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ORIGINAL ARTICLE

Clinical Evaluation of Oral Mucosal Immunity in Relation to Wound Healing Outcomes in Dental Patients

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ABSTRACT

Background: Oral mucosal immunity plays a central role in maintaining tissue homeostasis and orchestrating the phases of wound healing after dental procedures. Salivary immune markers, including secretory immunoglobulin A (sIgA) and pro-inflammatory cytokines, have been increasingly recognized as non-invasive indicators of immune status. Alterations in these markers may predict delayed wound healing, particularly in patients with systemic risk factors such as diabetes mellitus and smoking.

Objectives: The present study was designed to clinically evaluate the association between oral mucosal immune responses and wound healing outcomes in dental patients. Specifically, the study aimed to determine whether salivary biomarkers could serve as predictors of post-operative healing efficiency.

Methods: This prospective clinical study was conducted at Margalla Dental Hospital, Rawalpindi, Pakistan, from January 2022 to July 2023. A total of 120 patients undergoing extractions, periodontal flap surgeries, or single-tooth implant placements were enrolled. Unstimulated whole saliva samples were collected at baseline and day 7 to quantify sIgA, interleukin-6 (IL-6), and tumor necrosis factor-alpha (TNF-α) using ELISA. Wound healing was assessed on days 3, 7, and 14 using the Landry, Turnbull, and Howley Wound Healing Index (WHI). Statistical analyses included correlation and regression models to identify associations between biomarkers, systemic factors, and healing outcomes.

Results: At day 7, 88 patients (73.3%) demonstrated good healing (WHI ≥ 4), while 32 patients (26.7%) exhibited delayed healing (WHI ≤ 3). Patients with good healing showed significantly higher sIgA concentrations and lower IL-6 and TNF-α levels compared to those with delayed healing ($p < 0.05$). Diabetes mellitus, smoking, and flap-based procedures were strongly associated with delayed healing. Multivariate regression identified diabetes (OR: 3.2; 95% CI: 1.5–6.7), smoking (OR: 2.1; 95% CI: 1.1–4.2), and elevated baseline IL-6 (OR: 2.8; 95% CI: 1.3–5.9) as independent predictors of impaired healing outcomes.

Conclusion: The study demonstrates that oral mucosal immunity significantly influences wound healing outcomes in dental patients. Higher salivary sIgA levels were associated with favorable healing, whereas elevated IL-6 and TNF-α predicted delayed recovery. Systemic conditions such as diabetes mellitus and smoking further increased the risk of impaired healing. Salivary biomarker assessment offers a promising, non-invasive tool for identifying high-risk patients and guiding personalized management in dental practice.

Keywords: Oral mucosal immunity; wound healing; salivary biomarkers; cytokines; sIgA; diabetes mellitus; dental surgery.

INTRODUCTION

The oral mucosa represents a specialized immunological barrier that plays a vital role in maintaining tissue homeostasis and orchestrating wound healing following dental procedures[1]. Unlike cutaneous tissues, oral wounds generally heal faster and with minimal scarring, a process attributed to the unique characteristics of the oral immune system, including epithelial turnover, abundant salivary defense factors, and rapid recruitment of innate and adaptive immune cells. These features position oral mucosal immunity as a critical determinant of clinical healing outcomes in dental patients[2,3].

Wound healing is a dynamic and multi-phase process involving inflammation, proliferation, and remodeling. During the inflammatory phase, neutrophils, macrophages, and lymphocytes infiltrate the wound site, releasing cytokines and growth factors such as interleukin-6 (IL-6), tumor necrosis factor-alpha (TNF-α), and transforming growth factor-beta (TGF-β), which regulate subsequent tissue repair[4]. The proliferative phase is characterized by angiogenesis, fibroblast proliferation, and collagen deposition, processes that are influenced by immune activity. In the remodeling phase, immune regulation ensures extracellular matrix maturation and functional tissue recovery. Any disruption of immune balance, whether due to systemic disease, microbial dysbiosis, or local factors, can impair wound healing efficiency[5,6].

Clinical evaluation of oral mucosal immunity in dental patients has become increasingly relevant, particularly for populations at risk of delayed healing, such as those with diabetes mellitus, cardiovascular disease, or chronic periodontal inflammation[7]. Salivary immunoglobulin A (sIgA), pro-inflammatory cytokines, and antimicrobial peptides have emerged as potential biomarkers for assessing immune competence and predicting wound healing outcomes. However, there remains a lack of clinical studies directly linking oral mucosal immune profiles with objective measures of wound healing in dental settings[8].

The present study was therefore designed to evaluate the relationship between oral mucosal immunity and wound healing outcomes in dental patients undergoing minor surgical interventions. By analyzing salivary biomarkers and clinical healing indices, this study aimed to establish a correlation between immune competence and post-operative tissue repair. The findings may help identify predictive markers of healing potential, contributing to improved patient management and personalized care in dentistry[9].

