not significant) and went from a production of 5.9 Nml/gVS at day 2 to a production of 129.7 Nml/gVS at the end of the project. Compared to the control sample, the test sample with a double amount of (NH₄)₂SO₄ had a CH₄ production that was 51.6% less at the end of the study. The test sample with a quadrupled amount of (NH₄)₂SO₄ had a CH₄ production that was 57.7% less compared to the control sample at the end of the study.

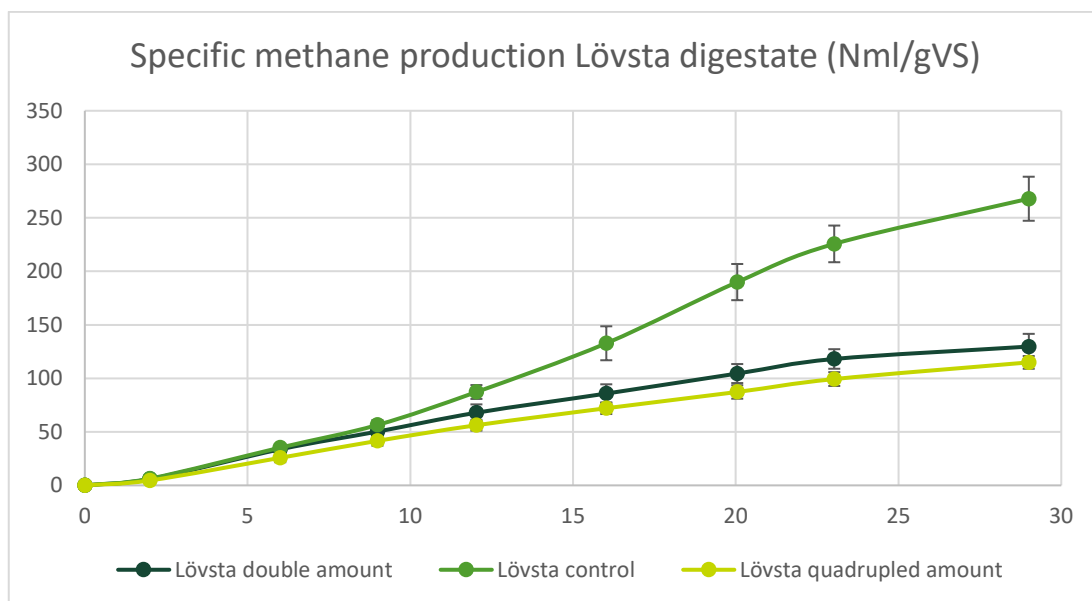


Figure 3. Accumulated values for specific CH_4 production (Nml/gVS, N standing for normal conditions) for Lövsta digestate with and without (control) added $(NH_4)_2SO_4$ in double and quadrupled amounts.

The standard deviation at the beginning of the study for the control sample was ± 0.0 and at the end ± 20.6 (Table 7). For the test sample with a double amount of $(NH_4)_2SO_4$, the standard deviation at the beginning of the study was ± 1.7 and for the sample with a quadrupled amount ± 2.2 . At day 29, the standard deviation was ± 11.9 for the sample with double amount and ± 5.9 , for sample with quadrupled amount.

Table 8. Accumulated values for specific CH_4 production (ml/gVS) for Lövsta digestate with the standard deviation

Days of the study	2	6	9	12	16	20	23	29
Lövsta digestate double amount AS	5.9 \pm 1.7	33.7 \pm 3.4	50.2 \pm 6.7	67.9 \pm 7.8	85.8 \pm 8.7	104.5 \pm 8.9	118.1 \pm 9.1	129.7 \pm 11.9
Lövsta control	6.3 \pm 0.0	35.3 \pm 1.2	56.4 \pm 4.6	87.2 \pm 6.5	132.8 \pm 15.9	189.9 \pm 16.9	225.6 \pm 17.1	267.8 \pm 20.6
Lövsta digestate quadrupled amount AS	4.6 \pm 2.2	25.7 \pm 4.1	41.6 \pm 4.5	56.2 \pm 5.0	72.1 \pm 5.4	87.3 \pm 6.3	99.3 \pm 6.4	114.9 \pm 5.9

The H_2S content increased steadily during the study for both test samples (Figure 4). The control sample reached its peak day 12 with 971 ppm and during the following measurements stayed stable at around 890 ppm. At day 23 it looked like the production of H_2S would start to decrease for the test samples, but at day 29 the highest amount of H_2S was measured, 2522 ppm for the test with an double amount of $(NH_4)_2SO_4$ and 2236 ppm for the test with a quadrupled amount.

