Methanosarcina barkeri in Biohybrid Systems for Carbon Dioxide Reduction: A Survey

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Abstract

This survey paper explores the pivotal role of Methanosarcina barkeri in biohybrid systems designed for carbon dioxide (CO) reduction. Methanosarcina barkeri, a versatile methanogenic archaeon, is distinguished by its metabolic flexibility and electron transfer capabilities, which are integral to microbial electrosynthesis processes. The organism's ability to perform direct and interspecies electron transfer enhances the efficiency of CO conversion into valuable chemical products. This survey also highlights the integration of advanced materials such as graphene to facilitate electron transfer and microbial attachment, thereby optimizing the performance of these biohybrid systems. Furthermore, innovative reactor designs, including zero-gap vapor-fed configurations, are discussed for their potential to enhance system efficiency by minimizing ohmic resistance and maintaining favorable environmental conditions. The paper addresses challenges in material and system integration, emphasizing the need for improved cathode designs and the understanding of syntrophic interactions within microbial communities. Future research directions include optimizing electron transfer mechanisms, exploring genetic modifications to enhance microbial capabilities, and integrating machine learning techniques to improve system adaptability. The survey concludes by acknowledging the ethical considerations of using living cells in technology and the potential for biohybrid systems to contribute to sustainable energy solutions and carbon sequestration efforts.

1 Introduction

1.1 Significance of Carbon Dioxide Reduction

The reduction of carbon dioxide (CO_2) emissions is a crucial environmental and economic necessity, driven by the urgent demand for sustainable alternatives to fossil-based production in the face of climate change [1]. The inherent inefficiency of natural photosynthesis in converting solar energy into chemical energy highlights the need for innovative CO_2 reduction strategies, particularly those that enhance solar-to-chemical energy conversion. Methanogens, including Methanosarcina barkeri, play a significant role in the global carbon cycle, underscoring the importance of CO_2 reduction efforts [2]. Furthermore, valorizing atmospheric CO_2 into useful compounds offers a sustainable solution to mitigate the environmental impacts of carbon emissions [3]. Economically, robust biohybrid systems can optimize resource allocation and minimize waste in distributed systems under varying workloads. Integrating biological entities with artificial materials in these systems presents a promising avenue to replicate and enhance biological functionalities, ultimately improving performance beyond that achievable with synthetic materials alone [4].

1.2 Role of Methanosarcina barkeri in Biohybrid Systems

Methanosarcina barkeri is essential in developing biohybrid systems due to its remarkable metabolic capabilities and ability to facilitate extracellular electron transfer, which is vital for enhancing CO_2

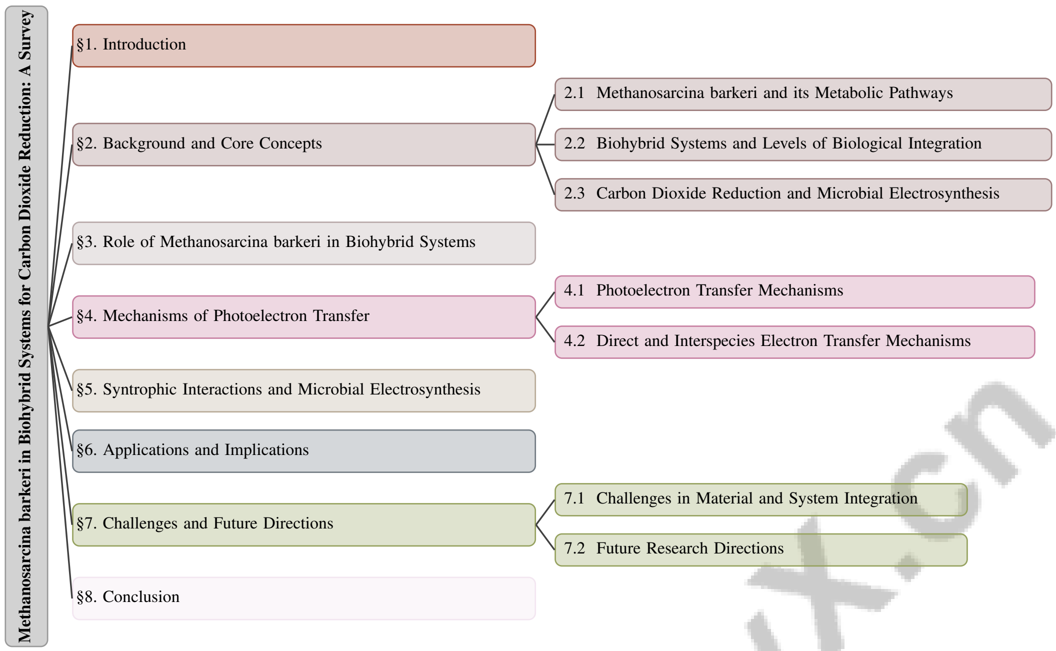

Figure 1: chapter structure

reduction efficiency [5]. By incorporating Methanosarcina barkeri into Photosynthetic Biohybrid Systems (PBSs), researchers aim to address the limitations of natural photosynthesis, thereby increasing solar fuel production through the synergistic interaction of biological catalysts and inorganic substrates [6]. Its proficiency in direct electron transfer (DET) and direct interspecies electron transfer (DIET) further emphasizes its adaptability and effectiveness within these systems [5].

