Article

Anti –bacterial study of polymeric blends for Oil well injection water application

Anmar S. Alabdy[1*], Yasin Y. Y. Al-luaibi[2], Nassir Abdullah Alyousif[3], Ali A. A. Al-Riyahee[1], Athir Haddad[1],

[1]Department of Chemistry, College of Science, University of Basrah, Basrah, Iraq.

pgs.anmar.oudah@uobasrah.edu.iq ali.abdulzahraa@uobasrah.edu.iq
athir.haddad@uobasrah.edu.iq

[2]Department of Biology, College of Science, University of Basrah. Basrah, Iraq,

yasin.yousif@uobasrah.edu.iq

[3]Department of Ecology, College of Science, University of Basrah. Basrah, Iraq.

nassir.hillo@uobasrah.edu.iq

Abstract

A total of thirty-three polymeric blends (A1-A33) were synthesised using xanthan gum, sodium alginate, carboxymethyl cellulose and polyacrylamide. The antimicrobial efficacy of all the synthesised polymeric blends was assessed against *Staphylococcus aureus*, a strain of gram-positive bacteria and *Escherichia coli*, a strain of gram-negative bacteria. The blends A9 and A12 exhibited antibacterial properties. A9 shown effective activity against both types of bacteria, but A12 exclusively displayed activity against Gram-positive bacteria. The two formulated polymeric blends were combined with varying quantities of *rhamnolipid (Rha2-C10-C10)* which are biosurfactants produced from *Pseudomonas aeruginosa*, a gram negative bacterium and evaluated against two strains of bacteria. However, no outcomes were seen. This study found that *(Rha2-C10-C10)* did not exhibit antibacterial properties when mixed with distilled water at lower concentrations. However, it was found to be effective as an antibacterial agent when not diluted with distilled water. Additionally, the study established the lowest concentration of polymer blend and rhamnolipid (Rha2-C10-C10) required for antibacterial activity.

Keywords antibacterial. biosurfactants, glycolipids, polymeric blends, Rhamnolipids, **w**ater-soluble polymers.

Introduction

The oil and gas industry's increased global water production necessitates environmental treatment and reuse of produced water, which contains high levels of pollutants [1]. Oil fields generate wastewater, posing environmental hazards to soil, air, and groundwater. To increase efficiency, "Enhanced Oil Recovery" involves injecting treated freshwater from rivers into wells. The Iraqi South Oil Company predicts a variable water production rate of 290,000 to 800,000 BBL/day from 2011 to 2028 for the North Rumaila field [2].

Polymer injection is widely used for its efficient oil recovery, utilizing water-soluble polymers in various water-related applications [3]. Crude oil classification based on sulphur content and density in reservoirs [4, 5].

Oil reservoir ecosystems' unique characteristics are determined by microbial population changes, allowing diverse microorganisms to thrive in challenging environments [6, 7]. Most oil field microorganisms can survive with or without oxygen due to their low redox potential in reservoirs. They prefer environments without oxygen [7].

The bacterial and archaeal communities present in oil fields consist of sulfate-reducing bacteria [8]. The microorganisms present include sulphur-oxidizing bacteria [9], methanogens [10], fermentative microorganisms [11], and acetogens [12], nitrate reducers [13], manganese and iron reducers [14], and hydrocarbon degraders [15].

Studies reveal that higher concentrations of $SO4^{-2}$ and Ca^{+2} ions change the wettability of rocks, leading to increased water wettability. [16]. Bacteria in water-injection systems can cause clogged wells and equipment, leading to H2S production and pitting corrosion, affecting the effectiveness of the enhanced oil recovery process [17, 18].

Surfactants, characterized by their amphiphilic nature, can be synthesized or microbially acted upon, exhibiting both hydrophilic and hydrophobic components in their molecular structure [19]. Lower-molecular-weight biosurfactants like glycolipids, lipopeptides, and phospholipids have industrial potential due to their ability to decrease surface and interfacial tension [20]. They are employed for improving solubility, combating bacteria and preventing adhesion, cleaning up contaminated areas, recovering crude oil, cleaning oil wells, and delivering medication [21].

Rhamnolipids (RHLs) are small amphiphilic glycolipid biosurfactants having anionic properties. They consist of rhamnose in the head and fatty acid chains in the tail [22, 23]. RHLs are significant environmentally friendly compounds synthesised by bacteria, known for their biocompatibility and nontoxicity [24]. RHLs interact with various bacteria, including Gram-negative and Gram-positive species, as well as fungi like Yarrowia lipolytica [25, 26].

Rhamnolipid, a key type of glycolipid, exhibits antibacterial properties due to its ability to permeabilize the bacterial plasma membrane, enhance hydrophobicity, and inhibit biofilm formation. [27,28]. Over 60 unique rhamnolipid variants have been identified, with microbial fermentation potentially generating a wide range of these, with variations in unsaturation degree, branching degree, and chain length. The molecular structure of mono- and di-rhamnolipids can be influenced by the number of rhamnose groups, as illustrated in Figure 1 [29].

