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Operative Dentistry publishes articles that advance the practice of operative dentistry. The scope of the journal includes conservation and restoration of teeth; the scientific foundation of operative dental therapy; dental materials; dental education; and the social, political, and economic aspects of dental practice. Review papers, book reviews, letters, and classified ads for faculty positions also are published.

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GUEST EDITORIAL

Where is the Next Generation of Dental Academics?

One of the critical points of discussion at the recent international dental schools and dental research meeting in Vancouver was the absence of new teachers and investigators entering dental academics. As a dean, and increasingly as an elder, I am most concerned about the diminishing pool of young people who choose careers in dental education and research.

The American Association of Dental Schools (AADS) estimates that there are 300 unfilled full-time and 100 unfilled part-time positions in US dental schools. For full-time positions, that is an average of about six core faculty for each school. Moreover, many of the searches for these core positions have been ongoing without success for several years. Rick Valachovic, Executive Director of AADS, reported that the three major concerns expressed at the meeting were faculty recruitment, faculty development, and faculty retention, and he noted a consensus among US and Canadian deans that this crisis of replacing our retiring faculty will have a significant effect on all aspects of dental practice, teaching, research, and service.

At the same time, Steven Bayne, President of the American Association of Dental Research (AADR), which represents a majority of dental faculty, reminded us that the average age of the Association's membership was 51 years. New dental graduates are not headed along the path of graduate training and competition for research funding, a fact that may reflect the $90,000 average burden of debt already carried by our students at time of graduation. In response, both AADS and AADR have assigned the highest priority to exploring ways that will encourage greater student exposure to teaching and research. Concurrently, the National Institute of Dental and Craniofacial Research is making a major effort to address the crisis with new approaches to training grants and novel mentoring programs with associated loan forgiveness. However, all such programs will surely fail if there are no applicants.

The number of new dental educators and scientists is a very small proportion of the national graduating class, but those few are essential to the future of oral health care. We must explore new training opportunities, enhance the oral health research environment, and increase the attraction for a career in dental research. We must facilitate the ability of dental students who are interested in an academic career to pursue those interests within the dental curriculum. We must sustain the growth of student research groups in all our dental schools. Most importantly, we must act as mentors. The primary responsibility for the next generation rests with those who can mentor those young people now in our dental classes, labs, clinics, and offices. Mentor is described in *The Odyssey* of Homer as a wise and trusted counselor. Barnet Levy, who is responsible for fundamental work in oral pathology but also known for his lifelong gift of mentoring, recently observed, "Most of my students went on to make a much greater contribution to Dental Science than I could have or did. That, I think, is what teaching is all about. Teachers bend so the student or young colleague can stand on their shoulders. Then the teacher straightens up so the student can reach the bottom rung of their ladder to success."

Like Barnet Levy, we must be mentors and counselors, and encourage students to participate in teaching through honors programs, community service, and involvement in established study clubs, as well as research of all kinds, be it biological, behavioral, or clinical. If we make a commitment to mentoring, most new graduates will still choose to enter full-time private practice, but they will be better dentists for that participation. Some will contribute to part-time teaching, an essential component of all dental training programs. A few will become the next generation dedicated to a career in education and research. In my view, the dental profession must intensify its efforts to mentor and support the next generation of leaders in dental academics, or the excellence in dental care so long enjoyed by our patients will be lost.

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Dean
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BUONOCORE MEMORIAL LECTURE

Michael Buonocore

Thoughts on Contemporary Restorative Materials

E STEVEN DUKE

INTRODUCTION

In the field of restorative dentistry, clinicians are faced with decisions regarding the selection of a vast number of materials to restore deteriorated dentitions. Never before in the history of the profession have so many different materials been presented as alternatives for very similar diseased conditions. For example, with direct esthetic restorative applications a clinician must decide whether to use a traditional composite resin, a new "packable" composite resin, a traditional glass-ionomer cement, a resin-modified glass-ionomer cement, a compomer restorative material, or a flowable composite formulation. In making these decisions, what criteria should be used? Obviously there is always the issue of form and esthetics, yet even when these are considered today the answer is often difficult. In some respects there exists a "revolution" when one discusses options available to clinicians. Procedures that have served the profession for over a hundred years find themselves at the edge of extinction. There is no question that material selection is changing away from metallic to more tooth-colored restoratives. While there has always been a balance between form and function in the profession and dental educational programs, decisions are becoming more frequently based on esthetics, with form and function moving to a secondary position. With this move a new meaning of the phrase "definitive" restorative procedure has developed. While in the past definitive care might have implied 25 to 30 years of service for a restorative treatment, some materials have demonstrated limited longevity, with a time frame of 3 to 5 years being quite common (Duke, Robbins & Treviño, 1994; Hume & others, 1996; Mjör & Qvist, 1997). As such, when a number of new materials becomes the treatment of choice, the term of definitive treatment, as we will see, undergoes significant change.

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E Steven Duke, DDS, MSD, professor and chair

MATERIALS UPDATES

A review of recent materials research data substantiates the issues raised above. In the area of adhesive restorative procedures advances are being made; however, studies are limited in number and the conclusions expressed by Van Meerbeek and others (1996) in a comparison of current adhesives seem very appropriate regarding modern adhesive systems: "A highly predictable level of retention was achieved with Clearfil Liner Bond System and Scotchbond Multi-Purpose, but *none* of the systems was able to guarantee *complete marginal sealing.*" Their paper highlights the advancements being made in the area of adhesives, yet further development is needed to reach the level of performance expected by most clinicians. Furthermore, the resin-based adhesive technology is the backbone of many modern esthetic procedures, such as ceramic inlays/onlays, laminate veneers, and complete ceramic crown restorations. If we are to progress in this area, then further durability needs to be achieved by adhesive systems, as they are the limiting factor of many of these procedures.

In the area of compomers, interest has risen sharply in recent years. This is despite unfavorable data that have surfaced in the literature (Roeters & others, 1998; Andersson-Wenckert, Folkesson & van Dijken, 1997). The compomer may very well be an excellent example of what trends have developed in the profession. On a global basis, the compomer is an easily placed esthetic restorative treatment that can be accomplished in a limited amount of clinical time. In spite of the inferior physical and mechanical properties of compomers, their ease of use has elevated their use in the profession. When you combine the financial compensation factor, which is equal to that of a more durable composite resin, the conclusions one draws are difficult to accept. In a 3-year study on children, Roeters and others (1998) demonstrated the extreme ease of use of a compomer. However, they also reported considerable wear of the material. In another study on children by Andersson-Wenckert and others (1997), similar findings were found with a basic conclusion that compomers had a higher failure rate than conventional composite resin materials in primary molars.

In the area of glass-ionomer technology, the resin-modified glass ionomer was introduced as an improvement over the original formulation. Very quickly dentists embraced the resin-modified glass-ionomer cements and their associated claims of improvement. In reality, the most significant improvement has been in the area of handling characteristics. Clinical trials have established that these materials are not competitive with the original formulation of glass ionomer (Matis, Cochran & Carlson, 1996; de Gee & others, 1996; Duke & Treviño, 1998). Again, ease of use has surfaced as a factor for material selection rather than durability and patient oral health interests.

The further evolution of resin-modified glass-ionomer formulations has resulted in a class of luting resins as alternative resin cements or alternatives to traditional zinc phosphate cement. An excellent example of the premature adoption of a technology prior to scientific validation is demonstrated with the resin luting cement Advance. Clinicians were reporting incidences of fracture with ceramic crown restorations from time to time. Yet, all reports were anecdotal without scientific validity. A recent publication (Leevailoj & others, 1998) examined Advance resin cement, relative to fracture incidence of ceramic crowns in the laboratory. It was found to result in 100% fracture after only a couple of months with porcelain fired full crowns stored in distilled water. It is truly unfortunate that such data were not available prior to the general use of this material on our patients.

If we examine the available evidence for selection of the "ceromer" or "packable" composite resin materials class of restoratives, one quickly concludes that results can equally be achieved with traditional direct placed composite formulation. Evidence is not available to support many of the claims made by clinicians and industry regarding these "alternatives to amalgam" and "alternatives to cast restorations" (Freiberg & Ferracane, 1998). Regardless, selection criteria appear to be focused more on esthetics rather than longevity or oral health parameters.

CONCLUSION

Presenting such a depressing platform for material selection and patient management is without question disturbing and disheartening. Yet, one cannot deny the trends that have developed in the profession. How does such a direction develop in a profession so deeply embedded in a field of science? An interesting question, and one that may be difficult to answer. My personal thoughts are that several factors have contributed to the state of affairs we find ourselves in today. First, clinicians do not have adequate resources available to make informed decisions on what is hype and what is valid when it come to new technologies. Second, keynote clinicians are presenting material without scientific evidence in a fashion that is accepted without question by clinicians. And finally, industry is providing the materials and technology that meet the demands of the profession. The 40 billion dollar market in dental materials and techniques was not introduced by industry, it was produced by industry. Until clinicians rely upon sound evidence-based clinical decision parameters,

the present trend will continue.

Advances in the field of dentistry are needed. There is no question that progress is needed in materials science. As a profession, let's move forward as our previous leaders, such as Dr Michael G Buonocore progressed. Yet, there is a scientific method for progress that has been the standard for examining new materials and procedures. While we look forward, maybe we should take a few minutes to look back. The future of the dental profession and the oral health of the public we have dedicated ourselves to serve are deserving of nothing less.

(Delivered 19 February 1999)

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ORIGINAL ARTICLES

Total Bonding vs Selective Bonding: Marginal Adaptation of Class 2 Composite Restorations

B THONEMANN • M FEDERLIN
G SCHMALZ • W GRUNDLER

Clinical Relevance
Although microleakage at the cervical margin was not prevented by either method tested, total bonding may be a suitable alternative to selective bonding when using certain materials.

SUMMARY

For improving the marginal integrity of composite restorations, a total bonding method has been advocated besides the conventional selective bonding method. Total bonding avoids the placement of a base. The entire internal cavity surface is available for the adhesive bond. Selective bonding involves the placement of a base covering the pulpal floor as well as the pulpoaxial wall. In this study, five dentin/enamel bonding system/composite combinations were used to restore 60 class 2 cavities with their cervical margins below the cementoenamel junction (CEJ); six teeth per material were restored according to the total bonding and the selective bonding methods. Before and after simultaneous thermocycling and mechanical loading (TCML) marginal adaptation was evaluated on replicas in the SEM. Microleakage was determined by dye penetration on the original samples after TCML. The data were statistically evaluated with the Mann-Whitney *U*-test and the Wilcoxon test. The error rates method was applied. In SEM analysis the error rates method indicated a significant difference between the two restoration methods in general. In the pairwise comparisons, no significant differences between the selective bonding and total bonding methods were found for Syntac/Tetric, Gluma 2000/Pekafill, and Gluma/Pekafill. With Scotchbond Multi-Purpose (SBMP) and All-Bond 2 (AB2), total bonding revealed significantly ($P \leq 0.01$) less gap formation before and after TCML than selective bonding. Accordingly, total bonding showed significantly less ($P \leq 0.01$) dye penetration with Scotchbond Multi-Purpose and All-Bond 2 compared to selective bonding. In conclusion, the reduction of microleakage by application of the

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total bonding method depended upon the bonding system used. Total bonding could be an alternative procedure for the adhesive restoration of class 2 cavities when their gingival margins are apical to the cementoenamel junction, provided the proper system is used and pulp damage is prevented.

INTRODUCTION

The complex adhesive bond between composite resin on the one hand and enamel and dentin on the other is mediated by bonding systems and a carefully applied adhesive restoration technique. However, gap formation and microleakage caused by polymerization shrinkage stresses and the different physical properties of tooth structure and restorative materials cannot be completely eliminated, especially when the gingival margins are located apical to the CEJ (Blaser, 1993; Derhami, Coli & Brännström, 1995; Prati & others, 1994). Material properties of the bonding systems and composite resins used, the cavity design and depth, the operative techniques, such as the mode of application of the bonding systems, and the insertion and curing techniques of the composite resin are factors that significantly influence the cavosurface bond between tooth structure and restorative material (Lutz, Krejci & Schüpbach, 1993). Fusayama and others (1979) advocated simultaneous etching of enamel and dentin. They claimed that "total etching" of enamel and dentin, along with the subsequent application of a bonding system, produced a protective resin-impregnated dentin layer, making the placement of a base material obsolete (Fusayama, 1990). Not all bonding systems include simultaneous total etching of enamel and dentin with one material but do advocate separate pretreatment of enamel and dentin. Lutz and others (1993) introduced the "total bonding method" in contrast to the conventional selective bonding method, which requires the placement of a base material.

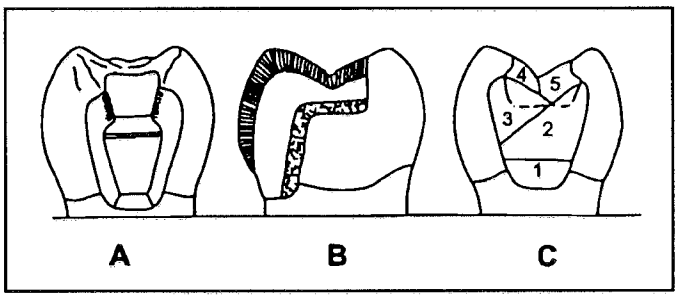

Figure 1. *A: Cavity preparation--occlusal-approximal, box-shaped, rounded inner angles, cervical margin approximately 1 mm below the cementoenamel junction; B: Selective bonding--placement of base material on the occlusal cavity floor and along the pulpoaxial wall; C: Placement of the composite increments--1-3 indicate approximal increments, 4 and 5 indicate occlusal increments. Dotted line indicates occlusal cavity floor.*

Selective bonding includes the placement of a glass-ionomer base material (Krejci, Lutz & Krejci, 1988; Blaser, 1993) in order to reduce the overall composite volume and allow free surface areas to compensate for the negative effects caused by polymerization shrinkage stresses (Davidson, de Gee & Feilzer, 1984; Feilzer, de Gee & Davidson, 1987). The glass-ionomer base remains unetched during the subsequent restorative procedures with the composite resin. The adhesive bond is selectively restricted to the enamel and dentinal margins not covered by base material.

Total bonding avoids the placement of a base material: the entire internal cavity surface is used to adhesively bond the composite resin to the tooth structure and thus achieve a good marginal adaptation. Instead of the base material acting as a stress breaker, it is proposed that the hybrid layer, the interdiffusion zone between bonding system and demineralized dentin, may act as well as an elastic buffer zone, thus compensating for thermal or mechanical stresses (Fusayama, 1990; Kemp-Scholte & Davidson, 1990; Haller, 1994).

The total bonding method has been reported to result in good marginal adaptation in class 3, 4, 5, and erosion lesions. For class 5 restorations, Krejci and Lutz (1991) and Krejci, Kuster, and Lutz (1993) demonstrated that the total bonding method gave the best marginal integrity and least microleakage. Baillod, Krejci, and Lutz (1994) confirmed these findings for class 3 and 4 composite restorations. By using one dentin adhesive system of the third generation and a fine hybrid composite, adhesive restorations in class 3 and 4 cavities were successfully placed with and without base material. Totally bonded restorations showed marginal integrity that was equal to that of selectively bonded restorations.

Only a few in vitro studies have reported on the influence of total bonding in class 2 restorations. Fusayama (1993) reported that a follow-up of posterior composite restorations placed according to the total bonding method involving simultaneous etching of enamel and dentin revealed excellent durability of the restorations. Blaser (1993) investigated the influence of total bonding and selective bonding in class 2 cavities using one bonding system and composite "base" build-ups to reduce the composite volume. He reported that restorations with total bonding to dentin had slightly lower percentages of marginal integrity than the selectively bonded restorations, especially following TCML (thermocycling and mechanical loading).

Total bonding seems to be of interest as an alternative operative procedure for class 2 composite restorations in order to reduce marginal leakage. Therefore, the purpose of the present in vitro study

was to compare the marginal integrity of totally bonded and selectively bonded class 2 composite restorations using five different bonding systems and corresponding composite resins.

METHODS AND MATERIALS

Sixty extracted human molars, which had been stored in 0.1 % thymol solution, were cleaned with pumice, mounted in acrylic resin (Paladur, Kulzer, Wehrheim, Germany) and stored in 0.9 % physiological saline solution 10 days prior to cavity preparation and restoration. Box-shaped, approximo-occlusal class 2 cavities (two surfaces) with rounded inner angles were prepared (Figure 1, A), using diamond burs and sufficient water cooling. The approximate dimensions of the cavities were: occlusal width—2 - 3 mm; occlusal depth—2 mm; width of approximal box—3 mm; height of approximal box—4 - 5 mm; and depth at gingival floor—1.5 - 2 mm. The cervical margin of the cavities was located approximately 1.0 mm below the cementoenamel junction.

The design of the investigation is summarized in Table 1. Five different composite resins and corresponding

Table 1. Investigation Design

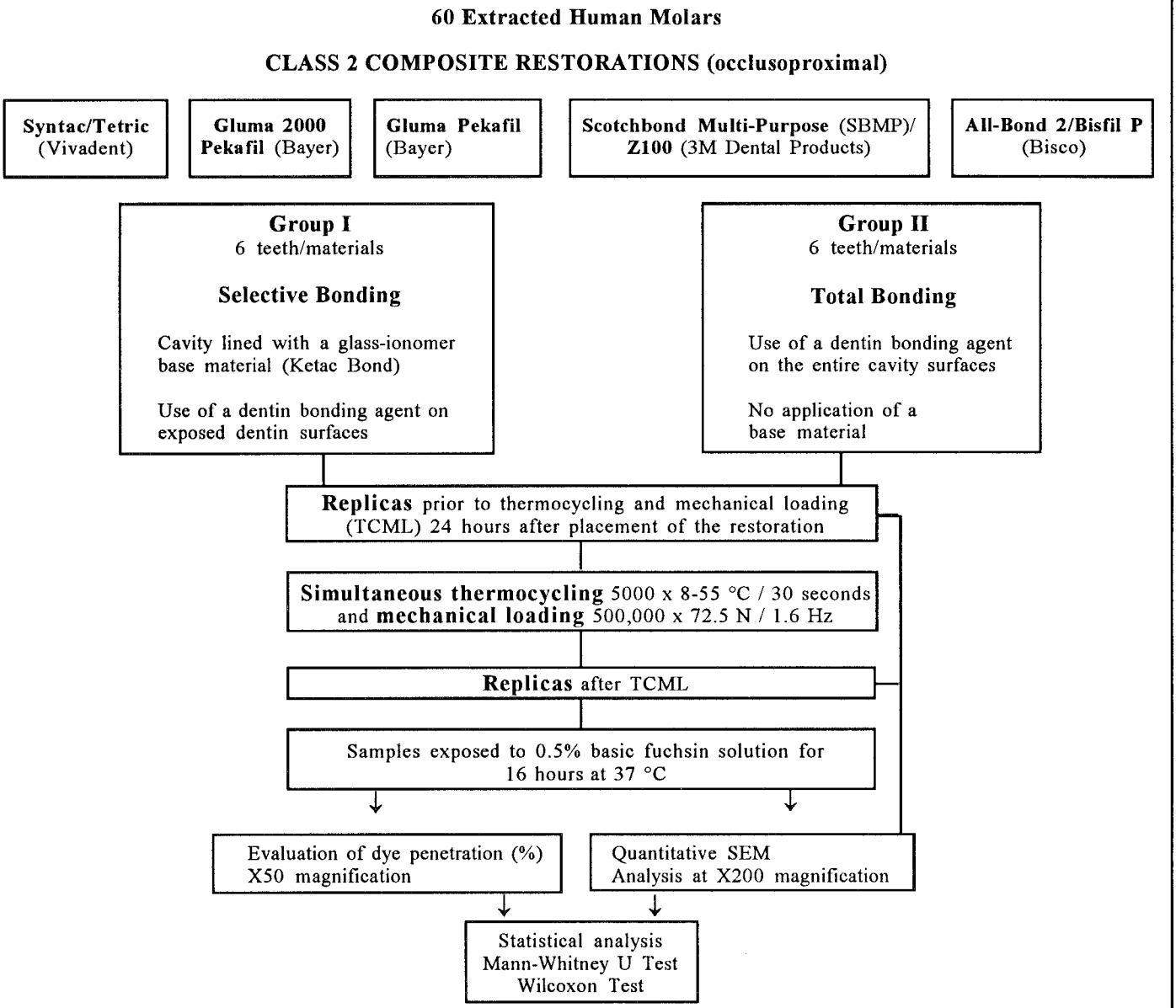

Table 2. Materials

Product	Type	Batch #	Manufacturer
Email preparator	Etchant		Vivadent
Syntac	Primer	4401731	Schaan,
	Adhesive	340274	Liechtenstein
Heliobond	Bonding Agent	261016	
Tetric Universal	Composite	460537	
Gluma 2000	Conditioner	1488W	Bayer Dental
	Adhesive Resin	1592W	Leverkusen,
Pekafill	Composite	1846K	Germany
Gluma	Etchant	1184H	Bayer Dental
	Cleanser	1107J	
	Primer	2086H	
	Sealer	1185H	
Pekafill	Composite	1146K	
Scotchbond Multi-Purpose	Etchant	7020100396-2	3M Dental Products St Paul, MN 55144
	Primer	7020100396-2	
	Adhesive	7020100396-2	
Z100	Composite	19930106	
All-Bond 2	All-Etch	099282	Bisco Dental
	Primer A	089312	Itasca, IL 60143
	Primer B	109162	
	Dentin/Enamel Bonding Resin	099212	
Bisfil P	Composite	059152	
Ketac Bond	Glass-Ionomer Base Material	00312018	ESPE Dental Seefeld/Oberbay, Germany

bonding systems (Table 2) were used to restore the cavities: Syntac/Tetric, Gluma 2000/Pekafill, Gluma/Pekafill, Scotchbond Multi-Purpose/Z100, and All-Bond 2/Bisfil P. For simplicity, the bonding system/composite combinations will be referred to by the bonding system.

Table 3 summarizes the operative procedures used. For each material, six teeth were restored according to the selective bonding method and the total bonding method. The selective bonding method included placement of a glass-ionomer base (Ketac Bond) covering the pulpal floor and axial wall (Figure 1, B) and selective application of the bonding system to the remaining, uncovered dentin along the cavity walls, the gingival floor (inclusive of eventual cement structures), and enamel surfaces. With the total bonding method, the bonding system was applied to the entire internal cavity surface after etching; no base material was placed. The manufacturers' instructions for each material were strictly followed. For all restorations, the incremental placement technique (three interproximal increments, two occlusal increments) (Figure 1, C) and the three-sited light-curing technique were applied (Lutz, Krejci & Oldenburg, 1986).

Following the finishing of the restorations with finishing diamonds and aluminum-oxide disks (Sof-Lex disks, 3M Dental Products), the samples were stored in physiological saline solution for 24 hours and then submitted simultaneously to thermocycling (8°C/55°C; 5000 cycles, 60 seconds each cycle) and mechanical loading (72.5 N, 500,000 loads, 1.6 Hz) (Figure 2). In the testing apparatus used for the investigation, six samples at a time can be thermocycled and loaded simultaneously. The sample holder moved from the 55°C water bath to the 8°C water bath, and at the same time the load was applied to the samples axially via a punch that rested on the occlusal surfaces of the teeth. Quantitative SEM analysis and dye penetration were evaluated in order to determine the marginal integrity at the surface and within the depth of the cavity.