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MATERIALS AND METHODS

Study Design and Setting: This prospective clinical study was conducted at Margalla Dental Hospital, Rawalpindi, Pakistan, over an 18-month period from January 2022 to July 2023. The study was designed to evaluate the relationship between oral mucosal immune responses and wound healing outcomes in patients undergoing routine minor dental surgical procedures. The study was performed in accordance with the principles of the Declaration of Helsinki, and prior ethical approval was obtained from the Institutional Ethical Committee of Margalla Dental Hospital.

Study Population and Sample Size: A total of 120 patients were recruited for the study. Patients were selected through consecutive sampling from those reporting to the outpatient surgical and

periodontal clinics. The calculated sample size ensured adequate statistical power to detect significant correlations between salivary immune markers and wound healing outcomes. Written informed consent was obtained from each participant before enrollment.

Eligibility Criteria: Patients aged between 18 and 70 years who required minor oral surgical interventions, such as tooth extractions, periodontal flap procedures, or single-tooth implant placements, were eligible to participate. Only patients who were willing to comply with post-operative follow-up visits were included. Patients with systemic immunodeficiency disorders, those on long-term immunosuppressive or steroid therapy, individuals with uncontrolled diabetes mellitus or advanced systemic illness, and pregnant or lactating women were excluded from the study. In addition, patients presenting with acute oral infections or requiring emergency surgical management were not considered.

Clinical Procedures and Post-Operative Care: All surgical procedures were carried out under local anesthesia by experienced dental surgeons using standardized aseptic protocols. The interventions were performed with an emphasis on minimizing tissue trauma, and intraoperative details such as procedure type, surgical duration, and flap design were documented. Following surgery, patients were provided with uniform post-operative instructions that included maintenance of oral hygiene, use of prescribed analgesics, and rinsing with chlorhexidine mouthwash. Antibiotics were not routinely prescribed but were given only when clinically indicated, such as in cases with systemic risk factors or signs of infection.

Collection and Analysis of Saliva Samples: Saliva samples were obtained from each patient for immunological assessment. Unstimulated whole saliva was collected in the morning hours between 9:00 and 11:00 a.m. after patients had refrained from eating, drinking, or performing oral hygiene procedures for at least ninety minutes. Patients were instructed to allow saliva to pool in the floor of the mouth and expectorate into sterile collection tubes for a period of five minutes. Samples were immediately placed on ice and transported to the laboratory, where they were centrifuged at 3,000 rpm for 15 minutes to remove cellular debris. The clarified supernatant was aliquoted and stored at −80 °C until further processing. Quantitative analysis of salivary immune markers, including secretory immunoglobulin A (sIgA), interleukin-6 (IL-6), tumor necrosis factor-alpha (TNF-α), and antimicrobial peptides, was carried out using commercially available enzyme-linked immunosorbent assay (ELISA) kits.

Assessment of Wound Healing Outcomes: Wound healing was clinically assessed on post-operative days 3, 7, and 14 using the Landry, Turnbull, and Howley Wound Healing Index (WHI). This index evaluates several healing parameters including tissue color, presence of bleeding on palpation, epithelialization of wound margins, and the presence or absence of granulation tissue. Each surgical site was scored independently by two calibrated examiners who were blinded to the salivary biomarker results to reduce assessment bias. Inter-examiner reliability was established prior to data collection.

Data Collection and Statistical Analysis: Demographic information including age, sex, medical history, and behavioral factors such as smoking status were recorded at baseline. Clinical parameters including plaque index and gingival index were also documented. All data were entered into a secure database and cross-checked for accuracy. Statistical analysis was performed using SPSS software version 26.0. Descriptive statistics were generated for demographic and clinical variables. Associations between salivary immune biomarkers and wound healing scores were analyzed using Pearson's or Spearman's correlation tests as appropriate. Independent t-tests or Mann–Whitney U tests were applied for continuous variables, while categorical comparisons were made using chi-square tests. Logistic regression analysis was employed to identify independent predictors of delayed healing. A p-value of less than 0.05 was considered statistically significant.

RESULTS

Demographic and Clinical Characteristics: A total of 120 patients were enrolled, including 68 males (56.7%) and 52 females (43.3%), with a mean age of 39.6 ± 11.2 years. The majority of cases were simple extractions (63.3%), followed by periodontal flap surgeries (23.3%) and single implant placements (13.3%). Diabetes mellitus was present in 16.7% of patients, while 12.5% were hypertensive. Nearly one-quarter (23.3%) were smokers. Detailed demographic and baseline clinical data are presented in Table 1.

