

Títol del Treball:

**OPTIMIZATION OF ELECTROMETHANOGENESIS: EFFECT
OF OPERATING CONDITIONS UNDER MESOPHILIC
CONDITIONS**

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RESUM

El 2019, més del 90% de la població mundial va viure exposada a una mala qualitat de l'aire. Als espais interiors, on les persones passen la major part del seu temps, es poden assolir concentracions més altes de contaminants com el CO₂, un gas d'efecte hivernacle que contribueix al canvi climàtic. Per això hi ha una necessitat creixent de desenvolupar tecnologies per mitigar el CO₂ atmosfèric i millorar la qualitat de l'aire interior, de gran rellevància per a la salut humana, alhora de ser capaços de transformar el CO₂ capturat en productes de valor afegit que es puguin utilitzar o introduir al mercat, generant beneficis econòmics.

Per abordar aquest problema, es proposa el procés MICRO-BIO com una plataforma integral per capturar CO₂ de l'aire interior i transformar-lo en productes químics valuosos i neutres en carboni, mitjançant la combinació de la captura directa de CO₂ de l'aire amb un sistema bioelectroquímic (BES). Els reactors de tecnologies d'electrosíntesi microbiana (MEST) es basen en microorganismes quimiolitotrofs que poden reduir el CO₂ mitjançant la via de Wood-Ljungdahl de tipus arqueà mediada per H₂.

Les taxes de producció als reactors MEST depenen de diversos factors, com ara els materials del reactor, les fonts d'inoculació i els paràmetres d'operació. Aquest projecte final de grau se centra en estudiar les condicions d'operació d'un BES per a la transformació bioelectroquímica del CO₂ d'espais interiors en CH₄. Per això, es van establir diversos experiments en què es van canviar el temps de residència hidràulic (HRT) i el temps de residència en llit buit (EBRT).

Els resultats obtinguts suggereixen que el BES estudiat va requerir un període d'adaptació de 50 dies perquè la biomassa ajustés el metabolisme al canvi entre el mode d'operació en fed-batch i continu. A més, es van avaluar els efectes de l'HRT i l'EBRT en la conversió de CH₄ i la taxa de productivitat.

Durant l'experimentació, es va subestimar la taxa de productivitat de CH₄ del biofilm, per la qual cosa es proposa l'ús de perles que continguin la biomassa per evitar-ne el rentat en futures optimitzacions. A més, els resultats obtinguts demostren que el reactiu limitant per a la reducció de CO₂ és l'H₂, per això es proposa augmentar el voltatge aplicat en futurs experiments per augmentar la hidròlisi de l'aigua, proporcionant més H₂ al sistema, augmentant la conversió i la taxa de productivitat.

La transferència de massa de CO₂ des de la fase gasosa fins a la fase líquida també va limitar la conversió de CH₄. Per millorar el sistema i evitar limitacions de transferència de massa, cal acoblar un mòdul capil·lar per a experiments futurs.

RESUMEN

En 2019, más del 90% de la población mundial vivió expuesta a una mala calidad del aire. En los espacios interiores, donde las personas pasan la mayor parte de su tiempo, se pueden alcanzar concentraciones más altas de contaminantes como el CO₂, un gas de efecto invernadero que contribuye al cambio climático. Por ello existe una creciente necesidad de desarrollar tecnologías para mitigar el CO₂ atmosférico y mejorar la calidad del aire interior, de gran relevancia para la salud humana, a la vez de ser capaces de transformar el CO₂ capturado en productos de valor agregado que se puedan utilizar o introducir en el mercado, generando beneficios económicos.

Para abordar estos desafíos, se propone el proceso MICRO-BIO como una plataforma integral para capturar CO₂ del aire interior y transformarlo en productos químicos valiosos y neutros en carbono, mediante la combinación de la captura directa de CO₂ del aire con un sistema bioelectroquímico (BES). Los reactores de tecnologías de electrosíntesis microbiana (MEST) se basan en microorganismos quimiolitautótrofos que pueden reducir el CO₂ mediante la vía de Wood-Ljungdahl de tipo arqueano mediada por H₂.

Las tasas de producción en los reactores MEST dependen de varios factores, como los materiales del reactor, las fuentes de inoculación y los parámetros de operación. Este proyecto final de grado se centra en estudiar las condiciones de operación de un BES para la transformación bioelectroquímica del CO₂ de espacios interiores en CH₄. Con este fin, se establecieron varios experimentos en los que se cambiaron el tiempo de residencia hidráulico (HRT) y el tiempo de residencia en lecho vacío (EBRT).

Los resultados obtenidos sugieren que el BES estudiado requirió un período de adaptación de 50 días para que la biomasa ajustara su metabolismo al cambio entre el modo de operación en fed-batch y continuo. Además, se evaluaron los efectos del HRT y EBRT en la conversión de CH₄ y la tasa de productividad.

Durante la experimentación, se subestimó la tasa de productividad de CH₄ del biofilm, por lo que se propone el uso de perlas que contengan la biomasa para evitar su lavado en futuras optimizaciones. Además, los resultados obtenidos demuestran que el reactivo limitante para la reducción de CO₂ es el H₂, por lo que se propone aumentar el voltaje aplicado en futuros experimentos para aumentar la hidrólisis del agua, proporcionando más H₂ al sistema, aumentando la conversión y la tasa de productividad.

La transferencia de masa de CO₂ desde la fase gaseosa hasta la fase líquida también limitó la conversión de CH₄. Para mejorar el sistema y evitar limitaciones de transferencia de masa, se requiere el ensamblaje de un módulo capilar para experimentos futuros.

ABSTRACT

In 2019, over 90% of the global population live exposed to poor air quality, which represents a significant threat to public health worldwide. In indoor spaces, where humans spend most of their time, can reach higher concentrations of pollutants such as CO₂, a greenhouse gas contributing to climate change. Therefore, there is an increasing need to develop technologies to mitigate atmospheric CO₂ and enhance indoor air quality, which has great relevance for human health, but also technologies capable of transforming the captured CO₂ into value-added products to be used on-site or to be introduced into the market, generating economic benefits.

To address these challenges, the MICRO-BIO process is proposed as a comprehensive platform to capture CO₂ from indoor air and transform it into valuable carbon-neutral chemicals by coupling CO₂ direct air capture to a bioelectrochemical system (BES). Microbial electrosynthesis technologies (MEST) reactors rely on chemolithoautotrophic microorganisms that can reduce CO₂ by the H₂-mediated archaeal-type Wood-Ljungdahl pathway.

The production rates in MES reactors are widely variable and dependent on several factors such as reactor materials, inoculum sources, and operation parameters. This final degree project focuses on studying the operating conditions of a BES bioelectrochemical transformation of indoor CO₂ into CH₄. For that purpose, several experiments are established changing the hydraulic residence time (HRT) and empty bed residence time (EBRT).

The acquired results suggest that the studied BES had a 50-day adaptation period of the biomass to adjust their metabolism to the switch between fed-batch and continuous mode operation. Also, HRT and EBRT effects on CH₄ conversion and productivity rate were evaluated.

During experimentation, the biofilm CH₄ productivity rate was underestimated, so to avoid washout, the use of pearls containing the biomass is proposed for future optimization. Furthermore, the obtained results demonstrate that the limiting reagent for CO₂ reduction is H₂, so increasing the applied voltage is proposed for future experimentation to increase water hydrolysis, providing more H₂ to the MES system, and subsequently increasing the conversion and productivity rate.

CO₂ mass transfer of CO₂ from gas to the liquid phase was also limiting the CH₄ conversion. To improve the system and avoid mass transfer limitations, capillary module assembly is required for future experimentation.

ETHICAL REFLECTION

Ethical concerns surrounding biotechnology include equitable access to information, potential ecological harm, the potential disruption of existing markets, and the ethical implications of interfering with nature. Industrial biotechnology finds applications in agriculture, healthcare, and energy sectors, having great potential, but ethical challenges must be addressed in order to harness it responsibly (Odongo et al., 2019).

SUSTAINABILITY REFLECTION

The sustainability of industrial biotechnology can be quantified by measuring the use of resources such as water, the release of toxic substances, the contribution to CO₂ reduction, and so forth (Fröhling & Hiete, 2020; Venkatesh et al., 2020).

Technologies are needed to reduce atmospheric CO₂ and improve indoor air quality (IAQ). One approach is to capture CO₂ and convert it into CH₄, which can be used as fuel for heating. This approach can eliminate the need to extract, transport, and store other fuels. CH₄ produces CO₂ when used, so CO₂ recycling after CH₄ use should also be assessed and considered to close the carbon loop.

All biotechnological applications should be evaluated based on a cost-benefit analysis with different perspectives to establish sustainability indicators. While experimentation in this field is expensive, the innovations resulting from it are more sustainable in the long term.

GENDER PERSPECTIVE REFLECTION

The integration of a gender perspective in the assessment of industrial biotechnology, has been explored through social life cycle assessment (SLCA). Utilizing national accounts' input-output tables, SLCA allows for predicting and quantifying socioeconomic impacts and identifying transmedia effects (Macombe, 2020).

Studies have revealed that gender does not affect scientists' entry into the biotech industry but is closely linked to promotion opportunities. Men tend to have a higher likelihood of obtaining early supervisory positions across various organizational settings, while female scientists are nearly eight times more likely to supervise in biotech firms compared to hierarchical settings. Different organizational forms, such as network-based structures and hierarchies, offer distinct employment experiences for female scientists (Smith-Doerr, 2004).

By incorporating a gender perspective into SLCA, we gain insights into how gender dynamics interact with industrial biotechnology, informing strategies for promoting gender equity and inclusivity in the biotech sector. This holistic approach ensures the responsible and equitable implementation of these technologies.

1. INTRODUCTION

1.1. Research motivation

One of the main courses of action in the near future is to face climate change. Climate change is caused by a wide range of factors, both natural and human-induced, however, one of the most important factors that have increased its effect is the atmospheric concentration increase of greenhouse gasses (GHG) such as methane (CH₄), nitric oxides (NO_x) and carbon dioxide (CO₂). The first two gasses have a 25 and 298-fold higher global warming potential than CO₂ respectively (IPCC, 2014). However, CO₂ emission is the largest contributor and human activities mainly release it. In Europe, the energy sector is responsible for roughly two-thirds of all GHG emissions, and the amount of CO₂ emitted from electricity production can greatly vary in time as a function of the sources used to generate it (Santos et al., 2021). Also, in 2019 the US CO₂ accounted for about 80% of GHG emissions (US EPA, 2021), which was 15% of the global emission, behind China with 30% (Olivier & Peters, 2020).

The prospect of a worsening climatic situation due to global warming is a subject of widespread public concern. In 2019, the Intergovernmental Panel on Climate Change (IPCC) emphasized the importance of CO₂ removal for keeping global warming within 1.5°C (Masson-Delmotte et al., 2021). However, the latest synthesis report of the IPCC (IPCC, 2022) suggests that unless immediate actions and deep emissions reductions across all sectors are performed, limiting global warming to 1.5°C will be unattainable, and even limiting warming to below 2°C will be difficult (Shukla et al., 2022). To redirect this tendency, in the last years several agreements have been signed by numerous governments in order to achieve climate neutrality. Climate neutrality implies bringing net carbon emissions to zero or below, balancing the amount of CO₂ released into the atmosphere with the same amount released in other ways (Leal-Arcas et al., 2023). In July 2021, the European Commission reached an agreement to reduce the GHG by 55% in 2030, and achieve climate neutrality in 2050 (European Commission, 2021).

This implies moving away from fossil fuels such as crude oil and coal, which are still the main energy sources (79%) (IEA, 2020; Singh et al., 2020) and the prevalent feedstock consumed in the chemical industry (85%) (Levi & Cullen, 2018; Skoczinski et al., 2021), towards clean energy sources such as wind, water, and solar power, among others. Thus, it is of high importance to find greener and sustainable carbon sources for a transition to renewable carbon in the next years. Carus et al. (2020) defined as renewable carbon all the carbon sources that avoid or substitute fossil carbon from the geosphere.

According to the Renewable Carbon initiative, there are only three sources of renewable carbon. The first one comes from the biosphere, and can be re-grown, such as all types of biomasses (food crops, non-food crops, side streams, by-products, and biogenic waste) (Kalt

et al., 2021; Serrano-Ruiz, 2020). The second source of renewable carbon comes from the techno-sphere and is obtained by recycling carbon-containing products such as plastics at the end of their life cycle (Bachmann et al., 2021; Shamsuyeva & Endres, 2021). The third source of renewable carbon is the CO₂ that comes either from the techno-sphere or the atmosphere, which can be captured from the exhaust gas of industries (Gabielli et al., 2020; Kätelhön et al., 2019), or directly from the atmosphere, providing an almost endlessly available resource (Goeppert et al., 2012; Marchese et al., 2021; Schellevis et al., 2021). The carbon cycle can be closed by converting industrially emitted or atmospheric CO₂ using carbon capture and utilization (CCU) and carbon capture and storage (CCS) technologies. CCS and CCU technologies could mitigate CO₂ emissions that are difficult to avoid (Bruhn et al., 2016; Wang et al., 2020). CCU could also reduce the extraction of carbon from the geosphere (Carus et al., 2020). CCS uses a combination of technologies to capture CO₂ and transport it to a safe and permanent storage location (Anderson & Newell, 2004; Zhang et al., 2014), which is particularly suited for CO₂ emissions from large point sources such as power plants and cement industries (Valentić et al., 2016; Yang et al., 2021). Despite the important role that CCS play in the mitigation of CO₂ emissions (Peridas & Schmidt, 2021; Tamme, 2021), it can only slow down the increase of CO₂ concentration in the atmosphere at best (Metz et al., 2005; Solomon et al., 2007).

