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Valorização do lodo excedente do tratamento de esgoto e de dejeto bovino através da co-digestão anaeróbia e recuperação de amônia

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O presente trabalho em nível de Mestrado foi avaliado e aprovado, em 17 de Dezembro de 2025, pela banca examinadora composta pelos seguintes membros:

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Certificamos que esta é a versão original e final do trabalho de conclusão que foi julgado adequado para obtenção do título de Mestra em Engenharia Química.

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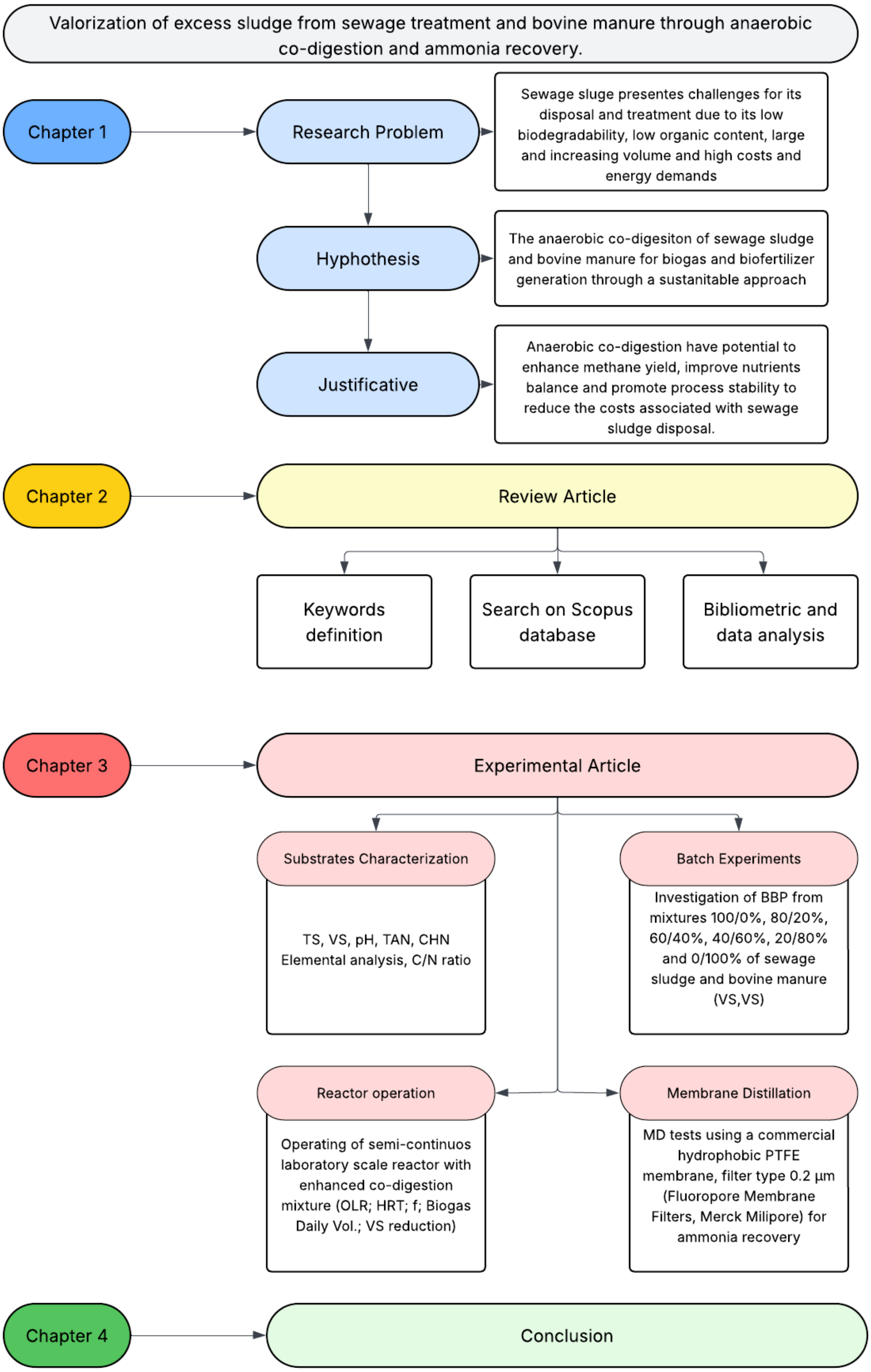
Ninguém ignora tudo. Ninguém sabe tudo. Todos nós sabemos alguma coisa. Todos nós ignoramos alguma coisa. Por isso aprendemos sempre. (Paulo Freire, 1989)

DIAGRAMA CONCEITUAL



Valorization of excess sludge from sewage treatment and bovine manure through anaerobic co-digestion and ammonia recovery.

Chapter 1

Research Problem

Sewage sluge presentes challenges for its disposal and treatment due to its low biodegradability, low organic content, large and increasing volume and high costs and energy demands

Hyphothesis

The anaerobic co-digesiton of sewage sludge and bovine manure for biogas and biofertilizer generation through a sustanitable approach

Justificative

Anaerobic co-digestion have potential to enhance methane yield, improve nutrients balance and promote process stability to reduce the costs associated with sewage sludge disposal.

Chapter 2

Review Article

Keywords definition

Search on Scopus database

Bibliometric and data analysis

Chapter 3

Experimental Article

Substrates Characterization

TS, VS, pH, TAN, CHN Elemental analysis, C/N ratio

Batch Experiments

Investigation of BBP from mixtures 100/0%, 80/20%, 60/40%, 40/60%, 20/80% and 0/100% of sewage sludge and bovine manure (VS,VS)

Reactor operation

Operating of semi-continuos laboratory scale reactor with enhanced co-digestion mixture (OLR; HRT; f; Biogas Daily Vol.; VS reduction)

Membrane Distillation

MD tests using a commercial hydrophobic PTFE membrane, filter type 0.2 µm (Fluoropore Membrane Filters, Merck Milipore) for ammonia recovery

Chapter 4

Conclusion

RESUMO

O crescimento populacional contínuo intensifica desafios globais, como o aumento na demanda por energia renovável e a maior geração de resíduos. Nesse contexto, a digestão anaeróbia se destaca como uma alternativa sustentável para o tratamento de resíduos e produção de combustível renovável, especialmente quando aplicada por meio da co-digestão anaeróbia, que combina diferentes substratos para elevar a produção de biogás, melhorar a estabilidade operacional e otimizar o balanço nutricional. O Brasil apresenta elevado potencial para a co-digestão devido à sua forte base agropecuária, aos grandes volumes de dejetos de bovinos, suínos e avícolas e à expansão das estações de tratamento de esgoto, que produzem consideráveis quantidades de lodo a serem manejados. O presente estudo teve como objetivo investigar a co-digestão anaeróbia do lodo de esgoto sanitário proveniente do tratamento secundário aeróbico com o esterco bovino proveniente de uma fazenda leiteira, a fim de aumentar a produção de biogás e recuperar a amônia resultante no digestato através da destilação por membranas. O Potencial Bioquímico de Produção de Biogás foi avaliado através do ensaio em batelada utilizando o teste de eudiômetros de acordo com a norma VDI 4630 (2006) e o ensaio semi-contínuo foi explorado através de um reator semi-contínuo em escala de bancada com progressão de carga. As frações de mistura investigadas nos ensaios de PBB foram de 100/0%, 80/20%, 60/40%, 40/60%, 20/80% e 0/100% (SV/SV) de lodo de esgoto e esterco bovino, respectivamente. Os parâmetros analisados foram pH, série de sólidos, alcalinidade e acidez e análise CHN. A recuperação de amônia foi analisada através da destilação com membranas utilizando membrana hidrofóbica PTFE na condição natural do pH da amostra e com o pH ajustado para 9,3. A codigestão anaeróbia apresentou desempenho superior à monodigestão, especialmente com a adição de 20% de esterco bovino (VS/VS), que resultou em um aumento de 53,07% no ganho de produção de biogás, alcançando um PBB padronizado de 260,97 ± 23,37 mL gSV^{-1}, em comparação com os 124,79 ± 28,47 mL gSV^{-1} obtidos na digestão isolada do lodo de esgoto. Com base nesses resultados, a mistura 80/20 foi selecionada para o experimento semicontínuo, operado sob cargas orgânicas progressivas de 1,0; 1,5 e 2,0 kgSV m^{-3} d^{-1}, correspondentes aos TRHs de 23, 16 e 14 dias, respectivamente. O reator demonstrou estabilidade operacional em todas as condições aplicadas e atingiu um volume diário máximo de biogás de 816,99 mL/dia, com teor de metano de 75,94%, valor superior ao normalmente observado na monodigestão de lodo. A remoção de sólidos voláteis variou entre 14,35% e 27,99%, destacando-se o estágio intermediário (1,5 kgSV m^{-3} d^{-1}), no qual o processo apresentou maior eficiência de degradação. Na etapa de recuperação de nitrogênio, o sistema de destilação com membranas recuperação máxima de 34,38% de amônia em pH 9,3. De forma geral, o processo demonstrou elevado potencial dentro dos princípios da economia circular, uma vez que integra a produção de biogás com a geração de biofertilizante proveniente do digestato, além de favorecer a recuperação de nitrogênio como insumo agrícola.

Palavras-chave: Biogás; Esterco bovino; Lodo de esgoto; Biofertilizante; Recuperação de amônia.

ABSTRACT

Continuous population growth intensifies global challenges, such as the increasing demand for renewable energy and higher waste generation. In this context, anaerobic digestion stands out as a sustainable alternative for waste treatment and renewable fuel production, especially when applied through anaerobic co-digestion, which combines different substrates to enhance biogas production, improve operational stability, and optimize nutrient balance. Brazil has high potential for co-digestion due to its strong agricultural base, the large volumes of cattle, swine, and poultry manure, and the expansion of wastewater treatment plants, which generate considerable amounts of sludge that require proper management. The aim of this study was to investigate the anaerobic co-digestion of sanitary sewage sludge from secondary treatment with cattle manure from a dairy farm in order to increase biogas production and recover ammonia from the digestate through membrane distillation. The Biochemical Biogas Potential was assessed through batch assays using eudiometer tests according to the VDI 4630 (2006) guideline, and the semi-continuous assay was conducted using a bench-scale reactor operated under progressive organic loading rates. The mixing ratios investigated were 100/0%, 80/20%, 60/40%, 40/60%, 20/80%, and 0/100% (VS/VS) of sewage sludge and cattle manure, respectively. The parameters analyzed were pH, solids content, alkalinity and acidity, and CHN analysis. Ammonia recovery was evaluated through membrane distillation using a hydrophobic PTFE membrane under the natural pH of the sample and with the pH adjusted to 9.3. Anaerobic co-digestion showed superior performance compared to mono-digestion, especially with the addition of 20% cattle manure (VS/VS), which resulted in a 53.07% increase in biogas production, reaching a BBP of 260.97 ± 23.37 mL gVS^{-1}, compared to 124.79 ± 28.47 mL gVS^{-1} obtained in the isolated digestion of sewage sludge. Based on these results, the 80/20 mixture was selected for the semi-continuous experiment, operated under progressive organic loading rates of 1.0, 1.5, and 2.0 kgVS m^{-3} d^{-1}, corresponding to HRTs of 23, 16, and 14 days, respectively. The reactor demonstrated operational stability under all tested conditions and reached a maximum daily volume of 816.99 mL/day, with a methane content of 75.94%, a value higher than typically observed in sludge mono-digestion. Volatile solids removal ranged between 14.35% and 27.99%, with the intermediate stage (1.5 kgVS m^{-3} d^{-1}) showing the highest degradation efficiency. In the nitrogen recovery stage, the membrane distillation system achieved a maximum ammonia recovery of 34.38% at pH 9.3. Overall, the process demonstrated strong potential within circular economy principles, as it integrates biogas production with biofertilizer generation from the digestate and enables nitrogen recovery for agricultural use.

Keywords: Biogas; Sewage Sludge; Livestock manure; Ammonia Recovery.

RESUMO EXPANDIDO

Introdução

O crescimento populacional global e a necessidade de ampliar a produção de alimentos impõem desafios significativos para a gestão sustentável dos sistemas produtivos. O Brasil destaca-se como um dos maiores produtores e exportadores de carne bovina e de frango do mundo, o que resulta na geração de grandes volumes de resíduos orgânicos, especialmente esterco bovino. O manejo inadequado desses resíduos pode causar impactos ambientais, como emissões de gases de efeito estufa, eutrofização de corpos d'água e contaminação ambiental. Nesse contexto, a digestão anaeróbia (DA) surge como uma alternativa promissora para a valorização desses resíduos, permitindo a produção de biogás e a recuperação de nutrientes. Paralelamente, o aumento da população também eleva a geração de lodo de esgoto nas estações de tratamento de águas residuárias, cujo manejo representa custos operacionais significativos. A co-digestão anaeróbia entre lodo de esgoto e esterco bovino pode melhorar o balanço nutricional do processo e aumentar a produção de biogás por meio de efeitos sinérgicos. Além disso, tecnologias de separação por membranas, como a destilação por membranas, apresentam potencial para recuperar amônia do digestato na forma de sais de amônio, possibilitando sua utilização como fertilizante. Entretanto, ainda existem poucos estudos que avaliem a integração entre co-digestão anaeróbia desses resíduos e a recuperação de amônia por processos de membranas.

Objetivo

Investigar o efeito da co-digestão anaeróbia de lodo de esgoto proveniente de tratamento secundário com esterco bovino em diferentes proporções na produção de biogás, bem como avaliar a viabilidade técnica da recuperação de amônia do digestato por meio de destilação por membranas visando a produção de biofertilizantes.

Materiais e Métodos

O esterco bovino foi coletado em uma fazenda leiteira localizada em Vacaria (RS, Brasil), enquanto o lodo biológico foi obtido de uma estação de tratamento de esgoto operada pela CASAN em Florianópolis (SC, Brasil). Os inóculos utilizados foram provenientes de uma lagoa anaeróbia adaptada à degradação de esterco bovino e de um digestor anaeróbio da estação de tratamento de esgoto. Para os experimentos de co-digestão anaeróbia, foi utilizada uma mistura de ambos os inóculos na proporção 50:50 (v/v).

Inicialmente foram realizados ensaios em batelada de acordo com a norma VDI 4630 (2006) para determinar a proporção ideal entre os substratos. Os digestores apresentavam volume de trabalho de 125 mL e foram mantidos em condições mesofílicas (37 ± 2 °C). As misturas de lodo de esgoto e esterco bovino foram avaliadas nas proporções (VS/VS) de 0/100, 20/80, 40/60, 60/40, 80/20 e 100/0. Os experimentos foram realizados em triplicata, sendo monitorada a produção diária de biogás através de sistema eudiométrico. Os resultados foram corrigidos para condições padrão de temperatura e pressão.

Com base nos resultados obtidos nos ensaios em batelada, foi operado um reator semi-contínuo em escala de bancada com volume de trabalho de 1,5 L, alimentado com uma mistura de 80% de lodo de esgoto e 20% de esterco bovino (VS/VS). O reator foi mantido em condições mesofílicas (37 ± 1 °C) e operado em três cargas orgânicas volumétricas (OLR): 1,0; 1,5 e 2,0 kgSV m^{-3} d^{-1}, com diferentes tempos de retenção hidráulica.

Para a recuperação de amônia, o digestato foi submetido a experimentos de destilação por membranas utilizando membranas hidrofóbicas de PTFE (0,2 µm). O processo ocorreu com solução de ácido sulfúrico (0,05 M) como fase receptora. Foram avaliadas duas condições de pH: pH natural do digestato e pH ajustado para 9,3 com NaOH. O processo foi conduzido durante três horas a 38 °C, com circulação da solução por bomba peristáltica.

Resultados e Discussão

A caracterização dos substratos indicou que o esterco bovino apresentou maior concentração de sólidos totais e sólidos voláteis quando comparado ao lodo de esgoto. Ambos os resíduos apresentaram pH próximo da neutralidade, condição favorável para a digestão anaeróbia. O esterco bovino também apresentou maior concentração de nitrogênio amoniacal, porém os valores permaneceram abaixo da faixa crítica de inibição relatada na literatura.

Nos ensaios em batelada, observou-se aumento no potencial bioquímico de biogás (BBP) com o aumento da proporção de esterco bovino. A digestão do esterco puro apresentou o maior BBP (353,32 ± 12,92 mL N biogás gSV^{-1}). Entretanto, todas as misturas avaliadas apresentaram efeito sinérgico positivo na co-digestão. A mistura contendo 80% de lodo de esgoto e 20% de esterco bovino apresentou o maior ganho percentual de biogás (53,07%), sendo selecionada para os experimentos em regime semi-contínuo.

A redução de sólidos voláteis foi significativamente maior nos experimentos de co-digestão (42–55%) em comparação com a monodigestão (34–36%), evidenciando a melhoria da biodegradabilidade do sistema. No reator semi-contínuo, operado por 95 dias, o sistema apresentou estabilidade operacional nas três cargas orgânicas aplicadas. O aumento da carga orgânica resultou em maiores taxas de produção de biogás, alcançando produção média diária de até 816,99 ± 237,25 mL na maior OLR aplicada.

Durante a operação do reator, o pH permaneceu dentro da faixa ideal para a digestão anaeróbia (6,5–7,5), enquanto a razão acidez/alcalinidade manteve-se entre 0,22 e 0,28, indicando estabilidade do processo. A redução de sólidos voláteis atingiu valores máximos de aproximadamente 28%, superiores aos relatados em estudos que utilizam apenas lodo de esgoto.

Nos experimentos de destilação por membranas, a recuperação de amônia foi fortemente influenciada pelo pH do digestato. Em pH natural (~7,9), a recuperação foi de aproximadamente 6,05%. Quando o pH foi ajustado para 9,3, a recuperação aumentou para 34,38%, evidenciando o efeito do ambiente alcalino na conversão de amônio em amônia gasosa e na eficiência de separação pela membrana. O processo resultou na formação de sulfato de amônio na solução receptora, composto que pode ser utilizado como fertilizante líquido.

Conclusões

A digestão anaeróbia apresenta grande potencial para a valorização de resíduos orgânicos e para o avanço de sistemas sustentáveis de gestão de resíduos no contexto da bioeconomia circular no Brasil. Os resultados experimentais demonstraram efeito sinérgico na co-digestão de lodo de esgoto com esterco bovino, com aumento de 53,07% na produção de biogás em relação aos valores teóricos. A operação do reator semi-contínuo indicou boa estabilidade do processo, alcançando

produção diária de biogás de até 816,99 ± 237,25 mL e teor de metano de 75,94%. Além disso, o processo de destilação por membranas possibilitou a recuperação de aproximadamente 34% da amônia presente no digestato em pH 9,3, evidenciando o potencial de integração entre produção de energia e recuperação de nutrientes. Apesar dos resultados promissores, ainda existem desafios técnicos e econômicos para a aplicação em larga escala, destacando-se a necessidade de estudos adicionais que integrem avaliação ambiental e econômica, como análises de ciclo de vida, para apoiar a implementação dessa tecnologia em sistemas reais.

