GLASS IONOMERIC RESTORATIVES: CONCEPTS ON SECONDARY CARIES INHIBITION AND ADHESION

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Introduction

Dental caries, by definition, is a multifactorial and contagious disease of the mineralized tissues of the teeth. It is caused by bacterial activity on carbohydrates resulting in fermentation, and is identified when the mineral content is demineralized followed by disintegration of the organic content. Basically, dental caries prevention can be assessed by eliminating plaque, reducing consumption of sugars, and increasing resistance of the tooth. However, secondary caries is still one of the main factors that afflict the longevity of the restorations, therefore prevention and assessment of secondary caries has become of great importance.

The development of new bonding restorative systems has revolutionized the concept of cavity preparation and restorative procedures. Extensive cavity preparations have been replaced by conservative ones by removing the carious tissue only, producing minimal reduction of the sound tooth tissue (Fusayama, 1979) and permitting an adequate surface for adhesion of restoratives to the tooth tissue. Adhesive restoratives are expected to promote enhanced short- and long-term bond strengths to tooth substrate, effectively seal the cavity margins while protecting against formation of secondary caries.

The advent of fluoride in the preventive dentistry as well as in restorative dentistry has yielded the development of a new concept of dental therapy. The role of fluoride in preventive dentistry is well known and has been widely reported in literature. Both systemic ingestion (e.g., fluoridated water, salt, or milk, fluoride tablets or drops) and topical applications (e.g., rinses, toothpaste, topical solutions and gels) are efficient for caries prevention. Restorative dentistry has also been revolutionized by the incorporation of fluoride ions to many of the recent adhesive and restorative materials, in the attempt to increase the longevity of restorations by protecting the restoration against secondary caries. The glass ionomeric materials, which include the conventional and resin-modified glass ionomer cements, and the so-called compomers, have been reported to promote an anticariogenic effect and an ion-exchange reaction with the tooth tissue (Mount, 1994).

Fluoride has also been incorporated to several recent adhesive resin systems and resin composites, with the intention to provide long lasting restorations with durable bonds, sealing of the cavity margins, and protection against secondary caries. However, consistent levels of fluoride release of these materials are still low, and extensive research is being conducted to improve these levels without weaken the physico-mechanical properties of these materials.

Since the development of the first glass ionomer cement (Wilson and Kent, 1972), a great variety of glass ionomeric materials have been marketed with purposes of restorative dentistry. Because of their potential anticariogenic capacity and bonding physico-chemically with the tooth tissue, these materials have become popular as esthetic
filling materials to restore carious lesions or for patients with a high caries risk. The conventional glass ionomer cements were initially placed directly into the cut cavity, without any prior surface treatment. However, the smear layer that covers the cut dental surfaces (Pashley, 1984), may break cohesively and fail during polymerization shrinkage (Tao and Pashley, 1988). Since bond strengths without pre-treatment were reported to be inconsistent (Hewlett et al., 1991) impairing clinical retention (Ngo et al., 1986), pre-treating surface with acid solutions became routine before restoring with a glass ionomer or a resin-modified glass ionomer cement. Polyacrylic acid is an effective pre-treatment agent, however, to date there is little evidence reporting to what extent the resinous monomers can penetrate the demineralized dentin.

Volumetric shrinkage of light-cured restoratives (e.g., resin composites, resin-modified glass ionomers, and compomers) during polymerization can be detrimental to the bonding properties of these materials to dentin (Feilzer et al., 1988). Similar to resin composites, resin-modified glass ionomers shrink approximately 3% in volume during setting (Feilzer et al., 1988). Although this intrinsic stress can be relieved by later water sorption (Davidson and De Gee, 1984; Feilzer et al., 1988), the initial polymerization shrinkage of the RmGICs may lead to adhesive failure at the tooth-filling interface compromising the longevity of the restoration (Ciucchi et al., 1997). Therefore, the use of adhesive bonding systems prior to filling with glass ionomeric materials could be a good alternative to increasing bond strengths to the tooth tissue. Studies are still required to determine if the anti-cariogenic potential of these materials and fluoride release are affected by the application of bonding systems.

With the increase of life expectancy and retained teeth in the elderly, as well as periodontal problems in later life, root surfaces become exposed to the oral environment increasing the risk of root surface caries. Root caries present specific operative difficulties such as retention and access of the restoration. Resin-modified glass ionomer cements are the materials indicated for restoration of these lesions, due to their ion-exchange adhesion to tooth tissue (Hinoura et al., 1991; Pawlus et al., 1994), and because of their anticariogenic potential to inhibit secondary caries by fluoride release.

Ideally, the restorative adhesive materials should be able to promote strong short- and long term bonding to the tooth tissues, undergo minimal contraction during light-curing, and possess anticariogenic properties by releasing and uptaking fluoride into and from the oral environment.
Chapter 1.

Introduction to glass ionomer materials

Conventional glass ionomer cements

The first glass ionomer cement for dental use was developed by Wilson and Kent in the early 1970s (Wilson and Kent, 1972). The conventional glass ionomer cement (glass polyalkenoate cement) contains a powder of fluoroaluminosilicate glass with high concentrations of aluminum, fluoride, calcium, sodium, and silica (Sidhu and Watson, 1995; Smith, 1992; Wilson and McLean, 1988); and a liquid that is normally polyalkenoic acid but may contain polymers and copolymers of polyacrylic acid, itaconic, maleic, or vinyl phosphonic acid (McLean, 1992; Smith, 1992). When the powder and the liquid are mixed together, the ion-leachable glass and the polyacid react forming overlapping acid-base reactions. In the first phase, the fluoroaluminosilicate glass particles in the powder are attacked by the hydrogen ions (H+) from the polyalkenoic acid, resulting in a limited degradation of the glass surface and releasing the simple metal ions calcium (Ca²⁺), aluminum (Al³⁺), sodium (Na⁺), and fluoride ions (F). Silicic acid is also released and a layer of silica gel is slowly formed around the surface of the unreacted glass powder. With rapid increase of the pH of the mixture, the metallic ions are progressively lost until complete decomposition of the glass particles occurs (Burgess, Norling, Summitt J, 1994). When the free calcium and aluminum ions reach saturation in the silica gel, they diffuse into the liquid and cross-link with two or three ionized carboxyl groups (COO⁻) of the polyacid to form a gel. The cross-linking increases through calcium ions being replaced by aluminum ions, hydrating sufficiently the gel, resulting in a precipitation of the cross-linked polyacrylate salt until the cement is solid (Burgess et al., 1994; Van Meerbeek et al., 1996). During the acid diffusion phase, sodium fluoride ions are released, however, because these are not a matrix forming species, it does not deplete the cement from its physical properties. As the cement matures over the first 24 hours, progressive cross-linking occurs possibly by hydrated Al³⁺ ions since the sensitivity to water decreases and the percentage of bound water and glass transition temperature increase (Wilson and McLean, 1988). The rate of reaction can be controlled by the powder/liquid ratio and the surface area of the powder, availability of fluoride ions and types of acids used (Walls, 1986; Wilson and McLean, 1988; Wilson, 1989).