Therefore, in the context of rising concerns about climate change and its mitigation efforts, developing green technologies capable of transforming waste into beneficial products has become a fundamental research challenge. In this scenario, a promising idea appears coupling of CO₂ Direct Air Capture (CO₂-DAC) to a bioelectrochemical system (BES). In the following sections the current state of the art of each technology will be discussed.

1.2. CO₂-Direct Air Capture

1.2.1. CO₂ as renewable carbon source

Although CCS and CCU technologies mostly aim at CO₂ emissions from large point sources (Valentić et al., 2016; Yang et al., 2021), it is estimated that a significant portion of total CO₂ emissions, ranging between one-third and one-half, originates from numerous distributed sources such as commercial and industrial buildings and small sources like transportation vehicles (Ghiat & Al-Ansari, 2021). Given these challenges, there is growing interest in the utilization of CO₂-DAC. CO₂-DAC is a promising approach for atmospheric CO₂ removal (Beuttler et al., 2019). CO₂-DAC technologies use chemical sorbents that are cycled through sorption and desorption for CO₂ removal from ultra-dilute gasses such as air and produce a

more concentrated stream of CO₂ for storage or utilization (McQueen et al., 2021). The sorbents include basic solvents, supported amine and ammonium materials, and solid sorbents that interact strongly with CO₂ (Kothari et al., 2020; Yu & Brillman, 2020).

CO₂-DAC is a challenging process that still needs to overcome several limitations to enhance its versatility and applicability. These limitations include the range of suitable locations for implementation, developing new materials, and establishing smart business cases to reduce operating costs (Beaumont, 2022). While CO₂-DAC is not restricted to specific locations, it does require sources of electrical or thermal energy, as well as water. Additionally, the concentrated CO₂ captured through CO₂-DAC, or conventional, needs to be stored in an appropriate outlet storage site (Jones, 2011). Deep geological formations accessed by a network of pipelines and injection wells have been considered optimal for CO₂ captured (Bui et al., 2018; Keith et al., 2018) followed by mineral carbonation (Gadikota & Park, 2015; Woodall et al., 2019). However, in 2023, as the urgency to minimize or to completely stop the extraction of carbon from the geosphere, it is important to view the CO₂ captured by means of CO₂-DAC as a renewable carbon source rather than a waste to be stored underground.

Another important drawback of CO₂-DAC is the thermodynamic challenge of capturing CO₂ from air at such low concentrations (Lackner, 2013). To capture the comparable amounts of CO₂ to flue gas processes, large air volumes need to be processed (Jones, 2011). However, this drawback can be mitigated if the CO₂-DAC process is installed in environments with considerably higher CO₂ concentrations than atmospheric, such as indoor environments.

Performing indoor CO₂-DAC can reduce the volume of gas that must be processed, thereby lowering the reactor size and operating costs. Increasing focus has been placed on indoor CO₂ direct air capture (iCO₂-DAC) as a tool to minimize the health impact of indoor CO₂ concentration in environments such as office buildings by coupling the CO₂-DAC process into Heating, Ventilation, and Air Conditioning (HVAC) systems (Han et al., 2016; Hu et al., 2017; López de León et al., 2022; Sodiq et al., 2023). This technology allows stabilizing CO₂ concentrations, as well as humidity levels, inside buildings allowing a higher indoor air recirculation rate, limiting the in-take of fresh air, and reducing the load on the HVAC system, so that HVAC/DAC-coupling in recirculation mode provides energy savings of 20-40 % through reduced air conditioning requirements (Beaumont, 2022). Furthermore, the CO₂ captured from the building exhaust air is a renewable carbon source that can be transformed into energy or materials (Baus & Nehr, 2022).

1.2.2. Indoor CO₂: from indoor air pollutant to renewable carbon source

It is important to highlight the relevance of improving indoor air quality (IAQ) by removing indoor CO₂, as the World Health Organization (WHO) identified exposure to poor-quality air as the most important threat to global public health, with more than 90% of the global population in 2019 living in areas where concentrations exceeded the 2005 WHO air quality guideline. For this reason, the new WHO air quality guidelines were released in September 2021 (WHO, 2021). It was the first update of WHO's guidelines since 2005 and takes into account recent evidence of the effect of air pollution on human health (Garland et al., 2021). The new guidelines recommend even lower concentrations of pollutants than previously understood, putting more pressure on nations to reduce air pollution levels (Carvalho, 2021; Garland et al., 2021).

Although indoor and outdoor pollutants are really similar, indoor air pollutants (IAP) concentrations are higher than outdoor air pollutants (Leung, 2015; Shen et al., 2020). IAPs of concern in indoor air include particulate matter, biological organisms, allergens, volatile organic compounds, and inorganic compounds, amongst others (López de León et al., 2018). The COVID-19 pandemic has increased public awareness of the effects of poor IAQ.

Therefore, due to the high concentrations that are reachable indoors, plus the high risk that contaminated air supposes for health, in addition, that humans spend most of their time indoors, there is an increasing need to develop technologies to mitigate atmospheric CO₂ and improve IAQ, but also technologies capable of transforming the captured CO₂ into value-added products.

Indoor CO₂ comes mainly from human metabolism and cooking, although outdoor CO₂ infiltrations contribute largely (Shen et al., 2018; Shen et al., 2020). Also, there is a correlation between air-conditioning and indoor CO₂ levels, related to insufficient performance of mechanical ventilation systems (Assimakopoulos et al., 2017). Indoor CO₂ has been used as an indicator of IAQ based on the direct relation between CO₂ and human occupancy (Olesen et al., 2021). Moreover, as CO₂ is always accompanied by other pollutants, their concentration can be estimated only by knowing the CO₂ concentration (Azuma et al., 2018). For this reason, most of the regulations regarding building ventilation standards are based on CO₂ concentration levels (Schibuola & Tambani, 2020). According to the guidelines set by the WHO, the maximum CO₂ concentration in indoor spaces should not exceed 1000 ppm, and it is recommended to keep it below 800 ppm in areas with significant aerosol generation (Chen et al., 2021). However, buildings with high occupancy density and inadequate ventilation frequently exceed these recommended levels of CO₂ (Rodero & Krawczyk, 2019).

In order to see the range of this problem, CO₂ concentrations from schools, offices, and public transport were briefly reviewed in the literature. In primary schools, a maximum concentration of 3284 ppm was registered in Spain, and in secondary schools, this maximum is even bigger, reaching 5366 ppm. Although these results significantly surpass the recommended thresholds, these elevated concentrations were observed during short periods of time (Becerra et al., 2020). In offices, which are typically well-regulated environments, indoor CO₂ concentrations reach values around 1000 ppm. While these values are considered legally acceptable in many countries, they are still a relatively high CO₂ concentration (Filo & Khammash, 2019). Finally, public transportation, known for its crowded conditions, often experiences increased CO₂ concentrations. In South Korea, p.e., the average CO₂ concentration in underground trains is approximately 1775 ppm (Park & Ha, 2008).

High air CO₂ concentration exposure for both short and long-term can lead to several diseases such as kidney failure, bone atrophy, neurological disorders, and hypercapnia, a condition of abnormally elevated CO₂ levels in the blood, causing respiratory acidosis (Bierwirth, 2018; Patel & Sharma, 2021). Due to the toxicity of CO₂, reducing it in habitable spaces is of great importance for human health. Indoor CO₂ capture generates several benefits. On the one hand, IAQ is improved which has great relevance for human health, and on the other hand, the captured CO₂ can be converted into valuable green chemicals and fuels to be used on-site or to be introduced into the market, generating economic benefits.

1.3. Microbial electrosynthesis technologies for CO₂ transformation

1.3.1. Fundamentals

Microbial electrosynthesis technologies (MEST) in bioelectrochemical cells are one of the most promising carbon capture and utilization technologies under development (Izadi et al., 2021). Among the available CO₂ conversion technologies, bioelectrochemical cells can be easily connected to the electric grid, making them the most suitable option for recycling indoor CO₂ (Grim et al., 2020). MEST like electromethanogenesis has multiple functions, including wastewater treatment, GHG reduction (e.g., CO₂), and renewable energy production (e.g., CH₄) (Van Eerten-Jansen et al., 2012; Lovley & Nevin, 2013).

The electromethanogenesis process is achieved in a bioelectrochemical system (BES), where CO₂ is directly reduced using renewable electricity as a reducing power at the biocathode (Cheng et al., 2009). MEST reactors rely on chemolithoautotrophic microorganisms that reduce CO₂ to carboxylic acids/alcohols and CH₄, respectively (Logan et al., 2019). CH₄ is

produced from CO₂ by Euryarcheota such as *Methanobacterium* and *Methanococcus* sp. mainly by the H₂-mediated archaeal-type Wood-Ljungdahl pathway (Borrel et al., 2016). As shown in Figure 1, methanogenic archaea can harvest the required electrons from the cathode electrode through two different electron transfer pathways (direct or indirect) (Cheng et al., 2009; Villano et al., 2010; Cai et al., 2016).

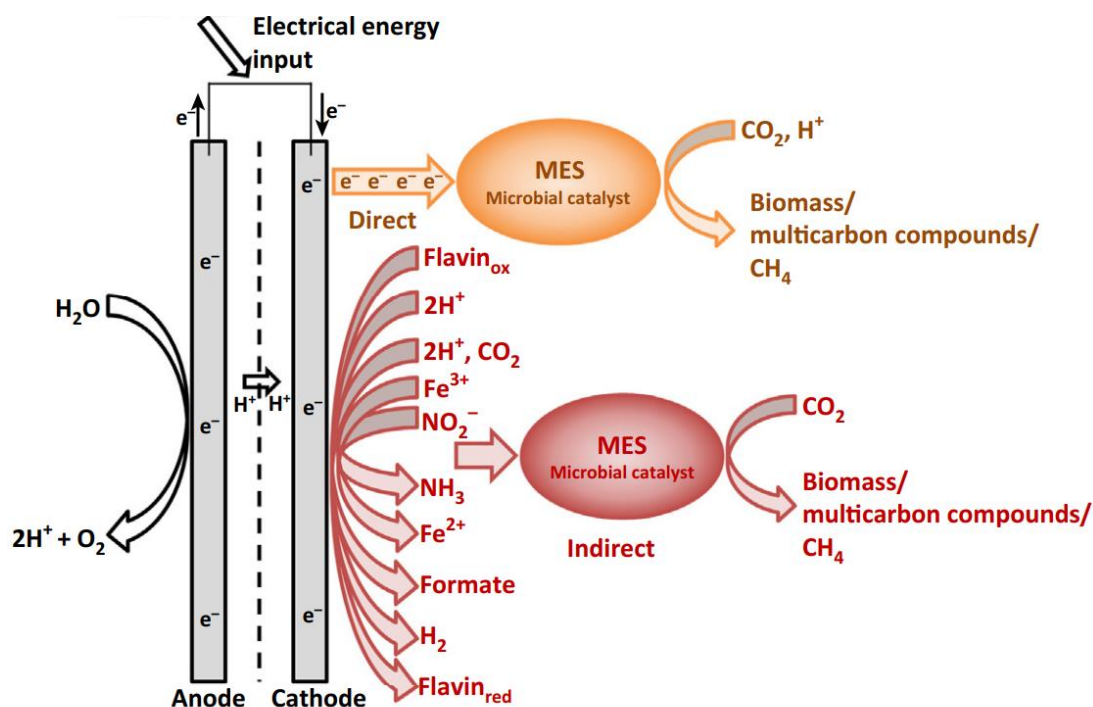


Figure 1. Direct and indirect electron transfer routes during microbial electrosynthesis (MES). The dashed line represents the ion-exchange membrane separating the anode from the cathode in MES. Oxidized (ox), reduced (red). Extracted from Tremblay et al. (2017).

As it appears in Figure 1, methanogenic archaea can take electrons directly from the cathode by direct contact via membrane-bound electron transfer proteins, such as c-type cytochromes, and use them to reduce CO₂ to CH₄ via (eq. 1). Alternatively, H₂ gas is first produced at the cathode, and then the hydrogenotrophic methanogens convert it with CO₂ into CH₄ via (eq. 2). This last via can be done as well by self-generated or added soluble electron shuttles (e.g., proteins such as flavins), or through electrochemically or bioelectrochemically produced mediators such as formate (Fu et al., 2015; Tremblay et al., 2017).



When methanogenic microorganisms are the principal catalyst in MEST, and when the cathode electrode is poised at a potential low enough to enable abiotic H₂ evolution (-0.414 V vs standard hydrogen electrode in theory under standard conditions, but often below -0.6 V due to overpotential) the H₂-mediated pathway often prevails (Tremblay et al., 2017).

1.3.2. Reactor design and operation conditions

Many efforts have been made to optimize the system. The product spectrum and production rates in MES reactors are widely variable and dependent on several factors, including the reactor configuration electrode materials, inoculum sources such as microbial activity, biomass quantity and availability, and operation parameters such as applied potential, pH, temperature, inorganic carbon source, and purity (Lovley, 2011; Dessì et al., 2021). Better microbial adhesion to the cathode and thick effective biofilm formation are also crucial for successful applications of electromethanogenesis (Blasco-Gomez et al., 2017). CH₄ has been produced with high selectivity and Coulombic efficiency (>90%) by mixed cultures at production rates generally higher than those obtained with pure cultures (Jiang et al., 2019). And also, it has been reported that BES using thermophiles have several advantages over mesophilic systems in performance, such as higher reaction activity, greater durability, and wider substrate range (Fu et al., 2015).

