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Comparative Study of CO_2 Dissolution in Aqueous Media for Applications in Bioelectrochemical Systems

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ABSTRACT

In alignment with the Sustainable Development Goals (SDG 13: Climate Action and SDG 7: Affordable and Clean Energy), the urgent need to mitigate the high atmospheric level of carbon dioxide (CO_2) has lead research toward sustainable carbon capture and utilization technologies. Bioelectrochemical systems (BESs), especially microbial electrosynthesis (MES), have emerged as promising technology for converting CO_2 into value-added products with the assistance of the electroactive microbial metabolisms. However, the efficiency of MES remains constrained by the inherently low solubility and limited mass transfer of CO_2 in aqueous systems, which minimizes the availability of CO_2 for microbial uptake and conversion. This study investigates the CO_2 dissolution behavior in various aqueous media—deionized (DI) water, reverse osmosis (RO) water, tap water, phosphate-buffered saline (PBS), PBS with anode additives, and microbial growth media including acetogen and sulfate-reducing bacteria formulations. Carbon dioxide (CO_2) dissolution was observed through changes of pH and conductivity to examine dissolution efficiency and media stability. Based on results achieved, this study highlights DI and RO water enhances CO_2 uptake but lack of buffering leads to pH stability, while PBS-based medium not only offers superior pH stability but also effectively support CO_2 dissolution, making them more suitable for long-term operation in microbial electrosynthesis systems. These findings provide crucial insights for selecting optimal aqueous environments to enhance CO_2 dissolution and support the design of more robust, efficient MES systems for future carbon-neutral technologies.

1. INTRODUCTION

Rising levels of atmospheric carbon dioxide (CO_2) have become a major threat to global climate stability. The burning of fossil fuels, deforestation, and industrial activities have driven CO_2 levels to record highs, leading to global warming, extreme weather events, rising sea levels, and ecosystem disruptions [1]. According to the NOAA's Global Monitoring Lab, CO_2 levels had reached a record of 419.3 ppm, with the largest one-year increase on record (3.75 ppm) [2]. In response to this alarming trend, there is a high demand for advanced carbon capture, utilization and storage (CCUS) technologies aimed to reduce emissions while promoting sustainable resource and recovery. Among these, microbial electrosynthesis (MES) a subfield of bioelectrochemical systems (BESs) has gained significant attention for their potential to convert CO2 into valuable-added products through the metabolic activity of electrogenic microorganisms [3], [4], offering a sustainable pathway for renewable chemical production and contributing to circular carbon economy initiatives. Despite that, one of the major limitations in MES systems is the restricted availability of dissolved CO_2 due to its inherently low diffusion and solubility rate in aqueous media [5], [6]. Addressing this problem is important for scaling MES from the lab to industry because better CO_2 dissolution can enhance microbial productivity, improve energy efficiency, and

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increase product yields, making the process more cost-effective and environmentally friendly.

Efficient microbial conversion of CO_2 is highly dependent on a consistent supply of its dissolved form and effective mass transfer to support microbial metabolism and electron transfer leading to a better system productivity [7]. While several operational parameters such as pressure, temperature, inflow rate, and agitation influence the dissolution rate, improving CO_2 inflow alone offers limited advantage beyond a certain point due to gas loss through outgassing or bubble formation. Additionally, the physicochemical characteristics of the media including the buffering capacity, ionic strength and presence of organic compounds play pivotal role in determining both the rate of rate and stability of CO_2 dissolution. Media with strong buffering systems may resist pH shifts but can also influence the types of carbonates formed, which alters how much dissolved carbon is available for microbes. On the other hand, low-ion media can promote rapid CO_2 absorption but often could not sustain for long-term.Therefore, it is crucial enough to optimize the CO_2 dissolution process for a better application in MES.