Palavras-chave: Biogás; Esterco bovino; Lodo de esgoto; Biofertilizante; Recuperação de amônia.

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1. CHAPTER 1

Introduction

This chapter introduces the anaerobic digestion context and defines the general and specific objectives.

1.1. INTRODUCTION

According to the United Nations (2019), by 2050, the world population is expected to surpass 9.7 billion, while 2.4 billion people currently lack access to adequate food (FAO, 2021). To ensure that the number of individuals in nutritional poverty does not increase and to meet the demand of a growing population, it is necessary to expand food production while generating the lowest possible environmental impact. Brazil is the second-largest global beef producer, behind only the United States, with around 11 million tons produced, representing 15.8% of global production in 2024 (EMBRAPA, 2024a). It is also the world's leading exporter of cattle, with approximately 2.536 million tons of fresh and processed beef exported in 2024 (MAPA, 2024). Regarding poultry production, Brazil remained the world's largest exporter of chicken meat in 2024, with an estimated 4.7 million tons, accounting for 21.6% of global production (EMBRAPA, 2024b).

However, this production potential generates significant environmental impacts that must be strategically managed. In livestock farming, cattle production is one of the sectors with the least control over environmental pollution, due to extensive farming practices, which result in high greenhouse gas emissions and inadequate management or treatment of animal waste. Nonetheless, this scenario has been changing as the adoption of confined and semi-confined cattle production systems has grown. In 2021, 6.5 million beef and dairy cattle were raised under these conditions in Brazil (Forbes Agro, 2021). Intensive cattle farming is considered a promising alternative as it reduces land use, minimizing the need for deforestation. It also relies on more advanced technologies, enabling higher-quality production through genetic improvement, better animal monitoring, and controlled feeding (Tecnologia no Campo, 2018). Currently, intensive farming in Brazil is more common in the dairy sector (Tecnologia no Campo, 2018), but it has also become a growing trend in beef production. In these systems, animal waste becomes more concentrated than in extensive production, requiring proper management. Cattle can produce up to 8% of their body mass in manure per day, and improper disposal can contribute to eutrophication of water bodies, ammonia volatilization leading to acid rain, and methane emissions to the atmosphere (Font-Palma, 2019).

Cattle manure contains high levels of organic matter (8,000 mg COD L^{-1}), approximately 20 g kg^{-1} of volatile solids (Bułkowska et al., 2022), as well as 7.89 g

kg^{-1} of phosphorus, 38.45 g kg^{-1} of potassium (Costa et al., 2015), and 2–8.1 g kg^{-1} of nitrogen (Bernal et al., 2009). Currently, in intensive systems, manure is typically stored in manure pits and, after stabilization, applied to soil without adequate control, which may cause nutrient imbalances and contamination of surface and groundwater (SEAPA, 2020). One strategy to reduce environmental impacts and add value to manure is anaerobic digestion (AD), which allows for the recovery of energy as biogas and the reuse of nutrients present in the waste. The biogas generated through AD can be used either in its raw form or upgraded to purified methane for electricity, heat production, or fuel applications.

However, one challenge in using manure for biogas production is its high ammoniacal nitrogen content, which can inhibit the microorganisms responsible for AD (Chen et al., 2008). To mitigate this risk, nitrogen can be removed using membrane-based processes. Membrane distillation is a viable alternative, as its separation mechanism captures ammonia in its gaseous state within the digester and its subsequent fixation in the permeate as ammonium, allowing its reuse in fertilizer production (Hu et al., 2024). Thus, energy recovery combined with ammonia recovery can provide both environmental and economic benefits by reducing energy costs and generating ammonium sulfate as a potential fertilizer.

At the same time, population growth demands an increase in wastewater treatment plants, leading to higher volumes of sewage sludge. The per capita production of primary and secondary sludge in wastewater treatment plants ranges between 0.9 and 2.0 L $inhabitant^{-1}$ day^{-1} and 3.1 and 8.2 L $inhabitant^{-1}$ day^{-1}, respectively (Von Sperling, 2014). In the metropolitan region of Florianópolis-SC served by CASAN, annual spending on transportation and landfill sludge disposal alone exceeded 5.5 million reais (CASAN, 2023). To reduce sludge disposal costs and maximize the generation of value-added byproducts, co-digestion of sludge with one or more substrates can be applied, enabling a more efficient process with increased methane potential. The co-substrate can supply nutrients lacking in the sludge, while promoting positive synergistic effects that improve process stability and enhance biogas/methane yields.

However, few studies have evaluated the synergistic effects of mixing cattle manure with biological sludge in anaerobic co-digestion. Therefore, this study aims to investigate the effect of different mixing ratios of these residues on biogas production and to assess the technical feasibility of ammonia recovery using a membrane process coupled to the reactor.

1.1.1 General Objectives

The objective of this work is to investigate the synergy and maximize biogas production from the anaerobic co-digestion of sewage sludge from secondary treatment and bovine manure, and recover ammonia present in the digestate using membrane distillation, aiming at the production of biofertilizers.

1.1.2 Specific Objectives

- Define the best ratio between the different mixtures of waste to maximize biogas production and quantify the biodegradation efficiency of the waste in co-digestion;
- Operate a semi-continuous bench-scale reactor to verify the efficiency of anaerobic digestion with the optimized mixture;
- To recover ammonia from the digestate by membrane distillation aiming at the production of ammonium sulfate.

1.1.3 Structure

This document was structured into 4 chapters that cover the dissertation’s development. Chapter 1 introduces the research context, the general and specific objectives, and the dissertation’s structure. Chapter 2 reviews the anaerobic co-digestion of sewage sludge with manure state-of-the-art. Chapter 3 presents the experimental investigation of the anaerobic co-digestion for biogas and fertilizer generation. Chapter 4 states the conclusions from the dissertation’s findings, proposing suggestions for future research on the theme.

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2. CHAPTER 2

State-of-the-art

This chapter reviews the anaerobic co-digestion of sewage sludge with manure state-of-the-art.

ARTICLE 1: CO-DIGESTION OF AGRICULTURAL AND URBAN WASTES: A SUSTAINABLE APPROACH FOR ENERGY AND NUTRIENT RECOVERY IN BRAZIL

Abstract

Constant population growth intensifies several global emergencies, including greater demand for renewable energy and higher waste production. Biogas, generated through anaerobic digestion (AD), is rich in methane, offering a versatile renewable energy source suitable for electricity, heat, and fuels. While traditionally produced from single substrates, co-digestion has emerged as an effective strategy to enhance biogas yield, improve process stability, and optimize nutrient balance. Brazil presents a great potential for anaerobic co-digestion (AnCod) due to its extensive agricultural sector, large livestock herds, and expanding wastewater treatment infrastructure. Municipal wastewater treatment plants generate substantial volumes of sewage sludge requiring sustainable disposal, while manure from cattle, swine, and poultry production represents an underutilized bioenergy resource. However, despite this potential, the implementation of AnCod in Brazil remains limited by technological gaps, and the scarcity of comprehensive national data. This review compiles and critically examines scientific studies focused on the anaerobic co-digestion of sewage sludge and livestock manure. A bibliometric analysis was conducted using RStudio and the Bibliometrix package, followed by a systematic content analysis of selected studies. In addition to technical and operational parameters, this article explores the current potential and policy framework for integrating co-digestion into Brazil's renewable energy strategy.

Keywords: Renewable energy; Livestock Waste; Sewage Sludge; Brazil.

2.1. INTRODUCTION

The global demand for renewable energy sources and sustainable waste management strategies has intensified in recent years, driven by efforts to mitigate climate change and population growth. Energy transition encompasses explicit changes such as shifts in energy types, structures, and technologies, and implicit changes, including transformations in governance, justice, and geopolitical dynamics (Yang et al., 2024).

One of the alternatives for renewable energy sources is biogas. Its usage as an energy resource is mainly due to the presence of methane (CH4), which can be used in multiple applications, such as electricity production, heating, and as a vehicle fuel. Although biogas production is concentrated primarily in Europe, its adoption has been steadily expanding in other parts of the world. Global energy production from biogas increased from 0.28 EJ in 2000 to 1.28 EJ in 2014, corresponding to approximately 59 billion m^3 of biogas (equivalent to 35 billion m^3 of methane). Of this total, around 18 billion m^3 methane equivalent originated from the European Union, accounting for roughly half of the global biogas production. And the global installed biogas capacity reached 15 GW in 2015, with 10.4 GW in Europe only, 2.4 GW in North America, 711MW in Asia, 147MW in South America and 33MW in Africa. (Scarlat et al., 2018).

Biogas is widely produced through anaerobic digestion (AD) technology using organic waste, generating energy while simultaneously supporting waste management. AD significantly reduces greenhouse gas emissions, mitigates waste accumulation, and supports energy sourcing (Timonen et al., 2019). However, economic feasibility depends on factors such as technology costs, policy incentives, and market conditions.

In an urban environment, one potential organic substrate for AD is the sewage sludge, produced in domestic sewage treatment plants, which is constantly generated and can be used to produce biogas. Municipal aerobic wastewater treatment plants (WWTPs) are among the most energy-demanding industrial facilities, with aeration processes accounting for the majority of their energy use (Maslon et al., 2024). Therefore, ensuring a sustainable supply of both electrical and thermal energy for their operations is essential and biogas generated through the AD of sludge represents a valuable renewable energy source that can be utilized to meet these demands (Sanaye et al., 2022). Whereas in a rural environment, animal manure, such as cattle, swine, and poultry waste is a potential resource in AD systems, since it has high values of organic content and nutrients and its improper disposal can lead to significant environmental problems. Animal manures are usually used in land application or lagoons, driven by the intensification of livestock production and the need to manage large volumes of waste. Although land application of manure is a common strategy worldwide, it often leads to nutrient overloading in soils and can cause environmental problems such as the eutrophication of water bodies, ammonia toxicity in aquatic ecosystems, significant greenhouse gas emissions, and the enrichment of heavy metals and pathogens in the soil (Qi et al., 2023). Therefore, AD becomes a potential method of livestock manure management and methane production.

However, mono-substrate for AD has limitations, as it often presents an unbalanced carbon-to-nitrogen (C/N) ratio, leading to suboptimal process performance and relatively low biogas yields. Anaerobic co-digestion (AnCod), which combines different organic residues such as sewage sludge and manure, has emerged as a promising strategy to improve process efficiency. This approach enhances the nutrient balance, dilutes potential inhibitors, and promotes synergistic effects between substrates, resulting in increased biogas production and improved economic feasibility of AD systems (Piñas et al., 2018).

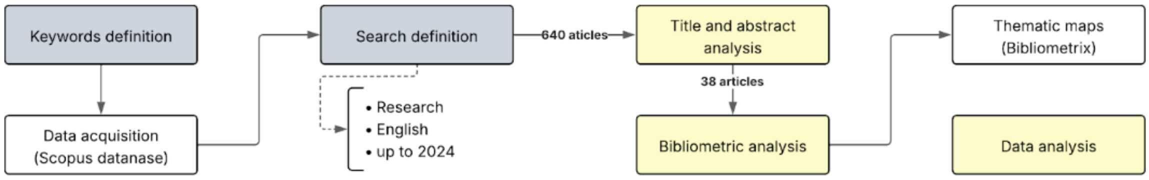
Brazil holds significant potential for anaerobic co-digestion due to its large-scale livestock production and the increasing wastewater treatment infrastructure. The country is the second largest beef producer, only behind the United States, with around 11 million tons produced, representing 15.8% of the world's production in 2024 (EMBRAPA, 2024a). It is the major exporter of cattle with approximately 2,536 million tons of fresh and processed beef exported in 2024 (MAP, 2024). Related to poultry production, Brazil remained the world's largest exporter of poultry meat by volume in 2024, with an estimated 4.7 million tons, accounting for 21.6% of the global production (EMBRAPA, 2024b). Therefore, Brazil generates vast quantities of manure that are often underutilized as a source of energy.

At the same time, municipal wastewater treatment plants (WWTPs) produce substantial amounts of sewage sludge that require proper management to avoid environmental impacts. Given this context, this review aims to compile and critically analyze studies addressing the anaerobic co-digestion of sewage sludge with livestock manure, with a particular emphasis on the Brazilian context. The paper discusses the current state of research, highlights technological and operational challenges, and identifies opportunities to enhance biogas production and resource recovery in line with national energy and sanitation strategies.

2.2. METHODOLOGY

A bibliometric analysis was carried out. The bibliometric review was based on bibliometric indicators by grouping the articles' information using the RStudio software and the Bibliometrix package. The systematic review was conducted through a content analysis of the selected articles on the central topic of this study. Figure 1 illustrates the methodological framework adopted for the literature search. Scopus (scopus.com) was chosen as the primary database due to its wide use in bibliometric and systematic reviews, and its integration with Bibliometrix. The search involved the use of relevant terms applied to article titles, abstracts, and keywords, restricted to research papers published in English up to 2024. The search terms included "CO-DIGESTION" AND "SEWAGE SLUDGE" AND "MANURE" and the review papers were excluded, resulting in a total of 640 articles. This collection went through a content analysis based on its title and abstract. The remaining 38 articles were examined in greater detail to explain, understand, and discuss the application of anaerobic co-digestion for treating sewage sludge and manure.

Figure 1 - Methodological framework for literature search.



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2.3. RESULTS AND DISCUSSION

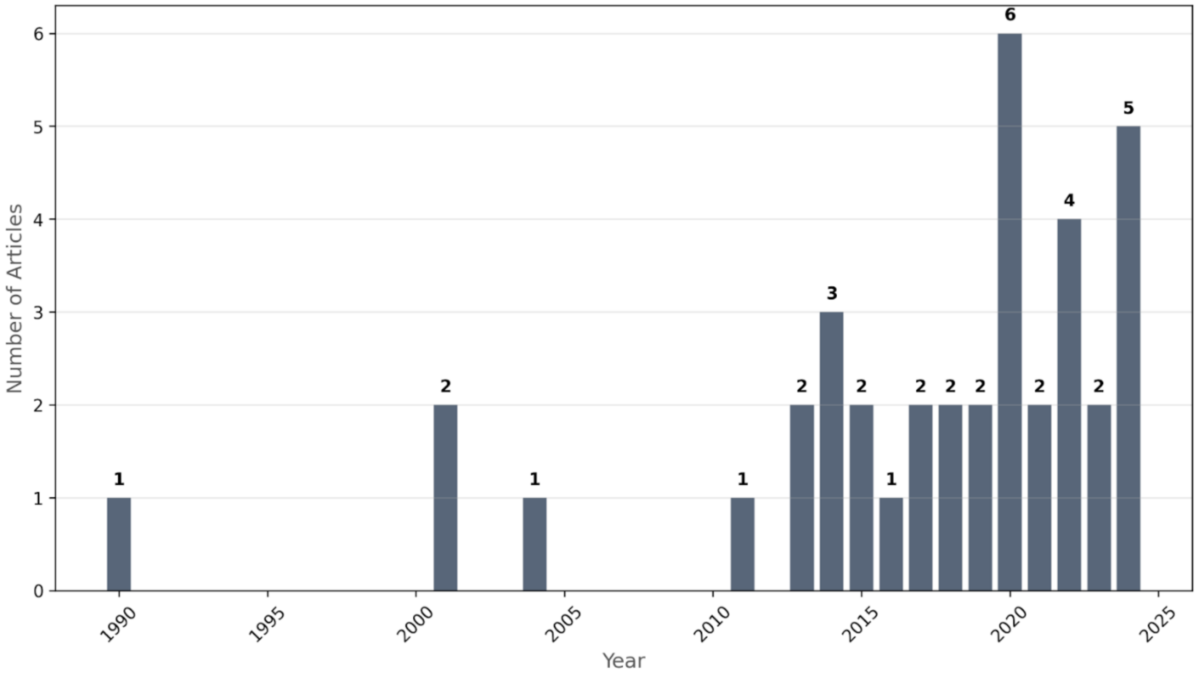
2.3.1. Bibliometric Analysis

A bibliometric study was conducted to analyze the relevance and trends in research about anaerobic co-digestion of sewage sludge with manures from the beginning to 2024. This study prioritizes the information obtained by the chosen database (Scopus). The open-source Bibliometrix R-package processed the data. For this review, the selected indicators were the number of publications, the most influential countries, and the most influential publications.

2.3.1.1. Number of Publications

The annual growth rate in the number of publications for AnCod treatment of SS and manures was 33.4% (Fig. 2). By analyzing Figure 2 extracted from Bibliometrix, it is explicit that the academic community’s interest in this topic and the possibility of the AnCod as a feasible treatment, however the number of articles is lower than other codigestion topics, with different substrates. The highest peak occurred in 2020, likely driven by growing concern over climate change and the urgent need to establish renewable energy sources with lower environmental impact within the global energy matrix. Within the collection of articles analyzed, only one study was conducted in Brazil: “Assessment of potential biogas production from multiple organic wastes in Brazil: Impact on energy generation, use, and emissions abatement”, which evaluates not only the biogas potential of sewage sludge and livestock manure but also includes several other substrates.

Figure 2 - Scientific production by year for anaerobic co-digestion treatment of sewage sludge and manure.



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2.3.1.2. *Most Influential Countries*

The analysis of the most influential country in the area was based on the criteria of the number of publications and citations within the selected period. China was the most influential country in terms of publications, with 12 articles and 575 citations. The second and third countries with the most publications were the United States and Spain, with 3 articles and 154 citations and 3 articles and 30 citations, respectively.

2.3.1.3. *Most Influential Publications*

The most influential articles were defined by the total number of citations, which indicates the papers likely to have the most significant relevance and impact on the research topic. The most three cited article on the collection were the study of the impact of food industrial waste on anaerobic co-digestion of sewage sludge and pig manure (Murto et al., 2004) with 350 citations, the study of anaerobic co-digestion of pig manure with dewatered sewage sludge under mesophilic conditions (Zhang et al.,

2014) with 244 citations and the study of anaerobic co-digestion of swine and poultry manure with municipal sewage sludge (Borowski et al, 2014) with 139 citations. The article from Brazil had 122 citations until the moment, a relevant number compared to other articles analysed, showing the relevance of the topic in the Brazilian context, and the interest in studying the country's biogas production potential using waste materials and promoting a circular economy.

