Effects of fluorosis on the shear bond strength of orthodontic brackets bonded with a self-etching primer

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SUMMARY The aim of this study was to compare the shear bond strength (SBS) of brackets bonded to fluorosed and non-fluorosed teeth with self-etching primer (SEP) and phosphoric acid (PA). The study involved 40 mildly fluorosed [Thylstrup–Fejerskov (TFF) Index = 1–3] and 40 non-fluorosed human premolar teeth. The fluorosed and non-fluorosed teeth were randomly divided into two subgroups. In the first subgroup, 37 per cent PA was applied for 30 seconds and in the second, a SEP (Transbond Plus) was used. The brackets were bonded with light-cure adhesive paste (Transbond XT) and cured for 20 seconds. The SBSs were measured after 1000 thermocycles. Two-way analysis of variance, Tukey’s multiple comparison test, and Weibull analysis were used for the evaluation of SBS values. Bond failure locations were determined with the adhesive remnant index (ARI) and were compared with the Kruskal–Wallis and Mann–Whitney U-tests.

The mean SBS was 9.01 MPa for the fluorosed teeth bonded with SEP. This value was significantly different from those of fluorosed teeth etched with PA (15.22 MPa) and non-fluorosed teeth conditioned with SEP (12.95 MPa) and PA (15.37 MPa). The ARI scores of the fluorosed teeth conditioned with SEP were significantly lower than those of non-fluorosed teeth conditioned with SEP or PA.

The results of this in vitro study suggest that there are no differences in the SBS of orthodontic brackets between mildly fluorosed and non-fluorosed enamel etched with 37 per cent PA for 30 seconds. The SEP showed lower SBS values for orthodontic brackets bonded to mildly fluorosed enamel. The findings provide some evidence that routine clinical use of a SEP to bond brackets to mildly fluorosed teeth cannot be supported.

Introduction

In orthodontic practice, it is essential to obtain reliable adhesive bonds between orthodontic brackets and tooth enamel (Grubisa et al., 2004). Bonding is based on the mechanical locking of an adhesive between the bracket base and irregularities in the enamel surface. Successful bonding, therefore, requires careful attention to all three components: the tooth surface and its preparation, the design of the attachment base, and the bonding material (Proffit et al., 2007).

Orthodontists working in endemic fluorosis regions (e.g. Colorado, USA; Sri Lanka; Isparta, Turkey; Hail Region, Saudi Arabia) may encounter difficulties in bonding brackets to fluorosed teeth (Adanir et al., 2009). Furthermore, it has been shown that the prevalence of dental fluorosis is increasing in many non-endemic fluorotic areas (Ratnaweera et al., 2007). This increase in the prevalence of dental fluorosis even in geographic areas without an overabundance of natural fluoride in the water has been attributed to high background exposure to fluoride from many sources, such as fluoride-treated water supplies and beverages, fluoridated salt and milk, the environment (e.g. air, soil), foods (e.g. tea, seafood), and fluoride-containing dental products (e.g. dentifrices, supplements; Miller, 1995; Clarkson et al., 2000).

Microradiography of fluorotic enamel showed that the thin outermost enamel surface layer was relatively highly mineralized and was accompanied by a subsurface hypomineralized area (Yanagisawa et al., 1989b). It has been reported that the highly mineralized surface layer contains hydroxyapatite and fluoridated-hydroxyapatite or fluorapatite, or both (Yanagisawa et al., 1989a). These fluoridated crystals are more acid resistant (Robinson et al., 2004). The reduction in acid solubility of enamel was attributed to larger apatite crystals, better crystallinity, and the buffering action of fluoride released from enamel crystals during the early stages of acid attack (Clarkson et al., 2000).