SEM Analysis

Qualitative SEM analysis was performed on replicas of the samples. Vinyl polysiloxane impressions (Permagum Garant Light/Putty, ESPE) were taken of the samples before and after thermocycling/mechanical loading (TCML), and replicas (Araldit epoxy resin, Ciba-Geigy, Wehr, Germany)

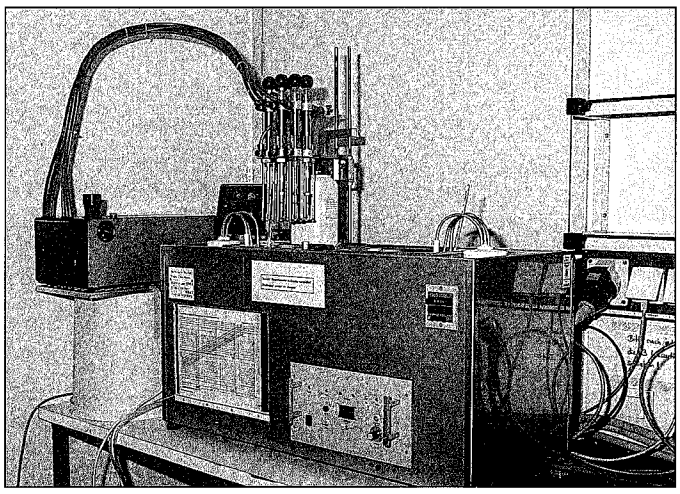

Figure 2. Testing apparatus

*Table 3. Operative Procedures: Selective Bonding**

Dentin Adhesive System	Base Material	Pretreatment of Enamel	Pretreatment of Dentin	Cavity Restoration
Syntac	The occlusal cavity floor and the pulpoaxial wall of all selectively bonded cavities are covered with a glass-ionomer base material (Table 2).	Email preparator (30 seconds), water spray, gentle blow drying	Syntac Primer (application + 15 seconds), gentle blow drying	Heliobond, gentle blow drying, light curing, placement of corresponding composite resin (Table 2)**
Gluma 2000		Gluma 2000 Conditioner (application + 20 seconds), water spray, gentle blow drying Gluma 2000 Adhesive (30 seconds), gentle blow drying		Placement of corresponding composite resin (Table 2)**
Gluma		Gluma Etchant (40 seconds), water spray, gentle blow drying	Gluma Cleanser (30 seconds), mechanical cleansing of dentin, water spray, gentle blow drying (10 seconds) Gluma Primer (30 seconds), gentle blow drying	Gluma Sealer, gentle blow drying, placement of corresponding composite resin (Table 2)**
Scotchbond Multi-Purpose		Etchant (application + 15 seconds), water spray, blot drying Primer (application + 15 seconds), gentle blow drying Adhesive, gentle blow drying, light curing (10 seconds)		Placement of corresponding composite resin (Table 2)**
All-Bond 2		All-Etch (application + 10 seconds), water spray, blot drying Primer (A and B, 1:1, five coats), gentle blow drying, bonding resin, light curing (20 seconds)		Placement of corresponding composite resin (Table 2)**

*Total bonding involves the same pretreatment procedures for enamel and dentin as selective bonding except that the entire internal enamel and dentin surfaces are pretreated and no base material is placed.
** The composite resin restorations were placed using the incremental technique and three-sited light curing.

were fabricated for SEM analysis. Marginal adaptation at the cervical margin was evaluated by quantitative SEM analysis using an image analyzing system (Kontron Bildanalyse, Eching, Germany) at X200 magnification on the Araldit replica. The entire length of the cervical cavosurface margin within the dentin was measured (100%); simultaneously, the marginal quality of the tooth/restoration interface was assessed and matching criteria were assigned to the corresponding lengths. The distribution of the criteria in percent (%) in relation to the entire cervical margin was calculated following the guidelines for quantitative SEM analysis as described by Roulet and others (1989). Morphologically, the following criteria were used to describe the marginal quality of the cavosurface dentin-restoration-interface: **GF** (gap formation: clearly visible loss of adhesion) (Figure 3A); **ME** (marginal expansion,

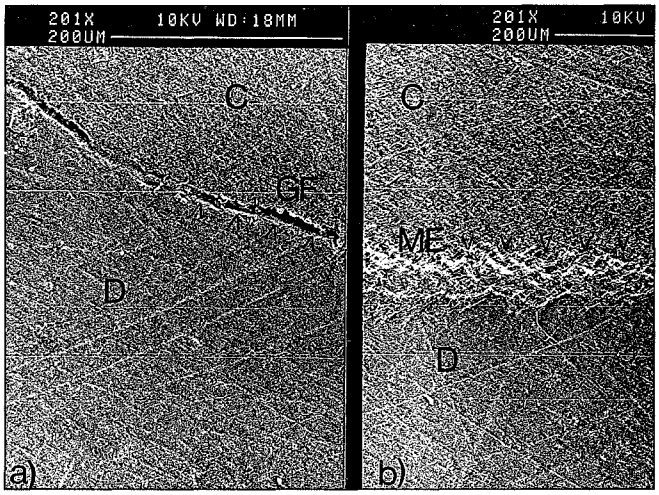

Figure 3. **A.** *Gap formation (GF) at the cervical tooth/restoration interface* **B.** *Marginal expansion (ME) at the cervical tooth/ restoration interface; (D = dentin; C = composite; original magnification X126.63).*

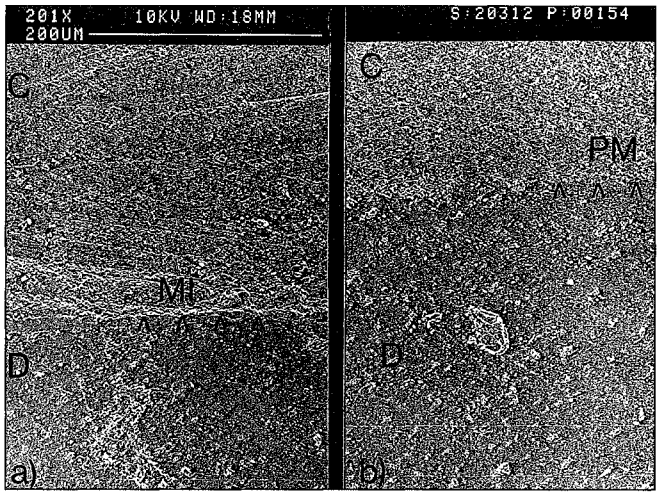

Figure 4. **A.** *Marginal imperfections (MI) at the cervical tooth/ restoration interface* **B.** *Perfect margin (PM) at the cervical tooth/ restoration interface; (D = dentin; C = composite; original magnification X126.63).*

morphological changes at the cervical restoration interface due to hygroscopic expansion of the composite) (Figure 3B); **MI** (marginal imperfections: no gap, but marginal imperfections—excess composite, positive or negative ledges) (Figure 4A); and **PM** (perfect margin, perfect adaptation between dentin and composite restoration) (Figure 4B).

Marginal expansion due to hygroscopic expansion of the composite resin was observed at the cervical margins of composite restorations below the CEJ (Kemp-Scholte & Davidson, 1989; Kunzelmann & others, 1993; Prati & others, 1994; Thonemann & others, 1997), and it may indicate an initial sign of insufficient marginal integrity, thus eventually adding to the deleterious effects. The results reported in the present study refer to the criterion "% gap formation," as a loss of adhesion that may eventually cause clinical failure of a restoration.

Dye Penetration

Microleakage at the cervical margin was determined by dye penetration. Dye penetration was performed on the original samples and after TCML only. Following TCML, all tooth surfaces were covered with nail varnish, except for the area approximately 1.0 mm away from the restoration margins. The samples were placed in a 0.5% basic fuchsin solution for 16 hours at 37°C. The teeth were sectioned 10-12 times (thickness of section approximately 200 μm) in the mesiodistal direction with a rotating diamond saw (Sägemikrotom, Leitz, Wetzlar, Germany), rendering approximately 20 sites (two sites/section) for evaluation per tooth. Thus the yield of the leakage test was increased (Gwinnett & others, 1995; Roulet, 1994). The extent of dye penetration site was expressed as a percentage of the penetration along the cervical cavity margin into the depth of the cavity (Figure 5). Each experimental group was comprised of six teeth/ material combinations. In order to create a characteristic descriptive value of dye penetration from the measurements obtained at the 20 sites/tooth, the median of all dye penetration measurements obtained at the cervical interface of a tooth was calculated. The medians for dye penetration of the six teeth per

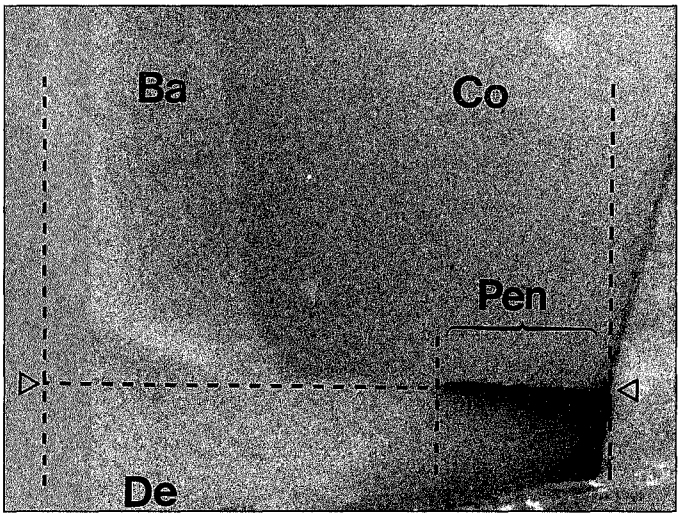

Figure 5. Tooth section of selectively bonded restoration with dye penetration. De = dentin, Co = composite; Ba = base material. Dotted line at base of restoration and arrows indicate possible path of dye penetration (100%); bracket indicates actual dye penetration (Pen) of 30%.

experimental group were used for statistical analysis.

Statistical Analysis

Medians and 25 and 75% quartiles were determined from the data obtained from quantitative SEM analysis (GF%) and evaluation of dye penetration (% dye penetration). Application of the Shapiro-Wilks test for normal distribution of the data and the Levene test for homogeneity of variance indicated that the data were not normally distributed and homogeneity of variance was not given. Thus statistical analysis was performed using nonparametric tests for pairwise comparisons at the 0.05 level of significance (α). The Mann-Whitney test was performed with the SEM and dye penetration data to determine statistically significant differences with respect to the variable "restoration method." With respect to the variable "time" that included repeated measurements on the same samples in the SEM, the Wilcoxon test was applied. In order to assess the influence of "restoration method" in general, the level of significance was adjusted to $\alpha^*(k)=1-(1-\alpha)^{1/k}$, where k = the number of performed pairwise tests, by application of the error rates method (Miller, 1981), which is a Bonferroni-type correction.

RESULTS

SEM Analysis

The results of the SEM evaluation referring to the criterion "gap formation" are summarized in Figure 6.

Selective Bonding

With the application of the selective bonding method, Gluma 2000 revealed the least percentage of gap formation (GF) before TCML (0.4% GF), as well as after TCML (3.8% GF), compared to all other bonding system/composite combinations used. All-Bond 2 revealed the highest percentage of gap formation with 12.4% GF before TCML and 28% GF after TCML. TCML significantly increased the loss of adhesion for all materials tested ($P \leq 0.05$).

Total Bonding

With the application of the total bonding method, Gluma 2000 revealed the least extent of gap formation, ranging from no gap formation before TCML to 1.9% GF after TCML. Before TCML, Syntac, Gluma, Scotchbond Multi-Purpose, and All-Bond 2 show loss of marginal integrity (GF) ranging from 1.3% - 10.7% GF. TCML significantly increased the loss of adhesion for all materials tested ($P \leq 0.05$).

Selective Bonding vs Total Bonding

The adjusted α^* for the comparison of the selective bonding vs the total bonding method was $\alpha^* = 0.0051$ for k = 10, indicating a statistically significant difference between the two restoration methods in general with respect to gap formation.

In the pairwise comparisons at the 0.05 level of significance (α), no significant differences between the selective bonding and the total bonding method were found for the bonding system/composite combinations Syntac/Tetric, Gluma 2000/Pekafill, and Gluma/Pekafill, before or after TCML. A significant difference between the selective bonding and the total bonding method was found for the bonding system/composite combinations Scotchbond Multi-Purpose/Z100 and All-Bond 2/Biscofil P before and after TCML. For Scotchbond Multi-Purpose, total bonding resulted in 1.3% GF before

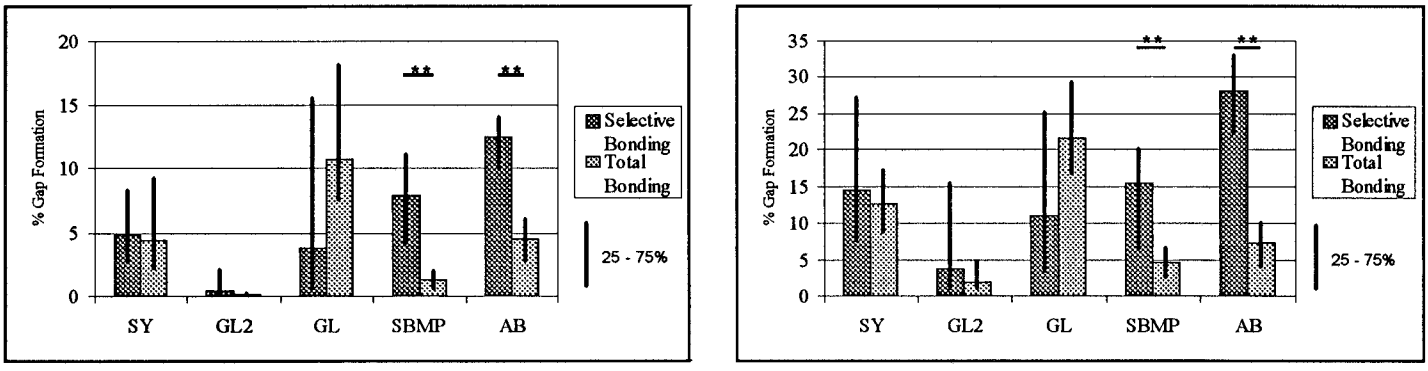

Figure 6. *Quantitative SEM analysis: selective vs total bonding before and after TCML; percentage of gap formation (GF) at the cervical tooth/restoration interface. Median, 25% and 75% quartiles; number of specimens/group = 6; SY = Syntac; GL2 = Gluma 2000; GL = Gluma; SBMP = Scotchbond Multi-Purpose; AB = All-Bond 2.* **P < 0.01; *There is a statistically significant difference between gap formation before and after TCML for all bonding agent/composite combinations with both restoration methods.*

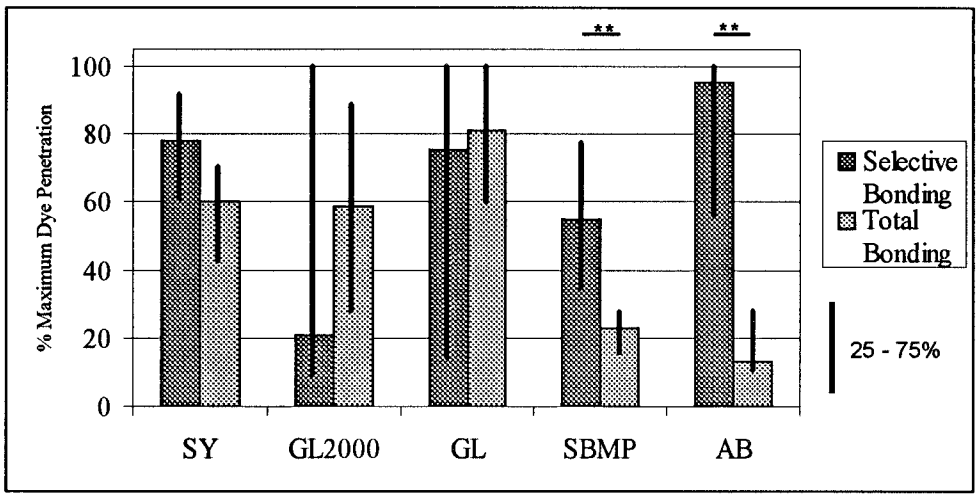

*Figure 7. Microleakage: maximum dye penetration recorded with selective bonding and total bonding at the cervical tooth/restoration interface after TCML. Median, 25%, and 75% quartiles; number of specimens/group = 6; SY = Syntac; GL2 = Gluma 2000; GL = Gluma; SBMP = Scotchbond Multi-Purpose; AB = All-Bond 2. **$P \leq 0.01$.*

TCML and 4.6% GF after TCML, which was significantly better ($P \leq 0.01$) than the results for the selective bonding method with 7.9% GF before TCML and 15.5% GF after TCML. For All-Bond 2, total bonding revealed significantly less ($P \leq 0.01$) gap formation before TCML (4.5%) and after TCML (7.3%) than the selective bonding method with 12.4% GF before TCML and 28% GF after TCML.

Dye Penetration

The results of the evaluation of dye penetration are summarized in Figure 7. Dye penetration was evaluated *after* simultaneous TCML only, as the method required sectioning of the samples to evaluate dye penetration.

Selective Bonding

Gluma 2000 exhibited the least dye penetration of 21%, whereas the highest degree of dye penetration (95.2%) was found with All-Bond 2. For Syntac, Gluma, and Scotchbond Multi-Purpose dye penetration ranged between 55%-78%.

Total Bonding

Syntac, Gluma, and Gluma 2000 showed dye penetration ranging from 59%-81%. With All-Bond 2 (23%) and Scotchbond Multi-Purpose (13%), less dye penetration was recorded than with Syntac, Gluma, and Gluma 2000.

Selective Bonding vs Total Bonding

The adjusted α^* for the comparison of the selective bonding vs the total bonding method was $\alpha^* = 0.01$ for $k = 5$, which indicated a statistically significant difference between the two restoration methods with respect to dye penetration.

In the pairwise comparisons, no statistically significant differences could be detected between the selective and the total bonding technique for Syntac and Gluma. With Gluma 2000 the total bonding technique (58.8%) exhibited more dye penetration than the selective bonding technique (20%). Scotchbond Multi-Purpose and All-Bond 2 revealed significantly less ($P \leq 0.01$) dye penetration when the total bonding technique (SBMP: 23%; AB2: 13%) was applied instead of the selective bonding technique (SBMP: 55%; AB2: 95%).

DISCUSSION

The purpose of the present in vitro study was to evaluate the marginal integrity of totally and selectively bonded class 2 composite resin restorations at cervical margins within cementum/dentin to determine whether the total bonding method, which has been successfully applied in class 3, 4, and 5 restorations (Baillod & others, 1994; Krejci & Lutz, 1991), was an acceptable alternative operative procedure to the selective bonding method used in class 2 restorations.

The conventional selective bonding method seemed to meet all requirements necessary to obtain a superior marginal quality: reduction of the overall composite volume due to the base build-up to reduce polymerization stresses (Feilzer & others, 1987) that might weaken the dentin bond, and protection of the pulp against adverse chemical and thermal effects (Baillod & others, 1994). On the other hand, placement of a base material reduced the overall bonding area, for no adhesion was established between base material and composite resin. Fusayama (1990) reported that placement of a lining cement increased the chance of marginal leakage.

With total bonding it was anticipated that due to the increase of the overall composite volume and the resulting unfavorable configuration factor of class 2 cavities, the cervical bond would not adequately resist polymerization shrinkage forces and would, therefore, fail. For one bonding system/composite combination Blaser (1993) reported that restorations

with total bonding to dentin revealed slightly lower percentages of perfect margins than selectively bonded restorations. Furthermore, biocompatibility of bonding systems is controversial. Clinically there are concerns about the compatibility of a bonding system placed in close vicinity to the pulp (Cox, 1992). However, the resinous layer resulting from simultaneous etching of enamel and dentin and total bonding without the placement of a base has been claimed to be a reliable protective liner (Fusayama, 1990).

The five bonding system/composite combinations investigated in the present study did not represent a random sample. They were selected because they were of interest themselves, e g, the number of steps needed for the application or whether the total etch technique or the wet bonding technique was applied. Thus, the results pertain only to the bonding used in this study.

In the literature it is reported, that if the restoration-tooth bond fails, it is most likely to fail at the cervical margin, for it is the weakest link compared to the bonding system/composite bond or composite/enamel bond (Haller, 1994; Pashley, Ciucchi & Sano, 1994). Polymerization shrinkage stresses induced by composite resin, insufficient penetration of the bonding agent into the demineralized dentin, or collapse of the collagen structure due to unintentional desiccation may be the reasons (Haller, 1994; Pashley & others, 1994). It should be taken into consideration, as well, that there are material interactions between the composites and corresponding adhesives, which may account for the occurrence of the marginal defects in terms of gap formation. In addition, photopolymerizing hybrid composite resins develop higher polymerization shrinkage stresses than microfilled composite resins (Feilzer, de Gee & Davidson, 1990; Haller, 1994).

In the present study SEM analysis revealed that total bonding significantly reduced gap formation when the bonding systems All-Bond 2 and Scotchbond Multi-Purpose were used along with their corresponding composite resins. The evaluation of microleakage by means of dye penetration confirmed the findings of quantitative SEM analysis regarding the superiority of Scotchbond Multi-Purpose and All-Bond 2 when used in combination with the total bonding method. The two materials involved a reduction of pretreatment procedures by applying the total-etch technique along with total bonding (Table 3), which may account for the superior results. Furthermore, the operative procedures for Scotchbond Multi-Purpose and All-Bond 2 omit air drying the tooth structure, favoring instead to "blot dry" the dentin. It is reported in the literature that All-Bond 2 and Scotchbond Multi-Purpose bond well to the dentin substrate in the presence of moisture, which may be explained by the properties of the acetone-water interactions (Kanca, 1992a,b). In the presence of moisture the primer can more readily wet the dentin surface and penetrate into the tubules and demineralized dentin (Kanca, 1992b). A collapse of the collagen structure is thus prevented. Along with the simultaneous etching procedures of enamel and dentin, this may account for the rapid development of a resin-impregnated layer over the entire internal dentin area that is able to resist polymerization shrinkage forces originating from the curing of the composite. With selective bonding, the bonding area to dentin compared to the bonding area to enamel may just be too small to resist polymerization shrinkage forces. Gap formation develops at the cervical cementum-dentin margins; whereas the enamel bond remains intact.

The combination Gluma 2000/Pekafil revealed the least gap formation with both restorative methods. With Gluma 2000 the operative procedures have been reduced to two steps. The SEM data indicated a good marginal seal when evaluating the surface of the tooth/restoration interface. However, the results of dye penetration were not in accordance with the SEM results: They revealed a loss of adhesion within the depth of the cavity when Gluma 2000 was used with the total bonding method. With the selective bonding method, the loss of adhesion within the depth of the cavity was not quite as severe, but the standard deviations varied considerably. The data indicated that the bond established with the Gluma 2000/Pekafil combination may not be as resistible to hydrolytic degradation (Prati & others, 1994) as suggested by the SEM data, especially with the total bonding method, where the entire internal dentin area was exposed. Insufficient demineralization of the dentin, insufficient penetration of the adhesive, as well as interactions with the composite resin may be the reason (Haller, 1994; Pashley & others, 1994).

Syntac showed comparable results with both restoration methods, whereas the Gluma/Pekafill combination resulted in better marginal adaptation with the selective bonding method. As with Gluma 2000, the dye penetration data revealed a loss of adhesion within the depth of the cavity. This increase in loss of adhesion after TCML was in accordance with the gap formation data determined by SEM analysis.