Advancements in electrode materials have bolstered the role of Methanosarcina barkeri by enhancing microbial attachment and facilitating efficient electron transfer, thereby optimizing CO_2 conversion rates [7]. The organism's metabolic diversity enables it to utilize a wide range of substrates, distinguishing it from other methanogens and expanding its applicability across various biohybrid frameworks [2]. Notably, its resilience to perchlorates, demonstrated by distinct transcriptional responses at varying temperatures, suggests potential applications in extraterrestrial environments, particularly on Mars, where perchlorates are common [8].

In microbial electrosynthesis systems, the interactions of Methanosarcina barkeri with other electron-uptaking and product-forming microorganisms are crucial for maximizing process efficiency [9]. The syntrophic relationships fostered by Methanosarcina barkeri in low-energy environments are instrumental in methane production and organic matter degradation, highlighting the organism's pivotal role in microbial partnerships [10]. These interactions, coupled with its unique metabolic pathways, position Methanosarcina barkeri as a cornerstone in the evolution of biohybrid systems aimed at sustainable CO_2 reduction and energy production.

1.3 Structure of the Survey

This survey is meticulously structured to provide a comprehensive exploration of Methanosarcina barkeri's role in biohybrid systems for CO_2 reduction. It begins with an **Introduction** that emphasizes the environmental and economic significance of CO_2 reduction and the central role of Methanosarcina barkeri in biohybrid systems. Following this, a detailed **Background and Core Concepts** section elucidates essential concepts, including the metabolic pathways of Methanosarcina barkeri, the intricacies of biohybrid systems, and the mechanisms of microbial electrosynthesis and biocatalysis.

The third section, **Role of Methanosarcina barkeri in Biohybrid Systems**, delves into the unique characteristics and metabolic pathways of Methanosarcina barkeri that facilitate its function in these systems. The **Mechanisms of Photoelectron Transfer** section examines the processes that enable effective electron transfer, highlighting the integration of synthetic and biological components.

Subsequently, **Syntrophic Interactions and Microbial Electrosynthesis** analyzes the cooperative interactions between Methanosarcina barkeri and other microorganisms, emphasizing their contributions to enhancing CO_2 reduction efficiency. The survey further explores the of innovative reactor designs, particularly the integration of photosynthetic biohybrid systems for solar fuel production, and examines potential extraterrestrial applications, such as utilizing methanogenic archaea in Martian environments to generate methane from perchlorates [6, 8, 5, 11].

The penultimate section, **Challenges and Future Directions**, identifies current challenges and proposes future research avenues to enhance system efficiency. Finally, the **Conclusion** synthesizes key findings, reinforcing the importance of Methanosarcina barkeri in advancing biohybrid systems for sustainable energy production.The following sections are organized as shown in Figure 1.

2 Background and Core Concepts

2.1 Methanosarcina barkeri and its Metabolic Pathways

Methanosarcina barkeri, a versatile methanogenic archaeon, plays a crucial role in energy conservation through its unique metabolic pathways, which accommodate a wide substrate range, distinguishing it from other methanogens. This flexibility is enhanced by cytochromes that facilitate essential electron transfer for methanogenesis [2]. Notably, M. barkeri can perform electromethanogenesis from a cathode at -400 mV, indicating its potential for direct electron uptake in bioelectrochemical systems [12].

The organism thrives in syntrophic interactions, collaborating with other microorganisms to degrade organic substrates under challenging thermodynamic conditions [10]. These interactions are vital for energy conservation, supporting M. barkeri's growth and methanogenesis in energy-limited environments. Computational models highlight the emergence of these syntrophic relationships, underscoring the importance of metabolic exchanges for optimized energy utilization [13].

M. barkeri's resilience to extreme environments, such as high perchlorate salt concentrations, demonstrates its adaptability and regulatory flexibility, essential for methane production under conditions resembling extraterrestrial environments like Mars [8]. Its ability to adjust metabolic pathways in response to environmental stressors reflects sophisticated energy conservation mechanisms crucial for survival across diverse ecological niches.

2.2 Biohybrid Systems and Levels of Biological Integration

Biohybrid systems combine biological entities with synthetic materials, utilizing the unique functionalities of living organisms for technological advancements. These systems are categorized by biological integration levels, ranging from nanoscale to macroscale, each with distinct advantages and challenges [4]. At the nanoscale, biohybrids exploit cellular molecular machinery, such as enzymes and proteins, for precise biochemical reactions, benefiting applications like targeted drug delivery and biosensing.

At the microscale, integration involves entire cells or components, enabling complex interactions and functionalities. Methanogens like M. barkeri are vital here due to their diverse metabolic pathways and robust energy conservation mechanisms [2]. They can be engineered into microbial electrosynthesis systems, leveraging metabolic flexibility and direct electron transfer capabilities to convert carbon dioxide into valuable compounds.