It has been emphasized that the most challenging surface for orthodontists is fluorosed enamel (Miller, 1995; Noble et al., 2008). There are limited in vitro studies in the literature concerning bond strength of brackets bonded to fluorosed
enamel. Opinya and Pameijer (1986) observed a significant difference between tensile bond strength (TBS) of normal and fluorosed teeth (moderate and severe according to Dean’s Fluorosis Index) after etching with 37 per cent phosphoric acid (PA). Similarly, Adanir et al. (2009) reported a significant difference between shear bond strength (SBS) of normal and moderately fluorosed teeth [Thylstrup–Fejerskov Index, (TFI) = 4] after etching with 37 per cent PA. However, Ng’ang’a et al. (1992) did not observe a significant difference between the TBS of non-fluorotic versus mild and moderately fluorotic teeth (TFI = 3 and 4) after etching with 40 per cent PA.

The introduction of self-etching primers (SEPs) has attracted considerable interest since they combine the etching and priming steps into one, eliminating the need for separate etching, rinsing, and drying. The effectiveness of SEP for orthodontic bracket bonding has been proven with numerous in vitro and in vivo studies (Larmour and Stirrups, 2003; Cal-Neto and Miguel, 2005; dos Santos et al., 2006; Turk et al., 2007; Elekdag-Turk et al., 2008).

Contradictory results were obtained in the in vitro studies (Weerasinghe et al., 2005; Ermis et al., 2007; Ratanweera et al., 2007; Shida et al., 2009) evaluating the bond strength of composite resin bonded to fluorosed enamel with self-etch adhesives. No significant difference was observed between the bond strengths of PA etching and self-etching bonding approaches for normal ground and fluorosed ground enamel (Ermis et al., 2007). Furthermore, no significant difference was observed between the bond strengths of PA etching plus SEP and self-etching bonding approaches for fluorosed ground enamel (Weerasinghe et al., 2005). In the study by Ratanweera et al. (2007), two all-in-one adhesive systems showed no significant differences in micro-SBS between fluorosed unground and non-fluorosed unground enamels. However, fluorosed unground enamel showed lower micro-TBS than normal unground enamel with two-step self-etch adhesives (Ermis et al., 2007; Shida et al., 2009). The aim of this study was to compare the SBS and bond failure site of brackets bonded to fluorosed and non-fluorosed teeth with SEP and PA.

Materials and methods
A power calculation showed that for a power of 0.982387, a sample of 20 teeth would be required.

Teeth
Forty fluorosed and 40 non-fluorosed fresh human mandibular premolar teeth were used in this study. All the teeth had been extracted from orthodontic patients aged between 12 and 18 years. The teeth were collected after receiving verbal consent to retain them for this study. The classification of fluorosed teeth was made by the consensus of two investigators (AMSS and DI) using the TFI (Thylstrup and Fejerskov, 1978). These teeth were mildly fluorosed (TFI = 1–3) and were obtained from an area endemic for dental fluorosis (Isparta, Turkey). The non-fluorosed teeth were acquired from a non-endemic area (Samsun, Turkey).

The fluorosed and non-fluorosed teeth were divided into two subgroups of 20 teeth each according to the bonding procedure. Each tooth was embedded in a cold-cure acrylic resin (Orthocryl; Dentaurum, Ispringen, Germany) cylindrical block. A jig was used to align the buccal surface of each tooth parallel to the base of the cylinder. The teeth were cleansed and polished with non-fluoride pumice and a rubber prophylactic cup for 10 seconds, washed with water, and dried.

Brackets
Stainless steel lower premolar brackets (Gemini bracket; 3M Unitek, Monrovia, California, USA) were used. The mean area of each bracket base according to the manufacturer was 10.61 mm².

Bonding procedure
The brackets were bonded according to one of the following two protocols:

In the first subgroup, the teeth were etched with 37 per cent PA for 30 seconds, washed for 20 seconds, and dried for 10 seconds. After etching, a thin uniform coat of primer (Transbond XT; 3M Unitek) was applied. The adhesive resin (Transbond XT Light Cure Adhesive Paste; 3M Unitek) was placed onto the bracket base and the bracket was positioned on the enamel surface. Excess adhesive resin was removed with an explorer. Adhesive resin was polymerized for a total of 20 seconds from two directions using a visible light-curing unit (Hilux 200; Benlioglu Dental Inc., Ankara, Turkey) with an output power of 600 mW/cm².

