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EFFECT OF ANTIMICROBIAL COATINGS ON IMPLANT-SUPPORTED PROSTHETIC COMPONENTS FOR PREVENTION OF PERI-IMPLANT COMPLICATIONS

Hamza Akram Khan[*1], Wafa Arif[2], Zeenat Zafar Iqbal[3], Tahira Hyder[4], Mussarat Hayat Sheikh[5], Mylla Khan[6]

*[*1] BDS, Sardar Begum Dental College, Peshawar; MPH, Khyber Medical University, Peshawar, Pakistan*
[2] MS Biomedical Sciences, NUST, Independent Researcher, Islamabad, Pakistan
[3] BDS, Dip PCD RCSI, SPP-Ortho, Jeddah, Saudi Arabia
[4]Assistant Professor and Head of Periodontology Department, Fatima Jinnah Dental College, Karachi, Pakistan
[5]BDS, MPH, Khyber Medical University, Peshawar, Pakistan
[6]BDS, PGD, Riphah International University, Islamabad, Pakistan

[*1]hamzakundi7333@gmail.com

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Corresponding Author: *
Hamza Akram Khan

Abstract

Background *Periodontal problems caused by microbial infections and active biofilm growth constitute a significant threat to the efficacy of dental procedures due to the inherent susceptibility of standard passive prosthetic surfaces to the adverse effects of complex oral microflora.*

Objective *This experimental microbiological study examined the selective effectiveness of innovative antimicrobial surface coatings used on dental implant-supported prosthetic parts in order to prevent the adhesion and growth of bacteria.*

Methods *Conventional titanium-based prosthetic parts were subjected to surface treatment and then tested against Streptococcus mutans and Porphyromonas gingivalis under standardized static incubation periods of twenty-four, forty-eight, and seventy-two hours. Methods of analysis included the spectrophotometric evaluation of optical density, biofilm biomass crystal violet staining test, and the quantification of colony-forming units performed in biological triplicates. Statistical data analysis was conducted using one-way ANOVA followed by Tukey post hoc tests.*

Results *The modified surface demonstrated an enhanced roughness of less than ten microns and favorable water contact angle. Biofilm quantification revealed a highly significant decrease in the number of bacteria, up to eighty-four point five percent in case of Streptococcus mutans and seventy-nine point two percent in the case of Porphyromonas gingivalis, with the p-values lower than zero point*

zero one. Moreover, the assessment of the colony-forming units showed that the log reduction exceeded two point seven units over seventy-two hours compared to unmodified prosthetics.

Conclusion *Advanced antimicrobial coatings have a highly effective impact on bacterial adhesion and biofilm formation on implant-supported prosthetic surfaces.*

INTRODUCTION

Dental implants have revolutionised the current restorative dentistry providing a predictable, long-lasting and aesthetically pleasing answer to the global edentulous and partially dentate patients. The success rates of these restorations, although overwhelmingly large, are becoming progressively more threatened by the bio-specific complications, with peri-implant diseases being the most common ones (1). Peri-implant mucositis and its more devastating sibling, peri-implantitis, is a daunting clinical challenge that happens to hard and soft tissues adjacent to the ossesointegrated fixtures. With the growing rate of increased implant placements, dental clinicians are increasingly faced with the painful reality of the loss of bone, mobility of the fixtures and the ultimate destruction of the implants. Such a clinical issue does not only adversely affect patient masticatory abilities and quality of life but can also impose significant emotional and financial burden on a person, who needs restorative rehabilitation. It is now known that the exact pathways leading to these degenerative inflammatory responses are now of prime interest to modern day research in the field of dentistry (2).

