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# Increase of Transforming Growth Factor-Beta 1 in Gingival Crevicular Fluid during Human Orthodontic Tooth Movement

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## Abstract

In this study, transforming growth factor-beta 1 (TGF-beta 1) was located and measured in human gingival crevicular fluid, and alterations during orthodontic tooth movement were investigated. A total of 87 patients, with a mean age of 19.58, participated. The central incisor of each patient getting one treatment for distal migration served as the experimental tooth. Before activation as well as after 7, 21, and 90 days after the onset of tooth movement in the experimental teeth, gingival crevicular fluid from each participant was taken. TGF-beta 1 was recognized utilizing an enzyme-linked immunosorbent assay. In the experimental teeth at 7 and 21 and 90 days compared to the baseline, which was before orthodontic therapy was performed, TGF-

-beta 1 concentration was significantly greater. Additionally, TGF-beta 1 levels in the gingival crevicular fluid were influenced by patient factors like age and sex. The use of A-wire and power elastic has an impact on TGF-beta 1, with power elastic having a greater effect than A-wire on TGF-beta 1 levels in GCF. According to these findings, TGF-beta 1 is likely involved in the remodeling of bone that occurs during orthodontic tooth movement.

## Keywords

Human gingival crevicular fluid (HGCF); Orthodontic tooth movement; Transforming growth factor-beta 1 (TGF-beta 1)

## Introduction

The quality of a person's life is greatly impacted by malocclusion. Yemen ranks third after caries and periodontal disease in terms of malocclusion prevalence, at almost 80% [1-4]. Orthodontic demands in the community have grown in demand along with the advancement of knowledge society and the desire to enhance quality of life. Orthodontic therapy aimed to correct jaw alignment issues and dental abnormalities. Through tissue remodeling, it makes use of the ability of the periodontal ligament and the alveolar bone to adjust to shifting mechanical conditions [5-8]. These adaptations allow for the movement of teeth through the alveolar bone as well as the influence on distant skeletal regions. These two methods can be used to provide a stable occlusion and a healthy jaw relationship [9]. Orthodontic tooth movement is achieved through mechanical pressure remodeling of the alveolar bone and periodontal ligament. The activation of many markers for bone remodeling mediates this remodeling. Numerous researches were conducted to provide us with more details regarding this molecular signaling, including those on TIMP-1, Col-1, RANKL, IL-6, sgp-130, and others [10–14]. Transforming growth factor (TGF-), one of the growth factors, is yet unknown in terms of its function in tooth mobility during orthodontic treatment. Others discovered it may help to induce bone resorption [17,18], whereas some authors identified it to be a mediator for decreased osteoclast activity [8,15,16]. There are three distinct isoforms of TGF: TGF-β1, TGF-β2, and TGF-β3. An osteoporosis-like phenotype was discovered to be caused by excessive TGF-1 expression [19]. Given that TGF-β1 plays a role in bone remodeling, the objective of this study was to look at how TGF-β1 levels changed as a result of mechanical stress during orthodontic tooth movement. This fluctuating level of TGF-β1 was examined soon before the application of mechanical load, 7 days, 21 days, and 90 days after the mechanical load.

## Materials and Methods

The study focused on patients who had received fixed orthodontic appliances in clinics run by the orthodontic department of Sana'a University's dental faculty and the Azal dental center in Sana'a city. Data on demographics and those related to fundamental management were gathered. Additionally, an

intra-oral examination, dentition evaluation, oral hygiene evaluation, and medical history evaluation were completed.

**Study Design:** This study compares the levels of pro-inflammatory (TGF- β1) cytokines in gingival crevicular fluid (GCF), and host and material factors may have an impact on these levels in addition to the effect of orthodontic treatment. It is a longitudinal prospective clinical randomized chosen cohort study.

**Inclusion Criteria:** Yemeni female or male, 13 to 35 years old, free of any obvious genetic disorders or dental anomalies, seemingly healthy, not pregnant, not smoking, not chewing khat, and free of any systemic or chronic diseases, diagnosed as being eligible for treatment with fixed orthodontic appliances, and not having received antibiotics, corticosteroid therapy, or anti-inflammatory drugs within the past month.

