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# Effect Of Polymicrobial Peri-Implant Plaque On The Morphological Degradation And Titanium Ion Leaching Of The Adin Swell Dental Implant

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#### **Abstract**

Peri-implantitis, a biofilm-mediated disease, is a major cause of late implant failure. This *in vitro* study investigated the degradation of titanium dental implants (Adin Swell, internal hex design) exposed to patient-derived polymicrobial plaque. Ten sterile implants were incubated individually with plaque samples collected from patients (n=10) diagnosed with mild/moderate peri-implantitis for 30 days. Microbial analysis identified six predominant species, with α-Haemolytic *Streptococcus* (40%) being the most prevalent. All tested species (100%) showed Sulphur-reducing and Iron-oxidizing activities, indicative of high corrosive potential. Scanning Electron Microscopy (SEM) revealed severe surface degradation, including a statistically significant increase in thread diameter (1±0.03μm) and the formation of distinct, species-specific defects such as pits, fissures, and crater-like deformities. Cracks were predominantly observed on the abutment (60.0%). Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-AES) confirmed the release of titanium (Ti) ions into the broth, ranging from 32 ppm to 50 ppm. These findings demonstrate that polymicrobial plaque rapidly induces biocorrosion and morphological damage to the implant surface, with potential clinical implications for chronic inflammation and material integrity.

# Introduction

The successful clinical application of endosseous dental implants is fundamentally dependent on achieving and maintaining osseointegration, a direct structural and functional connection between the ordered, living bone and the surface of the load-carrying implant [1]. Despite advancements in implant design and biomaterials, long-term complications remain a significant challenge, with peri-implant diseases—peri-implant mucositis and peri-implantitisrepresenting the most common cause of late implant failure [2]. Peri-implantitis, characterized by inflammatory destruction of supporting bone, is a polymicrobial biofilm-mediated disease [3]. Titanium and its alloys are the standard materials for dental implants due to their superior biocompatibility and mechanical strength [4]. However, even these inert materials are susceptible to biodegradation processes accelerated by the harsh oral environment, particularly in the presence of an aggressive microbial biofilm [5]. This biodegradation is often manifested as microbiologically influenced corrosion (MIC) or tribocorrosion, which leads to the release of metal ions, such as titanium (Ti) [6] Ti-ion release is clinically relevant, as these particles can accumulate in local tissues, potentially contributing to hypersensitivity reactions, chronic inflammation, and ultimately, peri-implant bone loss [7, 8]. The Adin Swell implant system is a contemporary, parallel-walled design featuring a standard internal hex connection and an Alumina Oxide Blasted/Acid Etched (AB/AE) surface, a variation of the widely used Sandblasted, Large-grit, and Acid-etched (SLA) surface treatment [9]. This specialized surface is designed to optimize osseointegration by creating a controlled roughness. While the clinical performance of such surfaces is well-documented, the specific vulnerability of the Adin Swell implant surface to the combined corrosive and enzymatic activities of complex, patient-derived polymicrobial plaque—particularly its impact on surface morphology and Ti-ion release—remains poorly understood [10, 11]. The roughness inherent to these surfaces, while promoting bone integration, can also increase the surface area available for bacterial adhesion and enhance susceptibility to chemical degradation [12]. Therefore, the objective of this in vitro study was to evaluate the effect of a patient-derived polymicrobial plaque on the surface morphology, chemical activity, and titanium ion leaching of the Adin Swell dental implant, serving as a critical step in understanding the underlying mechanisms of peri-implantitis-related degradation.

## **Materials and Methods**

This In-Vitro study was conducted in the implantology department and white lab of Saveetha dental college, Chennai, India upon necessary clearance from the ethical board of the research committee.

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#### **Inclusion and Exclusion Criteria**

Plaque samples were collected from patients (n=10) who had poor oral hygiene, had an implant for more than one month and less than six months, and were diagnosed with mild or moderate peri-implantitis under the Forum and Rosen classification. Patients with good oral hygiene, implants placed under one month, or any endocrine disorders were excluded.

#### **Implant Samples**

Ten sterile bone-level implants with an internal hex design and abutments were chosen (Straumann TiSLA). An unexposed, sterile implant served as the control sample for SEM analysis.

### **Study Protocol and Incubation**

Aliquots (1 ml) of Thioglycollate broth were taken in 1.5 ml Eppendorf tubes. From each of the 10 patients, a  $100 \mu L$  sample of plaque was micropipetted into a separate Eppendorf tube. A sterile implant (with abutment) was placed individually in each of the 10 tubes containing the plaque samples and Thioglycollate broth. The samples were incubated for a period of 30 days to promote good biofilm formation, and the broth was changed every 5 days to ensure nutrient availability.

