Bioscience Evidence 2024, Vol.14, No.4, 143-153
http://bioscipublisher.com/index.php/be

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# Process Study on Microbial Fixation of $CO_2$ and Its Conversion into Organic Acids

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Bioscience Evidence, 2024, Vol.14, No.4 doi: 10.5376/be.2024.14.0016
Received: 17 May, 2024
Accepted: 22 Jun., 2024
Published: 05 Jul., 2024

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**Preferred citation for this article**:
Li M.M., 2024, Process study on microbial fixation of $CO_2$ and its conversion into organic acids, Bioscience Evidence, 14(4): 143-153 (doi: 10.5376/be.2024.14.0016)

**Abstract** The study identified several natural and synthetic $CO_2$ fixation pathways, including the Calvin cycle, the Wood-Ljungdahl pathway, and the 3-hydroxypropionate cycle, among others. Key enzymes such as ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBisCO) and formate dehydrogenase were found to play crucial roles in these pathways. The research also highlighted the potential of specific bacterial strains, such as *Bacillus* sp. SS105, in enhancing $CO_2$ sequestration and lipid production for biodiesel applications. Additionally, the study demonstrated that metabolic engineering and optimization of microbial consortia could significantly improve the yields of organic acids like succinic acid and butyric acid. The findings of this study underscore the potential of microbial $CO_2$ fixation as a viable strategy for reducing greenhouse gas emissions and producing valuable organic acids. The identification of efficient microbial pathways and key enzymes, along with advancements in metabolic engineering, paves the way for future applications in sustainable chemical production and biofuel generation. Further research should focus on optimizing these processes to enhance their industrial applicability and economic feasibility.

**Keywords** $CO_2$ fixation; Microbial conversion; Organic acids; Metabolic engineering; RuBisCO; Formate dehydrogenase; *Bacillus* sp. SS105; Biodiesel; Succinic acid; Butyric acid

## 1 Introduction

The continuous rise in global $CO_2$ emissions has significantly contributed to climate change, leading to severe environmental consequences such as global warming, ocean acidification, and extreme weather events. The increasing concentration of $CO_2$ in the atmosphere is primarily due to human activities, including the burning of fossil fuels, deforestation, and industrial processes (Salehizadeh et al., 2020; Wang et al., 2023). These emissions have resulted in unprecedented levels of greenhouse gases, which trap heat in the atmosphere and disrupt the natural balance of the Earth's climate system (Salehizadeh et al., 2020).

Carbon fixation is a crucial process in mitigating the adverse effects of greenhouse gases. By converting $CO_2$ into organic compounds, carbon fixation helps reduce the overall concentration of $CO_2$ in the atmosphere. This process not only addresses the environmental impact of $CO_2$ emissions but also provides a sustainable approach to producing valuable chemicals and fuels. Microbial $CO_2$ fixation, in particular, has gained attention due to its potential to efficiently convert $CO_2$ into various organic acids and other value-added products (Salehizadeh et al., 2020; Chen et al., 2023; Wang et al., 2023).

Microbial $CO_2$ fixation involves the use of microorganisms to convert $CO_2$ into organic compounds through various metabolic pathways. Several natural and synthetic pathways have been identified, including the Calvin cycle, the Wood-Ljungdahl pathway, and the 3-hydroxypropionate/4-hydroxybutyrate cycle. These pathways enable microorganisms to assimilate $CO_2$ as a carbon source and produce a range of metabolites, such as organic acids, alcohols, and bioplastics (Salehizadeh et al., 2020; Zhang et al., 2020; Wang et al., 2023). Advances in genetic and metabolic engineering have further enhanced the efficiency and versatility of microbial $CO_2$ fixation processes (Salehizadeh et al., 2020; Zhang et al., 2020; Chen et al., 2023).

The conversion of $CO_2$ into organic acids is particularly relevant due to the high demand for these compounds in various industrial applications. Organic acids, such as acetic acid, succinic acid, and butyric acid, serve as key intermediates in the production of bioplastics, pharmaceuticals, and biofuels (Liu et al., 2018; Mateos et al., 2019;

Chen et al., 2023). Microbial electrosynthesis (MES) systems have shown promise in enhancing the conversion efficiency of $CO_2$ to organic acids by utilizing bioelectrochemical processes. These systems leverage the metabolic capabilities of microorganisms to produce organic acids with high selectivity and yield (Song et al., 2011; Liu et al., 2018; Mateos et al., 2019; Wang, 2024).

