

Fluid-to-Fluid Interaction of Rhamnolipid Biosurfactants with Divalent Ions: Investigation of Interfacial Tension and Emulsion Viscosity

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ABSTRACT - Rhamnolipid biosurfactants derived from microbial sources have gained substantial interest as environmentally sustainable alternatives to synthetic surfactants, particularly in the realm of Microbial Enhanced Oil Recovery (MEOR). Their biodegradability, low toxicity, and effectiveness under extreme conditions make them ideal candidates for improving oil displacement in reservoirs. However, the presence of divalent ions, specifically calcium (Ca^{2+}) and magnesium (Mg^{2+}), which are abundant in reservoir brine, can significantly affect the performance of these biosurfactants. This research investigates the influence of Ca^{2+} and Mg^{2+} ions on the phase behavior, stability, and interfacial properties of rhamnolipid-based microemulsion systems, which play a critical role in MEOR processes. A series of experiments was conducted to analyze the impact of varying concentrations of Ca^{2+} and Mg^{2+} ions on rhamnolipid microemulsions. The study evaluates phase transitions, stability, and microstructural characteristics of emulsions using a spinning drop tensiometer to measure interfacial tension (IFT) and rheological analysis to determine viscosity. The results demonstrate that both Ca^{2+} and Mg^{2+} ions influence the optimum salinity conditions required for microemulsion stability, with their presence causing shifts in the phase boundaries. Notably, Ca^{2+} ions exert a more pronounced effect on phase stability than Mg^{2+} , leading to increased IFT and viscosity at higher concentrations. These findings further elucidate the crucial role of divalent ions in governing the stability and functionality of biosurfactant systems under reservoir conditions and highlight the importance of controlling ion concentrations to achieve efficient MEOR applications. Overall, this research provides valuable insight for optimizing the formulation of rhamnolipid-based systems to enhance oil recovery performance while mitigating the adverse effects of high divalent ion content in brine. The research contributes to ongoing efforts to improve biosurfactant efficacy, offering a pathway toward refining MEOR strategies and advancing sustainable oil recovery technologies.

Keywords: rhamnolipid, MEOR, divalent ions, interfacial tension, microemulsion viscosity.

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INTRODUCTION

Microbial Enhanced Oil Recovery involves the use of microorganisms in petroleum reservoirs to increase the amount of recoverable oil (Ayirala et al., 2021; Kadarwati et al., 2002; Kusuryani, 2016; Nanda & Kussuryani 2013; Sheng 2013). These microorganisms utilize hydrocarbons as an energy source and produce metabolites such as biosurfactants and biopolymers (Kadarwati, 2018; Marhaendrajana et al., 2025; Nanda & Kussuryani 2013; Rita et al., 2025). Among these products, biosurfactants especially rhamnolipids, have attracted considerable attention due to their low toxicity, high biodegradability, and ability to remain effective under extreme temperature, pH, and salinity conditions (Awaludin & Sari 2017; Daulay et al., 2025; Esfandyari et al., 2021; Ganji et al., 2020; Hou et al., 2021).

Rhamnolipids are primarily produced by *Pseudomonas aeruginosa* and consists of one or two rhamnose sugars moieties linked to one or two β -hydroxy decanoic acid chains (Awaludin & Sari 2017). This molecular structure enables rhamnolipids to significantly reduce the interfacial tension (IFT) between oil and water, thereby altering rock wettability. Laboratory results have indicated that rhamnolipids can reduce IFT to approximately 10^{-2} mN/m. Furthermore, wettability tests conducted on sandstone substrates indicate that rhamnolipids shift the rock surface toward a more water-wet condition, with contact angles decreasing to below 75° (Akanji et al., 2021). Their amphiphilic nature of rhamnolipids

allows them interact effectively with both oil and rock surfaces. The hydrophilic groups, such as carbonyl and hydroxyl moieties, facilitate cations exchange, while the hydrophobic tails associate with hydrocarbon molecules. These interactions enhance the oil mobility within the reservoir by weakening the adhesion forces among oil, water, and rock (Megayanti et al., 2023; Udoh & Vinogradov 2019).

One of the primary challenges associated with synthetic surfactant in enhanced oil recovery is their tendency to adsorb onto rock surfaces. Surfactant adsorption is governed by several mechanisms including ion exchange, hydrophobic forces, electrostatic interactions, dispersion, and van der Waals forces (Hao et al., 2019; Mohammed & Babadagli, 2015). The application of biosurfactants, particularly rhamnolipids, can overcome these problems (Gazem et al., 2025; Sulistyarso et al., 2025; Tamil et al., 2020).

