Journal of Dentomaxillofacial Radiology, Pathology and Surgery

Spring 2018, Volume 7, Number 1

Research Paper: Comparing Streptococcus Mutans Adhesion by Using Different Orthodontic Bracket Ligations: An In Vitro Study

Majid Shalchi[1], Arefeh Hajian-Tilaki[2], Mehrdad Sadegh Khanjani[3], Peyman Sabzgolin[4], Reihaneh Aghajani Nargesi[5*]

1. Department of Orthodontics, Faculty of Dentistry, Guilan University of Medical Sciences, Rasht, Iran.
2. Postgraduate Student of Orthodontics, School of Dentistry, Guilan University of Medical Sciences, Rasht, Iran.
3. Doctor of Medical Laboratory Science, Reference Lab, Guilan University of Medical Sciences, Rasht, Iran.
4. Faculty of Dentistry, Guilan University of Medical Sciences, Rasht, Iran.
5. Department of Prosthodontics, Faculty of Dentistry, Guilan University of Medical Sciences, Rasht, Iran.

Use your device to scan and read the article online

Citation Shalchi M, Hajian-Tilaki A, Sadegh Khanjani M, Sabzgolin P, Aghajani Nargesi R. Comparing Streptococcus Mutans Adhesion by Using Different Orthodontic Bracket Ligations: An In Vitro Study. Journal of Dentomaxillofacial Radiology, Pathology and Surgery. 2018; 7(1): 7-12.

doi: http://dx.doi.org/10.32598/3dj.7.1.7

Article info:
Received: 10 Sep 2017
Accepted: 05 Jan 2018
Available Online: 01 Mar 2018

Keywords:
Adhesiveness, Orthodontic bracket, Streptococcus mutans

ABSTRACT

Introduction: Orthodontic treatments and brackets application with various ligature methods have increased worldwide, but the growth rate and mechanism of microorganism adhesion to these ligatures is not fully discovered. The current study aimed to compare the level of streptococcus mutans adhesion to three different ligation methods.

Materials and Methods: This in vitro study was performed on 30 samples. Three different ligature methods were used and 10 samples were used in each group. In group A, conventional brackets with elastomeric ligature, in group B, conventional brackets with steel wire ligature and in group C, self-ligated brackets were used. Resin composite was condensed on the mesh surface of the brackets and cured for 40 seconds. Then, the coated samples with saliva were put into glass vials, immersed in 2 mL of streptococcus mutans suspension (×109 CFU) and incubated at 37ºC for 24 h. Then, the samples were washed 3 times with normal saline, immersed into 2 mL of normal saline and shaked for 2 min. The obtained suspension was cultured on blood agar incubated at 37ºC for 48 h and the formed colonies counted. In analysis, we performed 1-way analysis of variance with Tukey test with multiple comparison in SPSS version 16.

Results: According to the results, streptococcus mutans growth rate showed statistically significant differences in the three groups. It was minimum in the steel wire (23.80±1.40), and maximum in elastomeric ligatures (38.60±1.84).

Conclusion: Steel wire ligatures had better effect on decreasing bacterial adhesion. The findings high light to decrease the use of elastomeric ligation brackets in patients with poor oral hygiene.

*** Corresponding Author:**
Reihaneh Aghajani Nargesi, PhD.
Address: *Department of Prosthodontics, Faculty of Dentistry, Guilan University of Medical Sciences, Rasht, Iran.*
Tel: *+98 (911) 1830644*
E-mail: *r.nargesi@yahoo.com*

Journal of Dentomaxillofacial Radiology, Pathology and Surgery

1. Introduction

Patients who seek orthodontic treatment to correct dental malocclusions often have difficulty in keeping their teeth clean because of crowding. After placement of fixed orthodontic appliances, these patients are at higher risk for caries owning to increased plaque retention around brackets and bands. Orthodontic treatment with fixed appliances often interferes with oral hygiene and may jeopardize dental health [1]. There is a rich ecosystem in the oral cavity, with a countless number of microorganisms. Although both periodontal disease and dental caries are considered multifactorial diseases, the bacteria in the dental plaque are the main factor in their onset and progression [2].

Increased oral microbiota of Streptococcus mutans and Lactobacillus is associated with the onset of tooth demineralization and periodontal disease. This condition is much more frequent in orthodontic patients with greater risk of colonization by these microorganisms [3]. Despite several research studies done to determine the amount of microbial growth in the patients under orthodontic treatment, the mechanisms of these microorganisms is still unclear [4]. However, the main factor presumably is the accumulation of dental plaque and inflammatory response in the retention around the components of fixed orthodontic appliance [5].