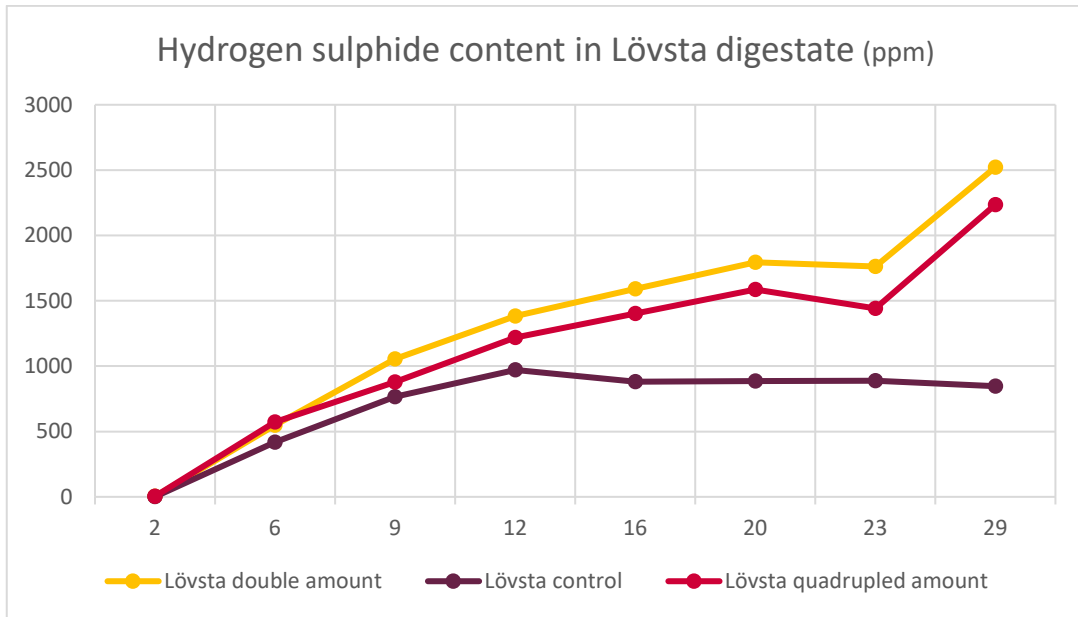


Figure 4. H_2S content for Lövsta digestate with and without (control) addition of ammonium sulphate.

3.3.3 The cattle manure

The cattle manure specific CH_4 production was relatively low for both the control sample and the test sample during the study, although the control sample had from day 6 a higher CH_4 production than the sample with added $(NH_4)_2SO_4$ (Figure 5). Between day 23 and day 29 the production increased significantly; the control sample went from a specific CH_4 production of 50 Nml/gVS to 164.5 Nml/gVS. The sample with added $(NH_4)_2SO_4$ increased gradually during the project and went from a production of 0.9 Nml/gVS at day 2 to the highest produced value day 29, 18.3 Nml/gVS. Compared to the control sample, the test sample CH_4 production was 88.9% less at the end of the study.