**Gingival Crevicular Fluids (GCF) Collection for Cytokine Detection:** Subjects were instructed beforehand that they should refrain from eating, drinking, chewing gum, or brushing their teeth for an hour before to sample collection. Sterile Paper-points strips (PAPER POINTS DIA-PROT, DiaDent Group, Choongchong Buk Do Republic of Korea) were inserted into the gingival fissure of the teeth and held there until a slight resistance was felt to collect gingival crevicular fluid. To avoid salivary contamination, sampling was limited to the gingival crevice of the tooth. dentist plaque was cleaned using cotton, and the tooth surfaces were dried using an air syringe attached to the dentist chair. Gently inserting paper-point strips into the sulcus with caution to prevent mechanical damage and bleeding, the strips were left in situ for 30 seconds to absorb the gingival fluids, and the gingival crevicular fluids were collected. Because contamination of strips with blood or saliva is significant and can lead to inaccurate results, contaminated samples were removed from the study. All gingival crevicular fluid samples were obtained in pre-labeled sterile Eppendorf tubes with 1.5 ml volumes from CITOTEST in China, which were then stored at -35 oC for ensuing testing and analysis.

**Finding and Measuring Pro-Inflammatory (TGF- β1) Cytokines:** To help with the elution of cytokines from each filter paper, each strip was eluted into 200 l of sterile Phosphate Buffered Saline PBS (pH 7.4). The samples were centrifuged at 1000 g for 20 minutes before the experiment was started. Following the instructions of the manufacturer (Wuhan Fine Biotech Co., Ltd. Wuhan, Hubei, China), the enzyme-linked immunosorbent assay (ELISA) was used to measure the concentration of the pro-inflammatory (TGF- β1) mediators present in the GCF.

**Data Analysis:** Epi-info (version 7) was used to enter and evaluate the data. The mean and standard deviation (SD) for the amounts of TGF- β1 during several time periods of GCF collection were used to express the data for this protein, which had a normal distribution. This method calculates the variance between the means found in two distinct samples. The difference between the mean of the reference (baseline) TGF- β1± SD and the mean of the TGF- β1 levels in GCF at 7, 21, and 90 days were calculated.
Ethical Consideration: Sana'a University's Faculty of Medicine and Health Sciences' Medical Ethics and Research Committee granted this study No. 699, dated January 24, 2021, their approval in accordance

with medical ethics. The review committee's ethical standards were followed in all procedures.

## Results

The levels of TGF-β-1 in human gums during orthodontic treatment were determined in 87 patients; 33 (37.9%) males, 54 (62.1%) females, and their ages ranged from 12 to 34 years, with a mean ± SD equal to 19.58 ± 4.4 years. TGF-β-1 levels differed from each other at different treatment periods as measures of TGF-β-1 central tendency was elevated after orthodontic treatment was applied. Whereas, the mean ± SD for TGF-β-1 increased from 47.83 ± 4.83 pg/mL at baseline to 50.5 ± 5.8 pg/mL after 7 days, to 52.3 ± 6.2 pg/mL after 21 days, and then slightly rise to 52.8 ± 7.1pg/mLl 90 days after orthodontic treatment (Table 1). Considering of sex in TGF-β-1 levels at different treatment periods as measures of TGF-β-1 were elevated in female patients comparing to male patients after orthodontic treatment was applied. Whereas, the mean ± SD for TGF-β-1 increased in females from 47.83 ± 4.83 pg/mL at baseline to 52.8 ± 6 pg/mL after 7 days, then decrease to 49.9 ± 4.9 pg/mL after 21 days, and then rise to 55.5 ± 7.4pg/mLl 90 days after orthodontic treatment, while male patients had lower values than these values (Table 1). Considering of age groups the TGF-β-1 levels at different treatment periods as were very low at the base line for <16 years group (26.83 ± 4.29 pg/mL) then sudden rise to 39.68 ± 10.74 pg/mL at after 7 days of applying orthodontic treatment, continue rise to 42.37 ± 10.81 pg/mL after 21 days, and then decrease to 32.43 ± 5.64pg/mL in 90 days after orthodontic treatment, while older age group (26-34 years) patients had higher values in the base line (37.27 ± 4.98 pg/mL ). The highest value was recorded after 90 days of orthodontic treatment (54.84± 11.92 pg/mL) for this age group (Table 1).

