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The effects of two soft drinks on bond strength, bracket microleakage, and adhesive remnant on intact and sealed enamel

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SUMMARY The purpose of this study was to evaluate the effects of Coca-Cola® and Schweppes® Limón on bond strength, adhesive remnant, and microleakage beneath brackets.

One hundred and twenty upper central incisor brackets were bonded to bovine incisors and divided into three groups: (1) Control, (2) Coca-Cola®, and (3) Schweppes® Limón. The teeth were submerged in the drinks three times a day for 15 minutes over a 15 day period. Shear bond strength (SBS) was measured with a universal testing machine, and adhesive remnant evaluated using image analysis equipment. Microleakage at the enamel–adhesive and adhesive–bracket interfaces was determined using methylene blue. One hundred and eight teeth were used for scanning electron microscopy to determine the effect of the drinks on intact and sealed enamel. SBS and adhesive remnant data were analysed using the Kruskal–Wallis test ($P < 0.05$) and microleakage using the Kruskal–Wallis and Mann–Whitney tests applying Bonferroni correction ($P < 0.017$).

No significant differences were found in SBS and adhesive remnant between the groups ($P > 0.05$). Microleakage at the enamel–adhesive interface for groups 2 and 3 was significantly greater than for group 1 ($P < 0.017$). At the adhesive–bracket interface, microleakage was significantly greater in group 2 than in group 1 ($P < 0.017$) while microleakage in group 3 did not differ significantly from either group 1 or 2 ($P < 0.017$). The drinks produced enamel erosion, loss of adhesive and microleakage. Coca-Cola® and Schweppes® Limón did not affect the SBS of brackets or the adhesive remnant.

Introduction

The term 'soft' drinks refers to all drinks except alcohol, mineral water, fruit juice, tea, coffee, or milk-based drinks, which may or may not be carbonated (Varnam and Sutherland, 1997). Recently, the consumption of soft drinks has increased (West *et al.*, 2000). They are damaging not only because of the high levels of sugar they contain but also because most have pH levels below the critical limit for enamel demineralization (pH <5.5; Dinçer *et al.*, 2002). Moreover, frequently consumed soft drinks have been shown to cause extreme dental erosion (Hunter *et al.*, 2000).

Dental erosion is defined as the acid-induced loss of hard tissue, a chemical process in which bacteria play no part; for this reason, it is not associated with dental plaque (Jensdottir *et al.*, 2004; Eygen *et al.*, 2005; Wongkhantee *et al.*, 2006; Barbería *et al.*, 2007). In an *in vivo* study, Jensdottir *et al.* (2006) found that the prevalence of dental erosion increased as the pH levels of the studied drinks decreased and as consumption increased. Other studies using scanning electron microscopy (SEM) have shown that soft drinks produce large areas of enamel decalcification (Rytömaa *et al.*, 1988; Meurman and Frank, 1991a; Grando *et al.*, 1996).

The appearance of white spot lesions caused by the demineralization of tooth enamel is a clinical problem associated with orthodontic treatment (Arikan *et al.*, 2006). Its prevalence is between 2 and 96 per cent in patients with fixed appliances (Arhun *et al.*, 2006) and is the result of demineralization processes occurring around and beneath the brackets due to a decrease in pH (Øgaard *et al.*, 1998). Various authors have suggested that microleakage around brackets might contribute to the formation of white spot lesions beneath the brackets (Arhun *et al.*, 2006; Arikan *et al.*, 2006). In restorative dentistry, microleakage is defined as the penetration of fluids and bacteria into the interface between the restoration and tooth. It has been shown that microleakage results in an increase in the probability of recurring caries and post-operative sensitivity (Mali *et al.*, 2006). However, the literature dealing with microleakage and its clinical consequences in orthodontics remains scarce (James *et al.*, 2003; Arhun *et al.*, 2006; Arikan *et al.*, 2006) and no study appears to have been carried out of the capacity of soft drinks to produce microleakage beneath orthodontic brackets. Nevertheless, studies that use SEM to evaluate the effect of soft drinks on enamel sealed with orthodontic adhesives have observed areas of enamel showing adhesive loss after exposure to soft drinks (Steffen, 1996; Dinçer *et al.*, 2002). This suggests that soft drink consumption may provoke an increase in microleakage beneath brackets and also compromise bond strength. The only study that deals with the influence of soft drinks on bracket bond strength showed a significant reduction in bond strength on exposure to this type of beverage (Oncag *et al.*, 2005).