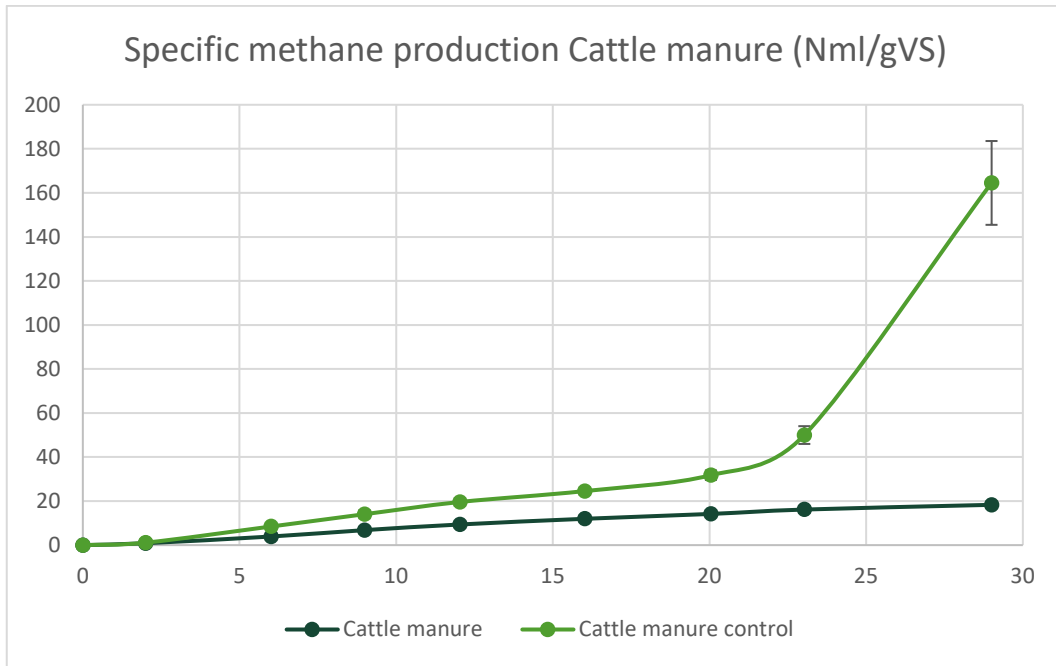


Figure 5. Accumulated values for specific CH_4 production (Nml/gVS, N standing for normal conditions) for the cattle manure with and without (control) added ammonium sulphate.

The standard deviation at the beginning of the study for the sample with added $(NH_4)_2SO_4$ was ± 0.2 and at the end ± 1.2 (Table 8). For the control sample, the standard deviation was ± 0.3 in the beginning and ± 19.0 in the end.

Table 9. Accumulated values for specific CH_4 production (ml/gVS) for the cattle manure with the standard deviation

Days of the study	2	6	9	12	16	20	23	29
Cattle manure AS	0.9 \pm 0.2	3.9 \pm 0.3	6.8 \pm 0.8	9.4 \pm 1.0	12 \pm 1.3	14.2 \pm 1.4	16.2 \pm 1.6	18.3 \pm 1.2
Cattle manure control	1.1 \pm 0.3	8.4 \pm 0.8	14 \pm 1.9	19.6 \pm 1.6	24.6 \pm 1.6	31.8 \pm 2.2	50 \pm 4.0	164.5 \pm 19.0

From day 6 the sample with added $(NH_4)_2SO_4$ reached the highest measuring value that Biogas5000 could obtain, namely 5000 ppm, and the H_2S production did not decrease during this study (Figure 6). The control sample had a consistent increase in its H_2S production and did also reach 5000 ppm as its highest value at day 16. At day 29 the production did decrease with the measured value being 4637 ppm.

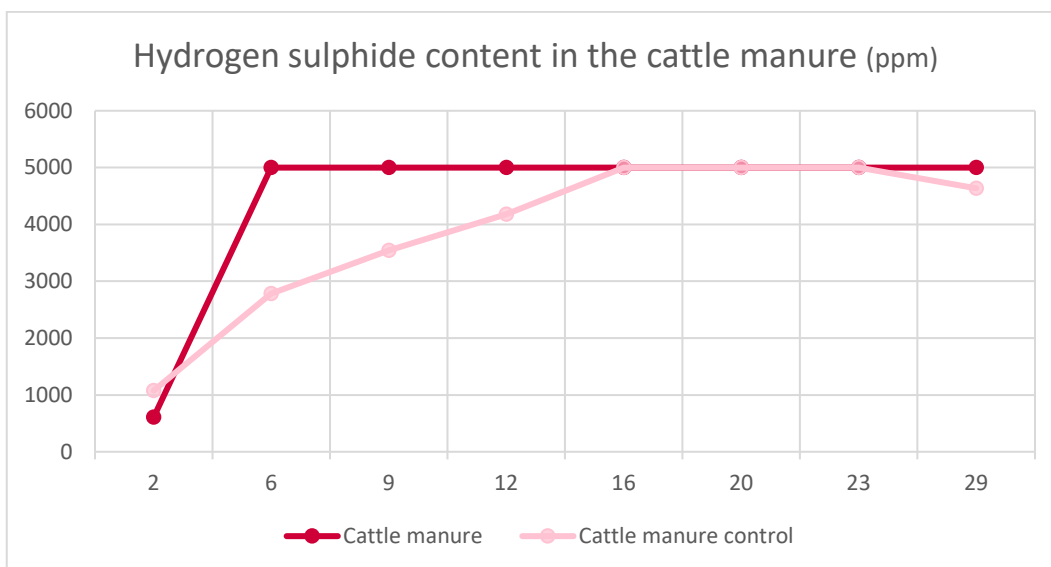


Figure 6. H₂S content for the cattle manure with and without (control) addition of ammonium sulphate.

