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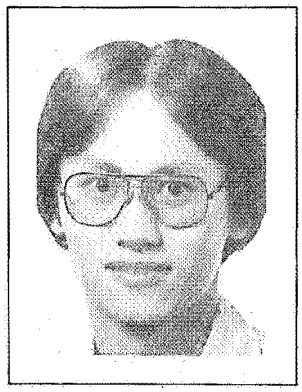
Microleakage at the etched enamel-resin interface with bonded orthodontic brackets



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Abstract:
This study was undertaken to investigate microleakage at the etched enamel-resin interface of directly bonded orthodontic brackets using a 2% aqueous fluorescein solution as a tracer. The buccal surfaces of 3 groups of 30 extracted premolar teeth were etched and Begg plastic brackets were directly bonded. One group was kept in Hanks' Balanced Salt solution for 24 hours at 37 degrees C (24 hour control group). Another group was thermally cycled between 4 degrees C and 60 degrees C for 200 minutes (thermal cycling group). The last group was aged in Hank's Balanced Salt solution for 3 months at 37 degrees C (3 months aging group). Each tooth was then sectioned bucco-lingually through the centre of the bracket and the section was examined with an ultraviolet light microscope. The depth of fluorescein penetration along the enamel-resin interface was measured using a calibrated eyepiece graticule.

The results of this study showed that: (i) microleakage occurred in 93.3% of the 24 hour control group, 91.6% of the 3 month aging group and 100% of the thermal cycling group, (ii) large variations in the microleakage depths were observed although, in the majority of cases, the fluorescein penetrated less than 0.27mm, (iii) a significant difference ($p < 0.01$) existed between the control teeth and the thermally cycled teeth with thermal cycling increasing the mean microleakage depth, and (iv) there was no significant difference between mean microleakage depth for the control teeth and teeth that were aged for 3 months but a tendency towards reduced microleakage after ageing was observed.

Reference: Aust.Orthod.J. 9(3): 270-275 (1986)

Key words: microleakage, bonding, enamel-resin interface, tracer

INTRODUCTION

Buonocore (1955) was the first to demonstrate that the bonding strength of acrylic materials to enamel could be substantially increased by conditioning the enamel surface with 85% phosphoric acid. The acid-etch technique is now an accepted procedure in dentistry.

The purpose of this study was to investigate and measure the depth of microleakage along the etched enamel-resin interface of directly bonded orthodontic brackets using a fluorescent dye technique. The effects of thermal cycling and ageing for 3 months were also evaluated.

Various ingenious techniques have been developed to study marginal permeability at the interface between the tooth and the bonding material. *The margins of restorations are* not fixed, inert and impenetrable borders, but rather are *'dynamic microcrevices which contain a busy traffic of ions and molecules'* (Myers 1966).

Penetration Markers

The use of organic dyes as tracers is one of the oldest and most popular methods of detecting microleakage (Going 1972, Shortall 1982). Christen and Mitchell (1966) studied microleakage around dental restoration and found that fluorescent dyes were particularly useful for demonstrating leakage. Their study also showed that fluorescein was superior to rhodamine B because fluorescein appeared brighter, was not quenched and could be detected in lower concentrations. In 1969, Loiselle et al. reported the use of fluorescein dye in the in vivo study of microleakage in the teeth of golden Syrian hamsters and in humans thus demonstrating the potential of fluorescein as a tracer in microleakage studies.

Radioactive salts have been used extensively as marked substances (Going et al. 1960; Phillips et al. 1960; Barber and Massler 1964; Rudolph et al. 1974; Hembree and Andrews 1976; Crisp and Wilson 1980). Their widespread use is based partly on the inherent ability of the isotopes to penetrate more deeply than the dye tracers, and partly on the fact that the autoradiographic techniques permit the detection of minute amounts of tracer that otherwise could not be seen (Going 1972).

A microleakage experiment involving bacteria was developed as early as 1929 when Fraser tested cements and filling materials placed in glass ampoules to determine whether they would allow bacteria to pass through or around the specimens. Seltzer (1955) and Rose et al. (1955) substituted teeth for glass and studied the seal of acrylic restorations. Mejare et al. (1979) investigated the occurrence, viability and identification of bacteria beneath unlined composite resin restorations, placed in vivo, using anaerobic culturing as well as histobacteriological methods of assessment.

Air pressure was introduced for the detection of marginal leakage by Harper in 1912. Pickard and Gaylord (1965) adapted the method to study the marginal seal of amalgam restorations placed in human teeth. Recently, Granath and Svenson (1970) studied microleakage of restorative materials with a more refined air pressure method. The main limiting factor of this method of study is that it is confined to use in vitro, and it does not simulate conditions in vivo.

