

Evaluation of Salivary Cortisol Levels and Periodontal Inflammation during Fixed Orthodontic Treatment: A Prospective Clinical Study

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ABSTRACT

Background: Orthodontic treatment is associated with both physical and psychological stress, which may influence periodontal health through inflammatory changes. Salivary cortisol is considered a reliable biomarker for evaluating stress responses during orthodontic therapy. **Aim:** To evaluate salivary cortisol levels and their association with periodontal inflammation during fixed orthodontic treatment. **Materials and Methods:** This prospective clinical study was conducted among 30 patients undergoing fixed orthodontic treatment. Periodontal parameters including Plaque Index (PI), Gingival Index (GI), Bleeding on Probing (BOP), and Probing Pocket Depth (PPD) were assessed at baseline, 24 hours after appliance activation, and one month after treatment initiation. Unstimulated saliva samples were collected at each interval and analyzed for salivary cortisol levels using Enzyme-Linked Immunosorbent Assay (ELISA). Statistical analysis was performed to evaluate changes in cortisol levels and periodontal parameters and their correlation. **Results:** An increase in salivary cortisol levels and periodontal inflammatory parameters was observed following orthodontic appliance activation, particularly during the initial phase of treatment. A positive correlation was noted between elevated cortisol levels and periodontal inflammation. **Conclusion:** Fixed orthodontic treatment may induce transient stress-related increases in salivary cortisol levels, which may be associated with mild periodontal inflammatory changes. Assessment of stress biomarkers alongside periodontal monitoring may aid in improving orthodontic treatment outcomes.

Keywords: Salivary cortisol; Orthodontic treatment; Periodontal inflammation

INTRODUCTION: ORTHODONTIC TREATMENT INVOLVES

the application of controlled mechanical forces to achieve tooth movement and improve esthetics and function.¹ Although fixed orthodontic appliances provide effective treatment outcomes, they may also contribute to physical discomfort, difficulty in maintaining oral hygiene, and psychological stress during the initial phases of therapy. These factors can influence periodontal health and inflammatory responses in the oral cavity.²⁻⁴

Periodontal inflammation during orthodontic treatment is commonly associated with increased plaque accumulation

around brackets and wires, leading to gingival irritation and bleeding.⁵ In addition, stress-related hormonal changes may further modulate the inflammatory response. Cortisol, a glucocorticoid hormone released by activation of the hypothalamic-pituitary-adrenal axis, is considered a reliable biomarker for assessing physiological and psychological stress. Salivary cortisol measurement is a non-invasive and convenient method widely used in clinical research.⁶⁻⁸

Previous studies have suggested that orthodontic treatment may lead to transient elevations in salivary cortisol levels, particularly during appliance activation and the early stages of treatment. Elevated cortisol levels may influence

immune response and periodontal status by altering inflammatory mediator activity. However, limited studies have evaluated the association between salivary cortisol levels and periodontal inflammation during fixed orthodontic therapy.

Therefore, the present study was designed to evaluate salivary cortisol levels and their correlation with periodontal inflammatory parameters in patients undergoing fixed orthodontic treatment.

Materials and

METHODS

Study Design and Ethical Approval: The present prospective clinical study was conducted in the Department of Orthodontics in collaboration with the Department of Periodontology after obtaining approval from the Institutional Ethical Committee. The study was carried out in accordance with the ethical principles outlined in the Declaration of Helsinki. Written informed consent was obtained from all participants prior to inclusion in the study.

Study Population: A total of 30 patients requiring fixed orthodontic treatment were selected from the outpatient department. The study population consisted of systemically healthy individuals aged between 18 and 30 years.

RESULTS: THIRTY PATIENTS UNDERGOING FIXED

orthodontic treatment were evaluated for salivary cortisol levels and periodontal inflammatory parameters at baseline (T0), 24 hours after appliance activation (T1), and one month after treatment initiation (T2).

Table 1 demonstrates the comparison of mean salivary cortisol levels and periodontal parameters at different time intervals. A statistically significant increase in salivary cortisol levels was observed at T1 compared to baseline, followed by a mild reduction at T2, although the values remained higher than baseline levels. The difference was statistically significant ($p < 0.001$).