| Characteristics | Number | (%) |
|---|---|---|
| Gender | | |
| Male | 33 | 37.9 |
| Female | 54 | 62.1 |
| Age groups in Years | | |
| <16 | 16 | 18.4 |
| 16 -25 | 62 | 71.3 |
| 26 -34 | 9 | 10.3 |
| Total | 87 | 100 |
| Mean | 19.58 years | - |
| SD | 4.4 years | |
| Mode | 17 years | |
| Median | 18 years | |
| Minimum -Maximum | 12-34 years | |

**Table 1:** Characteristics of patients, tested for TGF-β-1 Levels in the Human Gingival Sulcus during Orthodontic Treatment.

Regarding the impact of the wire types used in orthodontic treatment, patients who used A wire had greater TGF- β -1 levels than those who used power elastic (54.5 ±7.4 pg/mL in the 90 days following treatment against 52± 6.5 pg/mL in the same period for power elastic-using patients). Additionally, in

each of the three intervals, A wire patient values were higher than power elastic patient values (Table 2). Given the impact of gum bleeding following orthodontic treatment, there were greater TGF- β-1 levels in individuals with bleeding compared to non-bleeding patients (55.9 ±11.28 pg/mL vs. 44.65± 9.42 pg/mL after 21 days). Additionally, in the three intervals following orthodontic treatment, all bleeding patient values were higher than those of non-bleeding patients (Table 2).

| - | Baseline | 7 d | 21 d | 90 d | Baseline | 7 d | 21 d | 90 d | Baseline | 7 d | 21 d | 90 d |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Characters | - | - | - | - | - | - | - | - | - | - | - | - |
| Sex | Total n=87 | - | - | - | Males n=33 | - | - | - | Female n=54 | - | - | - |
| Mean | 47.83 | 51 | 52 | 52.8 | 47.9 | 50 | 48 | 52 | 47.9 | 52.8 | 50 | 56 |
| SD | 5.2 | 5.8 | 6 | 7.1 | 5.7 | 5.7 | 5.7 | 6.5 | 4.9 | 6 | 5 | 7 |
| Type of wire | Total n= 87 | - | - | - | Power elastic n= 39 | - | - | - | A wire n= 63 | - | - | - |
| Mean | 47.83 | 51 | 52 | 52.8 | 47.9 | 50 | 48 | 52 | 47.9 | 51.8 | 52 | 55 |
| SD | 5.2 | 5.8 | 6 | 7.1 | 5.7 | 5.7 | 5.7 | 6.5 | 4.9 | 6 | 5 | 7 |
| Age groups | < 16 years group n=16 | - | - | - | 16-25 years n=62 | - | - | - | 26-34 years n=10 | - | - | - |
| Mean | 26.83 | 40 | 42 | 32.4 | 32.43 | 33 | 43 | 45 | 37.3 | 33.77 | 52 | 54..84 |
| SD | 4.29 | 11 | 11 | 5.64 | 5.64 | 4.1 | 8.6 | 9.6 | 4.98 | 3.96 | 11 | 12 |
| Gam bleeding | Total n=87 | - | - | - | Bleeding n=12 | - | - | - | Non-bleeding | - | - | - |
| Mean | 47.83 | 51 | 52 | 52.8 | 35.42 | 54 | 56 | 41 | 31.7 | 41.93 | 45 | 36 |
| SD | 5.2 | 5.8 | 6 | 7.1 | 4.64 | 13 | 11 | 6.9 | 4.67 | 8.79 | 9 | 5 |

**Table 2:** TGF-β-1 concentrations (pg/ml) for total patients and the effect of sex, type of wires, age groups and gum bleeding in TGF-β-1 Levels in human gingival Sulcus during orthodontic treatment.

