



ISSN Print: 2394-7489
ISSN Online: 2394-7497
IJADS 2025; 11(1): 81-86
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www.oraljournal.com
Received: 02-10-2024
Accepted: 16-11-2024

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Effects of fluoride releasing bonding material on salivary microbial colonisation, plaque index and gingival index in orthodontic patients

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DOI: <https://doi.org/10.22271/oral.2025.v11.i1b.2104>

Abstract

Objective: To evaluate the effects of a fluoride releasing orthodontic bonding material on the colonisation of SM and LB in saliva, on plaque index and gingival index. The null hypothesis assumed that fluoride releasing orthodontic bonding material have no effect on these parameters.

Design and Methods: Randomised, prospective, controlled trial. The 45 patients were randomly divided into 3 equal groups according to bonding material type - Group 1: Transbond™ Plus SEP and Transbond™ Plus colour change adhesive (fluoride releasing bonding material, compomer); Group 2: Transbond™ XT primer and Transbond XT adhesive (conventional composite); and Group 3: untreated control group (no brackets were bonded, negative control group).

Results: SM level showed a significant increase in the Transbond XT group only. LB level showed a significant increase in the Transbond Plus, Transbond XT groups ($p < 0.05$). There was no statistical differences in LB level between the groups at T₀, T₁ and T₂. PI and GI scores showed a significant increase in all groups, except the negative control ($p < 0.05$). There were no statistical differences in PI and GI scores between the groups, except the negative control.

Limitations: The study only covers the period of one month. Blinding of the clinician was also lost.

Conclusion: Fluoride releasing orthodontic bonding material could prevent SM increase, but had no effect on LB increase, plaque index and gingival index.

Keywords: Orthodontic bonding material, compomer, microbial colonisation, plaque

Introduction

In patients undergoing orthodontic treatment, there is an increase in retentive spaces for dental plaque accumulation on the oral environment [1]. Following the insertion of orthodontic appliances, there is an increase in the level of acid-producing microorganisms such as *Streptococcus mutans* and *Lactobacillus* [2, 3]. While *Streptococcus mutans* (SM) is regarded as a primary pathogen in dental caries [4] and causes initial caries to form, *Lactobacillus* (LB) plays a role in the formation of active caries [5]. Increased plaque accumulation and bacterial acid production leads to the dissolution of calcium and phosphate ions from the tooth's structure and so results in dental caries [6].

The chemical structure and surface characteristics of orthodontic attachments and composites can affect plaque retention. Irregular composites around the orthodontic brackets provide a favourable environment for bacterial colonisation [7, 8]. It has been reported that orthodontic bonding materials have more retention capacity than bracket materials for SM and LB [9, 10]. Composite resins and glass ionomer cements are the two main classes of orthodontic bonding materials. Hybrid bonding materials, comprising glass ionomer and composite resin are resin modified glass ionomer cement and polyacid modified composite resin. Polyacid modified composite resins are called compomers. They have been developed [11] for sustained fluoride release and greater bonding strength because resin modified glass-ionomer cements were reported to have unreliable bonding strength [12].

In vitro studies have reported that plaque grown on a compomer does not contain SM and so the compomer could reduce the cariogenic effect of SM biofilms [13, 14]. Another study reports that the amount of demineralisation is less around a tooth bonded with compomer [15].

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In the literature, there is currently no study concerning the effects of compomer orthodontic bonding materials on salivary oral microbiota, plaque index and gingival index. Thus, the aim of this study is to evaluate the effects of a compomer orthodontic bonding material on the colonisation of SM and LB in saliva and on plaque index and gingival index.

The null hypothesis assumed that compomer orthodontic bonding material has no effect on these parameters.

Subjects and Methods

Study Group

Some 45 patients (24 girls, 21 boys) aged between 12-18 years were included in this study. Ethical approval was obtained from Selcuk University Faculty of Medicine Ethics Committee on 29th March 2013 (#2013/19). Before beginning the study, informed consent was obtained from the patients and their guardians. The inclusion criteria were: mild or moderate crowding, permanent dentition, and good oral

hygiene. Patients who had cleft lip and palate, systemic illness, deformities, were prescribed medication, used antibiotics and antibacterial mouthwash in the last month, topical fluoride treatments within four weeks or who had decalcification and caries cavities were excluded from the study. None of the subjects used fluoride mouthwash or fluoride varnish during the study.

