ISSN 2664-8075 (Print) & ISSN 2706-5774 (Online)
South Asian Research Journal of Applied Medical Sciences
Abbreviated Key Title: South Asian Res J App Med Sci

| Volume-7 | Issue-2 | Mar-Apr- 2025 | **DOI: https://doi.org/10.36346/sarjams.2025.v07i02.003**

Review Article

Ultrasound-Mediated Attenuation of Quorum Sensing and Its Impact on Biofilm Structure

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Article History
Received: 19.02.2025
Accepted: 27.03.2025
Published: 14.04.2025

Abstract: Bacteria use quorum sensing (QS) and biofilm formation as essential adaptation and persistence strategies in harsh settings, which contribute to the development of antibiotic resistance and persistent infections. Through signaling molecules, QS allows bacteria to coordinate group behaviors, and biofilms improve microbial resistance and offer structural protection. A potentially effective physical intervention for interfering with QS signaling and biofilm integrity is ultrasound. The modulation of QS pathways by ultrasound, including signaling molecule degradation and interference with QS receptor binding, is examined in this article. The physical disruption of biofilm structures by ultrasonic via cavitation and shear stress is also investigated. Important discoveries demonstrate how well ultrasound attenuates QS and encourages biofilm disintegration, which in turn increases bacterial sensitivity to antimicrobial drugs. The possible uses of ultrasound in both medical and environmental contexts are discussed, emphasizing its role as a non-invasive, targeted strategy for controlling biofilm-related infections and microbial persistence. Despite its promise, the purpose of this review is to provide a brief background on biofilms, and implants, and advance approach for treatment. Further research is needed to refine ultrasound parameters and optimize its integration with other therapeutic approaches for broader clinical and industrial use.

Keywords: Ultrasound, quorum sensing, biofilm attenuation.

INTRODUCTION

Quorum Sensing and Its Role in Biofilm Formation

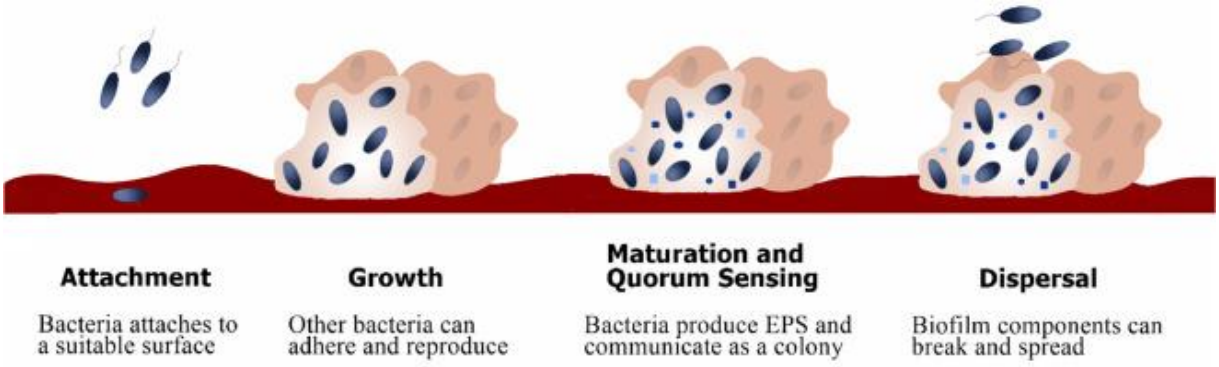
Quorum sensing (QS) is a cell-to-cell communication mechanism used by bacteria to coordinate collective behaviors as a result to response for changes in population abundance and community structure (Zeng *et al.*, 2023). This activity engages production, and reaction to extracellular indicating particles known as autoinducers (AIs). Recently studies have reported microorganisms such as bacteriophages, viruses that infect bacteria, also involve in utilizing chemical connection to arrange communication action (Sheraz *et al*., 2025). Quorum sensing mechanism: signaling molecules (autoinducers) accumulate to threshold levels, leading to coordinated gene expression among microbial populations (Xu, Lu *et al.*, 2020).

QS-mediated communication is wide common through the bacterial species systems leading to a different collective performance such as competition for DNA uptake, production of various virulence factors, biofilm production, and development of different antiphage protection defense mechanisms. Bacteria often collect signals from vary auto triggers, which establish communication within and between species and genera, acyl-homoserine lactones (AHLs) well known AIs. These AHLs are naturally synthesized by LuxI-type enzymes and identified by LuxR-type receptor-transcription factors inside the cytoplasm (Jiahui Li 2020). The objective of this review to evaluate the effect of sonication on the quorum sensing and their role in biofilm structure.

