The Role of Juniperus Communis Essential Oil in Novel Nanofiber Bandages and Antimicrobial Resistance

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Infectious diseases remain a significant cause of morbidity and mortality worldwide, particularly in low- and middle-income countries. The increasing resistance to antimicrobial drugs poses a growing challenge to the treatment of these infections. The World Health Organization (WHO) has identified antimicrobial resistance (AMR) as one of the top ten global public health threats. AMR is associated with high mortality rates, increased healthcare costs, and reduced treatment efficacy, primarily due to the misuse and overuse of antimicrobial drugs. The crisis is particularly concerning in nosocomial infections caused by the ESKAPE pathogens: *Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Enterobacter cloacae*. As antimicrobial therapies become increasingly ineffective, the urgency to develop alternative treatments has intensified.

The alarming rise in AMR has revived the search for plant-derived antimicrobial therapies. Essential oils (EOs) extracted from various plants have long been recognized for their antimicrobial and antibiofilm properties, contributing to symptom alleviation and bacterial growth inhibition. In addition to their antimicrobial effects, EOs have been successfully used to treat conditions such as chronic pain, pediculosis in children, and postoperative nausea in cancer patients. Biofilm formation on medical devices such as catheters, ventilators, and contact lenses presents a significant challenge in clinical settings, further complicating the treatment of infections. Given the rising resistance of bacterial biofilms to conventional antimicrobials, natural therapeutic agents capable of eradicating biofilm-forming pathogens have become necessary.

A pioneering research titled “Antibacterial and Antibiofilm Activity of Novel Nanofiber Bandages Formulated with *Juniperus communis* Essential Oil Targeting Antibiotic-Resistant Bacterial Strains” was conducted to investigate the potential of *Juniperus communis* essential oil (JEO)-infused nanofiber bandages in combating AMR. This research, conducted by Mohannad Abu Haweeleh and colleagues under the supervision of Dr. Susu Zughaier, Associate Professor of Microbiology and Immunology at Qatar University, was presented at the American College of Surgeons’ Clinical Congress in Boston, where it secured second place in the Basic Science category.

The research focused on developing polycaprolactone (PCL) nanofiber bandages infused with JEO to assess their antibacterial, antibiofilm, and immune-modulating properties against ESKAPE pathogens. The research involved collaboration with Dr. MD Anwarul Hasan, Associate Professor of Mechanical and Industrial Engineering at Qatar University, for nanofiber formulation and with Dr. Nahla Eltai, Research Associate at the Biomedical Research Center, for bacterial testing.

The *Juniperus communis* plant is widely distributed across Europe, Asia, and North America and possesses various medicinal properties, including diuretic, anti-inflammatory, antiseptic, antioxidant, and hypoglycemic effects. Different parts of the plant have demonstrated efficacy against various diseases. Studies have shown that methanolic extracts from *J. communis* exhibit dose-dependent analgesic effects, while its EO has demonstrated effectiveness in water disinfection against *Mycobacterium* species. Given its antimicrobial properties, JEO holds potential for integration into nonwoven polymer nanofibers for use in treating surgical site infections (SSIs) and skin ulcers.

The antibacterial efficacy of JEO was tested using bacterial growth curves, microtiter dilution assays, and agar diffusion methods. Additionally, its antibiofilm activity was evaluated through biofilm formation assays in 96-well plates, followed by staining and quantification using crystal violet assays. The findings revealed that JEO exhibited significant antibacterial and antibiofilm activities against ESKAPE pathogens. Growth curve analyses showed dose-dependent inhibition of bacterial proliferation, while bactericidal assays confirmed the oil's ability to eliminate bacterial colonies. The antibiofilm assays demonstrated a substantial reduction in biofilm formation in the presence of JEO, highlighting its potential to prevent infections related to medical devices. The research also investigated the anti-inflammatory effects of JEO on human monocyte (THP-1) and murine macrophage (RAW 264.7) cell lines. The results indicated that JEO modulated immune responses by reducing pro-inflammatory cytokine release, suggesting its potential role in mitigating excessive inflammation in infected wounds.