In the second subgroup, SEP (Transbond Plus Self Etching Primer; 3M Unitek) was applied to the enamel surface and rubbed for 3 seconds. A gentle burst of dry air was then delivered to thin the primer. Five teeth were etched with one SEP unit. The applicator was re-dipped into the reservoir for saturation between individual teeth. The bonding procedure with Transbond XT adhesive resin was performed as in the first subgroup.

Debonding procedure
Two minutes after bonding, the samples were placed in distilled water (37°C) for 24 hours to prevent dehydration. Subsequently, the samples were subjected to thermocycling testing of 1000 cycles. Thermocycling was performed between 5 and 55°C with a dwell time of 30 seconds as advised by the International Organization for Standardization (2003).

Shear bond testing was performed with a universal testing device (Lloyd LRX; Lloyd Instruments Ltd, Fareham,
Hants, UK) at a crosshead speed of 1 mm/minute. The bond strengths were calculated in megapascals (MPa).

**Residual adhesive**

The enamel surfaces were examined with a stereomicroscope (Stemi 2000-C; Carl Zeiss, Göttingen, Germany) at a magnification of ×10 to determine the amount of composite resin remaining according to the adhesive remnant index (ARI; Årtun and Bergland, 1984).

**Statistical analysis**

Two-way analysis of variance (ANOVA) was used to determine significant differences among teeth affected by fluorosis, bonding protocol, and their interactions. All treatment combination means for bond strength values were compared using the *post hoc* Tukey’s multiple comparison test ($P < 0.05$).

A Weibull analysis was performed, and the Weibull modulus, characteristic bond strength, correlation coefficient, and the stress levels at the 5 and 10 per cent probability of failure were calculated.

Kruskal–Wallis and Mann–Whitney U-non-parametric tests were used to determine whether there were any significant differences in the ordinal ARI values. In order to avoid an accumulation of errors due to multiple comparisons (to compare groups two by two, six comparisons were made), a Bonferroni correction was carried out. The significance level ($P < 0.05$) was modified by dividing by the number of comparisons made, and therefore, $P < 0.0083$ was considered significant.

**Results**

The results of two-way ANOVA, used to determine significant differences among fluorosis, bonding procedure, and their interactions, are shown in Table 1. The main effects show a significant difference for fluorosis ($P < 0.01$) and bonding procedure ($P < 0.001$) on the bond strength values. Furthermore, a significant interaction between fluorosis and bonding procedure was observed ($P < 0.01$).

The mean SBS, minimum and maximum values, and standard deviations (SD) for each group are given in Table 2. The results of the Tukey’s multiple comparisons test to compare the mean SBS are presented in Table 2 together with the parameters of the Weibull analysis for each group. The Weibull distribution plots of the probability of failure at a certain shear stress level for the two groups are depicted in Figure 1.

The lowest SBS was observed when bonding brackets to fluorosed teeth with SEP (Figure 2). This bond strength value (9.01 MPa) was significantly different from those of fluorosed teeth etched with PA (15.22 MPa) and non-fluorosed teeth conditioned with SEP (12.95 MPa) and PA (15.37 MPa).

The median, mean, SD, and range of the ARI scores are given in Table 3. The Kruskal–Wallis test indicated that there were significant differences between the groups ($chi$-square $= 14.423$, $P = 0.002$). The Mann–Whitney U-test showed that ARI scores of fluorosed teeth conditioned with SEP were significantly different from those of non-fluorosed teeth conditioned with SEP ($P = 0.005$) or PA ($P = 0.007$).

**Table 1** Two-way analysis of variance of force (Megapascals) required to debond metal brackets from the enamel surface. df, degrees of freedom.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Sum of Squares</th>
<th>df</th>
<th>Mean square</th>
<th>$F$ ratio</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fluorosis</td>
<td>83.834</td>
<td>3</td>
<td>83.834</td>
<td>16.584</td>
<td>0.006</td>
</tr>
<tr>
<td>Bonding protocol</td>
<td>371.984</td>
<td>1</td>
<td>371.984</td>
<td>35.082</td>
<td>0.000</td>
</tr>
<tr>
<td>Fluorosis × bonding protocol</td>
<td>71.724</td>
<td>1</td>
<td>71.724</td>
<td>6.764</td>
<td>0.010</td>
</tr>
<tr>
<td>Error</td>
<td>805.837</td>
<td>76</td>
<td>10.603</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Corrected total</td>
<td>1333.379</td>
<td>79</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Table 2** Mean shear bond strengths, standard deviations (SDs), minimum and maximum values, and Weibull parameters for each group ($n = 20$).