In vitro studies can only simulate the clinical conditions as closely as possible. Identical simulation cannot be performed and pulpal considerations cannot be taken into account. Questions regarding the durability of the composite-dentin bond and the relationship between marginal adaptation in vitro and bacterial contamination and development of secondary caries in vivo require further study (Haller, 1994).

The results of the present study indicated that total bonding may be an acceptable alternative to selective bonding, depending upon the bonding

system used. Further clinical study is needed to prove the effectiveness of total bonding in class 2 adhesive restorations.

CONCLUSIONS

SEM evaluation and dye penetration showed that out of five bonding system/composite combinations tested, two combinations (All-Bond 2, Scotchbond Multi-Purpose) resulted in significantly better cervical marginal adaptation with the total bonding method than with the selective bonding method.

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Water Storage Effect on the Marginal Seal of Resin-modified Glass-Ionomer Restorations

M IRIE • K SUZUKI

Clinical Relevance
Polishing glass-ionomer restorations at the second appointment appears to be most appropriate, since the marginal gap is significantly reduced after 1 day in water storage for both of the two glass-ionomer materials used in this study.

SUMMARY

The effect of storage in water on the marginal adaptation of glass-ionomer restorations placed in extracted human premolars and Teflon molds was studied. When the cavity preparations were filled with a glass ionomer and polished immediately after setting, a marginal gap of approximately 15 μm was formed. In contrast, when polishing following storage in water the marginal gap was 0 to 2 μm for the resin-modified and the conventional glass ionomers respectively. The bond strength of the glass ionomer to enamel and to dentin and the flexural strength of the glass ionomer increased after storage in water. Therefore the marginal seal of glass-ionomer restorations may not only be improved due to hygroscopic expansion during storage in water but also may exhibit greater bond strength.

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INTRODUCTION

Resin-modified glass ionomers are used not only as base/liner materials but also as filling materials (Mitra, 1991; Crispin, 1994; Croll & Helpin, 1995). However, in dental clinics the clinical procedure of filling a cavity with a resin-modified glass ionomer is often improperly performed due to insufficient understanding of the properties of the material. For example, little attention has been paid to the marginal gap between the glass ionomer and cavity preparation, even though it is known that a resin-modified glass-ionomer filling material produces a greater marginal gap, measured immediately after light activation, in cavity preparations than does a resin composite (Irie

& Nakai, 1993; Sidhu, 1994). Clinicians often complete a restorative procedure at one appointment out of consideration for the patient. However, the resulting potential for damage to the pulp should not be overlooked. For example, it was reported that early large gaps exposing the freshly cut dentin could facilitate bacterial penetration and pulpal damage (Brännström, Torstenson & Nordenvall, 1984; Torstenson & Brännström, 1988). Also, it was reported that many restoration failures occur in the earliest stage of restoration (Prati, 1989). Therefore, when conventional glass ionomers are used for clinical restorations, it may not be advisable to complete the restorations at one appointment (Irie & Nakai, 1987).

When a filling material is placed in the oral cavity, hygroscopic expansion of the material can be expected to compensate for marginal gap formation when either resin composite (Asmussen & Jörgensen, 1972; Hansen & Asmussen, 1990) or conventional glass ionomer (Irie & Nakai, 1987) is used. Although the setting reaction of the resin-modified glass ionomer is not the same as that of conventional glass ionomer, it is likely that some degree of hygroscopic expansion also occurs in the resin-modified glass ionomer, thus contributing to reduction of the marginal gap between the cement and cavity preparation (Sidhu, Sherriff & Watson, 1997). The resin-modified glass ionomer, unlike a light-cured resin composite, has a dual setting reaction consisting of an acid-base reaction and a photochemical polymerization process (Wilson, 1990; Mitra, 1991).

Flexural testing has previously been carried out to evaluate the mechanical strength of brittle restorative materials (Momoi & others, 1995; Irie & Nakai, 1995). The relationship between shear bond strength of silver-added glass ionomers and flexural strength was found to be statistically significant (Irie & Nakai, 1988).

In this study, therefore, the effect of storage in water on the marginal gap formation using two glass ionomers, a resin-modified glass ionomer and a conventional glass ionomer, was evaluated. The bond and flexural strengths of the glass-ionomer materials were examined to determine the influence that marginal gap formation had on retention strengths of the restorations.

METHODS AND MATERIALS

The basic properties of the two glass ionomers used in this study are summarized in Table 1. The resin-modified glass ionomer (Fuji II LC; GC Corp, Tokyo, Japan) was compared with the conventional glass ionomer (Fuji II; GC Corp), which was used as the control. All procedures were accomplished in accordance with the manufacturer's instructions. Human premolars, extracted for orthodontic reasons, were used for this project. After extraction, the teeth were immediately stored in cold, distilled water at about 4 °C for 1 to 2 months before testing. Ten specimen restorations were produced using each restorative material.

Each premolar was embedded in slow-setting epoxy resin (Epofix Resin; Struers, Copenhagen, Denmark). A flat surface of enamel was obtained by grinding the tooth with wet silicon carbide paper (#220). Then a cylindrical cavity was prepared to a depth of approximately 1.5 mm with a diameter of 3.5 mm with a tungsten carbide bur (200,000 rpm) and a fissure bur (8,000 rpm) using water spray. One cavity preparation was prepared in each tooth on the mesial surface of the crown. In total, mesial cavity preparations in 60 teeth were prepared for this study. Conditioning the surfaces of the preparations was not done, because bonding was expected without conditioning. Each cavity preparation was filled with glass-ionomer material using a syringe tip (Centrix C-R Syringe System; Centrix, Shelton, CT 06484), then covered with a plastic strip until set. The Fuji II LC was exposed to a visible light source (Luxor, Model 4000; ICI, Cheshire, England) with irradiation time of 20 seconds. The Fuji II was stored in an incubator at 37 °C and 100% relative humidity for 4 minutes after mixing. Surfaces were polished immediately after light activation or setting, or after storage in distilled water at 37 °C for 1 day or for 1 week. The excess filling material was removed by wet grinding with silicon carbide paper (#1000), followed by polishing using an aqueous slurry of 0.3μm aluminum oxide (Alfa Micropolish; Buehler Ltd, Lake Bluff, IL 60044) and thorough rinsing with distilled water. Each restoration margin was inspected under a light microscope (X1000, Measurescope, MM-11; Nikon, Tokyo, Japan). The maximum gap width between the material and the cavity wall was measured by using an optical microscope, as previously described (Munksgaard, Irie & Asmussen, 1985).

To estimate the degree of setting shrinkage and hygroscopic expansion and to compare the marginal gap width in restorations placed in the teeth, a Teflon

Table 1. Materials Used

Material	Type	Batch #	Powder/Liquid
Fuji II LC	resin-modified	P: 061111 L: 071011	3.0g/1.0g
Fuji II	conventional	P: 300802 L: 120901	2.7g/1.0g

Table 2. Marginal Gap in Tooth Cavity

Time Passed	Mean (μm)				Alpha Value
	Fuji II LC		Fuji II		
Immediately after light activation or setting	14.1 (0)	(10-20)*	14.8 (0)	(11-19)*	NS
1 day	0 (10)	(0)*	1.8 (5)	(0-5)*	NS
1 week	0 (10)	(0)*	1.2 (7)	(0-5)*	NS

Number of specimens = 10; ( ) = number of specimens with no gaps; ( )* = range of gap width; NS = not significantly different by Mann-Whitney *U* test ($\alpha > 0.05$). Values joined by vertical lines were not significantly different by Duncan's new multiple range test (Conover & Iman, 1981) ($P > 0.05$).

mold of the same diameter and depth was prepared. The prepared Teflon mold was placed on a silicone oil-coated glass plate, because a glass plate would not react or bond to the filling material. The degree of the setting shrinkage (immediately after set) and the hygroscopic expansion (after storage in water for 1 day or 1 week) was determined.

Bond strengths to enamel and to dentin were measured to evaluate the bonding effect between the filling material and the cavity preparations. Specimens were human premolars embedded in slow-setting epoxy resin (Epofix Resin; Struers). Flat surfaces of enamel and dentin were obtained by grinding the teeth with wet silicon carbide paper (#1000). The enamel and dentin surfaces were not conditioned. Each material was placed into a Teflon mold (3.6 mm in diameter, 2.0 mm in height) set on the enamel or dentinal surface, and hardened as described above. The specimens thus obtained were then mounted on a testing machine (Autograph, DCS-2000; Shimadzu, Kyoto, Japan), and shear stress was applied at a crosshead speed of 0.5 mm/minute. In this experiment as well, measurements were carried out under three different conditions: (1) immediately after the setting procedure, and after the specimen was kept in distilled water at 37 °C; (2) after 1 day or (3) after 1 week. After the shear bond strength measurements, all the failed specimens were analyzed utilizing a light microscope (X4) (SMZ-10; Nikon) to determine the nature of fractures.

Teflon molds (15 mm x 4 mm x 2.5 mm) were used as the specimens for determining the flexural strength measurements. Fuji II LC was cured in three overlapping sections, each cured for 30 seconds. Fuji II specimens were stored at 37 °C and 100% relative humidity for 4 minutes after mixing. The flexural strength was measured using the three-point bending method with a 10 mm span and load speed of 0.5 mm/minute. Flexure strengths were determined for restorations using both materials under the same three conditions described for marginal gap determination, i e, immediately after the setting procedure, after storage of the specimen in distilled water at 37 °C for 1 day, or 1 week.

All procedures, except for cavity preparation and mechanical testing, were performed in a thermohygrostatic room kept at 23 ± 0.5 °C and 50 ± 2% relative humidity. The results were analyzed statistically using the Mann-Whitney *U* test, Duncan's new multiple-range test (Conover & Iman, 1981), or *t*-test.

RESULTS

Table 2 summarizes the data for the marginal gap observed in the tooth restorations under various conditions. When the specimens were polished immediately after the setting procedure, a marginal gap of approximately 15 μm was observed regardless of the type of glass ionomer. In contrast, gaps of 0 to 2 μm were observed when the specimens were polished after storage in water for 1 day (Fuji II LC) or 1 week (Fuji II) respectively. No statistical

Table 3. Marginal Gap in Teflon Cavity

Time Passed	Mean (μm)		*P* Value*
	Fuji II LC	Fuji II	
Immediately after light activation or setting	29.1 ± 4.0	35.1 ± 6.6	< 0.05
1 day	6.2 ± 1.3	9.3 ± 1.4	< 0.05
1 week	6.9 ± 1.7	7.1 ± 1.5	NS

Number of specimens = 10; * = *t*-test; NS = not significantly different ($P > 0.05$).
Values joined by vertical lines were not significantly different by Duncan's new multiple range test ($P > 0.05$).

Table 4. Shear Bond Strength to Enamel

Time Passed	Mean ± SD (MPa)		*P* Value*
	Fuji II LC	Fuji II	
Immediately after light activation or setting	2.26 ± 0.40	0.71 ± 0.23	< 0.05
1 day	3.97 ± 2.55	3.78 ± 0.63	NS
1 week	6.54 ± 5.45	3.63 ± 1.65	NS

Number of specimens = 10; * = *t*-test; NS = not significantly different ($P > 0.05$).
Values joined by vertical lines were not significantly different by Duncan's new multiple range test ($P > 0.05$).

difference was observed between the specimens stored in water for 1 day and 1 week.

Table 3 summarizes the marginal gap width between the glass ionomer and Teflon mold under various conditions. The gap width under each condition was wider than that obtained using the tooth cavity. However, an apparent correlation between the marginal gap in tooth restorations and the marginal gap in Teflon mold restorations was observed for the two products (Fuji II LC: $r = 0.9996, P < 0.05$ and Fuji II: $r = 0.9995, P < 0.05$) when analyzed by the Pearson Product-Moment Correlation.

Tables 4 and 5 summarize the shear bond strength to the enamel surface and the mode of fracture respectively. Greater bond strengths were obtained after the storage periods of 1 day and 1 week than immediately after setting. The Fuji II LC showed larger values than did the Fuji II under all conditions, although no significant difference was observed for the specimens kept in water. The mode of all fractures was adhesive or mixed adhesive/cohesive. No cohesive-only fracture surfaces were observed. The proportion of mixed fractures increased as the shear bond strength increased for Fuji II LC.

Tables 6 and 7 summarize shear bond strengths to dentin surfaces and the mode of fracture respectively. Shear bond strengths to dentin showed essentially the same pattern as that to enamel surfaces. When specimens were stored in water for 1 day or 1 week, the shear bond strengths were significantly greater than that measured immediately after setting. All fractured surfaces to dentin were cohesive or mixed fractures. The proportion of cohesive fractures increased as the value of the bond strength increased.

Table 8 summarizes the flexural strength under various conditions. When specimens were stored in water for 1 day or 1 week, significantly greater flexural strength values were observed than when the specimens were measured immediately after setting. Fuji II LC specimens exhibited greater flexural strength than the Fuji II under all conditions.

DISCUSSION

This study clearly demonstrated that polishing a glass-ionomer restoration should not be performed immediately after the filling and setting procedure but should be delayed at least 1 day. In contrast to

Table 5. Analysis of Fracture Mode Data Corresponding to Those in Table 4

Time Passed	Number with Each Fracture Mode	
	Fuji II LC	Fuji II
Immediately after light activation or setting	N = 10 (AD: 10, MF: 0, CF: 0)	N = 10 (AD: 4, MF: 6, CF: 0)
1 day	N = 10 (AD: 9, MF: 1, CF: 0)	N = 10 (AD: 4, MF: 6, CF: 0)
1 week	N = 10 (AD: 4, MF: 6, CF: 0)	N = 10 (AD: 0, MF: 10, CF: 0)

N = number of specimens; AD = adhesive fracture at bonding site; MF = mixture fracture; CF = cohesive fracture.

Table 6. Shear Bond Strength to Dentin

Time Passed	Mean ± SD (MPa)		*P* Value*
	Fuji II LC	Fuji II	
Immediately after light activation or setting	1.38 ± 0.74	0.50 ± 0.11	< 0.05
1 day	8.51 ± 2.51	4.25 ± 1.05	< 0.05
1 week	11.79 ± 4.00	3.13 ± 0.79	< 0.05

Number of specimens = 10; * = *t*-test.
Values joined by vertical lines were not significantly different by Duncan's new multiple range test ($P > 0.05$).

Table 7. Analysis of Fracture Mode Data Corresponding to Those in Table 6

Time Passed	Number with Each Fracture Mode	
	Fuji II LC	Fuji II
Immediately after light activation or setting	N = 10 (AD: 0, MF: 2, CF: 8)	N = 10 (AD: 0, MF: 10, CF: 0)
1 day	N = 10 (AD: 0, MF: 2, CF: 8)	N = 10 (AD: 0, MF: 0, CF: 10)
1 week	N = 10 (AD: 0, MF: 2, CF: 8)	N = 10 (AD: 0, MF: 0, CF: 10)

N = number of specimens; AD = adhesive fracture at bonding site; MF = mixture fracture; CF = cohesive fracture.

the marginal gap of approximately 15 μm observed when specimens were polished immediately after setting, a gap between 0 and 2 μm was determined when the restorations were polished after storage in water for 1 day or 1 week. One reason for this significant difference in marginal gap formation may be the hygroscopic expansion of the glass ionomer. Glass ionomer shrinks during the setting reaction (Feilzer, de Gee & Davidson, 1988). Therefore, a marginal gap will form if the adhesion between the tooth and glass ionomer does not compensate for the stress exerted by cement shrinkage. Water absorption is useful in reducing this stress (Feilzer & others, 1995). This effect was reported as the uptake of water by the matrix of resin-modified glass ionomers forming a poly-HEMA complex (Wilson, 1990). Also, conventional glass ionomers form a hydrogel of calcium and aluminum polyacrylates by water uptake (Wilson & McLean, 1988). Although the hygroscopic expansion may not be enough to compensate for the setting shrinkage, it plays an important role in reducing the shrinkage caused by the cement setting reaction and thus improves the marginal seal (Sidhu & others, 1997).

Marginal gaps that were measured using Teflon molds showed similar patterns when comparing polishing conditions or the type of glass ionomer to that obtained using the restorations placed in teeth. However, marginal gaps were wider than the corresponding marginal gaps obtained using natural teeth. Cement fillings in Teflon molds are not susceptible to interaction with the cavity walls due to the nonreactivity of Teflon. Marginal gaps observed even after specimens were stored in water for 1 week indicated that the hygroscopic expansion did not fully compensate for the shrinkage caused by the setting reaction. The smaller marginal gap observed in the natural teeth than in the Teflon mold clearly demonstrated that adhesion between the cement and cavity walls was an important influence on the marginal gap.

It has been reported that the bond strength of a glass ionomer is closely related to its mechanical strength (Mitra, 1991; Hinoura, Miyazaki & Onose, 1991). The shear bond strength of a silver-added glass ionomer was correlated to its flexural strength (Irie & Nakai, 1988). Therefore, flexural strength has an influence on marginal gap formation. In this experiment it was found that there were close relationships among the flexural strength of the cement, the shear bond strength to enamel and to dentin, and the fracture mode, such that the marginal gap became smaller with increasing bond strength, proportioned rate of mixed and cohesive fractures, and flexural strength of the cement. It was not unexpected that the cement showed higher bond and mechanical strengths when fully set rather than during the setting reaction. It appeared that the bonding ability to the tooth structure increased with the development of the glass ionomer/tooth marginal interaction during storage in water, and that the cohesive strength of the cement itself improved as the setting process advanced. It is reported that pH, an index of the degree of hardening reaction, of set glass ionomer is lower at the initial stage regardless of the type of cement, i e, conventional or resin-modified glass ionomers. The pH value of the set cement gradually increased for 24 hours (Tosaki & Hirota, 1994; Anusavice, 1996). Therefore it could be presumed that completion of the setting reaction of a glass ionomer required 24 hours. Thus 24 hours are required for a glass ionomer to obtain adequate mechanical strength, which is closely related to its

Table 8. Flexural Strength

Time Passed	Mean ± SD (MPa)		*P* Value*
	Fuji II LC	Fuji II	
Immediately after light activation or setting	30.45 ± 2.30	6.31 ± 0.89	< 0.05
1 day	43.39 ± 8.64	12.23 ± 0.89	< 0.05
1 week	46.77 ± 7.87	15.80 ± 4.33	< 0.05

Number of specimens = 10; * = *t*-test.
Values joined by vertical lines were not significantly different by Duncan's new multiple range test ($P > 0.05$).

bond strength. It should be noted that the Fuji II LC, a resin-modified glass ionomer, also has a minimum setting time of 24 hours even after visible light curing. The results of this investigation corroborated previous reports that the resin modification of glass-ionomer cement leads to higher bond and flexural strength to enamel and to dentin (Mitra, 1991; Hinoura & others, 1991; Irie & Nakai, 1995).

As discussed above, at least two factors, hygroscopic expansion and greater bond strength, play important roles in reducing the marginal gap at the tooth/glass ionomer interface. The hygroscopic expansion helped to compensate for the shrinkage of the cement due to setting reaction, and the bond strength to the tooth increased due to the uninterrupted progress of the setting reaction. Therefore it appeared advisable not to complete restoration polish at the placement appointment.

This study was carried out without use of a conditioning agent because it was felt that adhesion to tooth structure would occur in spite of using a tooth conditioner. However, the lack of using a tooth conditioner did not demonstrate sufficient bonding efficacy to tooth substrate. A more valid approach to an evaluation of bonding efficacy with glass ionomers requires further investigation of enhanced adhesion with conditioning. It also requires further investigation of tested results with many kinds of resin-modified glass ionomers, since only one resin-modified glass ionomer product was tested in this study.

CONCLUSIONS

The cavity preparation/insertion appointment for glass-ionomer restorations and polishing procedure should not be performed at the same appointment.

Acknowledgments

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Evaluation of Antibacterial Activity of Three Dentin Primers Using an in Vitro Tooth Model

K OHMORI • N MAEDA • A KOHNO

Clinical Relevance
Although all dentin primers showed antibacterial activity, ED primer exhibited the strongest effect among three primers on both the in vitro tooth model and disk diffusion method.

SUMMARY

This study compared the antibacterial activities of three dentin primers and investigated a newly designed experimental system using a bovine tooth model method for evaluating antibacterial activity by comparing this method with a conventional disk diffusion method.

The antibacterial activities of SA primer in Clearfil Liner Bond, LB primer in Clearfil Liner Bond II, and ED primer in Panavia 21 were evaluated using the conventional disk diffusion method. The 50 µl aliquot of each primer was applied to three sterilized paper disks, then placed onto Tryptic Soy agar plates already inoculated with *Streptococcus mutans*. After anaerobic incubation for 48 hours, the diffusion of antibacterial components was determined using the inhibition zone produced around the paper disk. The diameter of the inhibition zones was measured and the average calculated.

Standardized cavities (diameter 5.0 mm, depth 3.0 mm) were prepared on the labial surfaces of bovine teeth and inoculated with *S mutans* (10^6 CFU/µ l) following sterilization by ^{60}Co γ rays (50 KGy). The teeth were divided into four groups: SA primer, LB primer, ED primer, and a control group. Except for the control teeth, the cavity preparations were treated with the respective dentin primers, and then firmly sealed with a temporary sealing material. The teeth were placed in bottles containing melted Tryptic Soy agar. Five ml of Tryptic Soy broth was then added to the surface of the hardened Tryptic Soy agar. After 1 week's incubation of the teeth in the bottles at 37 °C, the number of bacteria remaining in each cavity was counted, except for eight specimens, which were used for SEM observation.

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The ED primer showed the widest inhibition zone in the disk diffusion test, which was significantly different from the other primers. Using the bovine tooth model, all dentin primers showed antibacterial activity, with significant differences found among the four groups. The results indicated that ED primer had the strongest antibacterial effect among the three primers.

INTRODUCTION

Recent improvements in dentin adhesion effectively enable the dentist to preserve sound dentin, while facilitating an increase in the use of composite resin. The presence of cariogenic bacteria beneath resin composite restoratives has been shown to adversely affect the pulp (Stanley, Going & Chauncey, 1975). Thus, after removal of carious dentin, it is important to eliminate any remaining bacteria in the dentinal tubules. In vivo studies (Qvist & Qvist, 1977; Bränn-ström & Nordenvall, 1978; Anderson & Charbeneau, 1985) have shown that bacteria may exist on cavity walls, in the smear layer, or at the dentinoenamel junction. Bergenholtz and others (1982) and Kaketa (1984) reported the presence of bacteria under composite resin restorations. In a comparison between the conventional visual and tactile method for detecting carious dentin and a dye-enhanced method, Kidd and others (1989) found that out of 100 cavities assessed to be caries-free using the conventional method, 57 cavities, after being stained using caries-detecting dye, indicated the presence of infected dentin at the dentinoenamel junction.

Many adhesive systems use an acid agent for etching. CA Agent, which is 10% citric acid and 20% calcium chloride, is used as the acid agent in the original Clearfil Liner Bond (Kuraray, Osaka, Japan). Settembrini and others (1997) reported that the etchants removed bacteria on the cavity walls. However, Clearfil Liner Bond II (Kuraray) and Panavia 21 (Kuraray) do not use such acid etching systems, because LB primer and ED primer contain acidic monomers and are thus self-etching primers. Consequently, when these systems are used, the cavity needs only to be air blown and not rinsed with water, which decreases clinical chair time, but may increase the chance for bacteria to survive, if they still remained in the smear layer.

If tooth conditioners such as acid agents and primers possessed antibacterial activity, these bacteria could be eliminated, thereby preventing the onset of secondary caries.