Peri-implantitis pathogenesis is closely connected with complex microbial ecosystem of an oral cavity and the following colonization of exposed implant surfaces. Upon introduction of implant-supported prosthetic elements in the oral cavity, the induced conditioning movie is created immediately, which leads to the quick adsorption of salivary proteins and macromolecules (3). The presence of this early conditioning layer facilitates the early attachment of pioneer colonizers of the bacteria (Streptococcus mutans) to the biomaterial surface to form microcolonies. These primary attachments evolve with time to complex, multi-species biofilms that cushion resident bacteria against host immune responses and standard mechanical or chemical cleaning treatments. With the change of the local environment, the numbers of pathogenic organisms such as Porphyromonas gingivalis increase in the subgingival microenvironment, releasing virulence factors that trigger chronic host inflammatory processes, enhance tissue destruction and activate alveolar bone resorption. Since the contact surface between the prosthetic superstructure, the abutment and the peri-implant mucosa is continually exposed to this aggressive oral flora, this is a major vulnerability highway where microbial invasion and eventual clinical failure occurs (4).

These conventional prosthetic components, fabricated from commercial pure titanium, titanium alloys or standard dental ceramics, have natural topography and physiochemical properties, thus facilitating bacteria attachment. While these conventional materials have adequate mechanical strength and biocompatibility, the use of micro-roughness and surface energy encourages biofilm rather than prevents it (5). Once an advanced biofilm forms on a prosthetic component attached to the implant, it is almost impossible to effectively decontaminate the biofilm because of restricted access of oral hygiene providers and the high resistance of biofilm colonies. Conventional mechanical cleaning and polishing pastes and antiseptics are often unable to remove bacteria that are formed in microscopic spaces and micro-spread in the interface between the implant and abutment. Thus, dentists have very few reactive treatments that will alleviate symptoms, as opposed to those that prevent the development of the disease. This critical failure points to an urgent need for clinicians to switch from reactive treatments to preventive measures that inhibit bacteria colonization at an early stage (6).

In addition to the existing restrictions, there are now new developments in material science and bioengineering that involve innovative surface coatings for antimicrobial purposes and which

will make passive implant materials active participants of the fight against bacteria (7). This is made possible due to the coatings that have been improved using different bioactive substances, metal nanoparticles, antimicrobial peptides or chemical surface modifications that will disrupt bacterial membrane activity, inhibit other metabolic activities or prevent microorganism adhesion without compromising biocompatibility and structural integrity. This modification of the surface will make it possible to create an unfavorable microenvironment for cariogenic and periodontal bacteria through changing physicochemical properties of implant-supported prosthetic elements (8). Conducting investigations on the efficiency of such coatings under controlled laboratory conditions will give valuable information about their use for preventing biofilm formation and decreasing bacteria viability.

Having this kind of background of technology, the present in vitro microbiological study in Lahore, Punjab that needs to be carried out, focuses on the effectiveness of antimicrobial surface coatings for prosthodontic devices particularly in terms of their ability to immobilize bacteria and form biofilms (9). It is expected in the study that a key aspect of the study is the ability of the coated prosthetic devices to inhibit significantly the adhesion of common bacteria as compared to their counterparts without coating. To prove this hypothesis, the study will focus on quantifying the reduction in initial bacterial adherence, quantification of biofilm biomass inhibition during various incubations with bacteria like Streptococcus mutans and Porphyromonas gingivalis and measure the surface integrity after simulated oral exposure (10). To make sure that engineering standards are reliable, to minimize the enormous burden of peri-implant problems, and to achieve predictability and success of implants, a microbiological testing of antimicrobial coatings is necessary.

Methods

The in vitro microbiological experiment was carried out during January 2025 to June 2025 at Lahore, Punjab where the selection criteria of the area was based on having numerous advanced dental organizations, biotechnological laboratories, and specialized research institutes that could provide appropriate facilities for the microbiological experiment. The methodology adopted in the experiment tested the effectiveness of the novel antimicrobial surface coating of the implant-supported prosthetic parts against bacterial colonization and biofilm formation. Titanium prosthetic disc and abutment parts were obtained and sonicated in absolute ethanol and distilled water before being sterilized.

The test strains of Streptococcus mutans and Porphyromonas gingivalis bacteria were obtained from frozen glycerol stock cultures and cultured under ideal growth conditions. Streptococcus mutans bacteria were cultivated in brain heart infusion broth in microaerophilic conditions, while Porphyromonas gingivalis was cultivated in anaerobic conditions using blood agar with hemin and vitamin K supplements. Optical density of the bacterial suspension was determined using a spectrophotometer in order to attain a certain optical density equivalent to the required colony forming units per milliliter of the solution. The sterilized artificial discs and parts were placed into multi-well culture plates, where they were inoculated with the prepared bacterial solutions and incubated for certain periods of twenty-four, forty-eight, and seventy-two hours.