The primary objective of this study is to investigate the microbial fixation of $CO_2$ and its subsequent conversion into organic acids. This research aims to explore the various microbial pathways involved in $CO_2$ fixation, evaluate the efficiency of different microbial systems, and identify potential strategies to enhance the production of organic acids. By understanding and optimizing these processes, this study hopes to contribute to the development of sustainable and economically viable methods for reducing $CO_2$ emissions and producing valuable biochemicals.

## 2 Mechanisms of Microbial $CO_2$ Fixation

### 2.1 Overview of $CO_2$ fixation pathways

Microbial $CO_2$ fixation involves several distinct biochemical pathways that convert atmospheric $CO_2$ into organic compounds. The most well-known pathway is the Calvin-Bassham-Benson (CBB) cycle, which is prevalent in many autotrophic organisms and involves the enzyme ribulose-1,5-bisphosphate carboxylase/oxygenase (RubisCO) (Dangel and Tabita, 2015; Xiao et al., 2020; Asplund-Samuelsson and Hudson, 2021). Other pathways include the reductive citric acid cycle, the reductive acetyl-CoA pathway (Wood-Ljungdahl pathway), the 3-hydroxypropionate bicycle, the dicarboxylate/4-hydroxybutyrate cycle, and the 3-hydroxypropionate/4-hydroxybutyrate cycle (Sánchez-Andrea et al., 2020; Xiao et al., 2020; Chen et al., 2021). Recently, a seventh pathway, the reductive glycine pathway, has been identified in Desulfovibrio desulfuricans, which is highly ATP-efficient (Sánchez-Andrea et al., 2020).

### 2.2 Key microorganisms involved

Various microorganisms are involved in $CO_2$ fixation, including both photoautotrophic and chemoautotrophic bacteria. Cyanobacteria are well-known for their role in the CBB cycle, while other bacteria such as *Clostridium ljungdahlii* utilize the Wood-Ljungdahl pathway (Schuchmann and Müller, 2014; Zhang et al., 2020). Sulfate-reducing bacteria like *Desulfovibrio desulfuricans* employ the reductive glycine pathway (Sánchez-Andrea et al., 2020). Additionally, heterotrophic microorganisms have been engineered to enhance $CO_2$ fixation capabilities, leveraging their fast growth and ease of genetic modification (Hu et al., 2022).

### 2.3 Genetic and enzymatic basis

The genetic basis for $CO_2$ fixation involves a variety of genes encoding enzymes that catalyze the key steps in these pathways. For instance, the cbbL genes encode the large subunit of RubisCO in the CBB cycle, which is a critical enzyme for $CO_2$ fixation (Xiao et al., 2020; Asplund-Samuelsson and Hudson, 2021). Other important enzymes include formate dehydrogenase in the reductive glycine pathway and acetyl-CoA synthase in the Wood-Ljungdahl pathway (Sánchez-Andrea et al., 2020; Zhang et al., 2020). Regulatory proteins such as CbbR play a crucial role in controlling the expression of these genes, ensuring the efficient operation of the $CO_2$ fixation pathways (Dangel and Tabita, 2015).

### 2.4 Challenges in $CO_2$ fixation

Despite the potential of microbial $CO_2$ fixation, several challenges remain. One major issue is the efficiency of $CO_2$ fixation, which can be limited by the availability of energy and reducing equivalents (Gong et al., 2019; Hu et al., 2022). Additionally, the integration of $CO_2$ fixation pathways into heterotrophic microorganisms requires careful optimization to balance metabolic fluxes and avoid the accumulation of toxic intermediates (Hu et al., 2022). Environmental factors such as the presence of pollutants can also impact the efficiency of microbial $CO_2$ fixation (Chen et al., 2021). Addressing these challenges through metabolic engineering and synthetic biology approaches is crucial for enhancing the viability of microbial $CO_2$ fixation as a sustainable solution for carbon capture and conversion (Gong et al., 2019; Salehizadeh et al., 2020; Hu et al., 2022).

## 3 Conversion of Fixed $CO_2$ into Organic Acids

### 3.1 Pathways for organic acid production

Microbial fixation of $CO_2$ and its subsequent conversion into organic acids involves several metabolic pathways. Key pathways include the Calvin cycle, the reduced tricarboxylic acid (rTCA) cycle, the Wood-Ljungdahl (WL) pathway, the 3-hydroxypropionate/4-hydroxybutyrate (HP/HB) cycle, the dicarboxylate/4-hydroxybutyrate (DC/HB) cycle, and the 3-hydroxypropionate (3HP) cycle. Additionally, synthetic pathways such as the CETCH cycle, the MOG pathway, the acetyl-CoA bicycle, and the POAP cycle have been designed to enhance $CO_2$ fixation and conversion efficiency (Salehizadeh et al., 2020; Wang et al., 2023).