At the macroscale, biohybrid systems encompass whole organisms or ecosystems, facilitating large-scale applications such as bioreactors for industrial biocatalysis and environmental remediation. Designing advanced bioreactors for methanogenic communities is critical for optimizing carbon dioxide reduction processes, particularly in microbial electrosynthesis. Defined co-cultures convert CO_2 into valuable products like methane and acetate, utilizing cathodic electrons as an energy source, enhancing electromethanogenesis rates and related bioconversion processes [1, 9, 3]. Categorizing biohybrid systems by biological integration levels enables strategic design to address specific technological challenges, advancing sustainable energy production and environmental management.

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2.3 Carbon Dioxide Reduction and Microbial Electrosynthesis

Microbial electrosynthesis (MES) is a transformative technology that employs electroactive microorganisms to convert carbon dioxide (CO_2) into organic compounds, significantly contributing to CO_2 reduction [3]. This process captures CO_2 for electricity-driven bioproduction of organics like acetic acid, crucial across various industries. MES systems, however, face challenges such as low production rates and adverse pH changes affecting biocathodes, necessitating further optimization for enhanced efficiency [14].

Integrating Methanosarcina species in MES systems addresses these inefficiencies, as they can retrieve electrons from extracellular sources, including electrodes and electrogenic partners [5]. M. barkeri significantly enhances MES due to its unique metabolic pathways and direct electron transfer capabilities, improving CO_2 conversion selectivity and efficiency. Its metabolic flexibility allows utilization of various substrates, optimizing valuable chemical product production [7].

Innovative reactor designs have been proposed to improve biomass accumulation and overall system performance, which are critical for efficient CO_2 reduction [15]. Hydrogen-driven microbial electrosynthesis (HDME) further aids CO_2 reduction by combining hydrogen production and microbial CO_2 reduction in a single reactor, streamlining the process and enhancing efficiency [16]. Additionally, organic photocatalysts improve charge transfer and stability in biohybrid systems, supporting the MES process [17].

Despite these advancements, the inherent inefficiency of natural photosynthesis compared to inorganic semiconductor photovoltaics remains a significant challenge, highlighting the need for continued research and innovation in MES [6]. Transcriptomic analyses under experimental conditions simulating Martian environments have provided insights into methanogenesis under stress conditions, emphasizing MES systems' potential for both terrestrial and extraterrestrial applications [8]. These findings underscore the importance of MES in achieving sustainable CO_2 reduction and resource utilization.

In recent years, the exploration of biohybrid systems has garnered significant attention, particularly in the context of microbial electrosynthesis. A notable organism in this field is Methanosarcina barkeri, which exhibits a remarkable ability to adapt and thrive in various environments. Figure 2 illustrates the role of Methanosarcina barkeri in biohybrid systems, highlighting its unique characteristics, metabolic pathways, and energy conservation mechanisms. This figure emphasizes the organism's metabolic versatility and electron transfer capabilities, as well as its environmental resilience and syntrophic relationships. Furthermore, it showcases how Methanosarcina barkeri integrates with advanced materials and innovative reactor designs to optimize carbon dioxide reduction and enhance energy conservation in microbial electrosynthesis systems. By understanding these attributes, researchers can better leverage this microorganism's potential in sustainable energy solutions.

3 Role of Methanosarcina barkeri in Biohybrid Systems

3.1 Unique Characteristics of Methanosarcina barkeri

Methanosarcina barkeri is a pivotal organism in biohybrid systems, particularly for carbon dioxide reduction, due to its metabolic versatility and ability to utilize various substrates for methanogenesis [18]. Its proficiency in direct electron transfer (DET) and direct interspecies electron transfer (DIET) is crucial for microbial electrosynthesis (MES), enabling effective electron uptake and conversion processes [11]. M. barkeri's resilience in extreme environments, such as high salinity and variable pH, enhances its applicability in diverse settings, including extraterrestrial environments. This robustness is complemented by its capacity to form syntrophic relationships with microorganisms like Clostridium and Sporomusa, optimizing CO_2 conversion into valuable chemicals, thereby improving MES performance [19, 3]. The integration of advanced materials, such as graphene, enhances microbial attachment and electron transfer in MES, boosting chemical yields and system efficiency [11]. This synergy between biological and synthetic components underscores M. barkeri's potential to drive innovations in sustainable energy and carbon sequestration technologies, leveraging its unique metabolic pathways and electron transfer capabilities to advance next-generation biohybrid systems.