The aims of this study were to evaluate the effect of two soft drinks, Coca-Cola® and Schweppes® Limón (lemon drink), on bond strength, on the adhesive remnant on teeth

after debonding, and on microleakage beneath brackets, and to observe by means of SEM the effect of these drinks on intact and sealed enamel.

Materials and methods

Teeth

Two hundred and twenty-eight bovine lower central incisors freshly extracted and free from enamel cracks, caries, and fillings were used in this research: 60 for bond strength testing, 60 for microleakage analysis, and 108 for SEM observation. The teeth were washed in water to remove any traces of blood and then placed in a 0.1 per cent thymol solution. They were then stored in distilled water, which was changed periodically to avoid deterioration.

Brackets

One hundred and twenty upper central incisor metal brackets (Victory Series®; 3M Unitek Dental Products, Monrovia, California, USA) were used. The base area of each bracket was calculated (mean = 10.25 mm^2) using image analysis equipment (Sony DXC 151-AP video camera; Sony Corporation, Tokyo, Japan) connected to an Olympus SZ11 microscope (Olympus Corporation, Tokyo, Japan) and MIP 4 software (Micron Image Processing Software; Digital Image Systems, Barcelona, Spain).

Bonding procedure

The brackets were bonded on the buccal surfaces with Transbond® XT (3M Unitek Dental Products) according to the manufacturer's instructions. The buccal surfaces were polished with a rubber cup and polishing paste (Détartrine®; Septodont, Saint-Maur, France). They were then etched with 37 per cent *o*-phosphoric acid gel (Total Etch®, Ivoclar Vivadent, Schaan, Liechtenstein) for 30 seconds and the enamel was washed with water. After washing, the enamel surface was completely dried with compressed air. A layer of Transbond® XT primer was applied to the tooth and Transbond® XT paste to the base of the bracket and pressed firmly onto the tooth. Excess adhesive was removed with a probe from around the base of the bracket and the adhesive was light-cured, positioning the light guide of an Ortholux XT lamp (3M Unitek Dental Products) on each interproximal side for 10 seconds.

Storage of test specimens and experimental groups

The specimens were divided randomly into three groups:

Group 1. Control (n = 40): The specimens were submerged for 15 days in artificial saliva [400 mg NaCl, 1.210 mg KCl, 780 mg $NaH_2PO_4 \cdot 2H_2O$, 5 mg $Na_2S \cdot 9H_2O$, 1.000 mg $CO(NH_2)_2$, 1.000 ml of distilled water and 10 N sodium hydroxide], renewing the saliva daily.

Group 2. Coca-Cola® (The Coca-Cola Company, Madrid, Spain; n = 40): Over a period of 15 days, the specimens were submerged in Coca-Cola® for 15 minutes three times a day, separated by intervals of 2 hours. At other times they were kept in artificial saliva.

Group 3. Schweppes® Limón (Bebidas de España S.A., Madrid, Spain; n = 40): The teeth were submerged in Schweppes® Limón following the same procedures as for group 2.

While the artificial saliva was kept at room temperature, both Coca-Cola® and Schweppes® were stored at a temperature of 5°C. The pH of each medium was measured electronically (Schott®, Mainz, Germany). The values obtained were 6.75, 2.40, and 2.55 for groups 1, 2, and 3, respectively.

Bond strength test

Of the 40 specimens in each group, 20 were used to carry out shear bond strength (SBS) testing. SBS was measured with a universal testing machine (Autograph AGS-1KND; Shimadzu, Kyoto, Japan) with a 1 kN load cell connected to a metal rod with one end angled at 30 degrees. The crosshead speed was 1 mm/minute (International Organization for Standardization, 1994).

The teeth were set at the base of the machine so that the sharp end of the rod incised in the area between the base and the wings of the bracket, exerting a force parallel to the tooth surface in an occluso-apical direction.

The force required to debond each bracket was registered in Newtons (N) and converted into megapascals (MPa) as a ratio of Newtons to the surface area of the bracket (MPa = N/mm^2).