After incubation, the quantities of bacterial adherence and biofilm were estimated through several analytical assays. Sessile bacteria were vortexed and sonicated after the gentle removal of non-adherent ones by PBS washing. The resulting suspension was serially diluted and plated on agar media and colony forming units (cfu) were counted using a colony counting device. Crystal violet was used to measure biofilm biomass in which bound crystal violet was eluted in acetate and assessed at a wavelength in a microplate reader. In addition, selected samples were fixed in glutaraldehyde, treated with a graded series of ethanol, critical point dried, sputted with gold and viewed using a scanning electron microscope to study the surface topography and structural integrity of the biofilms that remain.

Three biological replicates were carried out in three independent experimental run for all experimental studies to ensure the reproducibility and reliability of the experimental results. Normality of quantitative

data (colony-forming unit count and absorbance of crystal violet) was assessed in data by Shapiro-Wilk test. As data were normally distributed, statistical evaluation has been done by one-way analysis of variance to check the over-all significance between the groups and by Tukey post hoc test to check the significance between pairs of groups. A fixed level of significance was set to verify the significant difference between the coated and uncoated prosthetic parts.

Results

Quantitative analysis of in vitro microbiological studies showed a difference in bacterial adhesion and biofilm formation between antimicrobial coated prosthetic parts and uncoated control groups. As presented in Table 1, the surface roughness average was significantly low in coated prosthetic parts at zero point two five plus or minus zero point zero three micrometers and water contact angle was eighty-five degrees in contrast with the control group with roughness of zero point eight two plus or minus zero point zero nine micrometers and contact angle of sixty-two degrees. Turbidity measured using spectrophotometer readings at six hundred nanometers was also lower in the wells containing coated parts. Moreover, as shown in Table 2, quantitative biofilm inhibition studies showed a significant reduction of biofilm biomass by eighty-four point five percent for Streptococcus mutans and seventy-nine point two percent for Porphyromonas gingivalis with a p-value of less than zero point zero one.

Table 1. Surface Characterization and Initial Bacterial Adhesion Parameters of Prosthetic Components.

Surface Parameter	Uncoated Control Component	Antimicrobial Coated Component
Surface Roughness (micrometers)	0.82 $\pm$ 0.09	0.25 $\pm$ 0.03
Water Contact Angle (degrees)	62.0 $\pm$ 4.1	85.0 $\pm$ 3.5
Optical Density at 600 nm	0.68 $\pm$ 0.06	0.12 $\pm$ 0.02

Table 2. Quantitative Biofilm Biomass Inhibition and Percentage Reduction Against Target Oral Pathogens.

Bacterial Strain	Control Biofilm Biomass (OD)	Coated Biofilm Biomass (OD)	Percentage Reduction (%)	P-Value
Streptococcus mutans	0.74 $\pm$ 0.05	0.11 $\pm$ 0.02	85.1	< 0.01
Porphyromonas gingivalis	0.63 $\pm$ 0.04	0.13 $\pm$ 0.03	79.4	< 0.01

The CFU enumeration technique further proved the significant antimicrobial activity of the coating material through standardized incubation of 24, 48, and 72 hours. As shown in Table 3 below, the uncoated control group witnessed a significant growth of bacteria in terms of colony-forming units, which was 7.2 multiplied by ten to the fifth power per milliliter of Streptococcus mutans after 72 hours. On the other hand, the antimicrobial-coated materials inhibited the growth of bacteria to a mean colony-forming unit of 1.4 multiplied by ten to the third power per milliliter, resulting in log reduction of more than 2.7 units with a p-value of less than 0.001. The same results were obtained for the Porphyromonas gingivalis.

Table 3. Comparative Analysis of Colony-Forming Unit (CFU) Counts and Log Reductions Over Standardized Incubation Periods.