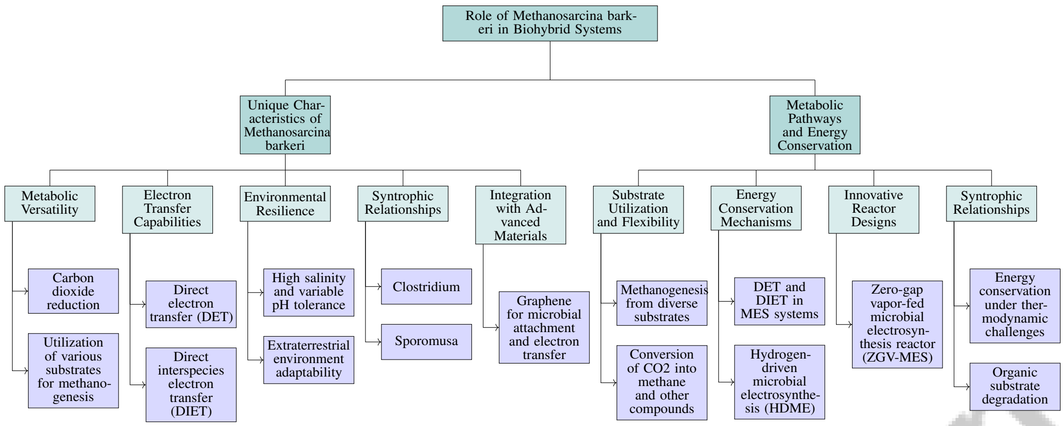

Figure 2: This figure illustrates the role of Methanosarcina barkeri in biohybrid systems, highlighting its unique characteristics, metabolic pathways, and energy conservation mechanisms. It emphasizes the organism's metabolic versatility, electron transfer capabilities, environmental resilience, and syntrophic relationships, as well as its integration with advanced materials and innovative reactor designs to optimize carbon dioxide reduction and energy conservation in microbial electrosynthesis systems.

3.2 Metabolic Pathways and Energy Conservation

Methanosarcina barkeri's metabolic pathways are integral to energy conservation and carbon dioxide reduction, making it a key organism in biohybrid systems. Its capacity to utilize diverse substrates for methanogenesis demonstrates its metabolic flexibility, facilitating the conversion of CO_2 into methane and other valuable compounds. This flexibility is enhanced by its adeptness in DET and DIET, crucial for energy conservation and CO_2 reduction in MES systems [19]. In bioelectrochemical systems, M. barkeri optimizes microbial electrosynthesis of acetate from CO_2 by enhancing biomass accumulation at the cathode, ensuring efficient electron transfer and nutrient flow [15].

Figure 3 illustrates the hierarchical structure of metabolic pathways and energy conservation strategies in Methanosarcina barkeri, focusing on metabolic flexibility, bioelectrochemical optimization, and environmental conditions critical for efficient CO_2 reduction and methane production. The incorporation of hydrogen-driven microbial electrosynthesis (HDME) further enhances energy conservation by using non-precious metal cathodes for in situ hydrogen production, streamlining the reduction process and improving system efficiency [16]. Innovative reactor designs, such as the zero-gap vapor-fed microbial electrosynthesis reactor (ZGV-MES), reduce ohmic resistance and maintain near-neutral pH, conditions conducive to higher methane and chemical production rates [14]. These advancements emphasize the importance of optimizing environmental conditions and material interfaces to maximize M. barkeri's metabolic potential in CO_2 reduction. Its metabolic pathways are bolstered by syntrophic relationships with other microorganisms, essential for energy conservation under thermodynamically challenging conditions, facilitating efficient organic substrate degradation and sustained methane production [19]. Through these advanced metabolic processes, M. barkeri continues to propel innovation in sustainable energy and carbon sequestration technologies.

4 Mechanisms of Photoelectron Transfer

4.1 Photoelectron Transfer Mechanisms

Photoelectron transfer mechanisms are pivotal for optimizing carbon dioxide conversion in biohybrid systems. Advanced materials like graphene significantly enhance electron transfer and microbial attachment, improving microbial electrosynthesis (MES) performance by facilitating electron flow and microbial loading [11]. This enhancement supports the growth of methanogens such as *Methanosarcina barkeri*, crucial for effective CO_2 reduction.

Bioelectrochemical reactors with reticulated vitreous carbon foam cathodes under galvanostatic control further enhance microbial growth and acetate production by mitigating product inhibition

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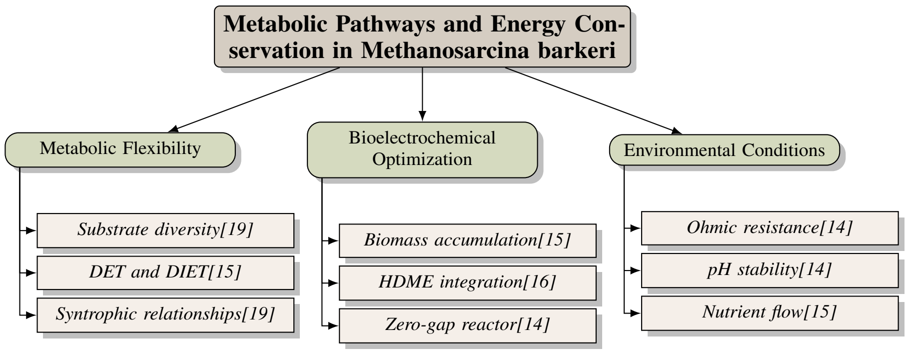
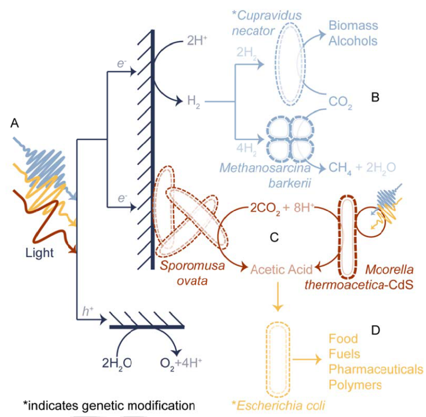