The main advantages of the conventional glass-ionomer cements over other esthetic restorative materials are short and long term release of fluoride with cariostatic potential, ion-exchange reaction with enamel and dentin, low setting shrinkage and thermal
expansion similar to the tooth structure (Burguess et al., 1994; Mount, 1994; Sidhu and Watson, 1995; Dunne et al., 1996; Forsten, 1991). However, practical difficulties such as a short working time and a slow setting reaction, high sensitivity to water and susceptibility to desiccation compromising the physical integrity and esthetic properties, and low adhesive properties compared with the resin composites have demanded modifications of the cement composition.

Resin-modified glass ionomer cements

In order to overcome the disadvantages of the conventional glass ionomer cements yet preserving the benefits of these materials, a new type of material that combines the acid-base reaction with the methacrylate resin technology has been developed (Antonucci et al., 1988). The resin-modified glass ionomer cements are set by both photoinitiator systems and conventional acid-base reaction. Light irradiation of the mixed cement results in fast initial hardening of the cement due to free radical polymerization of HEMA, forming a poly-HEMA matrix (Mitra, 1994; Tosaki and Hirota, 1994; Yoshikawa et al., 1994). The acid-base reaction starts slowly as the powder and liquid are mixed. This acid-base reaction continues up to 24 hours after the material has been light cured because the pH values of the surface of the light-cured cement gradually increases to 24 hours similarly to a conventional glass ionomer cement (Tosaki and Hirota, 1994).

The systems that undergo both acid-base reaction and free radical polymerization by light-curing are the so called dual-cure resin-modified glass ionomer cements (e.g., Fuji II LC, GC Corp.). One commercial product, 3M Vitremer™ has been reported to be a tri-cure system, since curing of the methacrylate group also occurs with reaction of redox initiator systems (e.g., amine/peroxide; ascorbic acid/persulfate) in the dark (Mitra, 1994).

The resin-modified glass ionomers provide a longer working time and improved esthetics, immediately hardens when light-cured, are less sensitive to water, possess improved mechanical adhesive properties, are radiopaque, and are biocompatible with the tooth structure (Momoi and McCabe 1993; Swift et al., 1995; Shono, 1995; Sidhu and Watson, 1995; Gladys et al., 1997). Furthermore, they preserve the clinical advantages of the conventional cements, which are potential chemical reaction with the tooth substance, fluoride release, and thermal expansion similar to the tooth structure (Burguess et al., 1994; Mount, 1994; Swift et al., 1995).

Because of their simple clinical application, the resin-modified glass ionomer cement has become very popular among the general practitioners. Although bond strengths were improved by applying conditioners/primers, bond-strengths of these materials are still low compared to the resin composite systems.
Compomers

Recently, a new class of hybrid resinomers (compomer) that combines the resin composite and glass ionomer technology has been developed (e.g., Dyract, DeTrey/Dentisply; Xeno, Sankin Kyogo). They are a one component system, containing basically aluminofluorosilicate glass, Bis-GMA, photoinitiators, and a reactive acid monomer. The compomers come in the anhydrous form preventing the initial acid/base reaction from taking place. These materials contain shorter chain monomers carrying both acidic and methacrylate groups, and are initially cross-linked through the methacrylate groups due to light activation. However, when it is placed as a restoration and the material becomes hydrated, the ionic reaction between the acid groups on the polymer and glass occurs. (Hammesfahr, 1994; Blackwell and Käse, 1996)

Differently to the resin-modified glass ionomers, the one component compomers have practically unlimited working time and are not sensitive to water. Their chemical properties are similar to those of the resin composites, however posses lower flexural strength and lower abrasion resistance of occlusal restorations. The compomers are less soluble in water than the ionomer cements, but have shown to be susceptible to degradation at low pH, releasing fluoride, metal ions, and silica (Blackwell and Käse, 1996; Watts, 1996).

A great variety of current resin-modified glass ionomer cements and compomers are available in the market, including novel two or single-step bonding systems (Nikaido et al., 1997). Although the future of these materials is still unclear, compomers may become a strong competitor to resin composite systems in preventive and general dentistry; for fissure pit sealing and for replacement of natural tooth structure, with anti-cariogenic properties and simple clinical application.

Release of fluoride and other elements from glass ionomeric materials

The release of fluoride from conventional and resin-modified glass ionomer cements has been extensively studied since the development of these materials (Forsten, 1977; Tveit and Gjerdet, 1981; Cranfield et al., 1982; Swartz et al., 1984, Meyron and Smith, 1984; Wilson et al., 1985; Forsten 1990; Mitra, 1991; Diaz-Arnold et al., 1995). Many studies have been conducted to compare the fluoride release from conventional and resin-modified glass ionomers (Forsten, 1977; Tveit and Gjerdet, 1981; Cranfield et al., 1982; Swartz et al., 1984, Meyron and Smith, 1984; Wilson et al., 1985; Forsten 1990; Mitra, 1991; Takahashi et al., 1993; Diaz-Arnold et al., 1995; Forss, 1995). However, the results varied when comparing both types of cements, leading to the conclusion that the release of fluoride can be easily affected by the measuring conditions and dissolving medium.
It is known that the formulation of the glass ionomer, initial fluoride content of the glass, pH changes of the environment, type and amount of resin incorporated to the material, differences in powder:liquid ratio, and mixing and setting times can influence the fluoride release (Thornton et al., 1986; Swift, 1988; Swift et al., 1990; Rezk-Lega et al., 1991; Forss, 1993; Momoi and McCabe, 1993; Takahashi et al., 1993). The pattern of fluoride release has been proposed (Forsten, 1977), and it was shown that after the initial fluoride ‘burst’ that occurs in the first 24 hours, the release rate will drop to lower levels by the end of 3 months, and traced up to 2 years (Forsten, 1990; Diaz-Arnold, 1995). The initial burst of fluoride that is released is probably due to initial ‘wash-off’ of fluoride from the surface of the cement. After this, the release rate seems to be controlled by dissolution of fluoride from the cement. This dissolution rate may dependable on the surface area available for dissolution rather than shape of the sample or of the immersion solution (Fukuzawa et al., 1987).

It has been reported that glass ionomer materials also release different chemical elements other than fluoride (Crisp et al., 1976; Wilson et al., 1985; Fukuzawa et al., 1987). This probably occurs because modified glasses have been included, and part of the calcium has been replaced by strontium in order to improve the radiopacity of the resin-modified glass ionomers. Crisp et al., (1976) have reported that the release of matrix forming elements such as calcium, strontium and aluminum leached in lesser amounts than the non-matrix forming elements silica, sodium and fluoride. It was further shown that the calcium released by the ionomeric materials may protect the adjacent enamel from demineralization and accelerate formation of calcium fluoride (Séppa et al., 1992). It has also been shown that release of these elements can be affected by the changes in pH (Forss, 1993), saliva proteins, and buffer systems (Rezk-Lega et al., 1991).