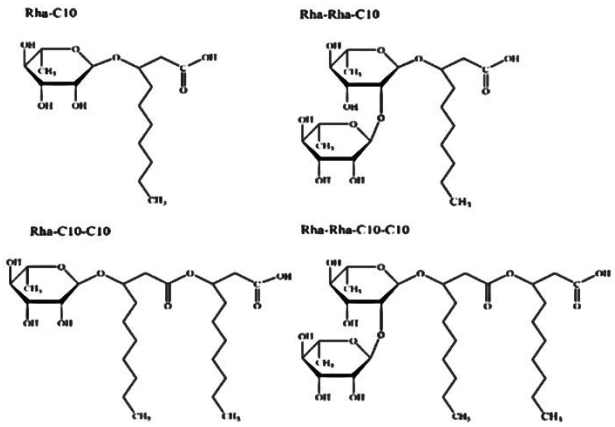


Figure 1. Common structures of rhamnolipids

In 1965, Cornell successfully synthesised the first antibacterial polymers by preparing homo- and copolymers of 2-methacryloxytroponone derivatives [30]. Polymers are increasingly used in green chemistry and environmental protection industries for antibacterial purposes, with some modified to include active function groups for enhanced germ-fighting properties. [31,32]. Physical and chemical techniques are utilized to modify polymers to possess antibacterial properties by inserting antibacterial elements or functional groups into the polymer matrix [33, 34].

In this study, we will prepare polymeric blends and assess their effectiveness against aerobic bacteria. We will also combine them with *Rhamnolipid (Rha$_2$-C10-C10)* as an antibacterial substance using a physical technique, in the form of mixtures, and study their antibacterial properties against two types of bacteria *Staphylococcus* is a type of gram-positive bacteria and *Escherichia coli*, is a type of gram-negative bacteria.

2. Materials and Methods

2.1 Chemicals.

Polyacrylamide and Carboxymethyl cellulose (CMC) were supplied from Alpha Chemika, while Sodium alginate and Xanthan gum were supplied from Hyper Chem.

2.2 The rhamnolipid biosurfactant Production

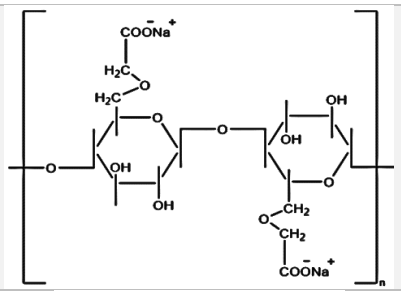
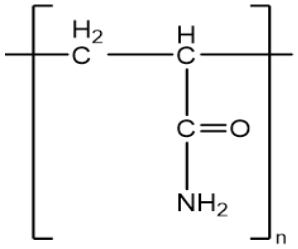
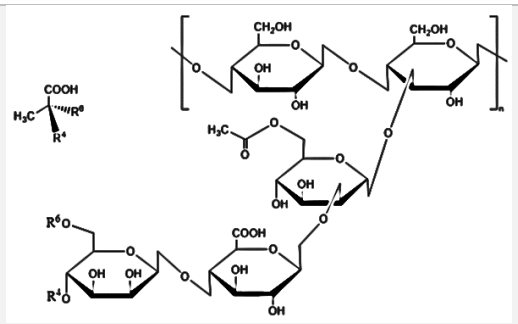
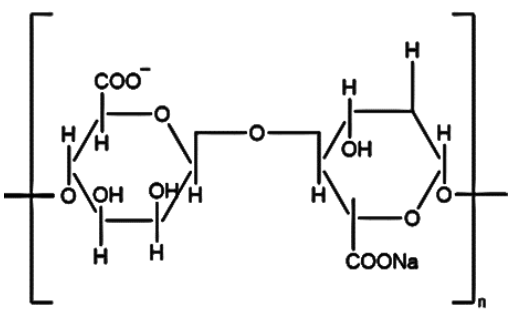
In a previous study involving hydrocarbon-contaminated soil, the biosurfactant rhamnolipid utilised in this research was produced from *Pseudomonas aeruginosa* [35]. Extracted, purified, and characterized [36].

2.3 Preparation of polymers blends.

Thirty-three polymeric blends have been prepared from different ratios of the water-soluble polymers carboxymethyl cellulose, xanthan gum, polyacrylamide and sodium alginate. Tables 1 and 2 present the chemical structures of the water-soluble polymers, as well as the symbols and composition ratios of the prepared blends, respectively.