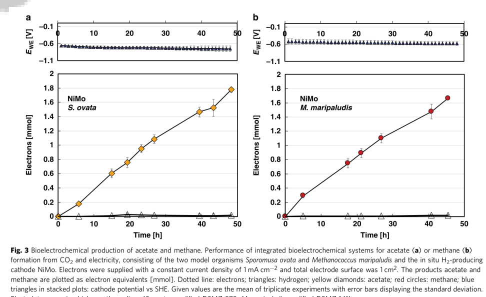
Figure 3: This figure illustrates the hierarchical structure of metabolic pathways and energy conservation strategies in Methanosarcina barkeri, focusing on metabolic flexibility, bioelectrochemical optimization, and environmental conditions critical for efficient CO2 reduction and methane production.

[15]. These reactors maintain electroautotrophic activity, enabling continuous organic compound production even during power interruptions [19]. Such resilience is vital for sustaining high microbial activity and consistent chemical production in biohybrid systems.

Innovative reactor designs, featuring closely spaced electrodes in a vapor-fed configuration, optimize photoelectron transfer by promoting effective proton transport to the cathode, minimizing pH fluctuations, and enhancing microbial efficiency in chemical production [14]. By maintaining optimal conditions and integrating advanced materials, biohybrid systems achieve superior photoelectron transfer performance, driving efficient CO_2 reduction and sustainable chemical production.



(a) Bioremediation and Bioprocessing: A Comprehensive Overview[6]



(b) Bioelectrochemical production of acetate and methane: Performance of integrated bioelectrochemical systems for acetate (a) or methane (b) formation from CO2 and electricity, consisting of the two model organisms Sporomusa ovata and Methanococcus maripaludis and the in situ H2-producing cathode NiMo.[16]

Figure 4: Examples of Photoelectron Transfer Mechanisms

As illustrated in Figure 4, photoelectron transfer mechanisms are exemplified in bioremediation and bioprocessing, and bioelectrochemical production of acetate and methane. The first scenario provides a comprehensive overview of photosynthesis techniques, illustrating the conversion of light energy into chemical energy. The second scenario emphasizes integrated bioelectrochemical systems for acetate and methane production, showcasing *Sporomusa ovata* and *Methanococcus maripaludis*, alongside an in situ hydrogen-producing NiMo cathode, for converting CO_2 and electricity into valuable chemicals. These examples highlight innovative approaches in photoelectron transfer

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mechanisms and their potential applications in sustainable energy and environmental remediation [6, 16].

4.2 Direct and Interspecies Electron Transfer Mechanisms

Method Name	Electron Transfer Mechanisms	Microbial Interactions	Efficiency Influencers
MES[19]	Bioelectrochemical Process	Direct Electron Transfer	Power Supply Disruptions
ZGV-MES[14]	Vapor-fed Anode	Direct Electron Transfer	Reactor Design
EMEDC[9]	Hydrogen Transfer	Interspecies Hydrogen Transfer	Cathode Potentials

Table 1: Overview of Electron Transfer Mechanisms, Microbial Interactions, and Efficiency Influencers in Microbial Electrosynthesis Systems. The table details various methods, including MES, ZGV-MES, and EMEDC, highlighting their electron transfer mechanisms, microbial interactions, and factors influencing efficiency, such as power supply disruptions, reactor design, and cathode potentials.

Electron transfer mechanisms are crucial for biohybrid system efficiency, particularly in microbial electrosynthesis involving *Methanosarcina barkeri*. Direct electron transfer (DET) and direct interspecies electron transfer (DIET) are primary pathways facilitating electron exchange between microorganisms and electrodes or among different microbial species [11]. These mechanisms optimize CO_2 conversion into methane and other valuable compounds by enabling efficient electron flow and energy conservation.

In DET, *Methanosarcina barkeri* directly receives electrons from an electrode, aided by its unique metabolic pathways and conductive materials like graphene, which enhance electron transfer capabilities and promote methanogen activity [11]. This direct interaction reduces energy losses and maximizes CO_2 reduction efficiency.

DIET, on the other hand, involves electron transfer between microbial species, often mediated by conductive pili or extracellular polymeric substances. *Methanosarcina barkeri* forms syntrophic partnerships with other microorganisms capable of producing hydrogen or formate, acting as electron carriers [19]. These interspecies interactions are essential for maintaining energy flow and stability in microbial ecosystems, particularly under thermodynamically challenging conditions.

The efficiency of DET and DIET is influenced by factors such as pH, temperature, and electron mediators. Innovative reactor designs optimizing these conditions can significantly enhance electron transfer mechanisms, improving the overall efficiency of biohybrid systems for CO_2 reduction [14].

