

# Evaluation of Triple Antibiotic Paste-modified Soft Liner on Antibacterial Activity and Wound Healing Following Dental Implant Placement: A Prospective Interventional Study

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## ABSTRACT

**Introduction:** Soft liners used in temporary prostheses during implant healing may harbour microbial biofilms, potentially impairing peri-implant tissue health. Incorporating antimicrobial agents into these liners could reduce bacterial colonisation and promote early wound healing.

**Aim:** To evaluate and compare the antibacterial activity and wound healing potential of a Triple Antibiotic Paste (TAP)-Modified Soft liner (TMS) versus a conventional soft liner in patients undergoing dental implant placement.

**Materials and Methods:** This prospective interventional study was conducted at the Department of Implantology, Saveetha Dental College and Hospital, Chennai, India, between April 2024 and July 2024. It included 40 systemically healthy participants with a single missing tooth and was randomly assigned into two equal groups. Group 1 (Control) received a conventional soft liner in a temporary partial denture post-implant placement, while Group 2 (Test) received TMS in the same prosthesis. Antibacterial activity was assessed on days 0, 3, and 7 against *Streptococcus aureus*, *Pseudomonas aeruginosa*, and *Enterococcus faecalis* using the agar well-

diffusion method. Wound healing was evaluated on days 7 and 14 using the Landry Wound Healing Index (WHI). Statistical analysis was performed using IBM Statistical Package for Social Sciences (SPSS) software, version 23.0 (Armonk, NY, USA), utilising an Independent t-tests, with significance set at p-value <0.05.

**Results:** The mean age of participants in the control group was 44.7±8.1 years, while the test group had a mean age of 45.3±7.5 years, with no statistically significant difference between the groups (p-value=0.78). Group 2 (TMS) demonstrated significantly larger zones of inhibition for all three test organisms on days 0, 3, and 7 (p-value <0.05), indicating superior and sustained antibacterial activity. Wound healing scores were significantly higher in group 2 at both day 7 (3.20±0.63 vs 2.00±0.82; p-value=0.002) and day 14 (4.70±0.48 vs 3.20±0.63; p-value <0.001), reflecting enhanced tissue healing.

**Conclusion:** TMS demonstrated superior antibacterial activity and enhanced early wound healing compared to conventional liners, indicating their potential as a bioactive adjunct to improve peri-implant healing. Further long-term studies are needed to confirm these findings.

**Keywords:** Dental prosthesis, Drug delivery systems, Peri-implant health, Tissue conditioners

## INTRODUCTION

Dental implants have revolutionised the field of prosthetic dentistry, offering a reliable and long-term solution for the replacement of missing teeth. Their capacity to restore both aesthetics and masticatory function, while preserving adjacent structures, has made them a preferred choice in contemporary oral rehabilitation [1-3]. Central to the success of dental implants is the phenomenon of osseointegration, a biological process wherein the implant achieves direct structural and functional connection with the surrounding alveolar bone [4]. This intricate integration is influenced by a combination of patient-related factors, surgical protocols, biomaterial properties, and most importantly, the biological events governing wound healing at the implant-tissue interface [5-10].

The post-surgical healing phase plays a pivotal role in determining implant prognosis. Healing, in this context, is a complex cascade of molecular and cellular events including inflammation control, angiogenesis, fibroblast proliferation, and new bone formation. These processes are tightly regulated and highly sensitive to both systemic and local factors. Systemic conditions such as uncontrolled diabetes, immunosuppressive states, or osteoporotic changes can adversely impact tissue regeneration, while local factors like surgical trauma,

microbial contamination, and inadequate soft tissue protection may compromise early healing and osseointegration [11].

To optimise healing outcomes, clinicians have increasingly turned their attention toward enhancing the peri-implant microenvironment. Soft-liners, resilient, tissue-friendly polymeric materials, are often employed in temporary prostheses to serve as a protective interface between the prosthesis and healing mucosa. Their cushioning effect helps distribute occlusal forces uniformly, reducing pressure-induced trauma, enhancing patient comfort, and promoting favourable wound healing. Additionally, their intimate adaptation to the mucosal surface minimises microbial ingress and mechanical irritation, thereby supporting the integrity of peri-implant tissues during early healing [12].

