

# In vivo plaque count of Streptococcus Mutans around orthodontic brackets bonded with two different adhesives

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## ABSTRACT

**Background:** The prevention of the enamel demineralization at the periphery of the brackets is a significant challenge to orthodontic professionals. The aim of this clinical study was to compare the Streptococcus mutans count in the plaque surrounding two orthodontic adhesive types, Fuji Ortho LC and Enlight (Ormco).

**Materials and methods:** A total of 13 patients (7 males and 6 females) needing fixed orthodontic appliance therapy were participated. A split mouth technique was followed with appliances bonded by two orthodontic adhesive types, Fuji Ortho LC and Enlight (Ormco). Saliva was collected before placement of appliances (T<sub>0</sub>) and again at three weeks (T<sub>1</sub>) and six weeks (T<sub>2</sub>) after placement of appliances. Plaque was collected from areas adjacent to brackets and buccal tubes at three weeks (T<sub>1</sub>) and six weeks (T<sub>2</sub>) after placement of appliances. The numbers (colony-forming units) of Streptococcus mutans were determined with the side-specific modified Strip-Mutans.

**Results:** No significant modification in the number of Streptococcus mutans CFU in saliva was observed at both time intervals (T<sub>1</sub>) and (T<sub>2</sub>) after placement of appliances. The number of Streptococcus mutans CFU in plaque at both time intervals (T<sub>1</sub>) and (T<sub>2</sub>) was statistically lower in sites adjacent to Fuji Ortho LC than in those adjacent to Enlight (Ormco) adhesive.

**Conclusion:** plaque surround brackets and tubes bonded with Fuji Ortho LC adhesive harbor less Streptococcus mutans and this will aid in prevention of enamel demineralization.

**Keywords:** Plaque, Streptococcus Mutans, Orthodontic Adhesive. (J Bagh Coll Dentistry 2014; 26(4):175-179).

## المخلص

**خلفية:** الوقاية من ظاهرة انتزاع معادن مينا السن حول اجزاء جهاز تقويم الاسنان الثابت يمثل تحديا كبيرا للمهنيين في اختصاص تقويم الأسنان. كان الهدف من هذه الدراسة السريرية لمقارنة اعداد بكتريا العقدية الطافرة في المادة الجيرية المحيطة بنوعين من المواد المثبتة لاجهزة تقويم الاسنان وهما: (Enlight (Ormco و LC Fuji Ortho

**المواد والطرق:** شارك في البحث 13 مريضا (7 ذكور و 6 إناث) من الذين يحتاجون إلى علاج تقويم الاسنان بواسطة الاجهزة الثابتة. تم تثبيت اجهزة الاسنان بواسطة المادتين اللاصقتين في فم المريض الواحد بقسمة الفم الى قسمين كل مادة تستخدم في قسم. وقد تم جمع عينات اللعاب قبل وضع اجهزة التقويم ومرة اخرى بعد ثلاث اسابيع ومن ثم ستة اسابيع بعد وضع اجهزة تقويم الاسنان. وقد تم جمع المادة الجيرية من المناطق المحيطة لاجهزة التقويم المثبتة على الاسنان بعد ثلاثة اسابيع ومن ثم بعد ستة اسابيع من وضع اجهزة تقويم الاسنان. وتم حساب عدد (الوحدات المكونة للمستعمرة) لبكتريا العقدة الطافرة في عينات اللعاب والمادة الجيرية

**النتائج:** لم يلاحظ أي تغيرات ذات اهمية احصائية في عدد العقدية الطافرة في عينات اللعاب في كافة فترات البحث. و كان عدد مستعمرات بكتريا العقدة الطافرة في المادة الجيرية المحيطة بمادة ال Fuji Ortho يفوق اعداد تلك المستعمرات في المادة الجيرية المحيطة بمادة ال Enlight Ormco

**الاستنتاج:** ان استخدام مادة ال Fuji Ortho لتثبيت اجهزة تقويم الاسنان الثابتة يساعد في الوقاية من ظاهرة انتزاع معادن مينا