The compomers were reported to release fluoride and metal ions, as well as silica once the acid-base reaction takes place after water sorption (Watts, 1996; Hickel, 1996). This fluoride release mechanism is a result of the reaction between acid and the amino-fluoro-silicate glass during setting. The compomers do not release as much fluoride as the conventional or resin-modified glass ionomers, however, to date the quantity of fluoride necessary to provide a suitable cariostatic effect has not been established. Therefore in vitro and in vivo studies are still necessary to clarify the effectiveness of fluoride-releasing materials.

Uptake of fluoride by glass ionomer materials

The concept that glass ionomers could also uptake fluoride as well as release it, was initially suggested (Walls, 1986) and it was later confirmed that these materials could act as rechargeable fluoride sources (Forsten, 1991). Previous studies have reported that
regular application of topical fluoride and regular use of fluoride toothpaste could result in uptake of fluoride into the conventional glass ionomer, and that this fluoride could be subsequently released to the adjacent tooth structure (Hatibovic-Kofman and Koch, 1991; Seppä et al., 1993). Another study has reported that resin-modified glass ionomer materials could also act as a short-term rechargeable fluoride sources (Diaz-Arnold et al., 1995). They reported that fluoride release after exposure to fluoride gels could only represent a wash-out of ions adsorbed to the surface of the material, rather than a diffusion into the matrix. Nevertheless, they recommended a 6 min daily use of fluoride gel for caries-prone patients, since most fluoride released after exposure to the gel occurred within two days.

The potential for surface degradation should also be considered after application of fluoride gels, since a decrease in microhardness of resin-modified glass ionomers has been reported (Diaz-Arnold et al., 1995; Billington et al., 1987) after long exposures to fluoride gels. This may surpass the cariostatic benefits of recharging the glass ionomeric restoration with fluoride gels.

**Uptake of fluoride by enamel and dentin**

The fluoride functions in several ways to increase resistance against enamel and dentin caries. The fluoride ion enhances remineralization of demineralized early lesions of enamel (Koulourides et al., 1961; Iijima and Koulourides, 1988; Featherstone et al., 1990), and increases enamel resistance to subsequent acid attack when incorporated to the crystals (Koulourides and Cameron, 1980; ten Cate and Duijsters, 1990a/b; Featherstone et al., 1990). The fluoride ion influences remineralization of enamel by acting as a catalyst. The size and electrostatic charge of the fluoride ion permits a favorable stereoscopic arrangement of calcium and phosphate on the crystal surface (Hatibovic-Kofman et al., 1997), and diminishes the energy activation required for the hydroxyapatite crystal growth.

Fluoride uptake by dentin is similar to that of the enamel, but may be greater because of its greater porosity, greater water content, and smaller size of the apatite crystallites (Mellberg and Singer, 1997, Scott and Symons, 1982). A low pH increases the uptake of fluoride by dentin, possibly due to the initial demineralization of the tissue and to deposition of greater amounts of fluoride into the hydroxyapatite crystals (Tveit et al., 1983). The possibility of secondary caries inhibition and remineralization of early carious lesions by fluoride release are the main advantages of glass ionomeric restoratives over other restoratives.

Topical applications of solutions containing high fluoride concentration promote the formation of calcium fluoride (CaF₂) on the sound tooth surface or on the surface of initial lesions. The CaF₂ will later dissolve and release the fluoride ions that will react
with the hydroxyapatite converting to fluorohydroxyapatite. This process may increase remineralization or decrease the demineralization process, depending on the pH, calcium, and phosphate concentrations of the environment (Rølla and Saxegaard, 1990). However, it has also been suggested that constant low levels of fluoride ions for at least 24 hours (Arends and Schuthof, 1975) were a favorable condition for formation of fluoroapatite, and that multiple applications of agents with low fluoride concentrations were more effective than a single, high concentration approach (Koulourides et al., 1975).

**Dental caries**

Dental caries, by definition is an infectious disease that afflicts the enamel, dentin, and cementum, caused by the production of acids as a result of the fermentation of carbohydrates by microorganisms. The caries lesion can be classified according to the anatomical site, to the severity of the lesion, or rapidity of the attack. When divided according to the anatomical region, the initial lesion can be a pit and fissure or smooth surface lesion, starting on enamel or root surfaces. It can also be secondary caries when located at a margin of a restoration. The classification for caries according to the rapidity of the attack is broader, since there is the individual variation of the teeth, and different degrees of carious challenge. The two extremes are namely the rampant caries, which occur as a result of a rapid destruction of numerous teeth, and the arrested caries, which is a carious lesion that does not progress due to changes in the environment (hygiene habits, fluoride applications, etc).

**Enamel caries lesion**

The histological appearance of the early human enamel carious lesion has been previously described (Darling, 1956; Gustafson, 1957; Darling, 1958; Silverstone, 1968), and divided into zones according to the histological appearance under the light microscope. The classical lesion has been divided into four zones (Silverstone, 1973). From the outer surface they are the surface zone, body of the lesion, dark zone, and translucent zone. The surface zone is a relatively unaffected region that is superficial to the lesion. The body of the lesion is markedly demineralized and is the greatest area of demineralization (Darling et al., 1961; Silverstone, 1970). As the caries lesion progresses, the body of the lesion increases in extension and cavitation of the surface occurs. Once the lesion becomes cavitated, it can no longer be remineralized (Limeback, 1996). The dark zone lies just under the advancing front of the body of the lesion. When examined under the light polarized microscope with quinoline as imbibing medium, the dark zone shows positive birefringence in contrast to the negative birefringence of the sound enamel (Hörsted-
Bindslev and Mjör, 1996). Studies have reported that the dark zone contained micropores that were easily penetrable to small molecules (Darling et al., 1961). The pores that were not penetrated with nonaqueous media remained filled with air, disclosing the positive birefringence zone characterized by a dark color. In contrast when observed with water as imbibing medium, the dark zone is not apparent. The translucent zone lies under the dark zone, at the advancing front of the lesion. This zone is a result of mineral loss (Hörsted-Bindslev and Mjör, 1996), and although well demarcated from sound enamel, it is not always present on the entire extension of the advancing front.

**Dentin caries lesion**

The dentin caries lesion in an early stage occurs when the caries progression invades the amelodentinal junction spreading the disease laterally. As the lesion progresses it follows the curvature of the dentinal tubules towards the dental pulp. At this early stage of lesion development, the caries lesion is still free of bacteria, however, the acids can easily diffuse to the underlying dentin because of the extremely porous enamel at this stage. At this stage, the dentin lesion can be classified microscopically in two zones (Hörsted-Bindslev and Mjör, 1996) from the pulp outwards: translucent zone and body of the lesion. The translucent zone (or zone of sclerosis) is formed by physicochemical precipitation of mineral salts within the tubules (Hörsted-Bindslev and Mjör, 1996). It is produced as result of mild stimulation of the dentin/pulp complex, and serves as a mineralized barrier against acids, enzymes and bacteria towards the pulp. The body of the lesion (also referred as ‘dead tract’) in this stage of lesion progression has not been yet affected by bacteria. However, the superficial peritubular dentin and intertubular dentin are partly demineralized, and the tubules possibly confine remnants of the odontoblastic process and air. This will disclose a dark zone when observed under a polarized light microscope (Hörsted-Bindslev and Mjör, 1996).