The combined use of rhamnolipids with divalent ions such as Ca^{2+} and Mg^{2+} has been reported to enhance emulsion stability by strengthening the interfacial film at the oil-water interface. This effect leads to increased emulsion viscosity and improved sweep efficiency during displacement process (Gandomkar & Rahimpour, 2017; Jha et al., 2018). In addition, these divalent ions could improve the performance of biosurfactants by promoting stronger hydrophobic interactions, hence improving their ability to reduce IFT (Chen & Lee 2022).

However, research that comprehensively investigates both the interfacial properties and rheological behavior of rhamnolipid emulsions under varying divalent ion conditions remain limited. Prior research has primarily focused on IFT measurements; while the combined evaluation of IFT and emulsion viscosity under reservoir-representative brine conditions remains underexplored.

This research introduces a fluid-to-fluid experimental framework to evaluate the interaction between rhamnolipid biosurfactants and divalent ions (Ca^{2+} and Mg^{2+}) in the presence of crude oil. The suggested methodology evaluates both interfacial tension and emulsion viscosity at the same time to find out how ion-specific interactions affect the effectiveness of biosurfactants. This integrated approach provides new insights into the dual functionality of rhamnolipids – namely their ability to reduce interfacial tension while also modifying emulsion rheology – which has not been comprehensively assessed in previous studies. Furthermore, this research offers practical guidance for evaluating biosurfactant compatibility prior to a large scale field application. This aspect is particularly relevant for Indonesian oilfields, where hard brines with high concentrations of Ca^{2+} and Mg^{2+} , coupled with elevated with high salinity and

various pH levels, can significantly affect biosurfactant stability, adsorption behavior, and interfacial performance.

METHODOLOGY

All laboratory experiments in this study are conducted at the enhanced oil recovery enhanced oil recovery (EOR) Laboratory, Department of Petroleum Engineering, UPN Veteran Yogyakarta, Indonesia.

Material and sample preparation

Crude oil sample

The crude oil samples used in this study were collected from two oil fields in Indonesia. The light oil properties had an API gravity of 43.45° , while the medium oil properties had an API gravity of 33.1° .

Rhamnolipid solution

This study employed a rhamnolipid biosurfactant purchased from Shanghai Yuchuang Chemical Technology Co., Ltd. The product appears as a light yellow to brown liquid, as shown in Figure 1 and is readily soluble in water. The detailed specifications of the rhamnolipid are provided in Table 1. Rhamnolipids consist of hydrophilic rhamnose sugar moieties covalently bonded to



Figure 1. Rhamnolipids solution samples at different concentrations prepared prior to IFT and viscosity measurements

hydrophobic fatty acid chains. Rhamnose is a deoxy sugar that typically occurs as one or two rhamnose units linked to one or two β -hydroxy fatty acid chains, forming mono-rhamnolipids or di-rhamnolipids (Xiao et al., 2020).

Table 1. Rhamnolipids specifications

Items	Specifications
Solid Content	298.76 %
pH	7.67
Solubility (Water)	Soluble

Experimental design

Fluid-to-fluid testing was conducted using synthetic brines with various compositions, as summarized in Table 2. A total of 90 experimental scenarios were designed, incorporating two of crude oil samples and rhamnolipid concentrations, 0 – 2 wt%.

Experimental procedure

Interfacial tension (IFT) test

The IFT was measured with a spinning-drop tensiometer (Figure 2) to evaluate the rhamnolipid efficiency in reducing the IFT

Table 2. Synthetic brine solutions in this study

Number	Synthetic Brine		
	NaCl (ppm)	CaCl ₂ (ppm)	MgCl ₂ (ppm)
1		-	-
2	5,000	2,000	-
3		-	2,000
4	10,000	-	-
5	10,000	2,000	-
6	10,000	-	2,000
7	15,000	-	-
8	15,000	2,000	-
9	15,000	-	2,000

between oil and brine. The test was performed for 30 minutes at a rotation speed of 6000 RPM and a controlled temperature of 60 ± 0.5 °C (representing the reservoir condition).

Viscosity measurement

Viscometer measurements were performed using a Brookfield viscometer, which determines viscosity by measuring the torsional resistance acting on a cylindrical spindle immersed in the fluid (Figure 3). In this study, the instrument was used to assess the viscosity of an oil-water emulsion. The



Figure 2. Spinning drop interfacial tensiometer used for oil-brine IFT measurements, with ± 3 RPM control accuracy



Figure 3. Brookfield DV3T rotational viscometer used to measure emulsion viscosity with instrument speed range is 0.01–250RPM

emulsion was prepared by mixing oil and rhamnolipid solution in a 1:1 ratio (5 mL of each component). Prior to measurement, the mixture was stirred for 10 minutes at reservoir temperature ($60 \pm 0.5^\circ\text{C}$) to ensure emulsion formation.