**الكلمات الرئيسية:** المادة الجيرية، العقدية الطافرة، المادة المثبتة لجهاز تقويم الاسنان

## INTRODUCTION

Biofilm formation on orthodontic adhesives is a serious clinical problem, as it leads to enamel demineralization around fixed orthodontic appliances, often leaving white spot lesions after their removal <sup>(1)</sup>. Clinical observation indicates that the most common site for bacterial adhesion and biofilm formation is at the bracket-adhesive-enamel junction, an area that is difficult to clean by daily brushing. Furthermore, the surface of an orthodontic adhesive is often rough, with a gap of around 10 µm at the adhesive enamel interface due to polymerization shrinkage <sup>(2,3)</sup>.

Streptococcus mutans has been considered as a major cariogenic bacterium involved in the initiation and progression of dental caries. The correlation between S. mutans counts in saliva or dental plaque and the incidence of dental caries has been postulated <sup>(5)</sup>. Placement of fixed orthodontic appliances leads to an increase in the level of Streptococcus mutans within dental plaque <sup>(6)</sup>. Orthodontic adhesives have a higher Streptococcus mutans -retaining capacity than bracket materials <sup>(7)</sup>.

For this reason, many studies have examined the antibacterial properties of orthodontic adhesives or the effect of antibacterial agents incorporated into orthodontic adhesives.

Enamel demineralization is a commonly recognized complication of orthodontic treatment with a fixed appliance. This enamel demineralization is principally Streptococcus mutans-associated disease <sup>(8)</sup>. It is caused by organic acids produced by mutans streptococci <sup>(9)</sup>. Preventing these lesions is an important concern for orthodontists because the lesions are unaesthetic, unhealthy and potentially irreversible <sup>(10)</sup>. In this sense, one of the effective methods for preventing enamel demineralization is to use orthodontic adhesives resistant to bacterial accumulation <sup>(11)</sup>.

Composite and glass ionomer are the two main classes among many commercially available orthodontic bonding adhesives. Their physical properties, surface characteristics, and fluoride-releasing capacities have been extensively studied; their biologic properties associated with adhesion of cariogenic streptococci also have been investigated. Differences in bacterial adhesion to the different orthodontic adhesives may be expected because of their different

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characteristics and the release of incorporated fluoride<sup>(9,12)</sup>. However, the effect of Fuji Ortho LC glass ionomer adhesive on the plaque *Streptococcus mutans* has not been clinically compared with that of Enlight (Ormco) composite adhesives. Therefore the aim of the present prospective clinical study was to compare the *Streptococcus mutans* counts in the plaque adjacent to Fuji Ortho LC -bonded brackets and that adjacent to brackets bonded with Enlight (Ormco).

## MATERIAL AND METHODS

Volunteer were recruited from patients about to start their treatment with maxillary and mandibular fixed orthodontic appliances in a private orthodontic clinic in Baghdad. A total of 13 patients (7 male and 6 female) were participated in this prospective clinical study after signing an informed consent according to the ethics of human research. The inclusion criteria were Class I malocclusion cases planned to treat with non extraction, good general health, no detectable carious lesions and faulty restorations, no signs of gingival inflammation before starting the study, no pregnancy, non-smokers, and no pharmacotherapy for at least 3 months.

Placement of maxillary and mandibular fixed orthodontic appliances were performed using stainless steel TruFit bondable maxillary and mandibular buccal Tubes (Ortho Technology Company, USA ) on the first molars and stainless steel BionicR Bracket System (Roth Prescription , Slot (.020" x .020") Ortho Technology Company, USA) on the maxillary and mandibular incisors, canines and premolars.

