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Evaluation of liver enzymes as biomarkers in serum and saliva among stage II generalized periodontitis patients with poor glycemic control

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Abstract:
Periodontitis and uncontrolled Type 2 Diabetes Mellitus share inflammatory pathways, but the diagnostic value of salivary liver enzymes as biomarkers of this interaction remains insufficiently established. This cross-sectional clinico-biochemical study evaluated 120 participants allocated into 4 groups of 30 each by periodontal and diabetic status, with clinical periodontal parameters and salivary and serum liver enzyme levels assessed. Uncontrolled diabetic patients with Stage II generalized periodontitis exhibited the highest clinical and biochemical values ($p < 0.0001$), while clinical attachment level differentiated controlled from uncontrolled diabetes ($p = 0.0429$) and salivary enzymes showed stronger correlations with glycated hemoglobin ($r = 0.863–0.962$) than serum enzymes ($r = 0.373–0.620$). All five liver enzymes increased significantly with periodontitis and diabetes, with salivary measurements showing superior discriminatory ability and strong positive correlations with corresponding serum levels ($r > 0.80$). Thus, data shows that salivary liver enzymes as reliable non-invasive biomarkers for assessing periodontal destruction and systemic inflammatory burden in individuals with Stage II generalized periodontitis and Type 2 Diabetes Mellitus.

Keywords: Aspartate transaminase, alanine transaminase, alkaline phosphatase, γ-glutamyl transpeptidase, lactate dehydrogenase, type 2 diabetes mellitus (T2DM), periodontitis, biomarkers, saliva

Background:
Periodontitis is a chronic multifactorial inflammatory disease characterized by the progressive destruction of the tooth-supporting tissues caused by dysbiotic biofilms and an exaggerated host inflammatory response **[1-3]**. It remains one of the leading causes of tooth loss in adults and has been increasingly recognized as a condition with significant systemic implications. Among the systemic diseases associated with periodontitis, Type 2 Diabetes Mellitus (T2DM) has the strongest evidence supporting a bidirectional relationship **[4].** Individuals with diabetes exhibit an approximately threefold higher risk of developing periodontitis because of impaired immune function, altered collagen metabolism and delayed wound healing **[5-9]**. Conversely, chronic periodontal inflammation contributes to increased systemic inflammatory burden, aggravates insulin resistance and compromises glycemic control in patients with diabetes **[10, 11]**. The biological link between periodontitis and diabetes is mediated by persistent dissemination of periodontal pathogens and their virulence factors, including lipopolysaccharides, into the systemic circulation **[12, 13]**. This inflammatory challenge stimulates the production of cytokines such as interleukin (IL)-1β and tumor necrosis factor (TNF)-α, together with excessive generation of reactive oxygen species, resulting in sustained oxidative stress and tissue injury **[14, 15]**. These inflammatory mediators can influence hepatic immune responses, promote hepatocellular inflammation and induce metabolic disturbances. Consequently, both diabetes and periodontitis have been associated with altered liver function, reflected by increased extracellular release of intracellular enzymes including Alanine transaminase (ALT), Aspartate transaminase (AST), Alkaline phosphatase (ALP), γ-glutamyl transpeptidase (GGT) and Lactate dehydrogenase (LDH). Serum has traditionally been regarded as the reference biological fluid for biochemical assessment of these enzymes. Recent advances in salivary diagnostics have further highlighted saliva as a non-invasive and accessible biofluid for disease screening and monitoring **[16]**. Saliva has emerged as a valuable diagnostic biofluid because it contains serum-derived constituents together with locally produced inflammatory mediators originating from gingival crevicular fluid and periodontal tissues **[17-20]**. Jeyasree *et al.* further showed that salivary and serum ALP levels decreased following non-surgical periodontal therapy, supporting their potential utility for monitoring periodontal disease activity **[21]**. Ridha & Kadri also reported elevated serum ALT, AST, ALP and GGT levels in patients with chronic periodontitis, particularly with greater disease severity, although only salivary AST showed a significant increase **[22].** Therefore, it is of interest to evaluate and compare the levels of serum and salivary liver enzymes as potential biomarkers among uncontrolled T2DM patients with Stage II generalized periodontitis.

Materials and Methods:
Study design:
This observational, cross-sectional, clinico-biochemical study was conducted at the Department of Periodontology, Rama Dental College Hospital & Research Center, Kanpur, over a period of 20 months. The study protocol was approved by the Institutional Ethical Committee and written informed consent was obtained from all participants.