Biofilm Structure and Clinical Implications

Biofilms as protective, structured communities of microorganisms encased in an extracellular polymeric substance (EPS) matrix.

Basically, Bacteria appear either in planktonic cells, which is highly sensitive to surrounding condition and antimicrobial agents; or biofilms, allow profile biological activity form biofilm providing better guard environment for resistant to different antimicrobial and disinfectants substance. Simply Biofilms are extracellular materials which allow the bacterial cells to strictly attached to the surfaces, the external surface composed of peptide, carbohydrates, and/or eDNA. The extracellular form due to a series of harsh condition that enhance the response to form the biofilm for example lack nutrition factors in the surrounding or exposure to subinhibitory levels of antibiotics or disinfectants (Zhao, Sun *et al.*, 2023).



During the transitions to biofilm growth, bacteria experience a phenotypical changes with various genetic factors being differentially organized. For instance, in *Escherichia coli* formation of biofilm involved modified expression of more than 200 genes, mostly those responsible for attachment, auto-aggregation, outer membrane proteins (OmpC, OmpF, OmpT, and Slp), and lipid A biosynthesis (Xiaoling Wang 2022). (Saipriya K 2019) investigated that in *Acinetobacter baumannii* biofilms, cans significantly affected by iron that subsequently has a great impact on efflux pump and quorum sensing.

Biofilm initiated when bacterial cells communicated between the genera and species or with surface. Primarily, free bacteria attached to the surface through weak, adjustable forces. As the cells aggregate, they produce an extracellular matrix that encases them, promoting communication through biochemical molecules which a clear as a signal which enabling genetic exchange within the community (Zhao A 2023).

The extracellular material of the biofilm mainly consists of DNA, RNA, and proteins, which consider nearly 2% of the biofilm's total mass. Dispersion is the final stage of biofilm development, where microcolonies experience cell death, allowing motile bacteria to actively disperse and leave behind hollow colonies. This process enables the bacteria to separately move from the biofilm and create new biofilm microcolonies in other environments. Biofilms are regulating a balance of growth and dispersal, which can occur either as single cells or small microcolonies (Campoccia, Montanaro *et al*., 2021). Biofilm structure is significantly different based on the organisms and the surrounding environmental conditions.

Stages of biofilm initiated with the hydrophobicity and electrokinetic potential of bacterial cells are influenced by their surface composition and structure. In Gram-negative bacteria, the presence of lipopolysaccharides can impact their ability to form biofilms. Increased hydrophobicity in bacteria reduces the repulsion between the bacterial cell and the extracellular matrix, facilitating biofilm formation (Flemming 2019).

Biofilms are considered as a more than 100 times resistance to antibiotics than the planktonic free bacteria, rendering a simple infection difficult to treat. To achieve an effective cure for any infection a higher antibiotic doses, combinations of antibiotics and natural products, or removal of foreign bodies in device-related infections are required. The main reason the insignificant response for the biofilm treatment using the antibiotics alone due to the inadequate antibiotic diffusion, nutrient limitations, slow bacterial growth, stress responses, and the formation of resist cells. The biofilm matrix plays a vital role in slowing antibiotic penetration by reacting with or binding to antibiotics. However, biofilm tolerance differs from antibiotic resistance, as bacteria become more susceptible once the biofilm is disrupted (Uruén C 2020).

Introduction to Ultrasound in Biofilm Management

Ultrasound can be defined as pressure waves with a frequency of 20 kHz or higher. Typically, ultrasound devices operate within the frequency range of 20 kHz to 10 MHz. ultrasound with low frequency has a higher power (20 to 100 kHz), often known "power ultrasound," can produce cavitation bubbles filled with high mechanical, shear force and free radicals, which make it more effective in food processing applications, medical field, and environmental application for microbial inactivation (H.A., H. Z. *et al*., 2020).

Ultrasonic transducers have several capabilities of generating ultrasonic waves, along with their equipment and specific functions, Growing concern about microorganism removal is reported, ultrasound represent as an effective microbial inactivation is essential in food processing. Even a single contamination report can significantly harm a manufacturer's reputation and long-term viability. To achieve a total microbial free food product, producers require to adopt inactivate existing microorganisms, and establish processes to prevent or slow the re-growth of any non-inactivated microbial populations. Ultrasonic wave gives a hopeful method for microbial prevention (Ali, Al-Haideri *et al*., 2022).