2.3.2. Potential and Scenario in Brazil

In 2024, the average natural gas consumption in Brazil reached approximately 52.47 million m³/day, representing a 0.7% increase compared to 2023, when the average consumption was around 52.07 million m³/day (ABEGÁS, 2025). These represent the total volume consumed by clients across various sectors, including industrial, automotive, commercial, residential, power generation, cogeneration, raw materials, and others, based on monthly statistical surveys conducted by ABEGÁS (Brazilian Association of Piped Gas Distribution Companies) in collaboration with piped gas distribution concessionaires throughout the country (ABEGÁS, 2025). As of 2025, natural gas is delivered to more than 4.7 million consumers through a network of 45,000 km of distribution pipelines across Brazil (ABEGÁS, 2025). The average supply of gas imported in the country from Bolivia between March 2024 and February 2025 was 18.35 million m³/day (MME, 2025)

Although natural gas is often presented as a cleaner energy source compared to other fossil fuels such as coal, bitumen, and diesel and has the ability to aid the integration of renewables, the infrastructure, transportation, and fracking-related impacts pose a significant environmental challenge (Mohammad et al., 2021). Thus, diversification remains a key strategy for ensuring a sustainable energy matrix in the long term, with a focus on renewable and regionally produced resources.

In this context, analyzing and understanding the energy potential of waste in Brazil is essential for defining priority actions to support the sector's development. Biogas production stands out as a relevant and promising alternative for replacing fossil fuels in an environmentally friendly manner, particularly given the availability of biomass and the necessity to manage agro-industrial waste, potentially replacing the gas imported (Ignatowicz et al., 2023). Despite Brazil being a major consumer of natural gas and possessing substantial biogas production potential, given its large territorial extent and prominent role in global agriculture, there is still a limited number of studies and scientific articles on the topic originating from the country. Brazil does not yet appear among the most influential contributors in this research field. Strengthening scientific investigation can generate more comprehensive results on co-digestion performance, offering a solid technical foundation to support practical applications and foster broader deployment of anaerobic digestion technologies nationwide.

2.3.2.1. Brazil's Livestock

There is a high potential for Brazil's biogas production. The country stands out as one of the world's leading agricultural producers, playing a strategic role in global food security. With a vast land area and favorable climate conditions, the country leads the production of several key commodities, including soybeans, corn, beef, poultry, and coffee. This extensive agricultural and livestock activity generates large volumes of organic residues, such as manure and agro-industrial waste, which present both an environmental challenge and an opportunity for sustainable energy production. In this context, biogas generation through AD emerges as a promising solution, enabling the recovery of energy from these residues while contributing to waste management and greenhouse gas mitigation. Salomon and Lora (2009) estimated a potential of about 1 GW (approximately 7 TW h/y) of energy generated from biogas in Brazil in 2009, when the population, livestock units, and landfills were lower than the current obtainable.

Brazil holds significant positions in the production and export of beef, pork, and poultry. Pig farming is one of the most prominent and profitable branches of livestock production, with Brazil ranking among the world's leading swine producers (USDA, 2020). The Brazilian swine industry is characterized by confined animal feeding operations (CAFOs), also known as intensive production systems, which generate large volumes of effluents. In 2023, the national swine herd reached 43 million heads, predominantly located in the southern region.

Egg-laying poultry production also holds a significant position within Brazilian agribusiness. Similar to swine farming, this sector is based on confinement systems, in which a high number of animals are raised in limited areas, resulting in substantial effluent generation. In 2023, the population of laying hens and other gallinaceous birds totaled 1.86 billion head, distributed throughout the country, with a concentration in the Central-West region. It is important to note that, for statistical purposes, only female birds of this species bred specifically for egg production are classified as hens. In contrast, the broader term "gallinaceous birds" encompasses not only hens but also other categories such as broilers raised for meat production.

Dairy and beef cattle farming are widespread across the entire national territory, playing a crucial role in both the economic and social dimensions. In 2023, Brazil's cattle herd reached 238.6 million head, concentrated in the North and Central-West regions (PPM, 2023). Brazil was the world's largest bovine meat exporter, the second-largest producer, and the third-largest consumer (MAPA, 2024). Other livestock sectors in the country also maintain large herds, including sheep, horses, buffaloes, and quails. However, these will not be addressed in this article . The chemical characteristics of the main types of livestock manure produced in Brazil will be further discussed in the following subsection.

In 2021, Institute 17 assessed Brazil's short-term potential for biogas supply (Institute 17, 2021), it was defined based on technical production criteria, excluding considerations of economic feasibility, operational and maintenance costs, or associated business models. According to the study, considering the swine herd value of 22,016 heads in 2021, the potential production of effluents was around 30,383 thousand m³/year. This effluent amount would result in a biogas production potential of 805,592 Nm³/year solely from swine waste, entailing a potential installation of 11,835 new biogas plants.

For laying hens, with a population of 68,739 heads in 2021, the biogas production potential was 175,631 Nm³/year, supported by approximately 2,259 new plants. Whereas considering only dairy cattle farming, the biogas production potential was estimated at 33,092 Nm³/year, with a projection of 11,959 potential new biogas plants (Institute 17, 2021). Although the study considered only dairy cattle, Santos et al. (2018) found that approximately 60% of the total potential energy from biogas comes from cattle manure, reflecting the country's high cattle population (over 200 million herds). It is important to highlight that these data may be underestimated, as breeding animals, such as sows or dairy cows used for reproduction, also generate potential effluents but are not accounted for within the slaughter or production herds.

In 2023, the number of animals in all cited sectors exceeds the 2021 values. Combined with population growth and the increasing demand for food, the forecast indicates an even greater potential volume in the coming years. In the same year, Brazil had 1,324 biogas plants in operation, with an installed production capacity of just over 4 billion Nm³/year, showing a significant increase compared to 2021, which had 755 plants and a production capacity of 2.3 billion Nm³/year (CIBIOGAS, 2021; 2023). Brazil has the potential to significantly reduce, or even eliminate, its dependence on natural gas imports from Bolivia, which average 6.7 billion m³/year, by fully utilizing its internal biogas generation capacity. Its installed biogas production corresponds to approximately 10.96 million m³/day. Although this volume does not yet completely replace imports, it demonstrates that with the expansion of biogas infrastructure and efficient management of organic residues, such as sewage sludge, livestock manure, and agro-industrial waste, Brazil could become self-sufficient in renewable gas production. Investing in biogas would decentralize energy sources, strengthening national energy security and sustainability. In January 2017, there were only 15 biogas power plants, which produced 114.7 MW of power (The Brazilian National Electric Energy Agency ANEEL, 2017). This represented only 0.83% of the national biomass capacity according to Santos et al. (2018), emphasizing the potential for utilizing the waste generated in the country and reducing dependence on external suppliers.

2.3.2.2. Brazil's Sewage Treatment

Regarding sanitation, Brazil has been progressively advancing its infrastructure, though significant challenges persist regarding sewage collection and treatment. According to the National Sanitation Information System (SNIS), in 2017, only 46% of the total wastewater generated in Brazil was treated, and this increased to 52.2% by 2022, with only 56% of the total population served. In comparison, the substantial investments in sewage infrastructure increased from R$7.35 billion in 2021 to R$9.95 billion in 2022 (SNIS-AE, 2022). Although still limited, this upward trend indicates growing investment in sanitation infrastructure and the volume of treated wastewater is expected to increase, leading to a greater generation of sewage sludge.

The predominant sewage treatment technologies in Brazil vary regionally. The “Australian system,” consisting of anaerobic lagoons followed by facultative lagoons, is widespread, particularly in the Southeast, while anaerobic reactors predominate in the Northeast, South, and Central-West regions. Conventional activated sludge processes, although fewer in number, serve approximately 24% of the population connected to treatment plants, mainly in the Southeast and Central-West regions. Primary treatment processes, often linked with submarine outfalls, cater to about 11% of the treated population. Other significant technologies include anaerobic reactors followed by aerobic filters and prolonged aeration activated sludge systems, collectively covering around 13% of the population served by sewage treatment (ANA, 2017). These diverse wastewater treatment technologies also influence the quantity and characteristics of the sewage sludge produced. As sanitation coverage grows and these treatment systems expand, the volume of sludge generated across the country will increase significantly, reinforcing the need for proper management and creating a substantial opportunity for valorization through anaerobic digestion and biogas production. Currently, final sludge disposal depends on waste classification and can include sanitary landfilling, landfarming, incineration, or reuse in agriculture, reforestation, land rehabilitation, and construction activities (SNIS, 2021).

According to data from the PROBIOGAS program (Brazilian Ministry of Cities, 2015), the potential biogas production from sewage sludge biodigestion is between 14.5 and 25 liters per person per day. Using the number of inhabitants served by collective aerobic wastewater treatment systems in 2021 (112.8 million people), based on the most recent data from the SNIS (2021), Brazil would have a production potential ranging from 598 million m^3 to 1.03 billion m^3 per year. Whereas the study by Santos et al. (2018) showed that potential biogas production, using sewage sludge residue in Brazil in 2018 was approximately 226.91 million Nm^3/year, with an electric potential of 0.352 TWh/year, and 0.144 Mt of CO2 emissions avoided. The estimates of biogas production from sewage sludge in Brazil are likely conservative due to incomplete data and limited sanitation coverage.

The potential of agropecuary sector waste is widely explored for biogas production due to its chemical characteristics that favor the biogas generation. In contrast, sewage sludge is less frequently utilized, as it is a residue with low organic matter concentration, a high presence of pathogens, and toxic substances, whereas manure has high organic content, buffer capacity and optimum C/N ratio, leading both wastes complementary in their characteristics (Santos et al., 2018; Zhou et al., 2023). The relatively low biogas yield from sewage sludge alone can be significantly enhanced through anaerobic co-digestion with animal manures, improving many aspects such as dilution of inhibitors, and carbon-to-nitrogen balance.

The biogas potential from sewage sludge not only represents an opportunity for renewable energy generation but could also contribute significantly to reducing the energy demand of wastewater treatment plants (WWTPs). Electricity is one of the main operating costs for water and sewage services. In 2022, electricity expenses for sanitation providers reached R$ 9.2 billion, representing a 2.2% increase compared to the R$ 9.0 billion recorded in 2021, and the energy consumption for sewage systems averaged 0.29 kWh/m³, totaling 1.7 TWh nationwide (Brazilian Ministry of Cities, 2022). By capturing and utilizing the methane produced during anaerobic digestion, WWTPs can partially or even fully supply their own electrical and thermal energy needs. This approach not only enhances the sustainability of treatment operations but also offers economic benefits by lowering energy costs and improving overall operational efficiency.

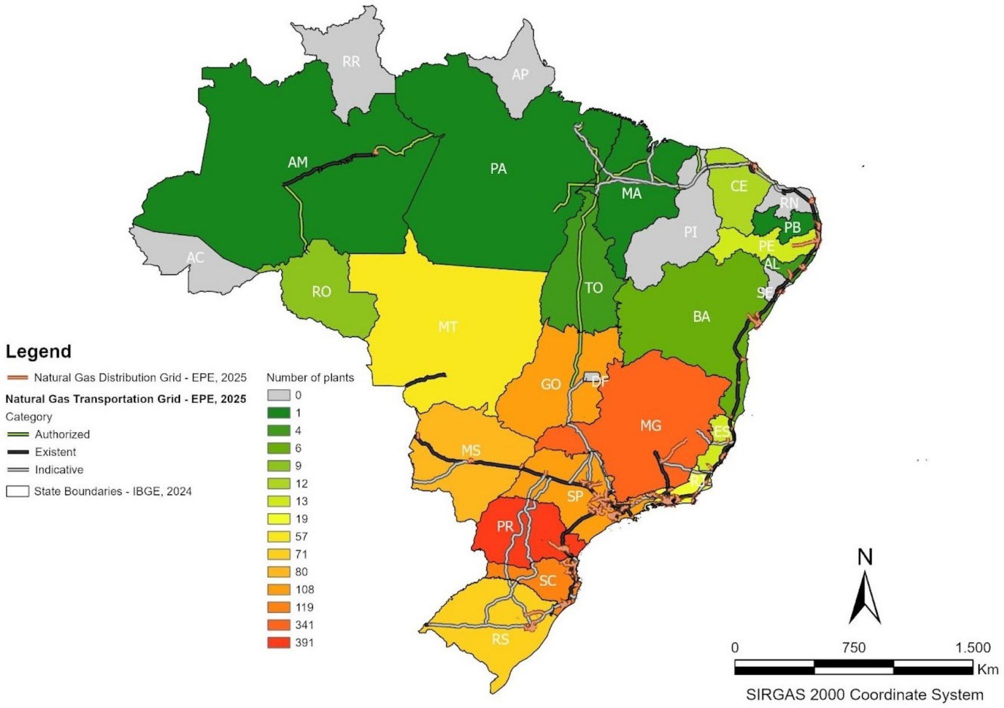
2.3.2.3. Biogas Purification and Infrastructure

Biogas injection into the natural gas grid is a key strategy for decarbonizing energy systems and increasing the share of renewable energy. This process involves upgrading biogas to biomethane, ensuring it meets strict quality standards, and then blending it with conventional natural gas for distribution and use in existing infrastructure.

The raw biogas upgrading involves removing CO_2, H_2S, and other impurities to achieve methane concentrations above 95% for grid injection, to avoid corrosion due to the H_2S and Lower Calorific Value and to comply with regulatory standards. Biogas upgrading technologies include membrane separation, pressure swing adsorption, chemical scrubbing, and novel mineralization methods, each with specific cost and efficiency profiles (Aghel et al., 2022; Rafiee et al., 2021; Guerin, 2022).

The main challenges are the cost factors and regulation considerations, the biomethane production and injection are generally more expensive than conventional natural gas due to the high costs associated with its production and preparation for grid injection, with costs influenced by plant size, upgrading technology, and local regulations. A study conducted by Paturska et al. (2015) in Latvia, shows that under the most favourable scenario the injected biomethane is approximately 19% more expensive than the natural gas.

Figure 3 - Natural gas grid extension in Brazil.



Source: Author

In Brazil, the current natural gas pipeline infrastructure primarily follows regions with the highest population density and energy demand, tracing the country's coastline from the South to the Northeast, with a single branch extending inland toward the state of São Paulo. Figure 3 presents the natural gas grid extension based on the Energy Research Company (EPE, 2025). Considering the country's significant potential for biogas production, this pipeline distribution does not favor the injection of biogas into the grid. Highlighted in green are the authorized lines, which are the lines that were planned and are already authorized to be installed. Highlighted in black are the existent lines, that represent the real and currently existenting lines. Highlighted in gray are indicative lines, which represent routes currently under study for potential implementation (EPE, 2025). The indicative and authorized lines represent a positive development, as it demonstrates the exploration of new gas transportation routes in Brazil. Such routes could enable the injection of biogas into the existing infrastructure and contribute to the advancement of biofuels in the country. This is important since the areas with the greatest potential biogas production are predominantly located in the western and inland regions of the country, due to favorable soil characteristics and the availability of space.

Hoo et al. (2019) investigated the optimization of biomethane injection into the natural gas grid. To address the logistical challenge of transporting biomethane from decentralized production sites to injection points, the study modeled a virtual pipeline system in Malaysia. This system would transport compressed biomethane using a modular storage box design and dedicated vehicles (e.g., fuel-powered trucks) to deliver the gas to the existing natural gas infrastructure. The total transport distance between production sites and injection points in the study was approximately 320 km and the study found that biomethane alone does not adequately capture the actual energy and economic landscape in Malaysia. A broader, integrated perspective is required to evaluate appropriate carbon taxation in the country, taking into account national energy planning, various resource options, conversion technologies, and their potential applications. Also, the authors emphasized that leveraging existing gas infrastructure is a key element in enhancing energy system resilience and supporting decarbonization efforts and that support from government agencies on providing accurate data is a key element for national bioeconomy planning.

Such an alternative is not well-suited to the Brazilian context as well. Given the country's vast territorial dimensions, the distance between the coastal pipeline infrastructure and regions with the highest biogas production potential can reach up to approximately 2,000 km, making virtual pipeline systems significantly less efficient and economically not viable.

The modernization of infrastructure and the installation of new connections require significant capital investments. Additionally, logistical challenges related to transporting biogas or biomethane from decentralized production sites to injection points and the need to comply with legislation and regulations, pose a considerable challenge in the Brazilian context. One of the primary challenges for electricity generation from biogas is its economic viability. Rangel (2016) estimated that the minimum cost of biogas-based energy production in Brazil is 105.3 USD/MWh, which is higher than the tariff applied to conventional thermoelectric power plants (86.9 USD/MWh). However, studies demonstrated that electricity generation from biogas can be economically feasible in regions with smaller populations when utilizing landfill gas and anaerobic sewage treatment plants (Barros et al., 2014; Santos et al. 2018).

One successful case in Brazil of anaerobic co-digestion is the Sanepar Bioenergy Plant of the Belém WWTP (USBio). The USBio is operated by the Paraná Sanitation Agency (Sanepar) adjacent to the Belém WWTP in Curitiba. The Belém

WWTP is the largest sewage treatment facility managed by Sanepar, producing approximately 110 tons of sludge per day (equivalent to 530,000 m³).

To maximize the energy efficiency of USBio, the sludge is co-digested with other organic residues from various sources, including food waste, fruit and vegetable residues, butcher and fish waste, brewery by-products, grease trap residues, and organic leftovers from industrial processes. These residues are received through a public call launched by Sanepar and must be accompanied by a technical report in accordance with NBR 10.004, which classifies waste according to potential environmental risks. The agency also requires analyses of volatile solids, BOD, COD, and toxic substances that could inhibit digestion, such as heavy metals, ammonia, surfactants, and phenols, which are not allowed in the received material. Currently, approximately 900 tons of sludge from the Belém WWTP and 50 tons of external organic residues are transformed into biogas, which is then converted into electricity. The energy generated supplies the plant itself, and the surplus is credited for use in other Sanepar facilities. The residues are classified according to the ratio of volatile solids to total solids, with Class A ≥ 90%, Class B ≥ 80% and < 90%, and Class C ≥ 65% and < 80%. This classification determines the maximum quantity of residues that can be accepted per month for co-digestion (GOVERNO DO ESTADO DO PARANÁ, 2025).