Table 1 Polymers Structures

No	Polymer name	Code	chemical structures

1	Carboxymethyl cellulose	CMC	
2	Poly acrylamide	PAcAm	
3	Xanthan gum	Xanthan	
4	Sodium alginate	SAlg	

The polymeric blend A1 was prepared by dissolving one gram of CMC polymer and one gram of polyacrylamide in 50 millilitres of distilled water at 24 °C, after ensuring that the polymers dissolved completely. The solution was dried at 40 °C, and the result was ground into powder. Further polymer blends (A2–A33) were prepared using the same procedure, but with varying ratios of polymers as indicated in Table 2.

Table 2: Ratios of polymers used in blend formulation.

Sample	CMC (g)	PAcAm (g)	Xanthan gum (g)	SAlg (g)	Ratio of blends
A1	1	1	--	--	1:1

A2	1	2	--	--	1:2
A3	2	1	--	--	2:1
A4	1	--	1	--	1:1
A5	1	--	2	--	1:2
A6	2	--	1	--	2:1
A7	--	1	1	--	1:1
A8	--	1	2	--	1:2
A9	--	**2**	**1**	--	**2:1**
A10	1	--	4	--	1:4
A11	1	--	5	--	1:5
A12	1	--	6	--	1:6
A13	--	3	1	--	3:1
A14	--	4	1	--	4:1
A15	--	5	1	--	5:1
A16	--	1	3	--	1:3
A17	--	1	4	--	1:4
A18	--	1	5	--	1:5
A19	--	1	--	1	1:1
A20	--	2	--	1	2:1
A21	--	3	--	1	3:1
A22	--	1	--	2	1:2
A23	--	1	--	3	1:3
A24	1	--	--	1	1:1
A25	2	--	--	1	2:1
A26	3	--	--	1	3:1
A27	1	--	--	2	1:2
A28	1	--	--	3	1:3
A29	--	--	1	1	1:1
A30	--	--	2	1	2:1
A31	--	--	3	1	3:1
A32	--	--	1	2	1:2
A33	--	--	1	3	1:3

2.4 Preparing of sample for anti-bacterial study

In this study, two types of samples were investigated. In the first experiment, polymer blends were dissolved in various concentrations of distilled water in separate tubes. The second experiment, different concentrations of polymer blends had dissolved in distilled water and then mixed with *rhamnolipids (Rha2-C10-C10),* an antibacterial substance that was dissolved in DMSO. The most straightforward approach is considered to be the method of blending, which entails combining organic and inorganic antibacterial compounds with the polymers [37- 40]. The polymeric compositions and biosurfactant blends were thoroughly mixed for 20 minutes to achieve homogeneity. Afterwards, the solutions were allowed to fully dissolve at room temperature for a duration of 24 hours. DMSO and water are the controlling samples.

2.5 Biological activity

The antibacterial properties were evaluated against two different kinds of bacteria *Staphylococcus aureus* and *Escherichia coli*. The efficacy of polymeric blends and mixtures of *rhamnolipids (Rha2-C10-C10)* with polymer blends in inhibiting the growth of *Staphylococcus aureus* and *Escherichia coli* was evaluated by incubating them at 37 °C for 24 hours. Disk Diffusion Testing on Mueller-Hinton Agar method was used [41].

3. Results and discussion

The bacterial activity was monitored and analyzed as follow; Initially, the bacteria were exposed to polymer blends (A1–A33) and original polymers, each at a concentration of 7.5 mg, in separate culture dishes containing either *E. coli* or *Staphylococcus aureus* the culture were incubated for 24 hours. Among these, only two blends demonstrated antibacterial activity, as detailed in Table 3.

Table 3. The inhibition zone diameter of polymers against *Staphylococcus aureus* and *Escherichia coli*

Sample	Concentration mg in 1ml water	Inhibition Zone (mm)	
		S aureus	***E.coli***
`A9	7.5	24	16
A12	7.5	12	R

R- Resistant.

The subsequent experiment evaluated the antibacterial efficacy of combinations of *rhamnolipids (Rha2-C10-C10)* with polymer blends A9 and A12 at various concentrations. The findings of this experiment against *S. aureus* and *E. coli* are presented in Table 4 and Table 5, respectively.

Table 4. The inhibition zone diameter mixes of *rhamnolipids (Rha$_2$-C10-C10)* with polymer blends A9 and A12 against bacteria *S aureus* and *E coli*

Sample	Concentration Mg			Inhibition Zone (mm)	
	Polymers in 1 ml water	Rha$_2$-C10-C10 in DMSO	Ratio	***S aureus***	***E.coli***
`A9	7.5	10	1:1	R	R
A9	7.5	20	1:1	R	R
A12	7.5	10	1:1	R	R
A12	7.5	20	1:1	R	R
Rha$_2$-C10-C10 in DMSO	--	10	--	16	R
	--	7.5	--	R	R
	--	5	--	R	R

DMSO and Distilled water	--	--	--	R	R

R- Resistant.