RESULT AND DISCUSSION

Interfacial tension

Interfacial tension (IFT) plays a crucial role in oil recovery, as it governs the capillary forces acting between oil and water phases. A reduction in IFT facilitates the mobilization of trapped oil and enhances displacement efficiency. IFT behavior is influenced by several factors, including salinity, ionic composition, and the presence of surfactants or biosurfactants. Monovalent ions such as Na^+ can alter electrostatic interactions at the interface, whereas divalent ions such as Ca^{2+} and Mg^{2+} typically exert stronger effects due to their ability to bridge polar compounds at the oil–water boundary. Biosurfactants such as rhamnolipids further reduce IFT by adsorbing at the interface and stabilizing micellar structures, thereby the attainment of ultralow

IFT values required for enhanced oil recovery (EOR) applications.

For medium oil (Figure 4a), IFT decreased as brine salinity increased from 5,000 ppm to 15,000 ppm, dropping from 13.37 mN/m to 2.83 mN/m. This trend indicates that increased salinity enhances ionic interaction at the oil water contact, lowering interfacial energy. In contrast, light crude oil (Figure 4b) exhibited the opposite behavior was observed, with IFT increasing from 1.00 mN/m at 5,000 ppm to 2.53 mN/m at 15,000 ppm. This response likely reflects weaker ionic interactions associated with the lower content of polar compound in light oil.

The addition of Ca^{2+} and Mg^{2+} further influenced IFT behavior, particularly in medium crude oil system (Figure 5a and b). At low salinity (5,000 ppm), Ca^{2+} increased IFT to 18.29 mN/m, suggesting stronger polar binding at the interface. However, this effect diminished at higher salinity due to ion crowding and interfacial saturation. Mg^{2+} also affected IFT, producing 12.94 mN/m at 5,000 ppm and decreasing to 2.58 mN/m at 15,000 ppm, showing a slightly stronger ability to reduce IFT at high salinity compared to Ca^{2+} .

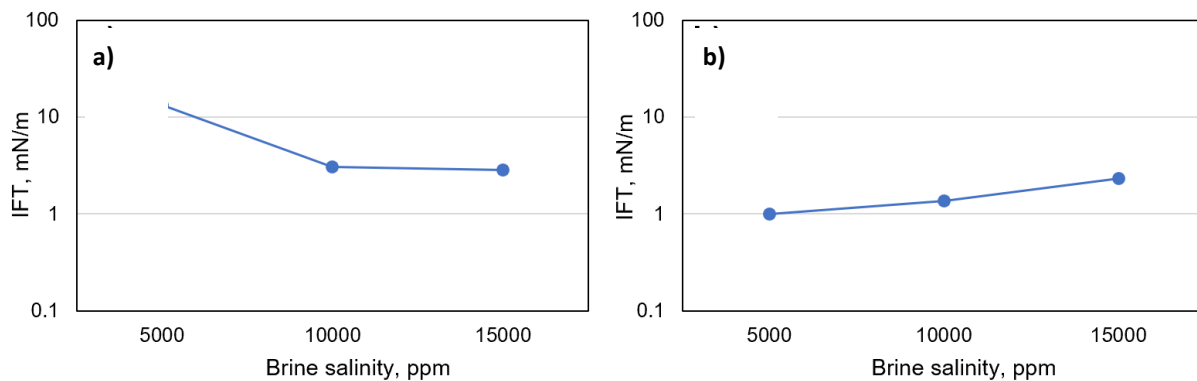


Figure 4. IFT vs brine salinity at medium (a); and light oil characteristic (b). IFT is plotted on a logarithmic scale and shows contrasting salinity dependence