Some composite resins, primers, and bonding agents have been shown by using the disk diffusion method to be antibacterial (Updegraff, Chang & Joos, 1971; Skjørland, 1973; Takemura & others, 1984; Prati & others, 1993; Onoe, 1994; Palenik & Setcos, 1996). However, a comparison of the antibacterial activity of different materials using this method is difficult, because the diffusion rate of such materials into the dentin may vary significantly.

The purpose of this study was to compare the antibacterial activity of three dentin primers, and to investigate an in vitro tooth model method for evaluating antibacterial activity by comparing it to a conventional disk diffusion method.

METHODS AND MATERIALS

Disk Diffusion Method

The antibacterial activity of each dentin primer was evaluated using the disk diffusion method.

Table 1 shows the dentin primers used in this study. SA primer, which is from the Liner Bond system, contains mainly N-methacryloyl 5-aminosalicylic acid (5-NMSA) as a salicylic acid derivative monomer and ethanol; LB primer in Liner Bond 2 contains phosphoric ester adhesive monomer (Phenyl-P) and 5-NMSA as well as a hydrophilic monomer (HEMA); and ED primer in Panavia 21 contains methacryloxydecyl-dihydrogen phosphate (MDP), HEMA, 5-NMSA, and polymerization accelerators. LB primer and ED primer consist of two bottles (Liquid A and Liquid B), which act as both conditioner and adhesion primer, while SA primer is a one-bottle system.

Table 1. Components of Three Primers

Product	Components	Batch #
SA primer	N-methacryloyl-5-aminosalicylic acid (5-NMSA), ethanol	0023
LB primer	N-methacryloyl-5-aminosalicylic acid (5-NMSA), 2-methacryloyloxyethyl phenyl hydrogen phosphate (Phenyl-P), 2-hydroxyethyl methacrylate (HEMA)	A: 00009 B: 00013
ED primer	N-methacryloyl-5-aminosalicylic acid (5-NMSA), 10-methacryloyloxydecyl dihydrogen phosphate (MDP), 2-hydroxyethyl methacrylate (HEMA)	A: 00012 B: 00017

All primers were produced by Kuraray, Osaka, Japan.

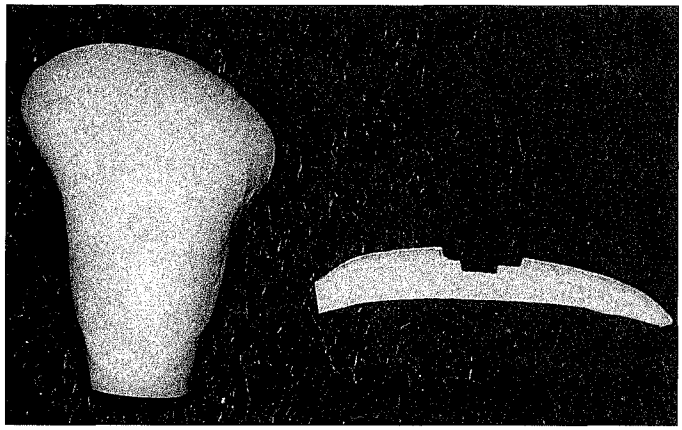

Figure 1. *Standardized cavity prepared for the experiment. Outer cavity (5 mm in diameter, 1.5 mm deep) and inner cavity (2.5 mm in diameter, 1.5 mm deep) were prepared on the labial surface of the bovine tooth.*

Streptococcus mutans ATCC25175 was grown in Tryptic Soy broth (Difco Laboratories, Labonia, MI 48152) for 24 hours, harvested by centrifugation, and resuspended with sterile phosphate-buffer (pH7.2) to an absorbance at 650 nm of 1.4 (approximately $1x10^9$ CFU/ml).

The 50 µl aliquot of each primer was applied to sterilized paper disks (8 mm in diameter, 1.5 mm thick) in triplicate. The paper disks were placed onto Tryptic Soy agar plates already plated with the 100 µl aliquot of *S mutans*. After anaerobic incubation of the plates at 37 °C for 48 hours, the diffusion of antibacterial components was determined using the inhibition zone produced around the paper disk. The diameter of the inhibition zones was measured in three locations and the average calculated.

Bovine Tooth Model

Twenty-eight extracted bovine teeth were stored at -80 °C until use. Standardized cavities (outer cavity: 5.0 mm in diameter, 1.5 mm deep; inner cavity: 2.5 mm in diameter, 1.5mm deep) were prepared on the labial surfaces of bovine teeth using a water-cooled diamond point (Figure 1). All teeth were stored in water and sterilized by ^{60}Co γ rays (50 KGy). The cavities were inoculated with 1 µl aliquot of *S mutans* following the removal of water using sterilized paper.

The teeth were divided into the following four groups (n = 7): SA Primer group (Group 1), LB Primer group (Group 2), ED Primer group (Group 3), and Control group (Group 4).

Teeth in all groups were treated with the test primers according to the manufacturers' instructions, except for the control group, which was not treated. The teeth from all four groups were firmly sealed with Fermit (Vivadent Ivoclar, Schaan, Liechtenstein) and

Figure 2. *Method of cultivation. The tooth was placed into the bottle containing melted Tryptic Soy agar. Five ml of Tryptic Soy broth was then added to the hardened agar, which was cultivated at 37 °C for 1 week.*

cured for 40 seconds using a light source (Optilux 400, Demetron Research Corp, Danbury, CT 06810).

The teeth were placed into bottles that contained melted sterilized Tryptic Soy agar (121 °C, 15 minutes). The bottles were stored in an incubator at 37 °C for 1 week following the addition of 5 ml of Tryptic Soy broth to the hardened Tryptic Soy agar (Figure 2). Following cultivation at 37 °C for 1 week, five teeth from each group were taken out of the bottle, and temporary sealings removed from the cavities, after which the teeth were put into another 10 ml of Tryptic Soy broth. To collect any *S mutans* remaining in the cavities, the teeth in the Tryptic Soy broth were treated with ultrasonic waves (Branson B-52, Branson Cleaning Equipment Company, Danbury, CT 06813) in water at 4 °C for 1 minute. After the serial 10-fold dilution of the Tryptic Soy broth that contained the remaining *S mutans* (10^0, 10^{-1}, 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5}), 100 µl of each dilution was placed on Tryptic Soy agar plates and cultivated anaerobically in an anaerobox (MIP-1025, Forma Scientific, Marietta, OH 45750) for 48 hours at 37 °C. The number of viable *S mutans* that remained in each cavity was calculated as log 10 colony forming units (CFU) per ml. The two remaining teeth in each group were used for scanning electron microscope observation. After the 1 week of incubation, the teeth were sectioned through the sagittal plane and fixed in 1N phosphate buffer containing 2.5% glutaraldehyde. Specimen blocks were fixed in 2% osmic acid-phosphate buffer for 2 hours. The specimens were then dehydrated in a graded series of ethanol (70, 80, 90,

Table 2. Diameter of Inhibition Zones in mm Produced by Each Experimental Group (Mean ± SD)

Group	Diameter
SA primer	11.6 ± 0.5
LB primer	25.7 ± 1.2
ED primer	33.6 ± 0.7

The average diameter of the inhibition zone for each sample was significantly different (ANOVA, Student-Newman-Keuls, $P < 0.05$).

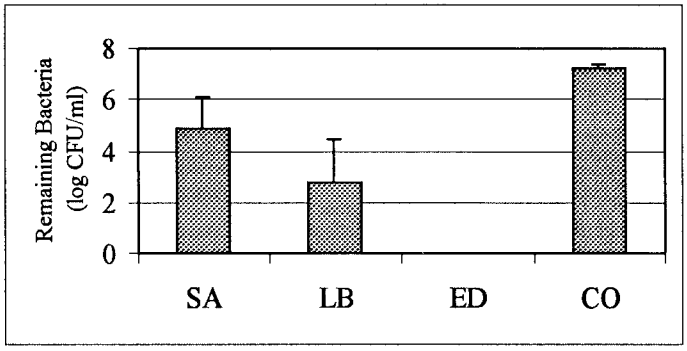

Figure 3. *Number of* S mutans *that remained in the cavity of each experimental group after the 1-week cultivation. SA = SA primer; LB = LB primer; ED = ED primer; CO = control.*

95, 100%, each for 15 minutes), followed by dehydration in t-butyl alcohol for 15 minutes, and the whole procedure was repeated. A t-butyl alcohol freeze-drying device (ID-2; Eico Engineering, Tokyo, Japan) was used to freeze-dry the specimens, which were coated with gold using the standard evaporation technique. The cavity walls were then observed under a scanning electron microscope (FE-8000; Elionix Inc, Tokyo, Japan).

The antibacterial activity of each dentin primer using the disk diffusion method was analyzed using ANOVA and the Student-Newman-Keuls test. Significance was determined at the $P < 0.05$ level. The number of bacteria remaining in each cavity was statistically analyzed using the nonparametric Mann-Whitney U test ($P < 0.05$).

RESULTS

Table 2 shows the diameter of inhibition zones in mm produced by each experimental group. All primer groups produced inhibition zones. The diameter of SA primer was 11.6±0.5 mm, LB primer's was 25.7±1.2 mm, and ED primer's was 33.6±0.7 mm. The average diameter of the inhibition zone for the samples treated with ED primer was significantly different from the samples treated with SA or LB primer (ANOVA, Student-Newman-Keuls, $P < 0.05$).

Figure 3 shows the number of *S mutans* that remained in the standardized cavity of each experimental group after 1 week of cultivation. The results were calculated by taking the log 10 of the sum of the organisms detected per milliliter plus one [log 10 (number of organisms +1)] (Conlon, Hepper & Teresa, 1977).

All of the primers exhibited antibacterial activity. The numbers of *S mutans* for the samples treated with SA and LB primer were 4.91(1.16) log CFU/ml and 2.77(1.66) log CFU/ml. *S mutans* was completely undetected in the ED primer group. There were significant differences among all primer groups, including the control group (Mann-Whitney U test, $P < 0.05$).

Figures 4 and 5 show the SEM view of the cavity wall treated with LB primer and ED primer respectively. A substantial amount of *S mutans* was observed on the cavity wall of the LB primer group. In contrast, little *S mutans* was observed on the cavity wall of the ED primer group. However, the morphological alterations of *S mutans* by the primers were not clearly visible under SEM observation.

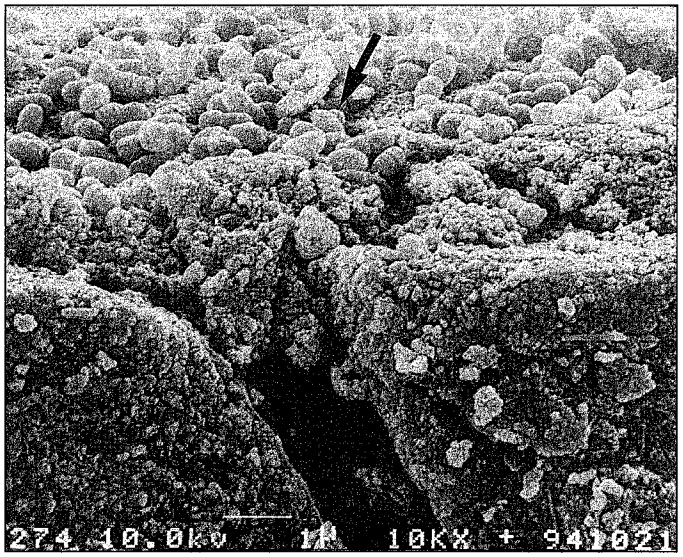

Figure 4. *SEM observation shows the cavity wall treated with LB primer (original magnification X6400). A substantial amount of* S mutans *was seen.*

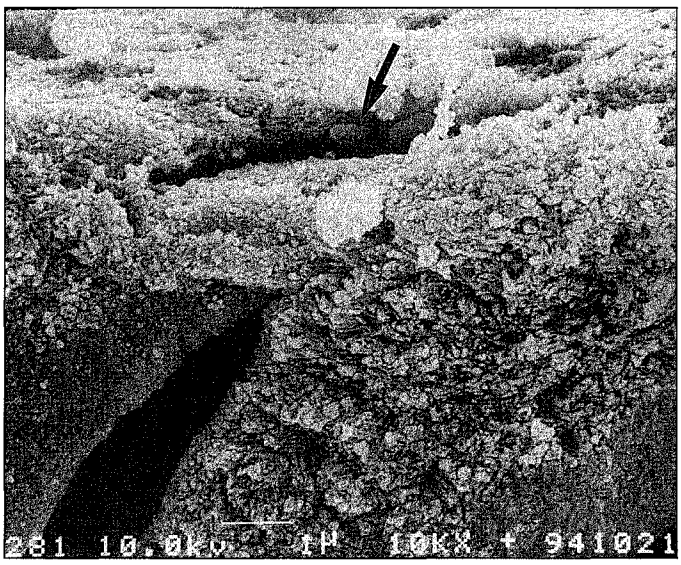

Figure 5. *SEM observation shows the cavity wall treated with ED primer (original magnification X6400). Little* S mutans *was seen.*

DISCUSSION

The need for restoratives with a potential antibacterial effect has been continuously discussed in dentistry. Glass-ionomer cements, for example, which release fluoride, also exhibit antibacterial activity, and chlorhexidine (Cawson & Curson, 1959) has been added to mouthrinse (Vaahtoniemi & others, 1995), cements (McComb & Ericson, 1987; Ribeiro & Ericson, 1991) and composite resin (Addy & Handley, 1981; Jedrychowski, Caputo & Kerper, 1983). Chlorhexidine is strongly bactericidal, especially for Gram-positive bacteria (Emilson & others, 1972). Chlorhexidine may also cause pulpal reaction. Many dentin adhesive primers and materials contain fluoride (Rawls & Zimmerman, 1983), tannic acid, and Ag zeolite because of their antibacterial effects.

Methacryloyloxydodecylpyridinium bromide (MDPB) (Imazato & others, 1994; Imazato, Russel & McCabe, 1995), which is an antibacterial monomer, was recently developed and tested for antibacterial activity. This monomer is co-polymerized with other monomers; therefore, the antibacterial agent is not released after curing.

The bonding strength of the various adhesive resin systems on the market has been improved not only to enamel but also to dentin (Sugisaki, 1991). Newly developed systems like Liner Bond II and Panavia 21 do not require rinsing with water during the bonding procedure. Although this reduces clinical chair time, there would be more risk of surviving cariogenic bacteria.

All three primers evaluated in the present study contained 5-NMSA, which is a highly antibacterial salicylic derivative. The main ingredient of SA primer is 5-NMSA, while LB primer and ED primer specifically contain a phosphoric adhesive monomer. Onoe and others (1993) suggested that the antibacterial activity of 5-NMSA against *S mutans* decreased when diluted. His findings agreed with our results, for the SA primer did not exhibit a strong antibacterial effect against *S mutans*. ED primer was found to be more effective than LB primer in this study. This might be explained by the fact that these two primers differ only in the type of adhesive monomer they contain, which suggests that the MDP in ED primer is more strongly antibacterial than the phenyl-p in LB primer.

The antibacterial activity of dentin primers is usually evaluated using the disk diffusion method. The inhibition zone observed in the disk diffusion method indicated bacteriostatic action of the specimens in the agar. However, this is not always indicative of bactericidal action, because the diffusion rate in the agar of specimens, such as primers, is normally different. Materials that do not release after curing, such as MDPB, could not be assessed by the disk method. Therefore, the bovine tooth model method was developed for evaluating the antibacterial activity of all dentin primers.

Prior to the present investigation, two preliminary examinations were performed. In the first, it was determined whether or not *S mutans* inoculated in a standardized cavity of a bovine tooth remained viable. In this model, the sterilized bovine teeth were inoculated with *S mutans*, sealed, and then cultivated for 1 week at 37 °C in bottles of Tryptic Soy broth. The second day after inoculation, bacterial growth was visible to the naked eye. The bacteria in the cavity were thought to have advanced toward the pulp, because the pulp chamber was filled with Tryptic Soy agar, *S mutans* is a nonmotile bacteria, and no microleakage from the cavosurface margin occurred. The results from the first preliminary examination showed that *S mutans* inoculated in teeth in this manner was a viable method.

To confirm the validity of bacteria collection from the cavity preparations following the 1-week incubation at 37 °C, the second preliminary experiment was performed. After cultivation, the teeth were divided into three groups to recover the remaining bacteria: A, from the whole tooth; B, from the tooth cut through the sagittal plane; C, from dentinal swabs of the cavity. No significant difference in the number of recovered *S mutans* was observed among the three methods. Thus, method A was chosen for the present study, because it was less complex than the other two methods.

Examination of the inhibition zones of the three primers, and the results of the disk diffusion tests in which the inhibition zone of SA primer was the smallest and the inhibition zone of ED primer was wider than that of LB primer, confirmed the results of the in vitro tooth model test developed in the present study.

CONCLUSION

The efficiency of the antibacterial effect of dentin primers was determined by the newly designed bovine tooth model test. All the primers showed antibacterial activity, with significant differences found among the groups of SA primer, LB primer, ED primer, and control. The new bovine tooth model method presented in this study proved to be an effective method to evaluate the antibacterial effect of dentin primers.

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Antibacterial Activity of Glass-Ionomer Restorative Cements Exposed to Cavity-producing Microorganisms

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Clinical Relevance
The confirmed antibacterial action of glass-ionomer cements may provide protection from the microorganisms involved in enamel caries as well as root caries.

SUMMARY

The antibacterial activity of the glass-ionomer restorative cements Ketac-Fil, Ketac-Silver, Fuji II LC, and Vitremer was studied in vitro, in conjunction with a total of 32 strains of five bacterial genera that may be associated with dental caries: *Streptococcus* spp, *Lactobacillus* spp, *Actinomyces* spp, *Porphyromonas* spp, and *Clostridium* spp. Agar plate diffusion was the method used for the bacterial cultures, which included a chlorhexidine control. All four glass-ionomer cements were found to inhibit bacterial growth, though with noteworthy differences in their spheres of action. Vitremer was the cement determined to have the greatest antibacterial effects, whereas Ketac-Silver presented the least inhibitory action.

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INTRODUCTION

The therapeutic procedures used in the treatment of caries do not always eliminate all the microorganisms in the residual tissues (Kidd, Joyston-Bechal & Beighton, 1993; Boston & Graver, 1994). The persisting bacterial presence, together with the lack of a thoroughly hermetic seal between the filling and the cavity walls, thus allowing bacterial leakage, may be involved in the development of recurring caries (Bergenholtz & others, 1982; Qvist, 1993). This is the most frequent cause for the replacement of restorative materials, whether amalgams (Qvist, Qvist & Mjör, 1990a; York & Arthur, 1993) or resin composites (Qvist, Qvist & Mjör, 1990b; York & Arthur, 1993). One possible solution

for this serious problem is to use dental materials with a bacteriostatic capacity.

Glass-ionomer cements are already widely used in restorative dentistry due to their pulpal biocompatability and adherence to dental structure. Several authors (DeSchepper, White & von der Lehr, 1989; Scherer, Lippman & Kaim, 1989; Behnen & others, 1990; Svanberg, Mjör & Orstavik, 1990; Benderli & others, 1997; Meiers & Miller, 1996) have determined the antibacterial action of the conventional glass-ionomer cements using different methodologies.

Photopolymerizable glass cements such as Vitrebond were created by adding resins to the composition of glass-ionomer cements, a modification that may affect their antibacterial properties. This fact, together with their growing use, justified researching their inhibitory capacity.

Although previous authors have studied these materials using *Streptococcus* spp (Barkhodar & others, 1989; Loyola-Rodríguez, García-Godoy & Lindquist, 1994) and *Lactobacillus* spp (Palenik & others, 1992), this study included three more bacteria that may be associated with cariogenic processes: *Porphyromonas* spp, *Actinomyces* spp, and, though to a lesser degree, *Clostridium* spp.

The antimicrobial properties of the different glass-ionomer cements currently available are due to their ability to release flouride (Barkhodar & others, 1989; DeSchepper & others, 1989), their low pH while setting (Fischman & Tinanoff, 1994; Fraga, Siqueira & De Uzeda, 1996) and/or the presence of certain cations, such as strontium and zinc, in some cements (Prati & others, 1993; Duke. Chan & Duke, 1995).

Chlorhexidine is an antiseptic with a wide spectrum of action, and its use has been generalized over the past two decades for the chemical control of bacterial plaque and the disinfection of therapeutic cavities. For these reasons, it is adopted as the positive control for studies on bacterial growth or antibacterial activity (Emilson & Bergenholtz, 1993).

The purpose of our study was to examine the in vitro response of five complete bacterial groups associated with the development of caries—*Streptococcus* spp, *Lactobacillus* spp, *Actinomyces* spp, *Porphyromonas* spp, and *Clostridium* spp (a total of 32 strains)—to the presence of four different glass-ionomer cements: Ketac-Fil, Ketac Silver, Fuji II LC, and Vitremer.

METHODS AND MATERIALS

Table 1 indicates the basic composition of each of the glass-ionomer cements (Ketac-Fil, Ketac-Silver, Fuji II LC, and Vitremer) tested. For Vitremer, the antibacterial properties of the different components of the system were studied separately: the primer was referred to as Vitremer A, the cement itself as Vitremer B, and the finishing gloss as Vitremer C. The bacteria used were autochthonous strains of international reference, in this case from the collection of the Dentistry Section of the Department of Microbiology, University of Granada, Spain. The bacteria pool consisted of 32 strains belonging to the genera *Streptococcus, Lactobacillus, Actinomyces, Porphyromonas,* and *Clostridium* (Table 2).

The antibacterial activity of the four cements was observed by means of agar plate diffusion, a method that has been described previously (Tobias, 1988; Prati & others, 1993; Meiers & Miller, 1996).