Acetic Acid: Acetic acid production is prominently facilitated by acetogenic bacteria such as Clostridium scatologenes and Moorella thermoacetica through the Wood-Ljungdahl pathway. These bacteria can convert $CO_2$ into acetic acid under anaerobic conditions, often using $H_2$ as an electron donor (Song et al., 2011; Liu et al., 2018). Microbial electrosynthesis (MES) systems have also been shown to enhance acetic acid production by improving $CO_2$ availability and electron transfer efficiency (Mateos et al., 2019) (Figure 1).

Lactic Acid: Lactic acid production from $CO_2$ is less common but can be achieved through engineered microbial strains. Genetic modifications in lactic acid bacteria can enable the utilization of $CO_2$ as a carbon source, although this area requires further research and development (Salehizadeh et al., 2020).

Succinic Acid: Succinic acid is another valuable product derived from $CO_2$ fixation. The production of succinic acid involves the reductive branch of the TCA cycle. Enhancing $CO_2$ utilization efficiency through genetic and metabolic engineering has been a focus to improve succinic acid yields. Strategies include optimizing $CO_2$ supply methods and employing advanced biotechnological approaches such as micro-nano bubbles and $CO_2$ adsorption materials (Liebal et al., 2018; Chen et al., 2023).

### 3.2 Microbial engineering for enhanced production

Genetic and metabolic engineering play crucial roles in enhancing the microbial conversion of $CO_2$ into organic acids. Key strategies include:

Enzyme Optimization: Improving the efficiency of carbon fixation enzymes such as ribulose-1,5-diphosphate carboxylase/oxygenase (RuBisCO), pyruvate carboxylase, and formate dehydrogenase (FDH) can significantly boost $CO_2$ fixation rates. For instance, the regulation of FDH1 by lysine acetylation and transcriptional factors in Clostridium ljungdahlii has been shown to enhance $CO_2$ metabolism (Zhang et al., 2020).

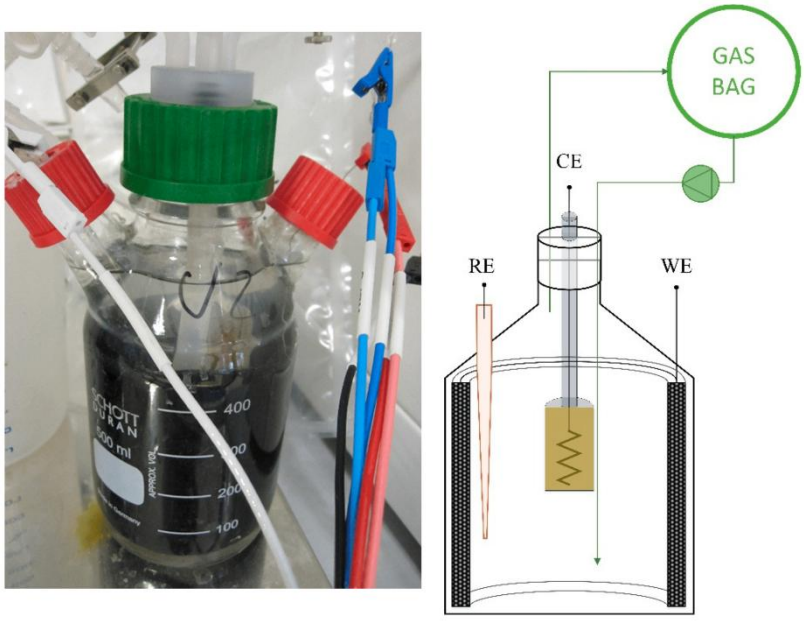


Figure 1 Image and diagram of reactor setup (Adopted from Mateos et al., 2019)
Image caption: CE: counter electrode; RE: reference electrode; WE: working electrode (Adopted from Mateos et al., 2019)

Pathway Engineering: Introducing and optimizing synthetic pathways like the CETCH cycle and the MOG pathway in model organisms such as *Escherichia coli* and yeast can lead to higher yields of target organic acids. These pathways are designed to be more efficient than natural $CO_2$ fixation routes (Wang et al., 2023).

Electrochemical Systems: Utilizing microbial electrosynthesis (MES) systems can improve the conversion efficiency of $CO_2$ to organic acids. By optimizing the bioelectrochemical conditions, such as cathodic potential and electron transfer mechanisms, the production of acetic acid and other organic acids can be significantly enhanced (Liu et al., 2018; Mateos et al., 2019) (Table 1).

Mixed-Culture Biocathodes: Employing mixed microbial cultures in bioelectrochemical systems can stabilize reactor performance and enhance $CO_2$ reduction. For example, a mixed-culture biocathode containing Sporomusa and Clostridium species has been shown to effectively convert $CO_2$ to acetate (Mateos et al., 2019).