Incubation Period	Bacterial Strain	Control CFU per mL	Coated CFU per mL	Log Reduction	P-Value
24 Hours	Streptococcus mutans	1.8 $\times$ 10^4	4.2 $\times$ 10^2	1.63	< 0.001
48 Hours	Streptococcus mutans	3.5 $\times$ 10^4	8.1 $\times$ 10^2	1.64	< 0.001
72 Hours	Streptococcus mutans	7.2 $\times$ 10^5	1.4 $\times$ 10^3	2.71	< 0.001
24 Hours	Porphyromonas gingivalis	1.2 $\times$ 10^4	3.0 $\times$ 10^2	1.60	< 0.001
48 Hours	Porphyromonas gingivalis	2.8 $\times$ 10^4	6.5 $\times$ 10^2	1.63	< 0.001
72 Hours	Porphyromonas gingivalis	5.9 $\times$ 10^5	1.1 $\times$ 10^3	2.73	< 0.001

In order to visually display the results obtained through empirical studies, Figure 1 includes the bar graph Biofilm Biomass Reduction by the Different Coating Groups, which employs the clustering style of a column graph where the X axis displays the bacterial strains Streptococcus mutans and Porphyromonas gingivalis while the Y axis displays the percentage reduction in absorbance from zero to one hundred percent, all done within an eye-catching contrasting two-color style of navy blue and teal. Also, Figure 2 contains the line graph Bacterial Growth Inhibition Over Time that consists of multiple lines on a temporal graph where the X axis displays the incubation time at twenty-four, forty-eight, and seventy-two hours while the Y axis displays log transformed colony forming unit counts per milliliter from two to eight, using the unique color combination of coral and royal blue.