Gas diffusion electrodes (GDEs) present a practical strategy for overcoming CO_2 mass transfer limitations in aqueous systems. By creating a gas-liquid interface, GDEs facilitate more effective CO_2 transfer from the gas phase into the liquid medium, thus increasing its bioavailability for microbial reduction reactions. This improvised transfer is relevant for cathodic processes in MES where dissolved as the main carbon source for the microbes. However, the performance of GDEs is mainly affected by the physicochemical properties of the liquid medium including ionic strength, pH buffering and chemical reactivity. Although various studies have explored on the microbial aspects of MES, a detailed understanding on the interaction of CO_2 with various liquid media, particularly those relevant to microbial electrosynthesis (MES) environments remains limited. The dissolution behaviour of CO_2 is not uniform and can be significantly influenced by several factors including buffering capacity, ionic strength and presence of organic compounds in the medium. These parameters affect both the initial rate of CO_2 dissolution and its long-term stability in the solution. Hence, a comprehensive study on how different types of solution such as deionized (DI) water, tap water or microorganism media influence CO_2 solubility is required for the optimization on the design and operation of gas diffusion electrode (GDE)-based MES systems.

Therefore, a study on the CO2 dissolution behaviour across commonly used aqueous systems in MES is very essential. Deionized water (DI), reverse osmosis (RO) water, tap water, phosphate-buffered saline (PBS), and microbial culture media (e.g., acetogen medium and sulfate-reducing bacterial medium) each represent different chemical environments relevant to real-world MES operations. This study hence focuses on investigation on the rate of CO_2 dissolution in different aqueous solutions, tap water, deionized (DI) water, reverse osmosis (RO) water, phosphate-buffered saline (PBS) and microbial medium such as Acetogen medium through pH and conductivity profiling over a 72-hour period. The main aim is to characterize on the rate of CO_2 dissolution in the respective medias and these results also help understanding how medium composition affects CO_2 mass transfer. Moreover, as a preliminary study, it offers fundamental understanding of the behaviour of CO_2 mass transfer and helps in determining which aqueous environment promote more stable and effective CO_2 dissolution. By identifying optimal media for CO_2 dissolution and pH control, this study supports the development of more efficient and stable microbial electrochemical systems capable of transforming CO_2 into other products. Further research is still required to fully understand long-term retention, microbial compatibility, and real-world application. These results serve as a foundation for future work in optimizing GDE-based systems for sustainable CO_2 capture and conversion technologies.

2. METHODOLOGY

2.1 Medium Preparation

First, Following are the samples that are prepared for the CO_2 dissolution study: deionized water (DI), reverse osmosis water (RO), tap water, phosphate-buffered saline (PBS), PBS supplemented with anode additives (ammonium chloride and sodium acetate), Acetogen medium, and Postgate's Medium B for Sulfate-Reducing Bacteria (SRB)in accordance in a previous study [8]. These media were selected to represent a broad range of aqueous environments, varying in ionic strength, buffering capacity, and organic content to study their influence on CO_2 solubility and stability.

The PBS solution (100 mL) was prepared by dissolving 0.53518 g of disodium phosphate (Na_2HPO_4) and 0.16967 g of monosodium phosphate (NaH_2PO_4) in deionized water with continuous stirring to ensure homogeneity. For the PBS with the anode additive elements, 0.5349 g of ammonium chloride (NH_4Cl) and 0.13608 g of sodium acetate (NaAc) were added to the PBS solution prior to dissolution. Acetogen and SRB media were prepared according to their specifications as shown in Table 1 and distributed into serum bottles using the same procedure.

Each medium sample (20 mL) was transferred into a 100 mL labelled serum bottle by using aseptic techniques. For each sample type, three serum bottles were prepared to represent sampling points at 24, 48, and 72 hours. Bottles were sealed using aluminium crimp caps to ensure an airtight environment throughout the experiment. All sealed serum bottles were then placed in a water displacement setup (Figure 1) submerged in a container filled with 100 mL of hydrochloric acid (HCl) solution adjusted to pH 2 to maintain an acidic environment throughout the experiment. Subsequently, each serum bottle was purged with 100% CO_2 gas for 10 minutes using a stainless-steel needle connected to a pressurized tank to saturate the liquid phase and ensure uniform CO_2 dissolution. After purging, all bottles were place at the room temperature (approximately 27°C) in a static condition without agitation. This setup enabled the study of CO_2 dissolution and retention purely under the influence of chemical interactions between CO_2 and the aqueous medium, without external mixing.