2.3.2.4. Relevant Regulatory Framework in Brazil

The regulatory framework for biogas-based energy in Brazil is relatively recent and continues to evolve. A key milestone in this process was the approval of ANP Resolution Nº 08 in 2015, by the Brazilian National Agency of Petroleum, Natural Gas, and Biofuels (ANP), which authorized the injection of biomethane derived from agroforestry residues into the natural gas distribution grid (ANP, 2015). Given the high concentration of contaminants commonly found in biogas produced from landfills and wastewater treatment plants, as well as the limited availability of domestic technologies for efficient purification, the use of such sources for biomethane production was only regulated later, through ANP Resolution Nº 685 in 2017 (ANP, 2017).

In the same year, the National Biofuels Policy (RenovaBio) was established through Federal Law Nº 13,576/2017 (BRASIL, 2017). This document defines the Fuel Certification as a process through which it is verified and certified that a given fuel meets specific criteria for quality, energy efficiency, and environmental sustainability.

Producers who meet these criteria receive Biofuel Certificates (CBios), which can be traded on the financial market and are used to demonstrate compliance with the government's decarbonization targets.

In 2018, ANP Resolution Nº 734 amended ANP Resolution Nº 685 and established procedures for the certification of biofuels.

There are also some state-level laws, such as Law No. 24,396/2023 of the state of Minas Gerais and Ordinary Law No. 20,710/2020 of the state of Goiás, both of which establish and regulate state policies on biogas and biomethane for their respective states. The state of São Paulo introduced a regulatory mechanism for the commercialization and injection of biomethane into the gas network, via Deliberation Nº. 744/2017, which established procedures for public tenders and quality assurance standards (São Paulo, 2017).

Regarding the quality of the biogas, the ANP Resolutions No. 886 of 2022 and No. 906 of 2022 establish the specifications and rules for approving quality control of biomethane derived from landfills and wastewater treatment plants, as well as biomethane from agro-industrial and livestock organic residues. Both resolutions set a minimum methane content of 90% and a maximum hydrogen sulfide (H_2S) concentration of 10 mg/m³ of biogas. They serve as important guidelines for the production and upgrading of biogas for energy use.

Overall, Brazil has made significant progress in establishing a regulatory framework for biogas and biomethane. However, there is still room for improvement, particularly in harmonizing regulations across states, expanding clear guidelines for all biogas distribution and enabling technologies for efficient purification and upgrading, which would facilitate broader and safer integration of biogas into the national energy system.

2.3.3. Wastes and Substrates

2.3.3.1. Sewage Sludge

In Brazil, gathering data on the current final disposal, treatment, and inadequate destination of sewage sludge can be very challenging due to the lack of a single and accurate official source.

Sewage sludge (SS) is a by-product generated during biological treatment of municipal and industrial wastewater. The SS is a solid or semi-solid waste, and its production has a significant and constant increase worldwide. Feng et al. (2023) estimated that the annual global production of sewage sludge may rise from around 53 million tons of dry solids, in 2023, to around 160 million tons if global wastewater were to be treated to a similar level as in Europe. In Brazil, according to the study, the estimated total annual production of SS is 2.5 million tons, reported as dry solids. The volume of treated wastewater, population demographics, and the specific technology used in the Wastewater Treatment Plant (WWTP) primarily influences the generated volume. The SS is primarily characterized by the presence of harmful and toxic substances, such as heavy metals and pathogens (Zhou et al., 2020). In Brazil, gathering data on the current final disposal, treatment, and inadequate destination of sewage sludge can be very challenging due to the lack of a single and accurate official source. Currently, the main disposal method for SS in Brazil is landfilling (SNIS, 2021).

The SS is primarily characterized by the presence of harmful and toxic substances, such as heavy metals and pathogens (Zhou et al., 2020). Sewage sludge waste generated through aerobic WWTP can be divided into two main products: primary sludge and secondary sludge. Sewage sludge is a heterogeneous waste whose chemical composition varies depending on location, cultural habits, and operational conditions, as well as the source and technology of wastewater treatment (Jasinska et al., 2024).

Primary sludge is generated during the initial sedimentation stage, where suspended solids are physically separated from raw sewage. Its organic matter includes lipids, proteins, and carbohydrates, with proteins being a significant component. As a result, it retains a high concentration of biodegradable organic matter and easily settleable inorganic solids. The secondary sludge is produced in the secondary treatment stage, where microorganisms break down dissolved and suspended organic matter. The excess microbial biomass that forms is referred to as secondary sludge, excess activated sludge, or waste activated sludge, and it is often thickened using mechanical thickeners and polymers (Sanaye et al, 2022). This process converts a significant portion of the available organic content into microbial biomass, thereby substantially reducing the amount of readily degradable organic material. Consequently, the lower energy potential of activated sludge, compared to primary sludge, limits its efficiency for biogas production (Nielfa et al., 2015). Table 1 summaries the main characteristics of different types of sewage sludge from the collection of articles studied.

Table 1 - Summary of sewage sludge composition characteristics.

Sewage Sludge	C/N	COD (mg/L)	TAN or FAN (mg/L)	TS (%)	VS (%)	TS/VS (%)	TS (g/kg)	VS (g/kg)	TKN (gN/kg)	pH	Ref
Dewatered Sewage Sludge	-	-	-	16.02 ± 0.35	-	-	-	470.30 ± 5.64	-	6.3 ± 0.1	Dai et al., 2016
Excess activated sludge	-	2151	185	5.2	38.1	-	-	-	-	7.36	Qin et al., 2024
Excess sludge	-	-	-	10.14 ± 0.05	2.70 ± 0.12	26.51	-	-	-	7.00	Wang et al., 2022
Excess sludge	6.51	-	-	6.78 ± 0.03	2.36 ± 0.08	34.80	-	-	-	07.04	Zhou et al., 2023
Municipal Sewage Sludge	8.29	1200 ± 20	-	27	-	40	269.58 ± 0.1	403.80 ± 0.15	-	7.78 ± 0.05	Akbay et al., 2024
Primary + Secondary sludge	-	44,740 ± 8,160 mg/kg	-	-	-	74.05 ± 4.20	46.90 ± 7.64	34.71 ± 5.90	2.85 ± 1.65	-	Borowski et al., 2013
Primary sludge	-	44,200 ± 13,500	-	-	-	-	30.5 ± 2.1	20.7 ± 1.6	1.4 ± 0	7.3 ± 0.3	Maragkaki et al., 2018
Primary Sludge + Excess Sludge and dewatered with the aid of a high-molecular flocculant based on Polyacrylamide (PAM).	6.76	20,769± 316 mg/kg	342.4 ± 36.2 mg/kg	17.69 ± 0.16	11.86 ± 0.06	67.04 ± 0.21	-	-	-	7.46 ± 0.03	Zhang et al., 2014
Primary sludge + Secondary Sludge	5.58	-	-	4.19 ± 0.01	3.30 ± 0.08	78.75	-	-	-	5.86 ± 0.12	Xu et al., 2020
Primary sludge + Waste Activated Sludge (1:1)	-	41,060 ± 13,300 mg/kg	-	-	-	75.20 ± 3.33	48.56 ± 7.98	36.70 ± 7.35	2.82 ± 0.61	-	Borowski et al., 2014
Sewage Sludge	-	2,305	62.5	6.9	13.5	-	-	-	-	7.27	Qin et al., 2024
Thickened Primary Sludge	-	188,000 mg/kg	-	-	-	-	198	132	-	-	Nielfa et al., 2015
Waste-Activated Sludge - 50% Efficiency	558	-	-	-	-	-	44.91 ± 0.056 g/L	11.56 ± 0.346 g/L	-	7.30 ± 0.014	Cruz-Azuara et al., 2024
Waste-Activated Sludge - 90% Efficiency	3.08	-	-	-	-	-	35.58 ± 0.056 g/L	24.24 ± 0.339 g/L	-	7.04 ± 0.084	Cruz-Azuara et al., 2024
Waste-Activated Sludge and Primary Sludge	-	-	138.34–299.47	3.08 – 4.71	2.22 – 3.21	66 - 74	-	-	-	5.54 – 5.97	Jasińska et al., 2024

Source: Author

Legend: C/N (carbon and nitrogen balance), COD (Chemical Oxygen Demand), TAN (Total Ammonium Nitrogen), FAN (Free Ammonium Nitrogen), TS (Total Solids), VS (volatile solids), TKN (total Kjeldahl nitrogen). REF (References).

Parameters such as TAN and TKN indicate nitrogen availability, TAN concentrations reached up to 342.4 mg/L and TKN values ranging from 1.4 to 2.85 gN/kg. The pH of the sludge samples remains relatively neutral to slightly alkaline (5.54 to 7.78), which can influence both biological treatment efficiency and subsequent sludge valorization processes. These physicochemical properties are essential for evaluating the potential of sewage sludge for resource recovery, such as biogas production and nutrient recycling.

Variations in the characteristics of SS can also be observed in relation to the plant's treatment efficiency. In the study by Azuara et al. (2024), two SS samples were analyzed: one from a treatment system with 50% efficiency in removing BOD and another with 90% efficiency. The sludge from the 50% efficient system had a higher concentration of TS, approximately 44.91 ± 0.056 g/L, whereas the sludge from the 90% efficient system showed a lower TS concentration, around 35.58 ± 0.056 g/L. However, the latter exhibited a higher VS content, approximately 24.24 ± 0.339 g/L, indicating greater biodegradability, probably due to the greater portion of soluble organic matter eliminated during the biological process in the plant with higher treatment efficiency (90%) for BOD removal. As a result, the generated sludge contains lower total solids content but is richer in volatile organic matter.

Several conventional methods are being used to treat SS, such as landfilling, composting, land application, and incineration (Zhou et al., 2023; Zhang et al., 2014) The disposal in landfills presents several disadvantages, including greenhouse gas emissions, the need for large land areas, and the devaluation of surrounding areas, as well as soil and groundwater pollution. Incineration is also associated with greenhouse gas emissions and high operational costs. The land application and composting methods recycle nutrients but raise concerns about heavy metals, organic pollutants, pathogens, and antibiotic resistance, necessitating careful monitoring and risk assessment to protect soil and human health (Wang et al., 2023).

The use of AD is noted as an alternative to treat SS and to enhance hygiene by lowering pathogens, reducing the volume of sewage sludge, degrading VS and reducing COD. The process can significantly reduce the load of total coliforms and fecal coliforms in bioslurry samples compared to bio waste samples (Maragkaki et al., 2018). Georgali and others (2022) employed Life Cycle Assessment (LCA) to compare sewage sludge treatment via AD versus open-air composting. They reported that the environmental impact of anaerobic digestion is 6.5% to 7.5% lower than that of composting, across all evaluated environmental impact categories.

Thus, AD is recognized as an alternative for treating sewage sludge due to its various environmental, economic, and operational advantages, especially when compared to other disposal methods, like incineration and landfilling. AD enables energy recovery, reduces sludge, and stabilizes biomass, and it is recommended in many countries. It is estimated that 66% and 90% of sewage sludge in the UK and Germany, respectively, is treated by anaerobic digestion (Tao et al., 2017). In the USA, over 1,200 wastewater treatment facilities have installed anaerobic digesters for sludge treatment and biogas production (Wang, 2023).

The SS disposal and management account for a significant portion of the total operations costs in WWTPs. In the United States, many wastewater treatment systems allocate between 40% and 50% of their budget on the treatment and disposal of waste-activated sludge (WAS), with approximately 62% of this being treated in landfills or incinerated (Cruz-Azuara et al., 2024).

2.3.3.2. Manure

Manure is an organic waste product from farmed animals, including excrement and urine, often mixed with bedding, with distinct characteristics that make it both a valuable resource and a management challenge (Jasinska et al., 2024).

Its composition is rich in biodegradable organic matter and contains valuable nutrients, especially phosphorus and nitrogen. However, it can also contain contaminants such as pathogens, organic pollutants, and heavy metals, as well as an inert fraction like lignin and non-degradable cellulose, depending on the animal diet and the farm location. Usually, it has high micronutrient, buffering capacity, and biodegradability. Indicative macromolecule analysis for livestock manure waste shows high ranges of carbohydrates (44.06-90.2%), proteins (8.3-23%), lipids (1.5-4.9%), and lignin (0.3-56%) (Zouaghi et al., 2020; Fierro et al., 2013; Fang et al. 2020; Toufexis et al., 2024).

From the database acquired through the collection of articles, the three types of manures most common in Brazil have been chosen to be cited in this study and are summarized in Table 2: cattle, poultry and pig manure.

Table 2 - Summary of manures composition characteristics.

Manure	**C/N**	**COD (mg/L)**	**TAN or FAN (mg/L)**	**TS (%)**	**VS (%)**	**TS/VS (%)**	**TS (g/kg)**	**VS (g/kg)**	**NTK (gN/kg)**	**pH**	**Ref**
Cattle manure	-	5,298	137.5	10.7	-	54.5	-	-	-	7.30	Qin et al., 2024
Cattle manure	-	259	-	-	-	-	222	209	-	-	Nielfa et al., 2015
Cattle manure	-	-	-	17.54 ± 0.21	-	-	-	758.60 ± 6.09	-	8.2 ± 0.2	Dai et al., 2016
Poultry manure	-	-	-	59.16 ± 0.06	48.19 ± 0.24	80.15	-	-	-	8.66	Wang et al., 2022
Poultry manure	-	201.39 ± 18.86	-	-	-	73.95 ± 5.55	277.18 ± 20.24	205.32 ± 25.57	13.09 ± 4.16	-	Borowski et al., 2014
Poultry manure	-	234,3000 ± 53,960 mg/kg	-	-	-	72.65 ± 6.22	288.55 ± 47.38	215.27 ± 37.29	14.07 ± 2.54	-	Borowski et al., 2013
Poultry manure	-	-	7,000 ± 12.3	27.44 ± 0.24	20.19 ± 0.81	0.74	-	-	-	5.41 ± 0.01	Jasińska et al., 2024
Swine manure	19.02	-	1,741.7 ± 128.0 mg/kg	37.60 ± 0.13	34.09 ± 0.27	90.66	-	-	-	8.25 ± 0.03	Xu et al., 2020
Swine manure	12.96	54,691 ± 3,887 mg/kg	-	29.96 ± 0.26	20.89 ± 0.23	69.73 ± 0.33	-	-	-	8.41 ± 0.02	Zhang et al., 2014
Swine manure	-	119.50 ± 7.95	-	-	-	72.20 ± 3.76	123.96 ± 28.20	90.12 ± 24.37	6.40 ± 2.17	-	Borowski et al., 2014

Source: Author

Legend: C/N (carbon and nitrogen balance), COD (Chemical Oxygen Demand), TAN (Total Ammonium Nitrogen), FAN (Free Ammonium Nitrogen), TS (Total Solids), VS (volatile solids), TKN (total Kjeldahl nitrogen). REF (References).

The cattle manure is characterized by relatively low TS, ranging from 10.7% to 17.54%, and SV representing 54.5% of TS content (TS/VS), corresponding to 209 - 758.6 g/kg TS. The COD of cattle manure varies considerably in the literature, ranging from 5,298 mg/L to 259 g/kg. Dai et al. (2016) found a lignin concentration of 75.01 ± 2.41(g/Kg-TS) and cellulose concentration of 254.31 ± 2.19 (g/Kg-TS). The ammoniacal nitrogen concentration reported in the literature was 137.5 mg/L and the pH levels range from neutral to slightly alkaline (7.3–8.2).

Swine manure presents high content of TS and VS. TS concentrations vary from 29.96% to 37.60% with TS/VS ratios ranging from 69.73% to 90.66%. COD values differ significantly, ranging from 119.50 to 54,691 mg/kg. Total ammoniacal nitrogen (TAN) concentrations are also elevated, reaching 1,741.7 mg/L, which may impose inhibitory effects if not adequately managed. The pH of pig manure is typically alkaline (8.25–8.41).

The poultry manure is notable for its high dry matter and organic content, with TS/VS ratio ranging from 72.20 ± 3.76 to 80.15%. In terms of COD, poultry manure exhibits values between 201.39 and 234.30 g/kg, highlighting its high biodegradability. It is widely recognized that poultry manure presents higher nitrogen content compared to other types of manure, as hens are classified as monogastric animals. The ammoniacal nitrogen concentration found in the literature was 7,000 ± 12.3, resulting in an inappropriate carbon-to-nitrogen ratio in the substrate. The intense accumulation of ammonia released by the decomposition of uric acid and undigested proteins usually results in the inhibition of methane-forming microorganisms (Borowski et al 2013).

Proper management and disposal of manures are essential, since inadequate practices may lead to significant environmental impacts, including soil and water contamination, unpleasant odors, and the release of greenhouse gases (Xu et al., 2020). Liu et al. (2019) studied the excessive application of livestock manure in paddy soils. The application significantly increases availability of phosphorus (P) in topsoil, intensifying the P leaching, an undesirable process of soil “washing,” in which nutrients and other chemical elements are transported to depths beyond those occupied by plant roots, which can cause economic and environmental losses for farmers and also contamination of aquatic habitats and other water resources. The application also leads to the release and enhanced migration of toxic metals. Table 2 presents different characteristics of animal manure, which chemical composition varies depending on the animal’s diet and farm system.

2.3.4. Anaerobic Digestion

Anaerobic digestion (AD) is a well-established biological process widely applied for treating organic waste and wastewater, enabling both the stabilization of residues and the generation of renewable energy in the form of biogas. The process occurs in an oxygen-free environment and involves a complex microbial consortium that acts synergistically through four main stages: hydrolysis, acidogenesis, acetogenesis, and methanogenesis (Appels et al., 2008).

In hydrolysis, complex organic matter, such as carbohydrates, proteins, and lipids, is broken down into soluble and simpler monomers by hydrolytic bacteria through extracellular enzymes. Lignocellulose and lignin are hydrolyzed more slowly, often incompletely. During acidogenesis, these monomers are then fermented by acidogenic bacteria into short-chain volatile fatty acids (VFA) (butyric, propionic, and acetic acids), alcohols, nitrogen oxides, hydrogen sulfide, hydrogen, and carbon dioxide. Acetogenesis is the third stage of anaerobic digestion and is considered critical to the process, being carried out by a group of bacteria known as acetogenic bacteria. During acetogenesis, VFAs are converted into acetic acid, hydrogen, and carbon dioxide. Finally, methanogenic archaea convert these products into methane and carbon dioxide, completing the process with the methanogenesis phase.

The efficiency and stability of AD are heavily influenced by the characteristics of the substrate, including the carbon-to-nitrogen (C/N) ratio and presence of inhibitory compounds such as ammonia or heavy metals, organic matter content and composition, usually expressed in Total Solids (TS), Volatile Solids (VS), and Chemical Oxygen Demand (COD). The operational parameters also influence the AD performance, such as temperature, Hydraulic Retention Time (HRT), and Organic Loading Rate (OLR).