The scanning electron microscope (SEM) provides a means of direct visual observation of the adaptation of restorative materials to cavity margins (Boyde and Knight 1969). Lee et al. (1970) attempted to correlate microleakage of radioactive isotopes with fissure size between tooth and restoration as seen in the SEM. No direct correlation was found, which led the authors to suggest that improved leakage tests were required. Grundy (1971) described a replica technique for the SEM and claimed that replica techniques did not change the structures being evaluated and could be repeated many times at different intervals, thus allowing one to follow changes in the size of a marginal defect on a longitudinal basis. Kidd (1976) commented that the use of the replica technique might have overcome the problem of introducing artifacts during specimen preparation, but the results were still difficult to quantify and not related to

diffusion and penetration. She felt that it would be unwise to equate cavity adaptation, as seen in the SEM, with microleakage and suggested that more work was needed.

Thermal Cycling

Nelsen et al. (1952) estimated the limits of oral thermal tolerance to be 60 degrees C and 4 degrees C, whereas a more recent study (Plant et al. 1974) found that while subjects would sip hot coffee at 60 degrees C, they would drink it comfortably between 50 and 55 degrees C. Under normal drinking conditions the temperature at the tooth surface ranges from 45 degrees to 15 degrees C. Kidd (1976) noted that results of temperature cycling experiments have almost invariably shown increased leakage after subjection of specimens to thermal treatment and concluded that it was obvious that some form of thermal stressing should be incorporated in microleakage study.

Factors affecting the bonding of orthodontic brackets

Moisture contamination (particularly saliva) is a potential problem during the etching procedure (Hormati et al. 1980), When saliva contacts the etched enamel, the salivary proteins clog the porosities of the etched enamel with subsequent chemical absorption. Diedrich (1981) suggested that a rapid wiping of the finger or cotton wool over the etched enamel surface could produce an almost total destruction of the etched structure. He studied the effect of moisture contamination and found that after salivary or mucosal contact (5 seconds), all etched surfaces were partially covered with a layer of salivary glycoprotein and the penetration of the adhesive material into the enamel structure was considerably reduced. Similar findings resulted from contamination by dispersed oil from the air compressor system of the dental unit. Young et al. (1975) studied the physical factors affecting adhesion of fissure sealant to enamel and found that moisture contamination of the etched enamel caused a 70% reduction in bond strength. These findings are in agreement with the observations of Hormati et al. (1980). Furthermore, etched enamel samples that were dried with air only after saliva contamination showed significantly less shear strength than etched enamel samples that were re-etched for 60 seconds, then washed and dried after the contamination with saliva. They concluded that should saliva contamination occur, the resin should not be applied directly to the contaminated surface. The surface should be dried, re-etched for 10 seconds, rewashed and redried.

MATERIALS AND METHODS

Sound human premolar teeth extracted for orthodontic reasons from patients of chronological age less than 16 years were obtained from the Children's Dental Hospital, Extraction Clinic of the University of Queensland Dental School and private dental practitioners. These teeth were cleaned and placed in Hanks' Balanced Salt* solution and stored at 4 degrees C until required, the solution being changed weekly.

Commercially available direct bonding orthodontic adhesives fall into three compositional categories: (a) the methyl methacrylate acrylic resins; (b) the polycarboxylate cements and (c) the dimethacrylates, of which the bisphenol A and glycidyl methacrylate (BIS-GMA) systems are a part (Johnson et al. 1976). The Right-On No Mix** adhesive bonding system was used. This is a difunctional methacrylate self-polmerising, single paste bonding system. The manufacturer's instructions were strictly followed during the investigation.

Begg plastic (Lexan)*** brackets with tunnelled bases were chosen for this investigation for ease of sectioning. The base of each bracket is circular and has a diameter of 3.5mm. A mild abrasive powder was used to clean the teeth. A mixture of sixteen grams of powder to 8.5 grams by weight of distilled water was prepared. The mixture was stored in a humidor until required.

Only teeth from patients who required extraction of all four premolars were used. Each of the premolar teeth was assigned at random to each of the treatment groups: (1) thermal cycling group, (2) 3 months ageing group, and (3) 24 hour control group. Each treatment group consisted of 30 teeth. The teeth were washed with distilled water and the buccal surface of each crown was pumiced for 30 seconds under a load of 450 grams. A weighing machine was used to indicate the load. A rubber cup was used for the pumicing procedure. The rotational speed of the rubber cup was kept constant.