Study Design

The study was a randomized, controlled clinical trial. The 45 patients were randomly divided into 3 equal groups according to bonding material type - Group 1: Transbond™ Plus SEP and Transbond™ Plus colour change adhesive (compo-mer); Group 2: Transbond™ XT primer and adhesive (conventional composite); and Group 3: untreated control group (no brackets were bonded, negative control group). The contents and chemical compositions of the bonding materials are shown in Table 1.

Table 1: Orthodontic materials used in this study

Brand	Components	Chemical Composition	Manufacturer
Transbond Plus	Transbond Plus Self Etching Primer	Water, Methacryloyl Phosphate Monomer, Phosphine Oxide, Fluoride Complex	3M Unitek, Monrovia Calif, USA
	Transbond Plus color change adhesive	Silane treated glass, silane treated quartz, polyethylene glycol dimethacrylate, 1,2,3-propanetricarboxylic acid, 2-hydroxy-, reaction products with 2-isocyanatoethyl methacrylate silane treated silica, bisphenol a diglycidyl ether dimethacrylate (bisgma), diphenyliodonium hexafluorophosphat	
Transbond XT	Transbond XT primer	Bisphenol a diglycidyl ether dimethacrylate, Triethylene glycol dimethacrylate 4 (dimethylamino)-benzeneethanol Camphorquinone, Hydroquinone	3M Unitek, Monrovia Calif, USA
	Transbond XT Adhesive	Silane treated quartz, Bisphenol a diglycidyl ether dimethacrylate (bisgma), bisphenola bis(2-hydroxyethylether) dimethacrylate, Silane treated Quartz	

Bonding Procedure

The enamel was roughened with 37% orthophosphoric acid (Pulpdent® Etch Royale™ Pulpdent Corporation Watertown, Massachusetts, USA) for 40 minutes until an opaque appearance occurred in Transbond XT group. The same procedure was conducted with Transbond Plus SEP for 20 seconds with some pressure in the Transbond Plus group. Enamel preparation continued with sealing of the primer corresponding to the Transbond XT group according to the respective manufacturer's instructions. Adhesive materials were installed to the bracket bases and bonded with Elipar™ S10 LED Curing Light (3M Unitek, Monrovia, California, USA).

Records

Microbial and periodontal records were obtained for all groups at T₀, T₁ and T₂. T₀: Before bonding in Transbond Plus and Transbond XT groups, as well as in the beginning for negative control group; T₁: Two weeks later; and T₂: Four weeks later.

All microbial and periodontal records were collected by one examiner (FBK). Periodontal examination was performed using a Williams probe and included plaque index (PI) and gingival index (GI) measurements at three sites (mesial, labial/buccal, and distal) for all teeth^[16]. Periodontal indices were calculated as a sum of the mean scores for each examined tooth divided by the number of evaluated teeth. A mean of all the measurements for each patient was considered.

Microbiological analysis was performed by obtaining a sample of stimulated saliva. The subjects had refrained from brushing their teeth, eating, drinking, and chewing gum for at least 2 hours before the saliva samples were taken. The saliva

samples was collected between 9:00 AM and 12:00 PM. Collection of saliva was performed before any oral examination or manipulation so as not to disrupt the oral microbiota. To evaluate the number of colony-forming units of SM and LB per millilitre of saliva, CRT bacteria kits (Ivoclar Vivadent AG, FL 9494 Schaan/Liechtenstein) were used. For collecting stimulated saliva, the patients were instructed to chew a paraffin pellet for 5 minutes. The agar carrier was removed from the test vial and protective foils were removed from the agar surface. Collected saliva was inoculated on an MSB agar surface for SM detection and a rogosa agar surface for LB detection. After adding a NaHCO₃ tablet, the CRT bacteria vials were incubated by Cultura incubator (Ivoclar Vivadent AG, FL 9494 Schaan/Liechtenstein) at 37°C for 48 hours.

