Creating Antibiofilm and Antibacterial Endotracheal Tubes Using Antimicrobial Peptides and Antibiotics

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Introduction

- Endotracheal tubes are a major site of microbial contamination, where bacteria rapidly adhere to PVC surfaces and initiate biofilm formation, contributing to airway infections during mechanical ventilation.
- Biofilms on PVC medical devices significantly reduce antibiotic effectiveness, enabling pathogens to survive, spread, and contribute to antimicrobial resistance in critical-care settings.
- Surface modification of PVC offers a promising strategy to prevent bacterial attachment, using chemical grafting methods (e.g., dopamine, diazonium chemistry) to introduce reactive groups for antimicrobial attachment.
- Antimicrobial peptides and antibiotics[1] can be immobilised onto modified PVC surfaces, creating coatings with potential antibiofilm and antibacterial activity while maintaining material integrity.

➢ Aims and Objectives

This study aimed to develop a surface-based strategy to reduce biofilm-associated antimicrobial resistance by attaching antimicrobial peptides and antibiotics to modified PVC endotracheal tube materials, and to determine surface modifications that influence antibiofilm efficacy against *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

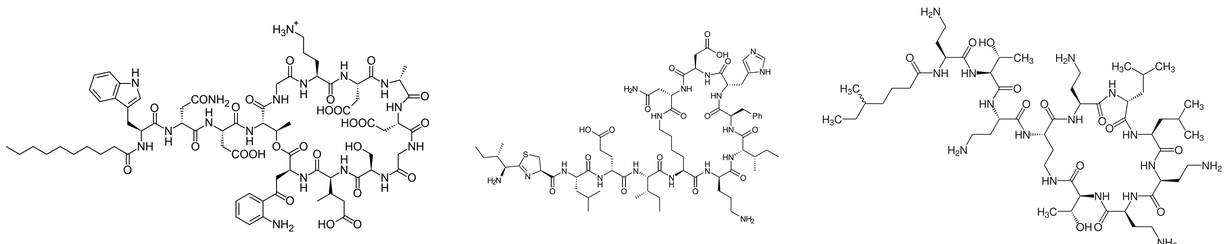


Figure 1: Chemical structures of the peptide antibiotics used in this study, including Daptomycin, Bacitracin, and Colistin.

Methods

a) **Samples & Pretreatment**: PVC discs (8 × 8 mm; 1 mm thick) were cut from medical-grade sheets, sonicated in dH_2O (3 × 5 min, 37 kHz), air-dried, and placed in sterile 24-well plates**.**

b) **Surface Modifications: i)** Polydopamine introduced amine groups. **ii)** Triple polydopamine (PDA) assembly (3 cycles of PDA), and **iii)** Diazonium chemistry: 4-carboxybenzenediazonium tetrafluoroborate was reduced with H_3PO_2/HBF_4 to graft aryl radicals, followed by NHS/EDC activation.

c) **Peptide & Antibiotic Attachment:** Ciprofloxacin was prepared at 10 mg·mL^{-1} in acetone: H_2O with HCl, and peptide solutions (Daptomycin, Bacitracin, Colistin) were prepared at 40 µg·mL^{-1}. Modified discs were immersed in peptide and antibiotic solutions for 24 h, rinsed, and dried.

d) **Surface Analysis: XPS and FT-IR** were used to confirm the introduction of functional groups and antimicrobial attachment. **Contact angle:** The sessile drop method was used to assess wettability**. Antimicrobial Assays:** Biofilm activity (TTC extraction) and planktonic growth (OD_{600}) were measured using a modified BOAT assay to assess antimicrobial efficacy.[2]

e) **Data Processing: Contact angle**: Welch's ANOVA with Dunnett's T3 post-hoc test (n = 9; α = 0.05). **Fold-change (OD_{600}):** One-way ANOVA with Dunnett's T3 post-hoc test (n = 3; α = 0.05).