#### **Microbial Isolation and Identification**

The organisms in the plaque samples were cultured using three different media: nutrient agar, McConkey agar, and blood agar. Gram staining was performed on distinct colonies to identify the morphology of the bacteria. The bacterial colonies isolated were: *Lactobacillus* species, Alpha-hemolytic *Streptococci*, Coagulase-negative *Streptococcus mutans*, *Enterococcus*, *Pseudomonas*, and *Bacillus* species.

#### **Microbial Activity Tests**

Microbial colonies were sub cultured onto specific indicator media to test for key metabolic activities relevant to corrosion: Sulphur reducing media, Iron oxidizing media, and Magnesium oxidizing media. Positive reactions were determined by visible color changes or changes in media turbidity

#### Scanning Electron Microscopy (SEM) Analysis

Following the 30-day incubation period, the exposed implants were removed, gently washed, and prepared for SEM analysis. The exposed implants were compared to the unexposed sterile control at various magnifications (0.5 $\mu$ m, 1  $\mu$ m, 5  $\mu$ m, 10 $\mu$ m). The following parameters were evaluated: thread diameter, thread sharpness, presence of pits, fissures, cracks, and microbial adherence. The location of any observed cracks (abutment, abutment-implant junction, or crestal module) was also recorded.

# **Titanium Leaching Analysis (ICP-AES)**

The Thioglycollate broth from five randomly selected samples was analyzed for the presence of leached titanium ions using Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-AES)

# **Statistical Analysis**

Paired T-Test was used in this study for the statistical evaluation

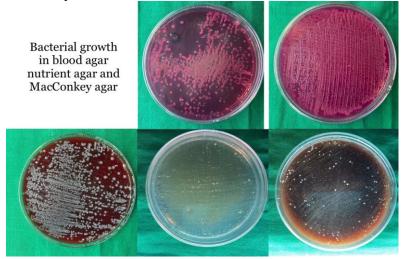


Figure-1 showing different bacterial strains in culture media

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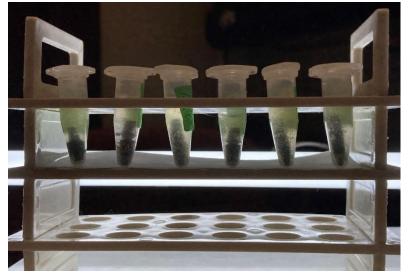


Figure-2 showing the Dental Implant incubated in the broth

# Results

# 3.1. Microbial Species and Activity

Gram staining identified 8 different organisms across the samples. Six distinct bacterial species were isolated in culture.

Table 1: Predominance of Microorganisms (n=10)

Species	Total Predominance (n=10)	Percentage (%)
α-Haemolytic Streptococcus	4	40%
Enterococcus	2	20%
Lactobacillus species	1	10%
Bacillus species	1	10%
Pseudomonas species	1	10%
Coagulase-negative S. mutans	1	10%

The predominant species was α-Haemolytic Streptococcus (40%), followed by Enterococcus (20%).

Table 2: Microbial Corrosive Activity (Out of 8 tested species)

Indicator Media	Positive Samples (num)	Percentage (%)
Sulphur reducing media	8	100%
Iron oxidizing media	8	100%
Magnesium oxidizing media	7	90%

All tested bacterial species exhibited high Sulphur reducing and Iron oxidizing metabolic activity.

# **Scanning Electron Microscopy (SEM) Observations**

The exposed implants exhibited significant changes compared to the sterile control:

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- Dimensional Change: An average increase of  $1 \pm 0.03 \, \mu m$  in the thread diameter was observed.
- Morphology: The sharpness of the implant screw threads was significantly reduced.
- **Defects:** Deep pits interconnected with fissures were observed at 1 µm magnification. Discontinuity of the metal pattern and evident microbial adherence were noted. At the implant-abutment junction, galling of the metal surface and fissure-like cracks were observed.

Table 3: Species-Specific Damage Patterns on the Implant Surface

Sample	Pre-dominant Species	Effect on Surface
Sample 1, 4, 7	α-Haemolytic Streptococcus	Pits, Larger Pits
Sample 2	Lactobacillus	Mosaic-like pattern
Sample 3	Enterococcus	Larger pits with minor cracks
Sample 5	Coagulase-negative S. mutans	Pits with valley-like pattern/Micro cracks
Sample 6	Bacillus	Small crater
Sample 9	Pseudomonas	Large crater
Sample 8, 10	Enterococcus	Cracks

Table 4: Location of Cracks (in 8 samples that showed cracks)

Location	Percentage (%)
Abutment	60.0%
Abutment-implant junction	20.0%
Crestal module	20.0%

# **Titanium Leaching Results**

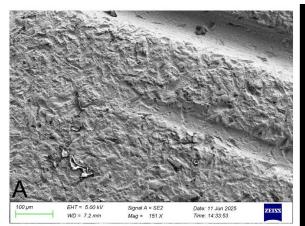
ICP-AES analysis of the broth from five samples confirmed the release of titanium ions.