Evaluation of adhesive remnant on teeth after debonding

The percentage of the surface of the bracket base covered by adhesive was determined using image analysis equipment and the MIP 4 software.

The percentage of the area still occupied by adhesive remaining on the tooth after debonding was obtained by subtracting the area of adhesive covering the bracket base from 100 per cent.

Microleakage testing

Twenty teeth were used to carry out microleakage testing. The teeth were dried with a dental air jet and covered with two coats of nail varnish (Resist and Shine; L'Oreal, Paris, France), leaving 1 mm around the edges of the bracket base uncovered. Afterwards, the specimens were submerged in a 1 per cent solution of methylene blue for 24 hours. In order to avoid penetration by the methylene blue through the apical foramen, the teeth were placed vertically in a container, fitting the roots into a metal grid so that the methylene blue only covered the crown of the tooth and the gingival third of the root.

The teeth were sectioned longitudinally in inciso-cervical direction with a water-cooled diamond saw (Horico®, Berlin, Germany) thus providing three sections per tooth. Each section was examined on both sides, so that each specimen underwent six examinations.

The percentage of microleakage for each interface was determined using image analysis equipment and the MIP 4 software, at the enamel–adhesive and adhesive–bracket interfaces, both on the gingival and incisal edge at ×100 magnification. The percentage of microleakage for the enamel–adhesive and adhesive–bracket interfaces was obtained by adding the percentages of microleakage observed at the incisal and gingival edges of each interface. All observations were carried out by the same researcher (RN).

Scanning electron microscopy

One hundred and eight teeth were divided into two groups: (1) intact enamel ($n = 54$) where the buccal surfaces were polished with a rubber cup and polishing paste, and (2) enamel etched and sealed with Transbond® XT primer ($n = 54$) where the buccal surfaces were polished with a rubber cup and polishing paste, etched with 37 per cent *o*-phosphoric acid gel and primed with Transbond® XT, which was light-cured for 20 seconds.

Of the 54 specimens that made up each group, 18 were placed in artificial saliva, 18 in Coca-Cola®, and 18 in Schweppes® Limón. The immersion cycles described above for storage were followed.

All specimens were cleaned in distilled water with ultrasonic agitation for 30 minutes and gently air-dried. Then they were affixed to SEM stubs, sputter coated with gold, and examined with a Jeol 6100 SEM (Tokyo, Japan) operating at 20 kV, at ×100 magnification. Images representative of the different surface treatments were captured and stored digitally.

Statistical analysis

The Kolmogorov–Smirnov normality test and the Levene variance homogeneity test were applied to the bond strength data and to the data for percentage of area of adhesive remaining on the tooth. As the data did not show a normal distribution, significant differences were evaluated using the Kruskal–Wallis test ($P < 0.05$).

As the data for the percentage of microleakage at the enamel–adhesive interface did not show homogeneity and the data for the percentage of microleakage at the enamel–adhesive interface were not normally distributed, significant difference between groups for each interface was evaluated using the Kruskal–Wallis test ($P < 0.05$), finding those groups that were significantly different by means of the Mann–Whitney test for two independent samples. In order to avoid an accumulation of errors due to multiple comparisons, the significance level was modified by dividing it ($P < 0.05$) with the number of comparisons made (Bonferroni correction). $P < 0.017$ was considered significant.

Results

There were no significant differences between the three groups for either bond strength ($P = 0.89$; Table 1) or the percentage of area occupied by adhesive after debonding ($P = 0.11$; Table 2).

For microleakage testing, at the enamel–adhesive interface, the Kruskal–Wallis test showed significant differences between the three groups ($P = 0.00$) and the Mann–Whitney test that both groups 2 and 3 had levels of microleakage that were significantly greater than group 1 ($P = 0.002$ and $P = 0.012$, respectively). The percentage of microleakage for groups 2 and 3 was similar ($P = 0.84$; Table 3). Figure 1a shows an example of microleakage at the enamel–adhesive interface.

At the adhesive–bracket interface, significant differences were observed in microleakage between the three groups (Kruskal–Wallis $P = 0.01$). Group 2 showed significantly greater microleakage than group 1 (Mann–Whitney $P = 0.00$). Microleakage in group 3 did not differ significantly from that in groups 1 and 2 ($P = 0.52$ and $P = 0.03$, respectively; Table 3). Microleakage at the adhesive–bracket interface is shown in Figure 1b.