The electrode design is a key factor for achieving high production rates in MES reactors. Optimal electrode materials should be cheap, scalable, biocompatible, and conductive, with high surface area, chemical and mechanical strength (Santoro et al., 2017). Flat carbon-based electrodes are cheap and easily scalable but engineering the biotic-abiotic interface for biocompatibility, adhesion, electron transfer, and maximum surface area remains a challenge (Tseng et al, 2022). In fact, the highest CH₄ production rates so far have been achieved by using 3-D structured electrodes, however, the production rate per volume of catholyte was relatively low (0.37 g/L·d) suggesting that further improvement is necessary to make such a design scalable (Jourdin et al., 2016).

So far, the highest CH₄ production rate was 202 L/(m²·d) obtained in an electromethanogenic reactor in which the cathodic chamber was filled with anaerobic granular sludge in contact with a Pt-Ti mesh current collector (Zhou et al., 2021). The granular sludge proved to be a low-cost and easily scalable catalyst for high-rate CH₄ production and demonstrated high tolerance to pH and oxygen disturbances. Customized 3-D printed biocathodes based on conductive carbon hydrogen coated with NiMo-alloy have been recently demonstrated to facilitate electromethanogenesis, reaching an unprecedented specific volumetric production of 2.2 L/(L·d) with 99% coulombic efficiency, showing great promise for scale-up (Kracke et al., 2021).

1.4. Project's global scheme

This research project has been carried out under the framework of the MICRO-BIO process. The MICRO-BIO process is proposed as a comprehensive platform to capture CO₂ from indoor air and transform it into valuable carbon-neutral chemicals by developing a scalable, modular, multi-unit platform process. To do so, this work aims to assess the coupling of CO₂-DAC and MEST as shown in Figure 2. In the set-up of the MICRO-BIO process, three modules can be easily differentiated, which are:

1. CO₂ micro concentrator module (CO₂-MCM), where adsorption and desorption of indoor CO₂ take place.
2. Microbial electrosynthesis module (MESM), where H₂ is electrochemically produced from water hydrolysis, and mixed with CO₂ from CO₂-MCM to produce biomethane (CH₄) through methanotrophic bacteria action.
3. Microbioreactor module (MBM), where the mass transfer capacity of CO₂ from the gaseous to the liquid phase is increased, to enhance the CH₄ production rate.

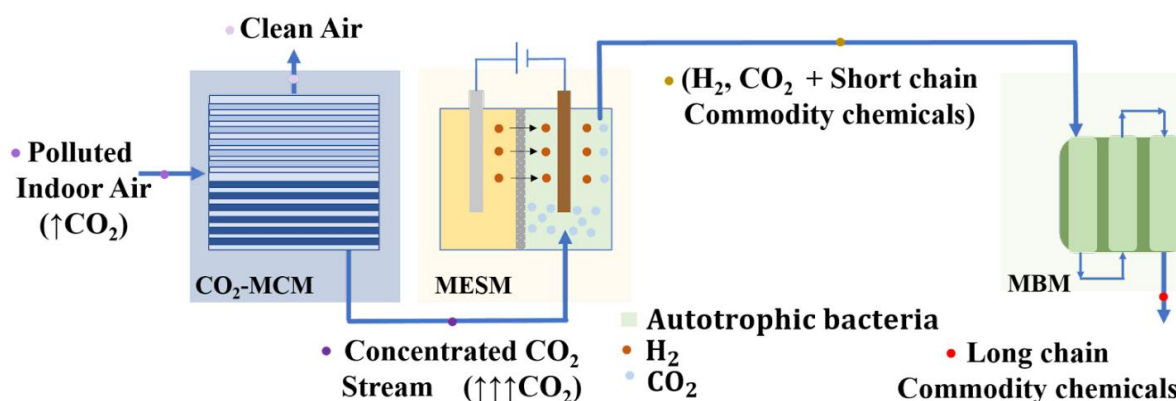


Figure 2. Diagram of the three serial modules of the MICRO-BIO process: CO₂-MCM (CO₂ adsorption-desorption), MESM (bioelectrosynthesis) and MBM (CO₂ mass transfer).

The overall goal for this process is to obtain a product stream at the end of the process mainly composed of biomethane (~90-100% CH₄). Before coupling the three modules, it is important to study and optimize the performance of each individual module. For this reason, secondary objectives are established for each individual module.

The goal of the first module (CO₂-MCM) is to adsorb and desorb CO₂ from indoor air and supply a concentrated CO₂ stream sufficient for the optimal performance of the second module (MESM).

The goal of the second module (MESM) is to perform a 50 to 70% conversion of CO₂ to CH₄. To achieve that, two hydraulically independent compartments of cells are built up, one of these being the anode and the other acting as a cathode. In the anode chamber, water hydrolysis

occurs due to the application of an electric potential (eq. 3). From this reaction, protons and electrons are obtained, being those last ones transferred to the cathode via electrodes. In the cathode, H₂ gas is bioelectrochemically produced (eq. 4) reducing the protons transferred from the anode and using the electrons received from the anode generated during the water hydrolysis (eq. 3). In the same cathode compartment, the H₂ gas bioelectrochemically produced plus the concentrated CO₂ stream from the CO₂-MCM, enable the CO₂ conversion to CH₄ thanks to the cathode's methanotrophic bacteria (eq. 5). The obtained stream is composed of CH₄, unreacted CO₂ and H₂, and is sent to the third module (MBM).



The goal of the last module (MBM) is to concentrate the CH₄ proceeding from the MESM. To achieve that, a capillary microbioreactor with the same methanotrophic bacteria and a huge contact area is set up, causing a yield increase of the CH₄ conversion reaction, resulting in a highly concentrated bio CH₄ outlet.

Is important to highlight that this final degree project is focused on the study of the optimization of the bioelectrochemical transformation into CH₄ using CO₂, specifically on the study of the operating conditions of the BES cell (MESM). Although the goal of the project is to use indoor CO₂, at the moment of presenting this work, the coupling of the first module (CO₂ capture) and the second module (CO₂ conversion) was still not performed, although it is planned to occur in the following weeks after the presentation of this work.

2. OBJECTIVES

This final degree project has three main research objectives (RO):

RO1: To identify the optimal operating parameters of a bioelectrochemical cell for CH₄ production to optimize the performance of the system in terms of methane productivity and methane purity of the cell effluent. This information can be later applied in a future prototype for the MICRO-BIO process. Several experiments are established changing the hydraulic residence time and empty bed residence time.

RO2: To evaluate the optimal CO₂ needs during CH₄ production in the bioelectrochemical cell and evaluate the capability to couple the CO₂-MCM. Information obtained from this second goal is key to design and define the operating conditions of the CO₂-MCM.

RO3: To verify the repeatability of the process through the assessment of the productivity and

selectivity of CH₄ produced by the BES.

Nevertheless, the global project goal of the MICRO-BIO process is to adsorb CO₂ and drive it to different phases to convert it to added-value by-products. The project is also focused on the trial of manufacturing a prototype containing all the needed phases to adsorb CO₂ and obtain bio CH₄.

3. MATERIALS AND METHODS

3.1. Analytical methods

The analysis methods used were those of an analytical chemistry laboratory. The voltage across the resistance was collected by a NEV 3.2 potentiostat (Nanoelectra, Madrid, Spain). Optical density was measured with a Cary Compact UV-Vis spectrophotometer (Agilent Technologies, California, USA) and the pH was measured with a pH110 pH-meter (VWR International, Pennsylvania, USA). The volatile fatty acids (VFAs) were tested in a 7890A GC with FID detection (Agilent Technologies, California, USA). Total inorganic and organic carbon were measured with a TOC-VCSH with combustion catalytic oxidation at 680°C and NDIR (Shimadzu Corporation, Kyoto, Japan). The gas sample was analysed in a 490 Micro GC (Agilent Technologies, California, USA).

3.2. Experimental procedure

The MES reactor in continuous mode was operated for 118 days and three times per week liquid and gaseous sampling was done. Medium is periodically added to both the catholyte and anolyte to equalize the extracted volume, and if the pH of the cathode was below 6, 3 mL of 5 M sodium hydroxide was added manually to ensure that enough CO₂ was dissolved in the liquid. Liquid samples that could not be analysed on the same day of extraction were stored in a refrigerator and analysed within less than a week. The gaseous samples were stored at room temperature. In the following sections details of experimental procedure for the sampling and analysis for liquid and gaseous samples are provided.

3.2.1. Liquid samples preparation and analysis

To obtain the liquid samples, 10 mL of anolyte and catholyte were extracted. Sample collected was used for monitoring the following variables: optical density (OD), pH, and VFAs, for both anode and cathode compartment. In addition, for the sample obtained from the cathode, the

total inorganic carbon (TIC) and total organic carbon (TOC) was also performed. The following protocols were implemented. Except of the OD analysis, all samples were filtered using a 0.22 μm filter.

OD analysis: cathode and anode sample preparation for OD reading using a spectrophotometer.

1. Blank the spectrophotometer, placing 4 mL of milli Q water in a spectrophotometer cuvette.
2. Sample preparation by placing 4 mL into a spectrophotometer cuvette.
3. Depending on the OD, extra dilution was applied accordingly. First samples were tested directly into the spectrophotometer without any dilution, if the readings were below 0.6, then no extra dilution was performed. If extra dilution was needed, a 50:50 dilution (2 mL milli Q water and 2 mL sample) was applied. Spectrophotometer cuvette was covered with parafilm paper to mix properly.
4. Once the OD measurement was finished, the cathodic and anodic solution was discarded.

pH measurement: using the portable pH-meter, read the pH value for the cathodic and anodic solution.

1. Start up the portable pH-meter, pressing the ON button.
2. Clean the tip of the pH-meter before reading the first sample, use distilled water of milli Q water for cleaning the tip.
3. Read the pH of the cathode/anode solution. Press the button M (Measure) within the portable pH-meter.
4. Between readings, always clean the tip of the pH-meter before reading the next sample, use distilled water of milli Q water for cleaning the tip.
5. Once finished, clean again the tip of the pH-meter, use distilled water of milli Q water for cleaning the tip.

VFA sample preparation: cathode and anode sample preparation for GC analysis.

1. Prepare two glass vials of 2 mL for GC for VFA analysis for the cathode and anode sample.
2. Add 1 mL of milli Q water using a micropipette.
3. Add 100 μL of a 1 M solution of phosphoric acid (H_3PO_4) using a micropipette.
4. Add 85 μL of a 1 M solution of crotonic acid ($\text{C}_4\text{H}_6\text{O}_2$) using a micropipette.
5. Add 0.5 mL of the corresponding sample already filtered.
6. Close the vial and mix to homogenize the content of the sample.
7. Store in the fridge before GC analysis.

TIC and TOC sample preparation: cathode samples preparation for TIC and TOC analysis.

1. Sample dilution was defined according to the TIC/TOC analysis system. In our case, a dilution of 1:10 and 1:100 was selected.
2. Filtered sample was diluted using milli Q water according to the corresponding dilution.
3. Samples were stored in the fridge before TIC/TOC analysis.

3.2.2. Gas samples preparation and analysis

Gaseous samples were analysed for gas composition (CO₂, CH₄, N₂, O₂, and H₂). Gas samples were collected using either 1, 3 or 5 L Tedlar bags connected directly to the BES setup. Gas bag volumes were selected as needed depending on the duration and conditions of the experiments.

3.3. Calculations

3.3.1. Operational variables

The current and the cell power in the cell were collected every 30 seconds by a potentiostat. The cell voltage was calculated according to Watt's law (eq. 6), and once the cell voltage was known, the anode voltage was obtained from eq. 7 and eq. 8, current demand using eq. 9, and power consumption using eq. 10.

$$\Delta V_{cell} = \frac{P}{I} \quad (\text{eq. 6})$$

$$\Delta V_{cell} = V_{cathode} - V_{anode} \quad (\text{eq. 7})$$

$$V_{anode} = V_{cathode} - \Delta V_{cell} \quad (\text{eq. 8})$$

$$J = \frac{I}{A_{cathode}} \quad (\text{eq. 9})$$

$$W = I \cdot \Delta V_{cell} \cdot t \quad (\text{eq. 10})$$

Where: ΔV_{cell} is the electric potential difference (V); $V_{cathode}$ and V_{anode} are cathode and anode voltages respectively (V); I is the circuit current (A); P is the power of the cell (W); J is current demand (A/m²); $A_{cathode}$ is the projected cathode electrode surface area (4.3·19 = 81.7 cm²); t is the time (h); and W is the power consumption of the cell (kW·h).