Ultrasound radiation act on cell membrane penetrating, localized heating, and free radical production (Karthikesh MS 2021) In this process, sonic waves produce and excessive of compression and expansion waves in a liquid to create a small bubbles medium (cavitation). This cavitation filled with gas subsequent expansion until the ultrasonic energy cannot equilibrium within all the vapor inside leading to bubble collapse, resulting in high temp. reach more than 5000˚ C highly shear force and pressures (up to 50,000 kPa), which directly inactivate the bacterial cells (Ali, Al-Haideri *et al*., 2022).

Formation of the cavitation bubble within the liquid medium depends on many factors such as dissolved gas, hydrostatic pressure, heat, and tensile strength (Sherif S. Rashwan 2021).

According to Chuah *et al*., [2], ultrasound is classified by frequency into high-frequency ultrasound (HFU) at 1000–10,000 kHz and low-frequency ultrasound (LFU) at 20–100 kHz. While high frequency ultrasound is primarily used in medical applications, its unique properties have expanded its use across various fields,

1. **Biological Effects**: Ultrasound can interrupt biological structures, such as algal cell membranes, enabling cellular component extraction
2. **Physical Effects**: Ultrasound's physical impacts include applications like ultrasonic cleaning, emulsification, and atomization. In atomization, micro-droplets are emitted once the acoustic intensity surpasses a liquid-specific threshold (E., A. *et al.*, 2011)
3. **Chemical Effects**: Ultrasound can produce highly reactive free radicals, including OH•, HO2•, and H•, through the implosion of bubbles created during irradiation, contributing to its chemical effects

Application of Ultrasound radiation has demonstrated a significant benefit throughout various field, such as in water treatment plant and degradation of many endocrine disruptor pollutants (Karthikesh MS 2021). The sonication Parameters including frequency, power operation mode , vibration influence, the intensity of the ultrasonic treatment and treatment time has a great impact on bacterial inactivation, for example the lower frequency has a direct role in the bubble size, which allow to produce a large number of bubbles and stronger shear forces, while high-frequency ultrasound yields smaller, more stable bubbles (stable cavitation) (Sherif S. Rashwan 2021). At lower frequencies, bubble implosions are more intense; higher frequencies generate more cavitation bubbles but with less collapse intensity, requiring greater power for active bubble formation (Ko and Bai 2022).

Ultrasound disrupts QS signals, impacting biofilm development and enhancing antimicrobial penetration.

Quorum sensing (QS) known as a controlling method that bacteria employe to accommodate to the surrounding harsh condition via enhance the expression of numerous genes in a population-reliant on [9]. During the QS bacteria can controlling a several biological function such as metabolism, protein synthesis, virulence factor expression, antibiotic resistance, biofilm formation, maintenance, dispersal, and the transition to stationary phase, which making it a hopeful target for substitute antimicrobial approaches (Saipriya K 2019).

Mechanisms of Quorum Sensing in Biofilm Formation

It has been reported that biofilm is controlled by the quorum sensing (QS) system. QS enable communication among bacteria through tiny biochemical molecules known as autoinducers (AIs) (Sahreen S 2022). By the time the population density increase resulting to rather increase these signaling molecules level in the environment, influence the binding to receptor proteins, triggering the expression of genes involved in biofilm formation (Williams 2007).

It is well established there is a various QS disrupting agents, including QS inhibitors (QSIs) and quorum-quenching (QQ) enzymes, disrupt this cell communication through various mechanisms, thereby preventing biofilm formation (Sutherland 2001). Additionally, QS-different QS inhibiting approaches can significantly enhance bacterial susceptibility to antibiotics (Ozcan *et al*., 2019). Consequently**,** employing QS-inhibiting agents offers a hopeful strategy for controlling bacterial infections. QS systems are classified according to the classification of the autoinducers (AIs) they use: the AHL and AIP systems

The auto induced system vary between the gram negative and positive bacteria. The Gram-negative bacteria, uses N-acyl homoserine lactones (AHLs) as signaling molecules (Papenfort K 2016). While, the AIP system, which uses autoinducing peptides (AIPs), is specific to Gram-positive bacteria (AI-2 and AI-3 systems, though less studied, are found in both Gram-positive and Gram-negative bacteria and facilitate interspecies signaling. AI-2 involves furanosyl borate

diesters derived from DPD (Kai Papenfort 1 2016), while AI-3 molecules, recently identified as pyrethroids, are also part of interspecies signaling. The specific mechanisms by which QS regulates biofilm formation differ between Gram-negative and Gram-positive bacteria(Majdura *et al*., 2023) (Ng & Bassler, 2009) (Costa TR 2015).