Each tooth crown was then washed with distilled water and dried with oil-free compressed air. A masking tape with a 3.5mm diameter hole was centred over the buccal surface of the tooth. The margin of the hole was lightly burnished to provide a reliable seal. The exposed crown area was etched with the conditioning solution for the recommended 60 seconds. The reaction products and excess acid were removed with distilled water and dried again for a period of 20 seconds. The tooth was then transferred to and secured onto a specially constructed acrylic holder.

A thin coat of activator was placed on the etched tooth surface using a disposable brush provided in the kit. With a pair of tweezers a plastic bracket was removed from the sealed envelope and its base was coated thinly with the activator. A small amount of adhesive paste was dispensed directly from the syringe onto the activated bracket base and the bracket was pressed immediately to the tooth with a slight rotational action. A seating pressure of 450 grams for 5 seconds was applied. Any excess of adhesive was removed from the masking tape. This method met with the requirements suggested by Mizrahi and Smith (1969) for standardising experimental techniques. After the brackets had been bonded, the teeth were subjected to one of three treatments.

Thermal Cycling

The thermal cycling device included two temperature controlled water baths, one capable of maintaining water temperature at 4 degrees C and the other at 60 degrees C. A beaker containing 2% aqueous fluorescein solution was immersed in each of the water baths. Fifteen of the 30 teeth in this treatment group were placed in a stainless steel wire basket which was attached to one end of an immersing arm. The remaining 15 teeth were placed in another similar basket suspended at the other end of the arm. The baskets were then lowered into fluorescein solution for 30 seconds. The baskets were raised at the end of 30 seconds, rotated through 180 degrees and lowered into the fluorescein solutions again. This procedure was repeated for 200 cycles. The total immersion time was 200 minutes.

Three Months Aging

The 30 teeth in this group were stored for 3 months in Hanks' Balanced Salt solution maintained at 37 degrees C, the solution being changed twice weekly. At the end of the 3 months the teeth were immersed in 2% aqueous fluorescein solution for 200 minutes.

24 Hour Control

The 30 teeth in this group were stored for 24 hours in Hanks' Balanced Salt solution maintained at 37 degrees C. At the end of the treatment the teeth were immersed in 2% aqueous fluorescein solution for 200 minutes.

After each treatment, the teeth were removed from the fluorescein solution, thoroughly washed and allowed to dry for 6 hours (Christein and Mitchell, 1966).

The root of each tooth was removed. Sectioning of the crown was carried out with the Bovis Plano-Matic# hard tissue sectioning machine. Each tooth was sectioned

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** T.P. Laboratories, Inc., LaPorte, Indiana.
*** T.P. Laboratories, Inc., LaPorte, Indiana.

Allied Fluid Power Limited, Birmingham, England.

bucco-lingually through the centre of the bracket into halves with a diamond edged blade at 400 r.p.m. under a continuous water spray *(Figure 1.)*.

The left and right sections of each tooth were examined under ultraviolet light using an Olympus BHA microscope with epi-illumination and two BG 12 excitor filters at 100 x magnification. A calibrated eyepiece graticule was used to measure the depth of fluorescein penetration along the etched enamel-resin interface to the nearest 0.01 mm (T1), both occlusally and gingivally. There were four measurements for each tooth, left-occlusal (LO), left-gingival (LG), right-occlusal (RO), and right-gingival (RG). All specimens were measured twice. The second measurement (T2) was carried out after an interval of two weeks.

A paired t-test between the first and second sets of measurements was carried out to determine the consistency of the measuring method. A split-plot analysis on the combined measurements was used to evaluate the significance of the effects produced by the experimental treatments and to determine if there were differences in the depth of fluorescein penetration between the occlusal and gingival measurements as well as between the left and right sections of each tooth.

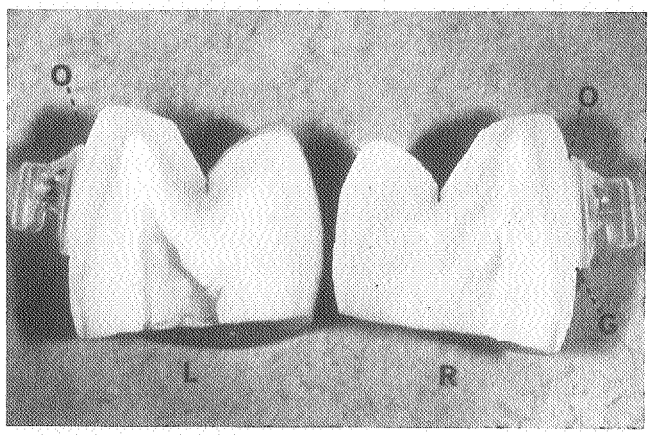

Figure 1. Sectioned tooth crown showing left (L) and right (R) sections. (O) and (G) indicate fluorescein penetration from occlusal and gingival directions.