Two commercially available adhesives which is widely used in orthodontic clinic for brackets bonding were evaluated in the present study, resin-reinforced glass-ionomer cement (Fuji Ortho LC, GC Corporation, US) and Enlight (Ormco, USA) light cured composite resin adhesive. Before bonding each tooth was polished, etched and prepared according to manufacturer's instructions for each product. A split mouth technique was followed with slight modification of previous method by Mota et al.<sup>(13)</sup>. Brackets and buccal tubes were bonded with a Fuji (Ortho LC) in one side of the dental arch, and with Enlight (Ormco) in the other side. The bonding material used in each quadrant was: maxillary left = Fuji Ortho; maxillary right = Ormco; mandibular left = Ormco; and mandibular right = Fuji Ortho. Excess adhesive was removed from around the margins using dental probe. Immediately after bonding, .014" TruFlex NiTi arch wire (Ortho Technology Company, USA )

were inserted into brackets and molar tubes slots and elastomere ligatures Power Sticks™ (Ortho Technology) were used for arch wire ligation into the brackets. The patients were given oral hygiene instructions, fluoridated toothpaste, and an orthodontic toothbrush, and asked not to use other oral hygiene supplements during the study.

Analysis of the number of *Streptococcus mutans* was performed by using Dentocult SM Strip mutans test. The patient previously instructed not to eat or brush their teeth two hours before the sampling appointments. Saliva specimens collected before fixed appliance placement (T0), after three weeks (T1) and after six weeks (T2) of appliance placement. Plaque specimens collected, after three weeks (T1) when arch wire changed to 0.018-inch NiTi in all patients and after six weeks (T2) of appliance placement. Selected teeth for plaque sampling were isolated with cotton rolls and dried. Plaque specimens were collected from the labial surfaces immediately surrounding the orthodontic brackets and buccal tubes with a sterilized dental scaler with the same tip dimensions.

The plaque and saliva *Streptococcus mutans* number of colony-forming units (CFU) were determined with the site-specific modified Strip-mutans® technique (Orion Diagnostica, Finland) according to Wallman and Krasse<sup>(14)</sup>. For the saliva collection, the participants were instructed to chew a paraffin pellet for 1 minute and to swallow any excess saliva and then press the rough surface of the round-tipped strip from the Strip-mutans® kit (Orion Diagnostica, Finland) against saliva remaining on the patient's tongue and then removed gently from the patient mouth.

Saliva sample were incubated in a selective culture vial at 37° C for 48 hours in a liquid medium. Sampled plaque was immediately spread in a thorough and gentle manner on the rough surface of the square-tipped strip from the kit. Strips were allowed to dry for 5 minutes at room temperature and then incubated in a selective culture vial at 37° C for 48 hours in a liquid medium.

Results were presented as colony-forming units (CFU). The data was analyzed via student paired t test.

## RESULTS

The mean value and standard deviation of *Streptococcus mutans* CFU counts in saliva are given in Tables 1. There was no significant difference in *Streptococcus mutans* CFU counts in saliva between the values obtained at T0, T1 and T2 as shown in Tables 2.

Regarding the effect of the tested materials on CFU formation, it was observed that CFU means in the plaque adjacent to Fuji Ortho LC at three and six weeks after the beginning of the treatment was significantly lower than the CFU means in the plaque adjacent to Enlight (Ormco) as shown in Tables 3 and 4.

**Table 1: Descriptive statistics of the number (CFU) of Streptococcus mutans in saliva at the beginning of treatment (T<sub>0</sub>) and three weeks (T<sub>1</sub>) and six weeks (T<sub>2</sub>) after placement of the appliances**

Time	Mean	S.D.
(T <sub>0</sub> )	6.308	1.032
(T <sub>1</sub> )	6.154	0.899
(T <sub>2</sub> )	6	0.817

**Table 2: Paired t-test for comparison of means for the number (CFU) of Streptococcus mutans in saliva obtained at T<sub>0</sub>, T<sub>1</sub> and T<sub>2</sub>**

Time	Mean diff.	S.D.	t-test	p-value
T <sub>0</sub>	0.154	0.376	1.477	0.165
T <sub>1</sub>				
T <sub>0</sub>	0.308	0.630	1.760	0.104
T <sub>2</sub>				
T <sub>1</sub>	0.154	0.555	1.000	0.337
T <sub>2</sub>				