A total of 120 participants (aged 35–65 years) were selected and equally divided into four groups (n=30):

[1] Group I (study group) – Consisting of 30 systemically healthy subjects with Stage II Generalized Periodontitis.
[2] Group II (study group) - Consisting of 30 controlled type 2

Diabetes Mellitus subjects with Stage II Generalized Periodontitis.

[3] Group III (study group) - Consisting of 30 uncontrolled type 2 Diabetes Mellitus subjects with Stage II Generalized Periodontitis.

[4] Group IV (control group) – Consisting of 30 clinically healthy periodontium subjects with no gingival inflammation, good oral hygiene and no history of Periodontal disease.

Inclusion criteria:

Participants with a minimum of 20 remaining teeth were included. Stage II Generalized Periodontitis was defined per the 2017 World Workshop classification **[23, 24]** (PPD ≤ 5mm, mostly horizontal bone loss, CAL 3–4 mm).

The full study flowchart is given in **Figure 1.**

Exclusion criteria:

Patients who received antibiotics within the past two months, pregnant/lactating women, smokers/tobacco chewers, alcoholics, patients with a history of periodontal surgery and those on NSAIDs or oral contraceptives.

Clinical examination:

Periodontal status was assessed utilizing the Plaque Index (PI), Gingival Index (GI), modified Sulcular Bleeding Index (mSBI), Probing Pocket Depth (PPD) and Clinical Attachment Level (CAL) using a UNC-15 periodontal probe.

Biochemical evaluation:

Participants were instructed to fast for 2 hours prior to sample collection. Between 9:00 AM and 10:00 AM, 5 mL of unstimulated whole saliva was collected via the spitting method. Concurrently, 5 mL of venous blood was drawn from the antecubital vein. Samples were centrifuged at 3000 rpm for 5 minutes. The serum and salivary supernatants were preserved at -80°C until further analysis. Levels of AST, ALT, ALP, GGT and LDH were quantified utilizing the International Federation of Clinical Chemistry (IFCC) kinetic method on a semi-auto analyzer (ERBA Chem 5 plus V2).

Statistical analysis:

Data were analyzed using SPSS version 26.0. Continuous data were expressed as mean ± standard deviation (SD). One-way ANOVA followed by Tukey's HSD post-hoc test was utilized to compare mean values across the four groups. Pearson's correlation coefficient (*r*) was employed to evaluate the relationship between HbA1c, clinical parameters and enzyme levels. A p-value < 0.05 was considered statistically significant.

Results:

A consistent, highly significant stepwise deterioration ($p < 0.0001$) in all clinical parameters (PI, GI, mSBI, PPD, CAL) was observed moving from Group IV to Group III as shows in **Table 1** and **Figure 2**. Tukey's post-hoc analysis revealed that while PPD did not differ significantly between controlled (Group II) and uncontrolled (Group III) diabetics ($p = 0.4713$), CAL showed a statistically significant difference between the two ($p = 0.0429$), indicating that CAL is a more sensitive indicator of cumulative tissue destruction mediated by dysglycemia. All five liver-related enzymes (AST, ALT, ALP, LDH, GGT) were significantly elevated in both serum and saliva across the disease groups compared to healthy controls ($p < 0.0001$). Peak values were consistently recorded in Group III (uncontrolled T2DM with periodontitis). Notably, the F-values for salivary enzymes from the ANOVA (ranging from 911.66 to 6792.79) were dramatically higher than those for serum enzymes (74.07 to 422.10), indicating that salivary enzymes possess a vastly superior discriminatory capacity between healths, periodontitis and varying degrees of glycemic control. Pearson's correlation analysis showed very strong positive correlations between matched serum and salivary levels for all five enzymes: AST ($r = 0.961$), ALT ($r = 0.948$), ALP ($r = 0.897$), LDH ($r = 0.843$) and GGT ($r = 0.810$) (all $p < 0.0001$) shows in (**Table 2 and Figure 3).** Among the diabetic cohorts (Groups II and III), worsening glycemic control (higher HbA1c) strongly correlated with elevated salivary enzymes: LDH ($r = 0.962$), ALP ($r = 0.954$), ALT ($r = 0.953$), AST ($r = 0.910$) and GGT ($r = 0.863$). These correlations were substantially stronger than those observed between HbA1c and serum enzymes ($r = 0.373$ to 0.620). A complete overview of the results for all clinical and biochemical parameters is provided in (**Table 3 and Figure 4)**. For each outcome, this table shows which group had the highest mean, which had the lowest mean and whether the between-group difference was statistically significant. For every single one of the 16 outcomes examined in this study, Group III (uncontrolled T2DM + Stage II periodontitis) had the highest values and Group IV (periodontally healthy controls) had the lowest values. Every between-group comparison was highly significant ($p<0.0001$), whether tested by one-way ANOVA (for four-group comparisons) or Welch's t-test (for the two-group HbA1c comparison). This perfect consistency across all outcomes — clinical indices, serum enzymes, salivary enzymes and glycemic markers — provides strong and unified support for the central idea of this study: periodontitis and type 2 diabetes mellitus, especially when diabetes is poorly controlled, are linked to significantly elevated levels of liver-related enzymes in both blood and saliva. The extent of elevation is proportional to the severity of both gum disease and metabolic imbalance.