QS-regulated genes involved in EPS production, virulence factor expression, and biofilm maturation

Quorum sensing (QS) is a crucial regulatory mechanism that controls gene expression associated with extracellular polymeric substance (EPS) production and bacterial virulence. QS-driven EPS synthesis plays a fundamental role in biofilm formation, ensuring structural stability and protection against environmental challenges (Flemming *et al*., 2016). Key QS-regulated genes involved in EPS production encode enzymes responsible for polysaccharide biosynthesis and transport. Furthermore, QS modulates the expression of various virulence factors, including proteases, toxins, and hemolysins, which facilitate host colonization, immune evasion, and tissue damage (Ng & Bassler, 2009). In *Pseudomonas aeruginosa*, QS systems such as Las and Rhl regulate genes linked to alginate biosynthesis, pyocyanin production, and elastase activity, all of which contribute to its pathogenicity. Investigating QS-regulated genes enhances our understanding of bacterial survival mechanisms and provides potential targets for anti-virulence therapeutic strategies.

Quorum Sensing in Biofilm Maturation and Antibiotic Resistance

QS-regulated Genes Involved in EPS Production, Virulence Factor Expression, and Biofilm Maturation

Quorum sensing plays a crucial role in controlling the genes responsible for extracellular polymeric substance (EPS) production and virulence factor expression in bacteria. EPS production, governed by QS, is essential for biofilm formation, providing structural integrity and protection against environmental stresses (Frederick *et al.,* 2011). Key QS-regulated genes in EPS synthesis include enzymes encoding for polysaccharide biosynthesis and transport. Additionally, QS regulates the expression of other virulence factors, such as the secretion of enzymes like proteases, toxins, and hemolysins, which facilitate host colonization, immune evasion, and tissue damage. For instance, in *Pseudomonas aeruginosa*, QS systems like Las and Rhl regulate genes involved in alginate production, pyocyanin synthesis, and elastase expression, all contributing to its pathogenicity. Understanding QS-regulated genes provides insights into bacterial survival strategies and offers targets for anti-virulence therapies (Haidar *et al*., 2023).

Targeting QS provides promising methods for biofilm removal. Recently a new term appeared for QS obstruction approaches, known as (quorum quenching), involve interfering with chemical signal production, damaging signaling molecules, or blocking receptor binding, thereby halting biofilm formation and maintenance. These can be achieved through various strategies, such as using inhibitors for chemical signals, enzymes that degrade autoinducers, and artificial analogs that competitively bind QS receptors. By blocking QS signaling, bacteria within the biofilm matrix become more accessible to treatments and immune clearance. Recently, these methods have emerged as a novel avenue for addressing biofilm-related infections and antibiotic resistance, emphasizing the therapeutic potential of QS disruption (Zhao, Sun *et al*., 2023).

Ultrasound as a Modulator of Quorum Sensing and Biofilm Structure

Ultrasound is emerging as a promising tool for modulating quorum sensing and disrupting biofilm structures in microbial communities. The physical and chemical effects of ultrasound on microbial cells primarily arise from acoustic cavitation, which occurs when low-frequency ultrasound generates oscillating bubbles in a liquid medium (Xia *et al*., 2025). The violent collapse of these bubbles produces localized shock waves, high shear forces, and microjets that disrupt bacterial cells and their environment. Cavitation can physically damage bacterial cell membranes, leading to increased permeability and impaired cellular function. Additionally, the chemical effects of ultrasound, such as the generation of reactive oxygen species (ROS), further contribute to bacterial stress and disruption of QS signaling pathways (Yousf *et al*., 2016).