3.4 Anova analysis

The Anova analysis showed that the difference between Lövsta digestate with the double amount and with the quadruple amount of (NH₄)₂SO₄ did not have a significant difference (Table 9). Same goes for Lövsta digestate with a quadruple amount of (NH₄)₂SO₄ and the sample of More Biogas with added (NH₄)₂SO₄. The other test samples did have a statistically significant difference when compared to respective control sample.

Table 10. The significant difference between the mean values of respective sample specific CH₄ production. Levels connected with the same letter are not significantly different. Levels not connected with the same letter are significantly different

Level		Mean
More Biogas control	A	345.40211
Lövsta digestate control	B	267.81782
Cattle manure control	C	164.50969
Lövsta digestate double amount AS	D	129.67484
Lövsta digestate quadruple amount AS	D E	114.91173
More Biogas AS	E	91.69997
Cattle manure AS	F	18.30129

4. Discussion

The results of this study were conclusive - when $(\text{NH}_4)_2\text{SO}_4$ was added to the respective fertilizer, CH_4 emission was reduced compared to the control samples. The samples containing $(\text{NH}_4)_2\text{SO}_4$ did have some CH_4 emission, although minuscule. At the same time, H_2S emission was greater for all the samples containing $(\text{NH}_4)_2\text{SO}_4$ compared to the control samples.

4.1 Ammonium sulphate's effect on More Biogas digestate

In the results for More Biogas digestate, the difference in CH_4 emission was statistically significant when comparing the sample containing $(\text{NH}_4)_2\text{SO}_4$ with the control sample, being 73.5% less in the test sample compared to the control sample at the end of the study. This reduction is slightly higher than the result from a previous study where CH_4 emission was reduced by 60-70% (Denier van der Gon et al. 2001). This could indicate that the methanogens are being efficiently outcompeted by SRB for existing substrates in the digestive when $(\text{NH}_4)_2\text{SO}_4$ is added, as shown in previous studies (Jarvis et al. 2009; Lindau et al. 1993; Denier van der Gon 2001).

More Biogas digestate had ferric chloride added to it, a substance intended to decrease the H_2S emission during the digestion process in the biogas plant by making sulphur less accessible for SRB (Edström et al. 2013; Zhou et al. 2016). In this study, the emission of H_2S increased significantly when $(\text{NH}_4)_2\text{SO}_4$ was added to the More Biogas digestate. Compared to the Lövsta digestate with no ferric chloride, the H_2S emission was much higher. There could be several reasons for this. Previous studies have shown that methanogens active in a thermophilic biogas plant are more sensitive to temperature changes than those found in mesophilic biogas plants (Jarvis et al. 2009). This suggests that the methanogens in the More Biogas digestate may have been inhibited by the temperature change that this study entailed. Combined with the fact that SRB are usually more dominant in mesophilic environments than methanogens (Chen et al. 2007), SRB might have had greater access to available substrates, resulting in higher H_2S emissions.

Another reason could be the difference in the composition of the digestates, More Biogas contained poultry, cattle, and swine manure, while Lövsta contained solely cattle and swine manure. The nutrient content analysis showed that More Biogas had a higher amount of sulphur content than both Lövsta digestate and the cattle manure, which could have affected the H₂S emission results. However, this is speculative, as the composition of the different digestates was not thoroughly studied due to limitations in this study.

Considering that the prevailing concentration of ferric chloride in the fertilizer is intended for a lower concentration of sulphur and not an increase as the addition of (NH₄)₂SO₄ entailed, the increased H₂S emission could perhaps be due to the iron becoming unavailable in the form of some precipitation during the study (Edström et al. 2013). In the control sample, the H₂S emission remained very low and stable, likely due to the buffering effect of ferric chloride. This contrasts with the Lövsta digestate control sample, where H₂S emission increased over time and became less stable.