When the enamel lesion becomes cavitated, and bacterial invasion occurs, the progression speed of the lesion increases. The primarily acidogenic bacteria produce acid, demineralizing the dentin tissue as deep as the translucent zone (Hörsted-Bindslev and Mjör, 1996). In this stage of caries progression, the lesion can also be divided microscopically in two zones: translucent zone and body of the lesion. However, because the pulp may show some inflammatory reaction to chemical stimuli, the irregular dentin may increase in thickness. Since the tissue changes at this stage are more complex, the body of the lesion can be subdivided microscopically in three other zones leading to a total of four zones: zone of demineralization, zone of penetration, and the zone of destruction (Hörsted-Bindslev and Mjör, 1996). The translucent zone lies deepest to all, separating
the body of the lesion from the sound dentin and is bacteria-free. The zone of
demineralization is the deepest part of the body of the lesion, and is usually bacteria-free. The zone of penetration contains bacteria and is situated in the middle part of the body of the lesion. The intertubular dentin is markedly demineralized, however the collagen fibers still present the typical banding (Hörsted-Bindslev and Mjör, 1996). The zone of destruction is the outermost zone, and lies at the amelodentinal junction. This zone has been described as the 'outer carious dentin' (Fusayama, 1979), since the dentin substance is completely destroyed and the bacteria spread throughout the entire zone. It is clinically recognized by a greatly softened and discolored dentin, and is stainable by fuchsin or acid red dye. The ultra-morphological analysis of this lesion showed a demineralized intertubular dentin with few collagen fibers, and absence of peritubular dentin and odontoblastic processes (Ohgushi and Fusayama, 1975). The hardness of this zone of destruction or outer carious dentin has been reported to be approximately 4.4 Knoop Hardness Number (KHN) in acute lesions, and approximately 61.0 KHN in chronic lesions (Fusayama et al., 1966).

What was microscopically described as the body of the lesion may correspond to the 'inner carious dentin' described by Fusayama et al. (1972). Ultramicroscopic observations of the inner carious dentin disclosed a partly demineralized intertubular dentin with apatite crystals bound to collagen fibers, that possessed apparently normal crossbands (Ohgushi and Fusayama, 1975). It was observed that the peritubular dentin was partially demineralized and with sound odontoblastic processes (Ohgushi and Fusayama, 1975). Furthermore, biochemical studies have reported that the crosslinks and precursors of the outer carious dentin were dramatically lesser than those of the inner carious dentin or sound dentin (Ohgushi and Fusayama, 1975).

Root caries lesion

With the increase of life expectancy and retained teeth in the elderly, as well as periodontal problems in later life, root surfaces become exposed to the oral environment increasing the risk of root surface caries. The process of root caries formation appears to be similar to that of enamel caries, in which alternating cycles of demineralization and remineralization occur constantly (Wefel, 1994). Root surface lesions initially occur at or below the cementoenamel junction with exposition of the root surfaces to the oral environment. It may involve cementum only, cementum and dentin, or dentin alone when the cementum has been lost.

As for enamel carious lesion, the root lesions have been also divided into zones under the microscope. The surface zone lies superficial to the lesion, and can be apparently intact or eroded. The body of the lesion represents the considerably demineralized dentin, and lies under the surface zone. The translucent zone of enamel
has been termed frontal zone for root surface lesions, and it lies deep in the body of the lesion denoting the advancing front of the lesion (Featherstone et al., 1987).

More advanced lesions have shown loss of surface contour and cementum, or a radiopaque surface layer (Wefel, 1994). This radiopaque surface layer may be a sign of an inactive root surface lesion, since a hard mineral surface is the requirement for a clinical classification of inactive root caries (Schüpback et al., 1992; Nyvad and Fejerskov, 1987). It has been also hypothesized that inactive root surface caries often show surface loss and later rehardening of the remaining exposed dentin, by means of oral hygiene and fluoride treatments (Nyvad and Fejerskov, 1986).

**Enamel and dentin as adhesive substrates**

Because restorative materials do not adhere effectively to dental tissues, bonding to tooth structure has become one of the greatest expectations of the general practitioner in the last decade. The development of the bonding technology and new bonding systems have caused great advances in adhesive dentistry and changes in the concept of cavity preparation. Box-shaped extensive cavity preparations have been replaced by conservative ones, cutting the carious tissue only while preserving the intact tooth structure (Fusayama et al., 1986).

The enamel is basically a dry tissue that comprises approximately 96% by weight of hydroxyapatite crystals [Ca_{10}(PO_4)_6(OH)_2] that are arranged in a rod structure, and 4% water and organic material (Scott and Symons, 1982). It was initially proposed that etching the enamel surface with phosphoric acids would increase adhesion of acrylic filling materials (Buonocore, 1955; Hotta et al., 1992). Since then, phosphoric acid has been widely used to etch enamel and successfully bond restorative materials. When the enamel surface is etched by acids, the hydroxyapatite crystals are selectively removed increasing the surface porosity and energy, allowing the monomers of the adhesive systems to provide appropriate wettability and to penetrate into the microporosities. With the polymerization of these monomers and formation of resin tags in the enamel microporosities, a strong micromechanical bond is formed and is the predominant mechanism of enamel bonding (Gwinnett and Matsui, 1967; Hotta et al., 1992). However, recent milder etching agents have been introduced as a substitute to the phosphoric acid, and several in vitro reports exist in literature demonstrating that bond strengths of different strong and mild etchants differ only slightly. However to date there are little clinical studies proving clinical efficacy of the mild etchants.

On the other hand, bonding to dentin is a greater challenge and has been extensively studied because of the difficulty of a less reliable substrate (Fusayama, 1988; Douglas, 1989). The dentin is composed of a heterogeneous structure containing
approximately 70% hydroxyapatite, 18% organic material (mainly type I collagen) and 12% water (Linde, 1985; Scott and Symons, 1982). The organic and inorganic components are unevenly distributed in the inter- and peritubular dentin. Moreover, the dentin is a highly permeable tissue with numerous dentinal tubules that extend radially from the pulp throughout the entire thickness of the dentin (Gaberoglio and Bränström, 1976). Therefore, several factors account for the difference in bonding mechanism of enamel and dentin.

When the tooth structure is cut by rotary instruments, a cutting debris is created over the dentin surfaces and is called the smear layer. This layer covers the dentin surface and occludes the entrance of the dentinal tubules. The thickness of this smear layer normally varies from 1 to 5 μm and depends on the type of instrument used and the irrigation condition (Eick et al., 1970; Pashley, 1984). Although the smear layer diminishes the dentin permeability, it may interfere impeding the direct contact of the bonding material with the dentin (Pashley, 1984; Pashley, 1990). In order to obtain a good adhesion to dentin, the smear layer should be removed or modified with conditioners, such as acid solutions or EDTA.