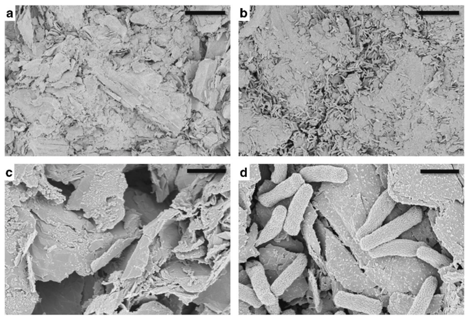
As illustrated in Figure 6, the hierarchical structure of electron transfer mechanisms in microbial electrosynthesis is highlighted, with DET and DIET as key pathways. This figure emphasizes the role of graphene in facilitating DET, the significance of conductive pili in DIET, and various factors influencing efficiency, such as pH and innovative reactor designs. By understanding and leveraging these pathways, researchers can develop more effective biohybrid systems that harness the potential of *Methanosarcina barkeri* and other electroactive microorganisms for sustainable energy production and environmental remediation. Table 1 provides a detailed comparison of different methods employed in microbial electrosynthesis, focusing on electron transfer mechanisms, microbial interactions, and factors influencing system efficiency.

Together, these examples provide a comprehensive overview of microbial electron transfer processes in diverse environments [9, 5].

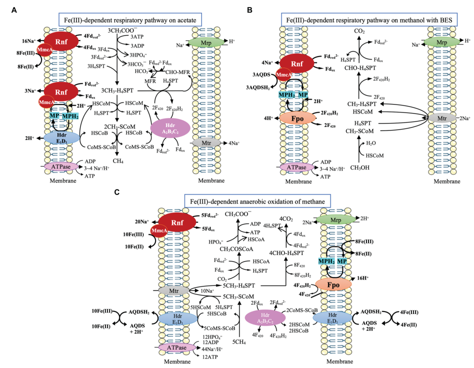
5 Syntrophic Interactions and Microbial Electrosynthesis

5.1 Syntrophic Interactions in Microbial Communities

Syntrophic interactions play a pivotal role in enhancing microbial electrosynthesis, especially in systems involving *Methanosarcina barkeri*. These interactions involve cooperative metabolic exchanges that facilitate the breakdown of complex organic substrates and the conversion of carbon dioxide into valuable products [13]. The ecological and evolutionary resilience of these mechanisms is crucial for maintaining system stability and performance under diverse environmental conditions [20].



(a) Microbial Colonization on a Synthetic Polymer Surface[9]



(b) Fe(III)-dependent respiratory pathways on acetate and methanol with BES[5]

Figure 5: Examples of Direct and Interspecies Electron Transfer Mechanisms

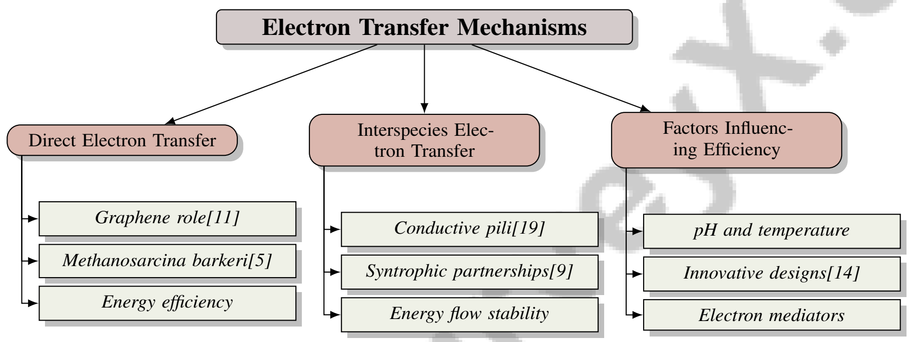


Figure 6: This figure illustrates the hierarchical structure of electron transfer mechanisms in microbial electrosynthesis, highlighting direct electron transfer (DET) and interspecies electron transfer (DIET) as key pathways. It emphasizes the role of graphene in DET, the significance of conductive pili in DIET, and factors such as pH and innovative reactor designs influencing efficiency.

These metabolic interdependencies can emerge independently of co-evolution, highlighting their adaptability to environmental pressures [13]. This adaptability is vital for optimizing microbial electrosynthesis, allowing dynamic adjustments of metabolic pathways to maximize energy conservation and product yield. *Methanosarcina barkeri* is central to these interactions, utilizing its metabolic flexibility and electron transfer capabilities to partner with microorganisms that produce hydrogen or formate, thereby effectively converting carbon dioxide into methane [8]. This cooperation not only boosts microbial electrosynthesis efficiency but also enhances the resilience and adaptability of the microbial community.

Understanding syntrophic interactions and their impact on microbial electrosynthesis is key to advancing biohybrid systems. By leveraging the cooperative dynamics of microbial communities, researchers can engineer more resilient and efficient systems, promoting sustainable energy production and carbon dioxide reduction. Defined co-cultures have been shown to enhance microbial electrosynthesis, with specific strains synergistically optimizing electron uptake and improving methane and acetate production rates [9, 20].

5.2 Syntrophic Interactions in Methane Production

Syntrophic interactions are essential for efficient methane production in microbial communities, particularly through the cooperation between *Methanosarcina barkeri* and other microorganisms. These interactions involve the exchange of metabolites like hydrogen and formate, crucial intermediates in methanogenesis. *Methanosarcina barkeri*'s metabolic versatility enables it to utilize these intermediates to convert carbon dioxide into methane, thereby enhancing production efficiency [13].