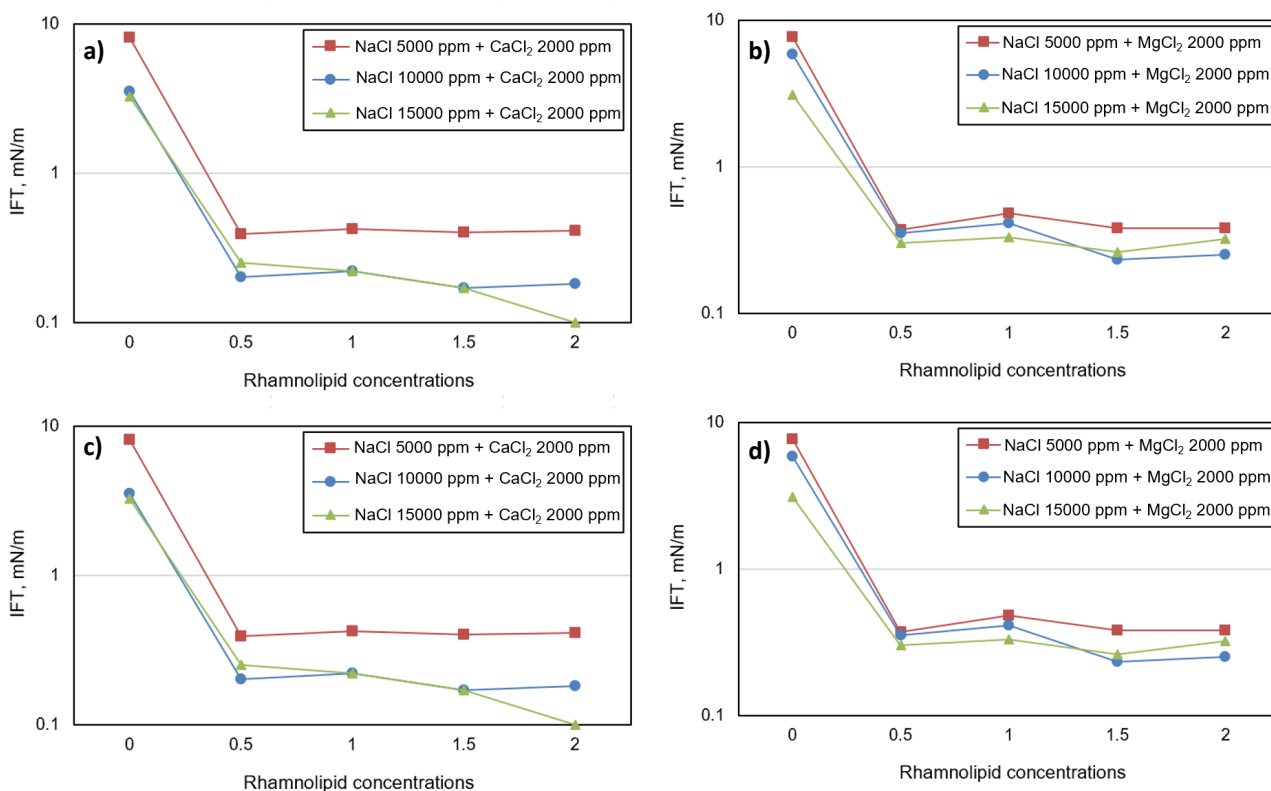


Figure 5. IFT vs rhamnolipid concentration at medium oil (a – b); and light oil (c – d) with varying brine salinity adding CaCl_2 and MgCl_2 at 2000 ppm.

In contrast, for light crude oil (Figure 5c and 5d), Ca^{2+} for low salinity (5,000 ppm) sharply reduced IFT to 0.07 mN/m, although its effect weakened as salinity increased. Mg^{2+} also reduced IFT in light oil systems, although with less consistency. The effect of rhamnolipids on IFT is shown in Figure 5. For medium oil (Figure 5a and b), IFT decreased rapidly as rhamnolipid concentration increased, reaching

ultralow values below 0.1 mN/m at 0.5%. Beyond this concentrations, IFT stabilized indicating saturation of the oil-water interface by surfactant molecules and the formation of micellar structures. The addition of Ca^{2+} and Mg^{2+} enhanced IFT reduction, with Ca^{2+} exhibiting a slightly stronger effects at higher rhamnolipid concentrations. For light oil (Figures 5c and d), a similar trend was