When the smear layer is removed and the superficial dentin is demineralized by acids, the resin monomers of the adhesive resin infiltrate and polymerize in situ, producing a hybrid structure of collagen fibrils surrounded by resin and residual hydroxyapatite crystals (Nakabayashi, 1982; Wang and Nakabayashi 1991; Sugizaki, 1991; Gwinnett and Kanca, 1992; Van Meerbeek et al., 1992; Eick et al., 1993). Several dentin bonding mechanisms have been proposed, however, the hybridization theory, which consists of micromechanical interlocking between the adhesive resin monomers and the network of exposed collagen fibrils, has been generally accepted (Bowen et al., 1982; Nakabayashi et al., 1982; Erickson, 1989; Inokoshi et al., 1990; Pashley, 1990; Asmussen et al., 1991; Harniratissai et al., 1991; Erickson, 1992; Van Meerbeek et al., 1993).

Adhesion of glass ionomeric materials to enamel and dentin

Although the exact mechanism of bonding conventional glass ionomer cements to enamel and dentin is still unknown, it is believed that it involves the wetting capacity of the surface by these materials and subsequent formation of ionic bonds by ion exchange with the hydroxyapatite (Wilson and McLean, 1988). It has been proposed that the polyacrylic acid will soften the surface of the tooth and the polyacid chains will diffuse into the tooth structure displacing the calcium and phosphate ions (Akinmade and Nicholson, 1993). The ion exchange has been traced by employing a fluorescent dye technique and confocal microscopy (Watson et al., 1991). It has also been suggested that there is a degree of
adhesion of the glass ionomer cement to the collagen of the dentin in addition to the ion exchange reaction (Akinmade, 1994).

For many years, the glass ionomer restoratives have been used as direct bonded materials without prior acid etching of the cavity. However, surface conditioners have been found to improve the bond strengths. Although many types of acids have been tested as conditioners, the polyacrylic acid has been found to be the most adequate, because it acts as a weak etching agent removing the smear layer, but does not remove completely the smear plugs from the dentin tubules (Powis et al., 1982). The polyacrylic acid will also alter the surface energy of the tooth surface, exposing the highly mineralized substrate to the diffusion of the acid and exchange of ions (Mount, 1994). In order to define these two actions, and to distinguish the behavior of the conditioner from that of the acid, the term “conditioning” was originally used by McLean and Wilson (1977). Therefore, it was suggested that the polyacrylic acid conditioner acts as a self-etching primer, because it demineralizes and probably penetrates the surface promoting a surface receptive for the cement. It is also chemically compatible with the components in the liquid phase of the cement, and may as well improve the wetting ability and surface integrity of the cement (Erickson and Glasspoole, 1994).

Although the exact bonding mechanism of the resin-modified glass ionomer cement is still unclear, adhesion of these materials probably occurs both chemically and micromechanically. As the conventional glass ionomer cements, they undergo an acid-base reaction as soon as the powder and liquid are mixed to later form ionic bonds. Additionally, a mechanism similar to those of the adhesive systems has been suggested, because they contain monomers such as HEMA that penetrate the exposed collagen network or enamel, bond micromechanically when light-cured (Carvalho et al., 1995), and are influenced by different light-curing intensities (Erickson and Glasspoole, 1994). It has been shown that light-curing directly influences the bond strengths of resin-modified glass ionomer cements, since bond strengths have decreased when the material thickness was increased or when the light-curing intensity was decreased (Erickson and Glasspoole, 1994). Because the resin bonds tend to decrease over time (Burrow et al., 1993), and probably due to degradation of the resin (Sano et al., 1998), the bonding mechanism of the resin-modified glass ionomer cements may be advantageous because ions may diffuse from the cement filling the spaces between the collagen fibrils. Long term studies on bonding properties of glass ionomer cements are still limited in literature, and are essential for a full understanding of the bonding mechanism.
Chapter 2

In vitro secondary caries inhibition around fluoride releasing materials

Introduction

Clinical dentistry aims to provide long lasting restorations that produce durable bonds and sealing of the cavity margins. However, the longevity of a restoration may be compromised due to many factors such as secondary caries, microleakage, and failure of the bond at the tooth-restoration interface. Volumetric shrinkage during light curing of the restorative material may stress and break the bond with the tooth, forming a marginal gap (Feilzer et al., 1988; Ciucchi et al., 1997). One of the main factors that induce secondary caries and failure of a restoration is microleakage. This gap between the restoration and the cavity wall permits invasion of fluids and bacteria leading to secondary caries.

With the increase of life expectancy and retained teeth in the elderly, as well as periodontal problems in later life, root surfaces become exposed to the oral environment increasing the risk of root surface caries. Root caries present specific operative difficulties such as retention and access of the restoration. Adhesive restorative materials which release fluoride such as resin-modified glass ionomer cements are the materials indicated for restoration of these lesions, because of their good adhesion to both dentine and enamel substrates (Hinoura et al., 1991; Pawlus et al., 1994; Pereira et al., 1997) and because they possess an anticariogenic effect and the capacity to inhibit secondary caries by fluoride release (Griffin et al., 1992; Prado et al., 1994; Souto and Donly, 1994).

Fluoride has been also incorporated in resin composites with the purpose of inhibiting secondary caries at the cavity margins by fluoride release. Additionally, newer bonding systems adhere much better to the cavity walls preventing separation of the restorative material from the cavity walls. Therefore it would be reasonable to speculate that restorations with recent bonding systems would prevent formation of artificial secondary caries especially when restored with a fluoride releasing resin composite.

The aims of this study were to compare the capacity of two resin-modified glass ionomer cements, a conventional glass ionomer cement, and a fluoride-releasing adhesive resin composite to inhibit in vitro secondary caries, as well as to measure the width and height of the inhibition zone and the depth of the demineralized outer lesion.
Materials and Methods

Sample preparation for artificial caries lesions

Bovine incisors from 20-24 month old cattle were used within 24 hours after extraction. Oral tissues and the cementum were manually removed using scalers to expose the underlying dentine. The roots were obtained by separating them from the crowns at the cementum-enamel junction with a low-speed diamond saw (Bronwill, NY, USA) under water spray coolant. In order to prevent dehydration of the dentine during the restorative procedure, the pulp tissue was left in situ, and the cut surface and root apex sealed with wax and coated with two layers of nail varnish.

Two box-shaped cavities approximately 3 mm long, 2 mm wide, and 1.5 mm deep were prepared on both buccal and lingual dentine surfaces of each root, using a diamond bur (ISO # 106) mounted in high speed turbine with air-water coolant. The cavity margins were finished with a straight fisher steel bur (ISO # 010) in a slow speed handpiece under copious water spray, to achieve a cavosurface angle as close as possible to 90°. Twelve cavities were prepared for each material.