2.2 Sample Collection and Analysis

Following each incubation period (24, 48, and 72 hours), liquid samples from each serum bottle were transferred into clean vials using sterile syringes and then the pH and conductivity were measured immediately. The pH value of each sample was measured using a calibrated pH meter, ensuring that the probe was rinsed with deionized water between measurements to prevent any contamination. Each measurement was recorded in triplicates to ensure accuracy.

Conductivity was measured with a conductivity meter following the same procedure. All readings were taken within 1-2 minutes of samples withdrawal to minimize changes that might occur due to gas escape.

Table 1. Composition and Preparation of *Acetogen* and *Sulfate Reducing Bacteria* Enrichment Media.

***Acetogen* medium**		**Postgate's Medium B for Sulfate Reducers**	
Chemicals	**Weight**	**Chemicals**	**Weight**
$NaHCO_3$	2.4g	$MgSO_4 \cdot 7H_2O$	2.0g
NH_4Cl	0.2g	NH_4Cl	1.0g
Yeast extract	0.2g	$CaSO_4$	1.0g
Stock salts solution #1	40.0mL	Yeast extract	1.0g
Potassium phosphate buffer	20.0mL	KH_2PO_4	0.5g
Clarified rumen fluid	20.0mL	$FeSO_4 \cdot 7H_2O$	0.5g
Stock salts solution #2	4.0mL	Ascorbic acid	0.1g
Trace minerals solution	4.0mL	Thioglycollic acid	0.1g
Vitamin solution	4.0mL		
Reducing agent solution	4.0mL		
Tungstate solution	0.4mL	—	
Resazurin (0.1% solution)	0.4mL		
Preparation of Medium: Add components, except NaHCO3 and reducing agent, to distilled/deionized water and bring volume to 417.8mL. Mix thoroughly. Gently heat and bring to boiling under 80% N2 + 20% CO2. Cool to 45°–50°C. Add NaHCO3 and reducing agent. Distribute into tubes or flasks under 80% N2 + 20% CO2. Autoclave for 15 min at 15 psi pressure–121°C. After inoculation, exchange headspace with 80% H2 + 20% CO2.		**Preparation of Medium:** Add components, except ascorbic acid and thioglycollic acid, to tap water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.0–7.5. Thioglycolate and ascorbate should be added immediately prior to sterilization. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.	

Fig. 1. Experimental setup for CO_2 dissolution by a water displacement method

3. Results and Discussion

In this study, due to the CO_2 dissolving from the atmosphere, the pH of all the samples all showed clear patterns of decreasing over time. Based on Figure 2(a), initially, the pH of tap water was 7.55 then decreased in a slow rate up to 24 hours then continued to decrease to 5.25. Although tap water consists of dissolved minerals which may act as buffer for the water, the constant absorption of CO_2 eventually reduces the pH. This gradual trend highlights the role of bicarbonate-carbonate equilibrium in natural water systems, where the earlier buffering capacity leads to the delaying in the pH changes. In practical systems, such buffering can aid maintaining microbial performance [10]. Starting at 7.33, the pH of Deionized (DI) water dropped to 5.63 after 72 hours. Since DI water lacks any dissolved ions and has no buffering capacity, it is extremely sensitive to CO_2 absorption. As CO_2 dissolves in the ion-free water, carbonic acid is immediately formed, causing a rapid decrease in the pH compared to tap water. This steep pH drop in DI water is likely due to its ion-free nature, which results in near-linear acidification rates in response to gas absorption, confirming its suitability for rapid saturation tests but not for systems requiring long-term chemical stability. Likewise, Reverse Osmosis (RO) water's pH started at 6.07 and gradually decreased to 4.21 after 48 hours. RO water similarly like DI water also has few ions due to its filtering process, making it more prone to CO_2 absorption. The pH of RO water drops dramatically due to a lack of buffering chemicals, which is the same trend observed in DI water [11], [12]. However, there is a slight increase in the pH was observed in the RO water after 48 hours, suggesting that experimental errors or sample contamination may have occurred. Possible causes may include either minor gas loss during sample handling or accidental entry of other substances, both of which could shift the carbonate equilibrium and result in a slight rise in the pH. Such anomalies highlight the significance of maintaining strict control over sampling and storage conditions when measuring CO_2 dissolution effects.