As illustrated in Figure 7, the hierarchical structure of syntrophic interactions in methane production highlights key factors, ecological influences, and future research directions that are critical to understanding these complex relationships. The effectiveness of these interactions is influenced by ecological factors such as substrate availability, environmental conditions, and the presence of electron donors and acceptors. Future research should focus on these factors to better understand the conditions that promote and sustain syntrophy, ultimately optimizing methane production processes [13]. By elucidating these complex interplays, researchers can develop strategies to enhance cooperative dynamics within microbial communities, thereby improving biohybrid systems for sustainable methane production and carbon dioxide reduction.

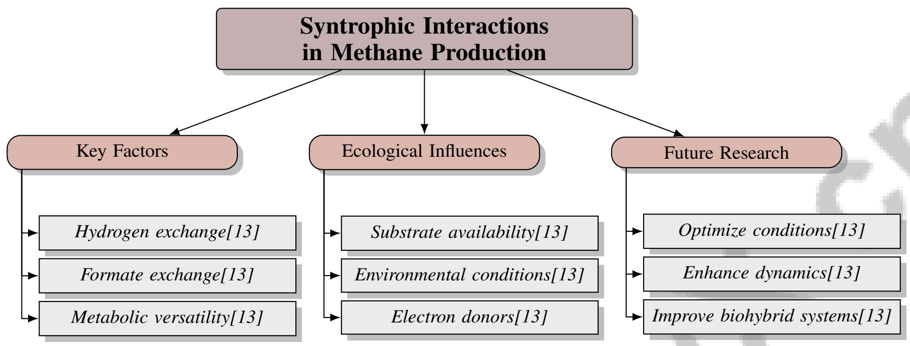


Figure 7: This figure illustrates the hierarchical structure of syntrophic interactions in methane production, highlighting key factors, ecological influences, and future research directions.

6 Applications and Implications

6.1 Innovative Reactor Designs for Enhanced Efficiency

Innovative reactor designs are pivotal for optimizing the efficiency of microbial electrosynthesis (MES) systems, which convert carbon dioxide into valuable compounds such as acetate and butyrate. These designs enhance electron transfer and microbial interactions, significantly boosting MES performance and contributing to greenhouse gas reduction [7, 9, 16, 3, 19]. Recent advancements in reactor design have demonstrated significant improvements in productivity and sustainability, highlighting the importance of maximizing chemical production rates.

A significant development is the zero-gap vapor-fed MES reactor, achieving methane production rates of 2.9 ± 1.2 L/L-d at a low voltage of 3.1 V [14]. This reactor design optimizes electrode arrangements and minimizes ohmic resistance, improving electron transfer efficiency and chemical output, with the low voltage requirement underscoring potential energy savings.

Integrating advanced materials and strategic reactor configurations can notably enhance biomass accumulation and product yield. For example, a study reported a space time yield of 0.78 g/L/h for acetate production, underscoring the crucial role of reactor design in sustaining high microbial activity [15]. Addressing inefficiencies in biohybrid systems, such as low solar-to-chemical energy conversion rates, through innovative designs can leverage biological and synthetic material synergies to improve light harvesting and metabolic efficiency. These advancements support more effective and sustainable energy production methods [6, 17, 4], contributing to the broader goal of developing scalable technologies for environmental and energy applications.

6.2 Extraterrestrial Applications

Photosynthetic Biohybrid Systems (PBSs) hold significant promise for extraterrestrial applications, where traditional chemical manufacturing is challenged by limited resources and harsh environments [6]. PBSs, which integrate biological components with synthetic materials, offer versatile solutions for in-situ resource utilization and sustainable production of essential compounds in space.

In settings like Mars, PBSs efficiently capture solar energy and convert carbon dioxide into valuable chemicals, essential for life support and fuel production, addressing the energy and resource needs of space missions. By combining inorganic materials with biological catalysts, PBSs offer a viable solution for reducing greenhouse gas emissions and supporting human habitation on other planets [6, 1, 11, 3, 19]. The resilience and metabolic flexibility of *Methanosarcina barkeri* make it an ideal candidate for these systems, enhancing its suitability for extraterrestrial biohybrid applications through direct electron transfer and syntrophic relationships.

Harnessing the unique characteristics of PBSs, which integrate efficient solar energy capture and biological catalysts for CO2 conversion, alongside *Methanosarcina barkeri*'s capabilities, researchers can develop advanced biohybrid technologies tailored for space environments. These innovations address energy and resource challenges in space exploration [6, 8], potentially revolutionizing resource management and utilization in extraterrestrial settings, paving the way for sustainable exploration and habitation beyond Earth.