Significance level: p < 0.05

**Table 3: Descriptive statistics of the number (CFU) of Streptococcus mutans in plaque for type of material and time of treatment**

Time	Adhesives	Mean	S.D.
(T <sub>1</sub> )	Fuji LC	43.308	1.601
	Ormco	53.769	1.423
(T <sub>2</sub> )	Fuji LC	43.154	1.345
	Ormco	53.615	1.325

**Table 4: Paired t-test for comparison of means for the tested material at three and six weeks after placement of the appliances**

Time	Adhesives	Mean Diff.	S.D.	t-test	p-value
(T <sub>1</sub> )	Fuji LC	-10.462	2.634	-14.322	0.000*
	Ormco				
(T <sub>2</sub> )	Fuji LC	2.222	.616	-16.978	0.000*
	Ormco				

\* Significance level: p < 0.05

## DISCUSSION

An essential event in the initiation of enamel demineralization is microbial adhesion to the teeth and/or orthodontic appliances. Once adhesion has occurred, cell proliferation can lead to increase of

the density of MS in plaque, which is the main cause of enamel demineralization. Researches show that one of the most potent risk factors for enamel demineralization on during orthodontic treatment are the orthodontic bonding adhesives. These adhesives have higher retaining capacity of cariogenic streptococci than bracket materials (2,3,10,15)

Composite resin, frequently used for the fixation of fixed orthodontic appliances has been reported to increase the accumulation of plaque (16), as well as the proportion of *mutans streptococci* in plaque (17-19). Therefore, many modifications and alternate materials have been advocated in an attempt to prevent enamel demineralization from occurring (20). These attempt focused on the control of the cariogenic streptococci adhesion around the brackets as an important factor for the success of orthodontic treatment (10,15). Therefore interest has developed in the use of glass ionomer cements (GICs) as orthodontic bonding agents. Glass ionomer cements (GICs) reduce or prevent decalcification of dental enamel (21).

Careful selection of the orthodontic bonding agent is one of the important factors for the success of orthodontic treatment. In the present study two commercially available bonding agents have been used resin-reinforced glass-ionomer cement (Fuji Ortho LC, GC Corporation, US), and Enlight Bonding system (Ormco, USA) light cured composite resin adhesive with fluoride-release. Both of them classified as good bonding material regarding their bond strength (22). Therefore the present study compared their antimicrobial activity against *mutans streptococci* in plaque.

Based on the findings obtained in this study the number of *Streptococcus mutans* CFU in saliva, showed no significant difference at all the experimental periods (T<sub>0</sub>), (T<sub>1</sub>) and (T<sub>2</sub>). This finding inconsistency with Mota et al. (13) and may be explained by the suggestion of Scheie et al. (23).

Concerning the number of *Streptococcus mutans* CFU in plaque the present study observed significant reduction in plaque adjacent to brackets bonded with Fuji Ortho than in plaque adjacent to brackets bonded with Enlight (Ormco) at (T<sub>1</sub>) and (T<sub>2</sub>). This in accordance with the result of McNeill et al. (24) who concluded that glass ionomer cement is more effective than composite resin in preventing white spot formation. Another explanation is the result of Gorton and Featherstone (25) who observed that the resin modified GIC showed a cariostatic effect around brackets up to 4 weeks after appliance

placement and assumed that this cariostatic effect may be due to the slow fluoride release, which results in the presence of fluoride in enamel or in plaque fluid. On the other hand Fishman and Tinanoff<sup>(26)</sup> suggested that the bacterial growth inhibiting effect seemed to be associated with GIC acid release. Moreover Cacciafesta et al.<sup>(27)</sup> suggested that Fuji Ortho LC recommended as suitable fluoride-releasing orthodontic adhesives because of their high fluoride releasing capacity than the other adhesives.

The significant increase in the number of *Streptococcus mutans* in plaque adjacent to brackets bonded with Enlight (Ormco) may be explained by the in vitro studies of Quirynen et al.<sup>(28,29)</sup> that showed surface roughness of orthodontic adhesive have a significant impact on the bacterial adhesion and colonization

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