<table>
<thead>
<tr>
<th></th>
<th>Mean (MPa)*</th>
<th>SD</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Weibull analysis</th>
</tr>
</thead>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Weibull modulus</td>
</tr>
<tr>
<td>Phosphoric acid</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-fluorosed teeth</td>
<td>15.37 A</td>
<td>3.40</td>
<td>10.70</td>
<td>20.58</td>
<td>5.00</td>
</tr>
<tr>
<td>Fluorosed teeth</td>
<td>15.22 A</td>
<td>3.07</td>
<td>9.72</td>
<td>20.05</td>
<td>5.35</td>
</tr>
<tr>
<td>Self-etching primer</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-fluorosed teeth</td>
<td>12.95 A</td>
<td>3.25</td>
<td>9.47</td>
<td>22.96</td>
<td>4.71</td>
</tr>
<tr>
<td>Fluorosed teeth</td>
<td>9.01 B</td>
<td>3.31</td>
<td>4.12</td>
<td>16.10</td>
<td>3.05</td>
</tr>
</tbody>
</table>

*Means for groups having the same letters show homogeneous subsets.
time with 37 per cent PA for fluorosed teeth. These prolonged etching times were chosen since the etching pattern of fluorosed teeth for 120 and 180 seconds appeared to cause a similar etch pattern to that of normal teeth etched for 60 seconds. On the other hand, Ng’ang’a et al. (1992) reported that etch depth and etch pattern of non-fluorotic and fluorotic teeth did not demonstrate any significant difference after 60 seconds etching with 40 per cent PA. According to Ng’ang’a et al. (1992) such contradictory results may stem from factors such as the evaluation criteria for ‘resistance’ to etching, differences in the age and relative concentrations of fluoride in the experimental samples, the types and concentrations of acids, and the etching times.

Before bracket bonding, grinding or micro-abrasion of fluorosed enamel surface has been recommended by several authors to increase bond strength (Opinya and Pameijer, 1986; Miller, 1995; Duan et al., 2006). After mechanical grinding of 100 μm of the enamel surface, no significant difference between the TBS of normal and ground fluorosed teeth was observed (Opinya and Pameijer, 1986). In a study evaluating failure rate of brackets bonded to fluorosed teeth, Noble et al. (2008) reported that additional micro-mechanical abrasion with 50 μm of aluminium silicate was not necessary to increase micro-mechanical bracket retention when an adhesion promoter was applied. Elimination of micro-mechanical abrasion results in the preservation of enamel, prevents a roughened enamel surface adjacent to the bracket, and allows for a bonding appointment that is more time-efficient, less complicated, and more comfortable for the patient and the orthodontist (Noble et al., 2008).

Richards et al. (1989) found that the concentration of fluoride in the outermost enamel increased with increasing TFI score. Regardless of the severity of fluorosis, the fluoride concentration profiles were similar and the highest concentration of fluoride was in the outer 200 μm of enamel.

### Discussion

In the present study, SBS values obtained with 37 per cent PA etching for 30 seconds did not show any significant difference between fluorosed and non-fluorosed teeth. This result is in agreement with the findings of Ng’ang’a et al. (1992). However, Opinya and Pameijer (1986) and Adanir et al. (2009) observed significant differences between bond strengths of fluorosed and non-fluorosed teeth with PA application.