-- No concentration of the substance was applied

Table 5. The inhibition zone diameter for mixes of *rhamnolipids (Rha$_2$-C10-C10)* with polymer blends A9 and A12 against *S aureus* and *E coli.*

Sample	Concentration Mg			Zone Inhibition (mm)	
	Polymer blends in water	Rha$_2$-C10-C10 in DMSO	Ratio	*S aureus*	*E.coli*
Rha$_2$-C10-C10 in DMSO	--	20	--	16	R
A9	7.5	20	1:1	R	R
A12	7.5	20	1:1	R	R
A9	7.5	--	--	24	16
A12	7.5	--	--	12	R
A9	15	--	--	R	R
A12	15	--	--	R	R
DMSO and Distilled water	--	--	--	R	R
A9	15	20	1:1	R	R
A12	15	20	1:1	R	R

R- Resistant.

-- no concentration of the substance was applied

Table 6 identified the minimum polymer concentration in the prepared blends. Inoculation was carried out using original polymers and blends prepared with varying concentrations (0.5–15 mg) on both bacteria, showing no activity against both of them bacteria except for A9 and A12, which yielded the same results as those mentioned above, which for polymeric blends was 7.5 mg while for Rha2-C10-C10 it was 10 mg.

Table 6. The inhibition zone diameter Initial Polymers and polymer blends (at different concentreation) against bacteria *Staphylococcus aureus* and *Escherichia coli*

Sample	Concentration Mg	Zone Inhibition (mm)	
	Polymer blends in water	*S aureus*	*E.coli*
CMC	7.5	R	R
PAcAm	7.5	R	R

Xanthan	7.5	R	R
SAlg	7.5	R	R
A9	7.5	24	16
A9	3.75	R	R
A9	2	R	R
A9	1	R	R
A9	0.5	R	R
A9	15	R	R
A12	7.5	12	R
A12	3.75	R	R
A12	2	R	R
A12	1	R	R
A12	0.5	R	R
A12	15	R	R

R- Resistant.

From the data presented in Tables 4 and 5 regarding the use of *rhamnolipids (Rha2-C10-C10)*, it is evident that Rha2-C10-C10 has demonstrated significant efficacy in inhibiting gram-positive bacteria, specifically *Staphylococcus aureus*. The inhibition zone measured 16 mm for both concentrations of 10 and 20 mg, as depicted in Figure 2. However, Rha2-C10-C10 did not exhibit any effectiveness against *Escherichia coli (E. coli).*

In order to ascertain the minimal concentration, the bacteria were inoculated with 7.5 and 5 mg of Rha2-C10-C10; however, no activity was observed against either species. In general, *glycolipids* exhibit stronger antibacterial properties against gram-positive bacteria compared to gram-negative bacteria, possibly due to variations in cell wall composition. Remarkably, the study demonstrated that gram-negative *E. coli* exhibited resistance to *rhamnolipids* at all concentrations that were studied.

Gram-negative bacteria have a more sophisticated and protective cell envelope than gram-positive bacteria, consisting of an outer layer (lipopolysaccharides and phospholipids), peptidoglycan, and an interior plasma membrane. The unique structure made it difficult for glycolipids to enter gram-negative bacteria, and it is widely accepted that the underlying mechanism of antibacterial activity involves reduced membrane permeability, loss of intracellular constituents, and death of cells caused by membrane lysis [42, 43]. *Rhamnolipids* target planktonic bacteria, altering and damaging the cell membrane, leading to higher cell permeability and lower cell surface hydrophobicity. Due to their amphiphilic nature, rhamnolipids can interact with phospholipids [44, 45].

The reduction in surface tension between the molecules allows the solid or liquid solute particles to form hydrophilic or hydrophobic contacts with the solvent, rendering immiscible fluids miscible through the production of new extra surfaces [46]. *Rhamnolipid* molecules are amphiphilic, which means they include both hydrophilic (water-attracting) and hydrophobic (water-repelling) sections. *Rhamnolipids* have the property of being amphiphilic, which means they can aggregate or form micelles when they are dissolved in water. *Rhamnolipids* that are dissolved in a hydrophobic

solvent, such as *dimethyl sulfoxide (DMSO)*, are better at lowering surface tension and creating emulsions than these aggregates.