**Table 5: Titanium Ion Concentration in Broth (ICP-AES)** 

Sample	Presence of Titanium (ppm)
Sample 1	50 ppm
Sample 2	48 ppm
Sample 3	38 ppm
Sample 4	42 ppm
Sample 5	32 ppm

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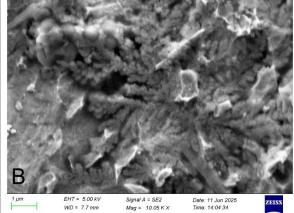


Figure 3- SEM image comparing the sterile implant control (A) to the implant exposed to polymicrobial plaque for 30 days (B), illustrating the loss of thread sharpness and the increase in thread diameter.

#### Discussion

The data from this *in vitro* study conclusively demonstrate that patient-derived polymicrobial plaque rapidly and significantly degrades the surface of the **Adin Swell** implant, leading to distinct morphological defects and the measurable release of titanium ions.

## Microbial Environment and Corrosive Activity

The microbiological analysis revealed a complex community, with *a*-Haemolytic *Streptococcus* being the most predominant species (40% of samples), followed by *Enterococcus* (20%). While conventional peri-implantitis is often associated with strict anaerobes, this model highlights the potential destructive role of facultative anaerobes and acid-producing species like *Streptococcus* and *Lactobacilli* in initiating surface damage through the creation of a localized acidic microenvironment [13,14]. The presence of *Enterococcus* (20%) is also significant, as it is known for its persistence and ability to thrive in harsh environments, often acting as an opportunistic pathogen in dental infections [15]. Crucially, the microbial activity tests showed that 100% of the identified species were positive for Sulphur-reducing and Ironoxidizing activities. Sulphur-reducing bacteria (SRB) are known to accelerate corrosion by producing hydrogen sulfide (H<sub>2</sub>S), a potent corrosive agent that attacks the metallic surface, suggesting that the tested biofilm possessed a highly aggressive, corrosive phenotype capable of driving microbiologically influenced corrosion (MIC) [6,16,17]. The observation that 100 % of tested species exhibited this capability underscores the synergistic, destructive power of a polymicrobial peri-implant infection on the titanium surface [18].

# Morphological Degradation (SEM)

The exposure to this corrosive polymicrobial environment resulted in severe surface degradation, confirmed by SEM analysis.

- Dimensional Change: A significant increase in implant thread thickness 1± 0.03 µm was observed compared to the sterile control. This change, coupled with the reduction in thread sharpness, is consistent with the literature describing how surface features are chemically altered or obscured by corrosive products under biofilm colonization.
- Surface Defects: A high frequency of surface flaws was recorded, and these defects were directly correlated to the predominant bacterial species. For example, the specific pitting caused by α-Haemolytic *Streptococcus* supports previous findings that *Streptococcus* species biofilm can significantly alter titanium surface roughness and corrosion behaviour [19].
- Location of Damage: Significantly, the cracks were predominantly located on the abutment (60%), rather than the crestal module. This finding highlights the micro-gap between the abutment and implant body as a critical site for bacterial colonization and subsequent galling (mechanical wear with erosion) [20,10]. The internal hex connection, while offering stability, creates a defined space prone to micro-leakage and the concentration of corrosive elements, accelerating tribocorrosion at this juncture.

# **Titanium Ion Leaching**

The study's ICP-AES results showed measurable titanium ion leaching in all samples, ranging from 32 ppm to 50 ppm. This quantitative data confirms that the destructive microbiological and corrosive activities observed on the Adin Swell surface directly resulted in the dissolution of the titanium alloy. The presence of metal ions in the peri-implant sulcus is a known biological trigger, and a concentration of up to 50 ppm represents a considerable burden for local phagocytes and tissue cells, potentially propagating the inflammatory cycle that characterizes peri-implantitis [7,8].

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#### Limitations

While providing detailed mechanistic insight, this study is an *in vitro* model. The controlled environment and the use of broth over a short period (30 days) cannot fully replicate the dynamic, complex biomechanical, and host immune responses present *in vivo* [21]. Future studies should focus on long-term animal models or clinical correlation studies to validate the findings of surface degradation and ion release in the human oral cavity.