Evaluation of Results

After incubation, SM was visible as blue transparent colonies, while LB was visible as white colonies. The growth density of bacteria was assessed according to the evaluation chart. The growth densities of SM and LB were categorised as follows: 1= <10⁴ CFU/mL, 2=10⁴ -10⁵ CFU/mL, 3= 10⁵ -10⁶ CFU/mL and 4= >10⁶ CFU/mL.

Statistical Analysis

Before starting the study, the number of required patients was determined with G* Power (Ver. 3.0.10, Franz Faul Universitat, Kiel, Germany). When the sample size was 45 patients, in three groups and with three different measurements (effect size=0.40, α=0.05), power was calculated as 94.97%. Statistical measurements were made using SPSS statistical software (SPSS 17.0 statistics, Chicago, USA) for Windows. Due to the categorical data and abnormal

distribution, nonparametric tests were used. The nonparametric Wilcoxon signed-rank test, following a significant Freidman ANOVA result, was used to determine significant differences between measurement times in each group. Differences between groups were analysed with the Mann-Whitney U pairwise comparison test, following a significant Kruskal-Wallis result. All statistical tests were performed at $p < 0.05$.

Results

Salivary SM Level

At the beginning (T₀), no significant differences were found in the SM levels between the groups. At week 2 (T₁), significant differences were found between the negative control and Transbond XT groups and between the Transbond Plus and Transbond XT groups. The Transbond Plus group was not different from the negative control. At week 4 (T₂), significant differences were found between the Transbond

Plus and Transbond XT groups; and between the negative control and Transbond XT groups ($p < 0.05$). The Transbond Plus group was not different from the negative control (Table 2).

Salivary SM levels showed a significant increase between T₀ and T₁ and between T₀ and T₂ in the Transbond XT group only ($p < 0.05$). In the Transbond Plus and negative control groups, no significant changes were found (Table 2).

Salivary LB Level

At the beginning (T₀), there was no significant difference in LB levels between the groups. No significant intergroup differences were found after two weeks (T₁) or after four weeks (T₂) (Table 3).

Salivary LB values showed a significant increase between T₀ and T₁ in the Transbond XT group only. Between T₀ and T₂, there were significant increases in the Transbond Plus and Transbond XT groups ($p < 0.05$) (Table 3).

Table 2: Intergroup and intragroup comparisons of SM Scores of the three group at each measurement time

	T0					T1					T2					Intragroup Differences /Wilcoxon	Intragroup Differences /Wilcoxon	
	Test Skoru(CFU/ml)					Test Skoru(CFU/ml)					Test Skoru(CFU/ml)							
	n	<10 ⁴	10 ⁴ -10 ⁵	10 ⁵ -10 ⁶	>10 ⁶	mean±SD	<10 ⁴	10 ⁴ -10 ⁵	10 ⁵ -10 ⁶	>10 ⁶	mean±SD	<10 ⁴	10 ⁴ -10 ⁵	10 ⁵ -10 ⁶	>10 ⁶			mean±SD
Transbond plus	15	3	9	3	0	2,00±0,65	0	14	1	0	2,06±0,25 (a)	2	8	5	0	2,20±0,67 (a)	NS	NS
Transbond XT	15	6	7	2	0	1,73±0,70	0	7	5	3	2,73±0,79 (b)	0	6	4	5	2,93±0,88 (b)	0,002	0,002
Negative Control	15	6	4	5	0	1,93±0,88	5	5	5	0	2,00±0,84(a)	5	4	6	0	2,06±0,88 (a)	NS	NS
Intergroup Differences		Kruskal Wallis/p value=0,296					Kruskal Wallis/p value=0,022					Kruskal Wallis/p value=0,006						

Different letters in the same coloumn indicates significant differences between the groups. NS,not significant

Table 3: Intergroup and intragroup comparisons of LB Scores of the three groups each measurement time