Table 1: Clinical periodontal parameters across study groups (Descriptive; Mean ± SD)

Parameter	Group I	Group II	Group III	Group IV
PI	2.02 ± 0.38	2.11 ± 0.28	2.98 ± 0.10	0.63 ± 0.49
GI	2.45 ± 0.46	2.46 ± 0.49	2.67 ± 0.48	0.00 ± 0.00
mSBI	2.03 ± 0.41	2.30 ± 0.53	2.67 ± 0.48	0.00 ± 0.00
PPD (mm)	4.27 ± 0.45	4.63 ± 0.49	4.80 ± 0.41	1.23 ± 0.43
CAL (mm)	3.27 ± 0.45	3.57 ± 0.50	3.83 ± 0.38	0.00 ± 0.00

PI = Plaque Index; GI = Gingival Index; mSBI = Modified Sulcus Bleeding Index; PPD = Probing Pocket Depth; CAL = Clinical Attachment Level; SD = Standard deviation

Table 2: Serum–saliva association for liver enzymes (Pearson correlation; pooled across all groups)

Enzyme	Serum–Saliva Pearson r	p-value	n
ALT	0.948	<0.0001*	119

AST	0.961	<0.0001*	120
ALP	0.897	<0.0001*	120
LDH	0.843	<0.0001*	119
GGT	0.810	<0.0001*	120

** p<0.05 considered statistically significant; r = Pearson's correlation coefficient; n = number of paired observations*

Table 3: Overall summary of group-wise differences (Highest vs lowest mean; inferential p-value)

Outcome	Highest mean group	Highest mean	Lowest mean group	Lowest mean	Test	p-value
PI	Group III	2.98	Group IV	0.63	One-way ANOVA	<0.0001*
GI	Group III	2.67	Group IV	0.00	One-way ANOVA	<0.0001*
mSBI	Group III	2.67	Group IV	0.00	One-way ANOVA	<0.0001*
PPD	Group III	4.80	Group IV	1.23	One-way ANOVA	<0.0001*
CAL	Group III	3.83	Group IV	0.00	One-way ANOVA	<0.0001*
ALT (Serum)	Group III	63.85	Group IV	11.63	One-way ANOVA	<0.0001*
AST (Serum)	Group III	63.63	Group IV	12.01	One-way ANOVA	<0.0001*
ALP (Serum)	Group III	259.98	Group IV	74.00	One-way ANOVA	<0.0001*
LDH (Serum)	Group III	554.20	Group IV	169.46	One-way ANOVA	<0.0001*
GGT (Serum)	Group III	62.49	Group IV	27.32	One-way ANOVA	<0.0001*
ALT (Saliva)	Group III	67.10	Group IV	13.04	One-way ANOVA	<0.0001*
AST (Saliva)	Group III	196.96	Group IV	55.23	One-way ANOVA	<0.0001*
ALP (Saliva)	Group III	263.08	Group IV	52.79	One-way ANOVA	<0.0001*
LDH (Saliva)	Group III	615.92	Group IV	268.06	One-way ANOVA	<0.0001*
GGT (Saliva)	Group III	64.56	Group IV	21.70	One-way ANOVA	<0.0001*
HbA1c	Group III	8.85	Group II	6.71	Welch's t-test	<0.0001*

Note: All statistical analyses were two-tailed; p<0.05 was considered statistically significant. ANOVA = Analysis of Variance; HSD = Honestly Significant Difference; SD = Standard Deviation; CI = Confidence Interval; η^2 = partial eta-squared; r = Pearson's correlation coefficient; df = degrees of freedom; T2DM = Type 2 diabetes mellitus; HbA1c = Glycated hemoglobin; PI = Plaque Index; GI = Gingival Index; mSBI = Modified Sulcus Bleeding Index; PPD = Probing Pocket Depth; CAL = Clinical Attachment Level; ALT = Alanine aminotransferase; AST = Aspartate aminotransferase; ALP = Alkaline phosphatase; LDH = Lactate dehydrogenase; GGT = Gamma-glutamyl transferase; U/L = Units per liter.