Beyond their mechanical function, soft liners can be functionalised to act as localised drug delivery vehicles. This therapeutic modification allows for sustained release of bioactive agents directly at the surgical site, thereby maximising clinical benefits while minimising systemic side effects. Among the various pharmacological agents studied, the TAP- a synergistic combination of metronidazole, ciprofloxacin, and minocycline has shown potent antimicrobial activity against pathogens commonly implicated in oral infections [13]. Incorporating TAP into soft-liners can potentially enhance

their therapeutic efficacy by targeting microbial colonisation and modulating local inflammation.

A previous in-vitro investigation confirmed that TMS demonstrated excellent antibacterial and anti-inflammatory properties, along with minimal cytotoxicity, indicating its potential clinical relevance [14].

In the present study, both ex-vivo microbiological analysis and in-vivo clinical evaluation were incorporated to validate these findings. Accordingly, this prospective interventional clinical investigation aimed to evaluate and compare the antibacterial activity and wound-healing potential of TMS versus conventional soft liners in patients undergoing dental implant placement. The null hypothesis is that there will be no statistically significant difference in antibacterial effectiveness or wound healing outcomes between the two groups.

## MATERIALS AND METHODS

This prospective interventional study was conducted at the Department of Implantology, Saveetha Dental College and Hospital, Chennai, India, between April 2024 and July 2024. It was designed in accordance with the principles of the Declaration of Helsinki and received approval from the Scientific Review Board of Saveetha University (SRB/SDC/IMPLANT-2209/23/TH-017). A total of 40 participants, aged between 25 and 60 years, who reported to the Department of Implantology for prosthetic rehabilitation of a single missing tooth between April and July 2024, were recruited.

**Inclusion criteria:** Eligible participants included those between 25-60 years of age with a single missing tooth and in good systemic and periodontal health.

**Exclusion criteria:** It included patients who were pregnant or lactating, current smokers, individuals who had received antibiotics or other medications within the preceding six months, patients with a history of systemic disease or periodontitis, and individuals with known allergies to any component used in the procedure.

**Sample size calculation:** Written informed consent was obtained from all participants after a thorough explanation of the procedure, associated risks, and anticipated benefits. The required sample size was determined using G\*Power Software Version 3.0, based on the effect size of 0.92 reported by Reddy GA et al., with a power of 80% and a significance level set at 0.05, which resulted in a total sample of 40 individuals [15].

Participants were assigned to groups using simple randomisation via the lottery method, resulting in two groups of equal size (n=20 each).

- Group 1 (Control) received conventional implant placement followed by a temporary partial denture lined with a commercially available soft liner material (GC Corporation, Tokyo, Japan), whereas
- Group 2 (Test) received the same surgical protocol followed by a temporary denture lined with 1:3 TMS [14].

## Study Procedure

All participants underwent implant placement following a standardised surgical protocol. A crestal incision was made, followed by the elevation of a full-thickness mucoperiosteal flap on the buccal and lingual or palatal sides. Site preparation was accomplished using a series of sequential drills of increasing diameters, as per the manufacturer's guidelines. Implant trajectory was confirmed with a 2 mm paralleling pin and further verified radiographically. Bone-level implants (TiUnite; Nobel Biocare®, Gothenburg, Sweden) were positioned 0.5 mm subcrestally, and the flap was sutured using 3/0 non absorbable black silk sutures.

Prior to implant placement, a trial of the temporary partial denture was conducted. After implant surgery, the inner surface of the prosthesis was reduced by 0.5-1 mm to eliminate direct contact

with the underlying soft-tissues. A tray adhesive was then applied to the trimmed inner surface. In group 1, the soft liner material was prepared in a standard powder-to-liquid ratio and applied to the denture base, followed by patient occlusion in centric relation for 10 minutes to allow setting. Excess material was carefully removed with a surgical blade. In group 2, the same protocol was followed using the TMS formulation instead of the standard soft liner.