SEM observations showed that the teeth with intact vestibular enamel that had been immersed in artificial saliva did not suffer any erosion, while those that had been immersed in the soft drinks showed significant erosion of the adamantine tissue (Figure 2). There was no change in the adhesive layer in teeth with etched and sealed vestibular enamel that had been immersed in artificial saliva. However, the specimens exposed to the soft drinks showed widespread loss of adhesive material, this being greater for group 2 (Figure 3).

Discussion

In recent years there has been an increase in the consumption of soft drinks among children and adolescents (Owens and Kitchens, 2007). In the UK in 1950, a thousand million

Table 1 Bond strength (MPa).

Groups	*n*	Mean	Median	SD	Range
Control	20	10.22	8.09	4.18	13.41
Coca-Cola®	20	9.90	8.75	3.94	14.68
Schweppes Limón®	20	10.11	9.95	3.31	11.80

The Kruskal–Wallis test did not show significant differences between the groups ($P = 0.89$).

Table 2 Percentage of area occupied by the adhesive on teeth after debonding.

Groups	*n*	Mean	Median	SD	Range
Control	20	62.19	66.24	18.18	74.02
Coca-Cola®	20	68.77	74.17	19.34	74.58
Schweppes Limón®	20	62.26	65.81	15.64	71.16

The Kruskal–Wallis test did not show significant differences between the three groups ($P = 0.11$).

Table 3 Percentage of microleakage at the enamel–adhesive and adhesive–bracket interfaces.

	Enamel–adhesive					Adhesive–bracket				
Groups	Mean	Median	SD	Range	*P**	Mean	Median	SD	Range	*P**
Control	3.08	2.18	2.67	11.49	A	1.59	1.51	1.89	7.89	A
Coca-Cola®	7.99	6.22	7.16	31.97	B	4.73	3.99	4.8	19.85	B
Schweppes® Limón	7.72	6.68	5.89	18.38	B	3.76	2.31	3.55	11.91	

*Different letters indicate values that are significantly different.

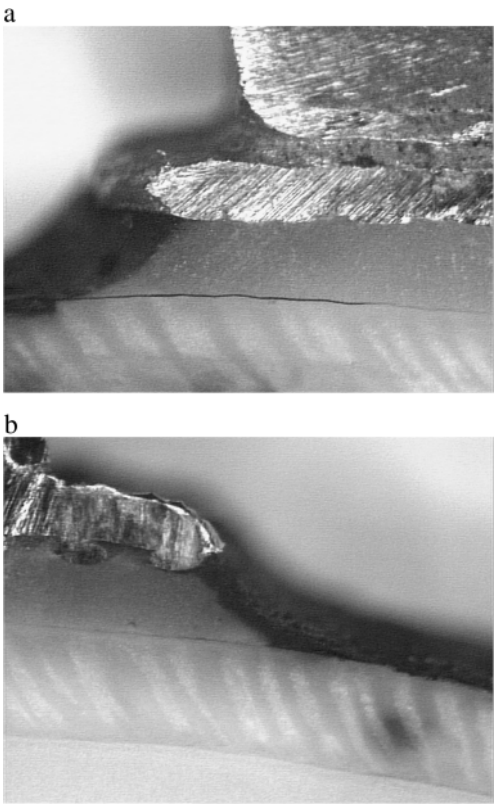

Figure 1 Microleakage at (a) the enamel–adhesive interface and (b) the adhesive–bracket interface of a specimen from the Coca-Cola® group (×100).

litres of soft drinks were sold; by 1990, this figure had increased 7-fold (West *et al.*, 2001).