Several experiments were established to identify the optimal operating parameters of the bioelectrochemical cell for CH₄ production, changing the hydraulic residence time (HRT) and

the empty bed residence time (EBRT). HRT is a term used in engineering to quantify the average time a liquid with a specific flowrate (Q_L) spends in a vessel of a certain volume (V). It is defined as the ratio between the volume and output flowrate of the system and is used to optimize reactors and to predict eutrophication levels of water bodies. The HRT (d) can be derived under the following assumptions: uniform velocity profile and perfect mixed system resulting in eq. 11, where V is the volume of the system (L) and Q_L is the liquid flow rate (L/d) (Bernardo & Bleninger, 2013).

$$\text{HRT} = \frac{V}{Q_L} \quad (\text{eq. 11})$$

EBRT is a term used in engineering to describe the average time a gas with a gas flowrate (Q_G) spends in a reactor bed (V). EBRT is equal to the volume of the empty bed of the reactor divided by the gas flow rate (Fundneider et al., 2021). For practical reasons, in eq. 12 we assumed that V is the volume of the system (L) and Q_V is the gas flow rate (L/d).

$$\text{EBRT} = \frac{V}{Q_V} \quad (\text{eq. 12})$$

The gas sample was taken from the reactors every 48h and analysed in a Micro GC. The CH_4 productivity (γ_{CH_4} ; L $\text{CH}_4/\text{m}^2 \cdot \text{d}$), CO_2 utilization efficiency (η_{CO_2} ; %), and coulombic efficiency (η_{CE} ; %) were calculated using the following equations:

$$\gamma_{\text{CH}_4} = \frac{Q_{\text{CH}_4}}{A_{\text{cathode}}} \quad (\text{eq. 13})$$

$$\eta_{\text{CO}_2} = \frac{\text{CO}_2 \text{ in} - \text{CO}_2 \text{ out}}{\text{CO}_2 \text{ in}} \cdot 100 \quad (\text{eq. 14})$$

$$\eta_{\text{CE}} = \frac{m \cdot n \cdot F}{\int_0^t I dt} \cdot 100 \quad (\text{eq. 15})$$

Where in equations 13, 14 and 15: Q_{CH_4} is the flow rate of CH_4 production in continuous mode (L/d); A_{cathode} is the projected cathode electrode surface area ($4.3 \cdot 19 = 81.7 \text{ cm}^2$); $\text{CO}_2 \text{ in}$ and $\text{CO}_2 \text{ out}$ are the CO_2 inlet and outlet respectively (mol); m is the total mole of CH_4 produced (mol); n is the number of electrons required for CH_4 formation (8 mol e^- for 1 mol CH_4); F is the Faraday constant (96,485 C/mol e^-); I is the circuit current (A); and t is the time (s) (Liu et al., 2017; Yang et al., 2018).

The biomass concentration was determined by measuring the OD of the catholyte in a 600 nm wavelength using the Lambert-Beer law (eq. 16) and a linear correlation between OD and total suspended solids (TSS in mg/L) (eq. 17).

$$A = \varepsilon \cdot b \cdot c \quad (\text{eq. 16})$$

$$OD = 2.1514 \cdot TSS - 0.1496 \quad (\text{eq. 17})$$

Where in equation 16: ϵ is the molar absorptivity of the absorbing species, b is the path length, and c is the concentration of the absorbing species.

3.3.2. Carbon mass balance

To evaluate the carbon distribution in the different species, a simplified carbon mass balance was studied. Eq. 18 describes the carbon mass balance within the cell:

$$E_C + G_C = S_C + A_C \quad (\text{eq. 18})$$

Where E_C accounts for all carbon species that enters the system and is described by eq. 19 and eq. 20:

$$E_C = \sum C_{in} \quad (\text{eq. 19})$$

$$\sum C_{in} = C_{in,G,CO_2} + C_{in,L,CO_2} + C_{in,L,HCO_3^-} \quad (\text{eq. 20})$$

Where C_{in,G,CO_2} , C_{in,L,CO_2} and C_{in,L,HCO_3^-} stands for the carbon that enters the system as CO_2 in the gas phase (100 % v/v), CO_2 dissolved within the liquid phase present in the mineral media and the HCO_3^- in equilibrium with the CO_2 in the mineral media according to Henry's law.

On the other hand, G_C stands for the generation term, which includes the carbon mass that appears or disappears within the system. Since we are performing an elemental mass balance, and there is no carbon generated (only transformed from carbon specie to another) within the system, then this term is zero (eq. 21).

$$G_C = 0 \quad (\text{eq. 21})$$

With regards of S_C , this term of the carbon mass balance is described by eq. 22 and eq. 23:

$$S_C = \sum C_{out} \quad (\text{eq. 22})$$

$$\sum C_{out} = C_{out,G,CO_2} + C_{out,G,CH_4} + C_{out,L,CO_2} + C_{out,L,HCO_3^-} + C_{out,L,VFA} + C_{out,L,X} \quad (\text{eq. 23})$$

Where C_{out,G,CO_2} , C_{out,G,CH_4} , C_{out,L,CO_2} and $C_{out,L,X}$ stands for the carbon that leaves the system as CO_2 in the gas phase, CH_4 (the main product of the reaction), CO_2 dissolved within the liquid phase present in the mineral media, HCO_3^- in equilibrium with the CO_2 in the mineral media according to Henry's law, and the organic carbon as volatile fatty acids (VFAs) and the biomass present in the liquid phase. The dimensionless Henry solubility (H_s^{cc}) can be expressed as the ratio between the aqueous-phase concentration (C_a) and its gas-phase

concentration (C_g) as shown in eq. 24:

$$H_s^{cc} = \frac{c_a}{c_g} \quad (\text{eq. 24})$$

The term A_C refers to the mass of carbon accumulated within the system along time. This term can be approximated to zero when the system is operated under steady state conditions, which means all the carbon entering the system leaves the system and there is no mass accumulation (eq. 25).

$$A_C = 0 \quad (\text{eq. 25})$$

To evaluate the mass balance closure (MBC) in the system eq. 26 is used.

$$MBC = \frac{\sum c_{in} - \sum c_{out}}{\sum c_{in}} \quad (\text{eq. 26})$$

3.4. Reactor set-up

Two-chamber electromethanogenesis reactor made of methacrylate with granular graphite and carbon felt was constructed (Figure 3).

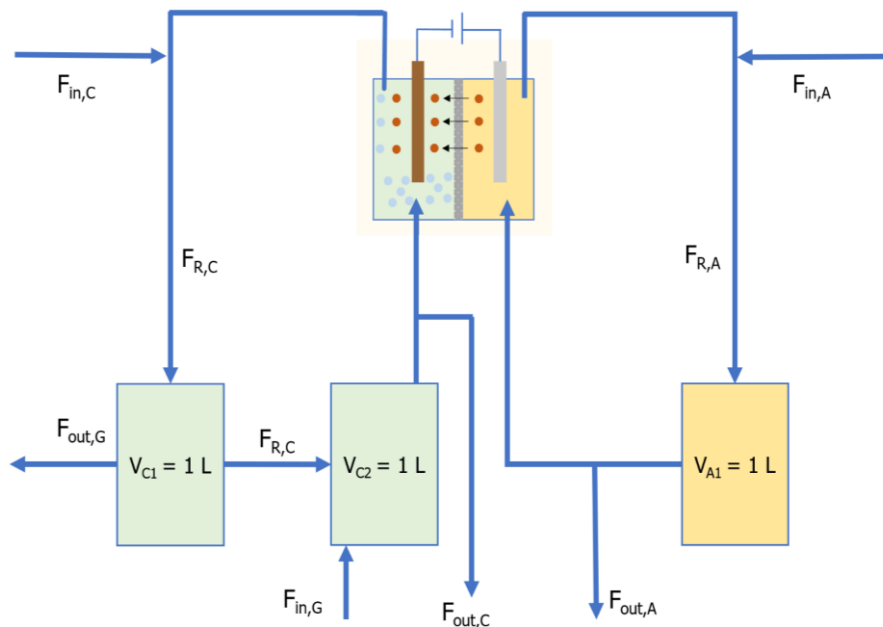


Figure 3. Schematic diagram of the MES. Anode chamber (yellow), cathode chamber (green). Inlet flow (F_{in}), outlet flow (F_{out}), recirculation flow (F_R), chamber volume (V), anolyte (A), catholyte (C), gas phase (G).

A cationic exchange membrane separated the anode and cathode chambers, and the volume of each chamber was 163.4 mL ($19 \cdot 4.3 \cdot 2 \text{ cm}^3$). The anode and cathode electrodes were

graphite rod electrodes. The anode and cathode electrodes were wired to the external circuit and operated under 997 mV of applied cathodic potential. Gas and liquid phases operate in continuous mode. The catholyte (total volume of 2 L) was recirculated via two 1 L glassware lab bottles (w/Blue GL45) by a pump with a 350 mL/min flow rate. The anolyte (total volume of 1 L) was recirculated by a pump with a 315 mL/min flow rate. The anode and cathode media were periodically renewed by a pump with different flow rates (0.06, 0.12, and 0.53 mL/min). Also, to operate in continuous mode, two gas bags were connected to decouple the inlet and outlet gasses, both by a pump with different flow rates (0.49 and 1.11 mL/min).

3.5. Reactor operation

Experimentation started transitioning the MES reactor from fed-batch mode into continuous mode. The BES reactor was operated for 241 days in fed-batch mode before the transition to continuous operation. The MES reactor transitioned to a continuous mode by adding inlet and outlet gas and liquid pumps in order to renew the liquid media continuously and to provide a continuous source of carbon (CO₂). The gas inlet pump provide a 100% CO₂ stream, and the liquid inlet pumps provided fresh anolyte and catholyte media. The MES reactor in continuous mode was operated at 30 ± 1°C in a water bath. To establish the optimal operation conditions, several experiments were established changing first the hydraulic residence time (HRT), and when selected the optimal one, changing the empty bed residence time (EBRT). The experimentation lasted 118 consecutive days, and 5 conditions were programmed (Table 1).

Table 1. Hydraulic residence time (HRT) and empty bed residence time (EBRT) for each condition and duration period, in continuous mode with an applied voltage of 997 mV.

Condition	Duration (d)	HRT (d)	EBRT (d)
1	24	11.94	2.81
2	24	2.63	2.81
3	23	23.15	2.81
4	23	11.94	1.25
5	24	11.94	2.81

As shown in Table 1, each experiment lasted between 23 and 24 days, and CH₄ conversion (%) and productivity rate (L CH₄/m²·d) were calculated for each condition.

3.6. Inoculum and mineral media

At the beginning of the fed-batch operation, the cathode chamber was inoculated with anaerobic sludge collected from an anaerobic digester of a wastewater treatment plant (Girona, Spain). The reactor was operated in fed-batch mode for 241 days and when transitioned to continuous mode, the biomass had already been selected/specialized for CH₄ production.

The anolyte was composed of mineral media solution (0.1 g/L of KH₂PO₄, 0.8 g/L of NaCl, 1 g/L of NH₄Cl, 0.2 g/L of MgCl₂·6H₂O, 0.1 g/L of KCl, 0.02 g/L of CaCl₂·2H₂O, 1.95 g/L of MES, and 0.4 g/L of cysteine HCl), adjusted at pH of 2.5 with HCl solution.

The catholyte was composed of the same mineral media, 1 mL/L of vitamin solution (20 µg/L of biotin, 20 µg/L of folic acid, 100 µg/L of pyridoxine hydrochloride, 50 µg/L of thiamine hydrochloride, 50 µg/L of riboflavin, 50 µg/L of nicotinic acid, 50 µg/L of DL-calcium pantothenate, and 1 µg/L of B12 vitamine), and 1 mL/L of trace metal solution (20 mg/L of nitrilotriacetic acid, 10 mg/L of MnSO₄·H₂O, 8 mg/L of Fe(SO₄)₂(NH₄)₂·6H₂O, 2 mg/L of CaCl₂·2H₂O, 0.002 mg/L of ZnSO₄·7H₂O, 0.2 mg/L of CuCl₂·2H₂O, 0.2 mg/L of NiCl₂·2H₂O, and 0.2 mg/L of Na₂MoO₄·2H₂O), adjusted at pH of 5.5 with NaOH solution.

4. RESULTS AND DISCUSSION

To develop a continuous electromethanogenic biocathode, the reactor was operated at a constant applied voltage of 997 mV in fed-batch mode for 241 days by a different lab member, and during the last 100 days with my support while performing an academic internship. That expertise allowed me to understand properly the operation of the cell and lead the transition to continuous mode. Images of the cell in batch mode and continuous mode are presented in Figures 4 and 5.

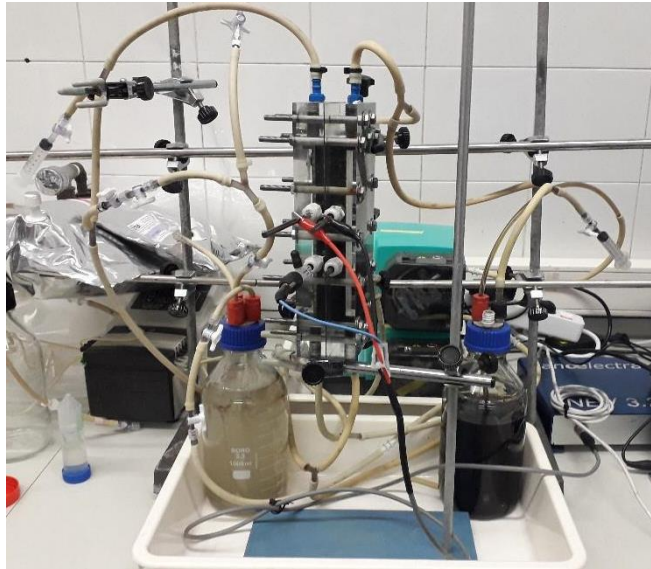


Figure 4. Electromethanogenic biocathode operating in fed-batch mode for 241 days. Catholyte (left bottle), cationic exchange membrane separating the electrodes (center of the image), and anolyte (right bottle).