Although the result showed an increase in H₂S emissions for More Biogas test sample, the accumulation of VFA and the measured pH indicated that the system maintained a stable buffering capacity in both the control sample and the sample with added (NH₄)₂SO₄. This since the measured VFA and pH did not vary greatly between the different measurements, indicating that although the test with added (NH₄)₂SO₄ led to some VFA accumulation, the system could buffer for this change.

4.2 Ammonium sulphate's effect on Lövsta digestate

The pH values of the Lövsta digestate control and test samples did not vary greatly from the beginning to the end of the study. This suggests that the digestate was able to buffer for the addition of (NH₄)₂SO₄ in the test samples. However, no results were obtained for the VFA composition in the sample, preventing further detailed commentary on this aspect.

Regarding specific CH₄ production, the result showed that the test sample with a quadrupled amount of (NH₄)₂SO₄ had the lowest CH₄ emission among the Lövsta digestates samples. Since this sample had the highest amount of (NH₄)₂SO₄ added, it was expected to have the highest amount of H₂S emission. This since according to previous studies, an abundance of available sulphur compounds should enable SRB to outcompete methanogens more effectively, resulting in lower CH₄ emission and higher H₂S emission (Lindau et al. 1993; Denier van der Gon 2001). In this case, the highest H₂S emission came from the sample with a double amount of (NH₄)₂SO₄. Nevertheless, there was no statistically significant difference in CH₄

emission between the two test samples, indicating that any measured difference is likely insignificant.

The CH₄ production in the samples containing (NH₄)₂SO₄ was not reduced to the same extent as reported in previous studies, which showed a reduction of 60-70% (Denier van der Gon et al. 2001). Instead, the sample with the double amount of (NH₄)₂SO₄ added had a CH₄ production that was 51.6% less than the control sample, while the sample with a quadrupled the amount had a reduction of 57.7% compared to the control sample. This difference could be attributed to the relatively high VS content of the Lövsta digestate at 75.8%. Previous studies have shown that materials with a high VS content typically contain more available degradable materials than those with a low VS content, leading to a higher gas exchange per unit volume (Swedish Waste Management 2009). Thus, the higher availability of degradable materials in Lövsta digestate may have allowed methanogens and SRB to efficiently compete in the samples, resulting in only a minor reduction in CH₄ production. The sample with a quadrupled amount of (NH₄)₂SO₄ showed a more significant reduction in CH₄ production than the sample with a double the amount, suggesting that for materials with high VS content, a larger amount of (NH₄)₂SO₄ is required to achieve a greater reduction in CH₄ production.

Similar to the cattle manure and More Biogas digestate, samples containing (NH₄)₂SO₄ exhibited reduced CH₄ emission along with increased H₂S emission compared to the control sample. This suggests that SRB could efficiently limit the available substrate in the digestate, thereby inhibit the methanogens to some extent.

4.3 Ammonium sulphate's effect on the cattle manure

The cattle manure with added (NH₄)₂SO₄ exhibited a clear accumulation of VFA, which affected both pH and FOS/TAC results measured on the last day of the study. Previous studies have shown that an increase in VFA often results in a lower pH value (Jarvis et al. 2009), a trend also observed in this study. The test sample went from a total VFA composition of 2.7 g/L and a pH of 7.5 without (NH₄)₂SO₄ added to it to 11.6 g/L and a pH of 6.8 after (NH₄)₂SO₄ was added.

The accumulated VFA primarily consisted of CH₃COO⁻. Since CH₃COO⁻ is one of the substrates that methanogens and SRB compete for, these results indicate an instability in the system in which the microbial organisms active in methanogenesis do not utilize the available substrates to the same extent as they are produced during acetogenesis. This was also confirmed by the FOS/TAC measurement. Initially, the sample already showed clear instability with a FOS/TAC ratio >0.5 before any (NH₄)₂SO₄ was added. This instability appeared more pronounced after (NH₄)₂SO₄

was added, as the control sample exhibited some buffering effects, with minimal variations in pH, accumulated VFA, or FOS/TAC from the beginning to the end of the study. When the $(\text{NH}_4)_2\text{SO}_4$ was added, this buffering capacity seemed disturbed, resulting in an acidification process and a decrease in pH. However, FOS/TAC measurements are typically conducted in an active biogas plant with regular substrate inflow, meaning that the measured FOS/TAC value in this study may not fully represent the dynamics in the process due to the lack of continuous substrate supply.