The C/N ratio indicated for AD is around 20 - 30 (Borowski et al., 2014), and mono-digestion often suffers from nutrient imbalances. It is possible to add nutrients; however, this introduces an additional cost to the process. This imbalance can be solved through co-digestion, where multiple substrates are treated simultaneously. The AnCod benefits include enhanced process performance by improving nutrient balance and microbial diversity, balanced C/N ratio, dilution of inhibitory or toxic compounds, economic and operational advantages such as energy recovery and biogas. Additionally, establishing a centralized facility for co-digesting multiple waste streams could address the challenge of the high implementation costs associated with anaerobic digestion systems (Fierro et al., 2014).

Different organic wastes have been used in the co-digestion process of sewage sludge, such as the organic fraction of municipal solid waste, agricultural waste, food waste, and fat, oil and grease (FOG) (Chow et al., 2020). In Brazil, the separation of municipal solid waste, and consequently the separation of food waste, still faces significant challenges, as a result of a lack of public knowledge, insufficient infrastructure, unequal cost-benefit distribution, and weak professional management that hampers the proper management of recyclable and organic materials (Conke, 2018). Consequently, a large portion of potentially recoverable waste is sent to landfills or, in some cases, to uncontrolled dumpsites, increasing treatment costs and limiting the recovery of materials and energy.

The vast amounts of biodegradable residues and byproducts produced by the livestock and agro-industrial sectors have significant value for biogas production through AnCod. Brazil has significant potential for the application of AnCod in livestock manure management, particularly due to the scale of its agricultural activities. Holding the first position in beef exports and the third in pork exports in the 2024/2025 global ranking, the country generates large volumes of animal manure, especially in intensive farming systems (Fierro et al., 2014; ABN, 2025).

In essence, organic matter serves as the fuel for anaerobic digestion; its quantity, quality, and composition dictate the efficiency of the biological process, the stability of the digester, and the composition of biogas produced. SS has low solids contents and thus poor biogas yields are reported, especially when treating waste activated sludge with little or no addition of primary sludge. Borowski et al. (2014) found that the addition of 30% of pig manure to SS (VS/VS) increased 60.2% gas yield from 274 d^3/kg SV (SS alone) to 439 d^3/kg SV. The amount of organic content in the substrate directly impacts the potential for methane production. The SS C/N ratio varies between 0.558 and 8.29 based on literature (Cruz-Azuara et al., 2024; Akbay, 2023). Thus, animal manure has been reported as an efficient co-substrate with SS due to its high concentrations of VS and COD. Aiming for the best organic matter concentration to enhance biogas production, Zouaghi et al. (2019) indicate a 25/75 ratio of organic substrate and SS, respectively, for the best C/N balance.

VS and COD are critical parameters for evaluating and optimizing AnCod processes, as they directly quantify the organic matter available for microbial degradation and subsequent biogas production. VS represents the biodegradable organic content within a substrate, and a high VS/TS ratio typically indicates a highly biodegradable material.

The solid fraction of manure is rich in trace elements and organic carbon, contributing more to biogas production than the liquid fraction (Zhang, 2011). Usually, high VS concentration leads to increased biogas production. However, careful management of VS and COD concentrations is necessary since high concentrations of readily biodegradable organic matter can lead to the accumulation of VFAs, which can affect the digester's pH and may inhibit methanogenic archaea that are highly sensitive to environmental pH and require a near-neutral pH (typically 6.5–8.5). Azuara et al. (2024) indicate the optimal TS concentration for continuous digesters is estimated to be 8–12%, while in discontinuous digesters it can range between 40% and 60%. However, Xu et al (2020) studied different SS and pig manure ratios to analyze initial TS concentrations (4%, 6%, 8%, and 10%), finding that the highest methane production of 226.1 CH4/g VS was obtained with an initial TS of 6%. The methane production with 10% TS was the lowest value, with 117.9 mL CH4/g VS. In this sense, it is possible to recognize that the quality of the TS is more important than the concentration, and different types of manures can perform differently.

The VS reduction rate is a key indicator of digester performance and efficiency. A relatively low VS reduction during the anaerobic digestion process may indicate suboptimal performance in one or more stages of the process and is often associated with a high fraction of slowly degradable or inert compounds, such as cell wall materials. In the case of sewage sludge, for instance, studies have shown that only up to 60% of its organic content is biodegradable (Mahmoud et al., 2004). Zhang et al. (2014) noticed that the VS reduction increased from 34.7% with monodigestion of SS to 55.20% with AnCod of SS with pig manure. Whereas Borowski et al. (2013) observed the VS reduction increase from 36.33 ± 8.72% for SS monodigestion to 49.35 ± 9.52% with the addition of 30% poultry manure (VS/VS).

Similarly, COD expresses the amount of organic matter in the substrate that can be oxidized, making it a crucial parameter for methane calculation during anaerobic degradation. The COD removal efficiency serves as another important metric for assessing the effectiveness of the anaerobic process.

Both parameters are important to understanding the hydrolysis phase, which is often considered the rate-limiting step in the breakdown of particulate organic matter into soluble substances. Increasing the solubilization of organic substances, as reflected by soluble COD (sCOD) levels, enhances the biodegradability of volatile solids, thereby greatly improving AD efficiency.

The removal of COD in conjunction with gas production in the reactor provided evidence of effective microbial activity by methanogenic archea. The COD level represents a dynamic balance between the degradation of solid organic matter and biogas production. An increase in biogas production alongside rising solids concentration and a stable COD level indicates a good balance between hydrolysis and methanogenesis. However, when an increase in COD is not accompanied by a corresponding peak in biogas production, it suggests an imbalance between these two stages. Xu et al. (2020) used the VFAs/sCOD ratio to evaluate acidogenesis and, consequently, to assess the potential methanogenic environment during the anaerobic digestion process. Overall, higher VFAs/sCOD ratios indicate a greater degree of acetogenesis, which is a prerequisite for methane conversion.

The main operational parameters of the collection of analyzed articles were compiled and are shown in Fig. 4. The optimal HRT depends on the properties of the substrates and temperature. In the case of co-digestion with sludge and manure, most studies reported HRT values of less than 30 days. This is likely due to the synergistic effect of manure with sludge, which facilitated the biodegradability of the manure, since longer HRT is required for lignocellulosic and recalcitrant substrates to ensure complete degradation and stable methane production (Shi et al., 2017).

The most frequent feed regime was batch tests, representing 50% of the studies, while the remaining 50% were divided between only semi-continuous and studies that conducted both tests, utilizing laboratory-scale digesters. Batch studies are commonly more frequent due to the operational limitations of running a reactor.

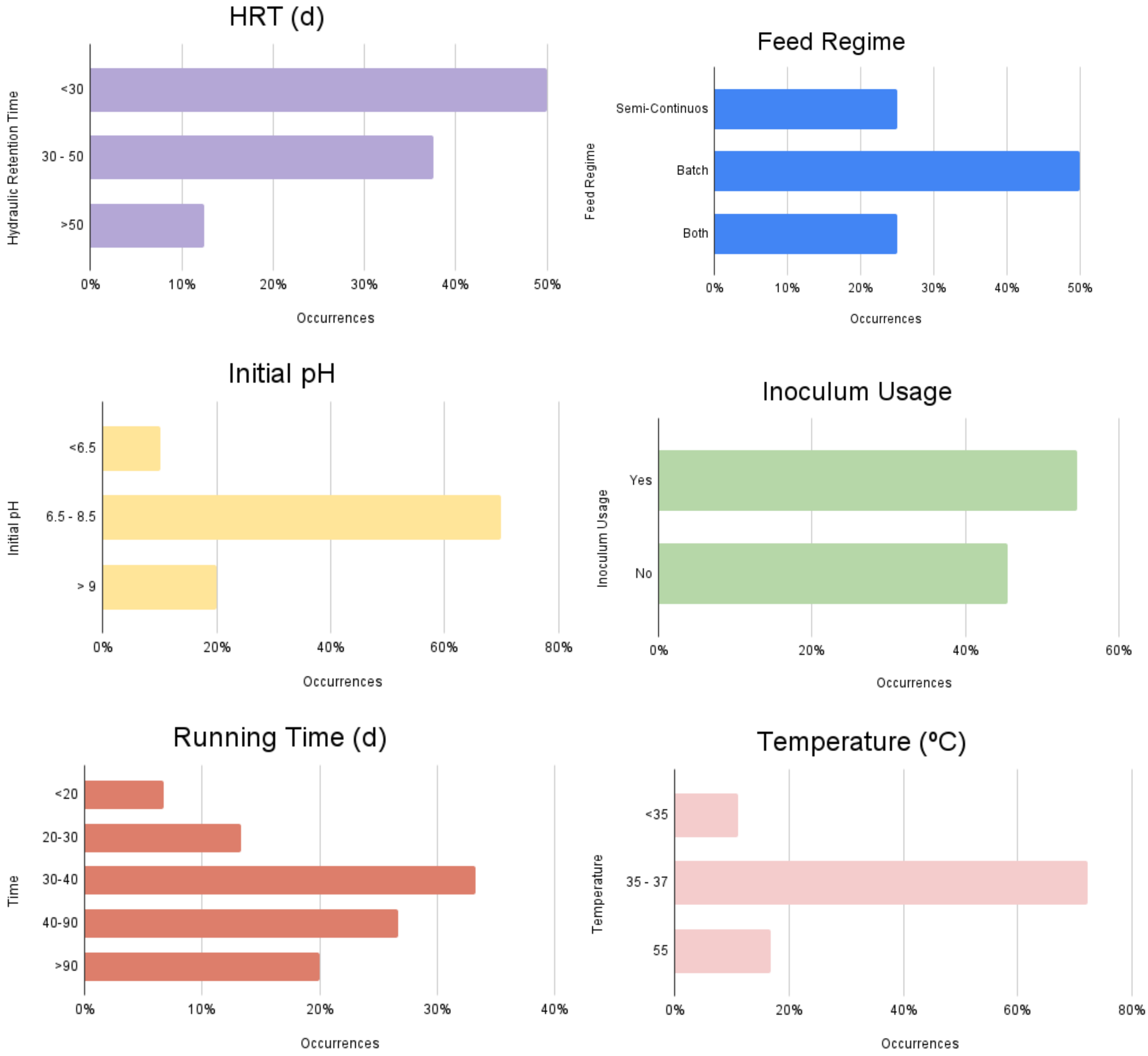
The initial pH of the substrates, in most studies (around 70%), was between 6.5 and 8.5, indicating that the majority of studies did not adjust the pH before the tests, using the natural pH of the substrates, which already presents a neutral characteristic and enables AD.

The use of inoculum was highly variable among the article collection; nearly half (55%) used inoculum to initiate the AD process, while the other half (45%) did not. The use of inoculum directly influences biogas yield, process stability, and microbial community dynamics. Inoculum with a stable active microbial community can effectively avoid AD initiation failure and establish a symbiotic network with more positive interspecific relationships to maintain AD stability and achieve efficient methane production (Chen et al., 2024). The studies that used inoculum did not describe the ratio between substrate and inoculum, and it was not possible to observe a direct outcome between the studies that used inoculum and a higher biogas production.

The running time was highly variable and strongly dependent on the operational conditions mentioned above. For most studies, the time was measured until biogas production was no longer relevant, and different parameters were used to determine when production ceased to be significant. Overall, the durations ranged between 30 and 90 days.

The operational parameters influence in different aspects, including the biogas production and content and the digestate characteristics. The following topics will discuss aspects that enhance biogas production and content such as operating temperature, pretreatments and toxic compounds dilution (Chen et al., 2024; Obileke et al., 2024).

Figure 4 - Summary of AnCod of sewage sludge with different manure operational parameters.



Source: Author

2.3.4.1. Operating Temperature

AD can be carried out under different temperature regimes; processes are commonly operated within mesophilic conditions, where digestion typically occurs at 37ºC or thermophilic conditions at temperatures of approximately 55ºC. There is also the psychrophilic condition that occurs at approximately 15ºC but is not widely used (Appels et al., 2008; Zhang et al., 2021). Each temperature range influences the microbial community structure, reaction kinetics, and overall process performance. The choice between temperature regimes is based on the balance between energy requirements and process efficiency.

Psychrophilic conditions are usually applied in cold-climate countries, wherein wastewater temperatures remain below 20 °C for most of the year and heating the wastewater would require significant energy input, increasing operational costs. The key benefit of this condition relates to its cost efficiency. Since all biological processes are temperature-dependent, lower operating temperatures negatively influence both microbial metabolism and reaction kinetics. In particular, psychrophilic ranges significantly reduce the hydrolysis rates of lipids and proteins, resulting in slower substrate degradation, usually leading to lower methane yields. Consequently, most of the AD research is conducted under mesophilic or thermophilic conditions, where process stability and biogas production are more favorable (Akindolire et al, 2022; Tiwari et al. 2021).

Mesophilic conditions are the most widely adopted in AD due to their operational stability and lower energy demand over thermophilic conditions. The lag phase observed in mesophilic digestion is shorter than that in thermophilic digestion, indicating that microbial communities require less time to adapt to the environment (Zhang et al., 2021). Also, the digestate produced under mesophilic conditions generally exhibits better dewaterability compared to that produced under thermophilic conditions (Zhou et al., 2023). However, pathogen removal under mesophilic conditions is often limited (Álvarez-Fraga et al., 2025). Specifically, the AD of SS at 35 ± 1 °C led to a decrease in *Enterobacteriaceae and E. coli* counts in the fiber fraction by only slightly more than one logarithmic unit (Borowski et al., 2013).

In thermophilic conditions, higher temperatures affect the partial pressure of hydrogen within the digester, thereby influencing the kinetics of syntrophic metabolism. There are notable advantages, including increased solubility of organic compounds, which enhances the rates of biochemical reactions and supports higher organic loading rates, higher methane production rates, and pathogen removal (Kunz and Saqib, 2016; Sun et al., 2013). Themophilic conditions offer better destruction of pathogens such as *Salmonella sp., E. coli, and Streptococcus faecalis*, which can be inactivated within 24 hours under thermophilic conditions and therefore is recommended for processes involving sanitary effluents, such as SS (Zhou et al., 2023; Borowski et al., 2014).

In thermophilic conditions, the increasing dissociation constant of volatile organic acids can make the digestion process more susceptible to inhibition, as well as the degradation of nitrogen-rich substances, contributing to the production of high concentrations of ammonium.

Zhang et al. (2021) investigated the AnCod of sludge and pig manure at thermophilic and mesophilic temperatures (55 ± 1ºC and 37 ± 1ºC, respectively). The methane content in the total biogas was greater at thermophilic temperatures than at mesophilic temperatures. According to the study, the methane production tends to be higher under thermophilic conditions than under mesophilic ones, a difference that may be primarily attributed to variations in microbial community composition and potentially due to more solubilization (Duan et al., 2012; Almeida Streitwieser, 2017). Furthermore, the thermophilic environment promotes hydrolysis, which facilitates the release of easily degradable substrates and, consequently, improves the efficiency of methanogenesis (Chen et al., 2020b). The highest Ultimate Methane Yield achieved in the study was 264.39 mL g^{-1} VSS added, obtained with a Pig/Sludge ratio of 2:1.

2.3.4.2. Dilution of Toxic Compounds

During AD, the accumulation of toxic compounds can significantly affect microbial activity and process stability. AnCod also contributes to the dilution of these inhibitory compounds, mitigating their negative effects on the anaerobic digestion process. In livestock manures, the main compound that could hinder AD is the ammonia. Ammonia is one of the most critical inhibitory agents, besides playing a crucial role in the AnCoD, acting as an essential nutrient, it can significantly influence process stability and biogas production. Ammonia is primarily released during the anaerobic degradation of proteins and nitrogen-rich organic matter especially in chicken manure due to the decomposition of uric acid and pig manure (Zhang et al., 2014; Borowski et al, 2013). In the literature, different ranges of ammonia concentrations that cause inhibition have been reported. Sung and Liu (2003) have reported that severe inhibition of methanogens is caused at total ammoniacal nitrogen (TAN) concentrations above 4,000 mg/L. Borowski et al. (2014) reported that the suggested values in literature vary between 70 and 250 mg/L whereas Xu et al. (2020) found a range of 1,700–6,000 mg/L.

Despite the different concentration ranges, the free ammoniacal nitrogen (FAN) is more toxic, and therefore, potential inhibition begins at lower concentrations compared to TAN. Concentration of FAN above 45–100 mg/L has been previously reported to have an inhibitory effect on the methanogenesis process. The high FAN concentration is usually due to the accumulation of VFA, and will entail an increase in pH and total alkalinity, which subsequently leads to changes in the hydrolytic bacterial community (Xu et al., 2020; Zhou et al., 2023).

The acetotrophic methanogens (i.e., *Methanothrix, formerly Methanosaeta*) are generally the dominant methanogens in AD reactors and are extremely sensitive to ammonia; the acetate metabolism can be inhibited and results in the massive accumulation of acetate and subsequent deterioration of the AD (Zhang et al., 2022). Thermophilic methanogens may be more tolerant to FAN than mesophilic methanogens, although lowering the temperature can decrease FAN to relieve ammonia inhibition (Wang et al., 2022).

Wang et al. (2022) evaluated AnCod with excess sludge and chicken manure in different ratios under thermophilic and mesophilic conditions. The sample with the higher portion of chicken manure (2:1 of chicken manure and SS) went to exorbitant TAN (approximately 10,000 mg/L) and FAN (>5,600 mg/L), and lead to ammonia inhibition, resulting in a low methane yield. The codigestion with SS promoted a more balanced C/N ratio and could alleviate the ammonia inhibition.

2.3.4.3. Pretreatments To Enhanced Biogas and Methane Production

Pretreatment strategies have been widely investigated as effective approaches to enhance biogas production in AnCod. These methods aim to break down complex materials to improve the hydrolysis step, which is often considered the rate-limiting phase of the process, by increasing the solubilization and availability of organic matter for microbial degradation.

Various pretreatments have been employed individually or in combination before AD. Moreover, previous studies have reported improvements in biogas production with thermal, physical, chemical, and biological pretreatments (Atelge et al., 2020). Neumann et al (2016) reviewed pretreatment methods applied to sewage sludge including mechanical, chemical and biological strategies.

Based on the authors’ review, mechanical approaches such as ultrasound and high-pressure homogenization (50–600 bar), have demonstrated biogas yield enhancements of 4–83% and 17–115%, respectively, with ultrasound also increasing methane content by approximately 14–260%. Chemical pretreatments have also shown promising results, including alkaline treatment, which can raise methane production by 13–120%, acid pre-treatment with biogas improvements of 12–32% and ozonation, which increases biogas production by 8–200% and methane content by 5–80%.