RESULTS

Duplicate measurements (T1 and T2) of microleakage depths for the 24 hour control, thermal cycling and 3 months ageing groups are tabulated in ***Tables 1, 2 and 3*** respectively. ***Table 4*** shows the mean microleakage of the combined first and second measurements. ***Table 5*** shows the overall mean microleakage depths for the treatment groups. Analysis of variance is shown in ***Table 6***. Examples of photomicrographs showing different degrees of fluorescein penetration along the enamel-resin interface are presented in ***Figures 2-4***.

Consistency of Measuring Method

The consistency of the measuring method was determined by performing a paired t-test between the duplicate measurements at time 1 and time 2 listed in ***Tables 1-3***. This gave a t value of 0.038 which was not significant. Confirmation of this result is shown in the analysis of variance *(Table 6)* where the method error mean square (0.0015) was very small in relation to the sub-plot error mean square (0.1371). Thus the measuring method was consistent.

Differences in Microleakage Depth

Table 4 shows the mean microleakage depths (first and second measurements combined) for the three groups. The analysis of variance showed no significant differences between mean microleakage depths for the left and right sections and for the occlusal and gingival measurements of all the treatment groups. More detailed inspection of the data revealed that the gingival mean microleakage values of both the control and ageing groups were slightly higher than the occlusal values. This indicated a slight tendency of deeper penetration along the enamel-resin interface by the fluorescein dye from the gingival direction. However, this finding was not reflected in the thermal cycling group and therefore care is needed in interpreting the differences in microleakage between occlusal and gingival measurements. In several samples fluorescein dye was seen to penetrate along the enamel prisms into the subsurface layer. This occured mainly in the gingival third region under the bracket, as indicated in ***Figure 3.***

Table 1. Duplicate measurements of microleakage depths (mm) for the 24 hour control group

PATIENT	LEFT				RIGHT			
	OCCLUSAL		GINGIVAL		OCCLUSAL		GINGIVAL	
NUMBER	T1	T2	T1	T2	T1	T2	T1	T2
1	0.22	0.22	0.48	0.50	0.10	0.11	0.31	0.30
2	0.61	0.63	0.62	0.62	0.40	0.42	0.61	0.62
3	0.60	0.62	0.15	0.14	0.55	0.52	0.10	0.10
4	0.20	0.22	0.16	0.18	0.40	0.40	0.10	0.11
5	0.33	0.33	0.33	0.31	0.20	0.20	0.25	0.27
6	0.15	0.15	0.06	0.06	0.65	0.66	0.08	0.08
7	0.25	0.23	0.40	0.45	0.25	0.21	0.17	0.16
8	0.10	0.11	0.17	0.20	0.28	0.25	1.05	1.05
9	0.00	0.00	0.52	0.50	0.11	0.10	0.05	0.03
10	0.29	0.30	0.23	0.23	0.20	0.15	0.27	0.26
11	0.00	0.00	0.20	0.25	0.06	0.07	0.05	0.05
12	0.13	0.15	0.17	0.16	0.21	0.22	0.41	0.41
13	0.50	0.50	0.17	0.17	0.54	0.52	0.21	0.22
14	0.31	0.30	0.08	0.08	0.27	0.26	0.35	0.35
15	0.20	0.20	0.61	0.60	0.42	0.42	0.25	0.24
16	0.70	0.70	0.09	0.08	0.28	0.28	0.00	0.00
17	0.24	0.23	0.00	0.00	0.45	0.45	0.00	0.00
18	0.18	0.16	0.30	0.32	0.43	0.43	0.18	0.18
19	0.08	0.08	0.37	0.36	0.21	0.22	0.09	0.10
20	0.16	0.16	0.00	0.00	0.13	0.13	0.13	0.13
21	0.09	0.09	0.24	0.22	0.14	0.14	0.85	0.85
22	0.26	0.26	0.15	0.15	0.15	0.15	0.35	0.35
23	0.26	0.26	0.23	0.22	0.26	0.26	0.10	0.10
24	0.57	0.56	0.25	0.25	0.25	0.96	0.37	0.37
25	0.25	0.23	0.15	0.15	0.00	0.00	0.05	0.06
26	0.14	0.13	0.10	0.10	0.25	0.24	0.10	0.10
27	0.13	0.12	0.10	0.14	0.31	0.30	0.36	0.35
28	0.10	0.10	0.24	0.23	0.19	0.18	0.10	0.11
29	0.19	0.18	0.00	0.00	0.10	0.10	0.42	0.40
30	0.25	0.25	1.25	1.25	0.20	0.20	1.40	1.40