By integrating these approaches, the microbial fixation of $CO_2$ and its conversion into valuable organic acids can be optimized, contributing to sustainable and economically viable bioprocesses.

## 4 Process Optimization for Industrial Applications

### 4.1 Fermentation process design

Designing an effective microbial fermentation process for $CO_2$ fixation and organic acid production involves optimizing several key parameters. These include substrate utilization, microbial community enrichment, and operational parameters such as pH, temperature, and partial pressure of gases. For instance, dark fermentation processes have shown that optimizing these parameters can significantly enhance biohydrogen yield and the production of valuable by-products like volatile fatty acids (Ghimire et al., 2015). Additionally, maintaining a sufficient $CO_2$ transfer rate and optimizing pH levels can improve succinate yield in bioreactors (Wu et al., 2012).

### 4.2 Bioreactor technologies

Various bioreactor types and configurations have been developed to optimize microbial $CO_2$ fixation. Continuous stirred-tank reactors (CSTR) and immobilized cell reactors are commonly used, with the latter showing advantages in mass transfer and cell density (Klasson et al., 1991). Hollow fiber membrane bioreactors (L/G MBR) have also been developed to combine biohydrogen production, in situ liquid-gas separation, and bacteria retention, which simplifies the fermentation process and enhances hydrogen yield (Renaudie et al., 2021). Additionally, bioreactors equipped with self-inducing agitators have been shown to reduce $CO_2$ waste and improve succinate production (Wu et al., 2012).

### 4.3 Scale-up challenges

Scaling up microbial $CO_2$ fixation processes from lab to industrial scale presents several challenges. One major issue is the mass transfer limitation due to low gas solubilities, which can hinder the efficiency of the process (Klasson et al., 1991). Additionally, maintaining consistent microbial activity and product yield at larger scales can be difficult. For example, the performance of syngas fermentation processes can be significantly affected by mass transfer rates and gas flow profiles, which need to be carefully managed in large-scale bioreactors (Benalcázar et al., 2020). Furthermore, the integration of gasification and fermentation processes remains an underdeveloped area that requires further research to achieve technological breakthroughs (Pacheco et al., 2023).

Table 1 Results of hydrogen autotrophic fermentation and microbial electrosynthesis experiments (Adopted from Liu et al., 2018)

| | $OD_{600,max}$ | Formate (g/L, max) | Acetate (g/L, max) | Butyrate (g/L, max) | Ethanol (g/L, max) | Rate of acetate (g/L/d) | $H_2$ (%) |
|---|---|---|---|---|---|---|---|
| 80%$H_2$–10%$CO_2$ | 0.120 | 0.023 | 1.250 | 0.320 | 0.192 | 0.170 | 80 |
| 10%$H_2$–10%$CO_2$–80%$N_2$ | 0.110 | 0.008 | 0.362 | 0.145 | 0.083 | 0.050 | 10 |
| − 0.6 V | 0.052 | – | 0.030 | 0.010 | 0 | 0.001 | 0 |
| − 0.8 V | 0.055 | – | 0.095 | 0.051 | 0.010 | 0.012 | 4.797 2 |
| − 1.05 V | 0.059 | – | 0.301 | 0.059 | 0.013 | 0.041 | 9.741 2 |
| − 1.2 V | 0.063 | – | 0.440 | 0.085 | 0.015 | 0.060 | 13.177 7 |

### 4.4 Economic and environmental implications

The economic viability of microbial $CO_2$ fixation processes depends on several factors, including the cost of substrates, bioreactor design, and operational efficiency. Utilizing by-products from fermentation processes, such as volatile fatty acids, can enhance the economic feasibility by providing additional revenue streams (Ghimire et al., 2015). Moreover, integrated systems that combine $CO_2$ fixation with other fermentation processes, such as ethanol production, can reduce costs by utilizing $CO_2$ produced during ethanol fermentation (Wu et al., 2012; Zhang et al., 2017).

Converting $CO_2$ into organic acids offers significant environmental benefits by reducing greenhouse gas emissions and producing valuable bio-based chemicals. For instance, the production of succinic acid from $CO_2$ can replace petroleum-based production methods, contributing to sustainability and reducing the carbon footprint (Ferone et al., 2019). Additionally, integrated fermentation processes that utilize $CO_2$ from ethanol production can further enhance environmental benefits by minimizing $CO_2$ emissions. The use of non-photosynthetic microorganisms for $CO_2$ fixation has also been shown to be more efficient than microalgae-based biofuels, providing a more effective pathway for carbon capture and utilization (Zhang et al., 2017).