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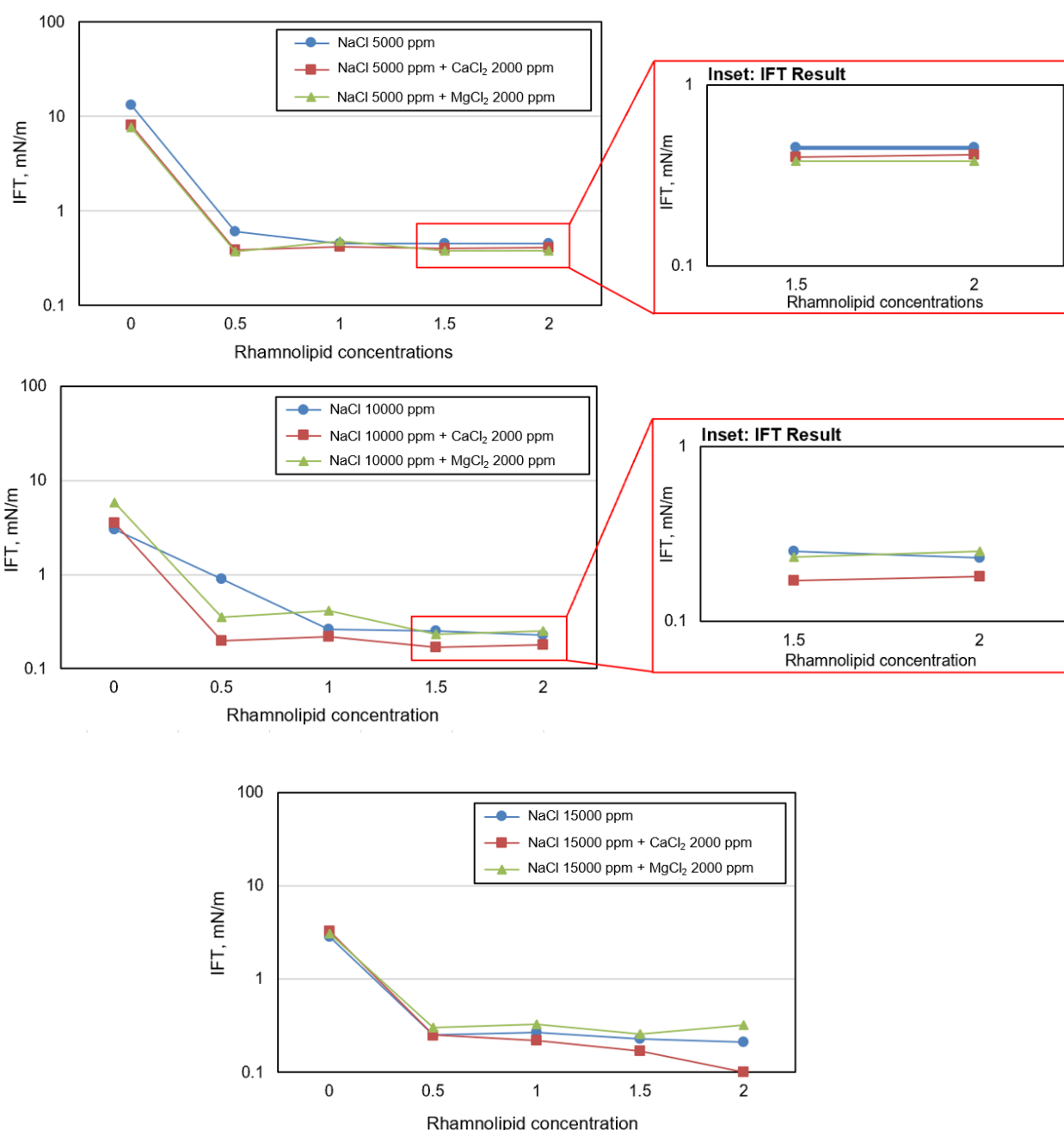


Figure 6. IFT vs rhamnolipid concentration at medium oil with varying brine salinity from 5000 ppm (a); 10000 ppm (b); to 15000 ppm (c) adding CaCl₂ and MgCl₂ at 2000 ppm.

observed with IFT decreasing below 0.1 mN/m at 0.5% rhamnolipids and remaining nearly constant at higher concentrations. The presence of Ca²⁺ and Mg²⁺ was less pronounced than in medium oil systems, since the biosurfactant alone was sufficient to dominate the interfacial properties. Overall, salinity and divalent ions affected medium and light oils differently,

however in both cases, rhamnolipids were highly effective in reducing IFT to ultralow levels. These results confirm that biosurfactants are powerful agents for interfacial modification, with stronger synergy observed in medium oils containing higher concentrations of polar components such as resins and asphaltenes.

Figures 6a – 6c demonstrate the critical role of rhamnolipid biosurfactant in lowering IFT under different salinity conditions. At relatively low salinity (5,000 ppm NaCl), IFT drops significantly as the concentration of rhamnolipid increases, eventually leveling off around 0.1 mN/m once it exceeds 1 wt%. The addition of divalent cations, especially calcium (Ca^{2+}), further enhances this effect at low to moderate levels, likely due to an electrostatic shielding, which promotes tighter surfactant alignment at the oil–brine boundary. Even at a higher salinity of 10,000 ppm NaCl, calcium maintains its advantage over sodium chloride alone, whereas magnesium (Mg^{2+}) produces a more variable performance.

As shown in Figures 7a – 7c, rhamnolipid biosurfactant effectively lowers IFT across a range of salinity levels. At low to moderate salinity (5,000–10,000 ppm NaCl), Ca^{2+} had

the strongest effect, lowering IFT values to significantly reduced levels (~ 0.01 mN/m). Mg^{2+} had less consistent effects. At higher salinity (15,000 ppm NaCl), the effect of divalent ions decreased, and all systems reached similar IFT values close to 0.1 mN/m after 1 wt% rhamnolipid. This behavior indicates that excessive ionic strength suppresses ion–surfactant synergy. Overall, these results highlight the potential of rhamnolipid as environmentally friendly surfactant for enhanced oil recovery, particularly under moderate salinity conditions where calcium ions significantly enhance interfacial performance.