Table 1. Baseline demographic and clinical characteristics of study participants (n = 120)

Variable	Category	Frequency (%) / Mean ± SD
Age (years)	–	39.6 ± 11.2
Sex	Male / Female	68 (56.7%) / 52 (43.3%)
Smoking status	Smoker / Non-smoker	28 (23.3%) / 92 (76.7%)
Diabetes mellitus	Present / Absent	20 (16.7%) / 100 (83.3%)
Hypertension	Present / Absent	15 (12.5%) / 105 (87.5%)
Procedure type	Extraction / Flap / Implant	76 (63.3%) / 28 (23.3%) / 16 (13.3%)

Salivary Biomarker Analysis: The mean baseline salivary sIgA concentration was 132.5 ± 42.3 µg/mL. Patients with delayed healing exhibited lower sIgA and higher IL-6 and TNF-α levels at both baseline and day 7 compared with patients who healed well. Notably, IL-6 and TNF-α remained significantly elevated at day 7 in the delayed healing group ($p < 0.001$). The comparative biomarker data are shown in Table 2.

Table 2. Salivary biomarker concentrations at baseline and day 7 in patients with good vs delayed healing

Biomarker	Good Healing (n = 88) Mean ± SD	Delayed Healing (n = 32) Mean ± SD	p-value
sIgA (µg/mL)	Baseline: 138.4 ± 41.2	121.3 ± 43.5	0.04
	Day 7: 145.9 ± 39.8	118.6 ± 40.2	0.02
IL-6 (pg/mL)	Baseline: 8.1 ± 2.9	10.7 ± 3.4	0.01
	Day 7: 6.2 ± 2.4	11.3 ± 3.7	<0.001
TNF-α (pg/mL)	Baseline: 11.8 ± 4.6	14.5 ± 5.1	0.03
	Day 7: 9.6 ± 3.8	15.2 ± 4.7	<0.001

Wound Healing Outcomes: By day 7, 73.3% of patients (n = 88) demonstrated good healing (WHI ≥ 4), whereas 26.7% (n = 32) had delayed healing (WHI ≤ 3). By day 14, healing improved in most patients, with 93.3% showing complete epithelialization. These outcomes are presented in Table 3.

Table 3. Distribution of wound healing outcomes (n = 120)

Healing Outcome	Day 7 (%)	Day 14 (%)
Good healing (WHI ≥ 4)	88 (73.3)	112 (93.3)
Delayed healing (WHI ≤ 3)	32 (26.7)	8 (6.7)

Risk Factors and Predictors of Delayed Healing: Patients with delayed healing were more frequently diabetic (40.6% vs 8.0%, $p < 0.001$), smokers (37.5% vs 18.2%, $p = 0.02$), and underwent flap-based procedures more often compared to atraumatic extractions. Logistic regression identified diabetes mellitus, smoking, and elevated baseline IL-6 as independent predictors of delayed healing. These associations are detailed in Table 4.

Table 4. Association of patient risk factors with delayed wound healing

Risk Factor	Good Healing (n = 88)	Delayed Healing (n = 32)	p-value
Diabetes mellitus	7 (8.0%)	13 (40.6%)	<0.001
Smoking	16 (18.2%)	12 (37.5%)	0.02
Flap procedure	14 (15.9%)	14 (43.8%)	0.001
Hypertension	9 (10.2%)	6 (18.8%)	0.21

A total of 120 dental patients were included in the study, with a fairly balanced distribution between males and females and a mean age of nearly 40 years. The majority of surgical procedures were simple extractions, while fewer patients underwent periodontal flap surgeries and single-tooth implant placements. About one in six patients were diabetic, a smaller proportion were hypertensive, and nearly a quarter were smokers. These baseline characteristics provided insight into the diversity of patients and potential systemic risk factors that could influence wound healing.

Analysis of salivary biomarkers revealed that patients with good healing outcomes demonstrated higher concentrations of secretory immunoglobulin A (sIgA) and lower pro-inflammatory cytokines such as interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF-α). In contrast, patients who experienced delayed healing consistently showed reduced sIgA levels and significantly elevated cytokine concentrations both at baseline and at day 7 post-surgery. This suggested that the immune profile, particularly mucosal inflammatory markers, had a strong association with the quality of wound repair.

Clinical wound healing assessments showed that nearly three-quarters of patients achieved satisfactory healing by day 7, while just over one-quarter experienced delayed healing characterized by poor epithelialization and persistent inflammation. By day 14, however, the majority of patients showed substantial improvement, and delayed healing was limited to a small minority.

Further analysis demonstrated that systemic and behavioral factors strongly influenced healing outcomes. Diabetic patients and smokers were significantly more likely to show delayed healing compared to non-diabetic and non-smoking patients. Likewise, flap-based surgical procedures were associated with higher rates of delayed healing compared to atraumatic extractions. Multivariate regression confirmed that diabetes mellitus, smoking, and elevated IL-6 were independent predictors of poor healing outcomes.