## 5 Case Studies

### 5.1 Case study 1: engineering *E. coli* for enhanced succinic acid production from $CO_2$

The engineering of *Escherichia coli* for enhanced succinic acid production from $CO_2$ involves several genetic modifications and process optimizations. One approach includes the heterologous expression of genes from the reductive tricarboxylic acid (rTCA) cycle. Specifically, ten genes encoding key rTCA cycle enzymes such as α-ketoglutarate:ferredoxin oxidoreductase, ATP-dependent citrate lyase, and fumarate reductase/succinate dehydrogenase were cloned into *E. coli*. This transgenic strain demonstrated enhanced growth and the ability to assimilate external inorganic carbon with a gaseous $CO_2$ supply (Lo et al., 2021). Additionally, metabolic engineering strategies have been employed to improve $CO_2$ utilization efficiency, including the use of micro-nano bubbles and $CO_2$ adsorption materials (Chen et al., 2023). Directed evolution of enzymes like propionyl-CoA carboxylase has also been applied to enhance the catalytic efficiency of $CO_2$ fixation pathways (Liu et al., 2020).

The genetically modified *E. coli* strains showed significant improvements in $CO_2$ assimilation and succinic acid production. For instance, the transgenic strain with rTCA cycle genes exhibited $CO_2$-enhanced growth and upregulation of genes involved in chemotaxis, flagellar assembly, and acid-resistance under anaerobic conditions (Lo et al., 2021) (Figure 2). Computational analyses have indicated that with optimized parameters, microbial succinate production processes could reach economically viable levels, making them promising alternatives to traditional sugar-based fermentations (Liebal et al., 2018). These advancements suggest that engineered *E. coli* could play a crucial role in sustainable industrial applications, potentially reducing greenhouse gas emissions and providing a cost-effective method for producing bio-based chemicals.

### 5.2 Case study 2: cyanobacteria as a platform for lactic acid production

Cyanobacteria have been explored as a platform for lactic acid production due to their ability to fix $CO_2$ and convert it into valuable chemicals. Strategies to enhance lactic acid yield include metabolic engineering to increase the flux through the reductive TCA branch and the use of nutritional supplements like corn-steep liquor (CSL) to boost production. For example, the recombinant strain PCCK of Synechocystis sp. PCC6803, which expresses foreign ATP-forming phosphoenolpyruvate carboxykinase (PEPck) along with overexpressed intrinsic phosphoenolpyruvate carboxylase (Ppc), showed increased production of C4 dicarboxylic acids, which can be further converted into lactic acid (Hidese et al., 2022).

The economic viability and scalability of using cyanobacteria for lactic acid production depend on several factors, including the efficiency of $CO_2$ fixation and the overall production costs. High-density cultivation and the use of non-sterile CSL have been shown to significantly enhance the production of malate, fumarate, and succinate, which are precursors for lactic acid production (Hidese et al., 2022). These findings suggest that with further optimization, cyanobacteria could provide a scalable and economically viable platform for lactic acid production, contributing to a sustainable bioeconomy.