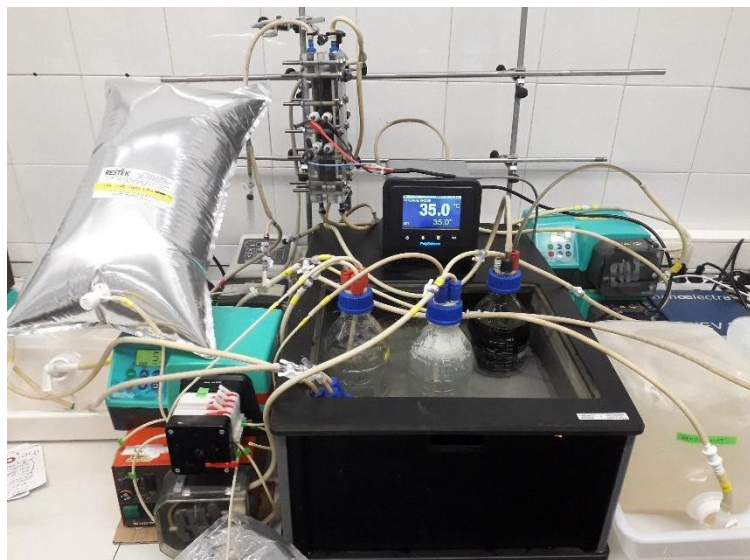


Figure 5. Electromethanogenic biocathode operating in continuous mode for 118 days. Catholyte (left and center bottle), cationic exchange membrane separating the electrodes (center of the image), anolyte (right bottle), inlet media (bottom right), outlet media (center left), inlet gas (top left), and outlet gas (bottom left). Initially, the temperature of the bath was 35°C, but eventually, it was changed to 30°C.

The experimentation in continuous mode lasted 118 consecutive days, and 5 conditions were programmed as described in Table 1. Figure 6 shows the evolution of gas composition (% CH₄, H₂, and CO₂) for each condition established. In the first 6 days of operation, the system CH₄ conversion was 18.78% since the system was still adapting to the new operating conditions. This can be observed by the remaining percentage of H₂ and CO₂, 49.67% and

31.54% that was not converted into CH₄. After that, almost all H₂ was consumed by the methanotrophic bacteria achieving a CH₄ conversion of $42.10 \pm 3.29\%$ and a productivity rate of $41.56 \pm 3.25 \text{ L CH}_4/\text{m}^2\cdot\text{d}$ in condition 1.

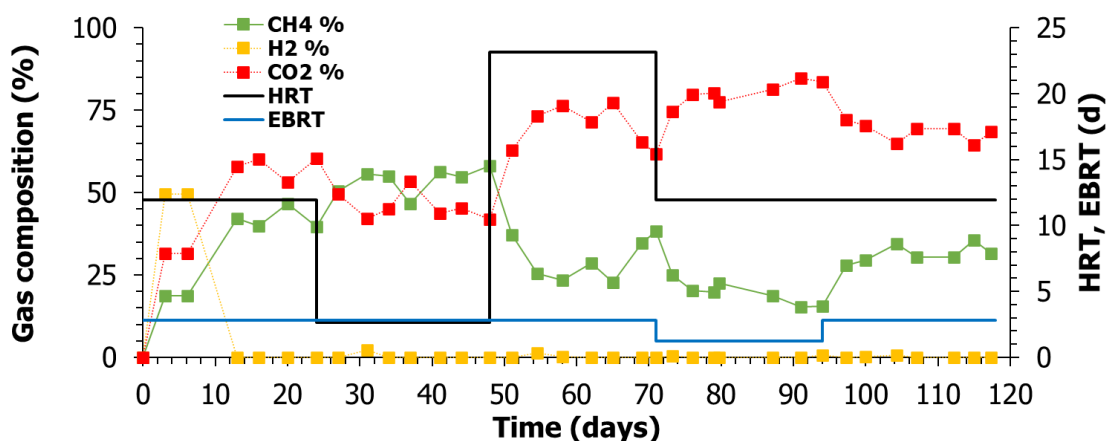


Figure 6. Gas composition (%) for the five different hydraulic residence time (HRT) and empty bed residence time (EBRT) conditions in continuous mode with an applied voltage of 997 mV.

To observe the effects of HRT on reactor performance, in condition two HRT was reduced one-fifth from condition one, by supplying more media to the bacteria, which increased both CH₄ conversion of $53.86 \pm 3.93\%$ and productivity rate of $53.17 \pm 3.88 \text{ L CH}_4/\text{m}^2\cdot\text{d}$. Although having an increase in productivity rate, the CH₄ conversion increase was not as high as expected since it only increased by 28% in CH₄ conversion. In practice, increasing media consumption by 500% is not feasible. For this reason, condition one HRT was selected for subsequent experiments of EBRT evaluation. To demonstrate that the increase in productivity was a result of the changes in HRT and not due to the increase of biomass in the system, which is expected to happen in biological systems, the HRT in condition three was doubled compared to condition one. The hypothesis was that an HRT increase (lower media renovation) would decrease both productivity and conversion rate, due to the decrease of media renovation. A decrease in media renovation would result in longer presence of the same media within the system, decreasing the supply of fresh nutrients to the microorganisms and thus affecting their metabolism and consequently the reactor performance (Cario et al., 2019). As observed in Figure 6, as predicted, both CH₄ conversion of $30.02 \pm 6.57\%$ and productivity rate of $29.64 \pm 6.49 \text{ L CH}_4/\text{m}^2\cdot\text{d}$ decreased, which represents a 44% drop in CH₄ conversion compared to the previous condition.

To observe the effects of EBRT on reactor performance, in condition four, HRT was set in the reference case (condition one), and EBRT was reduced one-half compared to condition one, thus reducing the time the gas was spending inside the cell. The hypothesis was that an EBRT

decrease on one side would increase the supply of CO₂ due to the increase of gas flow rate to the system, but at the same time would decrease the gas contact time and thus affect the mass transfer efficiency of the system. Thus, the expected result is less predictable than in the previous cases. As observed in Figure 6, CH₄ conversion decreased to 19.58 ± 3.51%, which represents a 53% drop in CH₄ conversion compared to condition one, but the productivity rate increased to 43.29 ± 7.75 L CH₄/m²·d, which represents a 5% increase from condition one, and a 46% increase from the previous condition (three).

To assess the cause of the increase in productivity but the decrease in the conversion rate, Figure 7 is presented. The decrease in CH₄ conversion can be explained by an increase in CO₂ content in the outlet gas composition, diluting the CH₄ produced under these conditions, while the increase in productivity can be explained by an increase in the liters of gas produced because of the increase in inlet gas flow rate.

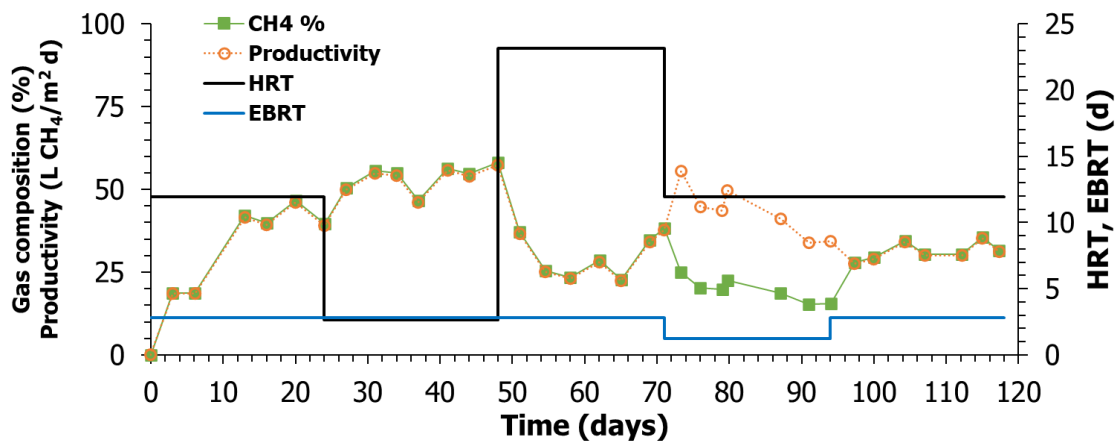


Figure 7. Catholyte productivity rate (L CH₄/m²·d) versus methane production (%) in five different hydraulic residence time (HRT) and empty bed residence time (EBRT) conditions in continuous mode with an applied voltage of 997 mV.

As shown in Figure 7, from day 72 to day 96, assessing the biological effects, in condition four the CH₄ productivity rate increased due to an increase of substrate (CO₂) for the methanotrophic bacteria, which allows more transformation of CO₂ into CH₄, but as mentioned before, the CH₄ conversion decreases through a dilution effect because too much CO₂ is provided and was leaving the system unreacted. These results demonstrate that in this case, the limiting reagent for CO₂ reduction is H₂.

Once experimentation of HRT and EBRT effects on gas conversion and productivity rate were evaluated, in condition five the system was set up in the reference condition (condition one) for both HRT and EBRT. This was done to see if, after stressing the system during condition two to four, the system could achieve again the same conversion and productivity rates from

condition one (reference conditions). Figure 7 shows that during condition five both CH_4 conversion of $31.43 \pm 2.74\%$ and productivity rate of $31.02 \pm 2.71 \text{ L CH}_4/\text{m}^2\cdot\text{d}$ were slightly lower compared to condition one. The production rates in MES reactors are widely variable and dependent on several factors, including the reactor materials, inoculum sources, and operation parameters (Lovley, 2011; Dessì et al., 2021). It must be taken into account that in condition five the reactor has been operating in continuous mode for 118 days, plus 241 days in fed-batch mode. As operation parameters were practically the same, the hypothesis presented is that after these 359 days, the performance of the cell could be affected by the deterioration of the cell materials, e.g., visual deterioration of granular graphite was observed, and for different biomass availability. To assess the effects of biomass availability, biomass concentration was registered and is presented in Figure 8.

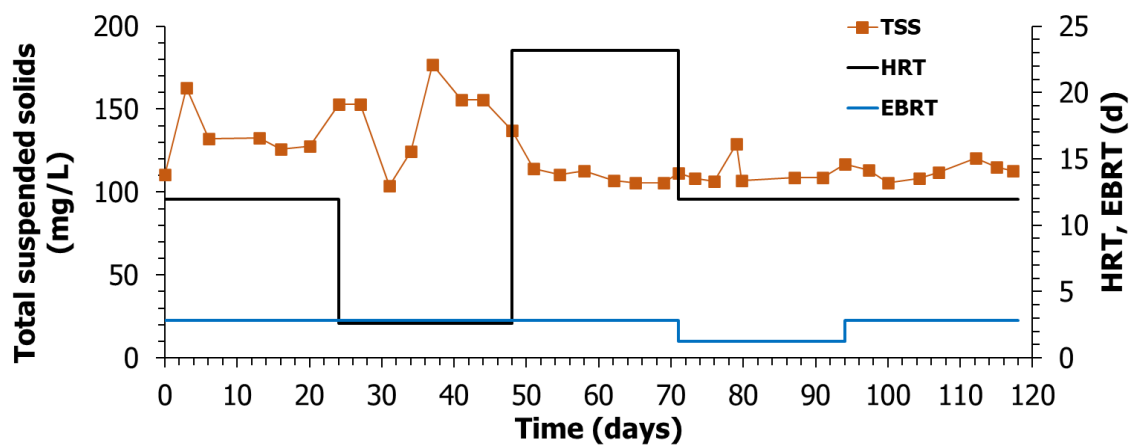


Figure 8. Total suspended solids (mg/L) in the catholyte for each condition in continuous mode with an applied voltage of 997 mV.

Figure 8 presents measurements of total suspended solids (TSS), which can be interpreted as predominantly consisting of biomass. Therefore, TSS can be considered an estimation of biomass concentration. This figure shows that the TSS of conditions one and two were the highest ones with high deviation, 130 ± 20 and $150 \pm 25 \text{ mg/L}$ respectively, and after that, in the following conditions, TSS became more stable (110 ± 10 , 110 ± 10 , and $115 \pm 5 \text{ mg/L}$ respectively). Figure 8 results suggest that the lower productivity observed in condition five, despite having the same operating parameters as condition one, could be explained by different biomass availability. Initially, we assumed that the system was adapted to the new operating conditions within the first 6 days of operation. However, the TSS results indicate that the system required an adaptation period of approximately 50 days for continuous operation. Also, optical density only could quantify the solubilized biomass and not the flocculated one or attached into the materials within the cell. In the cathode, during fed-batch operation, a lot

of biofilm was produced and not quantified, and during continuous operation this biofilm was washed out due to high shear stress conditions by having a liquid outlet flowrate. Hence, we can assume that CH₄ productivity rate of the biofilm was playing an important role that was changed when transitioning from fed-batch to continuous mode.

The presented hypothesis is that not quantified biofilm produced CH₄ and, conditions one and five differ on biomass availability. As mentioned before, operation parameters such as pH and media purity were also evaluated. In Figure 9, pH measurements for both anolyte and catholyte are represented, and Figure 10, present the VFAs produced in the cell. In Figure 9, pH remained mainly constant, 6.26 ± 0.22 and 2.13 ± 0.10 , for both cathode and anode media respectively. As observed in Figure 10, the only VFA produced was acetic acid, which achieved a maximum concentration of 79.15 mg/L, considered traces, and didn't affect the MES performance.