## Discussion

Orthodontic tooth movement is a useful way to learn how mechanical load causes bone remodeling. The tooth was displaced during orthodontic treatment as a result of the force used. According to the pressure and tension theory, this force will result in bone growth in the stretched area and bone resorption in the compressed area. The strength of the applied force and the biological reactions of the periodontal ligament (PDL) can regulate this movement. The stress exerted to the teeth will alter blood flow, which will lead to the release of several inflammatory mediators like growth factors, cytokines, colony-stimulating factors, arachidonic acid metabolites and neurotransmitters. These secretions lead to the remodeling of the bone [8,20]. The gap between basic research and clinical implications would be bridged by monitoring the biological system in clinical settings. If the biological system's reactions could be watched throughout therapy, it would be helpful for the doctor and the patient. This would make it possible to modify the treatment to fit the patient's biological needs. TGF-β, a growth factor with several

functions, was involved in bone remodeling. Along with activin, nodal, bone morphogenetic proteins (BMP), and others, TGF is a member of the TGF-super family. There are three different isoforms of TGF: TGF-β1, TGF-β2, and TGF-β3. Uncertainty exists regarding its function in orthodontic tooth movement [17,18]. It has the same level in both the compression and tension location, according to other researchers [12]. TGF- β 2 expressed later than TGF- β 1 and TGF- β 3 throughout development. [21]. However, tooth movement starts 7 days after the application of mechanical force as a result of osteoblast and osteoclast remodeling the bone socket [20]. This is the primary justification for the study's choice of time interval.

GCF sampling was taken from compression site because according to Erlebacher et al., [19] over expression of TGF-β1 will cause an osteoporosis-like phenotype. Thus, it can be concluded that TGF-β1 may play role in bone resorption. We have detected a level of TGF-β1 before application of mechanical load, this might be because there still remain a force used for leveling and aligning. At 7 days after mechanical load, there was a significant increase level of TGF-β1. Its increase was continued until 21 days and 90 days after mechanical load. Whereas, the mean ± SD for TGF-β-1 increased from 47.83 ± 4.83 pg/mL at baseline to 50.5 ± 5.8 pg/mL after 7 days, to 52.3 ± 6.2 pg/mL after 21 days, and then slightly rise to 52.8 ± 7.1pg/mLl 90 days after orthodontic treatment (Table 1). This result was different from Uematsu et al because they found that in 24 hours after mechanical loading, TGF-β1 was reached its peak level before continued to decrease at 7 days after mechanical loading [17]. However, it remains unknown whether TGF-β1 exhibits activity similar to that of TGF-β2 [22].

Considering of age groups the TGF-β-1 levels at different treatment periods as were very low at the base line for <16 years group (26.83 ± 4.29 pg/mL) then sudden rise to 39.68 ± 10.74 pg/mL at after 7 days of applying orthodontic treatment, continue rise to 42.37 ± 10.81 pg/mL after 21 days, and then decrease to 32.43 ± 5.64pg/mL in 90 days after orthodontic treatment, while older age group (26-34 years) patients had higher values in the base line (37.27 ± 4.98 pg/mL ). The highest value was recorded after 90 days of orthodontic treatment (54.84± 11.92 pg/mL) for older patient group (Table 1). In recent years, there has been an increase in the demand for adult orthodontic treatment. However, we still don't fully understand how effectively adult teeth move. The general consensus among orthodontists is that adult patients or patients who are not growing will require longer treatment times than children. Younger patients' teeth moved more than those of adults. Another investigation revealed that juvenile animals moved their teeth more quickly initially than adult animals [10]. This result differs from one made by Ren [11], who discovered that older rats' crevicular fluid cells responded to orthodontic force less favorably than their younger counterparts. This result contrasts with those observed by Yakovlev et al. [23], who examined cytokines in periodontal biopsies from various age groups and discovered higher cytokine levels in younger age groups as opposed to older age groups as shown by the current study's findings. The discrepancies from our results could be explained by gum tissue sample rather than GCF.

Regarding the impact of the wire types used in orthodontic treatment, there were greater TGF- β -1 levels in patients who used A wire versus power elastic type (54.5± 7.4 pg/mL against 52 ± 6.5 pg/mL after 90 days for patients using power elastic). Also all values of A wire patients were higher than that of power elastic patients in the 3 intervals (Table 1). The results were statistically significant after applying

the elastic force with a greater force rather than attempting to apply a slight flattening force via the arc wire, and the scores were generally high with some individual variances visible. This conclusion, however, contrasts with that reported by Basarana et al. [24] because there were no variations in the levels of cytokines produced when the various leveling forces were applied to individuals who had received orthodontic treatment [24]. Additionally, Tzannetou et al. [25] widened the palate by using the low and high powers of the upper teeth. Setting the spacer created the lesser forces, while the palatal expansion equipment provided the higher forces. With both levels of strength being equal, they noticed elevated TGF- β-1 levels. Additionally, Kee-Joon Lee et al. [26] shown that mean TGF- β--1 concentration rise in almost identical quantities in the first 24 hours following continuous and intermittent pressures. It is possible to say that low stresses can start the processes of bone resorption.