Oil-water emulsion viscosity

Viscosity is another key parameter in oil recovery, as it reflects the stability and flow behavior of emulsions formed during water

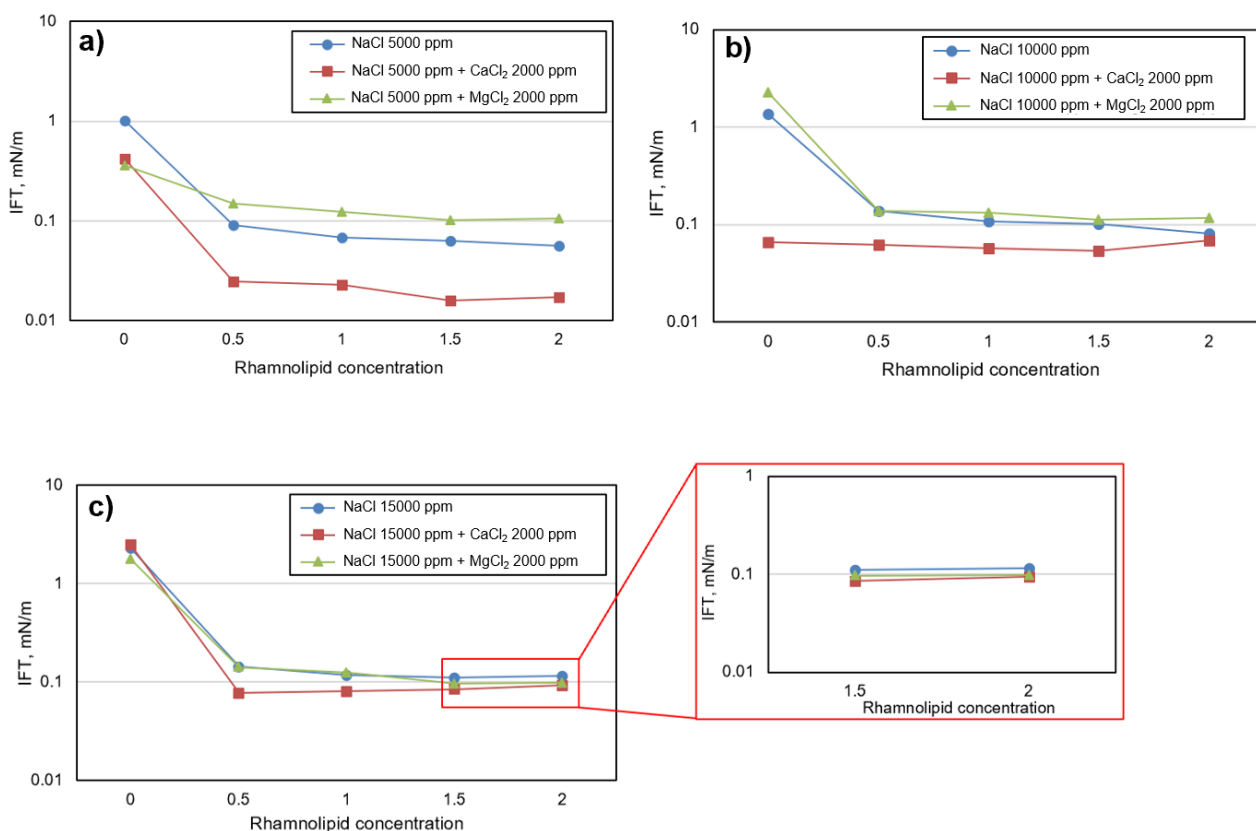


Figure 7. IFT vs rhamnolipid concentration at light oil with varying brine salinity from 5000 ppm (a); 10000 ppm (b); to 15000 ppm (c) adding CaCl_2 and MgCl_2 at 2000 ppm

flooding and chemical EOR. The presence of divalent ions such as Ca^{2+} and Mg^{2+} can significantly affect viscosity by promoting interactions among surfactants, water molecules, and polar components of crude oil. These interactions may lead to the formation of more complex molecular assemblies, including network-like or gel-like structures, which increase resistance to flow. Biosurfactants such as rhamnolipids further modify viscosity by stabilizing micelles at the oil–water boundary, thereby enhancing emulsion stability.