QS molecules, which mediate bacterial communication and regulate biofilm formation, are particularly susceptible to ultrasound-induced effects. Cavitation can degrade QS signaling molecules, such as AHLs in Gram-negative bacteria or peptides in Gram-positive bacteria, disrupting their ability to coordinate communal behaviors. Moreover, ultrasound can interfere with bacterial communication channels, preventing the expression of genes critical for biofilm assembly and maintenance. Studies have shown that ultrasound exposure reduces the density and structural integrity of biofilms by disrupting QS-regulated processes like EPS production and surface adhesion. This dual effect—physical disruption of biofilm architecture and chemical inhibition of QS signaling—underscores the potential of ultrasound as a non-invasive and efficient strategy to combat biofilm-associated infections, particularly in medical and industrial settings (Papenfort K 2016).

Impact of Ultrasound on QS Signal Production and Accumulation

Ultrasound significantly impacts QS signal production and accumulation, making it a valuable tool for disrupting bacterial communication in biofilm-forming bacteria. Studies have shown that ultrasound exposure can reduce the concentration of QS molecules, such as AHLs, which play a central role in coordinating biofilm formation and virulence in Gram-negative bacteria. This reduction may result from the physical effects of acoustic cavitation, including shear forces

and shock waves, which can degrade QS molecules in the extracellular environment. Additionally, ultrasound-generated reactive oxygen species (ROS) may chemically alter QS signals, further diminishing their efficacy in microbial communication (Jiahui Li 2020).

By disrupting QS molecule synthesis or reception, ultrasound can downregulate the expression of biofilm-promoting genes. QS systems typically regulate genes involved in processes such as EPS production, adhesion, and resistance to antimicrobials. When QS signaling is impaired, biofilm formation is hindered, and bacterial communities become less organized and more vulnerable to external stressors, including antibiotics and host immune responses. This dual impact of ultrasound—on both QS molecule levels and the regulatory pathways they control—highlights its potential as a non-invasive strategy to inhibit biofilm development and mitigate bacterial pathogenicity in clinical and industrial applications (Khadem, Tirtouil *et al*., 2020).

Ultrasound techniques have been developed as antimicrobial tools, either alone or in combination with other antimicrobial strategies. A promising method for enhancing efficiency and effectiveness is the combined application of low-frequency ultrasound with different treatment approaches to achieve biofilm disruption. For example, using low-frequency ultrasound with photodynamic therapy (aPDT) has shown promising results. The biological effects of ultrasound include thermal, mechanical, cavitation, and chemical effects. It is well known that the depth of invasion of microorganisms into dentinal tubules is much deeper than that of photosensitizer (PS) injection. Some investigations have successfully applied ultrasound techniques to activate PS, increasing penetration depth into dentinal tubules and enhancing antimicrobial efficacy (Faina Nakonechny Marina Nisnevitch 2021).

However, specific PS can be excited by ultrasound and light, leading to the concept of sonosensitizers. This new approach combines ultrasound and light to activate PS, referred to as sonophotodynamic therapy (SPDT). Research has explored the antifungal activity of aPDT, sonodynamic therapy (SDT), and SPDT in planktonic C. albicans suspension and biofilm. The study found that while the suspension was eradicated, the biofilm was minimally affected by aPDT or SDT application. However, SPDT treatment significantly reduced biofilm biomass and viability using photodithazine. In another study, the authors found that the reduction of S. aureus induced by Cur-mediated aPDT increased from 2.12 log10 to 4.33 log10 after activation with ultrasonic waves. Although invitro efficacy has been validated in published studies, the exact mechanisms underlying the enhancement effect of ultrasound on aPDT require further investigation.

Several hypotheses account for the possible mechanisms of the potentiated effect due to ultrasound. Studies have shown that ultrasound can generate transient pores in the cell membranes of microorganisms, enabling bacteria to absorb more photosensitizers. Additionally, ultrasound waves promote the circulation of microorganisms in the medium, making them more directly accessible to light. Currently, there are no SPDT treatments specifically targeting oral microorganisms or biofilms. The SPDT procedure shares features with aPDT, whose effects depend on frequency and energy. Future studies should focus on expanding the scope of SPDT and optimizing its parameters (Xu, Lu *et al*., 2020).

Research has shown that low-intensity ultrasound at physiotherapy levels can enhance the activity of antibacterial agents, resulting in a powerful biofilm removal effect. Moreover, ultrasound or ultrasound-targeted microbubble destruction (UTMD) can disrupt the matrix structure of bacterial biofilms by using cavitation to promote drug entry into the biofilm and can also directly destroy cells, increasing the metabolic activity of cells to enhance the bactericidal ability of antibiotics. These findings provide new insights into using ultrasound or UTMD to improve the effectiveness of antibiotic therapy for prosthesis- or catheter-related biofilms, with promising effects observed in vivo.