The lyophilized bacteria were rehydrated, and their purity and viability were confirmed in ideal culture media. They were then inoculated onto plates containing brain-heart infusion broth (BHI 0418-01-5; Difco Laboratories, Detroit, MI 48232) and incubated for 48-72 hours in the appropriate atmosphere: for *Lactobacillus*, aerobic conditions

Table 1. Dental Materials Used in Study

Materials	Symbol	Manufacturer (Batch #)	Principal Components
Ketac-Fil	KF	ESPE Seefeld/Oberbay, Germany (022A28)	F- aluminum silicate
Ketac-Silver	KS	ESPE (P064)	F- aluminum silicate and silver
Fuji II LC	FIILC	GC America Chicago, IL 60658 (130224)	F- aluminum silicate, HEMA
Vitremer		3M Dental Products	
Primer	VM-A	St Paul, MN 55144	HEMA, carboxylic acid copolymer
Complete cement	VM-B	(19930119)	F- aluminum silicate, HEMA
Finishing gloss	VM-C		Ascorbic acid, hydrogen peroxide

Table 2. Mean Diameters of the Inhibition Halos, in mm, Produced by Glass-Ionomer Cements

	KF	KS	F II LC	VM-A	VM-B	VM-C	CLX
S mutans ATCC 25175	18	12	17	23	26	21	17
S rattus ATCC 19645	16	13	21	17	32	18	15
S sobrinus NCFB 2724	16	13	12	21	25	15	15
S cricetus ATCC 19642	17	12	15	18	26	11	16
S sobrinus ATCC 33478	15	11	13	18	23	11	16
S sanguis ATCC 10556	15	12	20	17	30	19	20
S oralis NCTC 11427	16	13	19	27	32	21	13
S mitis NCTC 3165	15	12	25	23	30	23	16
S anginosus ATCC 33397	15	11	17	20	28	20	15
Inhibition percentage	100%	100%	100%	100%	100%	100%	100%
L salivarius ATCC 11741	11	11	24	0	0	0	22
L oris NCDO 2160	0	0	11	0	0	0	16
L plantarum ATCC 14917	0	0	0	0	0	0	14
L acidophilus ATCC 4356	0	0	0	0	0	0	15
L casei ATCC 7469	0	0	0	10	12	0	16
Inhibition percentage	20%	20%	40%	20%	20%	0%	100%
A odontolyticus ATCC 17982	26	27	30	31	36	30	33
A viscosus ATCC 15987	25	24	31	30	35	28	36
Inhibition percentage	100%	100%	100%	100%	100%	100%	100%
P gingivalis OGP-2	11	0	12	18	22	15	22
P gingivalis OGP-15	11	0	12	18	25	12	20
P gingivalis OGP-30	0	0	0	65	75	31	19
P gingivalis OGP-46	13	0	11	25	36	28	13
P gingivalis OGP-91	10	0	10	70	75	35	16
P gingivalis OGP-96	12	10	13	60	70	40	23
Inhibition percentage	83.3%	16.7%	83.3%	100%	100%	100%	100%
C malenominatum OGC-58	12	11	12	43	40	22	16
C malenominatum OGC-59	10	10	0	36	33	20	13
C malenominatum OGC-60	10	0	0	13	18	11	14
C malenominatum OGC-65	12	11	12	50	45	22	15
C ramosum OGC-66	12	10	13	40	37	22	15
C ramosum OGC-67	15	12	12	40	40	25	17
C ramosum OGC-68	12	11	11	36	37	20	15
C sporogenes OGC-70	10	0	0	12	14	0	13
C sporogenes OGC-71	11	0	0	36	33	20	16
C sporogenes OGC-72	11	0	11	10	15	11	18
Inhibition percentage	100%	60%	60%	100%	100%	90%	100%
Mean Inhibition Percentage	84.4%	59.4%	75%	87.5%	87.5%	81.3%	100%

with 5-10% CO_2; and anaerobic conditions for the rest of the bacteria. From these cultures, bacterial suspensions were prepared in a sterile isotonic saline solution, until obtaining a turbidity compatible with the 0.5 of MacFarland. This scale allowed the bacterial concentration of a suspension to be estimated by its turbidity; 0.5 corresponded to a concentration of 1.5×10^8 at an optic density of 550 nm. Then 150 µL of the standardized inocula were spread out evenly in 15 cm-in-diameter Petri dishes containing BHI agar (enriched with hemin and vitamin K for the *Porphyromona* spp inocula) to a thickness of 4 mm. Wells with a diameter of 6 mm were made in the agar with the blunt end of a sterile Pasteur pipette. The four cements (Ketac-Fil, Ketac-Silver, Fuji II LC, and Vitremer) were prepared following the respective manufacturer's instructions, and immediately were put into the wells, completely filling them. The wells containing Fuji II LC and Vitremer were photocured for 40 seconds using a polymerizing lamp (Heliolux 533505; Vivadent, Schaan, Liechtenstein).

In order to test the activity of the liquid elements of the Vitremer system (the primer and the finishing gloss), sterile paper disks with a diameter of 6 mm (54991 Disques nonimprégnés; bioMérieux SA, Marcy l'Etoile, France) were placed on the agar surface using sterile tweezers, and 10 µL of the primer or gloss respectively were added to the cultures and then photocured for 20 seconds.

Incubation of the dishes took place under aerobic or anaerobic conditions, depending on the type of bacteria in observation. After 48 hours of incubation at 36±1°C, readings were taken, measuring the diameter of the inhibition halos in millimeters.

The positive control used was a 0.2% aqueous solution of chlorhexidine added onto paper disks, as described above for the Vitremer components.

All the assays were carried out in triplicate, and results were recorded in terms of the average diameter of the positively inhibitory halos surrounding each material (halo ≥10 mm), rounded off to the nearest millimeter. The statistical analysis of results was based on the McNemar test for paired data. After determining the percentage of positive results for each material tested, this value was compared for each material (including chlorhexidine) in conjunction with each bacterial species.

Table 3. Intervals, in mm, of the Inhibitory Activity of the Glass-Ionomer Cements with Respect to the 32 Bacterial Strains Tested

	<10	10-15	16-20	≥ 21
KF	5 (15.6%)	20 (62.5%)	5 (15.6%)	2 (06.2%)
KS	13 (40.6%)	17 (53.1%)	---	2 (06.2%)
F II LC	8 (25.0%)	15 (46.8%)	4 (12.5%)	5 (15.6%)
VM-A	4 (12.5%)	4 (12.5%)	7 (21.8%)	17 (53.1%)
VM-B	4 (12.5%)	3 (09.3%)	1 (03.1%)	24 (75.0%)
VM-C	6 (18.7%)	7 (21.8%)	6 (18.7%)	13 (40.6%)

RESULTS

Table 2 gives the values, expressed in mm, obtained for the mean inhibition halos produced by each glass-ionomer cement studied and the chlorhexidine control, when exposed to the five groups of bacteria included in this study. Vitremer proved to be the most inhibitory material, particularly in conjunction with the bacterial strains of the *Porphyromonas* genus. The strains of genus *Lactobacillus*, on the other hand, were remarkably resistant not only to Vitremer, but to the other cements as well. The glass-ionomer cement showing the least antibacterial activity overall was Ketac-Silver (59.4%).

The inhibitory activity intervals, expressed in mm, of the studied materials with respect to the 32 strains tested are given in Table 3. The Vitremer cement produced the greatest inhibition halos, measuring 21 mm or more in 75% of the cases. Ketac-Fil and Ketac-Silver, cements with a conventional setting process, gave inhibition halos with diameters ranging between 10 and 15 mm in over 50% of the cases.

Table 5. Statistical Comparison of the Overall Antibacterial Activity of Each Glass-Ionomer Cement Tested with Respect to the Others and the Chlorhexidine Control

	KF	KS	F II LC	VM-A	VM-B	VM-C	CLX
KF							
KS				*	*		*
F II LC							*
VM-A							
VM-B							
VM-C							*

*significantly less inhibitory ($P < 0.05$) than the cement at the head of the column

Table 4 shows the results observed for chlorhexidine, which proved inhibitory in 100% of the instances. In most cases it produced halos with diameters measuring over 15 mm.

Finally, Table 5 offers the results of the statistical analysis, which compared the inhibitory action of the glass-ionomer cements and chlorhexidine with respect to each of the bacterial strains tested, and determined Vitremer A and Vitremer B to be particularly effective antibacterial materials.

DISCUSSION

The methodology used for the study of the antibacterial activity of dental materials is not standardized, and several different techniques have been proposed (Tobias, 1988; Meryon & Johnson, 1989; Loyola-Rodríguez & others, 1994). Agar plate diffusion was the method of choice for this study because it allowed both solid and liquid materials to be assayed (Emilson & Bergenholtz, 1993; Meiers & Miller, 1996). The micro-organisms used included the bacteria most frequently associated with the onset of caries: oral streptococci, *Lactobacillus* spp, and *Actinomyces* spp. The reasoning behind including *Porphyromonas* spp and *Clostridium* spp was based on substantiations that in dentin caries development, acidogenic bacteria are substituted by proteolithic bacteria such as these (Castillo & Liébana, 1995).

Table 4. Intervals, in mm, of the Inhibitory Action of Chlorhexidine on the 32 Bacterial Strains Tested

	<10	10-15	16-20	≥21
Oral streptococci	---	4 (44.4%)	5 (55.5%)	---
Lactobacillus spp	---	2 (40.0%)	2 (40.0%)	1 (20.0%)
Actinomyces spp	---	---	---	2 (100%)
Porphyromonas spp	---	1 (16.6%)	3 (50.0%)	2 (33.3%)
Clostridium spp	---	6 (60.0%)	4 (40.0%)	---

The present study is unusual in that it undertook the observation and comparison of an entire bacterial pool. For this reason, caution must be used when comparing these results with those of authors who have worked with a more limited variety of microorganisms (Svanberg & others, 1990; Meiers & Miller, 1996). The results obtained clearly showed Vitremer to be the glass ionomer with the greatest antibacterial activity (87.5%). The strains belonging to the genus *Lactobacillus* were the only ones that exhibited a resistance to this material, with the exception of the *L*

casei species, for which positive inhibition halos measuring an average of 12 mm were observed. Similar findings were reported by Fraga and others (1996), although their study did not include other *Lactobacillus* species.

The inhibitory effect produced on the majority of the strains by each of the three components of the Vitremer system was nearly always superior to the inhibition exerted by the other glass ionomers tested. In fact, Vitremer produced halos with remarkable diameters of up to 75 mm in some cases (Vitremer B with *P gingivalis* OGP-91). Our testing method did not allow us to surmise whether the sequential clinical application of these three components might enhance their individual antibacterial potential. This is a possibility to be explored by future research.

Except when exposed to *Actinomyces* spp and *Streptococcus* spp, Fuji II LC and Ketac-Fil showed limited antibacterial effectiveness. However, Ketac-Silver proved to be the least effective overall of the materials tested.

Streptococci results coincided with the findings of Loyola-Rodríguez and others (1994), who studied the growth of *S mutans* and *S sobrinus* in the presence of Ketac-Silver, Fuji II LC, Ketac-Fil, and Vitremer; they found all to exercise effective antibacterial action, the greatest inhibition halos corresponding to Vitremer and Fuji II LC, and the smallest to Ketac-Silver. Meiers and Miller (1996), in their study of Fuji II LC in conjunction with *L salivarius, A viscosus, S mutans,* and S *sobrinus*, obtained results similar to this study for the two streptococci strains, but observed less antibacterial action for *A viscosus* and *L salivarius*. Scherer and others (1989), with Ketac-Fil and Ketac-Silver, observed substantially less antibacterial action, especially for *A viscosus*. DeSchepper and others (1989) tested the antibacterial activity of Ketac-Fil on *S mutans*, and reported values lower than the ones obtained in this study.

A comparison of the antibacterial action of the different cements and the chlorhexidine control showed no statistically significant differences between chlorhexidine and Ketac-Fil or Vitremer; there were, however, significant differences between chlorhexidine and both Ketac-Silver and Fuji II LC (Table 5).

The discrepancies among the results documented by different authors may be influenced by the methodology applied: in some cases wells with varying diameters were made in the agar; whereas in the case of Palenik and others (1992) no wells were made, and the set materials were simply placed on the agar surface.

According to the results of this study and those of most other studies of these materials, a number of glass-ionomer cements exhibit antibacterial activity, to a greater or lesser degree. Attempts to describe the nature of this action, and to explain why one same group of materials can show such different degrees of effectiveness, have given rise to diverse interpretations. Some authors agree that the release of fluoride into the medium may be the cause (Chong & others, 1994; Eli & others, 1995). Other researchers point to the variations in pH and temperature that are produced in the medium (DeSchepper & others, 1989), or to the differing powder/liquid proportion of the cements (Perrin, Persin & Sarrazin, 1994). Yet another study focused on the amount of fluoride necessary to produce the inhibition of each bacterial group: Hamilton (1990) indicates, for example, that 50 to 300 ppm of fluoride were necessary to inhibit the development of *S mutans*.

CONCLUSIONS

In light of the reported results and those mentioned above, the use of glass-ionomer cements may be indicated in the restorative treatment of root surface caries. The confirmed antibacterial action of these materials is not limited to the microorganisms of the enamel, such as streptococci, but also may protect against the microorganisms involved in caries of the cementum, such as *Actinomyces* spp. In addition, these cements could prove beneficial for patients with periodontal disease who also suffer from root caries, where periodontopathogenic bacteria like *Porphyromonas* spp may be present.

It is the authors' hope that in vivo studies of dental restorative materials will soon come to support the in vitro antibacterial properties described here.

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Color of Restorative Materials after Staining and Bleaching

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Clinical Relevance
The bleaching agent tested removed stains from the composite and the hybrid ionomer but not the compomer.

SUMMARY

This study determined the effect of a 10% carbamide peroxide bleaching agent on the removal of stain from restorative materials. Color changes (ΔE*) of three restorative materials [compomer (Dyract); composite (TPH Spectrum); hybrid ionomer (Fuji II LC)] when exposed to juice/tea, chlorhexidine (CH), and water (control) for 120 hours were studied. Stained specimens were treated for two 2-hour periods with a bleaching agent (Platinum Tooth Whitening System) with and without the active ingredient. Color was measured at baseline, after staining, and after treatment using the CIE L*a*b* color system relative to CIE standard illuminant A (incandescent light) as measured by a reflection spectrophotometer. Means and standard deviations (n=5) were calculated and data were analyzed by four-way ANOVA. All variables and interactions were statistically significant. Color changes caused by CH and water were not perceptible (ΔE* < 3.3). After two 2-hour treatments, the following occurred with specimens stained with cranberry juice/tea: paste with and without active ingredient perceptibly changed color of stained composite. The stained hybrid ionomer perceptibly changed color after treatment with paste containing active ingredient but did not change after exposure to paste without active ingredient. The stained compomer was not perceptibly different with either treatment. Platinum successfully removed stains from the composite and hybrid ionomer tested.

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INTRODUCTION

Patients often seek dental treatment for esthetic problems such as discolored teeth. Methods including vital bleaching, porcelain veneers, resin veneers, and

ceramic crowns have been developed to treat discolored teeth. Of these, both in-office and nightguard vital bleaching are used because of their conservative approach to and their effectiveness in removing tooth discolorations. Numerous studies have shown bleaching to be effective in whitening certain types of discolored teeth (Matis, Cochran & Heinonen, 1997; Haywood, Leonard & Dickinson, 1997, Russell & others, 1996; Myers & others, 1995; Rosenstiel, Gegauff & Johnston, 1996; Simon & others, 1993; Gegauff & others, 1993; Haywood, 1992a,b; Wilson & Seale, 1985; Rosenstiel & others, 1991; Seale & Thrash, 1985). Nightguard vital bleaching was first introduced in the literature in 1989 (Haywood & Heymann, 1989). In clinical studies, nightguard vital bleaching has been known to have limited side effects (Haywood & others, 1994, 1997).

Questions have been raised regarding the effect of bleaching agents on restorative materials, particularly tooth-colored materials. Hunsaker, Christensen, and Christensen (1990) studied seven brands of bleaching gels and their effect on dentin, enamel, gold alloy, amalgam, porcelain, macrofill, and microfill restorative resins. Upon observation by scanning electron microscopy, they found no major changes in tooth structure or restoratives. Robinson, Haywood, and Myers (1997) studied the effect of 10% carbamide peroxide on the color of provisional restorations. They reported that an orange discoloration was present with methacrylate provisional materials, while resin composite provisional material was not affected by the carbamide peroxide. Cooley and Burger (1991) evaluated composites for changes in surface roughness, hardness, and lightness after exposure to carbamide peroxide and noted some statistically significant increases in all three measures. Bailey and Swift (1992) found similar slight roughening and softening of hybrid composites with one type of carbamide peroxide. Monaghan, Trowbridge, and Lautenschlager (1992) concluded that there were significant color changes in composite samples after 37% phosphoric acid etching and bleaching with 30% hydrogen peroxide with infrared light for 30 minutes. In a follow-up study, Monaghan, Lim, and Lautenschlager (1992) found no differences in composite color after exposure to carbamide peroxide with water as the control.

There are no reports of how bleaching agents affect restorations that have been stained. This study evaluated the effectiveness of a 10% carbamide peroxide bleaching agent in removing stains from three restorative materials: a composite, a compomer, and a hybrid ionomer.

METHODS AND MATERIALS

The materials tested were a compomer (Dyract; Dentsply/Caulk, Milford, DE 19963), a composite (TPH Spectrum; Dentsply/Caulk); and a hybrid ionomer (Fuji II LC; GC America, Chicago, IL 60658) as described in Table 1. Five specimens (10 x 2 mm disks) of each restorative material were made for each experimental and control group. Specimens were made by placing the restorative material in a polytetrafluoroethylene ring between glass slides and plastic strips and curing them on each side for 40 seconds with a light-curing unit (The Max; Dentsply/Caulk), which had been tested for proper output by a light meter (Curing Radiometer; Demetron/Kerr, Danbury, CT 06810). Specimens were roughened with silicon carbide paper (600-grit) to assist in stain retention and incubated in 100% humidity at 37° C for 24 hours, then a baseline measurement was taken.

Color was measured with the CIE L*a*b* color system relative to CIE standard illuminant A (incandescent light) against a white background on a reflection spectrophotometer (Color-Eye 7000; MacBeth Division of Kollmorgen Instruments Corp, Newburgh, NY 12550). Each group was exposed to cranberry juice/tea, chlorhexidine (Peridex; Proctor & Gamble, Cincinnati, OH 45201), or water for 120 hours, gently rinsed with water for 1 minute, and air dried. Chlorhexidine was included in this study to determine if its use would cause staining in the absence of plaque. Color of the stained specimens

Table 1. Products, Manufacturers, Shades, and Lot Numbers

Product	Manufacturer	Shade	Lot #
Fuji II LC	GC America	A2	031068
Dyract	Dentsply/Caulk	A2	9703000927
TPH Spectrum	Dentsply/Caulk	A2	9607162
Peridex	Procter & Gamble		H95019-052
Platinum (with active ingredient)	Colgate Oral Pharmaceuticals		604081 2079-10
Platinum (without active ingredient)	Colgate Oral Pharmaceuticals		
Cranberry Juice Cocktail	Ocean Spray (3.78 L)		
Instant Tea	Nestea (85 g)		

was then measured. Five specimens from each group were treated with 0.5 cc paste (Platinum Tooth Whitening System; Colgate Oral Pharmaceuticals, Canton, MA 02021) containing active ingredient 10% carbamide peroxide. In addition, five specimens from each group were treated with 0.5 cc paste (Platinum) that did not contain the active ingredient. The specimens were stored in 100% humidity at 37° C for 2 hours, gently rinsed with water for 1 minute, and air dried. Color was then remeasured. The process was repeated and color was measured after a total of 4 hours of treatment.

Color change was measured according to the following formula:

$$\Delta E^* = [(L^*_O - L^*_I)^2 + (a^*_O - a^*_I)^2 + (b^*_O - b^*_I)^2]^{1/2}$$

where ΔE^* = color change, L^* = luminance reflectance, a^* = red-green color coordinate, b^* = yellow-blue color coordinate, O = baseline, and I = treatment interval.

Color change (ΔE^*) data after 4 hours of treatment were analyzed by four-way analysis of variance for fixed effects (SuperANOVA; Abacus Concepts, Berkeley, CA 94704) for three materials, three staining solutions including water as a control, stained vs baseline conditions, and treatment with and without active bleaching ingredient. Tukey-Kramer intervals for comparison of means were calculated at the 0.05 significance level from the analysis of variance. Differences between means that were larger than the appropriate Tukey-Kramer intervals were considered significant.

RESULTS

Means and standard deviations (n=5) of ΔE^* after staining and after two 2-hour treatment periods are listed in Table 2. The results of the analysis of variance are shown in Table 3. The main effects except treatment and most interactions were statistically significant. Tukey-Kramer intervals ($P < 0.05$) for comparisons of ΔE^* among three materials, among three stains, between stained vs baseline conditions, and between treatment with and without active ingredient were 0.3, 0.3, 0.2, and 0.2 respectively. ΔE^* values greater than 3.3 were considered visually perceptible (Ruyter, Nilner & Moller, 1987).

Color changes caused by chlorhexidine and water were not perceptible ($\Delta E^* < 3.3$). There was no perceptible change in color after bleaching the specimens stored in water (controls) for two 2-hour treatments. Juice/tea perceptibly stained all three restorative materials. The hybrid ionomer was stained by juice/tea more than were the compomer or the composite.

After treatment of the composite, both the active treatment and control treatment reduced the stain such that there was not a perceptible difference between baseline and posttreatment color. The hybrid ionomer reverted to a color similar to baseline with the active treatment; it did not revert with the control treatment. The compomer, however, still showed a perceptible difference from baseline after both active treatment and control treatment.

Table 2. Color Change (ΔE^*) of Three Restorative Materials between Baseline and Staining with Juice/Tea, Chlorhexidine, and Water for 120 Hours and between Baseline and Treatment with or without Active Ingredient

	Compomer	Composite	Hybrid Ionomer
Platinum with active ingredient:			
Stained--juice/tea	5.6 (0.5)*	4.6 (1.5)	10.3 (1.6)
Bleached--4 hours, juice/tea	5.0 (0.9)	1.9 (1.1)	2.0 (0.5)
Stained--chlorhexidine	1.4 (0.7)	1.4 (0.9)	2.8 (0.5)
Bleached--4 hours, chlorhexidine	0.8 (0.4)	1.3 (0.1)	2.5 (0.4)
Stained--water	0.9 (0.6)	0.8 (0.3)	2.0 (0.6)
Bleached--4 hours, water	1.3 (0.6)	0.5 (0.3)	2.4 (0.5)
Platinum without active ingredient:			
Stained--juice/tea	5.9 (1.5)	3.9 (0.9)	10.1 (0.6)
Bleached--4 hours, juice/tea	5.6 (1.3)	2.2 (0.2)	7.2 (0.8)
Stained--chlorhexidine	1.6 (0.4)	1.3 (0.6)	3.0 (0.6)
Bleached--4 hours, chlorhexidine	0.5 (0.4)	1.0 (0.2)	1.8 (0.4)
Stained--water	1.6 (1.0)	0.8 (0.5)	1.7 (0.8)
Bleached--4 hours, water	0.7 (0.2)	1.0 (0.6)	1.5 (0.2)

*Means with standard deviations in parentheses. Tukey-Kramer intervals ($P < 0.05$) for ΔE^* among three materials, among three stains, between stained vs bleached conditions, and between active ingredient and control were 0.3, 0.2, and 0.2 respectively.

Table 3. Analysis of Variance of Color Change (ΔE) among Three Restorative Materials, among Three Staining Solutions, between Baseline and Staining for 120 Hours, and between Treatment with or without the Active Ingredient*

Source	df	Sum of Squares	Mean Square	*F*-Value	*P*-Value
Material (M)	2	152.60	76.300	137.00	0.0001
Condition (C)	1	58.80	58.800	106.00	0.0001
Stain (S)	2	616.00	308.000	554.00	0.0001
Treatment (T)	1	2.01	2.010	3.62	0.0590
MC	2	20.60	10.300	18.50	0.0001
MS	4	69.40	17.300	31.20	0.0001
MT	2	2.54	1.270	2.29	0.1050
CS	2	60.90	30.500	54.70	0.0001
CT	1	2.13	2.130	3.83	0.0520
ST	2	10.80	5.380	9.66	0.0001
MCS	4	50.70	12.700	22.80	0.0001
MCT	2	5.99	2.990	5.38	0.0056
MST	4	19.00	4.750	8.54	0.0001
CST	2	19.20	9.590	17.20	0.0001
MCST	4	13.90	3.470	6.23	0.0001
Error	144	80.10	0.557		

DISCUSSION

There was no perceptible difference in specimens stored in water for 120 hours. After 4 hours of chlorhexidine treatment, with and without the active ingredient, there was no perceptible difference in color from baseline measurements of the three materials. Therefore, the paste did not affect the color of the materials after 4 hours of exposure.

After 120 hours, chlorhexidine did not perceptibly stain the specimens. Even though there was a statistically significant change after treatment, a perceptible difference could not be noted.

Both active and control treatments removed juice/tea stains from two of the materials. The result that the paste without active ingredient removed stain from the composite and the hybrid ionomer suggested that other factors besides the carbamide peroxide may aid in stain removal. The surfactant effect of the sodium lauryl sulfate or addition of pluronic acid included in the bleaching agent may be additional factors influencing the stain-removal ability of this bleaching agent.