The addition of enzymes is a biological strategy that demonstrated a modest biogas yield increase of about 12% and a methane improvement of 3% in the study review. Also, the dual digestion process has been operated as an alternative to enhance biogas and methane production. Collectively, these pretreatment techniques improve sludge biodegradability, optimize hydrolysis, and significantly boost the overall performance of AD systems (Neumann et al., 2016).

Chen et al. (2022) investigated the impact of the acid, alkali, and thermal pretreatments on the AD of waste active sludge. The methane yield of the control sample, without any pretreatment, was 64.98 mLCH4/gVS, whereas the pretreated samples increased by 101.8% (thermal), 98.2% (alkali), and 53.7% (acid), with 130.78 mL/gVS, 128.44 mLCH4/gVS, and 99.64 mLCH4/gVS, respectively.

Al Ramahi et al. (2021) evaluated the effect of hydrothermal carbonization (HTC) as a pretreatment on the AD of dairy sludge. As a result, there was a significant increase in COD solubilization, indicating higher degrees of sludge biodegradability after HTC pretreatment. The biomethane potential in the control sample (without pretreatment) was 152 ± 30 mLCH4/gCOD due to the lack of easily accessible substrates. Whereas the pretreated essays' biomethane potential increased because of the higher availability of the organics, with potentials of 363 ± 10 mLCH4/gCOD (for 180ºC) and 444 ± 21mLCH4/gCOD (210º C). Higher methane production after HTC is a clear indicator of the increased availability of organic substrates within the biomass, which leads to an enhanced conversion of the organics during methanogenesis (methane production). However, the VFAs concentrations at 240 °C did not contribute to the methane yield potential, probably due to the inhibitory effects caused by the toxic compounds generated at high HTC temperatures, with a potential of only 158 ± 9 mLCH4/gCOD.

Not many studies on the AnCod of sludge and manure that used pre-treatments were found. Li et al. (2024) studied the AnCod of sewage sludge from activated sludge and cattle manure. Three digesters were operated under different conditions. Digester 1 served as the control, with no pretreatment applied. Digester 2 underwent ultrasonic pretreatment at 100 W and 100 kHz for 60 minutes, whereas Digester 3 was subjected to alkaline pretreatment for one week prior to digestion. The initial COD values in Digesters 2 and 3 were significantly higher than in Digester 1, reflecting the solubilization effects of ultrasonic and alkaline treatments. During operation, COD removal rates reached approximately 89% in Digester 2, compared to 61% and 59% in Digesters 1 and 3, respectively. Although Digesters 2 and 3 exhibited higher biogas production peaks during the initial stages, by day 45, biogas production had stabilized, and Digester 1 achieved the highest cumulative yield, exceeding 100 mL $(L \cdot d)^{-1}$, demonstrating that in the experiment the rate-limiting step of the AnCod was methanogenesis not the hydrolysis.

Pre-treatments applied prior to AnCod can significantly enhance biogas yields and methane production rates by improving substrate biodegradability and accelerating hydrolysis. However, depending on the operations parameters, conventional AD may be more cost-effective (Beinabaj et al., 2023) due to the implementation costs, especially at full scale. Thermal pre-treatments that rely on high temperatures tend to be more costly due to their substantial energy requirements, while chemical pretreatments may generate toxic intermediates associated with toxic compounds formation. Also, the recovery of these acids after pretreatment is a key for economic feasibility (Khan et al., 2022).

2.3.5. Digestate

The digestate is another byproduct of AD and can play a relevant role in the sustainability of biogas systems. It consists primarily of water and many inhibitory and value-added compounds such as ammonia, VFAs, medium-chain carboxylic acids, and lactic acids offering a significant potential for use as a biofertilizer in agriculture, contributing to circular economy practices by returning valuable nutrients to the soil (O’Connor et al, 2022). Unlike untreated organic waste, digestate is more stabilized and has reduced pathogen content and odor, making it safer and more environmentally acceptable for land application. Its composition varies depending on the type of substrate and digestion conditions, which may require posttreatment processes.

Zhang et al (2021) used the dewatering posttreatment to reduce total volume of digestate, which facilitates its further transport, management and final disposal. They studied the Normalized Capillary Suction Time (NCST) to evaluate the performance of the digestate’s solid-liquid separation during the dewatering process. In the case of SS and manures, other types of posttreatment may be required due to the pathogens and emerging contaminants from antibiotics used for medical and growth purposes (Nolvak et al, 2016). Depending on the AD conditions, most of the pathogens can be removed, but the number of fecal coliform bacteria may remain high and require post-treatment, since it would restrict agricultural use (Lamolinara et al., 2022). A promising method for transforming digestate into a nutrient-rich liquid free from pollutant particles is membrane filtration. This technique allows for the removal of pathogenic bacteria and viruses, depending on membrane pore size, and can be combined with processes such as leaching and acidification to further adjust the nutrient composition (Silkina et al., 2017).

Several techniques have been explored for ammonia recovery from digestate, such as ion exchange, gas stripping, membrane contactors, and electrochemical or bio-electrochemical systems. For the extraction of volatile fatty acids (VFAs), both non-membrane methods (including stripping, adsorption, and solvent extraction) and membrane-based processes (such as microfiltration, nanofiltration, electrodialysis, pervaporation, and membrane contactors) have been investigated (Aung et al., 2024, Yang et al., 2022) One example of membrane use for nutrient recovery is the ammonia stripping, one method that recover ammonium nitrogen from the liquid phase digestate by heating and increasing the pH. The free ammonium nitrogen is scrubbed into acid solution, usually with nitric or sulfuric acid, to form ammonium nitrate or ammonium sulfate that can be used as a fertilizer with relatively low organic contamination (Lu et al, 2021; O'Connor et al, 2022).

2.3.6. Challenges and future prospects

AnCoD plays a crucial role in sustainable waste management and in the transition toward circular bioeconomy systems in Brazil. The main barrier to generating electricity, gas injection, and other alternatives on a national scale is economic feasibility. This review highlights the potential of AnCod of SS with animal manure as a promising strategy for improving biogas production and promoting sustainable waste management in Brazil. The combined use of these residues not only enhances methane yield but also contributes to better nutrient balance, waste stabilization, and VS reduction. However, significant gaps remain. A relevant limitation is the lack of accurate and comprehensive data for Brazil, particularly regarding the management practices adopted in livestock production and the actual number of animals generating manure. For instance, official statistics such as those provided by IBGE (Brazilian

Institute of Geography and Statistics) focus primarily on cattle, swine, and poultry raised for meat production, but do not account for breeding farms, which also produce significant amounts of manure and could represent an important source of co-digestion feedstock. Another research gap is the scarcity of studies conducted under thermophilic conditions, as most experiments in the literature focus on mesophilic ranges. Comparative analyses evaluating the operational stability, biogas yield, and hygienization benefits under thermophilic versus mesophilic conditions are needed to determine the most advantageous operational regime depending on the type of manure and quality of SS. Furthermore, there is a lack of investigations combining co-digestion with pre-treatment techniques, which could potentially enhance substrate biodegradability and improve methane yields. Future research should also focus on region-specific assessments, techno-economic analyses, and pilot-scale applications to bridge the gap between laboratory results and practical implementation.

Addressing these gaps will contribute to a better understanding of the technical and economic feasibility of AnCoD in the Brazilian context and worldwide, supporting the development of scalable, optimized systems for renewable energy generation and sludge management.

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3. CHAPTER 3

Experimental Investigation

This chapter presents the experimental investigation of the anaerobic co-digestion for biogas and fertilizer generation.

ARTICLE 2: ANAEROBIC CO-DIGESTION OF ACTIVATED SLUDGE WASTE (ASW) AND BOVINE MANURE: ASSESSING BIOGAS PRODUCTION AND NUTRIENT RECOVERY

Abstract

This study explored the anaerobic co-digestion (AnCoD) of bovine manure (BM) and activated sludge waste (ASW) as a sustainable approach for biogas generation and nitrogen recovery via membrane distillation (MD). The rapid growth of population and livestock activities has intensified the production of both domestic sewage and livestock effluents, posing environmental challenges. AnCod offers an efficient treatment alternative, converting organic waste into biogas and biofertilizers, while co-digestion enhances process stability and methane yield. The Biochemical Biogas Potential was assessed through batch assays using eudiometer tests according to the VDI 4630 (2006) guideline, and the semi-continuous assay was conducted using a bench-scale reactor operated under progressive organic loading rates. The mixing ratios investigated were 100/0%, 80/20%, 60/40%, 40/60%, 20/80%, and 0/100% (VS/VS) of sewage sludge and cattle manure, respectively. A mixture of ASW with a 20% addition of BM yielded 53.03% gain in biogas production during the batch tests. The semi-continuous reactor produced up to 816.99 mL of biogas daily with Organic Loading Rate (OLR) of 2.0 kgSV/m^3.d and 14 days of Hidraulyc Retention Time (HRT), with a methane content of 75.94%, and achieved a 27.99% reduction in volatile solids. The MD recovered up to 34.38% of ammonia under the optimized conditions (pH adjusted to 9.3). The experiment demonstrated good potential for energy generation through a stabilized process, integrating ammonia recovery into the treatment chain strengthens circular-economy practices by converting digestate into a valuable nutrient source, reducing waste, and improving resource efficiency.

Keywords: Biogas; Sewage Sludge; Bovine Manure; Ammonia Recovery

3.1. INTRODUCTION

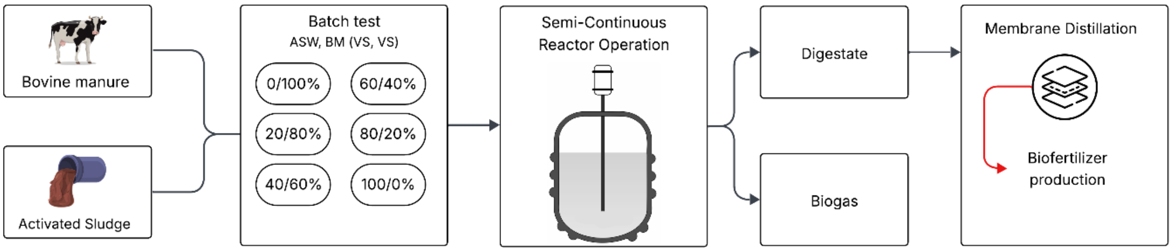
The steady expansion of the global population intensifies a variety of environmental challenges, including the growing demand for food and the rising generation of domestic sewage. One alternative to meet the increasing food necessity is the adoption of confined and semi-confined dairy farming systems, a practice that has been gaining ground in southern Brazil. Although essential for regional agricultural productivity, this production model generates concentrated effluents that may lead to adverse environmental effects, including atmospheric emissions, groundwater contamination, and eutrophication (Bulkowska et al., 2022). At the same time, population growth leads to a proportional escalation of domestic wastewater generation. Among the biological treatment processes applied in Wastewater Treatment Plants (WWTPs), the activated sludge system is one of the most widespread application (Inbar et al., 2023). However, this technology produces large volumes of ASW, which requires proper handling to avoid secondary environmental impacts. For example, in the metropolitan area of Florianópolis (Brazil), managed by the Catarinense Company of Water and Sanitation (CASAN), annual expenditures related to ASW transport and landfill disposal exceed 5.5 million BRL (CASAN, 2023).

Anaerobic Digestion (AD) is recognized as a mature and versatile technology for treating various types of organic waste. It promotes waste stabilization, reduces sludge volume, and converts organic matter into valuable by-products such as biogas and biofertilizers. However, when operated with a single substrate, AD performance can be limited by imbalances in nutrient composition and low biodegradability, particularly in the case of sewage sludge. AnCod, which combines two or more substrates with complementary characteristics, has proven effective in improving process stability, methane yield, and organic matter degradation (Fierro et al., 2014). Among potential co-substrates, BM stands out for its high carbon content, buffering capacity, and availability in large quantities in countries with strong livestock sectors, such as Brazil (Santos et al., 2018).

Globally, the fertilizer industry remains heavily reliant on synthetic fertilizers, and rising global demand continues to drive up their market prices. The industrial synthesis of ammonia through the Haber-Bosch process is highly energy-demanding and represents a significant source of greenhouse gas emissions, contributing to global warming (Dutta et al., 2022). Consequently, the recovery of ammonia from wastewater has gained increasing scientific and industrial attention as a potential alternative ammonia source for fertilizer production. This approach could replace approximately 20–30% of the ammonia currently derived from the Haber-Bosch process (Dutta et al., 2022; Pandey et al., 2021). Recovering nitrogen from AD digestate and transforming it into nitrogen-based fertilizers provides a more sustainable and cost-effective route compared to conventional synthetic fertilizer production, while also offering an additional source of revenue that may reduce wastewater treatment costs (Hu et al., 2024). The process operates through the transfer of NH_3 gas across a microporous hydrophobic membrane submerged in the digestate, where it reacts with a circulating sulfuric acid solution on the permeate side, forming an ammonium salt (Pandey et al., 2021; DUTTA et al., 2022).

Therefore, this study investigates the AnCod of BM and ASW, evaluating the synergistic effects on biogas production and process stability. Furthermore, the potential recovery of ammonia from the resulting digestate using MD is assessed, aiming to promote an integrated approach for energy generation and resource recovery. The summary of the research problem and methodological framework is illustrated in Figure 5.

Figure 5 - Summary of the research problem and methodological framework.



3.2. METHODOLOGY

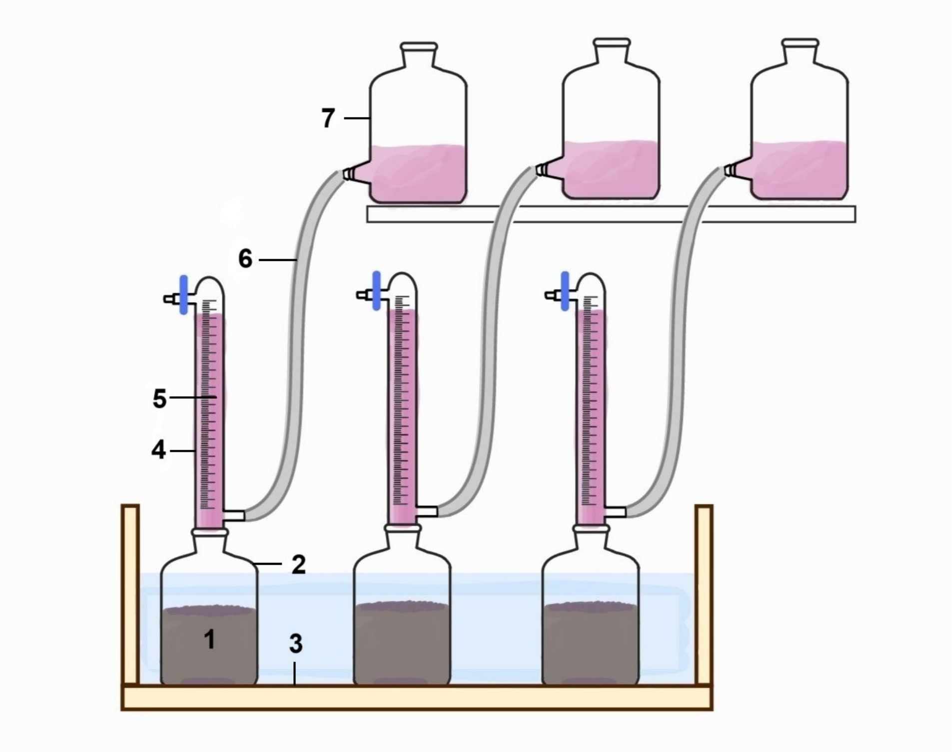
3.2.1. Substrates and Inoculum

The BM was obtained from a dairy farm located in Vacaria, Brazil. In the location, the inoculum was collected from an anaerobic lagoon system that had already been adapted to degrade BM. The ASW was collected from an aerobic biological treatment of the CASAN's WWTP located in Florianópolis/SC. In the same WWTP, the inoculum from the anaerobic digester was collected. The inoculum used in the AnCoD experiments was composed of a mixture of the two aforementioned inocula (50:50, v/v). The samples and respective inocula were characterized immediately upon arrival in the lab for volatile solids (VS) and pH (APHA, 2005).

3.2.2. Batch experiments

Batch experiments were carried out based on the VDI Standard - 4630 (2006) and the experimental apparatus is shown in Figure 6. Each digester had a working volume of 125 mL and was connected to an eudiometer system to measure daily biogas yields. The substrate-to-inoculum ratio followed the same standard, where the quotient of substrate VS to inoculum VS should be lower than or equal to 0.5 to ensure no inhibition due to excess substrate. To enable anaerobic conditions, digesters were flushed with N2 atmosphere for 1 min. The system was maintained at 37°C ± 2°C (mesophilic conditions) and tests were carried out with substrates in different mixture fractions of ASW and BM (VS, VS): 0/100, 20/80, 40/60, 60/40, 80/20, and 100/0. Each condition was performed in triplicate and followed by the blank (inoculum only). Experiments were terminated when the daily gas production was <1% of the total cumulative generation. The observed values were converted to standard temperature and pressure conditions (273 K, 1013 mbar) to eliminate systematic errors related to the quantification of gas production. Water vapor pressure based on tabulated data from the literature (SMITH, 2000) was also considered.