The viscosity profiles shown in Figures 8a and 8b indicate distinct system responses to variations in brine salinity. From Figure 8a viscosity increases significantly with salinity, reaching a maximum of around 16 cP at 10,000 ppm before declining at 15,000 ppm. This behavior suggests that optimal polymer-salt interactions occur at an intermediate ionic strength, resulting in chain expansion and increased solution viscosity. Conversely, Figure 8b illustrates that the viscosity remains rather constant throughout the same salinity range, maintaining a value about 1–2 cP. This indicates that the system exhibits minimal responsiveness to salinity, potentially due to limited interaction between the polymer and ions or the rigidity of the molecular structure. These contrasting trends demonstrate that polymer effectiveness in enhanced oil recovery applications is significantly influenced by

salinity, with optimal viscosity and mobility control achieved at moderate salinity levels.

The role of rhamnolipids is presented in Figure 9a–9d, which underscore the synergistic effects of rhamnolipid content, salinity, and divalent cations on solution behavior. In the presence of Ca^{2+} (Figure 9a and 9c), viscosity values exhibit a significant effect on both salinity and rhamnolipid concentration. At low salinity (5,000 ppm NaCl), viscosity remains consistently low across the concentration spectrum. At intermediate salinity (10,000 ppm NaCl), variations occur without a discernible monotonic pattern. At high salinity (15,000 ppm NaCl), an initial increase in viscosity at zero rhamnolipid concentration is succeeded by a notable decrease upon the introduction of rhamnolipid, signifying the instability of ion-surfactant interactions at increased ionic strength. This indicates that calcium ions augment viscosity more efficiently in elevated ionic conditions, yet may prove detrimental when paired with rhamnolipid beyond specific quantities.

In contrast, the addition of Mg^{2+} (Figure 9b and 9d) resulted the viscosity profile more equal across rhamnolipid concentrations, but the overall values were still substantially lower than those reported in Ca^{2+} systems. At 15,000 ppm NaCl with Mg^{2+} , the viscosity is always higher than that of Ca^{2+} systems. This illustrates that the thickening impact is stronger

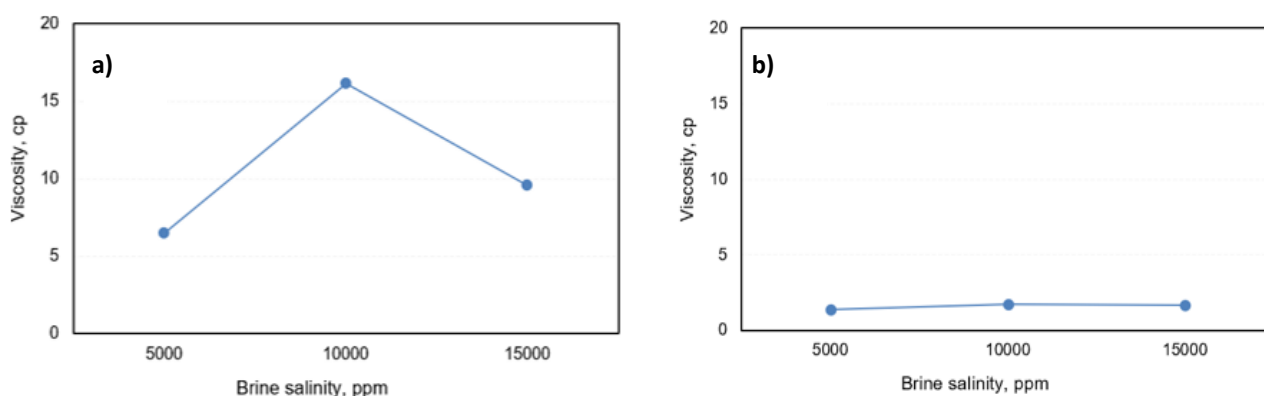


Figure 8. Viscosity vs brine salinity variation at (a) medium; and (b) light oil characteristic system