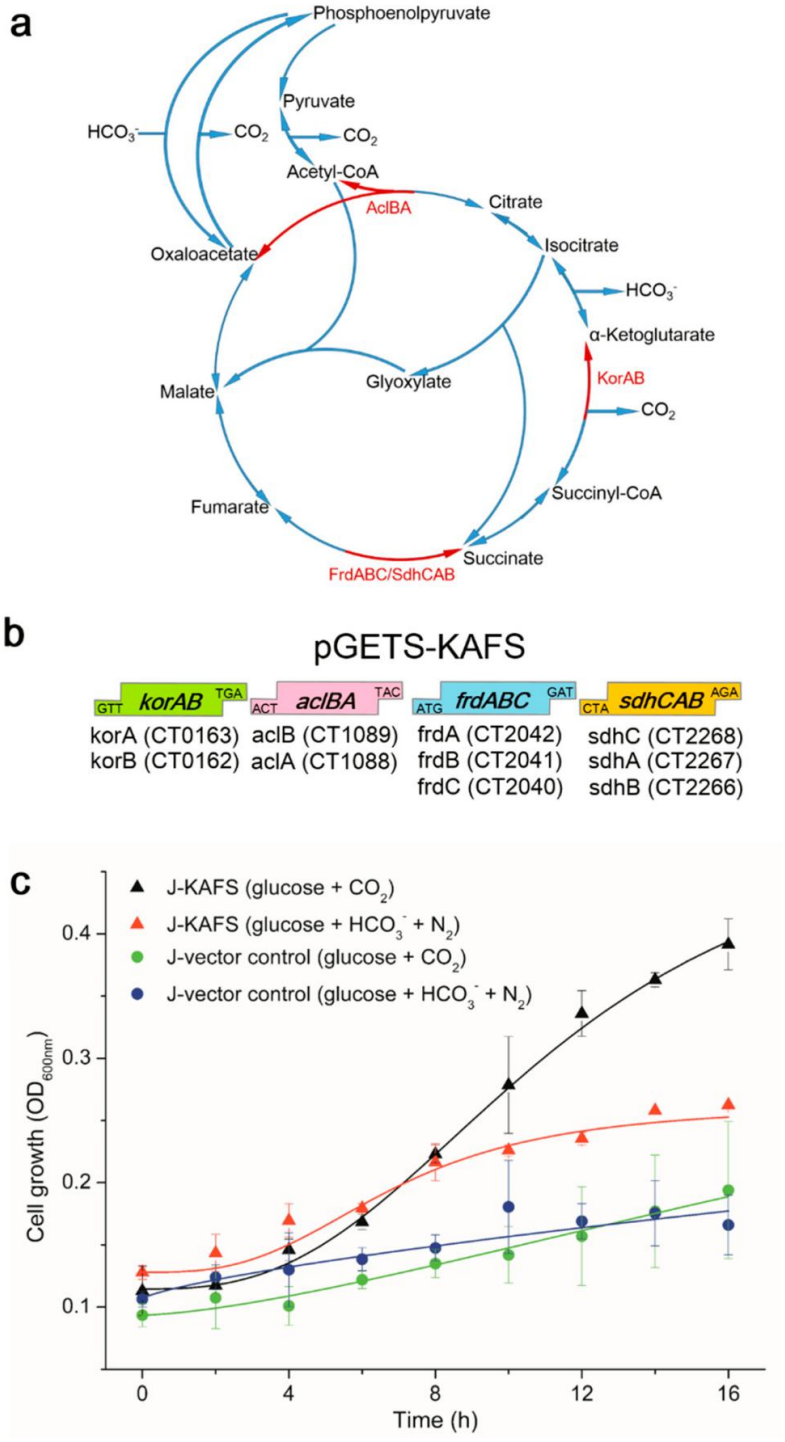


Figure 2 Designed genetic pathway and growth of transgenic *E. coli* strains with different inorganic carbon sources (Adopted from Lo et al., 2021)

Image caption: (a) Pathway map of $CO_2$ assimilation enzymes related to this study. The heterologously expressed enzymes are marked in red. AclBA, ATP-dependent citrate lyase; KorAB, α-ketoglutarate:ferredoxin oxidoreductase; FrdABC/SdhCAB, fumarate reductase. Blue arrows represent TCA cycle activities contributed by *E. coli* genes; arrowheads indicate oxidative (clockwise) or reductive (counter-clockwise) activities; blue/red activity lines are oxidative direction only for *E. coli* enzymes or reversible for cloned *C. tepidum* enzymes. (b) DNA cassettes used to construct the expression plasmid pGETS-KAFS, which encodes the korAB, aclBA, frdABC, and sdhCAB genes from *C. tepidum*. (c) Anaerobic growth curves of transgenic *E. coli* strains with $CO_2$ or bicarbonate present in glucose culture medium. Data are expressed as the means ± standard deviations, n = 3 independent biological samples (Adopted from Lo et al., 2021)

### 5.3 Case study 3: methanogens for acetic acid production

Methanogens have been investigated for their potential to produce acetic acid from $CO_2$ through various metabolic pathways. The exploration of these pathways involves understanding the enzymatic mechanisms and optimizing reactor designs to enhance $CO_2$ fixation and conversion efficiency. For instance, the use of bioreactors with optimized gas and photon transfer rates has been shown to improve the performance of microbial $CO_2$ assimilation processes (Liebal et al., 2018).

The sustainability and cost-effectiveness of using methanogens for acetic acid production are evaluated based on their ability to fix $CO_2$ and the overall production costs (Figure 3). Early assessments using stoichiometric metabolic modeling have indicated that while current microbial processes may not yet be competitive with traditional methods, optimized parameters could make them economically interesting alternatives (Liebal et al., 2018). The development of high-activity enzymes and efficient bioreactor designs are crucial for achieving sustainable and cost-effective acetic acid production from $CO_2$.

By leveraging genetic modifications, process optimizations, and innovative reactor designs, these case studies highlight the potential of microbial $CO_2$ fixation and conversion into valuable organic acids, paving the way for sustainable industrial applications.