In those studies (Opinya and Pameijer, 1986; Ng’ang’a et al., 1992; Adanir et al., 2009), different etching times were used, such as 30, 60, and 150 seconds. Opinya and Pameijer (1986) recommended 120 and 180 seconds etching time with 37 per cent PA for fluorosed teeth. These prolonged etching times were chosen since the etching pattern of fluorosed teeth for 120 and 180 seconds appeared to cause a similar etch pattern to that of normal teeth etched for 60 seconds. On the other hand, Ng’ang’a et al. (1992) reported that etch depth and etch pattern of non-fluorotic and fluorotic teeth did not demonstrate any significant difference after 60 seconds etching with 40 per cent PA. According to Ng’ang’a et al. (1992) such contradictory results may stem from factors such as the evaluation criteria for ‘resistance’ to etching, differences in the age and relative concentrations of fluoride in the experimental samples, the types and concentrations of acids, and the etching times.

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### Table 3  Frequency distribution of the adhesive remnant index (ARI scores)a.

<table>
<thead>
<tr>
<th>ARI scoresb</th>
<th>0</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>Median</th>
<th>Mean</th>
<th>SD</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Phosphoric acid</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-fluorosed teethc</td>
<td>—</td>
<td>13</td>
<td>2</td>
<td>5</td>
<td>1</td>
<td>1.60</td>
<td>0.88</td>
<td>1–3</td>
</tr>
<tr>
<td>Fluorosed teeth</td>
<td>1</td>
<td>18</td>
<td>—</td>
<td>1</td>
<td>1</td>
<td>1.05</td>
<td>0.51</td>
<td>0–3</td>
</tr>
<tr>
<td>Self-etching primer</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-fluorosed teethd</td>
<td>—</td>
<td>13</td>
<td>1</td>
<td>6</td>
<td>1</td>
<td>1.65</td>
<td>0.93</td>
<td>1–3</td>
</tr>
<tr>
<td>Fluorosed teethc,d</td>
<td>13</td>
<td>—</td>
<td>6</td>
<td>1</td>
<td>0</td>
<td>0.75</td>
<td>1.07</td>
<td>0–3</td>
</tr>
</tbody>
</table>

a Chi-square = 14.423, P = 0.002.
bARI scores: 0, no composite left on the enamel surface; 1, less than half of the composite remaining; 2, more than half of the composite remaining; and 3, all composite remaining.
cSignificant difference between the groups (P = 0.007).
dSignificant difference between the groups (P = 0.005).
The fluoride concentrations of teeth with TFI score 1 were higher than those of teeth with TFI score 0. The fluoride concentrations in teeth with TFI score 2 or 3 were significantly higher than those of teeth with TF score 0 or 1 up to a depth of 50 µm. Even though fluoride concentration increases with the severity of dental fluorosis, Al-Sugair and Akpata (1999) found that the mean depths ofetch for teeth with TFI score 0–3 were not significantly different from each other after 37 per cent PA application. Those authors recommended that teeth with TFI score 1–3 should be etched for the same time as non-fluorosed teeth. In the present study, the similarity of SBS values of fluorosed and non-fluorosed teeth, obtained with the application of 37 per cent PA for 30 seconds, corroborate the recommendation of Al-Sugair and Akpata (1999). On the basis of this result, etching of mildly fluorosed teeth with 37 per cent PA for 30 seconds without any additional mechanical abrasion might produce a clinically acceptable SBS.

In the present study, the SBS of fluorosed teeth conditioned with SEP was significantly lower than that of non-fluorosed teeth conditioned with SEP, and the values of non-fluorosed and fluorosed teeth etched with PA. This significant difference between SBS values of fluorosed and non-fluorosed enamel with SEP is in agreement with the result of Ermis et al. (2007), i.e. the lower micro-TBS value with the two-step self-etch adhesive was attributed to the fluoridated apatite, which is less soluble in acid. They concluded that the hypermineralized surface layer on the teeth with TFI score 5 affected the micro-TBS of the self-etch adhesive. Nevertheless, Ratnaweera et al. (2007) stated that the enamel bond strength of two all-in-one adhesive systems was not affected by mild to moderate fluorosis in unground enamel.