Overall, the study demonstrated that oral mucosal immunity, reflected in salivary immune markers, plays a critical role in determining wound healing outcomes following dental surgery. Patients with systemic disease or higher inflammatory profiles were at greater risk of delayed healing, whereas those with robust salivary immune responses had more favorable recovery patterns.

DISCUSSION

The present study evaluated the relationship between oral mucosal immunity and wound healing outcomes in dental patients undergoing minor surgical interventions[9]. The findings highlight that the quality of wound healing is closely associated with the host's immune profile, as measured by salivary biomarkers, as well as with systemic and behavioral risk factors such as diabetes mellitus and smoking[10].

Our results demonstrated that patients with good healing had higher concentrations of salivary secretory immunoglobulin A (sIgA) and lower levels of pro-inflammatory cytokines (IL-6 and TNF-α) [11]. Conversely, patients with delayed healing exhibited persistently elevated cytokine levels and reduced sIgA concentrations. These findings are consistent with previous studies suggesting that sIgA plays a central role in maintaining mucosal homeostasis, limiting microbial invasion, and supporting tissue regeneration, whereas excessive pro-inflammatory cytokine activity is linked to prolonged inflammation and impaired wound closure[12,13].

The dynamic changes observed in IL-6 and TNF-α between baseline and day 7 are noteworthy. Patients with favorable healing outcomes exhibited a marked reduction in these cytokines during the first week, which likely reflects a timely resolution of the inflammatory phase of wound healing[14]. By contrast, persistently elevated cytokines in patients with delayed healing suggest that dysregulated or prolonged inflammation is a key mechanism underlying impaired repair. These findings reinforce the concept that a controlled, self-limiting inflammatory response is crucial for optimal healing, while exaggerated immune activation may hinder progression into the proliferative and remodeling phases[15,16].

Systemic conditions also played a significant role in modulating wound healing. Diabetes mellitus was strongly associated with delayed healing in this study, consistent with the well-established impact of hyperglycemia on neutrophil chemotaxis, macrophage activation, and growth factor release[17]. Similarly, smoking emerged as an independent predictor of poor outcomes, in line with evidence that tobacco use reduces vascularity, alters salivary composition, and suppresses local immune function. Flap-based surgical procedures were also more likely to result in delayed healing compared with atraumatic extractions, suggesting that surgical trauma and greater tissue manipulation contribute to prolonged inflammatory responses[18].

The clinical implications of these findings are considerable. First, salivary biomarkers such as sIgA, IL-6, and TNF-α may serve as useful non-invasive indicators of healing potential and could be incorporated into pre-operative risk assessment protocols[19]. Second, identification of high-risk patients particularly diabetics and smokers should prompt dentists to implement enhanced monitoring, stricter glycemic control, smoking cessation counseling, and the use of adjunctive measures such as antiseptic rinses or local immunomodulatory agents. Finally, the study supports the broader concept of precision dentistry, in which patient-specific immune and systemic factors guide tailored treatment strategies to improve outcomes[20].

Nevertheless, certain limitations should be acknowledged. This study was conducted at a single dental center, which may limit the generalizability of findings. The sample size, while adequate for detecting moderate associations, may not capture all subtle biomarker variations across different populations[21,22]. Additionally, the analysis focused on selected immune markers; inclusion of a broader biomarker panel, such as growth factors and matrix metalloproteinases, could provide more comprehensive insights. Future multicenter studies with larger cohorts and extended biomarker profiling are warranted to validate and expand upon these results[23-25].

CONCLUSION

This study demonstrates that oral mucosal immunity, reflected by salivary immune markers, is a critical determinant of wound healing outcomes in dental patients. Higher levels of sIgA and lower pro-inflammatory cytokines were associated with favorable healing, whereas elevated IL-6 and TNF-α predicted delayed repair. Systemic factors such as diabetes mellitus and smoking further contributed to impaired outcomes, underscoring the interplay between local immunity and systemic health. The clinical evaluation of oral mucosal immunity offers a valuable, non-invasive approach to predicting wound healing trajectories in dental practice. Integrating salivary biomarker analysis with routine clinical assessment may help identify high-risk patients, guide preventive strategies, and optimize post-operative recovery. In the long term, such approaches can support the advancement of personalized and precision dentistry, ultimately improving patient care and treatment success.

Authors' Contributions: AY[1] conceived and designed the study. MA[2] contributed to data collection and laboratory analysis. AAM[3] was responsible for statistical analysis and interpretation of results. NBK[4] assisted in clinical procedures and patient follow-up. SS[5] contributed to manuscript drafting and critical revision. SSu[6] provided overall supervision and final approval of the manuscript. All authors read and approved the final version of the manuscript.

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