T1 = First measurement
T2 = Second measurement taken two weeks later

Table 2. Duplicate measurements of microleakage depths (mm) for the thermal cycling group

PATIENT	LEFT				RIGHT			
	OCCLUSAL		GINGIVAL		OCCLUSAL		GINGIVAL	
NUMBER	T1	T2	T1	T2	T1	T2	T1	T2
1	0.24	0.25	0.13	0.13	0.30	0.30	0.18	0.18
2	0.36	0.34	1.12	1.12	0.62	0.61	1.12	1.12
3	0.56	0.55	0.07	0.07	0.58	0.54	0.22	0.23
4	0.70	0.68	0.20	0.19	0.43	0.43	0.27	0.27
5	0.45	0.45	2.00	2.00	0.28	0.28	0.88	0.85
6	0.96	0.95	0.13	0.13	0.28	0.92	0.15	0.15
7	0.40	0.42	0.22	0.23	0.38	0.40	0.25	0.25
8	0.21	0.20	0.75	0.73	0.24	0.22	1.62	1.62
9	0.25	0.25	0.17	0.17	0.24	0.24	0.26	0.22
10	0.38	0.38	0.35	0.33	0.28	0.28	0.66	0.68
11	0.24	0.24	0.04	0.03	0.24	0.24	0.07	0.07
12	0.16	0.16	0.33	0.32	0.13	0.12	0.11	0.12
13	0.46	0.45	0.15	0.15	0.27	0.26	0.49	0.50
14	0.39	0.37	0.17	0.16	0.44	0.40	0.41	0.43
15	0.16	0.17	0.30	0.32	0.39	0.39	0.15	0.16
16	0.51	0.50	0.18	0.18	0.30	0.30	0.21	0.15
17	0.56	0.56	0.29	0.28	0.44	0.43	0.40	0.25
18	0.48	0.47	0.16	0.16	0.32	0.32	0.21	0.20
19	0.44	0.40	0.13	0.12	0.51	0.54	0.66	0.65
20	0.13	0.13	0.11	0.10	0.22	0.22	0.18	0.16
21	0.30	0.29	0.83	0.80	0.47	0.45	0.65	0.65
22	0.15	0.13	0.10	0.10	0.67	0.67	0.35	0.34
23	0.22	0.23	0.13	0.11	0.20	0.20	0.05	0.04
24	1.25	1.25	1.55	1.55	0.39	0.40	0.57	0.55
25	0.06	0.06	0.25	0.25	0.10	0.11	0.40	0.39
26	0.75	0.73	0.10	0.10	0.30	0.30	0.06	0.06
27	0.51	0.52	0.08	0.08	0.63	0.65	0.15	0.15
28	0.34	0.33	0.25	0.25	0.62	0.63	0.45	0.45
29	0.32	0.30	0.17	0.15	0.80	0.80	0.12	0.15
30	0.56	0.56	0.45	0.45	1.25	1.25	0.25	0.30

T1 = First measurement
T2 = Second measurement taken two weeks later

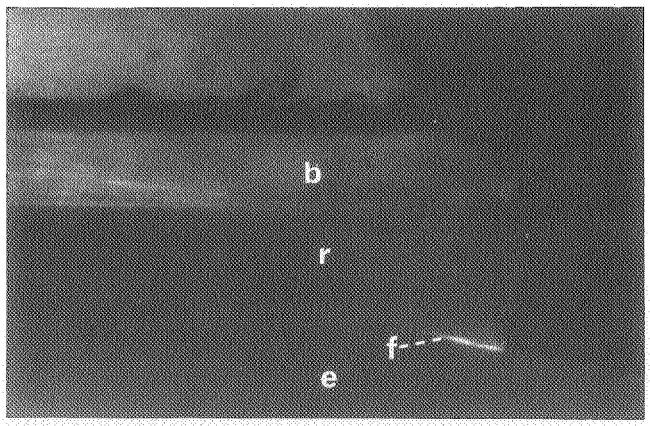

Figure 2. Occlusal third region showing slight fluorescein (f) penetration under the bracket (b = bracket; r = resin; e = enamel)

Table 3. Duplicate measurements of microleakage depths (mm) for the 3 months ageing group