The devices used in orthodontic appliances (bands, wires, ligatures or brackets) can promote changes in the oral environment, such as pH, amount of Streptococcus mutans, biofilm and enamel decalcification [6]. The clinical characteristics and the physical properties of the bracket types are very different and, thus, can directly influence the amount of biofilm and, consequently, gingivitis [7]. However the saliva composition and secretion rate also influence plaque formation [8]. The different components of the fixed orthodontic system may contribute to a shift in the balance of the oral ecology creating an ecological stress. Many studies have evaluated the effect of fixed orthodontic appliances on microbial flora and periodontal status; however, only a few studies have evaluated the type of ligation as a contributing factor [9].

Conventional brackets are introduced in orthodontics to use as either elastomeric or stainless steel ligature to keep the orthodontic wire inside the slot [10]. In addition, self-ligating orthodontic brackets have their own mechanism for opening and closing the slot, and do not need metal or elastomeric ligature [11]. However, several advantage and disadvantages are reported for both systems in the literature. Therefore, the current study aimed to compare three different orthodontic bracket ligations on Streptococcus mutans adhesion using in vitro conditions.

2. Materials and Methods

Thirty samples were used in this in vitro study with 3 different ligature methods. The samples were divided into 3 groups. In group A, conventional metal brackets with a 0.022-inch slot (American Orthodontics, Sheboygan, Wisconsin, USA) with elastomeric ligature (n=10), in group B, the same type of brackets with steel wires ligature (n=10), and in group C, 10 self-ligating brackets (ORMCO, Glendora, California, USA) with a 0.022-inch slot for the straight-arch technique. All brackets were maxillary first premolars brackets.

To reduce the effect of the rough surface of brackets on bacteria adhesion, Transbond XT resin composite (3M, Morovia, California, USA) was condensed on the mesh surface of brackets and exposed to light cure 850 mW/cm^2 for 40 s. To increase hygiene condition, all samples were cleaned by ultrasonic device (Mini Sono Cleaner CA 1470, KaijoDenki Co. Ltd., Tokyo, Japan) for 15 minutes and then transferred into 70% alcohol for 30 min. The sterile condition of the samples was tested by using Brain Hearth Infusion (BHI) condition for 24 h. Saliva samples were obtained from 2 healthy patients which had no medication for the last 3 months without dental caries or periodontal disease. The saliva samples were made sterile by using autoclave. Then samples coated with saliva, were placed in glass vials and immersed in 2 mL of streptococcus mutans (PTCCI 683) suspension (×109 CFU) and incubated in 37ºC and 5% CO_2 for 24 h. Samples were washed 3 times with normal saline, immersed into 2 mL of normal saline and shaken for 2 min. Obtained suspension cultured on blood agar and incubated at 37ºC with 5% CO_2 for 48 h and the colonies counted.

Bacteria load data were analyzed by 1-way analysis of variance (ANOVA) using SPSS 16.0 and presented as Mean±SD. For treatments showing a main effect by ANOVA, the study variable means were compared using Tukey HSD test. Results with P values less than 0.05 were considered as statistically significant.

3. Results

The comparison of Streptococcus mutans adhesion using different orthodontic bracket ligations is presented in Tables 1 and 2. The Streptococcus mutans counts in

Table 1. Streptococcus mutans count in different types of orthodontic bracket ligations

Type of Orthodontic Bracket Ligation	S. Mutans Count Mean±SD	P
Elastomeric ligation	38.60±1.84	
Steel wire ligation	23.80±1.40	0.001
Self-ligation	29.80±1.81	

* P was obtained from F-test using one-way ANOVA model.

Table 2. Comparison of Tukey HSD test on streptococcus mutans count of orthodontic bracket ligations

Ligation Method		Mean±SD	P	95% CI
Elastomeric Ligation	Steel wire ligation	14.80±0.76		12.92-16.68
	Self-ligation	8.80±0.76	0.001	6.92-10.68
Steel Wire Ligation	Self-ligation	-6.00±0.76	0.001	-7.88-4.12

*P values were obtained from Tukey HSD test.

different type of orthodontic bracket ligations are presented in Table 1. According to the obtained data, a significant difference was found between different types of orthodontic bracket ligations. Streptococcus mutans growth rate was 23.80±1.40 in steel wire, 29.80±1.81 in self-ligated bracket and 38.60±1.84 in elastomeric ligatures (P<0.001).

The comparison results of Tukey HSD test on streptococcus mutans count of orthodontic bracket ligations are presented in Table 2. According to the data, Mean±SD difference between elastomeric ligation vs. steel wire ligation was 14.80±0.76 (P<0.001). Also, the Mean±SD difference between elastomeric ligation vs. self-ligation was 8.80±0.76 (P<0.001). Additionally, the Mean±SD difference between steel wire ligation vs. self-ligation was -6.00±0.76 (P<0.001).