In addition, the type of restorative material was important. The hybrid ionomer had the greatest stain from baseline after staining with juice/tea. An earlier study indicated that glazing the surface of the hybrid ionomer made it more resistant to stain (Fay, Walker & Powers, 1998). A surface glaze was not used in the present study because the intended outcome was to stain the specimens. After treatment with the active ingredient, the color change from baseline was no longer perceptible. After treatment without the active ingredient, there was still a perceptible difference in color change from baseline. The composite had the least change from baseline after staining with juice/tea. After both active treatment and control treatment, the color change from baseline was no longer perceptible. ΔE* values of the compomer after staining were between those of the hybrid ionomer and the composite. After both active treatment and control treatment, there was still a perceptible color difference from baseline, indicating that treatment did not remove stain from the compomer.

The different effects on restorative materials may be due to different surface reactivity. Staining appeared to be on the surface of the composite. With the hybrid ionomer and the compomer, staining was observed to be deeper. Materials that contain glass ionomer tend to absorb water (Nicholson, Anstice & McLean, 1992), which may account for enhanced staining.

CONCLUSION

The bleaching agent used removed stains from composite and hybrid ionomer. It was not effective in removing them from the compomer. This finding is of clinical significance in that it can help determine which restorative material is best used in conjunction with bleaching for long-term success.

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Fluoride Release and Antibacterial Properties of New-Generation Tooth-colored Restoratives

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Clinical Relevance
For the materials investigated, the conventional glass-ionomer cement released significantly more fluoride than fluoride-releasing composites, compomers, or the resin-modified glass-ionomer cement. There was no correlation noted between fluoride-release potential and antibacterial properties.

SUMMARY

The aim of this study was to compare the amounts and pattern of fluoride release and antibacterial properties of new-generation restoratives over a 35-day period. Materials evaluated included fluoride-releasing composites (Tetric, Experimental X), compomers (Dyract, Compoglass), and a resin-modified glass-ionomer cement (Fuji II LC). A conventional glass ionomer (Fuji II Cap) was used as a control for fluoride-release testing. Five samples of each restorative material were evaluated for daily fluoride release over a 35-day period by means of ion chromatography. Ranking of materials from least to greatest total fluoride release over 35 days was as follows: Tetric < Experimental X < Dyract < Fuji II LC < Compoglass < Fuji II Cap. Fuji II Cap had significantly greater fluoride release than all other materials evaluated. Fuji II Cap, Fuji II LC, and Compoglass had similar patterns of fluoride release characterized by a high initial release that was many times that released later. The fluoride-releasing composites evaluated stopped releasing fluoride by day 14. Antibacterial testing was conducted using the agar diffusion inhibitory test. Five samples of each restorative were assessed at baseline and weekly intervals up to 35 days. The microorganisms used were *Lactobacillus casei*, *Streptococcus mutans,* and *Streptococcus sobrinus*. IRM, a zinc oxide/eugenol cement, was used as the baseline control. None of the restorative materials evaluated affected the growth of *L casei*, *S sobrinus*, or *S mutans* at all time periods including baseline, where fluoride was detected in the agar beneath the specimen disks. There was no correlation noted between fluoride-release potential and antibacterial properties.

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INTRODUCTION

Secondary or recurrent dental caries is by far the most frequent reason for replacement of restorations (Mjör, 1985). It is by definition found at the interface of tooth and restoration and is, in general, a result of microleakage (Arends, Dijkman & Dijkman, 1995). The breakdown of marginal integrity between restorative materials and cavity preparations can provide potential pathways for reinfection, as cariogenic microorganisms can easily penetrate into the underlying dentin through these defects (Brännström & Nordenvall, 1978; Brännström, 1984a). These microorganisms may also multiply in the smear layer on the prepared cavity walls (Brännström & Nyborg, 1973). Work on germ-free animals has shown that the healing capacity of injured pulp is impaired by bacteria contamination (Kakehashi, Stanley & Fitzgerald, 1965). In addition, pulpal damage during the restorative process is caused by bacteria and not by restorative materials (Brännström, 1984b). Reducing, or preferably the elimination of, microleakage is therefore an important element in reducing the incidence of secondary caries and subsequent pulpal damage. Although a good seal to enamel can be obtained with the acid-etch technique, the seal to dentin is still far from ideal. The marginal sealing abilities of tooth-colored restoratives, including new-generation restoratives like resin-modified glass-ionomer cements and polyacid-modified composite resins, were found to be significantly poorer in dentin than in enamel (Yap, Lim & Neo, 1995; Sjodin, Uusitalo & van Dijken, 1996). It would therefore be an advantage if restorative materials possessed antibacterial activity.

Glass-ionomer restorative materials and cements have demonstrated in vitro bactericidal abilities in published data (McComb & Ericson, 1987; Tobias, 1988; Scherer, Lippman & Kaim, 1989; DeSchepper, White & von der Lehr, 1989; Ribeiro & Ericson, 1991). Glass-ionomer cements have also been associated with reduced secondary caries development (Svanberg, Krasse & Ornerfeldt, 1990; Wood, Maxymiw & McComb, 1993). This favorable result has been attributed to the release of high concentrations of fluoride ions from the cement and the initially low material pH (McComb & Ericson, 1987; DeSchepper & others, 1989; Scherer & others, 1989). The presence of fluoride ions has been shown to inhibit the enolase enzyme, which converts 2-P-glycerate to P-enolpyruvate in the glycolytic pathway of carbohydrate metabolism, thereby inhibiting bacteria growth (Bibby & Van Kesteren, 1940).

Despite the in vitro efficacy of glass ionomers in reducing secondary caries, relatively little work has been done on new-generation fluoride-releasing restorative materials. These include resin-modified glass-ionomer cements, polyacid-modified resin composites (compomers), and fluoride-releasing resin composites. The purpose of this study was to determine the in vitro fluoride release and antibacterial properties of these new-generation tooth-colored restoratives. The relationship between fluoride-release potential (defined as the maximum possible fluoride release into an aqueous medium) and antibacterial properties was also investigated.

METHODS AND MATERIALS

Fluoride Release

The tooth-colored restorative materials tested are presented in Table 1. Five disk-shaped specimens, 6.0 ± 0.1 mm in diameter and 1.2 ± 0.1 mm thick, of each restorative material were formed in Teflon molds between glass plates. In each case, the mixing (where applicable) and curing procedures were those recommended by the respective manufacturers. The materials were then placed in a chamber of 37 °C and 100% relative humidity for 1 hour prior to separation from the molds. The actual dimension of each disk was measured using an electronic digimatic caliper (CD6BS; Mitutoyo Corp, Tokyo, Japan) to

Table 1. Tooth-colored Restorative Materials Tested

Restorative Material (Batch #)	Manufacturer	Material Type
Tetric Ceram (908369)	Vivadent Schaan, Liechtenstein	Fluoride-releasing resin composite
Experimental X (06972702)	Shofu Inc Tokyo, Japan	Fluoride-releasing resin composite
Dyract (9602940)	Dentsply Milford, DE 19963	Polyacid-modified resin composite (compomer)
Compoglass (912480)	Vivadent	Polyacid-modified resin composite (compomer)
Fuji II LC (250767)	GC Corp Tokyo, Japan	Resin-modified glass-ionomer cement
Fuji II Cap (091145)	GC Corp	Glass-ionomer cement

Table 2. Total Fluoride Release (ng/mm²) over 35 Days and Mean Daily Fluoride Release (ng/mm²) for Each Week

Material	Total Fluoride Release	Mean Week 1 (SD)	Mean Week 2 (SD)	Mean Week 3 (SD)	Mean Week 4 (SD)	Mean Week 5 (SD)
Tetric	9.77	0.77 (0.44)	0.62 (0.68)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
Experimental X	16.42	1.27 (0.94)	1.07 (1.18)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
Dyract	57.41	3.22 (1.53)	2.20 (1.32)	1.10 (0.47)	0.97 (0.12)	0.71 (0.12)
Compoglass	102.52	7.14 (4.72)	2.65 (1.78)	1.73 (0.58)	1.69 (0.25)	1.44 (0.22)
Fuji II LC	97.22	6.90 (5.85)	3.29 (2.20)	1.39 (0.52)	1.39 (0.92)	0.91 (0.33)
Fuji II Cap	442.64	37.00 (35.80)	10.73 (3.09)	6.40 (1.08)	5.24 (0.67)	3.85 (0.38)

ensure standardization of specimen size and for calculation of the exposed surface area.

After removal from the mold, each specimen disk was soaked in 9 ml of deionized water in a plastic container. Each disk, within its container, were placed in an orbital incubator (IOC400; Sanyo-Gallenkamp, Leicester, England) at 110 rpm and 37 °C. Twenty-four hours later, each specimen was rinsed with 1 ml of deionized water and transferred to fresh storage medium (9 ml of deionized water). The rinse water was added to the container with the storage medium, and the fluoride content was determined by means of ion chromatography (Waters 510 Ion Chromatograph; Waters Corp, Milford, DE 19963). This regimen of specimen transfer and fluoride analysis of storage medium was continued for 35 days.

The data for total weekly and total fluoride release over 35 days were subjected to a Kruskal-Wallis test at a significance level of $P < 0.05$. The total weekly fluoride release was the average fluoride released for five specimens for each 7-day period. The mean daily fluoride release for each week was also calculated and tabulated (Table 2). Intermaterial comparison was done using the Mann-Whitney Wilcoxson's Rank Sum test at a significance level of $P < 0.05$.

Antibacterial Properties

Bacteria

The microorganisms used were: (1) *Lactobacillus casei* (ATCC 4646), (2) *Streptococcus mutans* (ATCC 33535), and (3) *Streptococcus sobrinus* (ATCC 27607). All are human isolates and were obtained from the American Type Culture Collection (Rockville, MD 20852). These three microorganisms were selected as they are commonly associated with human dental caries (Hardie, 1992). Cultures were started from freeze-dried stocks into 10 ml of sterilized MRS broth (Becton Dickinson and Co, Franklin Lakes, NJ 07417) for *Lactobacillus casei* and Tryptic Soy Agar (TSA) broth with 5% sheep blood (Becton Dickinson and Co) for *S mutans* and *S sobrinus*. Aerobic incubation of *L casei* occurred at 37 °C (UL 40; Memmert, Schwabach, Germany). *S mutans* and *S sobrinus* were incubated in an infrared CO_2 incubator (Model 3194; Forma Scientific Inc, Marietta, OH 45750) at 37 °C in 5% CO_2. The microorganisms were allowed to grow for 48 hours prior to being used.

Specimen Preparation

In addition to the restorative materials in Table 1, IRM (Dentsply/Caulk, Milford, DE 19963), a zinc oxide/eugenol cement, was used as the baseline control. The specimen disks were prepared as described for fluoride release, and IRM was prepared according to the manufacturer's instructions. Pressure was placed on the glass plates during setting and curing to assure a flat contact surface. After separation from the molds, the specimen disks were transferred onto the bacterial plates. Five disks were tested for each microorganism. Templates, glass slides, and other necessary items were disinfected with methylated spirit (Septanol solution; ICM Pharma, Singapore) and subjected to 15 minutes of ultraviolet (UV) light sterilization between use. All procedures were conducted using aseptic techniques and carried out in a UV-light-sterilized Biohazard hood (MSC12; Jouan SA, Saint Herblain, France).

Agar Diffusion Inhibitory Test

While the restorative specimens (in their molds) were being stored in 100% relative humidity at 37 °C, the microorganisms were spread-plated onto appropriate agars with the recommended growth mediums (MRS agar for *L casei*; Tryptic Soy Agar with 5%

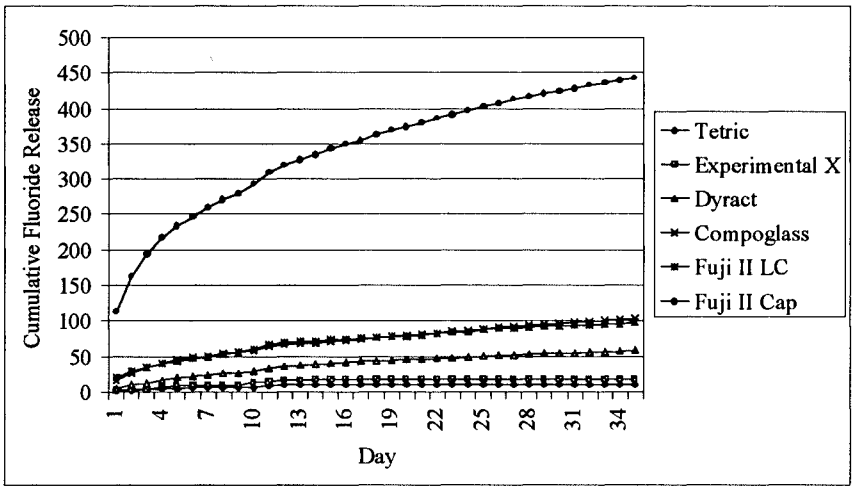

Figure 1. *Cumulative fluoride release over 35 days in ng/mm²*

sheep blood for *S mutans* and *S sobrinus*). Bacteria culture (0.1 ml) was pipeted from the bacteria broth and placed in the center of each plate. The bacterial culture was then spread evenly throughout the plate using a sterilized spreader. After the 1-hour storage period, the specimen disks were removed from their molds and placed onto the newly spread agar plates. The specimen disks were applied, three to each plate (i e, three disks of different materials per plate), to avoid congestion. The bacterial plates, with the specimen disks, were then incubated in their optimal growth conditions (as for the broth cultures) for 48 hours. After the incubation period, the plates were evaluated and the zones of microbial inhibition (inhibition halo effect) measured in millimeters by one examiner and confirmed by a microbiologist.

The specimen disks were subsequently removed aseptically from the bacterial plates and rinsed with sterilized deionized water to remove any attached bacteria. Each disk was then stored in 9 ml of sterilized deionized water for 24 hours. After 24 hours, each disk was rinsed with 1 ml of sterilized deionized water and transferred to a new medium of 9 ml sterilized deionized water as described for fluoride release. This process was continued daily until the next antibacterial testing, which was performed on fresh plates of microorganisms. The agar diffusion inhibitory test was done 1 hour after mixing/light polymerization (baseline), week 1 (day 7), week 2 (day 14), week 3 (day 21), week 4 (day 28), and week 5 (day 35) after specimen fabrication. To determine that fluoride had been leached out from the specimen disk into the medium, the areas of agar beneath the specimen disks at baseline were removed with a sterilized scalpel for fluoride analysis. The agar was mashed with 4 ml of deionized water and filtered. Deionized water (5 ml) was used to wash down the agar solution. The filtrate was collected and analyzed for fluoride using ion chromatography (Waters 510 Ion Chromatograph, Waters Corp).

RESULTS

Fluoride Release

The cumulative fluoride release of the various materials over 35 days is shown in Figure 1. Table 2 reflects the total fluoride release over 35 days and the mean daily fluoride release for the various weeks. The total weekly fluoride release is shown in Figure 2 and the results of the statistical analysis are reflected in Table 3. Ranking of materials from least to greatest total fluoride release over 35 days is as follows: Tetric < Experimental X < Dyract < Fuji II LC < Compoglass < Fuji II Cap. The conventional glass-ionomer cement had significantly greater fluoride release compared to all other materials evaluated. The resin-modified glass-ionomer cement and polyacid-modified resin composites released significantly more fluoride than the fluoride-releasing composites. When the two polyacid modified composite resins were compared, Compoglass exhibited significantly more fluoride release than Dyract.

When the total weekly release was compared, the conventional glass-ionomer cement again had significantly greater fluoride release than the other materials for weeks 1 to 5. The fluoride release of fluoride-releasing composites appeared to stop by week 2. For all materials, a progressive decrease in mean weekly fluoride release was noted.

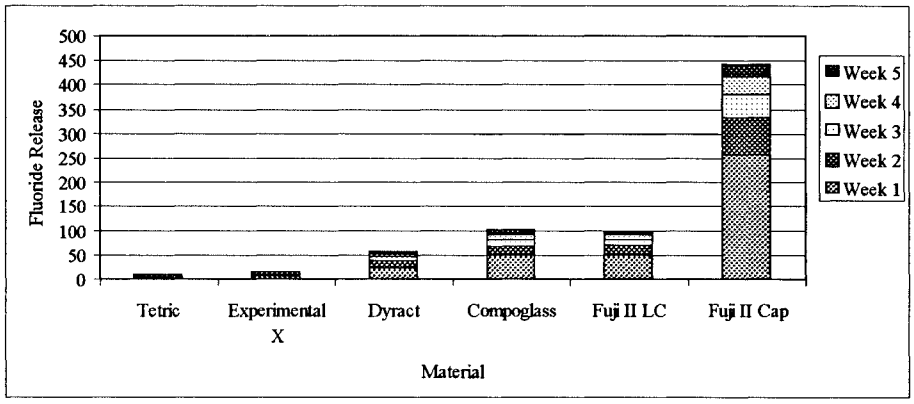

Figure 2. *Total weekly fluoride release in ng/mm²*

Table 3. Results of Statistical Analysis

Time	Differences
Total fluoride release over 35 days	Tetric, Experimental X, Dyract, Compoglass, Fuji II LC < Fuji II Cap Tetric, Experimental X < Dyract, Compoglass, Fuji II LC Dyract < Compoglass
Week 1 Total weekly fluoride release	Tetric, Experimental X, Dyract, Compoglass, Fuji II LC < Fuji II cap Tetric, Experimental X < Dyract, Compoglass, Fuji II LC Dyract < Compoglass
Week 2 Total weekly fluoride release	Tetric, Experimental X, Dyract, Compoglass, Fuji II LC < Fuji II Cap Tetric < Dyract, Compoglass, Fuji II LC Experimental X < Fuji II LC
Week 3 Total weekly fluoride release	Tetric, Experimental X, Dyract, Compoglass, Fuji II LC < Fuji II Cap Tetric, Experimental X < Dyract, Compoglass, Fuji II LC Dyract < Compoglass, Fuji II LC
Week 4 Total weekly fluoride release	Tetric, Experimental X, Dyract, Compoglass, Fuji II LC < Fuji II Cap Tetric, Experimental X < Dyract, Compoglass, Fuji II LC Dyract < Compoglass
Week 5 Total weekly fluoride release	Tetric, Experimental X, Dyract, Compoglass, Fuji II LC < Fuji II Cap Tetric, Experimental X < Dyract, Compoglass, Fuji II LC Dyract < Compoglass

Results of Kruskal-Wallis and Mann-Whitney Rank Sum Test ($P < 0.05$); < denotes significant difference in fluoride release.

Agar Diffusion Inhibitory Test (ADT)

The results of the agar diffusion inhibitory tests are shown in Table 4. The mean fluoride amounts detected in the agar beneath the specimen disks at baseline are shown in Table 5. The IRM control was found to inhibit the growth of all three microorganisms. None of the restorative materials evaluated affected the growth of *L casei*, *S sobrinus,* or *S mutans* at any time period including baseline, where fluoride was detected in the agar beneath the specimen disks. There was no correlation noted between fluoride-release potential and antibacterial properties.

DISCUSSION

Fluoride Release

Fluoride release from restorative materials is a complex process involving several phases, such as water diffusion into the material, dissolution of exchange of fluoride in the solid, and diffusion of fluoride ions out of the material. In this in vitro study, the fluoride release up to 35 days was assessed. This period was chosen because fluoride-releasing composites cause a substantial caries reduction related to fluoride release after 28 days (Dijkman & Arends, 1992). A relatively small specimen size was chosen to simulate the clinical dimensions of class 5 restorations. The experimental setup does not simulate the clinical situation correctly but gives an indication of the maximum amount of fluoride release possible; i e, fluoride release potential. In the mouth, fluoride is probably not washed away as completely as in this experiment. Biofilms and layers (e g, plaque) on the surface of restorations may reduce fluoride release, and part of the released fluoride will be accumulated in these layers. In addition, there is the possibility of an uptake of part of the released fluoride back into the restorative materials (Tam, Chan & Yim, 1997). Fluoride release may, however, be increased when the pH drops in plaque. Furthermore, fluoride release in water has been reported to differ from that in artificial saliva in that different products behave in different ways (Wandera, Spencer & Bohaty, 1996). Deionized water was selected for this experiment as more fluoride is released in deionized water than in artificial saliva (El-Mallakh & Sarkar, 1990).

The high release of fluoride by the conventional glass-ionomer cement, Fuji II Cap, observed during the first 24 hours is consistent with previous reports (Crisp, Lewis & Wilson, 1980) on the high early erosion of glass ionomers. This same trend was observed for the resin-modified glass-ionomer cement Fuji II LC and the polyacid-modified resin composite Compoglass. The amount of fluoride released was, however, much lower. The lower 24-hour fluoride release of Fuji II LC may be attributed to the occurrence of the photochemical reaction, which reduces early sensitivity to moisture (Wilson, 1990). The resin network could also reduce the diffusion of water into cement, thus reducing the elution of unbound fluoride in the material matrix (Mathis & Ferracane, 1989). Although Compoglass has been classified as a polyacid-modified resin composite or compomer, its fluoride release pattern is similar to that of conventional and resin-modified glass-ionomer cement—a large initial release of fluoride that was many times that released later. The mean weekly fluoride release by Fuji II Cap, Fuji II LC, and Compoglass had dropped by more than 50% in week

Table 4. Zone of Growth Inhibition in Millimeters (mm)

Material	Baseline 1 Hour (SD)	Week 1 (SD)	Week 2 (SD)	Week 3 (SD)	Week 4 (SD)	Week 5 (SD)
IRM		NA	NA	NA	NA	NA
L casei	9.85 (1.08)					
S sobrinus	12.00 (0.00)					
S mutans	10.00 (0.00)					
Tetric						
L casei	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S sobrinus	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S mutans	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
Experimental X						
L casei	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S sobrinus	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S mutans	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
Dyract						
L casei	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S sobrinus	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S mutans	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
Compoglass						
L casei	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S sobrinus	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S mutans	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
Fuji II LC						
L casei	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S sobrinus	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S mutans	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
Fuji II Cap						
L casei	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S sobrinus	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
S mutans	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)

2 and continued to decrease during the period of investigation. The pattern of release observed in this experiment was consistent with the results of other studies on conventional and resin-modified glass ionomers (DeSchepper & others, 1991; de Araujo & others, 1996). The clinical significance of this early release has not yet been clarified. A strong initial fluoride effect may inhibit bacterial activity and arrest the process in unintentionally left carious dentin. Furthermore, it may be expected that the fluoride will activate the remineralization of uninfected inner dentin and demineralized enamel. The polyacid-modified resin composite Dyract and the fluoride-releasing resin composites Tetric and Experimental X exhibited a more gradual release of fluoride. The flouridating composites, however, stopped releasing fluoride by week 2, while Dyract continued to release fluoride during the entire duration of the experiment.

When the materials were compared, the conventional glass-ionomer cement Fuji II Cap had significantly higher total fluoride release over 35 days than all the other materials evaluated, including the resin-modified glass-ionomer cement Fuji II LC. The total weekly fluoride release of Fuji II Cap for weeks 1 to 5 was also significantly greater. It is apparent that resin modification of the glass ionomer had resulted in a decrease in fluoride release. This finding is in agreement with other studies (Tam & others, 1997; Wandera & others, 1996), which found generally greater fluoride release from conventional glass ionomers and equivalent or less from resin-modified glass ionomers. The lower fluoride release may be attributed to decreased early erosion of glass ionomers due to resin-modification. It is, however, important to note that differences in fluoride release between different types of glass ionomers vary as a function of the media in which they are stored (Wandera & others, 1996).