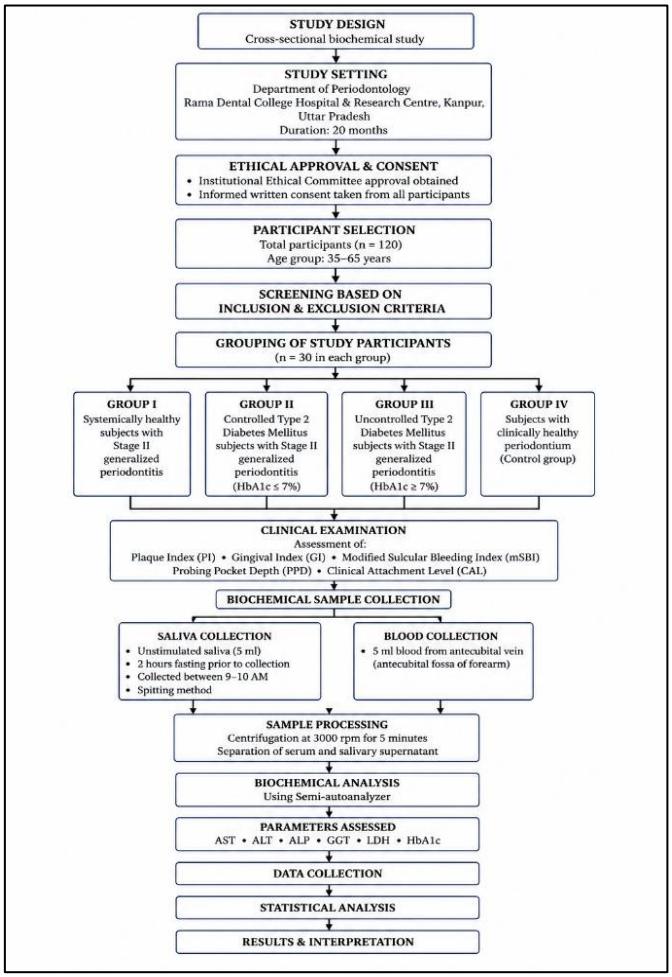


Figure 1: Flowchart of the study

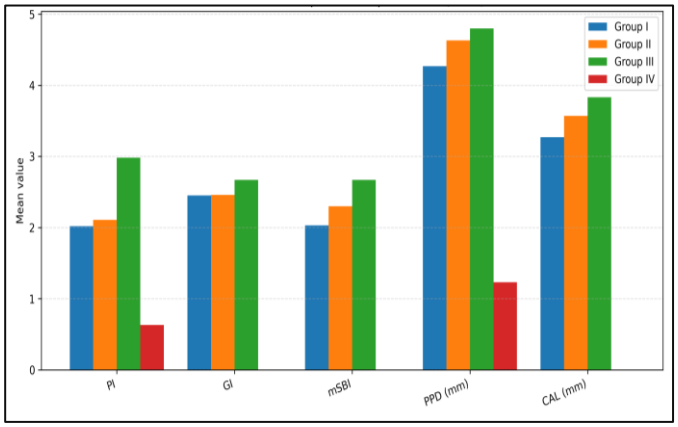


Figure 2: Grouped bar chart showing mean PI, GI, mSBI, PPD and CAL across Groups I–IV