7 Challenges and Future Directions

7.1 Challenges in Material and System Integration

Integrating materials and systems in biohybrid applications involving *Methanosarcina barkeri* presents notable challenges that must be addressed to enhance system performance. A significant issue is the high internal resistance in microbial electrosynthesis (MES) systems, which restricts current densities and CO_2 conversion efficiency [14]. This resistance impedes optimal electron flow and energy transfer. Moreover, integrating biological and inorganic components is often hindered by kinetic performance limitations, necessitating the optimization of synergistic interactions [6]. The susceptibility of biological systems to oxidative damage further complicates integration, highlighting the need for protective strategies to maintain system integrity.

Variability in electron transfer capabilities among *Methanosarcina* species, such as the differing electromethanogenesis performance between *Methanosarcina barkeri* and *Methanosarcina horonobensis*, underscores the necessity for a thorough understanding of the molecular mechanisms governing electron transfer in methanogens to improve biohybrid system design [12, 5]. Continuous flow in bioelectrochemical reactors can lead to planktonic biomass loss, reducing productivity and challenging cell retention strategies [15]. Prolonged operation may also decrease microbial activity due to nutrient depletion, critical for long-term functionality [16].

Insufficient characterization of methanogens' metabolic and regulatory responses to environmental stressors, such as perchlorate exposure, limits our understanding of their survival mechanisms under Martian-like conditions, hindering the development of robust biohybrid systems [8]. Additionally, the low efficiency of MES processes and high commercialization costs pose significant barriers to widespread adoption [3]. Addressing these challenges requires developing improved microbial strains and innovative reactor designs that enhance performance and reduce operational costs. Fluctuations or interruptions in electricity supply can damage microbial communities, further compromising system robustness and necessitating strategies for stable power supply [19].

7.2 Future Research Directions

Future research in biohybrid systems utilizing *Methanosarcina barkeri* should prioritize several strategic areas to overcome existing challenges and enhance capabilities. Optimizing cathode design is critical to minimizing unwanted product formation and improving reactor scalability for larger applications [14]. This involves exploring advanced materials and hybrid structures to enhance electrochemical properties, thereby increasing efficiency and reducing costs.

Investigating the effects of spatial structuring on the stability of syntrophic interactions is essential, as these significantly impact microbial electrosynthesis efficiency. Understanding conditions that foster successful endosymbiosis could optimize microbial community dynamics for enhanced performance [20]. Additionally, integrating advanced machine learning techniques with existing methodologies, such as ARAA, could further enhance the adaptability and robustness of biohybrid systems by enabling precise control over system parameters and performance [21].

Research should also focus on cooperative dynamics within microbial communities, particularly interspecies electron transfer mechanisms. A deeper understanding of interactions between synthetic materials and biological systems will facilitate the development of more efficient biohybrid systems, significantly increasing CO_2 conversion into valuable chemicals and fuels, thus contributing to sustainable energy solutions and climate change mitigation [17, 6, 3]. Exploring genetic modifications in *Methanosarcina* species to enhance electron uptake capabilities could broaden their applicability in diverse biohybrid frameworks.

Achieving an optimal balance between hydrogen production and microbial CO_2 reduction rates is pivotal in MES, which harnesses microbial activity for CO_2 conversion into valuable organic compounds, addressing energy sustainability and greenhouse gas mitigation [3, 19]. Optimizing this balance could significantly improve the overall efficiency and robustness of MES systems. Expanding analyses to include diverse microbial communities and operational conditions will deepen the understanding of interactions and processes involved, ultimately leading to more resilient and adaptable biohybrid systems.

A comprehensive understanding of the molecular mechanisms underlying electron transfer in methanogens, along with an exploration of diverse electron transfer strategies—such as direct interspecies electron transfer (DIET) and extracellular electron uptake—will enhance biohybrid system design and efficiency. These systems could leverage the unique metabolic pathways and electroactive properties of methanogens, which interact with other microorganisms and conductive materials to optimize energy conversion and carbon cycling in anaerobic environments [12, 2, 5, 10]. Enhancing microbial efficiency through genetic engineering, improving electrode design, and integrating microbial electrosynthesis with other renewable energy systems are essential steps toward sustainable and scalable solutions for CO_2 reduction and energy production.

8 Conclusion

This survey underscores the critical influence of *Methanosarcina barkeri* in advancing biohybrid systems for carbon dioxide reduction, highlighting its unique metabolic functions and electron transfer processes that enhance microbial electrosynthesis efficiency. Its metabolic adaptability and proficiency in both direct and interspecies electron transfer are instrumental in converting carbon dioxide into useful compounds. Additionally, the selection of appropriate carbon-based materials and reactor configurations plays a significant role in enhancing product selectivity and biocompatibility, thereby improving the overall performance of biohybrid systems.

The survey also addresses the ethical considerations of employing living cells in technological innovations, emphasizing the need to tackle issues related to animal welfare and social equity as biohybrid technologies progress. Furthermore, the adoption of advanced techniques such as ARAA has demonstrated considerable improvements in resource management and system agility, highlighting the capability of biohybrid systems to operate effectively in fluctuating environments.

Future progress in this domain is anticipated to focus on refining material and system integration, boosting microbial efficiency through genetic modification, and exploring novel reactor designs to enhance the scalability and sustainability of biohybrid systems. These advancements will be pivotal in addressing the challenges of carbon dioxide reduction and promoting the development of sustainable energy solutions.

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