Figure 6 - Experimental apparatus of the eudiometer system



Source: Author

Legend: 1 - substrate + inoculum, 2 - reactionary flask, 3 - thermostatized bath, 4 - eudiometer, 5 - sealing solution, 6 - connection, 7 - Mariotte flask

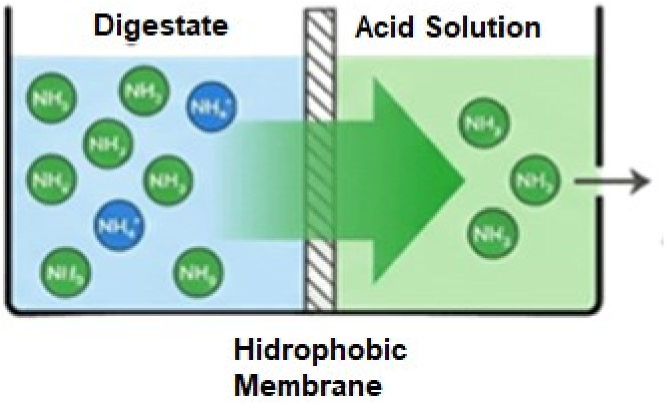
3.2.3. Semi-continuous experiments

The reactor was operated in semi-continuous conditions at 37 ± 1 °C (mesophilic conditions) and followed the same standard (VDI, 2006) to define the substrate-to-inoculum ratio (VS/VS). The reactor was made of acrylic (9.5 cm internal diameter, 29.5 cm in height) with a working volume of 1.5 L. Based on the batch experiments, AnCod reactor was fed with a mixture of 80% ASW and 20% BM (VS/VS). Anaerobic digestion performance was evaluated with three organic loading rates (OLR): 1.0, 1.5, and 2.0 kgSV/m^3.d. The substrates were added to the reactors and withdrawn three times a week and the digester was shaken by hand (about 1 min) daily. The reactor was flushed with N_2 atmosphere for 2 min to keep anaerobic conditions. The biogas readings were monitored by a real-time flow meter (MLima Flow). The observed values were converted to standard temperature and pressure conditions (273 K and 1013 mbar) to eliminate systematic errors related to the quantification of gas production. Water vapor pressure based on tabulated data from the literature was also considered (SMITH, 2000). The characterization of the produced biogas was carried out using a portable gas analyzer (Biogas 5000, Geotech). The gas was collected and stored in a gas sampling bag (BAG 10, Intrutherm), which was connected to the outlet of the gas measurement system. Analyses were performed in triplicate using three bags, each containing approximately 500 mL of biogas.

3.2.4. Membrane Distillation

The MD tests were carried out using a commercial hydrophobic PTFE membrane, filter type 0.2 µm (Fluoropore Membrane Filters, Merck Milipore). The digestate sample was centrifuged at 8,000 rpm for 5 min, and the liquid fraction was further filtered through a 14 µm paper filter to remove any solid particles. Ammonia separation occurred from a feed solution (digestate) at 38 °C into an acidic solution (0.05 M sulfuric acid) in a stripping process for three hours (Reis, 2025). The stripping process involves introducing an ammonia-rich solution into system, where its ammonia content is transferred to the gas. During the MD process, the ammonia nitrogen is converted from its dissolved state (NH4+) in an aqueous solution into its gaseous state (NH3). After volatilization, the ammonia is transferred from the feed side to the stripping solution side through the microporous membrane, based on a partial pressure difference. The stripping solution is designed to capture and trap the gaseous ammonia immediately upon contact, utilizing the acid solution (Dutta et al., 2022). The system consisted of a customized module that housed the membrane, where the feed solution entered and passed through the membrane, and the output was the ammonia, which was directed into the acid solution. The circulation was facilitated by a peristaltic pump. The process is illustrated in Figure 7. To evaluate the influence of pH, MD experiments were conducted using the natural pH of the liquid fraction and at pH 9.3. The pH was adjusted using a NaOH solution.

Figure 7. Illustration of Membrane Distillation Process



3.2.5. Analytical Methods

Total and volatile solids (TS, VS) were analyzed according to the Standard Methods for the Examination of Water and Wastewater (APHA, 2005). The pH was measured using the pH meter (Q402M, Quimis). The alkalinity and acidity were analysed through the Titrimetric/Potentiometric Method according to the Ripley et al., (1986) and Dilallo & Albertson (1961). The total ammonium nitrogen (TAN) was determined according to the Nessler method suggested by Vogel (1981) with a spectrophotometer (DR5000, Hach). The CHN was determined through the elemental analysis (PerkinElmer, model 2400 Series II). The Biochemical Biogas Potential (BBP) of the batch tests was calculated using Equation 1.

$$BBP = \frac{(VN(CumulativeYield)Substrate-(VN(CumulativeYield)Blank}{VS.mSample} \quad (1)$$

The BBP (mLN(Biogas).g.SV^{-1}) represents the relationship between the normalized volume of biogas produced (VN(Cumulative Yield)Substrate), minus the normalized volume produced by the blank (inoculum) (VN(Cumulative Yield)Blank), divided by the product of the mass of VS (gSV.kgsample^{-1}) the mass of the substrate (msample) added to the reaction flask. The possible synergistic effect of the substrates in different mixtures during the biodegradation process was evaluated through Equation 2, following Arribas et al. (2012).

$$a = \frac{Experimental\ production}{Theorical\ productioin} \quad (2)$$

The experimental production is the result of the BPP test on the different mixtures, whereas the theoretical production refers to the calculated production of AnCod based on the monodigestion of each substrate. The result of α indicates:

- $\alpha > 1$; the mixture has a synergistic effect in the final production;
- $\alpha = 1$; the substrates work independently from the mixture;
- $\alpha < 1$; the mixture has a competitive effect in the final production.

Also, the gain percentage (GP) of the mixtures with synergistic effect was calculated using the additional value of experimental BBP obtained in relation to the calculated theoretical BBP. The latter was calculated based on the experimental BBP results of the individually digested substrates (0/100% and 100/0% ASW/BM VS, VS). The conversion factor (f) represents the amount of VS in grams that was converted into biogas in mL per a period, standardized per day, indicating the stabilization of the process (Equation 3).

$$f = \frac{VN}{mSVsubstrato.\Delta t} \quad (3)$$

The VS removal was calculated according to the Kafle et al. (2013), using Equation 4, which considers the inoculum influence.

$$VS\ removal\ (\%) = \frac{(F+I)x\ a - I\ x\ b}{F}\ x\ 100\% \qquad (4)$$

Where F: total VS_{feed} added to reactor (g); I: total $VS_{inoculum}$ added to reactor (g); a: calculated VS removal of feed plus inoculum based on the total initial and final mass of VS present in the reactor (%); b: calculated VS removal of inoculum in blank reactor (%).

3.3. RESULTS

3.3.1. Characteristics of substrates

The characteristics of the substrates (ASW and BM) and inoculum used are described in Table 3.

Table 3 - Characteristics of bovine manure (BM), activated sludge waste (ASW), and mixed inoculum.

Parameter	Unit	ASW	BM	Mixed Inoculum (50:50)
pH	-	7.15	7.16	7.41
TS	g/kg	15.32 ± 0.01	82.28 ± 1.07	26.00 ± 0.29
VS	g/kg	12.46 ± 0.02	67.19 ± 1.13	17.63 ± 0.20
VS/TS	%	81.66 ± 0.00	81.35 ± 0.00	69.87 ± 0.01
TAN	mg/L	253.26 ± 0.90	1,782.36 ± 54.89	-
Carbon	%	43.71	43.48	-
Hydrogen	%	6.13	5.9	-
Nitrogen	%	8.65	1.84	-
C/N ratio	-	5.05	23.63	-

Source: Author

ASW showed an average TS content of 15.32 ± 0.01 g/kg with 81.35% of VS/TS ratio, whereas BM was far denser, with a TS content of 82.28 ± 1.07 g/kg, with 81.66% of these representing VS. The mixed inoculum showed 26.00 ± 0.29 of TS concentration with 69.87% of VS/TS ratio. The two residues and the inoculum presented neutral pH values, which is favorable in the AD process and similar to other studies that reached 7.3 and 7.78 for BM and ASW respectively (Qin et al., 2024; Akbay et al., 2024). The concentration of VS in the studied BM was 67.88 ± 0.09 g/kg, higher than the value reported in the literature of 19.40 g/kg (Bulkowska et al., 2022), possibly due to the presence of forage. Whereas the concentration of VS in the ASW was 9.09 ± 0.07 g/kg, below the reference from other studies, which reached 25 ± 2 g.kg-1, due to the residue origin (secondary treatment).

The TAN of ASW was 253.26 ± 0.90 mg/L, and the BM was 1,782.36 ± 54.89. There are different references in the literature regarding the ammonia concentration at which inhibitory effects begin to occur. Xu et al. (2020) report that the inhibitory effect of TAN on the AD process under mesophilic conditions was found in the range of 1,700–6,000 mg/L, and the BM falls next to the lower limit of this range . Thus, no TAN inhibitory effect was observed in the AD of the present study.

The carbon/nitrogen (C/N) ratio is a critical parameter in AD, directly influencing microbial activity, process stability, and biogas yield. The ASW C/N ratio found herein of 5.05 was similar to those reported in other studies. Xu et al. (2020) found a 5.8 ratio for the primary sludge plus secondary sludge sample, and Zhou et al. (2023) found 6.51 from excess sludge of a sewage treatment plant. The BM C/N ratio found was 23.63, similar to other types of manures, such as swine manure, with a 19.02 C/N ratio (Xu et al., 2020). Based on the literature, for experiemnts using sewage sludge and manure, an efficient ratio may range between 20–30 for stable operation of AD (Borowski et al., 2014). A low C/N (excess nitrogen) can cause ammonia inhibition; while a ratio that is too high (excess carbon) can limit microbial growth due to nitrogen deficiency, and the AnCod presents as a tool to increase this parameter. The ASW C/N value is relatively low and could lead to an unstable process, whereas the BM C/N ratio found in this study represents a good balance as recommended for the stable operation of anaerobic digestion.

3.3.2. Batch experiments

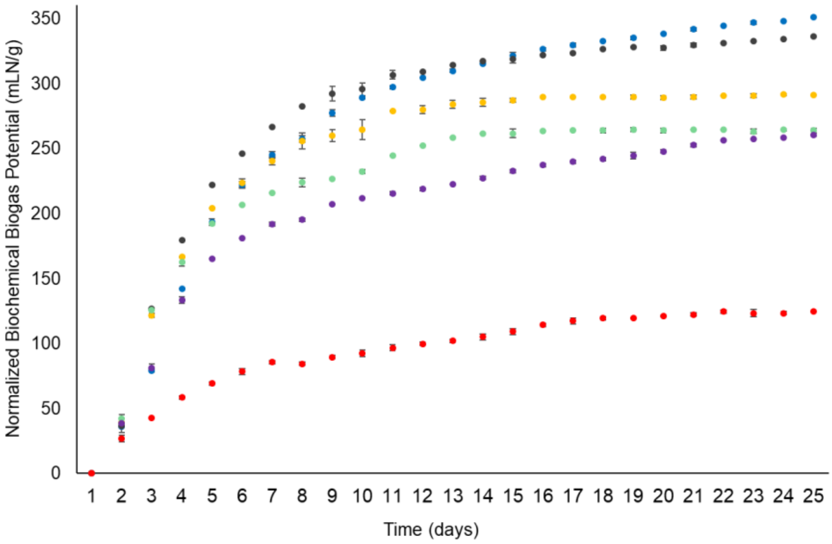
ASW was co-digested with BM in batch mode to establish the optimal composition of both substrates in terms of biogas production (gain percentage, GP) and volatile solids reduction. Data from these investigations are illustrated in Figure 7, showing the biogas curves accumulated over the test days for the six proportions analyzed. The blank tests were used to disregard the contribution of biogas production related to inoculum degradation and have already been discounted from the curves shown in the figure. The average BBP and GP values are shown in Table 4 and Figure 7.

Table 4 - Summary of the anaerobic codigestion batch experiments.

Parameter	Unit	Mixture fractions (ASW/BM%, VS/VS)					
		100/0	80/20	60/40	40/60	20/80	0/100
Duration time	d	28	28	25	25	25	28
Initial VS	g/kg	17.62	27.09	30.04	32.98	34.68	38.30
VS reduction	%	35.70 ± 0.15	42.16 ± 0.03	55.80 ± 0.04	47.82 ± 0.03	44.19 ± 0.05	34.30 ± 0.12
BBP	mL.g.SV^{-1}	124.79 ± 28.47	260.97 ± 23.37	263.70 ± 19.96	291.67 ± 24.32	336.98 ± 14.33	353.32 ± 12.92
Synergistic effect	-	-	$\alpha > 1$	$\alpha > 1$	$\alpha > 1$	$\alpha > 1$	-
Gain Percentage	%	-	53.07	21.97	11.36	9.55	-

Source: Author

Figure 8 – Biochemical Biogas Potential of ASW/BM (VS, VS) mixtures from batch test.



Source: Author

Legend: 100/0% (●), 80/20% (●), 60/40% (●), 40/60% (●), 20/80% (●), and 0/100% (●).

The mixture 0/100 (ASW/BM%, VS/VS) represents the AD of pure BM and presented the highest BBP of the experiments, with 353.32 ± 12.92 ($mL_{N(Biogas)}.g.SV^{-1}$), which is expected due to the higher concentration of VS in the substrate, which initiated the digestion with the concentration of 38.30 g/kg. It is observed that there is an increase in the BBP as the amount of BM increases. However, it should be noticed that the BBP is a measurement of the maximum biogas production capacity, and this value is not observed in semi-continuous systems, as the daily feeding of the system does not allow the complete degradation of the substrate. The AnCoD of the mixture exhibits an apparent synergistic effect in all experiments, benefiting the circumstances of lower nutrient concentrations and lower biodegradable organic matter content in the ASW, thereby improving its biodegradation. This result can be attributed to the increase in the production of volatile fatty acids resulting from the reaction with the insertion of BM, which subsequently increases the readily available biodegradable organic matter for use by microorganisms (Bulkowska et al., 2022). It is clearly seen that a 20% addition of BM (VS/VS) in the compound was sufficient to make such organic matter available, with the highest GP (53.07%), similar to the study of Borowski et al. (2014) that showed an increase of 40% in the biogas production of sewage sludge with the addition of 30% of swine manure. The C/N ratio of the mixture 80/20

(ASW/BM%, VS/VS) resulted in 8.77, an increased ratio due to the carbon content of the BM that contribute in an improved value, although not reaching the efficient ratio found in literature.

The mixture 60/40 (ASW/BM%, VS/VS) showed the highest VS reduction with 60% of ASW and 40% of BM. It is also clear that the synergetic reaction during the codigestion improved the VS percentage of reduction, with values between 42.16% ± 0.03 and 55.80% ± 0.04 whereas the monodigestion values varied between 34.30% ± 0.12 and 35.70% ± 0.15. Similar to the other studies that found a VS reduction of 55.20% during the AnCod of sewage sludge and swine manure, whereas the VS reduction during the monodigestion of sewage sludge was only 34.7% (Zhang et al., 2014). The low VS reduction during the digestion process may be attributed to the high content of slowly degradable and inert materials, such as cell walls in the sewage sludge that has only up to 60% biodegradability (Borowski et al., 2013).

The mixture 80/20 (ASW/BM%, VS/VS) reached the highest GP with the BBP 260.97 ± 23.37 mL.g.SV^{-1} and thus was the mixture chosen to conduct the semi-continuous experiments. This result corroborates with the findings that the proportion of 25/75 of organic substrate for sewage sludge improves the C/N ratio balance and, consequently, improves the biogas production (Xu et al., 2018).

3.3.3. Semi-Continuous Experiment

3.3.3.1. Co-digestion Operational Parameters

The operating parameters and the main results of the semi-continuous experiment are summarized in Table 5 and Figure 8.

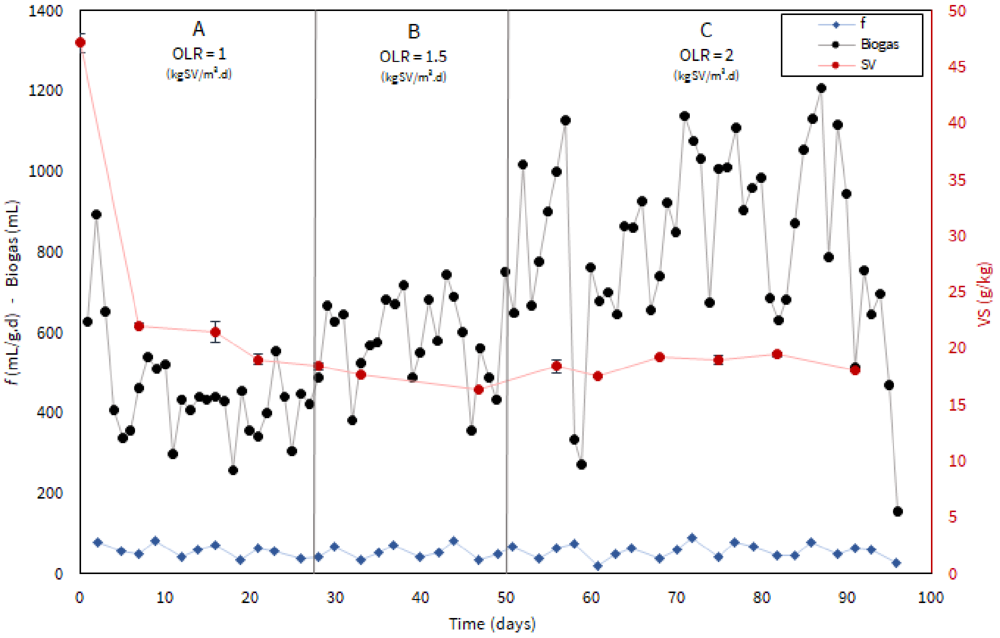
Table 5 - Operational Parameters and results of the semi-continuous experiment.

Parameter	Unit	Stage		
		A	B	C
Duration time	d	28	22	45
OLR	kgSV/m³.d	1.0	1.5	2.0
HRT	d	23	16	14
f	mL/g.d	293.47 ± 85.07	263.29 ± 46.49	284.86 ± 75.10
Biogas Daily Vol.	mL	451.78 ± 126.25	589.78 ± 111.39	816.99 ± 237.25
VS reduction	%	14.35 ± 0.08	27.99 ± 0.04	21.22 ± 0.03

Source: Author

The semi-continuous experiment lasted 95 days with Organic Loading Rate (OLR) increasing from 1.0 to 2.0 by decreasing the hydraulic retention time (HRT). Initially, the reactor was operated with the longest retention time of 24 days, with an OLR of 1.0 kgSV/m³.d to minimize the acclimation period (stage A). The HRT was shortened to 16 days with an OLR of 1.5 kgSV/m³.d to stage B, then during stage C, the HRT was 14 days with the highest OLR of 2.0 kgSV/m³.d. The conversion factor (f) ranged between 263.29 and 293.47 mL/g.d, varying according to the OLR and the feeding regimen.

Figure 9 - Semi-continuous Bench-Scale Reactor Operational Parameters.



Source: Author

The semi-continuous operations demonstrated good stability and high biogas production. The conversion performance also showed favorable values. The conversion factor remained relatively constant from the early days of operation under stage A, with an average of 293.47 ± 85.07 mL/g.d, producing an average daily biogas of 451.78 ± 126.25 mL. These findings align with those observed in the literature for similar substrates. For instance, Borowski et al. (2014) reported a conversion rate of 316 ± 120 dm³/kg·d with a HRT of 30 days in a reactor fed with 70% sludge and 30% swine manure, while Maragkaki et al. (2018) achieved 259 ± 21 mL/L·d using a mixture of 95% sludge and 5% sheep manure.