So, to achieve faster movement of the teeth, there is no need to use heavy forces, which can be harmful to other tooth structures (eg. root resorption). This means that levels of TGF-β-1 may return to baseline when a light straightening force is applied rather than a heavy retraction force and so it is recommended to use light force during orthodontic treatments [27]. After 21 days of orthodontic treatment, consider how TGF- β -1 (pg/ml) concentrations in human Sulcus gingiva are affected by bleeding during orthodontic treatment. TGF- β -1 (pg/) concentrations significantly increased in individuals who were bleeding (55.9 ±11.28 vs. 44.65± 9.42 pg/mL in the non-bleeding group). The current findings differ from those published by Grant et al. [28], in which no relationships between bleeding indicators for cytokine levels were discovered.

## Conclusion

TGF-β-1 (pg/ml) levels in periodontal tissues are raised by flattening and artificial dentition, which can be seen in GCF. Throughout orthodontic treatment, gum health is crucial. Changes in the gum tissue result from forces of various directions, lengths of time, and dimensions. During tooth movement caused by forced orthodontics, TGF-β-1 (pg/ml) levels are released. The findings of this study are consistent with the idea that pro-inflammatory cytokines are strongly involved in bone resorption following the application of orthodontic force, with a peak occurring in the fourth week after application and a subsequent fall.

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## Conflict of Interest

No conflict of interest associated with this work.

## Author's Contributions

Omar Ahmed Ismael Al-dossary, the researcher who oversaw the research for this study, had the original idea, produced the first draft of the article, and collaborated with other authors to examine the data, and writing the article.