Results and Discussion

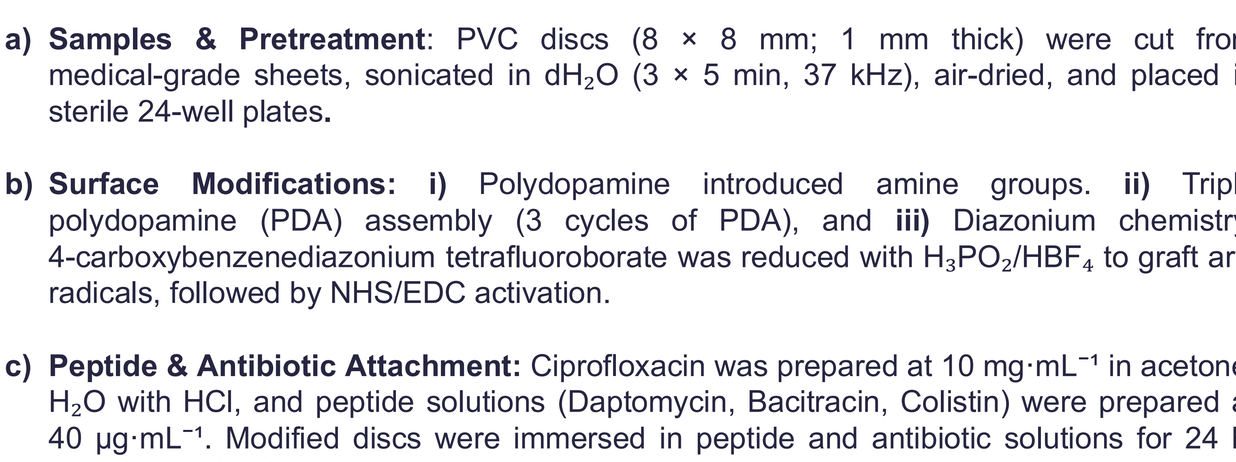


Figure 2: FTIR spectra of chemically modified PVC surfaces. (A) Dopamine-treated PVC: dopamine introduces aromatic and O–H/N–H features, while daptomycin adds amide I/II bands confirming peptide attachment. **(B) Diazonium-modified PVC**: diazonium grafting introduces new aromatic peaks, and subsequent daptomycin coupling produces characteristic amide bands, indicating successful peptide attachment.

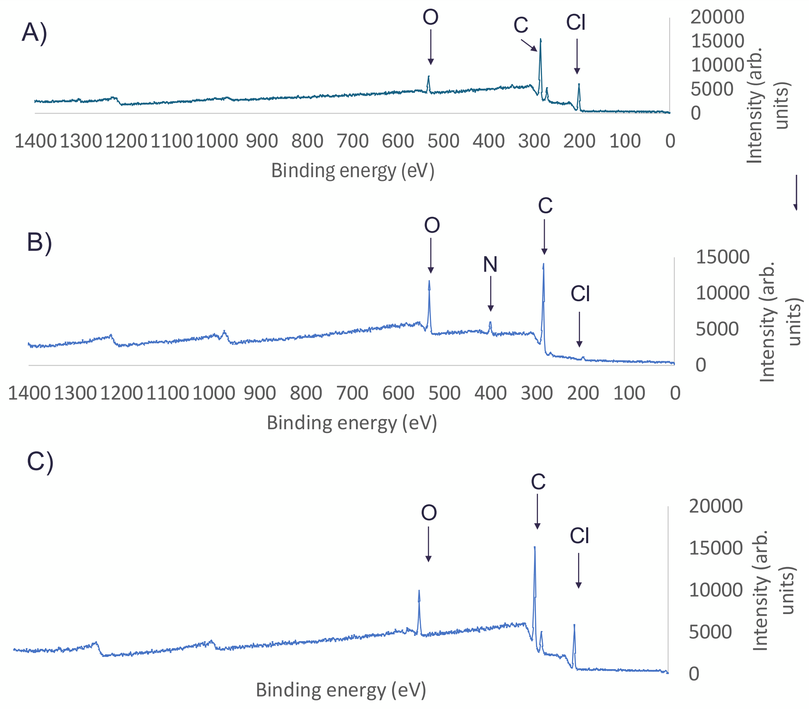


Figure 3: XPS survey spectra of PVC surfaces. A) Uncoated PVC showing characteristic carbon (C), oxygen (O), and Chlorine (Cl) peaks of the native polymer surface. **B) Polydopamine-coated PVC** displaying C, O, Cl, and an additional nitrogen (N) peak, confirming successful PDA deposition. **C) Diazonium-modified PVC** retaining the core PVC peaks: O, C and Cl.

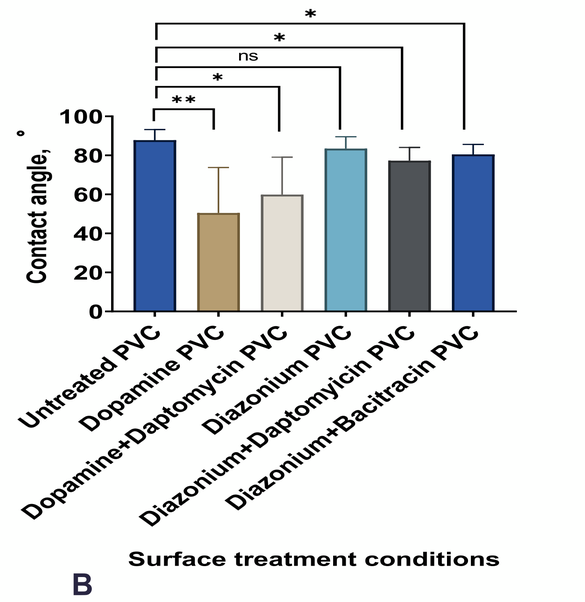


Figure 4: Contact angle measurements of PVC surfaces under different chemical treatments. Mean contact angles (n = 9) for untreated PVC, dopamine PVC, dopamine + daptomycin PVC, diazonium PVC, diazonium + daptomycin PVC, and diazonium + bacitracin PVC. Statistical analysis was performed using Welch's ANOVA with Dunnett's T3 post-hoc test (α = 0.05). * indicates significant differences compared with untreated PVC; "ns" denotes non-significant differences.