Rhamnolipids can interact with other molecules in the solution, influencing their capacity to inhibit bacteria. While DMSO reduces certain interactions or stabilises *rhamnolipid* molecules to prevent aggregation, it is efficient in reducing bacterial growth [47, 48]. This provides an explanation for the results obtained in tables 4 and 5, which were shown when *rhamnolipid* was mixed after dissolving it in DMSO with the prepared polymeric mixtures that were dissolved in distilled water, as it did not give any effectiveness in inhibiting gram-positive bacteria (*Staphylococcus aureus*) and gram-negative bacteria (*E. coli*). Furthermore, when *rhamnolipid* was dissolved in DMSO and then diluted by 50% with distilled water, it failed to exhibit any efficacy in inhibiting the growth of both gram-positive bacteria (*Staphylococcus aureus*) and gram-negative bacteria (*E. coli*). This is due to the fact that prior research has demonstrated that *rhamnolipids* possess an inherent tendency to aggregate, as well as engage in interactions with water and DMSO. There are three stable arrangements of water in dimethyl sulfoxide. Water forms a main OH···OS link with the oxygen atom and two weak CH···OH bonds with the methyl hydrogen atoms [48, 49].

Based on the data presented in tables 3 and 6, it became clear that only two blends, A9 and A12, exhibited antibacterial activity. The A9 blend exhibited significant activity against *Staphylococcus aureus*, with an inhibition zone of 24 mm at a concentration of 7.5 mg. It also showed activity against *E. coli*, with an inhibition zone of 16 mm at the same concentration. On the other hand, the A12 blend only displayed antibacterial activity against *Staphylococcus aureus*, with an inhibition zone of 12 mm at a concentration of 7.5 mg (Figure 2). It is possible that the discovery is a result of the unique characteristics of the surface of gram-negative bacteria, which is coated with a thick layer of *lipopolysaccharide* (LPS). The negative charge and non-polar structures on this surface could potentially hinder chemical interactions. Including certain antimicrobial properties [50, 51].

The A9 blend exhibits significantly stronger antibacterial activity against *S. aureus* compared to *E. coli*. This discrepancy in inhibitory impact on growth of the examined bacteria might be assigned to their distinct cell wall structural composition, and previous researchers found similar findings [52, 53]. The majority of *S. aureus's* thick cell wall is composed of 90% peptidoglycan and 10% teichoic acid, which have negative charges. Thus, there is a possibility of electrostatic interactions between polymer molecules with opposite charges and teichoic acid, which can disrupt the surface shape of bacterial cells and result in the leakage of cellular content [54].

The A9 blend was prepared by combining polyacrylamide and xanthan gum in a ratio of 1:2. Because of this, it has an amine group that has a positive charge on the nitrogen, as well as xanthan gum, which has a lot of hydroxyl groups. The antibacterial activity can be controlled by several factors such as the quantity of hydroxyl groups, percentage of glucose, viscosity, rate of solubility, alterations in configuration, molecular size, surface area, and others [55]. The antibacterial efficacy of LW-XG against S. aureus may be ascribed to its low molecular weight and the presence of hydroxyl groups in the molecules [56]. That's why the A9 blend demonstrated antibacterial activity against *S. aureus*, whose inhibition zone was 24 mm, and *E. coli*'s inhibition zone was 16 mm.

E. coli's cell wall differs from that of gram-positive bacteria, with an outer layer made of lipopolysaccharide, lipid bilayers, and lipoprotein and a thin peptidoglycan layer [57]. The complex bilayer cell structure may prevent A12 blend molecules from entering *E. coli* bacterial cells. The lack of effectiveness of blend A12 against *E. coli* (gram-negative bacteria) may be attributed to this

clear cause. Additionally, the lesser effectiveness of blend A9 against the same bacteria, compared to its effectiveness against *S. aureus* (gram-positive bacteria), can be explained.

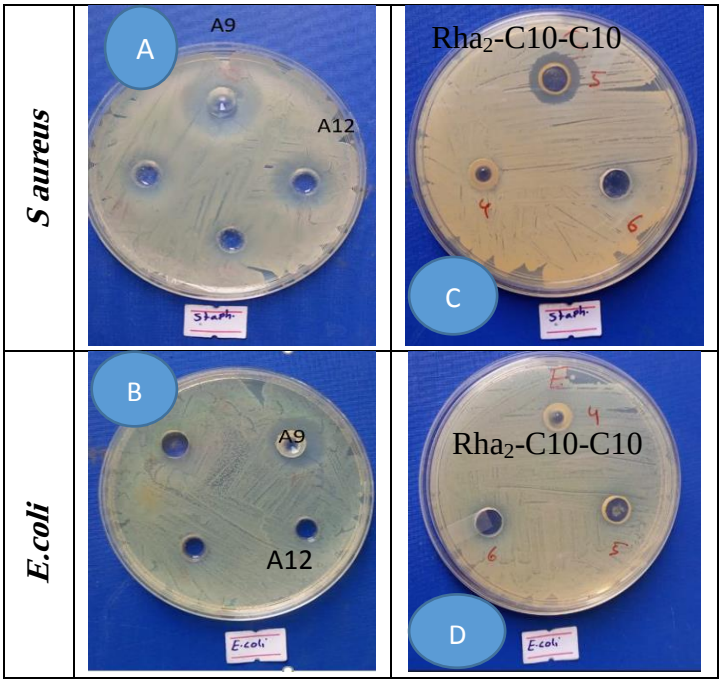


Figure 2. Showing inhibition zone A, A9 and A12 blends against *S aureus*, B against *E. coli*, C Rha_2-C10-C10 against *S aureus* and D Rha_2-C10-C10 against *E. coli*.