The aim of this study was to evaluate the effects of Coca-Cola® and Schweppes® Limón on SBS, the adhesive remnant on teeth after debonding, and the microleakage beneath brackets bonded with Transbond XT. A further aim was to observe, by SEM, the effect of these drinks on intact enamel and on enamel sealed with Transbond XT. These particular soft drinks were chosen for several reasons: firstly, because of their high levels of consumption in Spain [52 per cent Coca-Cola and 13 per cent Schweppes® Limón; Ministerio de Agricultura, Pesca y Alimentación (MAPA), 2004]; secondly, because they have a pH level below the critical limit for demineralization of tooth enamel (Dinçer *et al.*, 2002), and finally, because of the

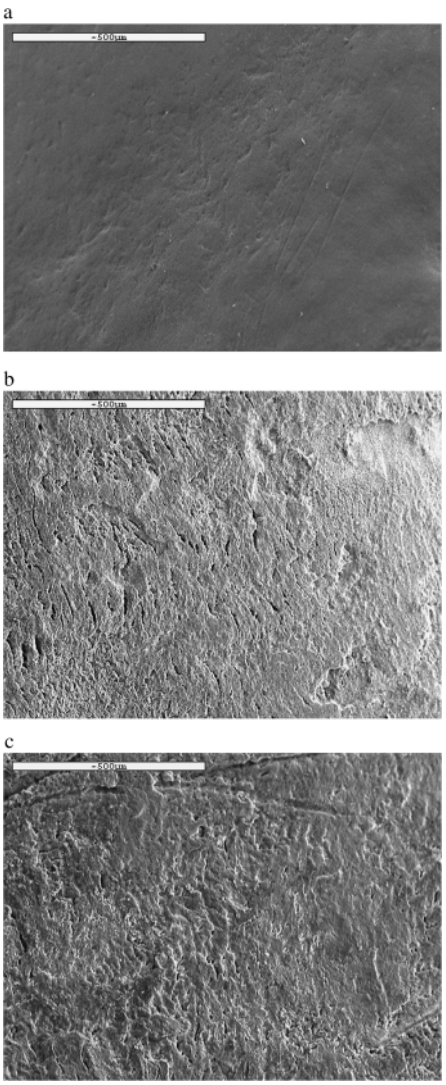

Figure 2 Scanning electron micrograph (×100) before immersion in the control (a), Coca-Cola® (b), and Schweppes® Limón (c) groups in specimens with intact vestibular enamel; bar = 500 μm.

type of acid they contain—phosphoric acid in the case of Coca-Cola® and citric acid in Schweppes® Limón, both of which are used in acid etching.

Lower bovine incisors were used in the study as it has been shown that bovine tooth enamel has similar properties to human enamel and achieves similar bond strengths (Nakamichi

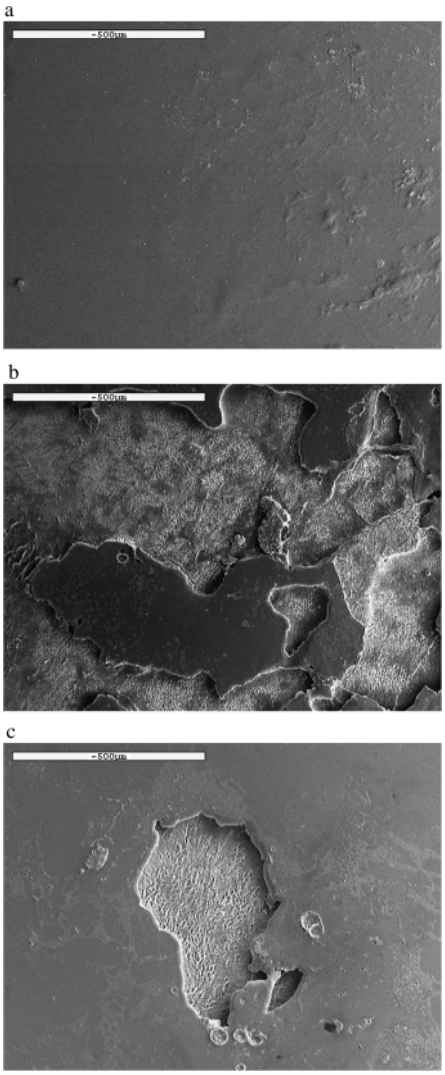

Figure 3 Scanning electron micrograph (×100) after immersion of a specimen in the control (a), Coca-Cola® (b), and Schweppes® Limón (c) groups in specimens with enamel etched and sealed with Transbond XT primer; bar = 500 μm.

et al., 1983; Fowler *et al.*, 1992; Baena *et al.*, 2003). These teeth have a nearly flat bonding surface, which avoids the difficulties of fitting a bracket base to a curved surface. In orthodontic bond strength tests, upper central incisor brackets are bonded to lower bovine incisors (Jaffer *et al.*, 2009; Minick *et al.*, 2009; Pithon *et al.*, 2009) because these teeth are closer to the size of upper human central incisors.