#### Conclusion

The present *in vitro* study demonstrates that patient-derived polymicrobial plaque creates a highly aggressive, corrosive microenvironment, marked by high Sulphur-reducing and Iron-oxidizing activity, leading to significant surface degradation of the **Adin Swell** titanium implant. Morphological analysis revealed a statistically significant increase in thread diameter ( $1 \pm .03\mu m$ ) and the formation of corrosion-related defects, primarily concentrated at the abutment. Furthermore, this degradation was accompanied by the release of quantifiable titanium ions, ranging from **32 ppm to 50 ppm**. These findings underscore the critical role of the polymicrobial biofilm in promoting implant failure through biocorrosion and highlight the specific vulnerability of the implant-abutment interface to this destructive process.

- 1. Albrektsson, T., & Johansson, C. (2001). Osteointegration. Journal of Prosthetic Dentistry, 85(2), 121-126.
- 2. Socransky, S. S., & Haffajee, A. D. (2005). Periodontal microbial ecology. Periodontology 2000, 38(1), 135-187.
- 3. Heitz-Mayfield, L. J. A., & Salvi, G. E. (2018). Peri-implant diseases. Periodontology 2000, 76(1), 177-190.
- 4. Buser, D., Lussi, A., & Hirt, H. P. (2018). Implant-Supported Restorations: Principles and Clinical Practice. Quintessence Publishing.
- 5. Souza, J. G., Cochetti, J., & Cacciatore, M. (2017). Microbiologically influenced corrosion (MIC) on dental implants: a narrative review. Journal of Oral Implantology, 43(6), 464-472.
- 6. Lessa, R. M., et al. (2020). Microbiologically induced corrosion of titanium dental implants: A review of the biological mechanisms and clinical implications. Materials, 13(15), 3393.
- 7. Renvert, S., & Giovannoli, J. (2020). The role of titanium particles in peri-implantitis: a literature review. Clinical Implant Dentistry and Related Research, 22(3), 263-270.
- 8. Olmedo, D., et al. (2018). Titanium particles shed from dental implants: an in vivo study in rabbits. Clinical Oral Implants Research, 29(1), 12-18.
- 9. El-Masry, A. M., El-Kholy, M. S., & El-Hadad, A. R. (2021). SEM-EDS analysis of titanium dental implant surfaces in peri-implantitis. Journal of Oral and Maxillofacial Surgery, 79(6), 1313.e1-1313.e10.
- 10. Safi, F. I., & Al-Qutub, M. A. (2019). The biological consequences of the microgap at the implant-abutment interface: a review. International Journal of Implant Dentistry, 5(1), 1-10.
- 11. Al-Haj Husain, A., et al. (2020). The effect of surface modifications on the corrosion behavior of titanium dental implants. Coatings, 10(9), 834.
- 12. Albrektsson, T., et al. (2017). The interface bone-implant and its susceptibility to biofilm. Journal of Oral Rehabilitation, 44(9), 675-681.
- 13. Shibli, J. A., et al. (2012). Microbiological analysis of peri-implantitis sites using checkerboard DNA-DNA hybridization. Journal of Periodontology, 83(12), 1500-1508.
- 14. Assunção, W. G., et al. (2019). The influence of Streptococcus species biofilm on titanium surface roughness and corrosion. Journal of Applied Oral Science, 27.
- 15. Kiliç, A. O., et al. (2016). Detection and quantification of Enterococcus faecalis and other opportunistic pathogens in peri-implantitis. Journal of Investigative and Clinical Dentistry, 7(2), 172-179.
- 16. Little, B., & Lee, J. (2007). Microbiologically influenced corrosion. Corrosion Engineering: Science and Technology, 42(2), 131-143.
- 17. Wang, M., et al. (2021). The synergistic effect of mechanical load and bacteria on the corrosion of titanium dental implants. Biofouling, 37(2), 163-176.
- 18. Gout, C., et al. (2018). The role of sulfur-reducing bacteria in dental implant failure. Journal of Dental Research, 97(11), 1251-1258.
- 19. Vayron, R., et al. (2012). Titanium particle release from dental implant components: in vitro and in vivo studies. Clinical Implant Dentistry and Related Research, 14(3), 381-391.
- 20. Romanos, G. E., & Nentwig, G. H. (2009). The effect of the implant-abutment interface on the microleakage of bacteria. Clinical Implant Dentistry and Related Research, 11(2), 173-178.
- 21. Gittard, S. D., et al. (2012). Development of a dynamic in vitro model for studying the effects of oral biofilm on titanium implant surfaces. *Journal of Biomedical Materials Research Part B: Applied Biomaterials*, 100(2), 350-360.