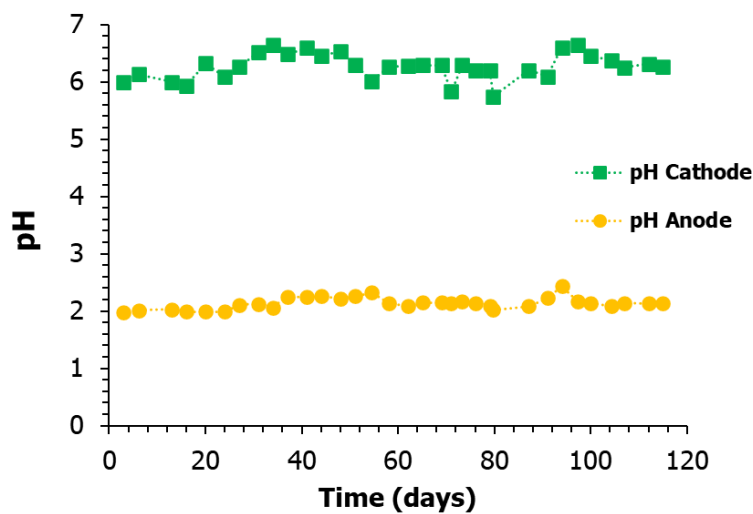


Figure 9. Anolyte and catholyte pH measurement in continuous mode with an applied voltage of 997 mV.

Figure 10 results on VFAs production support the hypothesis of a 50-day adaptation period for the system to operate in continuous mode, because after 50 days no more VFAs were produced, indicating that the microorganisms adapted their metabolism to the continuous conditions for methanogenic performance instead of fermentation metabolism.

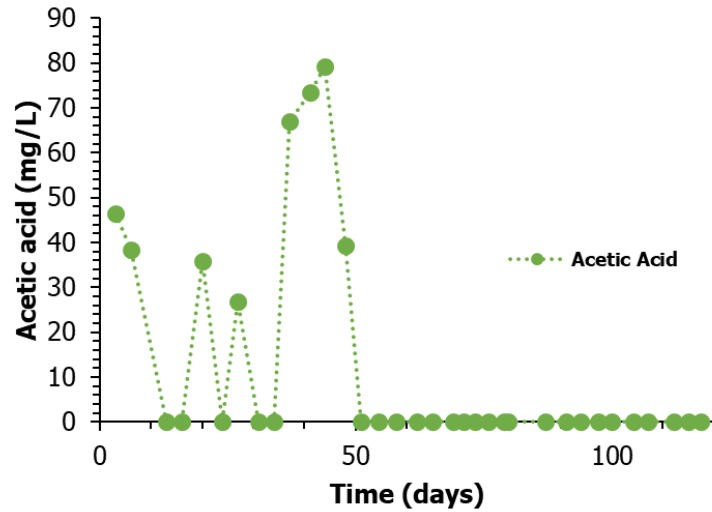


Figure 10. Cathodic acetic acid production (mg/L) in continuous mode with an applied voltage of 997 mV.

To evaluate the 50-day hypothesis, Figure 11 displays a linear regression between current demand and CH₄ productivity rate. Current demand is an important factor that affects the performance of MES systems, in which high current demand can lead to improved productivity.

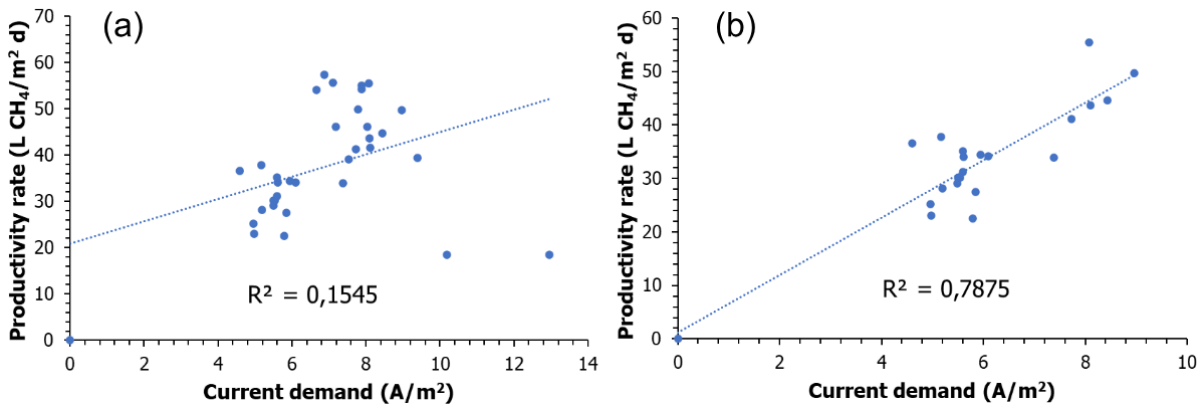


Figure 11. Linear regression between current demand (A/m²) and productivity rate (L CH₄/m²·d) in continuous mode with an applied voltage of 997 mV. Data obtained during all 118 experimentation days in continuous mode (a), data obtained from day 51 to 118 (b).

In Figure 11a no clear correlation is observed between current demand and productivity rate, but in Figure 11b, excluding the 50 days adaptation period, a clearer correlation is observed with a coefficient of determination of 0,7875. The findings from these experiments provide strong evidence in favour of the hypothesis that there is a 50-day adaptation period. During the initial 50 days following the switch from fed-batch to continuous mode, the microorganisms were still adjusting to the new operational conditions. The electrical current supplied to the system was being allocated to various metabolic pathways, such as growth or the production

of VFAs, rather than primarily focusing on CH₄ production. However, after the 50-day mark, the microorganisms successfully adapted their metabolism to the continuous conditions necessary for efficient methanogenic performance. Subsequently, a better correlation is observed between the supplied energy in the system and CH₄ production, indicating improved energy consumption efficiency.

As a summary, Figure 12 displays the mean productivity rate for each condition. In all conditions, there is a high deviation except for condition 5, which is more stable and seems to be the more adequate one for future experiments involving other operational parameters.

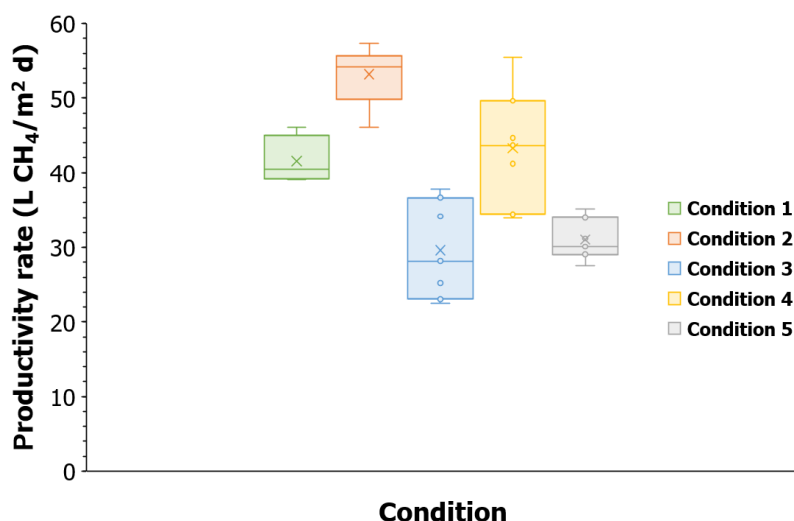


Figure 12. Mean productivity rate (L CH₄/m²·d) for each condition in continuous mode with an applied voltage of 997 mV.

With regards of the carbon mass balance, Figure 13 illustrates the evaluation of carbon distribution within the cell. The analysis reveals that 99.979% of the carbon source supplied to the cell comes from the fixed flow rate of 100% (v/v) CO₂ stream, as no bicarbonate was dosed in the liquid media. Adding bicarbonate is a common practice during the operation of bioelectrochemical systems with inorganic carbon demand, however in our case we avoided this practice to understand the effect of CO₂ supply from the gas phase, as this will be the operating conditions once the CO₂-microconcentrator module is coupled to the cell. Additionally, the liquid phase contributes 0.010% and 0.011% of dissolved CO₂ and HCO₃⁻, respectively, to the inlet carbon source.

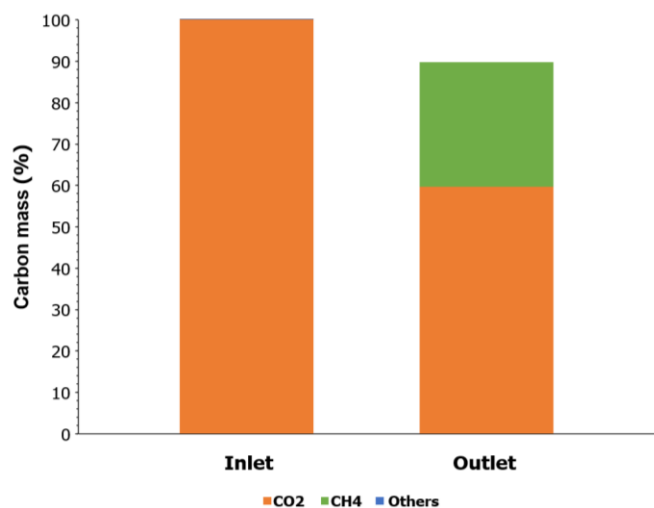


Figure 13. Mass balance for the inlet and outlet outputs during 118 days of continuous experimentation in the MES system with an applied voltage of 997 mV.

In terms of the outlet, the carbon source consists of 61.738% CO₂ stream, 28.934% CH₄ stream, and the sum of dissolved CO₂, HCO₃⁻, CO₃²⁻, total organic carbon, and biomass represented 0.002%. This accounts for a 90.674% closure in the carbon mass balance, indicating a significant level of accuracy. However, approximately 9.326% of the provided carbon mass remains unaccounted for, which can be accounted for mass accumulation inside the system. Furthermore, these findings clearly indicate a significant deficiency in the conversion of CO₂ to CH₄, suggesting that the optimization process is still far from being accomplished.

One of the research objectives of this work was to assess the carbon needs of the cell. Thus, the information obtained during the carbon mass balance, specifically the mass of CO₂ (in g of CO₂) that was supplied to the bioelectrochemical system, shown as “CH₄ cell” in Figure 14, was compared to the mass of CO₂ (in g) that was supplied by the CO₂-microconcentrator prototype. Results shown in Figure 14 indicate that the carbon supplied in the cell as CO₂ (solid green line) is comparable to the CO₂ that can be released from the CO₂-microconcentrator prototype under different release conditions (flow of nitrogen, release temperature, etc).

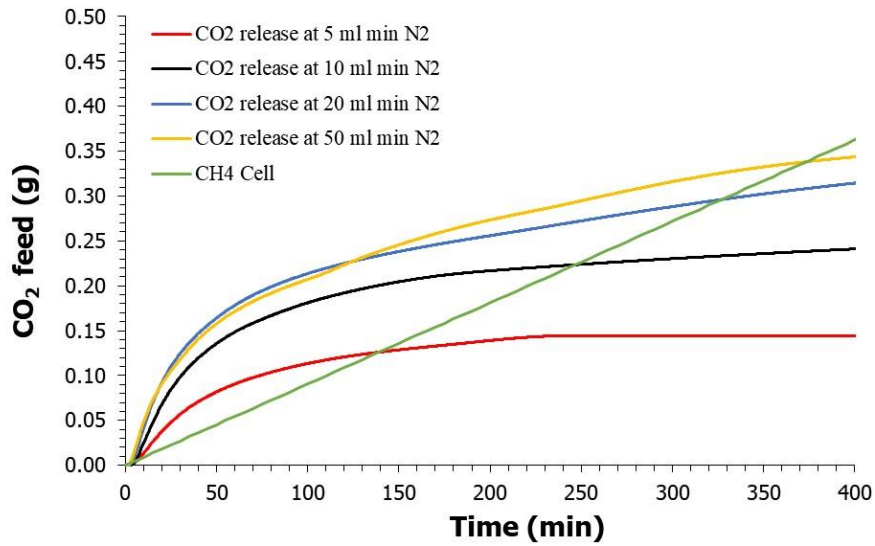


Figure 14. CO₂ desorption profiles of CO₂-MCM versus CO₂ stream demand for the MESM in continuous mode with an applied voltage of 997 mV.

These result supports the hypothesis that the carbon needs of the bioelectrochemical system can be supplied in the future by a CO₂-microconcentrator prototype that captures CO₂ from indoor air.

5. CONCLUSIONS

Once the results have been acquired, the next step is to analyse them and draw appropriate conclusions. This final degree project identified HRT and EBRT as the relevant operating parameters for electromethanogenesis optimization.

A decrease in HRT increases productivity and conversion rate, due to the increase of media renovation, but could lead to a washout of the biomass and excess of media needs which are unfeasible for a real application of this system.

During experimentation, the biofilm CH₄ productivity rate was underestimated, so to avoid washout, the use of pearls containing the biomass is proposed for future optimization.

EBRT effects are more complex since involves gas to liquid mass transfer and biological effects. As EBRT decreases, gas substrate availability increases, but in the MES system evaluated the limiting reagent for CO₂ reduction into CH₄ is H₂. So forth, when EBRT decreases leads to an increase in productivity, but the conversion rate decreases due to a dilution factor of the CH₄ produced from the not consumed CO₂.

All conditions were performed with an applied voltage of 997 mV. In this condition, H₂ was the limiting reagent, so increasing the applied voltage is proposed for future experimentation to increase water hydrolysis, providing more H₂ to the MES system, and subsequently an increase in conversion and productivity rate is expected.

Also, in the outlet mass carbon balance no dissolved CO₂ was observed, so CO₂ mass transfer of CO₂ from gas to the liquid phase is also limiting the CH₄ conversion. To improve the system and avoid mass transfer limitations, capillary module (third module of the MICRO-BIO process) assembly is required for future experimentation.