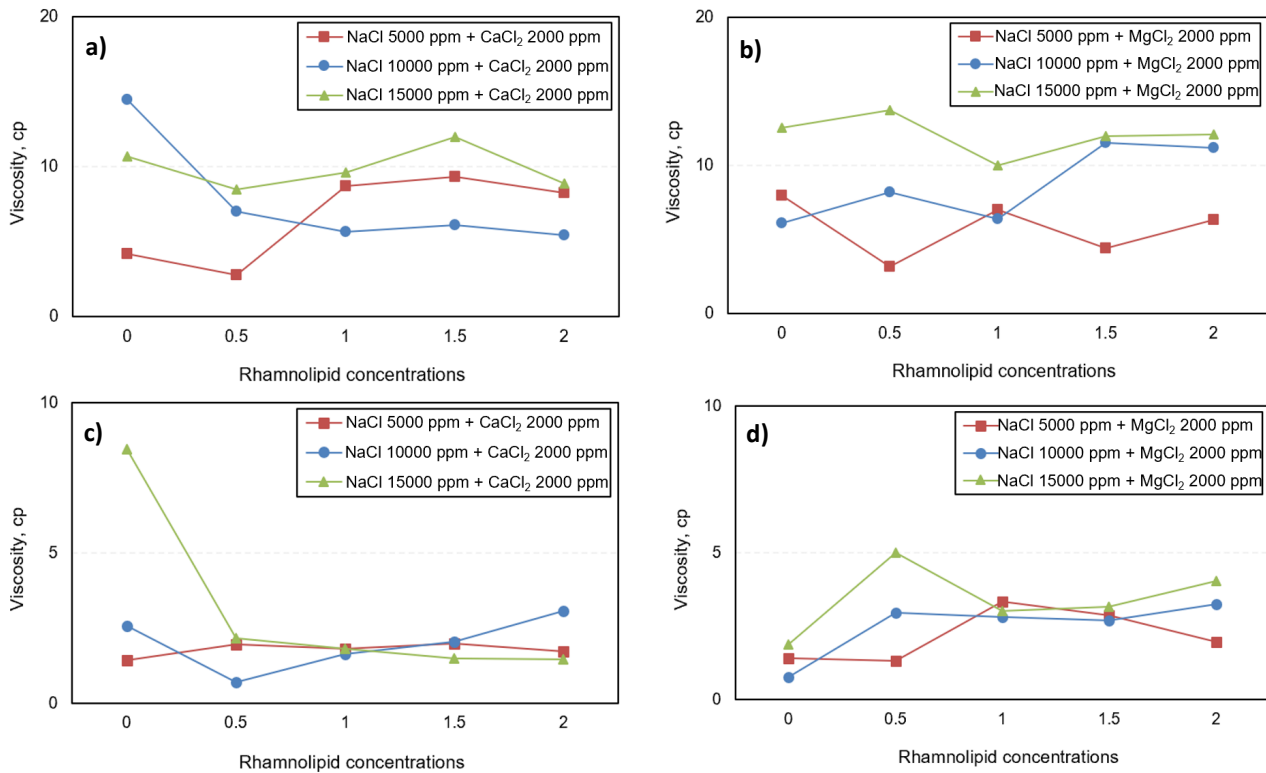


Figure 9. Mix viscosity at various rhamnolipids concentration for medium oil (a and b); and light oil (c and d) with adding CaCl_2 and MgCl_2

even when ionic conditions are strong. The lack of a clear rising trend in rhamnolipid concentration suggests that Mg^{2+} interacts with the biosurfactant, making it less sensitive to changes in concentration. The studies indicate that calcium induces more pronounced yet unstable alterations in viscosity, whereas magnesium results in more stable but less conspicuous modifications. This means that surfactant-brine mixtures for enhanced oil recovery applications need to be properly adjusted.

CONCLUSION

This study clearly demonstrates that the interplay among rhamnolipid, salinity, and divalent ions exerts a decisive impact on interfacial tension and viscosity, which are critical parameters for enhanced oil recovery. Calcium ions are generally more effective in reducing IFT and increasing viscosity under

moderate salinity conditions; however, their effects become less stable at higher salinity levels and concentrations. In contrast, magnesium ions tend to maintain moderate viscosity even under high ionic strength conditions, resulting in a more consistent but less pronounced response. Overall, rhamnolipid consistently achieve ultra-low IFT across a wide range of circumstances, highlighting their potential as environmentally friendly surfactants. Nevertheless, the results also indicate that optimal performance requires careful adjustment of salinity and divalent ion composition, as these factors can either enhance or hinder formulation effectiveness. Achieving this balance is essential for translating laboratory-scale findings into practical field application EOR.

Although these findings are promising, further research is required to optimize ion concentrations and evaluate their effects under practical reservoir conditions. Proper

management of divalent ions will be crucial for maximizing the efficiency of biosurfactant injection, improving oil recovery rates, and ensuring the sustainability of the EOR process in carbonate reservoirs.

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GLOSSARY OF TERMS AND SYMBOLS

Terms & Symbols	Definition	Unit
MEOR	Microbial enhanced oil recovery	
IFT	Interfacial tension	mN/m
API	American petroleum institute	°
w/w	Weight per weight	%
NaCl	Natrium chloride	ppm
CaCl ₂	Calcium chloride	ppm
MgCl ₂	Magnesium chloride	ppm
ppm	Parts per Million	
RPM	Rotation per Minute	
mL	Milliliters	

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