The H_2S emission was high for both samples of the cattle manure, as expected given that the manure was undigested. Previous studies have shown that due to the degradation of sulphur-containing amino acids, undigested manure can exhibit high H_2S emission (Ibid.). The cattle manure contained a substantial amount of organic matter, including sulphur-containing amino acids, which was reflected in its nutritional content with the sample containing the highest amount of Total-carbon. This means that smaller chemical compounds like CH_3COO^- and H_2 were not available in a larger concentration in the beginning. As the study progressed, larger organic materials were gradually decomposed, leading to the high H_2S emission observed. This also explains the significant increase in specific CH_4 production observed between Days 23 and 29, which was later compared to the more continuous emissions observed in the two digestates. By Day 23, enough larger chemical compounds had decomposed, enabling increased activity of methanogens in the sample. This is also confirmed in the measured H_2S emission, which decreased between the two sampling occasions, indicating more efficient competition by methanogens against SRB.

The sample containing $(\text{NH}_4)_2\text{SO}_4$ did not exhibit the same development in CH_4 emissions as the control sample. The test sample showed consistently low specific CH_4 production throughout the test period and did not peak in the same manner as the control sample, suggesting that the addition of $(\text{NH}_4)_2\text{SO}_4$ had an impact on emissions. Compared to the control sample, the test sample showed an 88.9% reduction in its CH_4 emission, significantly higher than in previous studies reporting reductions of 60-70% (Denier van der Gon et al. 2001). This might indicate that SRB effectively limited available substrates or, as shown in a previous study by Zhou et al. (2009), that the methanogens were inhibited by the high and toxic concentrations of H_2S . The sample with $(\text{NH}_4)_2\text{SO}_4$ reached the highest measured H_2S emission in this study at Day 6, the earliest among all organic fertilizer samples, indicating that the effect $(\text{NH}_4)_2\text{SO}_4$ had took place relatively early in the anaerobic digestion. Consequently, methanogens in the sample were exposed to H_2S for a longer duration than the methanogens in the other samples, potentially reducing CH_4 emissions due to gas toxicity.

Another notable result was the relatively large reduction in VS observed in the $(\text{NH}_4)_2\text{SO}_4$ test sample, decreasing from 82.4% to 75.5% compared to the control sample, which decreased to 79.3% VS content. This suggests that the sample with $(\text{NH}_4)_2\text{SO}_4$ underwent greater decomposition than the control sample. The reasons for this were not explored in this study, but the addition of $(\text{NH}_4)_2\text{SO}_4$ may have facilitated greater degradation due to its acidifying properties, with a pH of 5.28.

4.4 Final conclusions and suggestions for further research

The overall aim of the project, of which this study was part of, was to examine the possibilities of obtaining an organic fertilizer with high plant nutritional value while minimizing greenhouse gas emissions by adding $(\text{NH}_4)_2\text{SO}_4$ to digestates or livestock manures. Due to limitations in this study, specific plant nutrition content was not analyzed. However, based on $\text{NH}_4\text{-N}$ analysis results, the NH_4 content did increase in the fertilizers when $(\text{NH}_4)_2\text{SO}_4$ was added, which is expected given the addition of NH_4 -containing substances. Further research is needed to determine if the addition of $(\text{NH}_4)_2\text{SO}_4$ indeed enhances the overall plant nutritional value in an organic fertilizer and to assess which crops this type of fertilizer would be suitable for.

Regarding CH_4 emissions, this study's results demonstrated that adding $(\text{NH}_4)_2\text{SO}_4$ to the respective fertilizer reduced CH_4 emission. Therefore, adding $(\text{NH}_4)_2\text{SO}_4$ appears to be a method to mitigate CH_4 emissions during storage of digestates or stable manures, if they are being stored in mesophilic temperatures. Although, controlled temperatures in a livestock manure storage facility are rarely the case and usually the manure is stored during a longer period than 29 days. This means that to fully understand the effects of $(\text{NH}_4)_2\text{SO}_4$, further research needs to replicate farm conditions more accurately, including representative temperature profiles and longer study durations to monitor CH_4 emissions.

Adding $(\text{NH}_4)_2\text{SO}_4$ increases the risk of forming dangerous H_2S gas. This aspect also requires further investigation under realistic conditions, with measurements that can put the emission in relation to the risks of handling the fertilizer.

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