PATIENT NUMBER	LEFT				RIGHT			
	OCCLUSAL		GINGIVAL		OCCLUSAL		GINGIVAL	
	T1	T2	T1	T2	T1	T2	T1	T2
1	0.10	0.10	0.15	0.13	0.09	0.08	0.18	0.16
2	0.85	0.85	0.46	0.46	1.05	1.05	0.45	0.45
3	0.06	0.08	0.07	0.07	0.06	0.07	0.07	0.07
4	0.06	0.06	0.12	0.11	0.22	0.23	0.05	0.05
5	0.24	0.23	0.31	0.31	0.25	0.25	0.31	0.31
6	0.20	0.20	1.25	1.25	0.05	0.05	0.85	0.87
7	2.25	2.25	2.25	2.25	0.55	0.57	0.80	0.80
8	0.20	0.20	0.10	0.12	0.08	0.07	0.03	0.04
9	0.12	0.13	0.12	0.12	0.15	0.15	0.06	0.06
10	0.15	0.15	0.07	0.08	0.16	0.14	0.08	0.08
11	0.10	0.10	0.00	0.00	0.15	0.15	0.00	0.00
12	0.15	0.15	0.16	0.16	0.17	0.18	0.32	0.33
13	0.06	0.06	0.34	0.36	0.20	0.20	0.20	0.22
14	0.06	0.08	0.10	0.10	0.00	0.00	0.08	0.08
15	0.35	0.33	0.05	0.06	0.13	0.12	0.10	0.11
16	0.16	0.17	0.09	0.08	0.30	0.30	0.40	0.38
17	0.00	0.00	0.42	0.44	0.11	0.09	0.92	0.93
18	0.07	0.08	0.10	0.10	0.15	0.13	0.07	0.09
19	0.32	0.35	0.32	0.32	0.14	0.14	0.45	0.45
20	0.10	0.10	0.61	0.62	0.21	0.20	0.65	0.65
21	0.13	0.15	0.10	0.10	0.13	0.12	0.17	0.15
22	0.00	0.00	0.13	0.13	0.10	0.11	0.08	0.08
23	0.12	0.14	0.28	0.22	0.22	0.22	0.20	0.20
24	0.30	0.31	0.06	0.06	0.17	0.15	0.07	0.07
25	0.10	0.11	0.00	0.00	0.30	0.30	0.04	0.05
26	0.07	0.07	0.04	0.05	0.04	0.04	0.07	0.05
27	0.00	0.00	0.06	0.06	0.00	0.00	0.96	0.96
28	0.55	0.58	0.30	0.10	0.60	0.60	0.30	0.30
29	0.06	0.07	0.27	0.27	0.04	0.04	0.55	0.53
30	0.33	0.31	0.00	0.00	0.60	0.60	0.00	0.00

T1 = First measurement
T2 = Second measurement taken two weeks later

Table 4. Mean microleakage depths* (mm) for treatment groups

	24 HOUR CONTROL		THERMAL CYCLING		3 MONTNS AGEING	
	Left	Right	Left	Right	Left	Right
Occlusal	0.25	0.28	0.41	0.42	0.24	0.21
Gingival	0.26	0.29	0.36	0.38	0.27	0.28

Standard error of each mean = 0.048
* = Mean over the first (T1) and second (T2) measurements and the 30 patients from Tables 1, 2 and 3.

Table 5. Overall mean microleakage depths * (mm) for treatment groups

24 HOUR CONTROL	THERMAL CYCLING	3 MONTHS AGEING
0.27	0.39	0.25

Standard error of each mean = 0.035
* = The overall means were obtained by combining left-occlusal, left-gingival, right-occlusal and right gingival values from Table 4.

Differences Between Treatments

A. 24 Hour Control and Thermal Cycling

There was a statistically significant difference ($p<0.05$) between mean microleakage depths for teeth that were subjected to thermal cycling and teeth in the control group. The overall mean microleakage depth for teeth in the control group was 0.27 mm, whilst that for teeth subjected to thermal cycling was 0.39 mm (standard error of each mean = 0.035), indicating that thermal cycling increased the mean microleakage depth.

In the control group, microleakage depth ranged from 0.00 to 1.40 mm. No leakage was recorded in 8 specimens *(Table 1).* In contrast, all specimens in the thermal cycling group showed fluorescein penetration — the range was from 0.06 to 2.00 mm *(Table 2).* Moreover, 55.8 per cent of the measurements of the thermal cycling group were found to have a value greater than 0.27 mm whereas only 33.3% of the control group had a value greater than 0.27 mm. In general, the intensity of fluorescein dye was found to be stronger in specimens that were thermally cycled than in specimens of the control group.