Applications for Ultrasound in Biofilm Control

Medical Applications

As technological advances continue to push for innovative solutions to pressing health concerns, attention must also be given to roadblocks preventing further improvements. One of these obstacles are biofilms: potent invaders that continue to negatively impact wounds, medical devices, and patient health (Xu, Lu *et al*., 2020).

Given the consequences and prevalence of biofilm infections, it is important to keep up to date with research and to push for the advancement and application of novel treatments. The purpose of this review is to provide a brief background on biofilms, and implants, and advance approach for treatment.

Chronic and acute wounds are at risk of biofilm formation due to penetration of the protective dermal layers, the body's first nonspecific line of defense, and disruption of the innate immune system. Exposure to microorganisms living on the skin or other external sources can greatly impede wound healing if biofilms develop . In fact, Attinger and Wolcott suggest that 90% of chronic wounds and 6% of acute wounds are composed of bacteria within biofilms. This is concerning considering that chronic wounds affect 2% of the population in the United States (Yosman Dhar ∗ 2020).

There are some downsides to the current SOC as treatments are often not enough to eradicate all the bacterial colonies. In fact, biofilms have been shown to regain antibiotic resistance 72 h after debridement, suggesting reduced efficacy of treatment and delayed wound healing (Ko and Bai 2022).

Synergy with Antimicrobial Agents

Ultrasound debridement can treat biofilm-infected wounds, and has been reported to increase the effectiveness of antibiotics and promote wound healing. Recent studies on ultrasound for biofilm infections have focused on its use in chronic wounds. Studies on patients with diabetic foot ulcers and lower-extremity wounds reported decreased bacterial count, improved wound tissue, and reduction in wound size after ultrasound debridement (C.A. Murphy 2018). One study on patients with venous leg ulcers found that patients treated with ultrasound debridement ($N = 36$) had fewer treatments and healed faster than patients treated with sharp debridement ($N = 40$). This could argue for the use of ultrasound over sharp debridement, given that ultrasound is less painful for the patient. According to a review by Chang *et al*., which analyzes its use in chronic wounds, ultrasound debridement was most effective if applied three times a week (R. Chang and 2017).

Ultrasound may also be used to treat and increase the effectiveness of antibiotics in implant and implant-related biofilm infections. A study by Granick *et al*., which used metal discs made to mimic titanium and stainless steel implants, found that direct mid-level ultrasound exposure cleared *Staphylococcus epidermidis*, while any viable bacteria released from the biofilm was eradicated by a hypochlorous acid irritant (Hameister 2018). Ultrasound sonication was able to detect and eradicate biofilms in periprosthetic joint infections as well, but Hameister *et al*., suggest that it is only effective when used in conjunction with antibiotics (Hameister 2018).

Ultrasound-based treatments may be used to treat wounds and implants infected with biofilms; however, more clinical research should be considered for its large-scale implementation in a healthcare setting. Ultrasound debridement demonstrates great potential in wound care as it can be less painful than sharp debridement, leading to greater patient compliance and comfort. Studies comparing patients treated with ultrasound, sharp debridement, and other standards of care would be beneficial. Scholars should also examine factors such as pain, duration, ease of use, and include a follow-up period to determine wound progress and patient health (Cai 2017).

As mentioned earlier, ultrasound has been used with other conventional treatments such as antibiotics (urrent developments in biofilm treatments: Wound and implant infections.

Author links open overlay panel (Yosman Dhar and Yangha Han 2020). Beyond these treatments, however, ultrasound is now being examined with drug-delivery systems for greater biofilm eradication; for example, one study by Guo *et al*., found that acoustically-activated nanodroplets with vancomycin decreased *Staphylococcus aureus* biofilm viability and metabolic activity (H. Guo 2017). Within this area of combinational therapies, there has been a growing interest in the use of microbubbles.

Microbubbles are gaseous cores surrounded by stabilizing shells that can be acoustically activated to deliver drugs and to mechanically disrupt biofilms. Their recent emphasis in research can be attributed to their adaptable composition, size, and fabrication, which can control their reactiveness in the body (J. Owen 2018). Research on microbubbles appears to be primarily directed towards cancer therapies; however, studies on their application in biofilms are increasing. For more information about previous studies a review by LuTheryan *et al*., (2020) provides a comprehensive analysis of ultrasound mediated therapies, with detailed information related to microbubbles (G. LuTheryn 2020).