	T0					T1					T2					Intragroup differences /Wilcoxon	Intragroup differences /Wilcoxon	
	Test Skoru(CFU/ml)					Test Skoru(CFU/ml)					Test Skoru(CFU/ml)							
	n	<10 ⁴	10 ⁴ -10 ⁵	10 ⁵ -10 ⁶	>10 ⁶	mean±SD	<10 ⁴	10 ⁴ -10 ⁵	10 ⁵ -10 ⁶	>10 ⁶	mean±SD	<10 ⁴	10 ⁴ -10 ⁵	10 ⁵ -10 ⁶	>10 ⁶			mean±SD
Transbond plus	15	9	5	1	0	1,46±0,63	4	8	3	0	1,93±0,70	4	7	4	0	2,00±0,75	NS	0,021
Transbond XT	15	8	7	0	0	1,46±0,51	3	8	4	3	2,06±0,70	2	3	8	2	2,66±0,89	0,013	0,003
Negative Control	15	8	2	5	0	1,80±0,94	5	7	3	0	1,86±0,74	5	6	4	0	1,93±0,79	NS	NS
Intergroup Differences		Kruskal Wallis/p value= 0,787					Kruskal Wallis/p value= 0,945					Kruskal Wallis/p value= 0,064						

Different letters in the same coloumn indicates significant differences between the groups. NS,not significant

Plaque and Gingival Index

The plaque and gingival index values increased significantly at week 2 (T₁) and at week 4 (T₂) compared with the baseline (T₀) in two groups, except the negative control ($p < 0.05$) (Tables 4 and 5).

At the beginning (T₀), no significant differences were found in plaque index and gingival index values between the groups.

There was a significant increase in the plaque index and gingival index values between T₀ and T₁ and between T₀ and T₂ in all groups, except the negative control. No significant differences were found in the plaque and gingival index scores in the negative control group. After two weeks (T₁) and after 4 weeks (T₂), the negative control group was statistically different from the other groups ($p < 0.05$) (Tables 4 and 5).

Table 4: Intragroup and Intergroup Comparisons of Plaque Index values of the Three Groups at Each Measurement Time

	n	T ₀	T ₁	T ₂	Intragroup Differences /Wilcoxon	Intragroup Differences /Wilcoxon
		mean ± SD	mean ± SD	mean ± SD	T ₀ -T ₁	T ₀ -T ₂
Transbond plus	15	0,82±0,31	1,25±0,538 (b)	1,58±0,503 (b)	0,023	0,001
Transbond XT	15	0,83±0,55	1,3±0,581 (b)	1,55±0,749 (b)	0,005	0,004
Negative Control	15	0,74±0,46	0,68±0,44 (a)	0,68±0,4 (a)	NS	NS
Intergroup Differences/Kruskal Wallis		p=0,77	p=0,002	p<0,001		

Different letters in the same coloumn indicates significant differences between the groups. NS,not significant

Table 5: Intragroup and Intergroup Comparisons of Gingival Index values of the Three Groups at Each Measurement Time

	n	T ₀	T ₁	T ₂	Intragroup Differences /Wilcoxon	Intragroup Differences /Wilcoxon
		mean ± SD	mean ± SD	mean ± SD	T ₀ -T ₁	T ₀ -T ₂
Transbond plus	15	0,33±0,41	0,81±0,35 (b)	1,11±0,532 (b)	0,006	0,005
Transbond XT	15	0,38±0,431	0,74±0,434 (b)	1,05±0,483 (b)	0,003	0,001
Negative Control	15	0,52±0,38	0,49±0,28 (a)	0,43±0,3 (a)	NS	NS
Intergroup Differences/Kruskal Wallis		p=0,526	p=0,029	p<0,001		

Different letters in the same coloumn indicates significant differences between the groups. NS,not significant

Discussion

The results confirm that fluoride releasing (compomer) orthodontic bonding material could prevent an SM increase, but have no preventive effect on LB increase, gingival index and plaque index. Thus, the null hypothesis concerning SM was rejected.

In our study, the microbial and periodontal records were taken at the beginning, after 2 weeks and after 4 weeks. In the literature, in the studies that evaluated the effect of anti-caries applications on SM and LB colonisation, the records were taken at short intervals such as 1 or 2 weeks [17-20]. Similar to these studies, in our study the records were taken at short intervals to avoid ignoring the rapid effect of bonding materials on SM and LB colonisation.