4. Conclusion

Out of the thirty-three polymeric-prepared blends, two blends exhibit antibacterial activity. A9 has antibacterial activity against both *S. aureus* (a type of gram-positive bacteria) and *E. coli* (a type of gram-negative bacteria), but A12 exclusively shows antibacterial activity against *S. aureus* (a type of gram-positive bacteria). *Rhamnolipids (*Rha_2*-C10-C10)*, shown clear effectiveness against gram-positive bacteria *(Staphylococcus aureus)* only. The study determined that the combination of rhamnolipid, dissolved in DMSO, with polymer blends, dissolved in distilled water, did not effectively inhibit both gram-positive and gram-negative bacteria. Dissolving rhamnolipid in DMSO and combining it with 50% distilled water did not effectively suppress both gram-positive and gram-negative bacteria. The lowest concentrations of polymer-blends and Rha2-C10-C10 were identified.

References

1- Al-Ghouti, Mohammad A., et al. "Produced water characteristics, treatment and reuse: A review." Journal of Water Process Engineering 28 (2019): 222-239.

2- Kuraimid, Z. Kh, et al. "Paper Title Treatment of produced water in North Rumela oil field for re-injection Application." SPE Kuwait Oil and Gas Show and Conference. SPE, 2013.

3- Dwivedi, Sumant, et al. "High-temperature resistant water-soluble polymers derived from exotic amino acids." RSC advances 10.62 (2020): 38069-38074.

4- Romanow-Garcia, S., and H. L. Hoffman. "Petroleum and its products." Kent and Riegel's handbook of industrial chemistry and biotechnology. Boston, MA: Springer US, 2007. 801-842.

5- Speight, James G. Handbook of industrial hydrocarbon processes. Gulf Professional Publishing, 2019.

6- Varjani, Sunita J., and Edgard Gnansounou. "Microbial dynamics in petroleum oilfields and their relationship with physiological properties of petroleum oil reservoirs." Bioresource technology 245 (2017): 1258-1265.

7- Youssef, Noha, Mostafa S. Elshahed, and Michael J. McInerney. "Microbial processes in oil fields: culprits, problems, and opportunities." Advances in applied microbiology 66 (2009): 141-251.

8- Hussain, Ali, et al. "Exploited application of sulfate-reducing bacteria for concomitant treatment of metallic and non-metallic wastes: a mini review." 3 Biotech 6 (2016): 1-10.

9- Tian, Huimei, et al. "Compositions and abundances of sulfate-reducing and sulfur-oxidizing microorganisms in water-flooded petroleum reservoirs with different temperatures in China." Frontiers in Microbiology 8 (2017): 143.

10- Berdugo-Clavijo, Carolina, and Lisa M. Gieg. "Conversion of crude oil to methane by a microbial consortium enriched from oil reservoir production waters." Frontiers in microbiology 5 (2014): 83260.

11- Tüccar, Tuğçe, Esra Ilhan-Sungur, and Gerard Muyzer. "Bacterial Community Composition in Produced Water of Diyarbakır Oil Fields in Turkey: Bacterial communities in produced waters of south-eastern Turkey reported in detail for the first time." Johnson Matthey Technology Review 64.4 (2020): 452-466.

12- Begum, Sameena, et al. "Kinetics of Biochemical Degradation of Municipal Solid Waste in Landfills." Solid Waste Management. CRC Press, 2024. 48-64.

13- Okoro, Chuma Conlette, and Olukayode O. Amund. "Microbial community structure of a low sulfate oil producing facility indicate dominance of oil degrading/nitrate reducing bacteria and Methanogens." Petroleum Science and Technology 36.4 (2018): 293-301.

14- Tamazawa, Satoshi, et al. "Petrothermobacter organivorans gen. nov., sp. nov., a thermophilic, strictly anaerobic bacterium of the phylum Deferribacteres isolated from a deep subsurface oil reservoir." International Journal of Systematic and Evolutionary Microbiology 67.10 (2017): 3982-3986.

15- Van Hamme, Jonathan D., Ajay Singh, and Owen P. Ward. "Recent advances in petroleum microbiology." Microbiology and molecular biology reviews 67.4 (2003): 503-549.

16- A, Radhi. “Enhancement of Oil Recovery in West Qurna-1 Carbonate Reservoir by Injecting Seawater.” Petroleum & Petrochemical Engineering Journal, vol. 7, no. 2, Apr. 2023, pp. 1–15. https://doi.org/10.23880/ppej-16000353.