These divergent pH trajectories across tap water, DI water, and RO water collectively illustrate how initial ionic composition governs both the rate and the extent of CO_2-driven acidification. Because DI and RO water contain negligible dissolved solids, their carbonate–bicarbonate equilibria are established almost entirely by the CO_2 introduced during the experiment, making their pH highly responsive to even small changes in dissolved CO_2 concentration. Tap water, in contrast, already contains a baseline of dissolved minerals that partially buffers this response, delaying the onset of a measurable pH drop even though the medium eventually acidifies once its limited buffering capacity is exceeded. This distinction is particularly relevant when selecting a medium for rapid screening of CO_2 dissolution behaviour, since low-ion waters offer a more sensitive and immediate readout of gas uptake than naturally buffered water sources.

On the other hand, both PBS and PBS + anode solutions showed relatively stable pH values across the 72-hour period with minor decreases observed. This outcome is attributed to the strong buffer system as the phosphate buffer neutralizes the H^+ ions formed during CO_2 dissolution, helping to maintain a stable pH. The SRB medium exhibited a more rapid drop in pH from 8.74 to 5.78, compared to the acetogen medium, where pH decline was from 8.27 to 6.45, indicating a faster acidification response. This sharper pH change in SRB medium may be related to its composition, which, despite consisting of buffering agents, also includes components such as $FeSO_4$ and $CaSO_4$ that might be possibly interacting with dissolved CO_2 to form secondary products, thereby causing a change in the

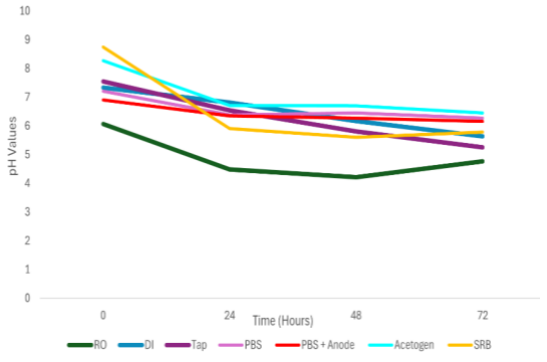
carbonate–bicarbonate balance. The acetogen medium, with different nutrient and salt profiles, likely provided a slightly stronger buffering action, moderating pH decline over the same time. Overall, these results highlight the distinct behaviours of each medium in response to CO_2 dissolution. Among all the samples, PBS and PBS + anode solutions demonstrated better pH stability, making them ideal for systems where pH maintenance is important like in controlled bioelectrochemical environments. The pattern observed SRB and acetogen media highlight their sensitivity to CO_2, which could be beneficial in applications aiming to accelerate microbial activity. For example, a quicker pH shift can help initiating certain metabolic pathways, such as those in acetogens that operate optimally under slightly acidic conditions, whereas in other cases, rapid acidification could disrupt microbial growth requiring a balance between dissolution rate and pH control in microbial electrosynthesis (MES) systems [13].