The materials used in this study were two resin-modified glass ionomer cements (Fuji II LC and Vitremer), a conventional glass ionomer cement (Fuji II), and a fluoride releasing adhesive resin system (Clearfil Liner Bond II, Protect liner F, and Clearfil AP-X) (Table 2.1). The cavities were treated according to the manufacturer’s instructions. The cements were mixed at room temperature, transferred into a C-R syringe tip (Centrix™, Connecticut, USA) and injected into the cavity. Fuji II LC and Vitremer were light cured for 60 s each, and Fuji II was allowed to set for 15 min in room temperature. The glass ionomer cement and resin-modified glass ionomer cement restorations were coated with the respective varnishes, and then stored for one week in tap water at 37 °C, because it has been shown that the greatest release of fluoride occurs within the first week (Takahashi et al., 1993). For the resin composite specimens, after application of the adhesive system, Protect Liner F was painted to the cavity walls, and light cured for 20 s. The cavity was then bulk filled with a resin composite, and light cured for another 60 s. Like the glass ionomer specimens, the resin composite specimens were stored in tap water for one week at 37 °C. All restorations were then finished and polished flat with polishing disks (Rainbow polishing Kit, Shofu Inc. Kyoto, Japan) under running water to expose the cavity margins. The integrity of each cavosurface margin was examined under a light microscope at 20x magnification. Two coats of acid-resistant nail varnish were then applied to the entire specimen surface, leaving a 1 mm-window around the cavity margins. Each specimen was stored for three days at 37 °C in individual bottles in 20ml of a buffered demineralizing solution previously described by Wefel et al., (1995), containing 2.2
mmol/L CaCl₂, 2.2 mmol/L NaH₂PO₄ and 50 mmol/L acetic acid adjusted to pH = 4.5.

**Specimen preparation for Polarized Light Microscopy**

The specimens were then removed from the demineralizing solution and thoroughly rinsed in running water. Longitudinal sections of approximately 150 μm thick were cut through each restoration, perpendicular to the long axis of the root by means of a water-cooled diamond saw microtome (Leitz 1600 Microtome, Wetzlar, Germany). The sections were reduced to approximately 100 μm thick by grinding and polishing on coarse and fine wetstones. The sections were then dehydrated in varying grades of ethanol up to 100% and then immersed in quinoline for 20 min. The specimens were observed under a polarized light microscope (PM-10AK, Olympus Inc., Tokyo, Japan) with quinoline as the imbibing medium (refractive index: 1.62), and photomicrographs of the cavity margins were taken at 40x magnification.

In both conventional- and resin-modified glass ionomer cement-cavity margins, a zone exhibiting the same birefringence as normal dentin was observed adjacent to the restoration. This zone was defined as an inhibition zone. Adjacent to this inhibition zone, a demineralized lesion with positive birefringence was defined as outer lesion.

The resulting photomicrographs were projected through a profile projector (V-16C Nikon, Tokyo, Japan), and evaluated for features such as: lesion depth and shape, and presence of an acid-resistant zone adjacent to the cavity wall. The contour of each outer lesion was traced to identify a possible correlation between depth and form of the lesion with tubule orientation and material. The depth of the outer lesion was measured from the top of the lesion to the deepest demineralized front, i.e., the limit between positive and negative birefringence fronts (Fig. 2.1a). Existence of wall lesion was determined when the demineralized front extended from the top of the dentin surface deeper than the outer lesion along the cavity margin (Fig. 2.1b). The inhibition zone was traced to determine differences in width and height of this zone according to the different restorative materials. The height of the inhibition zone was measured from the top of this zone to the deepest demineralized front of the outer lesion; the width of the inhibition zone was measured at the midpoint of this zone (Fig. 2.1a).

The depths of outer lesions, and widths and heights of the inhibition zones were analyzed by one-way analysis of variance (ANOVA) and Fisher’s PLSD test at 95% level of confidence.

**Results**

The outer lesions for all specimens showed similar features, which could be clearly
observed due to changes in birefringence (Fig. 2.2a-d). The top surfaces of the outer lesions were slightly concave due to loss of mineral and therefore surface shrinkage. Both conventional and resin-modified glass ionomer restorations showed a small separation at the cavity margins. This could have occurred during light-curing or setting of the cement due to volumetric shrinkage of the materials, or artificially created during the sectioning process by mechanical stress and material loss, or during dehydration process for microscopic observation by contraction of the demineralized outer lesion away from the restoration. Separation of the material from the cavity margin was not noted for the adhesive resin composite specimens.

The resulting mean values of the depths of the outer lesions and heights and widths of inhibition zones for the four materials are shown in Fig. 2.3a-c. The mean depths of the outer lesions did not show dependency on the direction of the dentinal tubules or differences among the four materials (p>0.05).

Nevertheless, the widths and heights of the inhibition zones were material dependent. The width of the inhibition zone adjacent to Fuji II was significantly greater than those created adjacent to Fuji II LC and Vitremer (p<0.001). The height of the inhibition zones created by Fuji II and Vitremer were statistically similar, and were significantly higher than the zone created adjacent to Fuji II LC (p<0.001). No inhibition layer was observed for the fluoride releasing low viscosity resin composite. In contrast, wall lesions were observed at the cavity margins in 80% of the resin composite restorations. A Wall lesion was not observed adjacent to the conventional or resin-modified glass ionomer cement restorations.

Discussion

Polarized light microscopy is a method used for assessment of mineralized dental tissues and of secondary caries inhibition. Estimation of the demineralized lesions and existence of inhibition layers have been previously reported using this method (Hicks and Silverstone, 1984; Griffin et al., 1992; Souto and Donly, 1994; Dionysopoulos et al., 1994). However, little is known about the consistency i.e., height and width of the inhibition zone adjacent to the restorative materials. This may be an important factor to the long-term protection against secondary caries, and longevity of a restoration. Therefore, in order to permit accurate quantitative analyses of the inhibition zones through a polarized light microscope, we traced all cavity margins and measured the outer lesions and inhibition zones.

No statistically significant differences among the contours and depths of the outer lesions for the four materials were found. These results are similar to those reported by Skartveit et al., (1991) who compared the depths at the midpoints of the lesions and found no significant differences between groups studied. The same results were found by
Dunne et al., (1996) who concluded that there was no significant difference in depth of the body of the outer lesion among fluoride containing and non-fluoridated materials.

Regarding the inhibition zones, Fuji II produced a zone with greater width and height when compared with those produced by Fuji II LC and Vitremer. Because of differences in the formulations of these materials, a difference in their respective capacity to inhibit artificial caries may also exist. Caries resistance and formation of the inhibition zone appears to be associated with the level of fluoride release from the glass-ionomer restorations (Swift, 1989; Dionysopoulos et al., 1990; Donly, 1994).

However, previous studies indicated varying results concerning the amount of fluoride released from conventional and resin-modified glass ionomer cements. Diaz-Arnold et al., (1995) observed that a conventional glass ionomer cement released greater amounts of fluoride than a resin-modified glass ionomer cement. Takahashi et al.; (1993) found no statistically significant differences in fluoride release between Fuji II and Fuji II LC. Forsten (1995) later observed that fluoride levels released by a resin-modified glass ionomer cement were higher or the same as that of the conventional glass ionomer cement. An explanation for the variation in results may be the different methods used to determine fluoride release. Moreover, other factors such as material composition and release of other elements from the ionomeric materials may be more significant and may have greater influence on artificial caries inhibition than fluoride release alone.