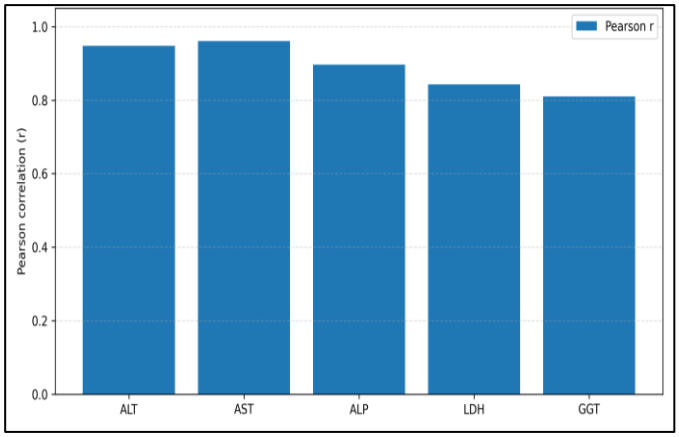


Figure 3: Bar chart showing pooled Pearson correlation coefficients (r) between matched serum and salivary enzyme levels

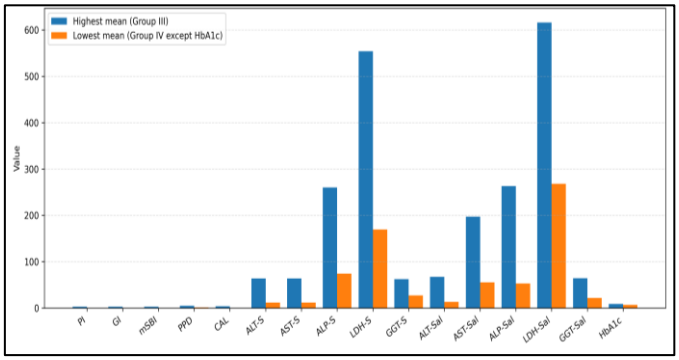


Figure 4: Grouped bars summarizing highest and lowest mean values for each outcome across study groups

Discussion:

The present study reinforces the well-established bidirectional relationship between periodontitis and Type 2 Diabetes Mellitus (T2DM) and further shows that both conditions are associated with alterations in hepatic enzyme levels **[25, 26]**. These findings support the concept that chronic periodontal inflammation extends beyond the oral cavity and contributes to systemic metabolic disturbances **[27]**. The progressive elevation of serum AST, ALT, ALP, LDH and GGT observed in periodontitis patients without diabetes (Group I) indicates that periodontitis itself may contribute to subclinical hepatic stress. Periodontal pathogens and their virulence factors can enter the systemic circulation and reach the liver through the portal venous system **[26]**. This process activates Kupffer cells and stimulates the production of pro-inflammatory cytokines, including TNF-α and IL-1β, resulting in mild hepatocellular injury and increased leakage of intracellular liver enzymes **[28]**. In uncontrolled diabetic patients (Group III), these inflammatory mechanisms are further amplified by insulin resistance and persistent hyperglycaemia **[26]**. In addition, the accumulation of Advanced Glycation End-products (AGEs) and diabetes-associated hepatic steatosis promotes oxidative stress and hepatocellular dysfunction, explaining the highest enzyme levels observed in this group **[29]**. A noteworthy finding of the present study is the superior discriminatory ability and stronger correlation of salivary enzymes with HbA1c when compared with serum enzymes. This observation may be explained by a mechanism of dual amplification. Hepatic injury associated with uncontrolled diabetes results in increased circulating liver enzymes that subsequently diffuse into saliva. Simultaneously, chronic periodontal inflammation causes extensive local tissue destruction, releasing intracellular enzymes such as LDH and AST, together with bone turnover markers including ALP, directly into the oral environment. Consequently, salivary biomarkers reflect both systemic metabolic alterations and local periodontal tissue destruction, thereby producing a more sensitive biochemical profile **[30]**. More recently, Desai *et al.* reported progressively higher salivary and serum ALP levels across healthy, diabetic and diabetic-periodontitis groups, with strong correlations between ALP and periodontal parameters such as probing depth and clinical attachment loss **[31].** The strong positive correlation observed between serum and salivary enzyme concentrations ($r > 0.80$ for all enzymes) has important clinical implications. These findings suggest that saliva may serve as a reliable surrogate for serum in evaluating liver enzyme alterations associated with periodontitis and uncontrolled diabetes **[25]**. A simple, painless and non-invasive salivary test could therefore facilitate simultaneous screening for periodontal disease activity and underlying metabolic disturbances while reducing the need for repeated venipuncture **[27]**. The present study has certain limitations. Its cross-sectional design precludes establishing a causal relationship between periodontal disease progression, glycaemic status and hepatic enzyme alterations. Furthermore, potential confounding factors such as body mass index, antidiabetic medications and the presence of hepatic steatosis were not evaluated. Future multicentre longitudinal studies incorporating imaging modalities and adjustment for these variables are required to validate the diagnostic and prognostic value of salivary liver enzymes in patients with periodontitis and T2DM.