## References


1. Sharfuddin AH, Alphameric BH, AL-Haddad KA, Al-Najhi MMA, Al-Shammah HA. (2023) The effect of dental implants on increasing the colonization rate of aerobic bacteria in the oral cavity. Universal Journal of Pharmaceutical Research. 8(3):28-33.
2. Alaklany BA, Almotareb F, Albaham H, Al-Shamahy HA, Al-hamzi AH. (2023) Deep bite malocclusion: Exploration of the skeletal and dental factors. Universal Journal of Pharmaceutical Research. 8(2)1-6.
3. Masdoose S M H, Nasher AT, El-Zine MA, Al-Akwa AAY, Al-Shamahy HA, et al. (2021) Histologic and radiographic study of pathologic change in complete impacted third molars dental follicles. Universal Journal of Pharmaceutical Research. 6(1):1-8.
4. Al-Shami IZ, Al-Shamahy HA, Abdul Majeed ALA, Al- Ghaffari KM, Obeyah AA. (2018) Association between the salivary Streptococcus Mutans levels and dental caries experience in adult females. Online Journal of Dentistary & Oral Health. 1(1):15.
5. Al-Motareb F, Al-Labanil M, Al-Zubair N, Dhaifullah E. (2017) Prevalence of impacted canine among Yemen population in Sana'a city. International Journal of Dental Research. 5(2):148-51.
6. Shumar A. (2021) Prevalence of impacted canine and its association with other dental anomalies among population in Sana'a city, Yemen. International Arab Journal of Dentistry. 12(1):5-16.
7. Alhadi YAA, Alrahabi LM, Shaalan MA, Al-Shamahy HA. (2023) Prevalence and localization of impacted canine teeth using panoramic radiograph in a sample of Yemeni adults in Sana'a, Yemen. Universal Journal of Pharmaceutical Research. 8(3):40-44.
8. Al-dossary OAI, Al-Kholani AIM, AL-Haddad KA, Al-Najhi MMA, Al-Shamahy HA, et al. (2022) Interleukin-1β levels in the human gingival sulcus: Rates and factors affecting its levels in healthy subjects. Universal Journal of Pharmaceutical Research. 7(5):42-48.
9. Uematsu S, Mogi M, Deguchi T. (1996) Increase of transforming growth factor-beta 1 in gingival crevicular fluid during human orthodontic tooth movement. Arch Oral Biol. 41(11):1091-5.
10. Ren Y, Maltha JC, Van 't Hof MA, Kuijpers-Jagtman AM. (2003) Age effect on orthodontic tooth movement in rats. J Dent Res. 82(1): 38-42.
11. Ren Y. (2002) Cytokine changes in GCF during orthodontic tooth movement. J Clin Perio.29: 757-62.
12. Garlet TP, Coelho U, Silva JS, Garlet GP. (2007) Cytokine expression pattern in compression and tension sides of the periodontal ligament during orthodontic tooth movement in humans. Eur J Oral Sci. 115(5): 355-62.
13. Faulkner MG. (2011) Gingival crevicular fluid (GCF) levels of interleukin-6 (IL-6), soluble glycoprotein 130 (SGP), and soluble interleukin-6 R during orthodontic tooth movement. UNLB Theses Disseratations, professional papers, and capstones.
14. Junior JC, Kantarci A, Haffajee A, Teles RP, Fidel R. (2011) Matrix metalloproteinases and chemokines in the gingival crevicular fluid during orthodontic tooth movement. Eur J Orthod. 35(6):705-11.
15. Janssens K, ten Djike P, Janssens S, Van Hul W. (2005) Transforming growth factor beta1 to the bone Endocr Rev. 26(6):743-74.
16. Kanaan RA, Kanaan LA. (2006) Transforming growth factor β1, bone connection. Med Sci Monit. 12(8): 164-9.
17. Uematsu S, Mogi M, Deguchi T. (1996) Increase of Transforming growth factor-β1 in gingival crevicular fluid during human orthodontic tooth movement. Arch Oral Biol. 41:1091-95.
18. Barbieri G, Solano P, Alarcón JA, Vernal R, Rios-Lugo J, et al. (2013) Biochemical markers of bone metabolism in gingival crevicular fluid during early orthodontic tooth movement. Angle Orthod. 83(1):63-9.

19. E leacher A, Deryck R. (1996) Increased expression of TGF-β2 in osteoblast result in an osteoporosis-like phenotype. J Cell Biol. 132(1-2):195-210.
20. Singh G. (2016) Textbook of orthodontics. 2nd ed. New Delhi: Jaypee Brothers Medical Publisher Ltd 216-20.
21. Blob GC, Schiemann WP, Lodish HF. (2000) Role of transforming growth factor β in human disease. N Engl J Med. 342(18):1350-58.
22. Nishimura R. (2009) A novel role for TGF-β1 in bone remodeling. IBMS Bone Key. 6(11):434-8.
23. Yakovlev E, Kalichman I, Pesante S, Shoshan S, Barak V. (1996) Levels of cytokines and collagen type I and type III as a function of age in human gingivitis. J Periodontal. 67(8):788-93.
24. Basaran G, Ozer T, Kaya FA, Kaplan A, Hamachi O, *et al.* (2006) Interleukine-1β and Tumor Necrosis Factor-α Levels in the Human Gingival Sulcus during Orthodontic Treatment. Angle Orthodontist. 76(5):830-36
25. Cumming BR, Leo H. 1973 Consistency of plaque distribution in individuals without special homecare instruction. J Periodontal Res. 8(2):94–100.
26. Chida M, Brun JG, Tinning T, Johnson R. (1995) Calprotectin levels in oral fluid: the importance of collection site. Eury J Oral Sci. 103(1):8–10.
27. Ulcer AE, Toluol O, Ozempic N, Can M, Demirtas S. (2008) The Evaluation of Cystatin C, IL-1b, and TNF-a Levels in Total Saliva and Gingival Crevicular Fluid From 11- to 16-Year-Old Children. J Periodontal. 79 (5):54-60.
28. Grant M, Wilson J, Rock P, Chapple I. (2013) Induction of cytokines, MMP9, TIMPs, RANKL and OPG during orthodontic tooth movement European Journal of Orthodontics. 35(5): 644–51.