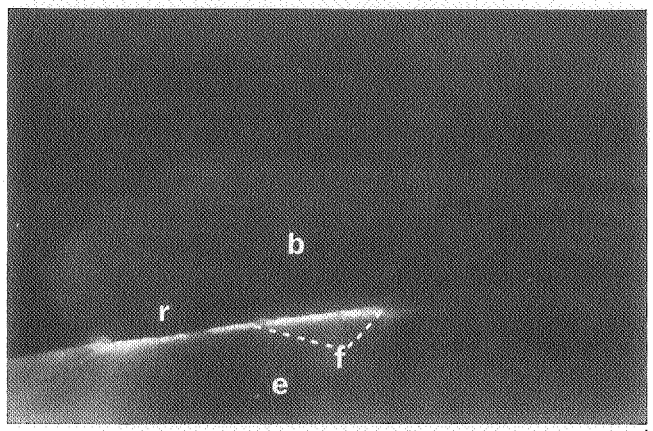

Figure 3. Gingival third region under a bracket showing fluorescein (f) penetration along the enamel-resin interface and into the sub-surface enamel layer (b = bracket; r = resin; e = enamel)

Table 6. Split-plot analysis of variance

Source	df	SS	MS	E	
Patients	29	15.3457	0.5292	1.770	ns
Treatments	2	2.8324	1.4162	4.737	*
Error (a)	58	17.3401	0.2990		
Teeth	89	35.5181			
Sites	3	0.0427	0.0142	0.104	ns
L v R	1	0.0184	0.0184	0.134	ns
O v G	1	0.0075	0.0075	0.055	ns
(L v R)*(O v G)	1	0.0168	0.0168	0.123	ns
Tr x Sites	6	0.3411	0.0569	0.415	ns
Tr x (L v R)	2	0.0466	0.0233	0.170	ns
Tr x (O v G)	2	0.2829	0.1414	1.031	ns
Tr *(L vR)*(O v G)	2	0.0116	0.0058	0.042	ns
Error (b)	261	35.7941	0.1371		
Sub Total	359	71.6961			
Method Error	360	0.5227	0.0015		
Total	719	72.2188			

ns = Not significant
R = Right
L = Left
* = $p \times<0.05$
O = Occlusal
G = Gingival

B. 24 Hour Control and 3 Months Ageing

No significant differences between mean microleakage depths were found in the control and ageing groups. However, as shown in *Table 5* the overall mean microleakage depth of the ageing group was 0.25 mm (S.E. = 0.035), slightly less than that of the control group (0.27 mm). In addition, no leakage was recorded in 10 specimens and only 30% of the measurements had a microleakage depth greater than 0.27 mm (compared with 33.3% of the control group). This indicated that the *ageing process tended to reduce microleakage.*

In one tooth of the ageing group the fluorescein dye penetrated the etched enamel-resin interface completely. The reason for this isolated instance is not known although contamination of the etched enamel surface is a possibility. The intensity of the fluorescein dye in the ageing group was similar to that in the control group.

Further Observations

Patient No. 2 showed mild fluorosis for all treatments. It was found that the mean microleakage measurements of this sample in the control, ageing and thermal cycling treatments were 0.57, 0.70, and 0.80 mm respectively, much higher than the respective treatment means. It was also noted that the fluorescein dye penetrated readily along enamel prisms into the subsurface layer. In a few teeth where cracks in enamel were present, dye penetration was observed following the fissures *(Figure 4).*

DISCUSSION

The penetration of adhesive material into the microscopic pores in the enamel is conducive to the retention of the materials. The etching of the enamel surface increases wettability and creates a large surface area for bonding and opens up spaces or pores in the enamel into which the adhesive can flow and ultimately polymerise (Gwinnett 1967), thus providing a good marginal seal.

In this study microleakage occurred along the etched enamel-resin interface of directly bonded orthodontic brackets in 93.3% of the 24 hour control group, 91.6% of the 3 months ageing group and 100% of the thermal cycling group. This indicated that the leakage incidence was high although in quantitative terms, the fluorescein dye penetrated only slightly (less than 0.27 mm) in the majority of specimens. This study also supports the general view that there is no material which will chemically bond to tooth substance to form a perfect seal capable of withstanding the moist oral environment and its attendant temperature changes.