The current study also evaluated the caries inhibition capacity of a fluoride releasing low viscosity resin composite. An inhibition layer was not identified adjacent to the Clearfil Liner Bond II system restoration. Similar results to ours have been presented regarding the lack of an inhibition layer adjacent to resin composites incorporated with fluoride (Griffin et al., 1992; Glasspoole and Erickson, 1993; Dionysopoulos et al., 1994). Although these materials release fluoride ions, the very low concentrations or slow release may not be sufficient to inhibit acid-attack demineralization.

Purton and Rodda, (1988) have reported that the demineralized outer lesion showed contraction away from the resin composite restorations. Because of the development of the dentin adhesive resins, the recent bonding system and fluoride releasing low viscosity resin that were used in this study bonded well to dentine and prevented separation of the material from the cavity margin (Kemp-Scholte et al., 1990 Burrow et al., 1994). The polarized light microscopy observations showed that the resin composite restoration was adhered to the artificially demineralized dentin even after the sectioning for sample preparation. Nevertheless, wall lesion was observed in 80% of the specimens. This suggests that wall lesion might occur even when separation at the cavity margin is not microscopically observed.

Dentine bonding is enhanced with the formation of a hybrid layer that is produced by demineralizing the dentine surface and later applying monomers that will infiltrate between the collagen fibers and will be polymerized within the subsurface of the
deminerlized dentin (Nakabayashi, 1984). Sano et al., (1995) reported that newer bonding systems that promote good adhesion to dentin showed penetration of silver nitrate through nano-sized spaces in the base of the hybrid layer (nanoleakage). Although the aim of this study was not to investigate leakage, it is possible to speculate that the demineralizing solution used in this study could have infiltrated through the base of the hybrid layer and be responsible for the formation of the wall lesions observed in this study (Figs. 2.1b and 2.2d). Therefore, recent adhesive resin systems may not be sufficient to inhibit secondary caries.

Because the current study is an in vitro evaluation, further in vivo studies on secondary caries inhibition around restorations with recent resin bonding systems are needed to clarify the relationship between adhesion, fluoride release, and caries inhibition.

Conclusions

Although all ionomeric restorative materials tested in this study produced an acid-resistant inhibition zone at the cavity margin, the dimensions of this zone were material dependent. The conventional glass ionomer cement produced the thickest zone, followed by the resin-modified glass ionomer cements. The recent bonding system that release fluoride failed to produce an inhibition zone and disclosed wall lesion at the cavity wall.

### Table 2.1. Restorative materials employed

<table>
<thead>
<tr>
<th>Material</th>
<th>Brand name</th>
<th>Content</th>
<th>Batch</th>
<th>Manufacturer</th>
</tr>
</thead>
<tbody>
<tr>
<td>Conventional glass-ionomer cement</td>
<td>Fuji Ionomer Type II</td>
<td>Conditioner: 10% polyacrylic acid P: Fluoro-aluminosilicate glass L: Acrylic-maleic acid copolymer, Polybasic carboxylic acid, Water P/L = 2.7 (g/g)</td>
<td>#071141</td>
<td>GC Corp., Tokyo, Japan</td>
</tr>
<tr>
<td>Resin-modified glass-ionomer cement</td>
<td>Fuji Ionomer Type II LC</td>
<td>Conditioner: 10% polyacrylic acid P: Fluoro-aluminosilicate glass L: Acrylic-maleic acid copolymer, HEMA, Water, CQ P/L = 3.0 (g/g)</td>
<td>#071241</td>
<td>GC Corp., Tokyo, Japan</td>
</tr>
<tr>
<td>Resin-modified glass-ionomer cement</td>
<td>Vitremer</td>
<td>Primer: 46% HEMA, 39% Ethyl Alcohol, P: Fluoro-aluminosilicate glass, potassium persulfate, ascorbic acid L: 50% Polycarboxylic acid copolymer, 20% HEMA, Water, 13% carboxylic acid copolymer</td>
<td>#3303L</td>
<td>3M Dental Products, MN, USA</td>
</tr>
<tr>
<td>Adhesive Resin</td>
<td>Clearfil Liner Bond II</td>
<td>Primers: Phenyl-P, 5-NMSA, HEMA, CQ, water Adhesive: Bis-GMA, MDP, HEMA, microfiller, CQ</td>
<td>#001 #0002</td>
<td>Kuraray Co., Osaka, Japan</td>
</tr>
<tr>
<td>Resin composite</td>
<td>Clearfil AP-X</td>
<td>Hybrid resin composite</td>
<td>#0029</td>
<td></td>
</tr>
</tbody>
</table>
Figure 2.1a. Schematic representation of the caries-like lesion and the inhibition zone around the conventional or resin-modified glass ionomer restoration.

Figure 2.1b. Schematic representation of the caries-like lesion adjacent to the fluoride-releasing adhesive composite restoration.
Figure 2.2a, b, c, d.  

a: Photomicrograph of a caries-like lesion and the inhibition zone formed adjacent to the conventional glass ionomer cement (Fuji II). Note that a thick inhibition zone (arrows) was formed despite the outer lesion (OL). Fuji II was lost during specimen preparation for polarized light microscopy. 

b: Photomicrograph of a caries-like lesion depicting the outer lesion (OL) and the inhibition zone (arrows) formed adjacent to Fuji II LC (FLC). 

c: Photomicrograph of a caries-like lesion and a thin inhibition zone (arrows) formed adjacent to Vitremer (V). In this sample the outer lesion (OL) seems shallower than the other samples, but not significantly different. Inhibition zone, OL = Outer lesion. 

d: Photomicrograph of a caries-like lesion and wall lesion (long black and white arrows) adjacent to the fluoride releasing adhesive resin composite (RC). Note that demineralization along the cavity wall extends beyond the length of the outer lesion (L) despite absence of gap at the cavity margin. Asterisk and white small arrow indicates bonding resin, and white long arrow indicates Protect Liner F.
Figure 2.3a, b, c. Graphs indicate mean depths of outer lesions, widths and heights of the inhibition zones, respectively. Bars indicate values that are not significantly different (p>0.05).
Chapter 3

Microhardness of *in vitro* caries inhibition zone adjacent to conventional and resin-modified glass ionomer cements

Introduction

The increase of retained teeth, particularly in the elderly, has accounted greatly for the development of dental materials designed for the treatment of root surface caries. Glass-ionomer cements are usually one of the restorative materials indicated for root surface lesions, due to their good adhesion to both enamel and dentin (Triana *et al.*, 1994), as well as fluoride release (Forsten, 1991; Takahashi *et al.*, 1993). However, due to poor mechanical and esthetic properties of conventional glass ionomer cements, these materials have been combined with resin components in order to improve these properties while still releasing fluoride (McCaghren *et al.*, 1990; Mitra and Kedrowski, 1994; Mount, 1994). The resin-modified glass-ionomer cements (McLean *et al.*, 1994) promote better esthetics, adhere better to tooth structure (Hinoura *et al.*, 1994; Pawlas *et al.*, 1994; Proado *et al.*, 1994), and possess the capacity to inhibit both *in vivo* and *in vitro* secondary caries (Souto and Donly, 1994; Griffin *et al.*, 1992).