The study ended in the fourth week. This was because longer periods of observation may change the results since, as noted by Turkkahraman *et al.* [21], oral hygiene, dietary habits and motivation can change over time.

Patients who had standard oral hygiene and gingival health criteria were included in this study [22]. Patients who had used antibiotics and antimicrobial mouthwash in the last month were not included in this study in order to prevent the effects of antimicrobial agents on plaque and oral microbiota [3, 23, 25]. In our study, all patients had upper and lower arch 7-7 bonding procedure. Scheie *et al.* [26] reported that the number of teeth bonded could affect oral microbiota. However, some studies have not mentioned the number of orthodontic attachments [24, 27, 28]. An equal number of teeth were bonded in all patients to provide standardisation.

In this clinical trial, a split mouth study design was not used because a slight crossover of fluoride could still occur with saliva, as reported by Wiltshire [29], so split mouth study design is not recommended.

In the present study, salivary samples were used to determine SM and LB. Using the salivary samples to determine SM and LB has some advantage: It is non-invasive, easy to collect and not limited to a specific area [30, 31]. In our study, the counts of salivary SM and LB were estimated using CRT bacteria kits. Previous studies showed that this method correlates well with conventional laboratory methods [32-35].

Fluoride releasing adhesive materials are conventional glass ionomer cement, resin modified glass ionomer cement and polyacide modified composite resins (compomer) [36, 37]. Chin *et al.* [36] reported that compomer bonding materials release 32% of the fluoride released by glass ionomer cement and 18% of the fluoride released by resin modified glass ionomer cement. Compomers are less hydrophobic than conventional composite resins and they absorb the moisture following initial polymerisation, which initiates the acid-base reaction between the acid groups and the glass filler of the functional monomer. This feature allows the fluoride release from the glass filler to the matrix and then the oral environment [37, 38]. A study has reported that the compomer has a strong antimicrobial activity against SM and LB, as well as having more antimicrobial efficacy than glass ionomer, although it releases less fluoride [39].

In the Transbond XT group, as with the findings of other studies [3, 24, 40, 41, 42, 43], there were significant increases in the SM and LB levels. In the other groups, there were no significant differences in SM level. The findings suggest that compomer orthodontic bonding material could prevent SM increase. It is known that the LB level can increase with a high dietary intake of carbohydrates and orthodontic treatment [44]. In previous studies, it has been reported that there is a positive correlation between SM and LB

colonisation and caries prevalence [4, 45]. Some researchers reported no significant increase in SM and LB level with orthodontic treatment [26, 46, 47].

Although our study is consistent with the results of some previous studies, there is currently no *in vivo* study in the literature comparing the effect of compomer bonding materials on salivary microbial colonisation, plaque index and gingival index.

In our study, the plaque and gingival index values increased significantly in all groups, except the negative control. The intergroup results of the present study concerning the plaque and gingival indexes are consistent with studies in the literature [14, 49, 50]. Badawi *et al.* [13] reported that supragingival dental plaque is not effected by orthodontic bonding material types that contain fluoride.

Conclusion

1. There is a significant increase in SM level at two weeks and at four weeks in patients for whom a conventional composite bonding material was used, but no significant difference was found in patients for whom compomer bonding material was used.
2. At two weeks, Transbond Plus group is significantly different from the Transbond XT group. At four weeks, the significant difference between the Transbond XT and Transbond Plus groups continues.
3. Intergroup differences in LB level were not significant at two weeks and at four weeks.
4. After orthodontic bracket placement, a significant increase in plaque and gingival index values occurred at two weeks and four weeks, and fluoride releasing orthodontic bonding material was found to have no effect on the plaque index and gingival index values.

Conflict of Interest

Not available.

Financial Support

Not available.

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How to Cite This Article

Kahraman FB, Demir A. Effects of fluoride releasing bonding material on salivary microbial colonisation, plaque index and gingival index in orthodontic patients. *International Journal of Applied Dental Sciences*. 2025; 11(1): 81-86.

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