4. Discussion

Orthodontic appliances can reduce the patient's oral hygiene because of the encroach of the appliances to the gingival sulcus. After placement of fixed orthodontic appliances, gingival inflammation is usually noticed and the degree of oral hygiene during treatment can affect the periodontal status of the patient. Even in patients who have perfect oral hygiene, moderate gingivitis is noticed a few months after placement of the appliances. It is believed that these changes in the periodontal status are reversible [12]. Orthodontic appliances will also induce oral ecologic changes such as decreasing salivary pH and increasing the retention of food particles that may lead to increase dental decay [7, 9, 13]. Therefore it is important to take preventive measures such as using appliances that may cause lower bacterial plaque retention.

Forsberg et al. conducted a study with two different types of ligation, with steel wires and elastomeric rings and their association with microbial colonization of Streptococcus mutans and lactobacilli [14]. The results showed that, in the majority of patients, the incisor which was attached to the arch-wire with an elastomeric ring, had greater number of micro-organisms in the plaque than the incisor ligated with steel wire. Therefore they recommended the use of steel wires in patients with bad to poor oral hygiene as elastomeric rings can increase the bacterial counts on tooth surface and cause gingivitis and dental caries.

Previously, Pithon et al. documented a study to verify the hypothesis that self-ligating brackets favour greater aggregation of microorganisms compared to conventional brackets [5]. This investigation showed that the self-ligating brackets had greater bacterial accumulation compared with the conventional brackets. In contrast with the results of our study, Tukkanraman et al. demostrated that teeth ligated with elastomeric rings showed slightly greater number of microorganisms than teeth ligated with steel ligature wires but the difference was not statistically significant [15]. Although teeth ligated with

elastomeric rings were more prone to bleeding in their investigation, no significant differences in the gingival index, bonded bracket plaque index, or pocket depths of the bonded teeth was noticed between the two archwire ligation techniques.

However, in our study, the microbial colonization in the self-ligating brackets was lower than elastomeric ligatures and more than steel wire ligation. This could be due to the opening and closing mechanism of the bracket slot such as clip in the self-ligating brackets that could create sites for plaque retention. Our findings are in agreement with the investigation conducted by Graces and associates [10]. In their study, they used Optical Coherence Tomography (OCT) for evaluating biofilm retention around orthodontic brackets. They showed that biofilm retention is around the elastomeric ring and in the slot and under the clip for self-ligating brackets. They also demonstrated that steel wire ligating bracket had the least biofilm retention.

Raja et al. in an in vivo investigation evaluated the bacteria on the metal, ceramic, and self-ligating orthodontic brackets by scanning electron microscopes [16]. This investigation demonstrates the higher retention of plaque in ceramic brackets ligated with elastomeric ring followed with metal brackets ligated with steel ligatures and comparatively less plaque retention in self-ligating brackets, which their findings were in contrast to the results of our study. One of the reasons for the different results can be attributed to the different method of this investigation. In their study, the bonded tooth went under the preparation procedure for scanning electron microscope after extraction, but in our investigation, the samples were washed three times with water and then cultured so that only the bacteria attached to the brackets and not the bacteria in the saliva would grow in the culture.

In the investigation conducted by Nascimento et al. the greatest colonization of Streptococcus mutans was found in active self-ligating bracket group and the fewest colonies were in a conventional bracket group [17]. One of the advantages of their research was in vivo condition of the study and using the both cultural and scanning electron microscope method.

5. Conclusion

The findings of the current study indicate better effects of steel wire ligatures on decreasing bacterial adhesion. The results suggest less use of elastomeric ligation brackets in patients with poor oral hygiene.

According to the results of the current study, we suggest that elastomeric ligation brackets are less used in patients with poor oral hygiene because elastomeric rings significantly increase the accumulation of microbial plaque adjacent to tooth surface.

Ethical Considerations

Compliance with ethical guidelines

There is no ethical principle to be considered doing this research.

Funding

This study was designed as a thesis for PhD. degree in General Dentistry and all the expenses were paid by the authors.

Authors contributions

All authors contributed in preparing this article.

Conflict of interest

The authors declare no conflict of interest.