During stage B, the conversion factor remained steady (f = 263.29 ± 46.49 mL/g·d), indicating that the microbial consortium had adapted well to the substrates. Meanwhile, the average daily biogas production rose to 589.78 ± 111.39 mL, consistent with the higher supply of organic matter.

After maintaining one complete HRT to confirm system stability, a second increase in organic loading was applied 22 days later, initiating stage C. At this stage, the OLR reached its highest value of 2.0 kgVS/m³·d, while the HRT was reduced to 14 days. Operating too short HRT can lead to a biomass washout and potentially methanogenic archea caused by too large dilution rate and therefore no further load increases were introduced (Attar, 2020). Instead, the feeding regime was maintained for three HRTs to monitor operational performance. As in the previous stages, the conversion factor remained stable, averaging 284.86 ± 75.10 mL/g·d, which demonstrated consistent reactor performance and a strong synergistic relationship between the two substrates and their microbial populations, with no inhibitory events. Biogas daily production continued to rise, averaging 816.99 ± 237.25 mL, confirming the positive effect of the increased organic load. The daily biogas volume increased in proportion to the organic loading rate, proving the system’s robustness and adaptability to higher substrate concentrations. The peaks in biogas production were related to the feeding day intervals, most likely due to the degradation of fast biodegradable compounds.

The VS reduction is a critical measurement used to evaluate the efficiency and performance of the AD process and directly affects the generation of biogas. During the digestion process, microorganisms break down part of the organic solids into biogas, which leads to a noticeable reduction in volatile solids (VS). A higher decrease in VS typically reflects more efficient substrate conversion and overall digestion effectiveness. In AnCod systems, the synergistic interaction between different feedstocks can further improve VS breakdown, resulting in increased biogas production and enhanced process stability. The VS reduction started with 14.35% during stage A, achieving the highest rate during stage B with 27.99% and then decreasing to 21.22% during stage C, with the greater value of OLR, probably due to the HRT reduction, which gave microbes less time to degrade solids, leading to incomplete VS reduction. When it comes to sludge treatment through AD, co-digestion has shown significant benefits in enhancing volatile solids reduction. Leite et al. (2023) operated an anaerobic digester, at mesophilic temperature, with only activated sludge waste and achieved a VS reduction of 9% with an HRT of 17 days, while the co-digestion experiment with BM in this study performed a VS reduction of 27.99% for 16 days of HRT. The AnCod of sludge with BM showed an improvement of around 211% in VS reduction, when comparing both studies, probably enabled by the biodegradable content of the manure, since sewage sludge has a high content of slowly degradable and inert material (e.g., cell walls), which has been reported to be up to 60% biodegradable (Mahmoud et al., 2014).

Other AnCod studies achieved higher VS reductions, with values around 42.93% (Jasinska et al., 2024) and 43.16% (Borowski et al., 2013) with poultry manure. The poultry manure contains a large fraction of easily degradable organic matter, including proteins and simple carbohydrates, leading to higher VS reduction compared to more fibrous manures like cattle or swine manure, especially due to the grass that is usually collected together with the manure, that has a high content of lignocellulose, with slow biodegradation (Zhang et al., 2014; Ziganshina et al, 2022).

Table 6 – Summary of the digestate characteristics

Parameter	Unit	Stage		
		A	B	C
Total Alkalinity	mgCaCO3/L	2,038.82 ± 30.81	1,739.48 ± 41.25	1,711.99 ± 26.41
Total Acidity	mg/L	441.36 ± 0.00	385.42 ± 61.49	486.00 ± 98.19
Acidity/Alkalinity ratio	-	0.22	0.23	0.28
Volatile Solids (VS)	g/kg	20.23 ± 1.77	15.89 ± 2.07	18.61 ± 0.73
pH	-	7.1 ± 0.05	7.3 ± 0.16	7.6 ± 0.13

Source: Author

The alkalinity functions as a buffering agent, neutralizing acids produced during digestion and preventing rapid pH drops that can inhibit methanogenic microbes. Adequate alkalinity supports a stable pH, which is crucial for efficient microbial activity, whereas low alkalinity can lead to acid accumulation and process failure, while excessive alkalinity may inhibit certain microbial groups (Xiao et al., 2024). The total alkalinity of the experiment decreased with the increase of OLR and reduction of HRT (from 2,038.82 to 1,711.99 mgCaCO3/L), similar to the Leite et al. (2023) study, which went from 1,772 to 1,275 mgCaCO3/L with the decrease of HRT from 50 to 9 days. Sewage sludge usually has low alkalinity. Chen et al. (2012) also reported that the highest alkalinity value around 2,740 mgCaCO3/L, was achieved at 25 days of solid retention time with a mesophilic anaerobic digester treating sewage sludge. Since the reactor was operated with 80% of sewage sludge (VS/VS), it was expected to have a low alkalinity.

Other studies of AnCod achieved higher alkalinity concentrations. Fierro et al. (2013) found a concentration between 7,000-8,750 mg CaCO3/L with a 10:1 ratio of poultry manure and sewage sludge (v/v). Jasinska et al. (2024) found a concentration around 11,080 mg CaCO3/L with 60% of poultry manure (VS/VS), both at mesophilic conditions, expected due to the higher buffer capacity of manures.

The acidity/alkalinity ratio is an indicator of the AD performance. The ratio value below 0.3 represents a good AD performance and operation. Values between 0.3-0.5 represent a deficiency in the digestion process, and values over 0.8 indicate an over-acidification and collapse risk (Andreoli et al., 2001). During all stages of the experiment, the ratio remained within stable limits (0.22 – 0.28); however, it was noticed that the value increased with the OLR, reaching a 0.28 ratio in stage C, close to the stable limit considered. The value reflects the possible destabilization of the reactor at low HRT.

The pH value is a critical parameter that determines the stability, efficiency, and overall success of anaerobic digestion due to the high sensitivity of the microbial communities involved in the process During the semi-continuous experiment, the pH was maintained within the optimal functional range, often cited between 6.5 and 7.5, since methanogenesis generally occurs most successfully in a neutral environment (Geißler et al., 2019; Sanaye et al., 2022). The pH, alongside other indicators, shows the process stability, and it resulted in a positive outcome, demonstrating the reactor robustness. The slight increase in pH, with increasing OLR, was probably due to the degradation of VFAs and the production of ammonia nitrogen.

3.3.3.2. Biogas Characterization

Regarding the biogas content, the literature reports that the methane percentage does not change significantly with increasing organic loading within the same anaerobic digester operating under mesophilic conditions (Borowski et al., 2014; Zhang et al., 2021). Therefore, in this study, biogas characterization was performed using the highest applied OLR (2.0 kgVS/m^3·d).

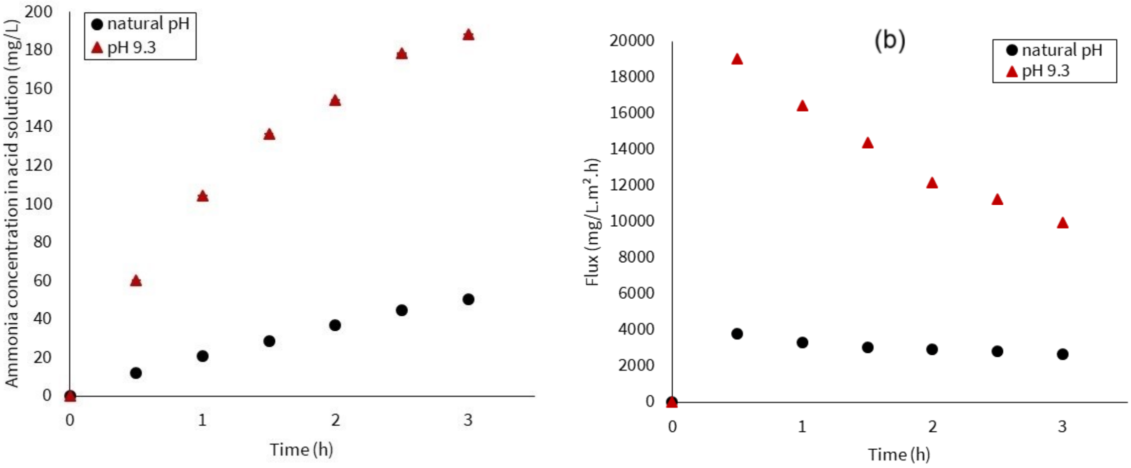
The quantified carbon dioxide (CO_2) was 24.04% and the methane concentration (CH_4) was 75.94%, highly favorable, particularly when compared to other studies involving livestock manure and sewage sludge. Borowski et al. (2014) reported values between 67 and 68% of methane concentrations for both reactors utilizing 70% of sewage sludge with 30% poultry manure and 70% of sewage sludge with 20% of pig manure and 10% of poultry manure (w/w). The methane concentration for sewage sludge with municipal solid waste was also similar. Ghosh et al. (2020) reported a methane yield of around 69% using batch experiments under an optimal ratio of 40% solid waste and 60% sludge (w/w). Leite et al. (2023) found a methane concentration of 64% for a mono-digestion reactor of activated sludge waste at mesophilic conditions, with a similar HRT of 17 days. Compared to Leite et al (2023) results with mono-digestion, the AnCod experiment increased the methane content by 18,66%. From a wastewater sludge management perspective, the AnCod results are highly favorable, as they substantially increase methane concentration. Stoichiometry schows that methane percentage can vary in the range of 50 to 75%, depending on the substrate composition. This improvement is particularly relevant because methane percentages in biogas from sludge mono-digestion are typically low due to low readily biodegradable organic matter and the presence of inhibitory substances (Dai et al., 2017).

The Hydrogen Sulfide (H_2S) concentration found in biogas was 0.0121% (121 ppm) and is a key parameter when analysing biogas characterization. The compound causes severe catalyst poisoning, equipment corrosion, reduced energy yield, and process inhibition, interfering with the biogas use for electricity, heat, and energy utilization. Thus, its monitoring is essential during AD, and regulatory limits have been set for the applications of biogas as an energy resource or transportation fuel. The limit for direct combustion is 1,000 ppm, whereas the limits for internal combustion engine fuel and compressed natural gas for transportation fuel are 100 and 16 ppm, respectively (Khanal and Li, 2017). The percentage of H_2S in the biogas in this study corresponds to 121 ppm, and complies with the limit for internal combustion engine fuel. The literature reports that the H_2S concentration is generally higher in sludge mono-digestion, with average values of 5.89 ± 7.83%, whereas in co-digestion it typically ranges between 0.02 and 1.00% (Jasinska et al., 2024).

3.3.3.3. Membrane Distillation

Figures 9 illustrate the behavior of the ammonia concentration in the stripping solution and the ammonia flux over the two membrane distillation experiment.

Figure 10 - Ammonia concentration of stripping solution with the feed solution at natural pH (7.9) and pH 9.3 (a), and Ammonia flux behavior at natural pH and pH 9.3



Source: Author

The digestate was originated from the semi-continuous experiment stage C, with 2.0 kgSV/m³.d OLR. The digestate natural pH was around 7.9, and the MD was conducted without any adjustments. At the end of the process using the natural pH (~7.9), the stripping solution reached ammonium concentrations around 50.23 mg/L, starting with sulfuric acid, representing an ammonia recovery of 6.05%. A similar result was found by Guo et al (2019), achieving 4.5% of ammonia recovery at pH 7 using a Nafion 8% membrane at 45°C.

In membrane separation processes, the flux is defined as the rate of mass transfer or flow of a substance across the membrane area over time, driven by concentration or partial pressure differences across the membrane, as well as pH, temperature, and other factors. The flux with natural pH reached an average of 3,781.89 mg/L.m².h and ended the process with 2,645.78 mg/L.m².h, an expected decrease due to fouling and membrane wetting, which can limit long-term flux performance (Ma et al., 2023).

In an alkaline environment (pH>9.3), a substantial portion of dissolved ammonia and ammonium ions rapidly dissociate in aqueous solution to form NH_3 molecules (Guo, 2019). Thus, ammonia flux was also higher for the higher pH, reaching an initial flux of 19,019.79 mg/L.m².h and 9,941.75 mg/L.m².h at the end of the process, which led to a better ammonia recovery of 34,38% at pH 9.3, with 188,73 mg/L of ammonia concentration in the stripping solution. The results found corroborate with other studies, Guo et al. (2019) reached an ammonia recovery of 27.2% using a Nafion 8% membrane at 45°C with pH of 9 and Xu et al (2021) also found an increasing recovery with a higher pH, from 16.0 ± 2.0% of recovery with no pH adjustment (7.44) to 84.2 ± 1.9% at pH 12, both at 60°C. Both the ammonia recovery efficiency and the flux were clearly determined by the environment pH, which had a positive effect, as evidenced by the results shown. Rivera et al (2022) also found higher ammonia recoveries by increasing the digestate pH, nevertheless, an increase in the pH above 10 does not entail large improvements in the ammonia removal. The MD at pH 11 resulted in a NH3 recovery of 82.8% ± 0.4%, only 10% higher than the recoveries recorded at pH of 10, both at 35°C, using synthetic digestate.

The membrane type also influences the process, along with the digestate content. Rivera et al (2022) studied PTFE and PVDF membranes in terms of ammonia recovery due to specific material properties and structural characteristics. Whereas the synthetic digestate demonstrated greater recoveries than the MD with real digestate.

Following the MD process, the resulting acid solution contains ammonium sulfate ($(NH_4)_2SO_4$), which can serve as an alternative source of liquid fertilizer (Dutta et al., 2022). This strategy offers economic benefits while preventing the direct application of raw digestate to the soil, thereby reducing the risk of introducing toxic substances and pathogens.

A preliminary economic feasibility analysis was conducted to compare the produced fertilizer with the commercial fertilizer. The main cost drivers that affect the process were utilized: sulphuric acid consumption and commercial membrane. The consumption of NaOH for pH adjustment was negligible and, therefore, was not included in the cost analysis. Given the minimal volumes required its contribution does not influence the economic assessment of this study. Because this study was conducted at laboratory scale, the economic costs may be overestimated compared to those expected in a full-scale implementation. To minimize this discrepancy and provide a more realistic economic perspective, a quotation for hydrophobic PTFE tubular membranes, representative of the type commonly used in full-scale membrane distillation systems, was adopted. This approach allows for a more accurate projection of the potential costs associated with large-scale operation.

Table 7 presents the costs per experiment and per kilogram of nitrogen recovered for both natural pH and pH 9.3 experiments utilizing regional dependent prices quoted at Florianópolis, Brazil in November, 2025.

Table 7 - Costs per item used in membrane distillation.

Compound	USD ($) Per experiment	USD ($) Per kg recovered (natural pH)	Per kg recovered (pH 9.3)	Reference
Sulphuric acid	0.021	418.2	111.3	Local price quote
Commercial Membrane	2.39	47,570	12,660	Manufacturer
Total	2.41	47,988	12,771	

Source: Author

The local price for ammonium sulfate fertilizer is around $2.05 per kilogram, according to the local price quote (2025), which contains 21% nitrogen. Considering the nitrogen content of the product, the cost would result in $9.76 per kg equivalent of the compound (AgroAdubo, 2025). Although the membrane separation process demonstrated effective nutrient recovery under laboratory conditions, its economic feasibility remains a significant challenge. The operational and maintenance costs associated with membrane systems, including membrane and reagent costs, are considerably higher than the cost of purchasing the nutrient in its commercial form based on the current prices for 2025 in Brazil.

Implementing the ammonia recovery process at full scale would naturally involve higher operational and capital costs; however, substrates with higher ammonia concentrations, such as poultry and swine manure, could substantially increase the mass of recoverable nitrogen, thereby improving process efficiency and reducing the unit cost of recovery (Jasińska et al., 2024; Xu et al., 2020). Although economic considerations remain relevant, the integration of ammonia recovery into AD systems strengthens circular economy strategies, enhances sustainability, and decreases dependence on externally sourced synthetic fertilizers. This broader environmental and resource-efficiency perspective supports the long-term value of adopting such technologies despite their initial financial demands. Therefore, despite its technical viability and potential environmental benefits, the MD process would require additional analysis for its sustainable and feasible application.

3.4. CONCLUSION

The anaerobic co-digestion process of sewage sludge waste with bovine manure can be implemented in existing wastewater treatment plants as a supporting process for sewage sludge disposal. The experiments resulted in a synergistic effect with a biogas gain of 53.07% with the addition of 20% of bovine manure. The operation of a bench-scale semi-continuous reactor demonstrated good potential for energy generation through a stabilized process, reaching a daily biogas production of up to 816.99 ± 237.25 mL with a methane percentage of 75.94% with the highest applied OLR. The MD process recovered approximately 34% of the total ammonia at pH 9.3 in 3 hours, a secondary by-product of AD. However, its related costs require additional process optimization studies. These results establish the basis for future research into applying an AnCoD system as a sustainable and low-energy-cost solution for waste management, energy supply for WWTPs, and stabilization/sanitation of sewage sludge.

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4. CHAPTER 4

Conclusion

This chapter states the conclusions to the findings of the dissertation, proposing suggestions for future research on the theme.

4.1. CONCLUSION

The anaerobic digestion (AD) process plays a crucial role in advancing sustainable waste management and supporting the transition toward circular bioeconomy systems in Brazil. Despite its promise, large-scale implementation still faces economic and technical barriers, including the limited availability of accurate national data on manure generation, the predominance of mesophilic studies over thermophilic investigations, the scarcity of research combining co-digestion with pretreatment techniques to enhance substrate biodegradability, and large-scale studies. Also, to develop a comprehensive sustainability evaluation, the life cycle assessment (LCA) is an important tool that provides a systematic approach to assess the environmental impacts of AD across its entire life cycle, from feedstock production to energy generation.

Experimental evidence from bench-scale semi-continuous reactors demonstrates the potential of this approach: co-digestion of sewage sludge with 20% bovine manure increased biogas production by 53.07%, comparing to the theorical values and achieved up to 816.99 ± 237.25 mL of daily biogas with a methane content of 75.94%, indicating strong operational stability and energy-generation potential. Additionally, the MD process recovered approximately 34% of total ammonia at pH 9.3 within 3 hours, suggesting opportunities to integrate nutrient recovery into AnCoD systems, although economic feasibility remains a challenge.

Overall, these findings reinforce the potential of AnCoD as a scalable sustainable solution for enhancing biogas generation, improving sludge stabilization, and reducing environmental impacts in wastewater treatment plants. The authors are already conducting studies on the anaerobic co-digestion of sewage sludge and bovine manure under thermophilic conditions, as well as developing a LCA of this operation in comparison with the current sewage sludge disposal practices adopted by CASAN.