This observation correlates with findings reported in a three-chamber microbial electrolysis cell (3cMEC), where CO_2 dissolution from the gas chamber adjacent to the cathode mitigated catholyte pH rise. The system exhibited a slower pH rise compared to a conventional two-chamber MEC, stabilizing at pH 8.50 instead of exceeding 9.75 microbial activity, retention behaviour, and operational scalability under continuous CO_2 supply [8]. This buffering affect is attributed to the formation of H^+ and HCO_3^- species during CO_2 dissolution, a buffering mechanism which resembles behaviours observed in PBS-based media in this study. Notably, while the present study involved static conditions without any applied potential, a normal MEC operated under voltage-driven electrochemical reactions that continuously generated hydroxide ions at the cathode. The fact that CO_2 diffusion still moderated pH under those dynamic conditions reinforces the applicability of our static findings to operational systems, specifically in showing how gas dissolution can lead to rising of pH value. While the overall pattern is comparable, the difference in operational context should be noted as this study involved a static, batch-based setup, whereas the 3cMEC involved a dynamic, voltage-driven system with continuous gas supply. Thus, the resemblance in pH stabilization behaviour suggests that well-buffered media can replicate some of the pH-regulating advantages observed in more advanced system configurations.

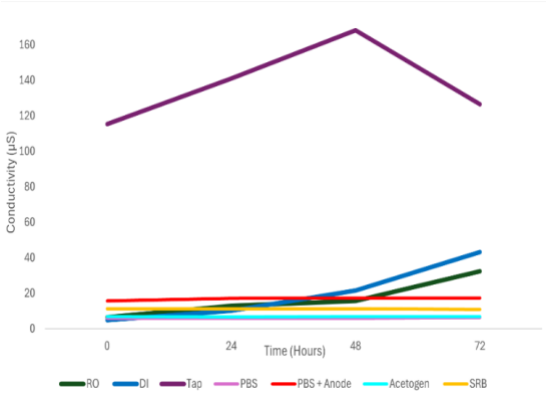
Beyond the immediate implications for pH stability, these findings carry practical significance for the design of gas diffusion electrode (GDE)-based MES reactors. In systems where the cathode operates in close contact with the liquid medium, a rapidly acidifying environment such as DI or RO water could locally lower pH beyond the tolerance range of electroactive microorganisms before dissolved CO_2 becomes fully available for reduction, whereas a well-buffered medium such as PBS would sustain a more consistent pH microenvironment around the electrode surface. This trade-off between rapid CO_2 uptake and pH stability suggests that medium selection in MES should be guided not only by the total quantity of CO_2 dissolved but also by the rate at which that dissolution proceeds relative to the buffering capacity of the surrounding solution. For applications where fast initial saturation is prioritised, such as short-duration batch tests, low-ion media may be advantageous; for continuous, long-duration operation where microbial viability must be preserved, buffered media are likely to offer superior operational resilience.

As shown in Figure 2(b), tap water consistently exhibited highest conductivity, with its value remaining above 110 µS over the 72-hour period. This result is expected due to its initial ionic composition, as tap water naturally consists of a variety dissolved mineral. In contrast, both DI and RO water showed a noticeable increase in conductivity over time. RO water started below 10 µS and increased slightly, while DI water resulted in more obvious rise in conductivity reaching 45 µS. These increases show that as CO_2 dissolved and dissociated in solution, ionic species including hydrogen and bicarbonate ions formed. This increase in conductivity shows that CO_2 is dissolving in low-ion media and suggests that conductivity measurements could be used to monitor gas transfer efficiency in real time without disrupting the system. For PBS and PBS + anode solutions maintain stable conductivity profiles. The PBS + anode medium consistently showed higher conductivity than PBS alone, possibly due to the extra elements (ammonium chloride and sodium acetate) added that contributes as well to the ionic compound. However, both remained similar, suggesting strong ionic stability provided by the phosphate buffer system. This ionic stability is critical for bioelectrochemical applications, because sudden conductivity changes can have an impact on the current distribution and electrode performance. By maintaining stable ionic conditions, PBS-based solutions can help ensure steady-state operation in systems such as MECs. Acetogen and SRB media displayed similarly steady conductivity values, further confirming that buffered and nutrient-rich media can maintain ionic balance despite exposure to CO_2. A comparable trend was also observed in the 3cMEC study, where conductivity in the catholyte increased proportionally with the applied voltage as CO_2 diffused from the gas chamber and accumulated as carbonate species [8]. While the rate of change was larger in the 3cMEC due to continuous electrochemical activity, the basic principle is on that CO_2 dissolution contributes to ionic enrichment remains consistent between the two conditions. In both scenarios, carbonate formation plays a key role, whether occurring under static saturation or in dynamic, voltage-driven processes. Although not directly equivalent, the similarity in outcomes supports the broader conclusion that controlled CO_2 delivery and proper buffering, whether through media formulation or system design can be able to enhance chemical stability in bioelectrochemical processes.