Table 5. Mean Fluoride (ng/mm²) Detected in Agar beneath Specimen Disks at Baseline

Materials	Mean Fluoride (SD)
Tetric	0.61 (0.49)
Experimental X	1.04 (0.49)
Dyract	0.72 (0.82)
Compoglass	1.02 (0.19)
Fuji II LC	0.71 (0.60)
Fuji II Cap	1.08 (0.37)

When the two polyacid-modified resin composites were compared, Compoglass was found to have a significantly higher fluoride release. The total fluoride release after 35 days and pattern of fluoride release for Compoglass were comparable to that of the resin-modified glass ionomer Fuji II LC. In typical polyacid-modified composites, the functional groups of polyacrylic acid and methacrylates are combined into one molecule. Light curing results in a setting process analogous to that of resin composites. Subsequent water sorption leads to an acid-base reaction, resulting in a partially ionic structure within the polymeric matrix. This results in fluoride release in a similar manner to that of glass ionomers. Compoglass, however, releases fluoride from two independent sources: the glass filler (barium-aluminum-fluorosilicate glass) and the fluoride-releasing filler (ytterbium trifluoride). The single-component adhesive used with Compoglass provides another source of fluoride release (ammonium fluoride), which was not assessed in our experiment. The addition of fluoride-releasing fillers may explain in part the difference in fluoride released between the two polyacid-modified resin composites.

Three different approaches to the development of fluoride-releasing composites have been reported. These are: addition of water-soluble fluoride salts, matrix-bound fluoride, and fluoride-releasing filler systems (Arends & others, 1995). The first approach is not ideal, because soluble fluoride salts easily wash out, causing a porous structure, which can compromise the physical and mechanical properties of the composite. The second group of materials has been intensely investigated, and fluoride-releasing resin systems are now used in some commercial composites. The fluoride-releasing filler system approach has been adopted by most commercially available fluoride-releasing composites, as in the case of the composites chosen for this experiment. The major advantage of these systems is that adequate physical and mechanical properties can be preserved despite substantial fluoride release (Danielson, 1990). Two filler types can be distinguished, namely: very sparingly soluble compounds such as ytterbium trifluoride (YbF_3) and leachable glass fillers. The authors do not know what type of fluoride-releasing filler Experimental X uses. Tetric, in which the radiopaque fluoride-releasing filler YbF_3 is used, most likely releases fluoride by means of an exchange mechanism (Arends & others, 1995). Water diffuses into the composite, reaches the YbF_3 filler, and causes fluoride release according to the reaction $YbF_3 + 3OH^- \leftrightarrow Yb(OH)_3 + 3F^-$. The fluoride ion diffuses out of the composite in the direction of the lowest fluoride concentration. As the existence of a concentration gradient is the actual driving force for fluoride release, it would be expected that the amount of fluoride release would decrease with time due to the diminishing gradient, as fluoride is leached out from the composite. This explains the decline of fluoride release observed in this experiment. The fluoride concentration gradient after 14 days may be too low to facilitate fluoride ion diffusion from the composite into the medium.

Agar Diffusion Inhibitory Test (ADT)

Many investigations have been carried out on the antibacterial actions of dental restoratives. Information in the area of antibacterial restorative materials in general has been limited both in the number of materials and variety of microorganisms tested. In addition, few attempts have been made to correlate fluoride-release potential and antibacterial properties, which was done in this experiment. One evaluation scheme included the use of acidified gel systems. Such systems have produced in vitro secondary caries around glass-ionomer restorations (Dérand & Johansson, 1984; Kidd, 1978). Acidified gels have limitations because they do not mimic well the dynamics of actual microbial growth and plaque formation. Alternative in vitro testing methods would include direct inhibition of microbial growth in the agar diffusion method, broth suspension, or through the use of a bacterial adherence assay system. The strength of these systems is that a decrease of microbial growth or activity could be directly determined. The microorganisms used in this project are cariogenic in humans (Hardie, 1992) and thought to be indicative of progressive lesions (Boyar & Bowden, 1985). There are, however, several problems associated with the agar diffusion inhibitory test (ADT). The greatest disadvantage is that it does not distinguish between bacteriostatic and bacteriocidal properties of dental materials, nor does it provide any information about the viability of the test bacteria.

IRM was used as a baseline control for ADT because zinc oxide is strongly antiseptic and, in combination with eugenol, has long-lasting, highly bactericidal properties (Turkheim, 1953). The results of this study showed that none of the restoratives evaluated possessed antibacterial activity. This was the case at baseline and all weekly time periods. The baseline experiment was repeated twice, with new batches of specimens, to confirm negative findings. No zones of inhibition were found for any of the restoratives, with the exception of IRM, for the second and third baseline experiment. Results appear to contradict those of Scherer and others (1989), who found that glass-ionomer cement materials produced measurable zones of inhibition. It is important to note that there were great disparities between the zones of inhibition produced by the different glass ionomers evaluated in their experiment. The glass ionomers, which do not contain zinc oxide, had minimal zones of inhibition compared to those that did. Furthermore, no zones of inhibition were found for Lactobacilli with the restorative glass ionomers tested. They concluded that the zones of bacteria inhibition produced with glass-ionomer cements might be attributed to the zinc oxide component of the material in addition to their fluoride-leaching ability. None of the restoratives evaluated in the present study had zinc oxide as a component. This, together with the fact that the restoratives were not allowed to set on the bacteria plates, may have accounted for the negative results observed. The objective of the experiment was to test the fluoride-release and antibacterial properties of the material and not their unreacted components. The hypothesis is supported by the work of Turkheim (1953), who found that most dental materials proved to be bactericidal while setting, i e, so long as a chemical reaction was proceeding. The low pH level of glass-ionomer cements while setting may contribute more to their antibacterial properties than their fluoride—leaching capabilities. This is supported by the fact that there was no antibacterial activity at baseline, despite the presence of fluoride in the agar beneath the specimen disks.

The results showed that there is no correlation between fluoride-release potential and antibacterial activity. Fluoride inhibition of carbohydrate metabolism by acidogenic plaque microflora is well established. Fluoride has a dual action of dissipating proton gradients and preventing their generation through its action on H^+/ATPase. The collapse of transmembrane proton gradients, in turn, reduces the ability of cells to transport solutes via mechanisms involving proton motive force. In spite of these known effects on bacterial cells, there is no general agreement that the antibacterial effects of fluoride contribute to the anticaries effect of fluoride (Hamilton, 1990). The critical amount of fluoride released from a restorative material to be effective in inhibiting caries has not yet been established and warrants investigation.

CONCLUSIONS

Under the conditions of this in vitro study:

Fuji II Cap had significantly greater fluoride release than all other materials evaluated.

Fuji II Cap, Fuji II LC, and Compoglass had similar patterns of fluoride release characterized by a high initial release that was many times that released later.

The fluoride-releasing composites stopped releasing fluoride by 14 days.

There was no correlation noted between fluoride-release potential and antibacterial properties.

Acknowledgments

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Polymerization Color Changes of Esthetic Restoratives

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Clinical Relevance
All materials evaluated undergo color changes during polymerization. Therefore, the clinical practice of polymerizing some material on, or adjacent to, the undried tooth to confirm shades of esthetic restoratives before restorative procedures is prudent.

SUMMARY

The color changes of three different types of tooth-colored restoratives during polymerization were investigated using colorimetry. L*, a*, b* color parameters of five different shades of Z100 (a mini-filled composite resin), Fuji II LC (a resin-modified glass-ionomer cement), and Dyract (a polyacid-modified composite resin) were taken precure and postcure. The results showed that the restoratives evaluated all underwent color changes during polymerization. The polymerization changes in color parameters were shade and not material dependent. Changes in L* parameter or lightness during polymerization were significant for all material and shade combinations and had the greatest influence on the overall polymerization color change. As the color change was perceivable by the human eye for most shades of materials, the clinical practice of polymerizing some material on, or adjacent to, the undried tooth to confirm shades of esthetic restoratives before restorative procedures is prudent.

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INTRODUCTION

Glass-ionomer cements and light-polymerized composite resins were developed as esthetic tooth-colored restorative materials in the 1970s. The recent introduction of light-polymerized resin-modified glass-ionomer cements and polyacid-modified composite resins now provide a whole spectrum of materials that combine the technologies of composites and glass ionomers.

Shade selection for these restoratives is frequently difficult, despite the increasing number of available shades. The majority of these tooth-colored materials are keyed to the Vita Lumin shade guide (Vita Zahnfabrik, Bad Säckingen, Germany), which is widely

used in porcelain shade selection. Such a practice reduces the need for different shade guides and improves interpractitioner communication. However, since none of the materials matches the Vita shade guide to which they are supposedly keyed (Yap, Tan & Bhole, 1997), Vita shade tabs are not recommended for clinical shade selection. Instead, the manufacturers' shade guides or a customized shade guide of the actual restorative material is recommended. The latter is not usually practical for conventional and resin-modified glass-ionomer cements due to material dehydration and subsequent color change when stored in a dry environment (Walls, 1986; Sidhu & Watson, 1995).

The variance of shades and the low reliability of shade guides (Wozniak & others, 1985) point to the importance of polymerizing some material on, or adjacent to, undried teeth to confirm shades of esthetic restoratives. Although polymerized composites are easy to remove from unetched teeth, polymerized resin-modified glass ionomers can achieve good bonding to enamel and dentin (Sidhu & Watson, 1995) and are harder to remove. The technique of polymerizing material on teeth also entails the use of materials, which has cost as well as clinical time implications. Initial shade matching of the uncured restorative material to the tooth is thus important, and once an acceptable shade is obtained, the retention of color match should ideally be maintained after the curing process is completed. Seghi, Gritz, and Kim (1990) evaluated the color of nine light-polymerized composites immediately before and 10 minutes after light curing and found a characteristic chromatic shift toward the blue-green region of color space. This resulted in a perceived decrease in yellow chroma. As a compensatory measure, they suggested an initial color choice more yellow or more chromatic than the desired final color. Eldiwany, Friedl, and Powers (1995) studied the changes in optical properties when curing resin composites. Light polymerization was found to cause color changes that were rated barely perceptible to perceptible for the composites studied. Once the composites were light polymerized, postcuring caused no further perceptible changes in shade.

Although the polymerization color changes of composite resins have been extensively studied, the effects of light polymerization on the color of resin-modified glass ionomers and polyacid-modified composite resins have not been widely investigated. Since restorative material formulations continually change, it was postulated that newer moieties used for new-generation restorative materials would be more color stable during curing. The objective of this study was to determine the polymerization color changes of different types of esthetic restorative materials. The variability in color changes among different types of restorative materials and shades was also evaluated.

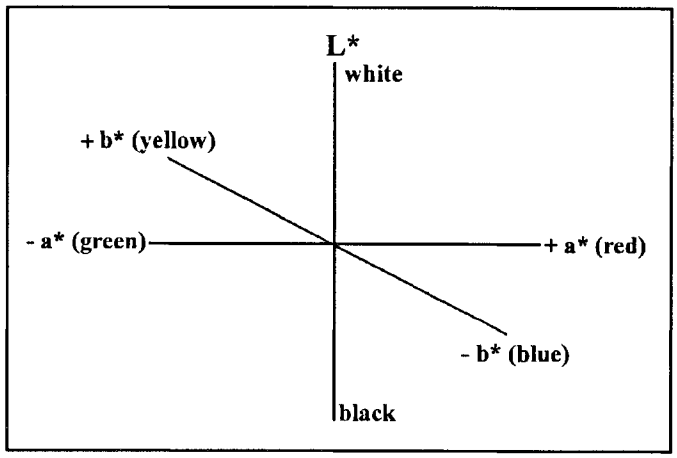

Figure 1. *Diagram of the CIELAB color space*

METHODS AND MATERIALS

Three different types of restorative materials representing the new spectrum of commercially available esthetic restorative materials were selected. These included Z100 (3M Dental Products, St Paul, MN 55144), a mini-filled composite resin; Fuji II LC (GC, Tokyo, Japan), a resin-modified glass-ionomer cement; and Dyract (De Trey/Dentsply, Surrey, England), a polyacid-modified composite resin or compomer. Common shades for these materials were selected so that intermaterial comparison could be done. The shades were Vita A2, A3, A4, B3, and C5.

Special grid molds were used for preparation of square specimens, which were 7 mm wide and 1.5 mm thick. Capsulated Fuji II LC was mixed for 10 seconds and placed directly into the molds. Z100 and Dyract were dispensed from syringes and compules respectively. The molds were slightly overfilled with material under evaluation and then sandwiched between two glass plates to extrude the excess material. A small-area (3 mm in diameter) colorimeter (Dental Colorimeter; Minolta Camera Co Ltd, Tokyo, Japan) was then used to determine the precure CIELAB coordinates of each specimen. The CIE color system (CIELAB) was determined by the International Commission on Illumination in 1978. The three attributes of color in this system are L*, a*, and b*, where L* is the lightness variable proportional to Value in the Munsell system, and a* and b* are chromaticity coordinates (Figure 1). The a* and b* coordinates designate positions on a red/green and yellow/blue axis respectively (+a = red, -a = green; +b = yellow, -b = blue).

Table 1. Mean L and ΔL Values for the Various Materials*

Shade & Material	Mean Precure L* Values (SD)	Mean Postcure L* Values (SD)	Mean DL Values (SD)
Shade A2			
Z100#	73.86 (0.82)	71.89 (0.80)	1.97 (0.33)
Fuji II LC#	77.53 (0.44)	72.42 (0.74)	5.11 (0.47)
Dyract#	72.13 (0.60)	69.24 (0.66)	2.89 (0.67)
Shade A3			
Z100#	72.80 (0.45)	70.60 (0.28)	2.20 (0.41)
Fuji II LC#	76.33 (0.69)	71.95 (0.98)	4.38 (1.05)
Dyract#	69.91 (1.19)	67.21 (0.98)	2.70 (1.33)
Shade A4			
Z100#	69.46 (0.74)	67.13 (0.74)	2.33 (0.34)
Fuji II LC#	70.72 (2.23)	64.98 (1.21)	5.74 (1.41)
Dyract#	66.06 (0.83)	62.61 (0.80)	3.45 (0.24)
Shade B3			
Z100#	72.70 (1.01)	70.18 (0.76)	2.52 (0.53)
Fuji II LC#	75.90 (0.65)	70.52 (0.91)	5.38 (1.29)
Dyract#	68.98 (1.38)	66.88 (0.72)	2.10 (1.19)
Shade C5			
Z100#	62.11 (0.49)	58.68 (0.67)	3.43 (0.69)
Fuji II LC#	68.09 (2.63)	62.33 (1.10)	5.76 (2.31)
Dyract#	65.27 (1.24)	62.74 (0.87)	2.53 (0.74)
All Shades			
Z100			2.49 (0.68)
Fuji II LC			5.27 (1.47)
Dyract			2.73 (0.99)

= statistically significant differences in pre- and postcure values (results of Mann-Whitney test at a significance level of 0.05).

Illumination corresponding to "average" daylight (CIE illuminant D65) from a pulsed xenon light source was used. The colorimeter was calibrated before each measurement session using the white calibration tile supplied by the manufacturer. After the precure L* a* b* values were obtained, the specimens were immediately light polymerized according to the exposure time recommended by the manufacturers. No overlapping irradiation was necessary, as the diameter of the exit window of the light source was larger than the specimen. Postcure readings were then taken with the colorimeter. All readings and light-curing procedures were done through the upper glass plate with a standardized white background below. Ten specimens were evaluated for each material and shade combination. The precure and postcure L* a* b* values were averaged to obtain a single set of values for each material and shade using SPSS for Windows version 7.5 (SPSS Inc, Chicago, IL 60611).

Significant differences between precure and postcure L*, a*, and b* values for the various materials and shade combinations were determined by the Mann-Whitney U Test at a significance level of 0.05. The polymerization color change (ΔE) was calculated using the equation:

$$\Delta E = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2}$$

ΔL, Δa, and Δb are the mathematical differences between precure L* a* b* and postcure L* a* b* values. These and ΔE values were subsequently used to compare the change in color parameters between shades and materials. Statistical analysis was done using Kruskal-Wallis and Mann-Whitney tests at a significance level of 0.05.

RESULTS

The mean precure L*a*b*, postcure L*a*b*, ΔL, Δa, and Δb values are reflected in Tables 1 to 3. The mean ΔE values for the various materials and shades are shown in Table 4 and Figure 2. There were significant differences between precure and postcure L* values for all materials and shades. Fuji II LC had significant differences in precure a* and postcure a* values for shades A2, A4, and C5. There was also a significant difference in a* values for Z100 and Dyract for shades B3 and C5 respectively. Four out of five shades tested for Dyract had significant differences between precure b* and postcure b* values. These shades were A2, A3, A4, and B3. For shade C5, both Z100 and Fuji II LC had a significant change in b* values after light polymerization.

There were significant differences in ΔL, Δa, and Δb values among the different shades evaluated for all three restoratives. The ΔE values among the different shades were also significant for Z100 and Dyract. There was, however, no significant difference in ΔE values among the various shades for Fuji II LC.

Comparison of ΔL, Δb, and ΔE values by shades revealed significant differences among materials. Significance differences in polymerization color change (i e, ΔE) of materials generally followed that of ΔL values. This was expected, as the changes in L* values after light polymerization were usually greater than that of a* and b*. For shades A2, A3, and A4, Z100 had significantly smaller ΔE than Fuji II LC and Dyract. For shade B3 both Z100 and Dyract had significantly smaller ΔE than Fuji II LC. For shade C5, Dyract had the least polymerization color change. ΔE of Dyract was significantly smaller than Z100 and Fuji II LC. Only two out of five

shades evaluated showed significant differences in Δa values. These were shades A2 and A4, where Z100 had significantly greater Δa compared to Fuji II LC. For shades A2, A3, and A4, Z100 and Fuji II LC had significantly smaller Δb values than Dyract. For shade B3 and C5, only Z100 had significantly smaller Δb values than Dyract. Results suggested that polymerization changes in color parameters are shade and not material dependent.

Table 2. Mean a and Δa Values for the Various Materials*

Shade & Material	Mean Precure a* Values (SD)	Mean Postcure a* Values (SD)	Mean Δa Values (SD)
Shade A2			
Z100	-3.03 (0.90)	-2.68 (1.18)	-0.35 (1.15)
Fuji II LC#	-0.18 (0.67)	1.28 (0.39)	-1.46 (0.94)
Dyract	-2.60 (1.26)	- 1.74 (1.13)	-0.86 (1.87)
Shade A3			
Z100	-0.30 (0.67)	-0.10 (0.64)	-0.20 (0.69)
Fuji II LC	2.20 (1.22)	2.84 (1.13)	-0.64 (0.51)
Dyract	-1.64 (1.04)	-1.18 (1.37)	-0.46 (1.33)
Shade A4			
Z100	-0.70 (0.80)	-0.28 (0.77)	-0.42 (0.74)
Fuji II LC#	3.19 (1.13)	4.65 (1.72)	-1.46 (0.88)
Dyract	-0.43 (1.29)	0.88 (1.60)	-1.31 (1.30)
Shade B3			
Z100#	-1.67 (0.59)	-0.55 (0.94)	-1.12 (0.56)
Fuji II LC	-0.14 (0.83)	0.75 (1.38)	-0.89 (1.33)
Dyract	-0.91 (1.21)	-0.50 (0.63)	-0.41 (1.14)
Shade C5			
Z100	-0.08 (1.05)	-0.08 (1.34)	0.00 (1.38)
Fuji II LC#	-0.32 (0.83)	0.37 (0.43)	-0.69 (0.89)
Dyract#	-0.58 (0.68)	0.30 (0.48)	-0.88 (0.92)
All Shades			
Z100			-0.42 (0.99)
Fuji II LC			-1.03 (0.98)
Dyract			-0.78 (1.33)

= statistically significant differences in pre- and postcure values (results of Mann-Whitney test at a significance level of 0.05).

DISCUSSION

All the restorative materials evaluated, regardless of shade, had a significant decrease in L* value after polymerization. The materials were thus significantly darker after polymerization. The range for ΔL was as small as 1.97 for Z100 A2 shade to as large as 5.76 for Fuji II LC shade C5. For all material and shade combinations, changes in the lightness variable (ΔL) or value was greater than for chromaticity (Δa and Δb) and had the greatest influence on polymerization color change (i e, ΔE). For all shades, Fuji II LC had the greatest difference between precure and postcure L* values. Selection of a prepolymerization shade that is lighter than the desired final shade is thus recommended for the materials evaluated. The values for a* were generally small because the color of teeth, to which materials were matched, had little red-green component. There was no significant difference change in a* values after polymerization for most shades of Z100 and Dyract. The exceptions were B3 for Z100 and C5 for Dyract. Fuji II LC, however, had more shades with significant changes in a* between precure and postcure. The shades with significant a* changes were A2, A4, and C5. Polymerization change in a* values were therefore shade and not material dependent. There was a general shift towards the red region of the color space for all three materials. With the exception of shade C5, both Z100 and Fuji II LC had no significant difference between precure and postcure b* values. Dyract had significant decreases in b* values after polymerization for four out of five shades. This meant that Dyract generally tended to become less yellow after polymerization. Seghi and others (1990) also reported a characteristic chromatic of resin-based materials towards the blue region of the color space, resulting in perceived decrease in yellow chroma after polymerization.

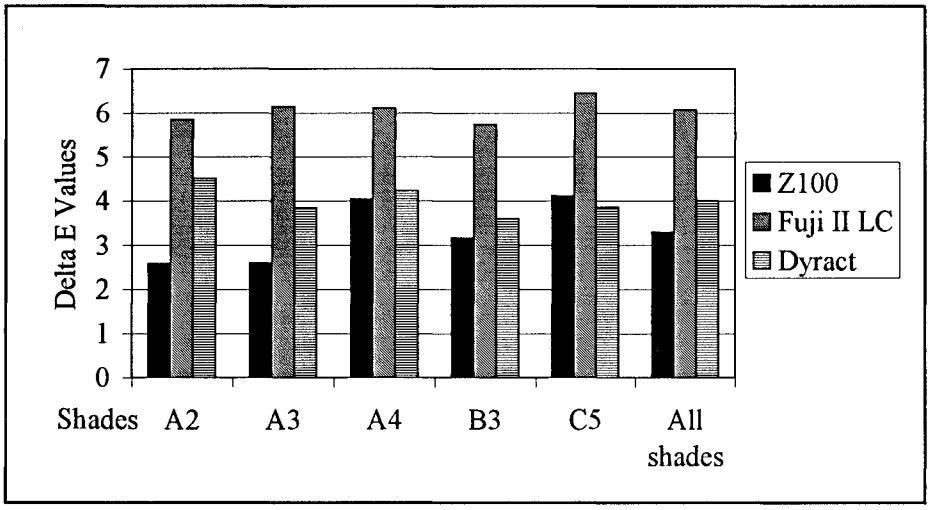

Figure 2. *Mean polymerization color change for the various materials*

Table 3. Mean b and Δb Values for the Various Materials*

Shade & Material	Mean Precure b* Values (SD)	Mean Postcure b* Values (SD)	Mean Δb Values (SD)
Shade A2			
Z100	14.90 (.088)	14.38 (1.92)	0.52 (1.30)
Fuji II LC	14.64 (1.16)	13.53 (2.32)	1.11 (2.17)
Dyract#	18.44 (0.66)	15.53 (0.82)	2.91 (0.75)
Shade A3			
Z100	16.73 (1.30)	16.95 (0.83)	-0.22 (1.22)
Fuji II LC	20.05 (2.88)	19.37 (2.45)	0.68 (5.10)
Dyract#	17.87 (1.12)	15.91 (1.14)	1.96 (1.72)
Shade A4			
Z100	15.13 (4.85)	17.28 (0.98)	-2.15 (4.70)
Fuji II LC	17.37 (0.93)	17.87 (0.68)	-0.50 (1.20)
Dyract#	18.43 (1.42)	17.45 (1.03)	0.98 (1.57)
Shade B3			
Z100	19.54 (0.87)	20.00 (1.32)	-0.46 (1.51)
Fuji II LC	15.98 (0.76)	15.24 (1.03)	0.74 (0.93)
Dyract#	22.26 (2.20)	20.29 (1.58)	1.97 (2.12)
Shade C5			
Z100#	10.75 (1.01)	12.41 (1.14)	-1.66 (0.85)
Fuji II LC#	9.73 (0.78)	11.43 (1.61)	-1.70 (1.30)
Dyract	17.69 (1.06)	18.73 (3.36)	-1.04 (3.57)
All Shades			
Z100			-0.79 (2.48)
Fuji II LC			0.06 (2.73)
Dyract			1.35 (2.47)

= statistically significant differences in pre- and postcure values (results of Mann-Whitney test at a significance level of 0.05).