Similarly, periodontal parameters including Plaque Index (PI), Gingival Index (GI), Bleeding on Probing (BOP), and Probing Pocket Depth (PPD) showed a progressive increase from baseline to one month after orthodontic appliance placement. The increase in PI, GI, and BOP scores was highly statistically significant ($p < 0.001$), whereas PPD also demonstrated a significant increase ($p = 0.032$) (Table 1).

The findings indicate that initiation of fixed orthodontic treatment was associated with transient elevation in stress-related salivary cortisol levels along with mild periodontal

inflammatory changes.

Table 2 shows the correlation between salivary cortisol levels and periodontal parameters. A statistically significant positive correlation was observed between salivary cortisol levels and Plaque Index ($r = 0.61$), Gingival Index ($r = 0.69$), Bleeding on Probing ($r = 0.58$), and Probing Pocket Depth ($r = 0.42$). These findings suggest that increased stress levels during orthodontic tre

Inclusion Criteria

Patients aged between 18–30 years

Patients indicated for fixed orthodontic treatment

Systemically healthy individuals

Presence of good periodontal health at baseline

Patients with complete permanent dentition excluding third molars

Patients willing to participate and provide informed consent

Exclusion Criteria

Patients with systemic diseases affecting periodontal status

Smokers and tobacco users

Patients with active periodontal disease

Patients on antibiotics, corticosteroids, or anti-inflammatory medications within the previous three months

Pregnant or lactating females

Patients with previous orthodontic treatment

Patients with poor oral hygiene compliance

Standardization of Orthodontic Procedure: To ensure uniformity, all participants were treated using conventional fixed orthodontic appliances with MBT prescription brackets having a 0.022-inch slot. Initial alignment was performed using standardized nickel-titanium archwires. Appliance placement and activation procedures were performed by the same orthodontist under standardized clinical conditions.

Calibration and Standardization of Examiner: All periodontal examinations were carried out by a single calibrated examiner to minimize inter-examiner variability. Calibration was performed before commencement of the study, and measurements were repeated in a subset of patients to assess reproducibility and reliability.

Clinical Periodontal Assessment

Periodontal parameters were recorded at three different intervals:

T0 – Before appliance placement (baseline)

T1 – 24 hours after appliance activation

T2 – One month after initiation of treatment

The following periodontal parameters were assessed:

Plaque Index (PI) by Silness and Loe

Gingival Index (GI) by Loe and Silness

Bleeding on Probing (BOP)

Probing Pocket Depth (PPD) measured using a UNC-15 periodontal probe

All measurements were recorded under standardized illumination and clinical conditions.

Saliva Collection Procedure: Unstimulated whole saliva samples were collected from all participants at T0, T1, and T2. Participants were instructed to refrain from eating, drinking, chewing gum, or performing oral hygiene procedures for at least one hour before sample collection.

To minimize circadian variation in cortisol secretion, all saliva samples were collected between 9:00 AM and 11:00 AM. Participants were seated comfortably and instructed to allow saliva to accumulate in the floor of the mouth and expectorate into sterile graduated collection containers. Approximately 2–3 mL of saliva was collected from each participant.

Sample Processing and Storage: The collected saliva samples were immediately transported to the laboratory and centrifuged at 3000 rpm for 10 minutes to remove debris and cellular components. The supernatant obtained was transferred into sterile microcentrifuge tubes and stored at –20°C until biochemical analysis.

Estimation of Salivary Cortisol: Salivary cortisol levels were estimated using commercially available Enzyme-Linked Immunosorbent Assay (ELISA) kits according to the manufacturer's protocol. Optical density was measured using an ELISA reader, and cortisol concentrations were calculated based on the standard calibration curve.

Oral Hygiene Instructions: All participants received standardized oral hygiene instructions at baseline, including proper brushing techniques and maintenance of oral hygiene during orthodontic treatment. No adjunctive mouthwash or periodontal therapy was provided during the study period.

Statistical Analysis: The collected data were tabulated and analyzed using Statistical Package for Social Sciences (SPSS) software version 25.0. Descriptive statistics including mean and standard deviation were calculated for all variables. Comparison of periodontal parameters and salivary cortisol levels at different time intervals was

performed using repeated measures ANOVA followed by post hoc analysis. Pearson's correlation coefficient was used to assess the correlation between salivary cortisol levels and periodontal parameters. A p-value of less than 0.05 was considered statistically significant.

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