# A Comparative Evaluation of Microbial Colonization with Different Ligation Techniques

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**Abstract:** Fixed or removable orthodontic appliances impede the maintenance of oral hygiene and result in plaque accumulation. The effects of fixed orthodontic appliances on microbial flora is well known. This study was done to determine the changes in microbial flora (Streptococcus) after orthodontic bonding; and to determine whether two different archwire ligation techniques affect these changes; and to find out whether self-ligation brackets and conventional brackets have a difference in aggregation of microorganisms. Pretreatment sample was collected from patient saliva in a sodium glycolate. Self-ligation (3M) bracket bonding was done on all the teeth, except 14: bonded with 3M conventional bracket and ligated with ligature wire; 24: bonded with 3M conventional bracket and stainless steel ligature. Similar result was found for Streptococcus count also.

Keywords: Orthodontic appliances; Self ligation, Elastomeric rings, Streptococcus

#### I. Introduction

The different components of the fixed orthodontic system may contribute to a shift in the balance of the oral ecology. Plaque retention surrounding orthodontic appliances leads to enamel demineralization caused by organic acids produced by bacteria in the dental plaque.

A large amount of research has dealt with the intimate contact of orthodontic materials with the tooth and periodontal tissue<sup>1</sup>. Oral microbiota attachment in orthodontic patients has been mainly associated with increased risk of *Streptococcus mutans* which may lead to the development of pathology of the hard tissues such as decalcification and, in specific cases, caries development <sup>1</sup>.

The placements of fixed appliances impede the maintenance of good oral hygiene. Components of the fixed appliance create new retention areas for Streptococcus mutans and impedes access to areas for optimal cleaning<sup>2</sup>. Clinicians may use either 0.008 to 0.014 inch stainless steel ligature wire, self-ligating springs, or circularsynthetic elastomers to secure arch wires to brackets<sup>2</sup>.

One of the proposed favorable aspects of self-ligating brackets is the elimination of elastomeric or stainless steel ligatures. It has been proposed that this feature brings two basic advantages: the eradication of cross-contamination, which may accidentally be involved in the process of ligature change, and the claimed improvement in the oral hygiene of patients. The latter has been attributed to the fact that the patient is given the opportunity to clean surfaces of reduced complexity and with less retentive sites for microbial colonization<sup>1</sup>. This study investigated the differences in microbial level of Streptococcus mutans self-ligation bracket, and, conventional bracket with the two methods of archwire ligation- i.e. the elastomeric module and ligature wire.

## II. Materials And Methods

The study was done by bonding all the teeth with self-ligation bracket (3M), except 14 (conventional 3M) and 24 (conventional 3M) (Fig 1). Informed consent was obtained from the patient.



#### **Inclusion Criteria:**

- 1. All the permanent teeth were erupted
- 2. Free of dental plaque
- 3. No signs of gingival inflammation before the beginning of study.

#### **Exclusion Criteria**

- 1. Patient undergone antibiotic therapy in last 6 months
- 2. History of systemic disease
- 3. Prior periodontal treatment

The subject was given no additional information on oral hygiene maintenance and was asked to be refrain from any other oral hygiene product for the duration of trial.

Before orthodontic appliance placement, the saliva sample (T1) was collected from the oral cavity (Fig.2) and stored in the media (Fig 3) and was transferred to the lab within 24 hours of collection.

Nitinol arch wire 0.014", was engaged in the appliance immediately on all the teeth, with elastomeric modules on 24, with stainless steel ligature wire on 14.

Fig.2 : Saliva collection





Fig.3 : saliva sample transfer into media

The sample of stainless steel ligature wires elastomeric modules accumulation along self-ligation brackets and salivary sample (T2) was collected after 21 days. The samples were stored in the media and transferred to the lab within 24 hours. The samples were cultivated and analyzed by the same examiner in the department of clinical microbiology.

For Streptococcus isolation, Haem Agar media (Table 1) and for Total microbial count, LB agar (Luria Bertini-Table 2) was prepared for required amount and was autoclaved. From the given samples 0.1ml was taken and made upto 1ml with 1% saline (Sodium chloride) and using pour plate method 0.1ml of suspension

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Fig.2 : Saliva collection

was taken and added onto the plate and media was poured upon Inoculum plates were incubate at 37<sup>o</sup>c for 17-24hrs. The colony forming unit (CFU) was calculated for per ml of sample.