Conclusion:

Stage II generalized periodontitis was associated with significantly elevated serum and salivary liver enzyme levels, with the highest concentrations observed in uncontrolled Type 2 Diabetes Mellitus. Salivary liver enzymes showed strong correlations with corresponding serum enzyme levels and glycated hemoglobin, indicating their value in reflecting periodontal and metabolic status. Thus, data support salivary liver enzymes as promising non-invasive biomarkers for periodontal assessment, warranting validation through larger longitudinal studies

References:

[1] Pihlstrom BL *et al. Lancet.* 2005 **366**:1809. [PMID: 16298220]
[2] Kinane DF *et al. Nat Rev Dis Primers.* 2017 **3**:17038. [PMID: 28805207]
[3] Page RC & Kornman KS. *Periodontol 2000.* 1997 **14**:9. [PMID: 9567963]
[4] Petersen PE & Ogawa H. *Periodontol 2000.* 2012 **60**:15. [PMID: 22909104]
[5] Lalla E & Papapanou PN. *Nat Rev Endocrinol.* 2011 **7**:738. [PMID: 21709707]
[6] Mealey BL *et al. J Periodontol.* 2006 **77**:1289. [PMID: 16881798]
[7] Preshaw PM *et al. Diabetologia.* 2012 **55**:21. [PMID: 22057194]
[8] Polak D & Shapira L. *J Clin Periodontol.* 2018 **45**:150. [PMID: 29280184]
[9] Casanova L *et al. Br Dent J.* 2014 **217**:433. [PMID: 25342350]
[10] Engebretson S & Kocher T. *J Periodontol.* 2013 **84**:S153. [PMID: 23631575]
[11] Borgnakke WS *et al. J Periodontol.* 2013 **84**:S135. [PMID: 23631574]
[12] Chapple IL. *J Clin Periodontol.* 1997 **24**:287. [PMID: 9178107]
[13] Gaffen SL & Hajishengallis G. *J Dent Res.* 2008 **87**:817. [PMID: 18719207]

[14] Graves D. *J Periodontol.* 2008 **79**:1585. [PMID: 18673014]
[15] Dixon DR & Darveau RP. *J Dent Res.* 2005 **84**:584. [PMID: 15972584]
[16] Xia X *et al. Coord Chem Rev.* 2026 **549**:217351. [DOI: 10.1016/j.ccr.2025.217351]
[17] Taba M Jr *et al. Dent Clin North Am.* 2005 **49**:551. [PMID: 15978241]
[18] Nomura Y *et al. Arch Oral Biol.* 2012 **57**:413. [PMID: 22030151]
[19] Ebersole JL *et al. Sci Rep.* 2024 **14**:30649. [PMID: 39730430]
[20] Ji S & Choi Y. *Front Cell Infect Microbiol.* 2015 **5**:65. [PMID: 26389079]
[21] Jeyasree RM *et al. J Indian Soc Periodontol.* 2018 **22**:487. [PMID: 30631226]
[22] Ridha HSH & Kadri ZHM. *J Biotechnol Res Cent.* 2020 **14**:20. [DOI: 10.24126/jobrc.2020.14.1.584]
[23] Tonetti MS *et al. J Periodontol.* 2018 **89**:S159. [PMID: 29926952]
[24] Papapanou PN *et al. J Periodontol.* 2018 **89**:S173. [PMID: 29926951]
[25] Lalla E *et al. Ann Periodontol.* 2001 **6**:113. [PMID: 11887453]
[26] Wiener RC *et al. Int J Inflam.* 2016 **2016**:3901402. [PMID: 26981311]
[27] Kim J *et al. J Periodontol.* 2020 **91**:903. [DOI: 10.1038/s41598-020-60797-7]
[28] Zhang J *et al. J Dent Sci.* 2025 **20**:1499. [PMID: 40654451]
[29] Fujii T *et al. J Clin Med.* 2023 **12**:4759. [PMID: 37510874]
[30] Maftei GA *et al. Antioxidants (Basel).* 2021 **10**:1741. [PMID: 34829612]
[31] Desai K *et al. Bioinformation.* 2025 **21**:4559. [PMID: 41907941]

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