To enhance the clinical applicability of JEO, PCL-based nanofiber membranes were developed with varying concentrations of the EO (2%, 4%, 6%, and 8%). The antibacterial efficacy of these membranes was evaluated using bacterial inhibition zone assays and broth-based viability assessments. The results confirmed that nanofiber membranes infused with higher concentrations of JEO exhibited stronger antibacterial effects, effectively inhibiting bacterial growth and reducing biofilm formation. To further validate the potential of JEO-infused nanofiber bandages, bacterial strains were cultured in the presence of the membranes, and their viability was assessed at multiple time points. The results consistently demonstrated a reduction in bacterial colony-forming units, indicating sustained antimicrobial activity over time **(Figure 1).** These findings support the potential use of JEO-infused nanofibers in clinical settings for preventing and treating infections caused by antibiotic-resistant bacteria.

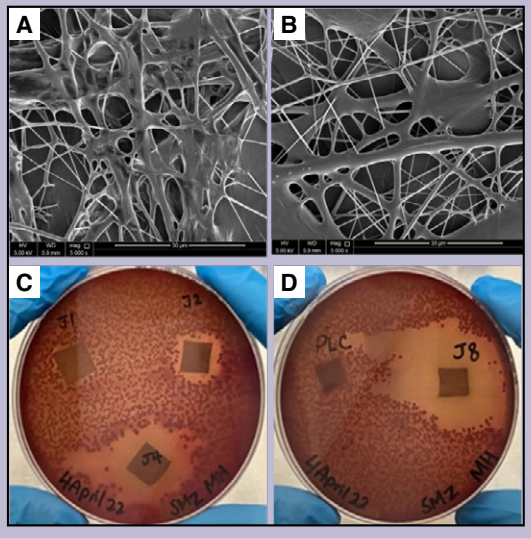

Figure 1: Scanning electron microscope (SEM) images demonstrate that incorporation of 8% Juniper essential oil (JEO) does not affect the morphology or porosity of polycaprolactone (PCL) nanofibers. **A)** PCL nanofibers without Juniper EO. **B)** PCL nanofibers with 8% Juniper EO. The antibacterial activity against methicillin-resistant Staphylococcus aureus (MRSA) was evaluated using sheep blood agar for bacterial growth, with zones of inhibition measured for nanofibers containing varying concentrations of Juniper EO (0%, 1%, 2%, 4%, and 8%). **C)** Inhibition zones for PCL nanofibers with 1%, 2%, and 4% Juniper EO. **D)** Inhibition zones for PCL nanofibers with 0% (control) and 8% Juniper EO.

AMR is an escalating public health concern, necessitating the urgent development of alternative therapeutic strategies. Research on JEO-infused nanofiber bandages highlights the potential of plant-based antimicrobials in addressing this challenge. By integrating the natural antimicrobial properties of JEO with advanced nanofiber technology, this innovative approach presents a promising strategy for managing antibiotic-resistant infections. Given the growing impact of AMR on global healthcare, plant-based antimicrobial solutions offer a viable alternative. This research underscores the potential of JEO-infused nanofiber bandages in preventing hospital-acquired infections and reducing reliance on conventional antibiotics. Future research should focus on optimizing JEO-infused nanofiber formulations, assessing their long-term stability, and conducting in vivo studies to evaluate their clinical efficacy.

The success of this research highlights the critical role of interdisciplinary collaboration in addressing AMR. Equipping future physicians with expertise in antimicrobial stewardship and translational research is essential for developing effective strategies to combat this global challenge. This research study aligns with Qatar's national priorities for biomedical innovation, emphasizing the pivotal role of physician-researchers in driving advancements in global healthcare solutions. The research is currently under review in the Chinese Herbal Medicines Journal, published by Elsevier.

Mohannad Abu Haweeleh (center) achieved second place at the American College of Surgeons Clinical Congress 2023.