Selection of a prepolymerized shade that is more yellow (i e, shade with greater chroma) than the desired final shade can thus be recommended for Dyract.

When ΔL, Δa, and Δb of the different shades were compared by material, it was apparent that polymerization change in color parameters were not material dependent. Significant differences existed between different shades of the same material. For example, the difference in ΔL for Z100 ranged from 1.97 for shade A2 to 3.43 for shade C5. Such discrepancies were also noted for Dyract and Fuji II LC where ΔL ranged from 2.10 for shade B3 to 2.53 for shade C5 and 5.11 for shade A2 to 5.76 for shade C5 respectively. Δa and Δb were also significantly different between shades for the different materials. When the polymerization color changes (i e, ΔE) of the different shades were compared, significant differences among shades were only noted for Z100 and Dyract. There was no significant difference in ΔE among the various shades for Fuji II LC as all shades underwent similarly large polymerization color changes.

Comparison of ΔL, Δa, Δb, and ΔE of materials by shades showed that differences among materials were shade dependent. When the data for all shades were pooled, the composite Z100 and polyacid-modified composite Dyract had significantly less polymerization color change than the resin-modified glass-ionomer cement Fuji II LC. When the light source from the colorimeter hit the surface of the material, transmission, absorption, or scattering could occur. As the materials evaluated all consisted of small particles of different refractive indices from the bulk of the material, some light was transmitted, and some was scattered. Composite resins and glass ionomers acquire their color through the addition of minute amounts of pigments, which often consist of the oxides of different metal. So it is conceivable that since these pigments are generally chemically inert, the visible-light-activated polymerization and/or acid-base reaction caused a change of the refractive index of the matrix phase, making the material less translucent due to increased light scattering. As changes of the matrix phase in resin-modified glass-ionomer cements are more substantial and dynamic after light polymerization, the polymerization color change is expected to be greater. There is minor postcuring of composites after light polymerization, and even under ideal conditions the conversion rate from monomer to polymer is rarely greater than 60% (Pearson, 1990). This decrease in translucency may in part account for the decreased L* values after polymerization and mixing, as a less translucent material appears darker in color (Combe, 1992).

Photodetectors can measure much smaller color changes than those detectable by the human eye.

Table 4. Mean ΔE Values for the Various Materials

SHADES	MATERIALS		
	Z100	**Fuji II LC**	**Dyract**
A2	2.58 (0.68)	5.84 (0.80)	4.51 (1.16)
A3	2.59 (0.45)	6.13 (2.80)	3.84 (1.65)
A4	4.04 (3.98)	6.11 (1.40)	4.24 (0.62)
B3	3.16 (0.66)	5.73 (1.21)	3.60 (1.51)
C5	4.12 (0.58)	6.45 (1.49)	3.86 (2.61)
All Shades	3.30 (1.91)	6.06 (1.63)	4.01 (1.61)

Change is perceptible to the human eye when the value of $\Delta E \geq 3.3$.

Ruyter, Nilner, and Moller (1987) showed that under clinical settings the human eye can sense ΔE values of 3.3 or greater. Under controlled environments, the human eye could perceive changes of color between ΔE of 1 and 2 (Seghi, Johnston & O'Brien, 1986; Seghi, Hewlett & Kim, 1989). Based on a ΔE value of 3.3 as the lower perceptibility for color changes, all shades for Fuji II LC and Dyract would have clinically detectable polymerization color changes. For Z100, only the darker shades A4 and C5 would have clinically detectable color changes. The clinical practice of polymerizing some material on, or adjacent to, the undried tooth to confirm shades of esthetic restoratives before restorative procedures is therefore prudent. The long-term color stability of these materials postcure warrants further investigation.

CONCLUSIONS

All tooth-colored restoratives evaluated underwent polymerization color changes. The changes in color parameters after polymerization were shade and not material dependent. Changes in L* parameter or lightness during polymerization were significant for all material and shade combinations and had the greatest influence on the overall polymerization color change. Based on a ΔE value of 3.3 as the lower perceptibility for color changes, all shades for Fuji II LC and Dyract would have clinically detectable polymerization color changes. For Z100, only the darker shades A4 and C5 would have clinically detectable color changes. The clinical practice of polymerizing some material on, or adjacent to, the undried tooth to confirm shades of esthetic restoratives before restorative procedures is therefore prudent.

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Digital Radiology and Image Analysis for Approximal Caries Diagnosis

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Clinical Relevance
Radiovisiography proved useful in the diagnosis of simulated early incipient approximal caries.

SUMMARY

The aim of this study was to determine if radiology combined with digital reception and image analysis, radiovisiography (RVG), is effective in the early diagnosis of simulated approximal carious lesions. An experimental lesion similar to the one produced by caries was made in 39 permanent molars and premolars. Radiographic images of these lesions were obtained under standardized conditions using RVG. The image obtained was magnified 700 times and the densities of the lesion, the enamel, the dentin, and the pulp were measured using an image analysis program (Visualdent). The results showed statistically significant differences between the density of the produced lesions and that of healthy enamel. This measurement was independent of the differing thickness of the approximal enamel surface in molars and premolars. These results indicated the potential usefulness of the system tested in the diagnosis of incipient approximal caries.

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INTRODUCTION

Radiographic imaging has become a viable and useful aid in the diagnosis of approximal caries. Different methods of improving this medium are continually being researched. Some of these are based on videographic recording of radiographic images in order to then obtain a digitized image, which can be manipulated using a conventional computer (Verrier & others, 1989). These systems have proven useful in the diagnosis of occlusal carious lesions, as they are capable of diagnosing a greater number of dentin lesions, without raising false positives, than other conventional diagnostic procedures such as radiology, xeroradiography, or visual examination (Wenzel, Larsen & Fejerskov, 1991). Similarly, the

diagnosis of approximal caries may be improved through the digitization of radiographic images (Heaven, Weems & Firestone, 1994; Duncan & others, 1995).

A significant improvement in caries diagnosis occurred with the introduction of digital image reception systems called radiovisiography (RVG) (Mouyen & others, 1989). RVG claims the fundamental advantages of speed, decreased radiation dose, and the absence of factors such as correct illumination. Minimal image distortion is experienced with RVG, and its use in endodontics has been described. Also, improvements for the system have been suggested (Horner & others, 1990).

The brightness and contrast of images obtained with RVG can be modified, and they may be magnified or used with various filters. However, each of these operations must be specifically selected for each distinct diagnostic situation (Dunn & Kantor, 1993). When conventional radiographs of approximal carious lesions are compared to the results of the basic mode RVG, they show the same specificity but slightly reduced sensitivity (Russell & Pitts, 1993). Continuous technological advances in this field of digital diagnostic systems and their association with image analysis programs show promise for further research.

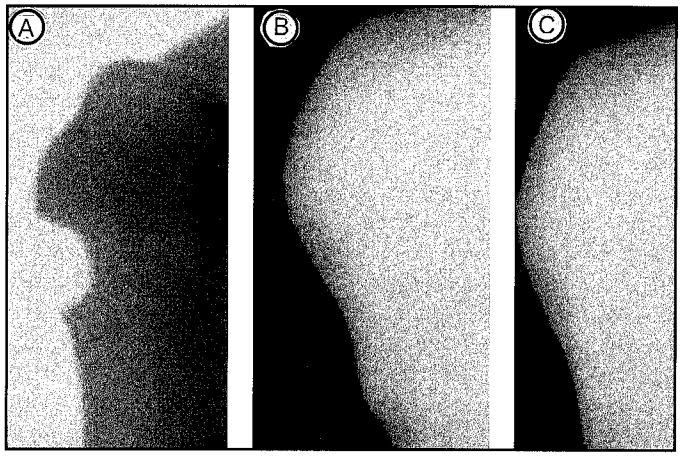
The purpose of this study was to determine if a digital image analysis software that is usually integrated into a RVG unit could effectively differentiate between the radiographic density of an experimentally produced lesion that simulates caries and the healthy dental structures surrounding it.

METHODS AND MATERIALS

A sample of 39 caries-free posterior teeth consisting of 18 premolars and 21 permanent molars were used. They were extracted due to periodontal or orthodontic reasons and were subsequently stored in a buffered 10% formalin solution prior to their use. Approximofacial and approximolingual angles were carefully examined for the presence of any enamel developmental defect or irregularity that could lead to diagnostic error by being confused with a cavitated lesion. Two cavitated lesions, similar to those caused by early caries, were made on each tooth; one of these lesions was in the mesial surface and the other in the distal one. Both were equidistant from the facial and lingual surfaces.

In order to simulate a minimal carious lesion (minimal cavitation) in the best way possible, a very small cavity was made with a high-speed handpiece with a Komet 009 diameter (code 806 314. 8804; Komet Dental/Brasseler, Lemgo, Germany) (figure). This bur incorporates a stop that impedes excessive depth beyond the stop in the cut produced. The resultant preparations were almost conical in shape, with the vertex directed towards the interior of the tooth, similar to those caused by caries. A new bur was used for each cavity preparation to achieve the same efficacy in the cut and therefore obtain a more homogenous cavity size in the lesions. In a pilot study, it was determined that the use of burs with a limited penetration and using them only once produced lesions that were very similar to each other and that were limited to the external half of the enamel layer.

Radiographic images of the lesions produced were obtained using the latest-generation Trophy RVG digital system (Trophy Radiologie, Vincennes-Paris, France). The generator of the X-rays was set at 65 kV (Orix-AET; Ardet, Corsica, Italy). The teeth were placed on the sensor of the RVG unit, covered by a disposable rubber protector, with the main occluso-apical axis parallel to the surface of the sensor. This was achieved using small soft wax supports, which were placed between the apices and the sensor. A dental surveyor was used to keep the X-ray tube perpendicular to the sensor. The distance between the source and the sensor was set at 10 cm. The exposure time was set at 1 second. These values, in the same way as the conditions under which the RVG unit shows the images produced (brightness and contrast), were selected after various tests and were set at the values that provided the best possible results for the conditions of this study. The next step was to magnify (X700) the image in the exact zone of the lesions produced. After fixing the image with the values previously stated, a Visualdent 1.1 image analysis program (Nemotec, Madrid, Spain) was used. One of the functions of this program is to produce image densitometries on a point or points on a determined line.



Representative sample of an experimental defect used as a simulated caries lesion used in the study. **A.** *Cavity produced by the bur* **B.** *RVG of the cavity shown in A* **C.** *Radiograph of the cavity produced in A*

Density in Lesion Zones, Enamel, Dentin, and Pulp

	Total	Premolars	Molars
Total	39	19	20
Lesion			
Mean	3.78	4.07	3.50
SD	0.73	0.72	0.64
Minimum	2.50	3.00	2.50
Maximum	5.50	5.50	5.25
Enamel			
Mean	7.65	7.62	7.68
SD	0.38	0.48	0.27
Minimum	6.75	6.75	7.25
Maximum	8.00	8.00	8.00
Dentin			
Mean	5.33	5.46	5.21
SD	0.66	0.72	0.59
Minimum	4.00	4.25	4.00
Maximum	6.50	6.50	6.00
Pulp			
Mean	3.65	3.42	3.88
SD	1.13	1.51	0.55
Minimum	0.00	0.00	2.75
Maximum	5.00	5.00	5.00

The density of the lesion and that of the enamel was statistically significantly different ($P = 0.0001$).

Over the magnified image, a straight line was traced that crossed over half of the cavity produced in the tooth from the internal vertex of the lesion to the external approximal surface, and in the center of this line the measurement of the density of the image at that point was obtained. The program has a scale of values from 0 to 8 that corresponds to a maximum represented in white and a minimum represented in black. The density of enamel, dentin, and pulp cavity was also measured.

The enamel density measurement was made by obtaining the average densities of a line in the zone of the lesion and the dentin. For the measurement of the dentin, a similar method was used, obtaining the average densities of a line in the zone of the lesion, which stretched from the enamel to the pulp cavity. Finally, the density of the pulp was obtained from the average of the densities of the zone adjacent to the pulp cavity. The absence of a normal distribution in the sample data was proven using the Kolmogorov-Smirnov test. The nonparametric Wilcoxon test was used with a 95% level of significance.

RESULTS

The table shows the average results with corresponding standard deviations (SD) of the measurements obtained of the densities of the radiographic images provided by the RVG unit, both in the lesion produced in the tooth and in adjacent tissues: enamel, dentin, and pulp. This information refers to both premolars and permanent molars. On the scale of 0 to 8, the average density obtained for the specimen, as a whole, was 3.78 in the zone of the defect, 7.65 in the zone of the enamel, 5.33 in the dentin, and 3.65 in the pulp. The Wilcoxon test revealed statistically significant differences ($P = 0.0001$) between the density of the defect and that of the enamel (the average density of the enamel doubled that of the defect). No differences were found between the measurement of the density of the cavity simulating the lesion and that of the dentin, although its average values were greater than those found for that of the simulated lesion and the pulp ($P = 0.95$). Similar results were noticed when the data obtained in molars were compared to those found in premolars, with no significant differences between both groups.

DISCUSSION

While it is true that no experimental defect (cavity) can exactly reproduce caries (Robinson, Weatherell & Kirkham, 1995), the simulation of minimal caries by means of cavitation is a procedure that is useful due to its capacity to be reproduced (Kang & others, 1996). The procedure employed in this study produced lesions radiographically comparable to those produced by caries.

The use of a digital radiographic system demands the calibration of the conditions under which the system offers the images in each case. In this way observations are always made in the best possible conditions. This is achieved by equalizing the image and maintaining the same conditions of brightness and contrast throughout the study. When the density of a lesion similar to caries is compared to that of neighboring tissue, mainly enamel, it can be seen if they are different, and thus if the system employed is effective in the diagnosis of these defects. As a radiographic image is a flat representation of a three-dimensional reality, the thickness of the enamel that surrounds the lesion and is superimposed to it on the image produced could alter the final image (Gröndal & Hollender, 1988). Due to this fact, the present study was conducted with teeth of varying volumes, one group of molars and another of premolars; although in view of the results, this circumstance was not relevant in the conditions of this study. The values of the measurement of the density of the simulated lesion were taken from the center of the

defect because of differences observed between the external surface and the internal extreme of the simulated lesion, and also as it is easily detectable due to the geometry of the simulated defect.

CONCLUSION

RVG, in combination with the described image analysis system, was able to distinguish incipient caries-like lesions in vitro. This type of diagnostic system, which is available to dentists, can help them improve early caries diagnosis.

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DEPARTMENTS

LETTERS

A DREAM OF PRESERVATIVE HEALTH

Your editorial in the May-June issue of *Operative Dentistry* [1999; 24(3) 129] touched a very sensitive nerve in me.

First of all, you are one of many who have "discovered" this idea that perhaps we should be looking in other directions for solutions to some of our dental and medical (systemic) health problems. I could not agree with you more. In fact, dentistry has had many pioneers in this area. However, the advent of third-party payment systems and their attendant bureaucrats/bean counters and a goodly number of greedy hostile dental academics, not to mention a certain part of the hierarchy of organized dentistry, have willfully "murdered" these ideas, especially in dentistry. Lip service is given to prevention and to hygiene, etc, but that is about where it ends.

A look at a partial list of the protagonists would include the following: Weston Price, Melvin Page, Sumter Arnim, Merrill Wheatcroft, Emmanuel Cheraskin, W Ringsdorff, Bruce Pascetti, Bob Barclay, James Clark, and CC Bass, MD. These are people who made a mighty effort to steer the thinking in some different areas. Many of them were teachers and researchers. Many also wrote extensively for a wide variety of journals and authored numerous books in their fields of study. Some of them were general practitioners who made significant contributions towards patient education and treatment. But many of them, especially Price and Page, were pilloried by the profession (ADA) for their revolutionary approaches to dental disease and the attendant possible treatment options.

As a former department head in a dental school, I can tell you that when I brought some of these controversial speakers into the school to address CE courses and even (especially) undergraduates, I was given a very hard time. I prevailed only because I was the Head with the most clout!

There is so much to talk about in this area and never enough time or energy. I fear that this new PEW report will put a further lid on any creative thinking by practitioners.

DAVID O MOLINE, DDS
3200 Southwood Drive
Philomath, OR 97370

TWO POST SYSTEMS

This letter to the editor will discuss the results in the fine article entitled "Retention and Shear Bond Strength of Two Post Systems" by Stockton and Williams [*Operative Dentistry* 1999; 24(4) 210-216]. In this article the retention and shear bond strengths of a stainless steel post were compared to that of a carbon fiber bound epoxy resin post (C-Post). Our research group at Essential Dental Laboratories has observed similar results regarding the physical properties of the C-Post. In our recent article entitled "Comparison of the Retentive and Photoelastic Properties of Two Prefabricated Endodontic Post Systems" by Cohen, Pagnillo, Musikant, and Deutsch [*Journal of Oral Rehabilitation* 1999; 26(6) 488-494], we clearly illustrated by photoelastic analysis that the C-Post displayed uneven and asymmetric patterns of stress when loaded in vertical and oblique states. These findings were consistent with that found in the fine article by Stockton and Williams. For example, Stockton and Williams found 12 root fractures for C-Post #1 and 11 root fractures for C-Post #2 compared to only six for the stainless steel post. They also concluded that the C-Post lacked stiffness, and this adversely affected the success of a restoration. Because of the high incidence of root canal fracture, the use of the C-Post may not be desirable for most cases.

If you have any questions about this letter to the editor, please feel free to call me at 1-201-487-9090.

BRETT I COHEN
Vice President of Dental Research
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The University of Washington Department of Restorative Dentistry is seeking applicants for a full-time position at the rank of assistant professor, academic (tenure) or clinician-teacher (nontenure) track. The position is available 1 July 2000. Responsibilities include predoctoral didactic and clinical teaching in Restorative Dentistry, along with research. Qualifications include a DDS or DMD degree from an accredited institution, licensure or eligibility to become licensed in Washington State, advanced education in prosthodontics or comprehensive restorative dentistry, and at least two years of clinical experience with an emphasis in comprehensive treatment. The University of Washington is building a culturally diverse faculty and strongly encourages applications from female and minority candidates. The University of Washington is an Equal Opportunity Employer. Submit a letter of application and curriculum vitae by 30 November 1999 to:

Dr Glen H Johnson
Department of Restorative Dentistry
Box 357456
University of Washington
Seattle, WA 98195-7456

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The Department of Restorative Dentistry is one of three departments in the Faculty and is responsible for the teaching of dental materials, endodontics, operative dentistry, and prosthodontics. In addition to teaching, staff members also contribute to clinical care of patients under the National University Hospital. The department is also active in research; the major research focus includes the development of biomaterials, clinical evaluation of new restorative materials, and dental implants. Strong support is given to research activities, and collaboration with other faculties for multidisciplinary research helps us to achieve these research goals. The department is forward-looking and innovative in its efforts to educate undergraduates and postgraduates in research and clinical care. With a staff strength of 10, the department works closely to achieve the Faculty's mission "to be a leading international dental institution in teaching, research, and clinical care." The successful candidate will be involved in the teaching of undergraduate students, research, and clinical practice. Visit our website at http://www.nus.edu.sg/NUSinfo/Appoint/den-resto.htm for links to information on the department, the university, terms and conditions of service, and the application form. Only shortlisted candidates will be notified. Send applications to:

Professor Chew Chong Lin
Head, Department of Restorative Dentistry
National University of Singapore
5 Lower Kent Ridge Road
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FAX: (65) 773-2603
E-mail: rsdhead@nus.edu.sg

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The University of Texas--Houston Health Science Center, Dental Branch seeks applicants for a full-time tenure-track position in the Department of Restorative Dentistry and Biomaterials. Applicants must have a DDS/DMD degree with prior teaching and/or private practice experience. Advanced training in operative dentistry, prosthodontics, or GPR/AEGD certification is preferred. Responsibilities include clinical and preclinical teaching to undergraduate and graduate dental students, research, and service. This position will be available 1 September 1999. Academic rank and salary are commensurate with qualifications and experience. The University of Texas--Houston Health Science Center is an EEO/AA employer. Women and minorities are encouraged to apply. Send letters of application and curriculum vitae to:

Dr Peter Triolo, Chair
Department of Restorative Dentistry and Biomaterials
Suite 452
6516 John Freeman Avenue
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ANNOUNCEMENTS

ACADEMY OF OPERATIVE DENTISTRY, EUROPEAN SECTION SECOND ANNUAL MEETING

1-2 October 1999
Munich, Germany

RESTORATION OF POSTERIOR TEETH: THE EUROPEAN VIEW

This meeting will feature a panel of international speakers, who will discuss restorative treatment on posterior teeth. Poster presentations will be given on Saturday, 2 October. This meeting immediately follows the First Munich Esthetic Symposium. Additional information may be obtained by contacting:

Dr Margaret A Wilson, Hon Sec AOD ES
Restorative Dentistry
University Dental Hospital of Manchester
Higher Cambridge Street
Manchester M15 6FH, UK
Telephone: 44 (0) 161 275 6619;
FAX: 44 (0) 161 275 6710
E-mail: Wilsonm@fs1.den.man.ac.uk

AMERICAN ACADEMY OF GOLD FOIL OPERATORS MEETING

27-30 October 1999
MEHARRY UNIVERSITY DENTAL CLINIC
NASHVILLE, TENNESSEE

Two half-day clinical sessions are being planned that will include both castings and direct gold restorations. The essay session is being coordinated by Dr Tim Carlson. Headquarters will be at the Sheraton Music City Hotel. Extracurricular events include an evening at the famous Grand Ole Opry.

For more details contact:

Dr Ronald Harris
AAGFO Secretary-Treasurer
17922 Tallgrass Court
Noblesville, IN 46060
FAX: (317) 867-3011

FUTURE MEETING: The meeting on 1-4 November 2000 will be held in Hawaii, so make your plans early to attend!

CHANGE OF EDITOR

As we begin the move of *Operative Dentistry* to Indiana University under the able leadership of Dr Michael Cochran, new manuscripts submitted on or after 1 June 1999 should be sent to the new Journal office at the following address:

Dr Michael A Cochran, Editor
Operative Dentistry
Indiana University School of Dentistry
1121 West Michigan Street
Indianapolis, IN 46202

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Reprints of any article, report, or letter published in 1999 or before can be ordered through the Seattle office: University of Washington, OPERATIVE DENTISTRY, Box 357457, Seattle, WA 98195-7457; (206) 543-5913, FAX (206) 543-7783; e-mail: rmccoy@u.washington.edu.

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