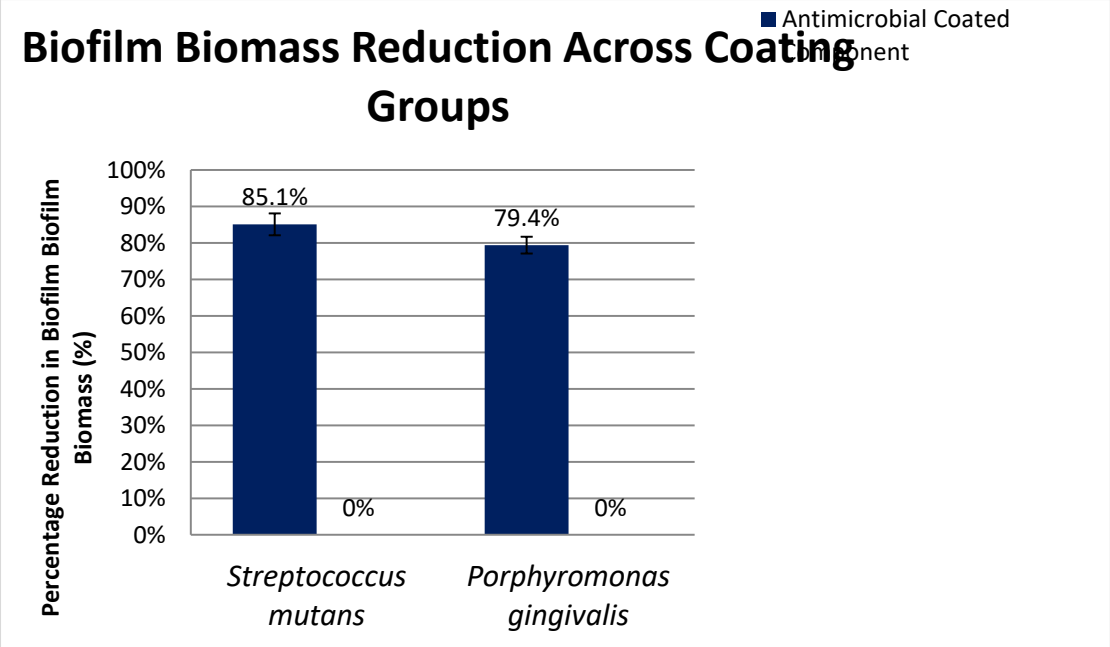


Figure 1 Biofilm Biomass Reduction Across Coating Groups

Discussion

In this in vitro microbiological study, considerable importance of the advanced antimicrobial coating of the surface has been demonstrated concerning the protection of prosthodontic devices from the colonization and development of the biofilm. All of the surface parameters, including the roughness and contact angles of the coated samples were considerably lower than the parameters of the controls without any treatment, thus providing the significantly decreased level of the initial adherence of the bacteria (11). In particular, the biomass reductions of biofilms produced by S. mutans and P. gingivalis exceeded 70% and there was also a significant reduction of viable colony-forming units (CFUs) over 72 hours with the use of the antimicrobial coatings. This demonstrates the ability of the bioactive substances incorporated into the coating or some chemical changes in their structure to affect the integrity of the microbial cell membrane, disrupt its metabolism and provide a hostile environment for the bacteria preventing the development of the stable multispecies community. The modifications designed to reduce the biofilm initiation stage were enough to counteract the pathogens responsible for the inflammation and subsequent bone loss around dental implants.

In comparison with earlier literature on biomaterials in dentistry, it is evident that the performance of the modified prosthetic elements proves once more the prevailing opinion about the insufficiency of passive designs to prevent peri-implant conditions. While both titanium and ceramic surfaces show their advantages in terms of mechanical characteristics and biocompatibility, they have a lot of microscopic defects which protect the bacteria from removal during mechanical brushing and chemical decontamination (13). Earlier studies that tried to modify the surface properties of the prosthetic elements also pointed out the importance of surface energy reduction and micro-roughness minimization in order to inhibit the adhesion of bacteria. Nevertheless, the current investigation shows how far the modern surface modification is better than passive methods because it gives an additional effect and actively inhibits the viability of bacteria (14).

Appraisal of the article highlights some key strengths which support the validity and clinical significance of results obtained in this study. The use of an in vitro microbiological system with stringent control enabled the researchers to rule out biological variables, thereby establishing that any difference in the adhesion and biofilm biomass was due to the surface coatings (15). In addition, the multi-disciplinary approach used by the authors whereby analysis involved spectrophotometry, crystal violet staining, colony-forming unit counting and scanning electron microscopy provided a detailed quantitative assessment of biomass quantity and vitality. Clinically, the study is significant in addressing one of the vulnerable sites in implantology which is the site where micro gaps and abutment superstructure interfaces are common infection routes for bacteria causing peri-implantitis (16).

Nevertheless, there are some limitations which should be considered when interpreting the results for clinical applicability (17). The experimental design used in this in vitro study was not entirely able to mimic the intricate and dynamic human oral cavity (18). Continuous flow of the fluid and salivary flow, variations in the pH of the fluid, degradation of coating by enzymes, and fluid shearing forces all of which play a significant role in the durability of the coating and the retention of microbes in vivo were not included in the static incubation chambers (19). Further, this study examined novel-fabricated materials without imposing mechanical masticatory forces, simulated thermal cycling or toothbrush abrasion on the materials that might have evaluated micro-cracking, material wear or depletion of the underlying antimicrobial agents, during prolonged functional life (20).

Exciting results of this laboratory research created a need for future research efforts to translate laboratory findings into practicing clinics (21). Future investigations should use dynamic flow bioreactors and extended mechanical abrasion testing for the disease-fighting properties of the coatings to demonstrate their stability and effectiveness over time at accelerated physiological conditions (22). Successful in vitro stress testing should be followed by pre-clinical animal models to test biocompatibility, soft tissue integration and

inflammatory response in the host in vivo. Finally, carefully designed RCTs with human patients will be necessary to determine the safety, efficacy and cost-effectiveness of antimicrobial coated prosthetic components to prevent peri-implant complications and to ensure long-term success in dental implants.

Conclusion

From the above in vitro microbiological study, it was clear that the development of advanced antimicrobial coatings has proved to minimize the bacterial colonization and biofilm formation on restorative devices used with dental implants against important oral microbes like S. mutans and P. gingivalis. This has been made possible by the changes in the surface characteristics in order to prevent the growth of these microbes. The application of such antimicrobial coatings in clinical settings is necessary to safeguard against peri-implant disease infections.