Challenges and Limitations

The use of ultrasound as a tool to disrupt quorum sensing (QS) and biofilm integrity is not without its challenges and limitations, particularly in terms of technical and practical constraints. One major challenge lies in the variability of bacterial responses to ultrasound, which depends heavily on parameters such as frequency, duration, and intensity. For instance, lower frequencies are typically more effective at generating cavitation, but they may also cause unintended damage to surrounding tissues or surfaces in clinical settings (Perez *et al*., 2022). Additionally, the optimal parameters for one bacterial species or biofilm type may not be effective for another, as differences in biofilm composition, thickness, and matrix density can influence susceptibility to ultrasound-induced disruption.

Another significant limitation is the difficulty of standardizing ultrasound applications across various biofilm types and clinical contexts. Biofilms in industrial pipelines, medical devices, or chronic infections often differ substantially in their physical and biological properties, making it challenging to design a one-size-fits-all ultrasound treatment. Moreover, achieving uniform ultrasound exposure in complex environments, such as within the human body or on irregular surfaces, adds another layer of complexity. Ensuring safety and efficacy in clinical applications also requires careful consideration to avoid adverse effects, such as overheating or collateral tissue damage. Addressing these challenges will

require further research to refine ultrasound protocols, optimize treatment parameters, and develop specialized equipment tailored for specific applications and microbial targets (Sue *et al*., 2022).

The application of ultrasound to disrupt quorum sensing (QS) and biofilm structures must address potential adverse effects, particularly in clinical and therapeutic settings. One major concern is the risk of collateral tissue damage or cytotoxicity in host cells at higher ultrasound intensities. Jiang *et al*., (2022) highlighted that intense ultrasound exposure can generate excessive heat, mechanical stress, and reactive oxygen species (ROS), which may inadvertently harm surrounding tissues or impair normal cellular functions. Such effects are especially critical when applying ultrasound near sensitive areas, such as internal organs or implanted medical devices, where unintended damage could exacerbate patient conditions. To mitigate these risks, optimizing ultrasound parameters, including intensity, frequency, and exposure duration, is essential to achieve effective QS attenuation without compromising safety. Balancing the mechanical disruption of biofilms and QS signaling with minimal adverse effects requires a tailored approach, accounting for the biofilm's location, composition, and host environment. Furthermore, integrating real-time monitoring systems to track ultrasound effects could enhance precision and reduce unintended harm. Continued research is necessary to refine these techniques, ensuring that ultrasound-based interventions are both safe and effective for managing biofilm-associated infections in clinical practice.

CONCLUSION

In conclusion, ultrasound has emerged as a promising and versatile tool for modulating quorum sensing (QS) and disrupting biofilm structures, offering significant potential for microbial control in clinical and industrial settings. Key findings from current research highlight ultrasound's ability to attenuate QS signaling, particularly through the degradation of quorum sensing molecules like acyl-homoserine lactones (AHLs), and by interfering with the synthesis and reception of QS signals. This disruption impairs biofilm formation, reduces virulence factor expression, and enhances bacterial susceptibility to antimicrobial agents, demonstrating its efficacy in targeting biofilm-associated infections.

- Ultrasound’s impact on biofilms extends beyond QS modulation, as it can physically disrupt biofilm architecture through cavitation, shear forces, and the generation of reactive oxygen species. The combination of physical disruption and chemical QS interference provides a multifaceted approach to biofilm management, which is crucial for addressing the challenges posed by biofilm-related antibiotic resistance. Moreover, ultrasound’s integration with novel technologies, such as ultrasound-responsive nanoparticles, further enhances its potential as an effective antimicrobial strategy.
- While ultrasound shows promise, there remain several areas for future research, including a deeper molecular understanding of its effects on QS pathways and the development of species-specific ultrasound protocols. Ultimately, advances in ultrasound technology and its integration with antimicrobial delivery systems will enhance its applicability for managing biofilms, opening new avenues for non-invasive treatments in diverse clinical

Conflict of Interest: Author has no conflict of interest**.**

Acknowledgement: Author would like to acknowledge the mustansiriyah collage of sciences/ biology department for their support.

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