Postoperative medications included Amoxicillin 500 mg three times daily and Zerodol-SP twice daily, both for three days. Participants were instructed to wear their dentures for a minimum of six hours daily and to maintain standard oral hygiene. Follow-up evaluations were scheduled on postoperative days 3, 7, and 14. Sutures were removed on the seventh day.

## Outcome assessment:

- **Antibacterial analysis:** On days 0, 3, and 7, samples of the soft liner from the external surface of the temporary dentures were retrieved using sterile scalpels. These samples were placed in labelled Eppendorf tubes containing sterile Phosphate-Buffered Saline (PBS) to preserve their integrity and minimise contamination. The tubes were stored at 4°C until analysis.

Microbiological evaluation was carried out using standard procedures. Strains of *Streptococcus aureus* (ATCC 25178), *Pseudomonas aeruginosa* (ATCC 9027), and *Enterococcus faecalis* (ATCC 29212) were procured from HiMedia Laboratories, Chennai. These strains were cultured in Brain Heart Infusion (BHI) broth and incubated at 37°C for 24 hours. The cultures were then adjusted to match a 0.5 McFarland standard, equivalent to approximately  $1.5 \times 10^8$  CFU/mL.

The agar well-diffusion method was used to assess antibacterial activity [16]. Mueller-Hinton Agar (MHA) plates were inoculated with standardised bacterial suspensions using sterile cotton swabs to ensure even distribution. Wells of 6 mm diameter were bored into the agar, and soft-liner samples were inserted into each well. TAP served as the positive control in the agar well diffusion assay and was used only to compare the antibacterial activity of the soft-liner samples. The plates were incubated at 37°C for 24 hours under aerobic conditions. After incubation, zones of inhibition were measured using a digital calliper, and diameters were recorded in millimeters.

- **Wound healing evaluation:** The wound healing outcome was assessed using the Landry WHI on postoperative days 7 and 14 [17]. This validated scoring system evaluates healing on a five-point scale, where a score of 1 indicates very poor healing and a score of 5 signifies excellent healing.

## STATISTICAL ANALYSIS

All collected data were statistically analysed using IBM SPSS software, version 23.0 (Armonk, NY, USA). The distribution of data was tested for normality using both the Shapiro-Wilk and Kolmogorov-Smirnov tests, which confirmed a parametric distribution. Consequently, an Independent t-tests were applied to compare the mean age, zone of inhibition and wound healing scores between the two groups. Chi-square test for gender comparison. A p-value of <0.05 was considered to indicate statistical significance.

## RESULTS

The mean age of participants in the control group was  $44.7 \pm 8.1$  years, while the test group had a mean age of  $45.3 \pm 7.5$  years, with no statistically significant difference between the groups (p-value=0.78). Gender distribution was comparable, with 11 males and nine females in the control group and 12 males and eight females in the test group, showing no significant difference (p-value=0.81).

## Antibacterial Assay

The antibacterial assay demonstrated significant differences between group 1 and group 2 in inhibiting microbial growth across all test organisms.

For *Streptococcus aureus*, the zone of inhibition at day 0 was  $10.90 \pm 1.73$  mm in group 1, while group 2 showed a significantly greater inhibition of  $16.40 \pm 1.65$  mm. On day 3, the antibacterial activity declined to  $8.30 \pm 1.34$  mm in group 1, but group 2 maintained a larger inhibition zone of  $14.00 \pm 1.56$  mm. By day 7, the zone was further reduced to  $6.80 \pm 1.23$  mm in group 1, whereas group 2 still showed a considerable inhibitory effect at  $10.90 \pm 1.37$  mm. This indicates the enhanced and sustained antibacterial potential of the TMS [Table/Fig-1].