Ermis et al. (2007) concluded that the preparation of enamel might be critical to obtain efficient adhesion to enamel surfaces for self-etch adhesives. Grinding of enamel removes the hypermineralized and acid resistant outer surface of fluorosed teeth (Weerasinghe et al., 2005; Ermis et al., 2007). After the grinding procedure, no significant difference between micro-TBSs of moderately fluorosed and non-fluorosed enamel was observed with the two-step self-etch adhesive (Ermis et al., 2007). Similarly, no significant differences were observed in the bond strengths between PA etching and self-etching bonding for normal and mildly fluorosed ground enamel (Weerasinghe et al., 2005). Even though grinding has been suggested to improve the bond strength of self-etch adhesives to fluorosed enamel, Shida et al. (2009) found a significant difference between the micro-SBSs of mildly fluorosed and non-fluorosed ground enamel with a two-step self-etching adhesive system. The comparison of bond strength measurements of different studies is complicated because of the variety of materials and methods, including variations in tooth type, storage conditions, method of debonding, analysis of the results, and the selection of products for comparison (Fox et al., 1994). A separate control for each study has been recommended because the SBS can differ significantly depending on the method used (Fritz et al., 2001).

Brackets are subjected to either tensile, shear, or torsion forces or a combination of these, which are difficult to measure (Movahhed et al., 2005). It was reported that clinically adequate TBSs for metal orthodontic brackets to enamel should range from 6 to 8 MPa (Reynolds, 1975). Although these values were suggested as adequate for most clinical orthodontic needs, the minimum clinically acceptable SBS is not known. In the present study, the SBSs were above these values suggested as adequate for all bonding procedures.

The Weibull analysis provides information concerning the probability of bracket failure and gives the clinician an indication of how the material is likely to perform in a clinical situation (Fox et al., 1994). Even for materials with a high mean bond strength, there is a finite measurable probability of failure occurring at relatively low stress levels (McCabe and Walls, 1986). In the Weibull plots in the present study, non-fluorosed and fluorosed teeth etched with PA were very similar initially, whereas fluorosed teeth conditioned with SEP significantly shifted to the left, indicating a higher probability of failure at low levels of stress. Littlewood et al. (2001) suggested using the 5 per cent chance of failure as a more appropriate level to assess bond strength. According to those authors, the bond strength of a material with a 5 per cent chance of failure should be at least 5.4 MPa. In the present study, the SBS of fluorosed teeth conditioned with SEP showed a lower shear stress level than 5.4 MPa at the 5 per cent probability of failure. These results provide some evidence that routine clinical use of SEP (Transbond Plus) to bond brackets to mildly fluorosed teeth is not recommended. Duan et al. (2006) reported the results of bracket failures bonded to severely fluorotic teeth (moderately severe or severe according to Dean’s Fluorosis Index) with different enamel preparations. Surface preparation via pumicing and etching with Transbond SEP showed the highest bond failure rate of 74.0 per cent. However, removing 0.1–0.2 mm of enamel before Transbond SEP application decreased the bond failure to 25.9 per cent.

For the fluorosed teeth conditioned with SEP, ARI scores were significantly lower than those for non-fluorosed teeth conditioned with SEP or PA. This lower ARI score indicated bond failure at the enamel–adhesive interface. This type of bond failure demonstrated that the bond between the adhesive and the bracket base and the cohesive strength of the adhesive were stronger than that between the adhesive and enamel. Weerasinghe et al. (2005) and Ratnaweera et al. (2007) observed a predominant adhesive failure with self-etch adhesives. Furthermore, enamel fracture or cohesive failure in enamel was observed for SEP and PA (Weerasinghe et al., 2005; Ermis et al., 2007; Ratnaweera et al., 2007). In the present study, there were no enamel fractures.
Conclusions

1. The results of this research indicate no differences in the SBS of orthodontic brackets between mildly fluorosed and non-fluorosed enamel etched with 37 per cent PA for 30 seconds.

2. SEP (Transbond Plus) showed lower SBS values for orthodontic brackets bonded to mildly fluorosed enamel. These results provide some evidence that routine clinical use of SEP (Transbond Plus) to bond brackets to mildly fluorosed teeth is not recommended.

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