Furthermore, the findings from different analyses provide strong evidence in favour of the hypothesis that there is a 50-day adaptation period of the biomass to adjust their metabolism to the switch between fed-batch and continuous mode operation.

Finally, it was demonstrated that the CO₂ needs of the bioelectrochemical system can be supplied in the future by a CO₂-microconcentrator prototype that captures CO₂ from indoor environments.

6. BIBLIOGRAPHY

- Anderson, S., & Newell, R. (2004). Prospects for carbon capture and storage technologies. *Annu. Rev. Environ. Resour.*, 29, 109-142. <https://doi.org/10.1146/annurev.energy.29.082703.145619>
- Assimakopoulos, M. N., Barmmparesos, N., Pantazaras, A., Karlessi, T., & Lee, S. E. (2017). On the comparison of occupancy in relation to energy consumption and indoor environmental quality: a case study. *Energy Procedia*, 134, 875-884. <https://doi.org/10.1016/j.egypro.2017.09.548>
- Azuma, K., Kagi, N., Yanagi, U., & Osawa, H. (2018). Effects of low-level inhalation exposure to carbon dioxide in indoor environments: A short review on human health and psychomotor performance. *Environment international*, 121, 51-56.
- Bachmann, M., Kätelhön, A., Winter, B., Meys, R., Müller, L. J., & Bardow, A. (2021). Renewable carbon feedstock for polymers: environmental benefits from synergistic use of biomass and CO₂. *Faraday discussions*, 230, 227-246. <https://doi.org/10.1039/d0fd00134a>
- Baus, L., & Nehr, S. (2022). Potentials and limitations of direct air capturing in the built environment. *Building and Environment*, 208, 108629. <https://doi.org/10.1016/j.buildenv.2021.108629>
- Beaumont, M. L. (2022). Making Direct Air Capture Affordable; Technology, Market and Regulatory Approaches. *Frontiers in Climate*, 4, 127. <https://doi.org/10.3389/fclim.2022.756013>
- Becerra, J. A., Lizana, J., Gil, M., Barrios-Padura, A., Blondeau, P., & Chacartegui, R. (2020). Identification of potential indoor air pollutants in schools. *Journal of Cleaner Production*, 242, 118420. <https://doi.org/10.1016/j.jclepro.2019.118420>
- Bernardo, J. W. Y., & Bleninger, T. (2013). *Interpretation and applicability of residence times in reservoirs* (Doctoral dissertation, Dissertação de mestrado). Setor de Tecnologia, Universidade Federal do Paraná, Curitiba.
- Beuttler, C., Charles, L., & Wurzbacher, J. (2019). The role of direct air capture in mitigation of anthropogenic greenhouse gas emissions. *Frontiers in Climate*, 1, 10. <https://doi.org/10.3389/fclim.2019.00010>
- Bierwirth, P. N. (2018). Carbon dioxide toxicity and climate change: a major unapprehended risk for human health. *Web Published: ResearchGate*, 10.
- Blasco-Gómez, R., Batlle-Vilanova, P., Villano, M., Balaguer, M. D., Colprim, J., & Puig, S. (2017). On the edge of research and technological application: a critical review of

electromethanogenesis. *International journal of molecular sciences*, 18(4), 874. <https://doi.org/10.3390/ijms18040874>

Borrel, G., Adam, P. S., & Gribaldo, S. (2016). Methanogenesis and the Wood–Ljungdahl pathway: an ancient, versatile, and fragile association. *Genome biology and evolution*, 8(6), 1706-1711. <https://doi.org/10.1093/gbe/evw114>

Bruhn, T., Naims, H., & Olfe-Kräutlein, B. (2016). Separating the debate on CO₂ utilisation from carbon capture and storage. *Environmental Science & Policy*, 60, 38-43. <https://doi.org/10.1016/j.envsci.2016.03.001>

Bui, M., Adjiman, C. S., Bardow, A., Anthony, E. J., Boston, A., Brown, S., & Mac Dowell, N. (2018). Carbon capture and storage (CCS): the way forward. *Energy & Environmental Science*, 11(5), 1062-1176. <https://doi.org/10.1039/c7ee02342a>

Cai, W., Liu, W., Yang, C., Wang, L., Liang, B., Thangavel, S., & Wang, A. (2016). Biocathodic methanogenic community in an integrated anaerobic digestion and microbial electrolysis system for enhancement of methane production from waste sludge. *ACS Sustainable Chemistry & Engineering*, 4(9), 4913-4921. <https://doi.org/10.1021/acssuschemeng.6b01221>

Cario, B.P., Logan, B.E., Pugh, E.R., Regan, J.M., & Gorski, C.A. (2019). Evaluating the impact of substrate composition, reactor configuration, and operating conditions on the performance of bioelectrochemical systems treating fermentation effluents.

Carus, M., Dammer, L., Raschka, A., & Skoczinski, P. (2020). Renewable carbon: Key to a sustainable and future-oriented chemical and plastic industry: Definition, strategy, measures and potential. *Greenhouse Gases: Science and Technology*, 10(3), 488-505. <https://doi.org/10.1002/ghg.1992>

Carvalho, H. (2021). New WHO global air quality guidelines: more pressure on nations to reduce air pollution levels. *The Lancet Planetary Health*, 5(11), e760-e761. [https://doi.org/10.1016/S2542-5196\(21\)00287-4](https://doi.org/10.1016/S2542-5196(21)00287-4)

Cheng, S., Xing, D., Call, D. F., & Logan, B. E. (2009). Direct biological conversion of electrical current into methane by electromethanogenesis. *Environmental science & technology*, 43(10), 3953-3958. <https://doi.org/10.1021/es803531g>

Chen, C. Y., Chen, P. H., Chen, J. K., & Su, T. C. (2021). Recommendations for ventilation of indoor spaces to reduce COVID-19 transmission. *Journal of the Formosan Medical Association*, 120(12), 2055. <https://doi.org/10.1016/j.jfma.2021.08.007>

Dessi, P., Rovira-Alsina, L., Sánchez, C., Dinesh, G. K., Tong, W., Chatterjee, P., & Puig, S. (2021). Microbial electrosynthesis: Towards sustainable biorefineries for production of green

chemicals from CO₂ emissions. *Biotechnology Advances*, 46, 107675. <https://doi.org/10.1016/j.biotechadv.2020.107675>

European Commission. (2021). *Fit for 55 - The EU's plan for a green transition - Consilium*.

Filo, M., & Khammash, M. (2019). Optimal parameter tuning of feedback controllers with application to biomolecular antithetic integral control. In *2019 IEEE 58th Conference on Decision and Control (CDC)* (pp. 951-957). IEEE. <https://doi.org/10.1109/CDC40024.2019.9029430>

Fröhling, M., & Hiete, M. (2020). *Sustainability and life cycle assessment in industrial biotechnology: a review of current approaches and future needs* (pp. 143-203). Springer International Publishing. https://doi.org/10.1007/10_2020_122

Fu, Q., Kuramochi, Y., Fukushima, N., Maeda, H., Sato, K., & Kobayashi, H. (2015). Bioelectrochemical analyses of the development of a thermophilic biocathode catalyzing electromethanogenesis. *Environmental Science & Technology*, 49(2), 1225-1232. <https://doi.org/10.1021/es5052233>

Fundneider, T., Alonso, V. A., Abbt-Braun, G., Wick, A., Albrecht, D., & Lackner, S. (2021). Empty bed contact time: The key for micropollutant removal in activated carbon filters. *Water research*, 191, 116765. <https://doi.org/10.1016/j.watres.2020.116765>

Gabrielli, P., Gazzani, M., & Mazzotti, M. (2020). The role of carbon capture and utilization, carbon capture and storage, and biomass to enable a net-zero-CO₂ emissions chemical industry. *Industrial & Engineering Chemistry Research*, 59(15), 7033-7045. <https://doi.org/10.1021/acs.iecr.9b06579>

Gadikota, G., & Park, A. H. A. (2015). Accelerated carbonation of Ca-and Mg-bearing minerals and industrial wastes using CO₂. In *Carbon Dioxide Utilisation* (pp. 115-137). Elsevier. <https://doi.org/10.1016/B978-0-444-62746-9.00008-6>

Garland, R., Wernecke, B., Feig, G., & Langerman, K. (2021). The new WHO Global Air Quality Guidelines: What do they mean for South Africa?. *Clean Air Journal*, 31(2). <https://doi.org/10.17159/caj/2020/31/2.12915>

Ghiat, I., & Al-Ansari, T. (2021). A review of carbon capture and utilisation as a CO₂ abatement opportunity within the EWF nexus. *Journal of CO₂ Utilization*, 45, 101432. <https://doi.org/10.1016/j.jcou.2020.101432>

Goeppert, A., Czaun, M., Prakash, G. S., & Olah, G. A. (2012). Air as the renewable carbon source of the future: an overview of CO₂ capture from the atmosphere. *Energy & Environmental Science*, 5(7), 7833-7853. <https://doi.org/10.1039/c2ee21586a>

- Grim, R. G., Huang, Z., Guarnieri, M. T., Ferrell, J. R., Tao, L., & Schaidle, J. A. (2020). Transforming the carbon economy: challenges and opportunities in the convergence of low-cost electricity and reductive CO₂ utilization. *Energy & Environmental Science*, 13(2), 472-494. <https://doi.org/10.1039/C9EE02410G>
- Han, G. B., Jang, J. H., Lee, T. J., & Choi, C. (2016). Performance test of PSA-type O₂ separator for efficient O₂ supply to room ventilation system combined with CO₂ adsorption module. *Korean Journal of Chemical Engineering*, 33, 1311-1317. <https://doi.org/10.1007/s11814-015-0289-2>
- Hu, S. C., Shiue, A., Chang, S. M., Chang, Y. T., Tseng, C. H., Mao, C. C., & Chan, A. (2017). Removal of carbon dioxide in the indoor environment with sorption-type air filters. *International Journal of Low-Carbon Technologies*, 12(3), 330-334. <https://doi.org/10.1093/ijlct/ctw014>
- IEA (2020). Key World Energy Statistics 2020.
- IPCC (Intergovernmental Panel on Climate Change). (2014). Anthropogenic and Natural Radiative Forcing. In *Climate Change 2013 – The Physical Science Basis: Working Group I Contribution to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change* (pp. 659-740). Cambridge: Cambridge University Press. <https://doi.org/10.1017/CBO9781107415324.018>
- IPCC (Intergovernmental Panel on Climate Change). (2022). SYNTHESIS REPORT OF THE IPCC SIXTH ASSESSMENT REPORT (AR6) IPCC. Eur. Univ. Inst. 2–5.
- Izadi, P., Fontmorin, J. M., Lim, S. S., Head, I. M., & Eileen, H. Y. (2021). Enhanced bio-production from CO₂ by microbial electrosynthesis (MES) with continuous operational mode. *Faraday discussions*, 230, 344-359. <https://doi.org/10.1039/D0FD00132E>
- Jiang, Y., May, H. D., Lu, L., Liang, P., Huang, X., & Ren, Z. J. (2019). Carbon dioxide and organic waste valorization by microbial electrosynthesis and electro-fermentation. *Water research*, 149, 42-55. <https://doi.org/10.1016/j.watres.2018.10.092>
- Jones, C. W. (2011). CO₂ capture from dilute gases as a component of modern global carbon management. *Annual review of chemical and biomolecular engineering*, 2, 31-52.
- Jourdin, L., Freguia, S., Flexer, V., & Keller, J. (2016). Bringing high-rate, CO₂-based microbial electrosynthesis closer to practical implementation through improved electrode design and operating conditions. *Environmental science & technology*, 50(4), 1982-1989. <https://doi.org/10.1021/acs.est.5b04431>
- Kalt, G., Kaufmann, L., Kastner, T., & Krausmann, F. (2021). Tracing Austria's biomass consumption to source countries: A product-level comparison between bioenergy, food and

material. *Ecological Economics*, 188, 107129. <https://doi.org/10.1016/j.ecolecon.2021.107129>

Kätelhön, A., Meys, R., Deutz, S., Suh, S., & Bardow, A. (2019). Climate change mitigation potential of carbon capture and utilization in the chemical industry. *Proceedings of the National Academy of Sciences*, 116(23), 11187-11194. <https://doi.org/10.1073/pnas.1821029116>

Keith, D. W., Holmes, G., Angelo, D. S., & Heidel, K. (2018). A process for capturing CO₂ from the atmosphere. *Joule*, 2(8), 1573-1594. <https://doi.org/10.1016/j.joule.2018.05.006>

Kothari, D.C., Chaudhari, G.N., & Thorat, P.V. (2020). Direct Capture of CO₂ from Air & Scale-Up it for Sustainable Developments. *Journal of emerging technologies and innovative research*.