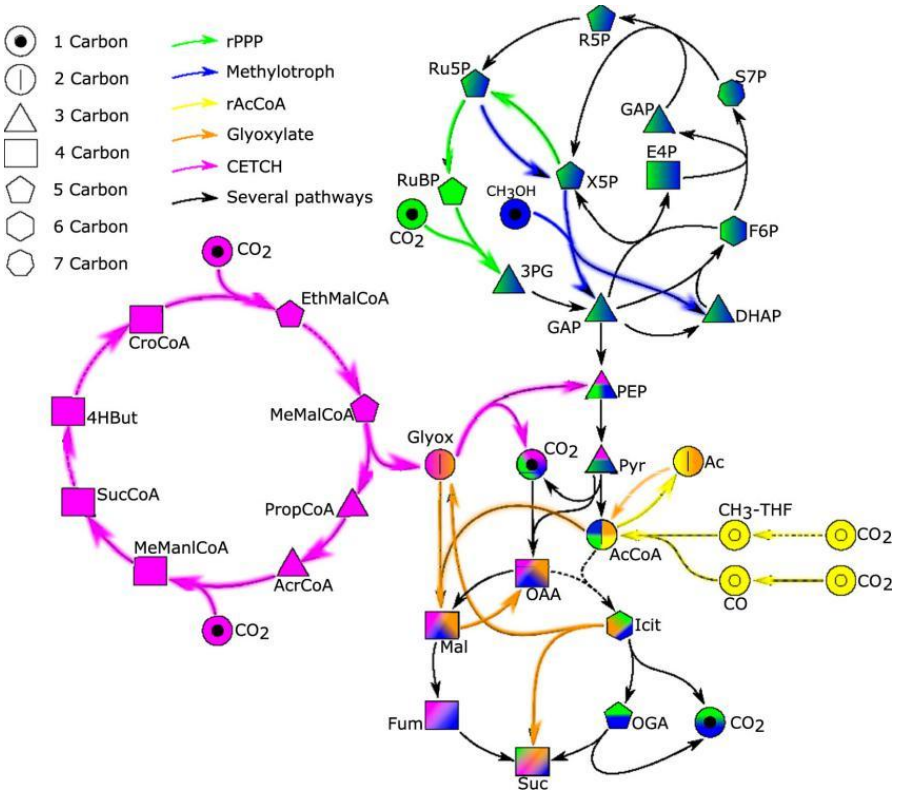


Figure 3 Pathways of carbon fixation to succinate investigated in this study for their economic potential (Adopted from Liebal et al., 2018)

Image caption: The reductive pentose phosphate pathway (green), DHAP pathway of methylotrophic yeasts (blue), reductive acetyl-CoA pathway (C. ljungdahlii, yellow), glyoxylate shunt (*E. coli*, orange), and CETCH pathway (purple). Ac, acetate; AcCoA, acetyl-CoA; AcrCoA, acrylyl-CoA; CH3-THF, methyltetrahydrofolate; CroCoA, crotonyl-CoA; DHAP, dihydroxyacetone phosphate; EthMalCoA, ethylmalonyl-CoA; E4P, erythrose-4-phosphate; Fum, fumarate; F6P, fructose-6-phosphate; GAP, glyceraldehyde-3; Glyox, glyoxylate; Icit, isocitrate; Mal, malate; MeMalCoA, methylmalyl-CoA; MeManlCoA, methylmalonyl-CoA; OAA, oxaloacetate; OGA, 2-oxoglutarate; PEP, phosphoenol pyruvate; PropCoA, propionyl-CoA; Pyr, Pyruvate; RuBP, ribulose-bisphosphate; Ru5P, ribulose-5-phosphat; R5P, ribose-5-phosphate; Suc, succinate; SucCoA, succinyl-CoA; S7P, sedoheptulose-7-phosphate; X5P, xylulose-5-phosphate; 3PG, 3-phosphoglycerate; 4HBut, 4-hydroxyburyrate (Adopted from Liebal et al., 2018)

## 6 Applications and Future Prospects

### 6.1 Industrial applications

Microbially produced organic acids have a wide range of applications across various industries, including pharmaceuticals, food, and biofuels. In the pharmaceutical industry, organic acids such as lactic acid and itaconic acid are used as building blocks for the synthesis of various drugs and medical products (Baumschabl et al., 2022). The food industry utilizes organic acids like citric acid and fumaric acid as preservatives, flavor enhancers, and acidulants, contributing to the taste and shelf-life of food products (Lorenzo et al., 2022). Additionally, the biofuel industry benefits from organic acids such as succinic acid, which can be converted into bio-based chemicals and fuels, providing a sustainable alternative to fossil fuels (Liebal et al., 2018; Reddy et al., 2020). The microbial production of these acids offers a more sustainable and environmentally friendly approach compared to traditional chemical synthesis, which relies on depletable petroleum resources and harsh reaction conditions (Li et al., 2021).