Timeline	Positive control (TAP) (Mean $\pm$ SD)	Group 1 (Mean $\pm$ SD)	Group 2 (Mean $\pm$ SD)	T value	p-value
Day 0	20.25 $\pm$ 2.30	10.90 $\pm$ 1.73	16.40 $\pm$ 1.65	-7.284	<0.001*
Day 3	-	8.30 $\pm$ 1.34	14.00 $\pm$ 1.56	-8.763	<0.001*
Day 7	-	6.80 $\pm$ 1.23	10.90 $\pm$ 1.37	-7.045	<0.001*

**[Table/Fig-1]:** Antibacterial effect of soft liner on *Streptococcus aureus*.  
aIndependent t-test Group 1 vs Group 2; \*Statistically significant

For *Pseudomonas aeruginosa*, group 1 exhibited a zone of inhibition of  $10.60 \pm 1.58$  mm on day 0, compared to  $15.30 \pm 1.42$  mm in group 2. On day 3, group 1 showed reduced activity ( $8.10 \pm 1.45$  mm), while group 2 maintained superior inhibition at  $10.00 \pm 1.41$  mm. At day 7, group 1 recorded  $7.00 \pm 1.94$  mm and group 2 recorded  $9.60 \pm 0.71$  mm, again highlighting the prolonged efficacy of the TAP formulation [Table/Fig-2].

Timeline	Positive control (TAP) (Mean $\pm$ SD)	Group 1 (Mean $\pm$ SD)	Group 2 (Mean $\pm$ SD)	T -value	p-value
Day 0	19.85 $\pm$ 2.46	10.60 $\pm$ 1.58	15.30 $\pm$ 1.42	-7.006	<0.001*
Day 3	-	8.10 $\pm$ 1.45	10.00 $\pm$ 1.41	-2.968	0.008*
Day 7	-	7.00 $\pm$ 1.94	9.60 $\pm$ 0.71	-3.973	0.001*

**[Table/Fig-2]:** Antibacterial effect of soft-liner on *Pseudomonas aeruginosa*.  
aIndependent t-test Group 1 vs Group 2; \*Statistically significant

For *Enterococcus faecalis*, on day 0, group 1 showed  $10.40 \pm 1.43$  mm while group 2 exhibited a superior  $16.30 \pm 2.00$  mm. On day 3, the zones were  $8.10 \pm 0.99$  mm and  $9.80 \pm 1.23$  mm for groups 1 and 2, respectively. At day 7, inhibition decreased to  $6.80 \pm 0.19$  mm in group 1 and  $7.50 \pm 0.57$  mm in group 2. These results support the enhanced antibacterial activity and sustained effectiveness of the TAP-modified liner compared to the conventional formulation [Table/Fig-3].

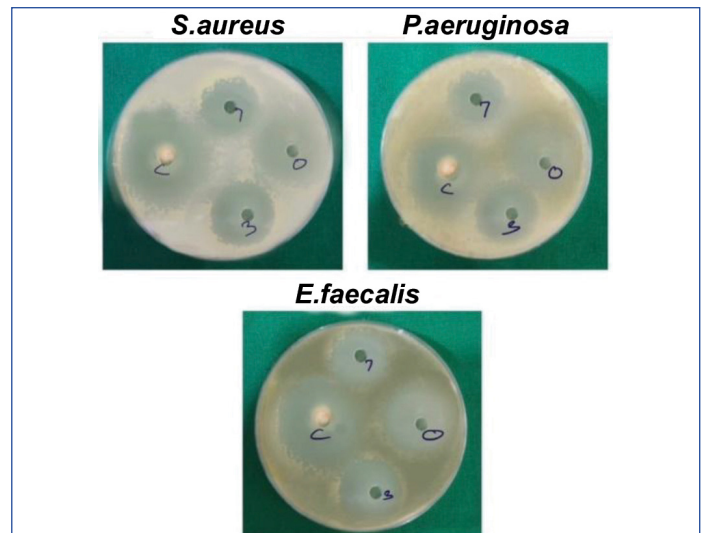
Timeline	Positive control (TAP) (Mean $\pm$ SD)	Group 1 (Mean $\pm$ SD)	Group 2 (Mean $\pm$ SD)	T -value	p-value
Day 0	20.65 $\pm$ 2.49	10.40 $\pm$ 1.43	16.30 $\pm$ 2.00	-7.581	<0.001*
Day 3	-	8.10 $\pm$ 0.99	9.80 $\pm$ 1.23	-3.401	0.003*
Day 7	-	6.80 $\pm$ 0.19	7.50 $\pm$ 0.57	-3.692	0.006*

**[Table/Fig-3]:** Antibacterial effect of soft liner on *Enterococcus faecalis*.  
aIndependent t-test Group 1 vs Group 2; \*Statistically significant