Kracke, F., Deutzmann, J. S., Jayathilake, B. S., Pang, S. H., Chandrasekaran, S., Baker, S. E., & Spormann, A. M. (2021). Efficient hydrogen delivery for microbial electrosynthesis via 3D-printed cathodes. *Frontiers in Microbiology*, 12, 696473. <https://doi.org/10.3389/fmicb.2021.696473>

Lackner, K. S. (2013). The thermodynamics of direct air capture of carbon dioxide. *Energy*, 50, 38-46. <https://doi.org/10.1016/j.energy.2012.09.012>

Leal-Arcas, R., Ulloa Martinez, L., Abang, V., Kapur, K., Greenwood, S., Chatzopoulos, K., & Schoettmer, L. (2023). Climate Neutrality and Sustainability in International Trade. *Forthcoming) University of Pennsylvania Journal of International Law*, 44(3). <http://dx.doi.org/10.2139/ssrn.4183881>

Leung, D. Y. (2015). Outdoor-indoor air pollution in urban environment: challenges and opportunity. *Frontiers in Environmental Science*, 2, 69. <https://doi.org/10.3389/fenvs.2014.00069>

Levi, P. G., & Cullen, J. M. (2018). Mapping global flows of chemicals: from fossil fuel feedstocks to chemical products. *Environmental science & technology*, 52(4), 1725-1734. <https://doi.org/10.1021/acs.est.7b04573>

Liu, D., Zheng, T., Buisman, C., & Ter Heijne, A. (2017). Heat-treated stainless steel felt as a new cathode material in a methane-producing bioelectrochemical system. *ACS sustainable chemistry & engineering*, 5(12), 11346-11353. <https://doi.org/10.1021/acssuschemeng.7b02367>

Logan, B. E., Rossi, R., Ragab, A. A., & Saikaly, P. E. (2019). Electroactive microorganisms in bioelectrochemical systems. *Nature Reviews Microbiology*, 17(5), 307-319. <https://doi.org/10.1038/s41579-019-0173-x>

López de León, L. R., Deaton, K. E., & Deshusses, M. A. (2018). Miniaturized biotrickling filters

and capillary microbioreactors for process intensification of VOC treatment with intended application to indoor air. *Environmental science & technology*, 53(3), 1518-1526. <https://doi.org/10.1021/acs.est.8b05209>

López de León, L. R., Dessì, P., Cabrera-Codony, A., Rocha-Melogno, L., Kraakman, B., Naddeo, V., & Puig, S. (2023). CO₂ in indoor environments: From environmental and health risk to potential renewable carbon source. *Science of The Total Environment*, 856, 159088. <https://doi.org/10.1016/j.scitotenv.2022.159088>

Lovley, D. R. (2011). Powering microbes with electricity: direct electron transfer from electrodes to microbes. *Environmental microbiology reports*, 3(1), 27-35. <https://doi.org/10.1111/j.1758-2229.2010.00211.x>

Lovley, D. R., & Nevin, K. P. (2013). Electrobiocommodities: powering microbial production of fuels and commodity chemicals from carbon dioxide with electricity. *Current opinion in biotechnology*, 24(3), 385-390. <https://doi.org/10.1016/j.copbio.2013.02.012>

Macombe, C. (2020). Social life cycle assessment for industrial biotechnology. *Sustainability and life cycle assessment in industrial biotechnology*, 205-230. https://doi.org/10.1007/10_2019_99

Marchese, M., Buffo, G., Santarelli, M., & Lanzini, A. (2021). CO₂ from direct air capture as carbon feedstock for Fischer-Tropsch chemicals and fuels: Energy and economic analysis. *Journal of CO₂ Utilization*, 46, 101487. <https://doi.org/10.1016/j.jcou.2021.101487>

Masson-Delmotte, V., Zhai, P., Pirani, A., Connors, S. L., Péan, C., Berger, S., & Zhou, B. (2021). Climate change 2021: the physical science basis. *Contribution of working group I to the sixth assessment report of the intergovernmental panel on climate change*, 2.

McQueen, N., Gomes, K. V., McCormick, C., Blumanthal, K., Pisciotta, M., & Wilcox, J. (2021). A review of direct air capture (DAC): scaling up commercial technologies and innovating for the future. *Progress in Energy*, 3(3), 032001. <https://doi.org/10.1088/2516-1083/abf1ce>

Metz, B., Davidson, O., De Coninck, H. C., Loos, M., & Meyer, L. (2005). *IPCC special report on carbon dioxide capture and storage*. Cambridge: Cambridge University Press.

Odongo, J., Elizabeth, C., Francis, O., & Erick, O. (2019). Synthetic Biology Industrial Revolution, Social And Ethical Concerns. *International Journal of Research and Innovation in Applied Science (IJRIAS)*, 4(8), 45-48. <https://doi.org/10.6084/m9.figshare.9975407>

Olesen, B. W., Bogatu, D. I., Kazanci, O. B., & Coakley, D. (2021). The use of CO₂ as an indicator for indoor air quality and control of ventilation according to EN16798-1 and TR16798-2. In *ROOMVENT2020 (A conference paper)*, Torino, Italy.

Olivier, J.G.J., Peters, J.A.H.W., 2020. Trends in Global CO₂ and Total Greenhouse Gas Emissions. PBL Netherlands Environ. Assess. Agency 2020, 70.

Park, D. U., & Ha, K. C. (2008). Characteristics of PM₁₀, PM_{2.5}, CO₂ and CO monitored in interiors and platforms of subway train in Seoul, Korea. *Environment International*, 34(5), 629-634. <https://doi.org/10.1016/j.envint.2007.12.007>

Patel, S., & Sharma, S. (2021). Respiratory Acidosis. In *StatPearls [Internet]*. StatPearls Publishing.

Peridas, G., & Schmidt, B. M. (2021). The role of carbon capture and storage in the race to carbon neutrality. *The Electricity Journal*, 34(7), 106996. <https://doi.org/10.1016/j.tej.2021.106996>

Rodero, A., & Krawczyk, D. A. (2019). Carbon dioxide human Gains—A new approach of the estimation. *Sustainability*, 11(24), 7128.

Santoro, C., Arbizzani, C., Erable, B., & Ieropoulos, I. (2017). Microbial fuel cells: From fundamentals to applications. A review. *Journal of power sources*, 356, 225-244. <https://doi.org/10.1016/j.jpowsour.2017.03.109>

Santos, T. M., Júnior, J. N., Bessani, M., & Maciel, C. D. (2021). CO₂ Emissions Forecasting in Multi-Source Power Generation Systems Using Dynamic Bayesian Network. In *2021 IEEE International Systems Conference (SysCon)* (pp. 1-8). <https://doi.org/10.1109/SysCon48628.2021.9447104>

Schellevis, H. M., van Schagen, T. N., & Brilman, D. W. F. (2021). Process optimization of a fixed bed reactor system for direct air capture. *International journal of greenhouse gas control*, 110, 103431. <https://doi.org/10.1016/j.ijggc.2021.103431>

Schibuola, L., & Tambani, C. (2020). Indoor environmental quality classification of school environments by monitoring PM and CO₂ concentration levels. *Atmospheric Pollution Research*, 11(2), 332-342. <https://doi.org/10.1016/j.apr.2019.11.006>

Serrano-Ruiz, J. C. (2020). Biomass: a renewable source of fuels, chemicals and carbon materials. *Molecules*, 25(21), 5217. <https://doi.org/10.3390/molecules25215217>

Shamsuyeva, M., & Endres, H. J. (2021). Plastics in the context of the circular economy and sustainable plastics recycling: Comprehensive review on research development, standardization and market. *Composites Part C: open access*, 6, 100168. <https://doi.org/10.1016/j.jcomc.2021.100168>

Shen, G., Aniwaeer, S., Zhu, Y., Zheng, S., Hou, W., Shen, H., & Tao, S. (2020). Quantifying source contributions for indoor CO₂ and gas pollutants based on the highly resolved sensor

data. *Environmental Pollution*, 267, 115493. <https://doi.org/10.1016/j.envpol.2020.115493>

Shen, G., Hays, M. D., Smith, K. R., Williams, C., Faircloth, J. W., & Jetter, J. J. (2018). Evaluating the performance of household liquefied petroleum gas cookstoves. *Environmental science & technology*, 52(2), 904-915. <https://doi.org/10.1021/acs.est.7b05155>

Shukla, P.R., Skea, J., Slade, R., Khourdajie, A. Al, Diemen, R. van, McCollum, D., Pathak, M., Some, S., Vyas, P., Fradera, R., Belkacemi, M., Hasija, A., Lisboa, G., Luz, S., Malley, J., (2022). IPCC, 2022: Summary for Policymakers. In: Climate Change 2022: Mitigation of Climate Change. Contribution of Working Group III to the Sixth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge, UK. <https://doi.org/10.18356/9789210012973c007>

Singh, D., Sharma, D., Soni, S. L., Sharma, S., Sharma, P. K., & Jhalani, A. (2020). A review on feedstocks, production processes, and yield for different generations of biodiesel. *Fuel*, 262, 116553. <https://doi.org/10.1016/j.fuel.2019.116553>

Skoczinski, P., Carus, M., de Guzman, D., Káb, H., Chinthapalli, R., Ravenstijn, J., & Raschka, A. (2021). Bio-based Building Blocks and Polymers—Global Capacities, Production and Trends 2020–2025—Short Version. *Language*, 16.

Smith-Doerr, L. (2004). Flexibility and fairness: Effects of the network form of organization on gender equity in life science careers. *Sociological Perspectives*, 47(1), 25-54. <https://doi.org/10.1525/sop.2004.47.1.25>

Sodiq, A., Abdullatif, Y., Aissa, B., Ostovar, A., Nassar, N., El-Naas, M., & Amhamed, A. (2023). A review on progress made in direct air capture of CO₂. *Environmental Technology & Innovation*, 102991. <https://doi.org/10.1016/j.eti.2022.102991>

Solomon, S., Qin, D., Manning, M., Averyt, K., & Marquis, M. (Eds.). (2007). *Climate change 2007-the physical science basis: Working group I contribution to the fourth assessment report of the IPCC* (Vol. 4). Cambridge university press.

Tamme, E. (2021). Brief-Carbon removal with CCS technologies.

Tremblay, P. L., Angenent, L. T., & Zhang, T. (2017). Extracellular electron uptake: among autotrophs and mediated by surfaces. *Trends in biotechnology*, 35(4), 360-371. <https://doi.org/10.1016/j.tibtech.2016.10.004>

Tseng, C. P., Liu, F., Zhang, X., Huang, P. C., Campbell, I., Li, Y., & Verduzco, R. (2022). Solution-deposited and Patternable conductive polymer thin-film electrodes for microbial bioelectronics. *Advanced Materials*, 34(13), 2109442. <https://doi.org/10.1002/adma.202109442>

- US EPA (2021). Inventory of U.S Greenhouse Gas Emissions and Sinks, EPA 430-R-21-005.
- Valentić, V., Žiković, S., & Višković, A. (2016). Can CCS save coal fired power plants—The European perspective. *International Journal of Greenhouse Gas Control*, 47, 266-278. <https://doi.org/10.1016/j.ijggc.2016.01.037>
- Van Eerten-Jansen, M. C., Heijne, A. T., Buisman, C. J., & Hamelers, H. V. (2012). Microbial electrolysis cells for production of methane from CO₂: long-term performance and perspectives. *International Journal of Energy Research*, 36(6), 809-819. <https://doi.org/10.1002/er.1954>
- Venkatesh, A., Posen, I. D., MacLean, H. L., Chu, P. L., Griffin, W. M., & Saville, B. A. (2020). Environmental Aspects of Biotechnology. *Advances in biochemical engineering/biotechnology*, 173, 77–119. https://doi.org/10.1007/10_2019_98
- Villano, M., Aulenta, F., Ciucci, C., Ferri, T., Giuliano, A., & Majone, M. (2010). Bioelectrochemical reduction of CO₂ to CH₄ via direct and indirect extracellular electron transfer by a hydrogenophilic methanogenic culture. *Bioresource technology*, 101(9), 3085-3090. <https://doi.org/10.1016/j.biortech.2009.12.077>
- Wang, H., Liu, Y., Laaksonen, A., Krook-Riekkola, A., Yang, Z., Lu, X., & Ji, X. (2020). Carbon recycling—An immense resource and key to a smart climate engineering: A survey of technologies, cost and impurity impact. *Renewable and Sustainable Energy Reviews*, 131, 110010. <https://doi.org/10.1016/j.rser.2020.110010>
- Woodall, C. M., McQueen, N., Pilorgé, H., & Wilcox, J. (2019). Utilization of mineral carbonation products: current state and potential. *Greenhouse Gases: Science and Technology*, 9(6), 1096-1113. <https://doi.org/10.1002/ghg.1940>
- World Health Organization (2021). *Air Quality Guidelines - Update 2021*. Copenhagen, Denmark: WHO Regional Office for Europe.
- Yu, Q., & Brilman, W. (2020). A Radial Flow Contactor for Ambient Air CO₂ Capture. *Applied Sciences*. <https://doi.org/10.3390/app10031080>
- Yang, H. Y., Bao, B. L., Liu, J., Qin, Y., Wang, Y. R., Su, K. Z., & Mu, Y. (2018). Temperature dependence of bioelectrochemical CO₂ conversion and methane production with a mixed-culture biocathode. *Bioelectrochemistry*, 119, 180-188. <https://doi.org/10.1016/j.bioelechem.2017.10.002>
- Yang, L., Xu, M., Fan, J., Liang, X., Zhang, X., Lv, H., & Wang, D. (2021). Financing coal-fired power plant to demonstrate CCS (carbon capture and storage) through an innovative policy incentive in China. *Energy Policy*, 158, 112562. <https://doi.org/10.1016/j.enpol.2021.112562>

Zhang, W., Liu, H., Sun, C., Drage, T. C., & Snape, C. E. (2014). Capturing CO₂ from ambient air using a polyethyleneimine–silica adsorbent in fluidized beds. *Chemical Engineering Science*, 116, 306-316. <https://doi.org/10.1016/j.ces.2014.05.018>

Zhou, H., Xing, D., Xu, M., Su, Y., Ma, J., Angelidaki, I., & Zhang, Y. (2021). Optimization of a newly developed electromethanogenesis for the highest record of methane production. *Journal of Hazardous Materials*, 407, 124363. <https://doi.org/10.1016/j.jhazmat.2020.124363>