Ingredients	Gms	
Haemoglobin	0.4g in 10ml	
Sodium Chloride	1	
Yeast Extract	0.2	
Peptone	1	
Agar	4	
Distilled water	200	

Table 2: LB agar st

Ingredients	Gms/liter	
Yeast Extract	5	
Tryptone	10	
Sodium Chloride	10	
Agar	20	
Distilled water	1000	

## III. Results

The Streptococcal count was 130 CFU/ml and total bacterial count  $2X10^3$  CFU/ml in the saliva sample. Streptococcal count along elastomeric module came to be 300 CFU/ml and total bacterial count  $1.4X10^4$  after 21 days.

Streptococcal count was 160 CFU/ml and total bacterial count 8 X  $10^2$  in the Self-ligation bracket after 21 days. Streptococcal count was 30 CFU/ml after 21 days in the stainless steel ligature wire sample (Table 3). Total bacterial count in saliva was found to be 2000CFU/ml, whereas it was 5000CFU/ml after 21 days of bonding. Total bacterial count in E-modules along conventional brackets was found to be 14000CFU/ml, which is tremendously increased as compared to pre-treatment values.

## TABLE 3

Sample (CFU/ml)	Streptococcous	Total bacterial count
Saliva	130	<b>2*10</b> <sup>3</sup>
Ring	300	1.4*104
Self ligation bracket	160	8*10 <sup>2</sup>
String	30	No growth

## IV. Discussion

The use of fixed appliances results in a significant challenge to the patient as far as maintaining good hygiene in order to avoid or minimize decalcification of enamel during treatment. A number of investigators reported a significant increase in salivary and plaque levels of Streptococcus mutans and lactobacilli in patients undergoing fixed appliance treatment<sup>12,13,14</sup>. The microbial count along ligature materials <sup>9,10,11</sup>; elastomeric rings <sup>3,11</sup>; self-ligation brackets <sup>4</sup>; has been measured in the past.

Although this study is agreeing to the results of previous ones that microbial count increases after bonding, and is also highest along e-modules, however rules out that self-ligation has least count. It is least for the stainless steel ligature wire. The methodology in this study of microbiologic evaluation of teeth with selfligation brackets, conventional bracket ligated with ligature wire and conventional bracket ligated with elastomeric modules in the same oral cavity was a method of reducing the individual variations as well as intragroup variations. The results obtained from this study indicated a tremendous increase in microbial count when compared between pre and post bonding. Also the total bacterial count was extremely high along emodules among all the four samples. There was decreased total bacterial count along self-ligation bracket in comparison to e-modules along conventional brackets but increased count in comparison to ligature wire. Total bacterial count was highest along e-modules among all four samples. Study by Forsberg et al showed that elastomeric rings exhibited a great number of microorganisms in the plaque around the bracket than the steel ligatures<sup>3</sup> which is also in agreement to the result of this study which shows the increases count of microorganisms in elastomeric rings.

Study by Pellegrini<sup>4</sup> etal showed that Self-Ligation appliances promote reduced retention of oral bacteria<sup>4</sup> this study stated that microbial colonization along self-ligation brackets is although lesser than e-modules but it is greater than stainless steel ligature wires.

Sukontapatipark et al <sup>11</sup> using scanning electron microscopy reported that the method of ligation did not appear to influence the bacterial morphotypes on either composite or enamel surfaces. But in this study, the microbial count showed an increase from ligature tie to self-ligation bracket and was highest in e modules.

The results obtained for streptococcal count indicated an increase in the number of streptococcus post bonding. Also the streptococcal count was extremely high along e-modules among all the four samples. There was decreased streptococcal count along self-ligation bracket in comparison to e-modules along conventional brackets but greater than ligature wire along conventional brackets. The streptococcal count was highest along e-modules among all the four samples.

Although the results indicate fewer plaque bacteria surrounding the self-ligation appliances, any mechanics with elastomeric chains or similar auxiliaries with self-ligation appliances will presumably negate the beneficial effects of self-ligation appliances, possibly also diminishing other purported benefit.

## V. Conclusion

- Total bacterial count and streptococcal count increases post bonding
- E-modules always show greatest total bacterial count and streptococcal count
- Stainless steel ligature wire shows the least total bacterial and streptococcal count
- Self-ligation bracket had a count less than e modules but stainless steel ligature showed the least microbial colonization among the three.

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#### **Figure Legends**

Figure 1: Bonding of entire maxillary arch Figure 2: Saliva collection Figure 3: Saliva sample transfer into the media **Table Legends** Table 1-Composition of HAEM agar Table 2-Composition of LB agar Table 3-Microbial count in CFU/ml