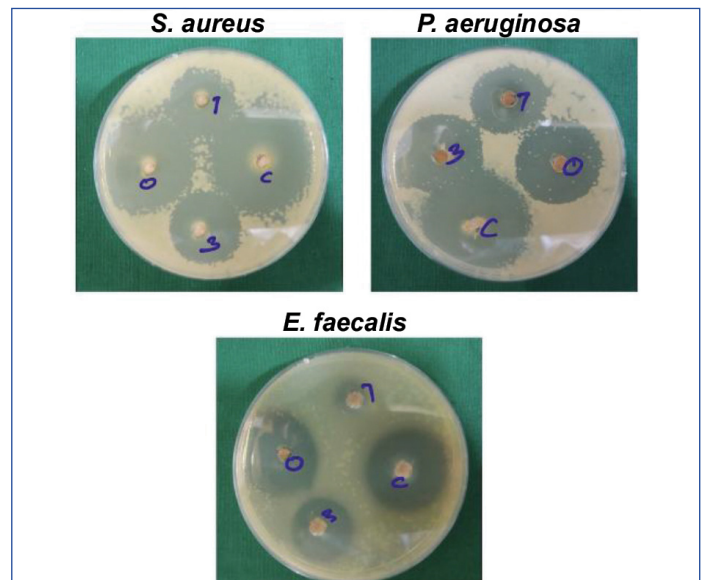
Zone of inhibition against *Streptococcus aureus*, *Pseudomonas aeruginosa* and *Enterococcus faecalis* for both control and test groups are shown in [Table/Fig 4,5].

## Wound Healing Assessment

On day 7, group 1 exhibited a mean WHI score of  $2.00 \pm 0.816$ , indicating poor healing, while group 2 showed a significantly higher score of  $3.20 \pm 0.632$  (p-value=0.002), reflecting better healing. By day 14, both groups improved; group 1 recorded  $3.20 \pm 0.632$ , while group 2 showed a notable increase to  $4.70 \pm 0.483$  (p-value <0.001), signifying excellent healing in group 2 [Table/Fig-6].



**[Table/Fig-4]:** Agar well-diffusion assay showing the antibacterial activity of soft-liner samples from the Control group (group 1 – conventional soft liner) against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Enterococcus faecalis* at Day 0, Day 3, and Day 7. The well-marked "C" denotes the Positive Control (Triple Antibiotic Paste, TAP).



**[Table/Fig-5]:** Agar well-diffusion assay showing the antibacterial activity of soft-liner samples from the Test group (Group 2 - TAP-modified soft-liner, TMS) against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Enterococcus faecalis* at Day 0, Day 3, and Day 7. The well-marked "C" denotes the Positive Control (TAP).

Timeline	Groups	Mean $\pm$ SD	T-value	p-value
Day 7	Group 1	2.00 $\pm$ 0.82	-3.677	0.002*
	Group 2	3.20 $\pm$ 0.63		
Day 14	Group 1	3.20 $\pm$ 0.63	-5.963	< 0.001*
	Group 2	4.70 $\pm$ 0.48		

**[Table/Fig-6]:** Comparison of WHI scores between groups.

## DISCUSSION

Building upon previous in-vitro findings that highlighted the biocompatibility, low cytotoxicity, and pronounced anti-inflammatory effects of TMS [14], the present in-vivo investigation sought to evaluate their clinical applicability in implant-supported provisional prostheses. In this clinical study, one group received a temporary partial denture lined with TAP-infused soft liner, while the control group was provided a conventional soft liner post-implant surgery. Microbiological sampling conducted on days 0, 3, and 7 assessed antibacterial activity against *Streptococcus aureus*, *Pseudomonas aeruginosa*, and *Enterococcus faecalis*, while wound healing was clinically evaluated on days 7 and 14.

The results revealed a marked reduction in microbial colonisation in the TAP group across all evaluated time points (p-value <0.05).



Enhanced wound healing was also observed, with the TAP group demonstrating significantly improved tissue response by day 14 (p-value 0.001), thus rejecting the null hypothesis. These outcomes suggest that incorporating TAP into soft liners offers a dual benefit-limiting early microbial load around the surgical site and promoting faster mucosal recovery.