The *large variation in microleakage depth* observed may be explained by the differences in enamel solubility. Silverstone (1957) and Diedrich (1981) have shown that in spite of an identical pretreatment, there are *significant variations and differences in the distribution and appearance of the etch relief from one tooth to another and also on the same tooth.*

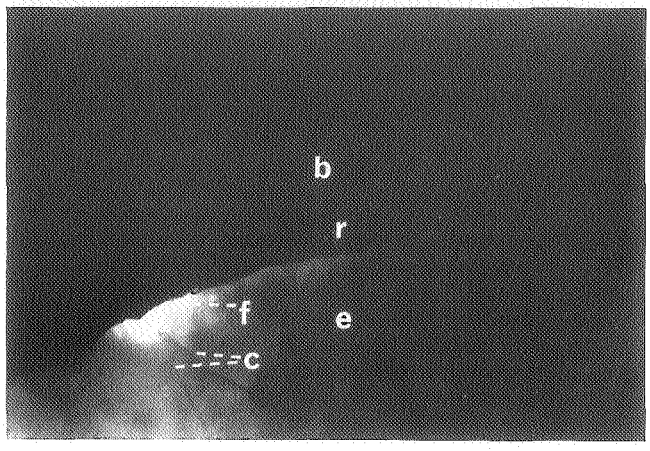

Figure 4. Photomicrograph showing fluorescein (f) penetration along cracks (c) in enamel (e). (b = bracket/ r = resin).

The selective solubility is due to structural variations in the superficial enamel layer — differences in the course of prisms and in crystallite orientation, anisothrophy of the chemical reaction of single crystallites, and local differences in the mineral content and in the organic/inorganic composition of prisms (Sharpe 1967).

The presence of a prismless layer on the enamel surface has been observed by several workers. Ripa et al. (1966) have reported that prismless enamel is present on all deciduous teeth and on 70% of permanent teeth. Prismless enamel is more resistant to attack by organic acids than regions where the prisms have reached the surface. This may be due to the fact that the crystallites are packed closely together because of their parallel arrangement, thus reducing the volume of pores or spaces. In several samples the fluorescein dye penetrated along enamel prisms into the subsurface layer of the gingival third region under the bracket.

Treatment with fluoride reduces the wettability of surface enamel and results in the formation of reaction products. These products can partly or completely fill the interprismatic and other spaces. (Kochavi et al. 1975). This interferes with resin penetration and tag formation and thus results in reduced bond strength (Sheykholeslam and Buonocore 1972). On the basis of the well-recognised resistance of fluoridated enamel to acid dissolution, one would expect to find *more leakage with teeth showing fluorosis.* This assumption is reflected by the severe fluorescein penetration along the enamel-resin interface observed in sample No. 2.

As the fluoride uptake, as well as the fluoride distribution, differs in each tooth Wickwire and Renz (1973) have suggested that *one should consider the history of enamel in regard to fluoride as a variable factor in etching time.* The omission of fluoride application sometime before a bonding procedure might also have a favourable effect (Lehman and Davidson 1981).

The effect of thermal cycling observed in this study supports the findings of microleakage studies carried out on resinous restorative materials and fissure sealants (Powell 1975). The increase in intensity of the fluorescein dye observed in specimens subjected to thermal cycling may be due to widening of microspaces between the bonding materal and the enamel surface because of differences in the coefficient of thermal expansion. This is particularly important with acrylic resins (Right-On bonding material is an acrylic resin) since its coefficient of thermal expansion is seven times greater than that of the tooth structure. A marginal space of 10 microns could develop when a restoration is subjected to temperature changes of 4 degrees C to 60 degrees C. *Although this is below the limit of visibility (50 microns) it is larger than the diameters of common bacteria found in the mouth* (for example, Streptococcus mutans, 0.8 to 1 micron). In Seltzer's (1955) study micro-organisms were found in 21.3% of restorations of thermally cycled teeth whereas no organism penetrated the margin of restoration of control teeth that had not undergone thermal changes.

Although not significant, a tendency towards reduced microleakage was observed in specimens that were aged for three months. This observation supports the findings of Going and Sawinski (1966) who reported a decrease in marginal leakage of composite restorations with ageing of 4 and 8 weeks. Powell (1975) also found a significant decrease in microleakage of pit and fissure sealants with ageing of 3 months.

The reduction in microleakage after ageing may be due to the tendency of acrylic resin to take up water by a process of imbibition.

SUMMARY AND CONCLUSIONS

The results of this study showed that:

Microleakage occurred along the etched enamel-resin interface of directly bonded orthodontic brackets in

93.3% of the 24 hour control group, 91.6% of the 3 months ageing group and 100% of the thermal cycling group.

Large variations in microleakage depths were observed and differences in enamel solubility was thought to be the cause.

A statistically significant difference ($p < 0.01$) between the means was found for the control teeth and the thermally cycled teeth. Thermal cycling increased the mean microleakage depth. This was thought to be due to differences in the coefficient of thermal expansion between the bonding material and the tooth structure.

No significant difference existed between the control teeth and teeth that were aged for 3 months. However, there was a tendency towards reduced microleakage after ageing. Water sorption was thought to be responsible for the reduction in microleakage.

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