17- Naser, Abuelfotouh A., and Fadel Dahab. "Cost-Effective Solution for Harsh Well Conditions Reciprocating Rod Pumping (RRP) Wells in Oman." Paper presented at the SPE Conference at Oman Petroleum & Energy Show, Muscat, Oman, April 2024. doi: https://doi.org/10.2118/218499-MS

18- Popoola, Lekan Taofeek, et al. "Corrosion problems during oil and gas production and its mitigation." International Journal of Industrial Chemistry 4 (2013): 1-15.

19- Bjerk, Thiago R., et al. "Biosurfactants: properties and applications in drug delivery, biotechnology and ecotoxicology." Bioengineering 8.8 (2021): 115.

20- Jahan, Ruksana, et al. "Biosurfactants, natural alternatives to synthetic surfactants: Physicochemical properties and applications." Advances in colloid and interface science 275 (2020): 102061.

21- Ali, Irfan, et al. "Biosurfactants: introduction and classification." Industrial Applications of Biosurfactants and Microorganisms. Academic Press, 2024. 1-23.

22- Raza, Zulfiqar Ali, et al. "Production of rhamnolipid surfactant and its application in bioscouring of cotton fabric." Carbohydrate research 391 (2014): 97-105.

23- de Jesús Cortés-Sánchez, Alejandro, Humberto Hernández-Sánchez, and María Eugenia Jaramillo-Flores. "Biological activity of glycolipids produced by microorganisms: new trends and possible therapeutic alternatives." Microbiological research 168.1 (2013): 22-32.

24- de Araujo, Livia Vieira, et al. "Rhamnolipid and surfactin: Anti-adhesion/antibiofilm and antimicrobial effects." Food Control 63 (2016): 171-178.

25- Arif, Muhammad, et al. "Chitosan-based nanoparticles as delivery-carrier for promising antimicrobial glycolipid biosurfactant to improve the eradication rate of Helicobacter pylori biofilm." Journal of Biomaterials Science, Polymer Edition 32.6 (2021): 813-832.

26- e Silva, S. S., et al. "Disruption of Staphylococcus aureus biofilms using rhamnolipid biosurfactants." Journal of dairy science 100.10 (2017): 7864-7873.

27- Alyousif, Nassir Abdullah, et al. “Antimicrobial and Antioxidant Activity of Rhamnolipids Biosurfactant is Produced by Pseudomonas aeruginosa.” Bionatura, vol. 8, no. 4, Dec. 2023, pp. 1–11. https://doi.org/10.21931/rb/2023.08.04.25.

28- da Silva, Anderson Ramos, et al. "Rhamnolipids functionalized with basic amino acids: Synthesis, aggregation behavior, antibacterial activity and biodegradation studies." Colloids and Surfaces B: Biointerfaces 181 (2019): 234-243.

29- Ławniczak, Łukasz, Roman Marecik, and Łukasz Chrzanowski. "Contributions of biosurfactants to natural or induced bioremediation." Applied microbiology and biotechnology 97 (2013): 2327-2339.

30- Cornell, Robert J., and L. Guy Donaruma. "2-Methacryloxytropones. Intermediates for the synthesis of biologically active polymers." Journal of medicinal chemistry 8.3 (1965): 388-390.

31- Parcheta, Monika, and Magdalena Sobiesiak. "Preparation and Functionalization of Polymers with Antibacterial Properties—Review of the Recent Developments." Materials 16.12 (2023): 4411.

32- Borjihan, Qinggele, and Alideertu Dong. "Design of nanoengineered antibacterial polymers for biomedical applications." Biomaterials science 8.24 (2020): 6867-6882.

33- Li, Fu, et al. "Synthesis, characterization and excellent antibacterial property of cellulose acetate reverse osmosis membrane via a two-step reaction." Carbohydrate polymers 216 (2019): 312-321.

34- Tsekova, Petya B., et al. "Electrospun curcumin-loaded cellulose acetate/polyvinylpyrrolidone fibrous materials with complex architecture and antibacterial activity." Materials Science and Engineering: C 73 (2017): 206-214.

35- Alyousif, N.A., Al-Luaibi, Y.Y.Y. & and Hussein, W. Distribution and molecular characterization of biosurfactant-producing bacteria. Biodiversitas, 21(2020): 4034-4040. DOI: 10.13057/biodiv/d210914.

36- Alyousif, N.A., Al-Tamimi, W.H. & Al-Luaibi, Y.Y.Y. Screening, enhance production and characterization of biosurfactant produced by Pseudomonas aeruginosa isolated from hydrocarbon contaminated soil. Eurasia Journal of Bioscience, 14(2020): 4377-4391.