The immersion times and schedules used in previous studies vary widely. In general, specimens were submerged in the soft drinks continuously for long periods (Rytömaa *et al.*, 1988; Meurman and Frank, 1991a,b; Steffen, 1996; Jensdottir *et al.*, 2005; Owens and Kitchens, 2007), whereas in the present research an immersion schedule was used that would reproduce as closely as possible the situation *in vivo*. In this way, assuming that these drinks are consumed three times a day and that it might take around 45 minutes to consume one drink, the specimens were submerged in the drinks for 15 minutes at a time and afterwards in artificial saliva, a procedure that was repeated three times a day. The teeth were kept in saliva between immersions in the drinks in order to reproduce normal oral environment conditions and also to allow the possible remineralizing effects of saliva on enamel to take place (Oncag *et al.*, 2005; Jensdottir *et al.*, 2006).

The SBS and adhesive remnant values did not show significant differences between the three groups. No other study has analysed the effect of soft drinks on adhesive remnants after debonding and only one has dealt with the effect on bond strength. In that study, Oncag *et al.* (2005) observed bond strength values for brackets immersed in Coca-Cola® and Sprite that were significantly lower than in the control group. The difference between their results and the present findings may be due to the fact that their specimens were thermocycled; thermocycling having been shown by some researchers to reduce bond strength between 20 and 70 per cent (James *et al.*, 2003; Mali *et al.*, 2006).

In addition, no study appears to have evaluated the effect of soft drinks on the incidence of microleakage in either orthodontics or conservatory dentistry and, in spite of the clinical relevance of this phenomenon, orthodontic studies concerning microleakage are scarce (James *et al.*, 2003; Arhun *et al.*, 2006; Arikan *et al.*, 2006). The presence of microleakage at the enamel–adhesive interface is critical for the appearance of white spot lesions resulting from demineralization and for the formation of caries, while microleakage at the adhesive–bracket interface is related to bond failure (Arikan *et al.*, 2006).

The present study, microleakage values at the enamel–adhesive interface were significantly higher for groups 2 and 3 than for group 1. Microleakage values for group 3 did not differ significantly from group 2. At the adhesive–bracket interface, microleakage was seen to be significantly greater for group 2 than for group 1, while microleakage in group 3 did not differ significantly from that in either group 1 or group 2.

These results do not seem to confirm that microleakage at the adhesive–bracket interface is a decisive factor for bond strength given that, in spite of the significantly greater microleakage at the adhesive–bracket interface in group 2 than in group 1, no significant difference in bond strength was observed between the two groups. James *et al.* (2003) also did not find any relationship between adhesive–bracket microleakage and reductions in bond strength; indeed, the highest levels of microleakage were observed in the group that showed the greatest bond strength.

Erosion observed using SEM was less for the intact enamel specimens in group 2 than that observed by other researchers (Rytömaa *et al.*, 1988; Meurman and Frank, 1991a; Grando *et al.*, 1996; Owens and Kitchens, 2007), probably due to the fact that in most studies the teeth were immersed continuously in the drink and without artificial saliva use, ignoring the

possible remineralizing effect of saliva. In agreement with the study of Dinçer *et al.* (2002), teeth etched and sealed with bond material and immersed in Coca-Cola® showed large areas lacking adhesive with the enamel beneath exposed.

The obtained images of the teeth in group 3 are not comparable with other studies as there has been no previous investigation of the effects of this particular drink. Even so, the results are similar to those of lemon Fruko® (Dinçer *et al.*, 2002). Those researchers also observed that the lemon drink affected intact enamel and sealed enamel to a lesser extent than Coca-Cola®.

Conclusions

1. No significant differences were observed in bond strength or adhesive remaining on the teeth after debonding in any of the three groups.
2. Significantly greater microleakage at the enamel–adhesive interface was found in groups 2 and 3 than in group 1. There were no significant differences between groups 2 and 3. However, at the adhesive bracket interface, significantly more microleakage was found in group 2 than in group 1, whereas microleakage in group 3 did not differ significantly from that in groups 1 and 2.
3. Both Coca-Cola® and Schweppes® Limón produced enamel erosion and loss of adhesive material.

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