References

[1] Lombardo L, Ortan YÖ, Gorgun Ö, Panza C, Scuzzo G, Siciliani G. Changes in the oral environment after placement of lingual and labial orthodontic appliances. Progress in Orthodontics. 2013; 14:28. [DOI:10.1186/2196-1042-14-28] [PMID] [PMCID]

[2] Harikrishnan P, Subha TS, Kavitha V, Gnanamani A. Microbial adhesion on orthodontic ligating materials: An in vitro assessment. Advances in Microbiology. 2013; 3(1):108-14. [DOI:10.4236/aim.2013.31017]

[3] Nascimento LE, Souza MM, Azevedo AR, Maia LC. Are self-ligating brackets related to less formation of Streptococcus mutans colonies? A systematic review. Dental Press Journal of Orthodontics. 2014; 19(1):60-8 [DOI:10.1590/2176-9451.19.1.060-068.oar] [PMCID]

[4] Brusca M, Chara O, Sterin Borda L, Rosa A. Influence of different orthodontic brackets on adherence of microorganisms in vitro. The Angle Orthodontist. 2007; 77(2):331-6. [DOI:10.2319/0003-3219(2007)077[0331:IODOBO]2.0.CO;2]

[5] Pithon MM, dos Santos RL, Nascimento LE, Ayres AO, Alvian D, Bolognese Am. Do self-ligating brackets favor greater bacterial aggregation? Brazilian Journal of Oral Sciences. 2011; 10(3):208-12. [DOI:10.20396/bjos.v10i3.8641633]

[6] Pejda S, Varga ML, Milosevic SA, Mestrovic S, Slaj M, Repic D, et al. Clinical and microbiological parameters in patients

with self-ligating and conventional brackets during early phase of orthodontic treatment. The Angle Orthodontist. 2012; 83(1):133-9. [DOI:10.2319/010412-8.1] [PMID]

[7] de Souza RA, de Araújo Magnani MB, Nouer DF, da Silva CO, Klein MI, Sallum EA, et al. Periodontal and microbiologic evaluation of 2 methods of archwire ligation: Ligature wires and elastomeric rings. American Journal of Orthodontics and Dentofacial Orthopedics. 2008; 134(4):506-12. [DOI:10.1016/j.ajodo.2006.09.067] [PMID]

[8] Gameiro GH, Nouer DF, Cenci MS, Cury JA. Enamel demineralization with two forms of archwire ligation investigated using an in situ caries model-a pilot study. The European Journal of Orthodontics. 2009; 31(5):542-6. [DOI:10.1093/ejo/cjn119] [PMID]

[9] Ahmed I, Saif ul Haque, Nazir R. Carious lesions in patients undergoing orthodontic treatment. Journal of Pakistan Medical Association. 2011; 61(12):1176-9. [PMID]

[10] Garcez AS, Suzuki SS, Ribeiro MS, Mada EY, Freitas AZ, Suzuki H. Biofilm retention by 3 methods of ligation on orthodontic brackets: A microbiologic and optical coherence tomography analysis. American Journal of Orthodontics and Dentofacial Orthopedics. 2011; 140(4):193-8. [DOI:10.1016/j.ajodo.2011.04.019] [PMID]

[11] Batoni G, Pardini M, Giannotti A, Ota F, Rita Giuca M, Gabriele M, et al. Effect of removable orthodontic appliances on oral colonisation by mutans streptococci in children. European Journal of Oral Sciences. 2001; 109(6):388-92. [DOI:10.1034/j.1600-0722.2001.00089.x] [PMID]

[12] Shashidhar EP, Sahitya M, Sunil T, Murthy AR, Rani MS. A comparative evaluation of adherence of microorganism to different types of brackets: A scanning electron microscopic study. Journal of International Oral Health. 2015; 7(9):25-30. [PMID] [PMCID]

[13] Gastel J, Quirynen M, Teughels W, Pauwels M, Coucke W, Carels C. Microbial adhesion on different bracket types in vitro. The Angle Orthodontist. 2009; 79(5):915-21. [DOI:10.2319/092908-507.1] [PMID]

[14] Forsberg CM, Brattström V, Malmberg E, Nord CE. Ligature wires and elastomeric rings: Two methods of ligation, and their association with microbial colonization of Streptococcus mutans and iactobacilli. The European Journal of Orthodontics. 1991; 13(5):416-20. [DOI:10.1093/ejo/13.5.416]

[15] Türkkahraman H, Sayın M, Bozkurt FY, Yetkin Z, Kaya S, Önal S. Archwire ligation techniques, microbial colonization, and periodontal status in orthodontically treated patients. The Angle Orthodontist. 2005; 75(2):231-6. [PMID]

[16] Raju AS, Hegde NA, Reddy VP, Chandrashekar B, Mahendra S, Harishkoushik S. An in vivo study on bacterial colonization with metal, ceramic and self-ligating brackets: A scanning electron microscopy study. Journal of Indian Orthodontic Society. 2013; 47(2):88-96 [DOI:10.5005/jp-journals-10021-1135]

[17] do Nascimento LE, Pithon MM, dos Santos RL, Freitas AO, Alviano DS, Nojima LI, et al. Colonization of Streptococcus mutans on esthetic brackets: Self-ligating vs conventional. American Journal of Orthodontics and Dentofacial Orthopedics. 2013; 143(4):S72-7. [DOI:10.1016/j.ajodo.2012.07.017] [PMID]

This Page Intentionally Left Blank