37- Gong, Yiyu, Hepeng Wang, and Jing Sun. "AMP-Mimetic Antimicrobial Polymer-Involved Synergic Therapy with Various Coagents for Improved Efficiency." Biomacromolecules (2024).

38- Abou-Zeid, N. Y., et al. "Preparation, characterization and antibacterial properties of cyanoethylchitosan/cellulose acetate polymer blended films." Carbohydrate Polymers 84.1 (2011): 223-230.

39- Beisl, Stefan, et al. "Synthesis and bactericide activity of nanofiltration composite membranes–Cellulose acetate/silver nanoparticles and cellulose acetate/silver ion exchanged zeolites." Water research 149 (2019): 225-231.

40- Jia, Leilei, Xinyu Huang, and Qiong Tao. "Enhanced hydrophilic and antibacterial efficiencies by the synergetic effect TiO2 nanofiber and graphene oxide in cellulose acetate nanofibers." International journal of biological macromolecules 132 (2019): 1039-1043.

41- Ooi, Shok Yin, Ishak Ahmad, and Mohd Cairul Iqbal Mohd Amin. "Cellulose nanocrystals extracted from rice husks as a reinforcing material in gelatin hydrogels for use in controlled drug delivery systems." Industrial Crops and Products 93 (2016): 227-234.

42- Shu, Qin, et al. "Contributions of glycolipid biosurfactants and glycolipid-modified materials to antimicrobial strategy: A review." Pharmaceutics 13.2 (2021): 227.

43- Zhang, Lijuan, et al. "Lighting up the interactions between bacteria and surfactants with aggregation-induced emission characteristics." Materials Chemistry Frontiers 1.9 (2017): 1829-1835.

44- de Freitas Ferreira, Jakeline, Estevão Alan Vieira, and Marcia Nitschke. "The antibacterial activity of rhamnolipid biosurfactant is pH dependent." Food Research International 116 (2019): 737-744.

45- Sánchez, Marina, et al. "Modulation of the physical properties of dielaidoylphosphatidylethanolamine membranes by a dirhamnolipid biosurfactant produced by Pseudomonas aeruginosa." Chemistry and physics of lipids 142.1-2 (2006): 118-127.

46- Abbot, Vikrant, and Poonam Sharma. "Thermodynamics and acoustic effects of quercetin on micellization and interaction behaviour of CTAB in different hydroethanol solvent systems." Zeitschrift für Physikalische Chemie 235.9 (2021): 1177-1207.

47- Esposito, Rodolfo, et al. "Rhamnolipid Self-Aggregation in Aqueous Media: A Long Journey toward the Definition of Structure–Property Relationships." International Journal of Molecular Sciences 24.6 (2023): 5395.

48- Lv, Dingding, et al. "Characterizing the interactions of dimethyl sulfoxide with water: A rotational spectroscopy study." The Journal of Physical Chemistry A 126.39 (2022): 6882-6889.

49- Kiefer, Johannes, Kristina Noack, and Barbara Kirchner. "Hydrogen bonding in mixtures of dimethyl sulfoxide and cosolvents." Current Physical Chemistry 1.4 (2011): 340-351.

50- Hsouna, Anis Ben, et al. "Chemical composition, cytotoxicity effect and antimicrobial activity of Ceratonia siliqua essential oil with preservative effects against Listeria inoculated in minced beef meat." International journal of food microbiology 148.1 (2011): 66-72.

51- Barreiros, Yuri, et al. "Xanthan gum-based film-forming suspension containing essential oils: Production and in vitro antimicrobial activity evaluation against mastitis-causing microorganisms." Lwt 153 (2022): 112470.

52- Su, Zhiwei, et al. "Preparation, characterization and antibacterial properties of 6-deoxy-6-arginine modified chitosan." Carbohydrate polymers 230 (2020): 115635.

53- Yue, Lin, et al. "Geraniol grafted chitosan oligosaccharide as a potential antibacterial agent." Carbohydrate polymers 176 (2017): 356-364.

54- Xu, Tao, et al. "Synthesis, characterization, and antibacterial activity of N, O-quaternary ammonium chitosan." Carbohydrate research 346.15 (2011): 2445-2450.

55- Mirzadeh, Monirsadat, Mohammad Reza Arianejad, and Leila Khedmat. "Antioxidant, antiradical, and antimicrobial activities of polysaccharides obtained by microwave-assisted extraction method: A review." Carbohydrate polymers 229 (2020): 115421.

56- Hu, Xiaolong, et al. "Characterization and antioxidant activity of a low-molecular-weight xanthan gum." Biomolecules 9.11 (2019): 730.

57- Palencia, Manuel, Tulio A. Lerma, and Álvaro A. Arrieta. "Antibacterial and non-hemolytic cationic polyurethanes with N-carboxymethyl-N, N, N-triethylammonium groups for bacteremia-control in biomedical-using materials." Materials Today Communications 22 (2020):10070