Taken together, the pH and conductivity profiles indicate that ionic buffering and dissolution kinetics are closely interlinked properties of the aqueous medium, rather than independent variables. Media that resisted large pH shifts also tended to display more stable conductivity, consistent with the interpretation that the phosphate buffer system simultaneously moderates proton activity and maintains a relatively constant ionic background even as CO_2 continues to dissolve and dissociate into carbonate species. This coupling has a direct bearing on electrochemical performance in MES, since fluctuations in either parameter can alter electrode overpotential, ohmic resistance, and the local microenvironment available to attached biofilms. From a system design perspective, these observations reinforce the value of pairing GDE-based CO_2 delivery with a buffered catholyte formulation, particularly for longer operational periods where cumulative CO_2 loading would otherwise be expected to drive larger swings in both pH and conductivity in poorly buffered media.



(a)



(b)

Fig. 2. pH (a) and conductivity (b) variations of the different aqueous media over 72 hours period.

4. CONCLUSION

This study highlights the importance of selecting appropriate aqueous media for optimizing CO_2 dissolution behavior in bioelectrochemical applications. From a practical perspective, the selection of medium is not only significant of achieving high initial CO2 dissolution rates but also making sure of the chemical stability throughout prolonged operation. Deionized (DI) and reverse osmosis (RO) water, with their low ionic content, are preferable for rapid and high CO_2 absorption, supporting efficient gas–liquid transfer. However, for systems requiring long-term chemical stability, phosphate-buffered saline (PBS) and PBS with anode additives are more suitable due to their strong buffering capacity, which effectively minimizes pH fluctuations during CO_2 exposure. Thus, they are appropriate because able to extend operational stability and reduce the need for frequent pH control interventions. The conductivity results further support this finding, with buffered solutions resisting major ionic shifts even under CO_2 exposure, a desirable behavior for maintaining consistent electrochemical conditions.

These findings also point to practical criteria that can guide medium selection at the design stage of a MES reactor, rather than being determined empirically after construction. For instance, applications prioritising fast CO_2 loading during start-up could employ a low-ion medium such as DI or RO water for an initial saturation phase before transitioning to a buffered operating medium once steady-state conditions are required. Such a staged approach would combine the rapid gas-liquid transfer advantages of ion-free media with the long-term pH and conductivity stability offered by phosphate-buffered or nutrient-rich formulations, potentially shortening start-up time without compromising the operational resilience of the system.

The findings also align with trends observed in three-chamber MEC systems, reinforcing the importance of combining buffered media with controlled CO_2 supply for enhanced system performance. While the results provide foundational insights into medium behavior under static conditions, future work should address microbial activity, retention behavior, and scalability under continuous CO_2 input and real-time electrochemical operation.

Moving forward, integrating these findings into scaled, real-time CO_2-fed MESCs will be essential to evaluate performance under actual operating pressures. Future studies should also examine the interaction between CO_2 dissolution, microbial metabolism, and electrode performance, including maximizing gas uptake and maintaining favorable biochemical conditions. Additionally, exploring alternative buffering systems could further optimize the balance between rapid CO_2 dissolution and pH stability. Ultimately, this work provides an experimental foundation for electrolyte selection in CO_2-utilizing bioelectrochemical systems, contributing to the broader goal of enhancing carbon capture, conversion efficiency, and operational durability in sustainable energy and resource recovery applications.

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