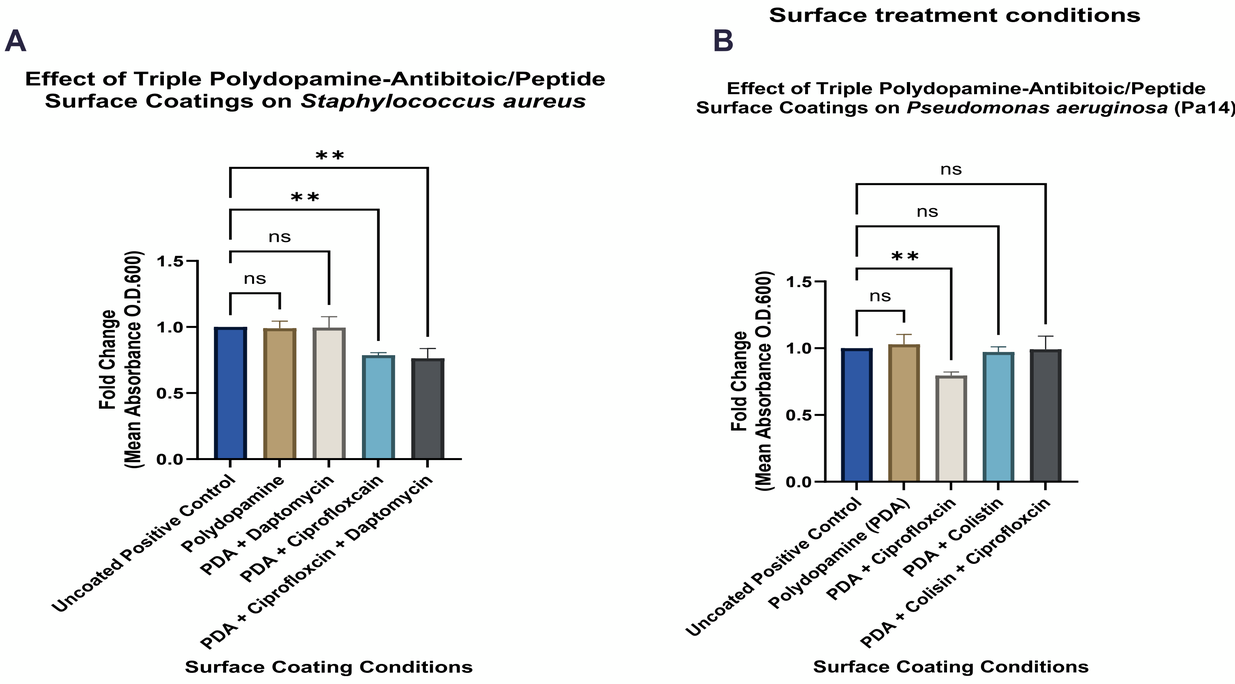


Figure 5: Effect of 3-fold polydopamine (PDA) coatings with antibiotics on biofilm formation. **(A)** ***S. aureus***: PDA + ciprofloxacin and PDA + ciprofloxacin + daptomycin significantly reduced biofilm biomass (OD_{600}) compared with uncoated PVC (One-way ANOVA, $p < 0.0001$). **(B)** ***P. aeruginosa*** **(Pa14):** PDA + ciprofloxacin significantly reduced biofilm biomass relative to uncoated controls (One-way ANOVA, $p = 0.0047$; $p < 0.01$).

Conclusion

- PVC surfaces were successfully functionalised using polydopamine and diazonium chemistry, as confirmed by XPS and FTIR analysis.
- Surface modification altered PVC surface wettability, indicating changes in surface energy necessary for antimicrobial attachment.
- PDA + ciprofloxacin and PDA + ciprofloxacin + daptomycin coatings produced a significant reduction in *S. aureus* biofilm biomass.
- PDA + ciprofloxacin also reduced *P. aeruginosa* biofilm formation, demonstrating strong Gram-positive and moderate Gram-negative antibiofilm activity.
- These findings show that combining surface chemistry with antimicrobial loading can create effective antibiofilm PVC surfaces suitable for improving endotracheal tube safety.
- Further optimisation of coating stability and peptide–antibiotic combinations could enhance performance and broaden clinical applicability.

References:

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