Several prior studies across various dental disciplines echo the observed benefits of TAP. In the field of oral surgery, Gupta M et al., evaluated TAP-soaked haemostatic agent as a local delivery system following third molar extraction and found a substantial reduction in postoperative pain and trismus compared to conventional haemostatic agent. Specifically, the TAP group exhibited lower Visual Analogue Scale (VAS) scores by day 3 and better mouth opening, both of which returned to baseline by day 7 -highlighting the early therapeutic impact of TAP on tissue inflammation and healing [18].

In endodontics, TAP has been extensively studied for its broad-spectrum antimicrobial effect. Abdel Hamid EA et al., demonstrated that TAP, when used as an intracanal medicament, significantly reduced microbial load in necrotic teeth and resulted in fewer postoperative flare-ups, thereby contributing to a smoother recovery phase [19]. These findings were corroborated by Khan AM et al., who found TAP to be more effective than calcium hydroxide in relieving pain and infection associated with symptomatic apical periodontitis. The anti-inflammatory and antibacterial synergy provided by the combination of ciprofloxacin, metronidazole, and minocycline is thought to underlie this clinical advantage [20].

Furthermore, Pai S et al., explored the application of TAP in medically compromised individuals, specifically diabetic patients undergoing endodontic therapy. Their study found that TAP led to the absence of inter-appointment flare-ups, unlike the calcium hydroxide and control groups, which showed higher complication rates [21]. Similarly, Sinhal TM et al., reported that both TAP and 2% chlorhexidine gel were superior to calcium hydroxide in reducing inter-appointment flare-ups in diabetic patients, with no statistically significant difference between the TAP and chlorhexidine groups [22]. These findings are particularly relevant as they underscore TAP's consistent performance across varying host immune conditions and microbial burdens.

The regenerative potential of TAP has also been highlighted in periapical healing. In a case series by Taneja S et al., teeth with persistent periradicular lesions unresponsive to conventional therapy showed marked healing when treated with TAP for extended durations (up to three months). Radiographic evidence demonstrated gradual lesion resolution, indicating TAP's role in eradicating entrenched infections and facilitating tissue regeneration [23].

The current investigation corroborates and extends this body of evidence by demonstrating, for the first time, the successful integration of TAP into soft liners used in provisional prostheses following implant placement. The reduced bacterial load and enhanced soft tissue healing seen in the present study underscore the potential of TAP as a bioactive additive that may significantly improve early post-surgical outcomes in implant dentistry. By mitigating microbial invasion and inflammation, TAP-modified liners could pave the way for safer healing environments around dental implants.

In summary, this pioneering clinical study demonstrates that incorporating TAP into soft liner materials in temporary partial dentures can offer considerable antibacterial and anti-inflammatory benefits following implant surgery. The observed benefits in both microbial control and soft-tissue healing reinforce the clinical value of TAP-modified liners. A key strength of the present study lies in its novel clinical translation of a well-documented endodontic medicament into the realm of implant prosthetics, potentially opening new frontiers for enhancing peri-implant healing.

Future research should aim to validate these findings over longer durations and in larger cohorts, and further investigate the optimal concentration and release kinetics of TAP when incorporated into various prosthetic materials. Assessing the effects on a broader range of periopathogens and evaluating patient-reported outcomes could also offer valuable insights into its real-world applicability.

## Limitation(s)

Limitations of the current study include the relatively short follow-up period and limited sample size. Additionally, the study did not assess long-term outcomes such as implant survival rates or changes in peri-implant bone levels. The controlled clinical setting may not fully capture the variability seen in routine clinical practice. Further multicentre randomised trials are warranted to generalise these findings and explore the long-term safety and efficacy of TAP in implant prosthodontics.

## CONCLUSION(S)

The present study demonstrated that TMS significantly enhanced the antibacterial activity and early wound healing around implants compared to conventional liners. As a novel bioactive approach in implant prosthodontics, TAP incorporation showed promise in reducing postoperative complications and improving peri-implant tissue health. Further long-term studies are needed to confirm these outcomes.

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