### 6.2 Integration with carbon capture technologies

The integration of microbial $CO_2$ fixation with carbon capture and storage (CCS) systems presents a promising approach to mitigate greenhouse gas emissions while producing valuable organic acids. Microbial $CO_2$ fixation pathways, such as the Calvin cycle and the Wood-Ljungdahl pathway, can be harnessed to convert captured $CO_2$ into organic acids, thus providing a dual benefit of carbon sequestration and production of industrially relevant chemicals (Salehizadeh et al., 2020; Wang et al., 2023). This approach can be further enhanced by coupling microbial and electrochemical methods, which have shown potential in producing carboxylic acids and alcohols from $CO_2$ using reducing power provided by electrodes (Vassilev et al., 2018). By integrating these microbial processes with existing CCS infrastructure, it is possible to create a more sustainable and economically viable system for reducing atmospheric $CO_2$ levels while generating valuable products (Chen et al., 2023).

### 6.3 Future research directions

Future research should focus on several key areas to improve the efficiency of microbial $CO_2$ fixation and expand the range of products that can be synthesized. One critical area is the enhancement of carbon fixation enzymes and metabolic pathways to increase the conversion rates and yields of organic acids (Salehizadeh et al., 2020; Wang et al., 2023). Genetic and metabolic engineering strategies can be employed to optimize microbial strains for higher productivity and broader substrate utilization (Reddy et al., 2020; Li et al., 2021). Additionally, exploring the potential of mixed microbial consortia and co-culturing schemes can lead to the discovery of new microbial interactions and pathways that enhance the production of target compounds (Konstantinidi et al., 2023). Another important research direction is the development of advanced bioreactor designs and process optimization techniques to improve the scalability and economic feasibility of microbial $CO_2$ fixation processes (Liebal et al., 2018; Lin, 2024). By addressing these challenges, it will be possible to create more efficient and versatile microbial cell factories capable of producing a wide range of valuable organic acids from $CO_2$.

## 7 Concluding Remarks

Microbial $CO_2$ fixation and its conversion into organic acids have emerged as promising strategies to mitigate greenhouse gas emissions and produce valuable biochemicals. Various natural and synthetic pathways have been identified for microbial $CO_2$ fixation, including the Calvin cycle, the Wood-Ljungdahl pathway, and the 3-hydroxypropionate/4-hydroxybutyrate cycle, among others. Key enzymes such as ribulose-1,5-bisphosphate carboxylase/oxygenase (RubisCO) and formate dehydrogenase play crucial roles in these processes. Microbial electrosynthesis (MES) has also shown potential in converting $CO_2$ into organic acids like acetic and butyric acid, with Clostridium scatologenes and other acetogenic bacteria demonstrating significant efficiency in these conversions. Genetic and metabolic engineering have further enhanced the efficiency of these microbial processes, making them viable for industrial applications.

The long-term impact of microbial $CO_2$ fixation technologies could be substantial in reducing global $CO_2$ levels. By leveraging the natural ability of microorganisms to assimilate $CO_2$ and convert it into valuable organic compounds, these technologies offer a sustainable and eco-friendly alternative to traditional $CO_2$ capture methods, which are often cost-inefficient and environmentally hazardous. The integration of microbial $CO_2$ fixation with

bioelectrochemical systems (BESs) and MES can further enhance the efficiency and scalability of these processes, potentially leading to significant reductions in atmospheric $CO_2$ levels. Additionally, the development of robust microbial strains through genetic and metabolic engineering could optimize $CO_2$ fixation rates and product yields, making these technologies economically viable for large-scale deployment.

The future of microbial processes in sustainable industrial practices looks promising, with microbial $CO_2$ fixation and conversion technologies poised to play a critical role in addressing climate change and promoting green manufacturing. The continuous advancements in understanding microbial metabolic pathways, coupled with innovations in genetic engineering and bioelectrochemical systems, are likely to drive the development of more efficient and cost-effective $CO_2$ fixation processes. As these technologies mature, they could be integrated into various industrial applications, from biofuel production to the synthesis of high-value chemicals, thereby contributing to a circular carbon economy and reducing our reliance on fossil fuels. The collaborative efforts of researchers, industry stakeholders, and policymakers will be essential in realizing the full potential of microbial $CO_2$ fixation technologies and ensuring their successful implementation in sustainable industrial practices.

## Acknowledgments

Sincere thanks to the peer reviewers for their detailed reviews and valuable guidance on this manuscript.

## Conflict of Interest Disclosure

The author affirms that this research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

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