Surface softening is one of the initial signs of dental caries. Hardness changes of the tooth surface may indicate the degree and extent of the carious lesion. Various studies have measured the microhardness of sound, demineralized, or remineralized enamel and dentin (Caldwell *et al.*, 1958; Craig and Peyton, 1958; Ryge *et al.*, 1961; Hegdal and Haebö, 1972; Davidson *et al.*, 1974; Arends *et al.*, 1979; Arends *et al.*, 1980; Herkströter *et al.*, 1989). However, micro hardness of the acid-inhibited zone in dentin adjacent to fluoride containing materials has not yet been reported.

The quality of the inhibition zone seems to be related to its degree of mineralization. Since mineral content contributes mainly to hardness of dental substrates (Davidson *et al.*, 1974), the micro hardness of this acid resistant zone may indicate the level of mineralization which could be an important factor in prevention of secondary caries. However, this hardness has not yet been measured, probably because of the minute width of the acid-inhibited layer, and large size of conventional hardness indenter tips. Since glass ionomer cements are the materials of choice for restoring root surface lesions, we focused on measuring the triangular hardness of the inhibition zones and lesions around these materials and correlating these with Knoop hardness numbers.
Materials and methods

Sample preparation for artificial caries lesions. Bovine incisors from 20-24 month old cattle were used within 24 hours after extraction. The cementum was manually removed using scalers to expose the underlying dentin. The roots were obtained by separating them from the crowns at the cementum-enamel junction with a low-speed diamond saw (Bronwill, NY, USA) using water coolant. In order to prevent dehydration of the dentin during the restorative procedure, the pulp tissue was left in situ, and the cut surface and root apex were sealed with wax and coated with two layers of nail varnish.

Two box-shaped cavities approximately 3 mm long, 2 mm wide, and 1.5 mm deep were prepared on both the buccal and lingual dentin surfaces of each root, using a diamond bur (ISO # 106) mounted in high-speed turbine with air-water coolant. The cavity margins were finished with a straight fisher steel bur (ISO #010) in a slow-speed handpiece under copious water spray, to achieve a cavosurface angle as close as possible to 90°. Twelve cavities were prepared for each material.

The materials used in this study were a conventional glass ionomer cement (Fuji II, GC Corp., Tokyo, Japan), and two resin-modified glass ionomer cements (Fuji II LC, GC Corp., Tokyo, Japan; and Vitremer, 3M Co., St. Paul, MN, USA) (Table 3.1). The cavities were conditioned according to each manufacturer's instructions. The cements were mixed at room temperature, transferred into a C-R syringe tip (Centrix™, Connecticut, USA) and injected into the cavity. Fuji II was allowed to set for 15 min, and Fuji II LC and Vitremer were light cured for 60 s each. The glass ionomer cement and resin-modified glass ionomer specimens were then coated with the manufacturer's recommended varnish. Because greatest release of fluoride has been shown to occur within the first week (Takahashi et al., 1993), the specimens were stored for one week in tap water at 37 °C. All restorations were then finished and polished flat with polishing disks (Rainbow polishing Kit, Shofu Inc., Kyoto, Japan) under running water. The integrity of each cavosurface margin was examined under a light microscope at 20x magnification. Two coats of acid-resistant nail varnish were then applied to the entire specimen surface, leaving a 1 mm-window around the cavity margins. Each specimen was stored in individual bottles, in which 20 mL of an acid buffer solution previously described by Wefel et al. (1995) was placed. The acid buffer contained 2.2 mmol/L CaCl₂, 2.2 mmol/L NaH₂PO₄ and 50 mmol/L acetic acid adjusted to pH = 4.5. Specimens were stored in the solution for three days at 37 °C.

The specimens were removed from the demineralizing solution, and thoroughly rinsed in running tap water. Longitudinal sections of approximately 150 μm thick were cut through each restoration, parallel to the long axis of the root by means of a water-cooled diamond saw microtome (Leitz 1600 Microtome, Wetzlar, Germany). The sections were ground to approximately 100 μm thick using coarse and fine wet stones.
All sections were dehydrated in gradations of ethanol, immersed in quinoline, and observed under polarized light microscope (PM-10AK, Olympus Inc., Tokyo, Japan) in order to confirm formation of outer lesions and inhibition zones.

**Procedures for Micro Hardness Determination.**
Five dentin slabs for each material were randomly selected and embedded in epoxy resin. Special care was taken so that the surface to be evaluated would remain free from the embedding material. The samples were polished manually to a high gloss using diamond pastes of successively smaller (6, 3, 1 and 0.4 μm) grits. Between each step, the samples were cleansed in ultrasonic bath for 1 min, in order to remove remnants of polishing debris and paste.

First, Knoop hardness number (KHN) of the outer lesion and underlying normal dentin was determined. Then, because the size of the Knoop indenter was greater than the width of the inhibition zone, triangular hardness (HT) of this layer was measured. The HT of the demineralized dentin adjacent to the knoop indentations was also measured (Fig. 3.1).

**Knoop Hardness.** The specimens were mounted on the stage of the Knoop Hardness apparatus (MXT70, Matsuzawa, Tokyo, Japan) in a horizontal position. The indentation process was made perpendicular to the surface with a 5 g force load for 15 s. The first indentation was made 100 μm from the top of the demineralized lesion. Subsequent indentations were made at 50 μm intervals parallel to the cavity margin and towards the normal underlying dentin. The surface area of the plastic deformation of the indentation was measured by means of a video control connected to the light microscope, and converted to KHN automatically. Fifteen indentations were performed on each lesion totaling 75 indentations for each material.

**Triangular Hardness.** A micro-hardness tester (DUH-200 Shimadzu, Kyoto, Japan) was used to determine the triangular hardness of both the demineralized lesion and inhibition zone. The micro-hardness tester apparatus consists of three major components: a triangular pyramidal indenter with an apex angle of 115°, an optical microscope with a maximum magnification of 500x, and a X-Y test piece stage that transports the specimen between the microscope and the indenter with a high lateral precision of 0.01 μm (information provided by the manufacturer).

In order to obtain homologous hardness at similar points (Fig. 3.1), the triangular micro indenter was positioned and loaded precisely adjacent to the knoop indentations (n = 15) on the demineralized lesion and on the underlying sound dentin. To measure the hardness of the inhibition zone, the indenter was first placed at a distance 100 μm from the top of this layer. Subsequent indentations (n = 15) were performed at 50 μm intervals
parallel to the long axis of the restoration wall. A load of 2 gf was applied to the triangular indenter tip, at a loading speed of 0.029 g/s. During the indentation process, the load, as well as the loading rate were continuously monitored. The diagonal length of the triangular indentation was determined through the microscope device, and the triangular hardness obtained according to the formula: \( HT = \frac{1569.7 \ p}{1^2} \) \( HT = \) triangular hardness, \( p = \) test load gf, and \( l = \) height of indentation (μm).

Specimen preparation for SEM and EDS Line-Analyses. Following hardness measurements, the samples were gold-coated, and the micromorphology of the inhibition layer and outer lesion were observed under a Scanning Electron Microscope (SEM; JXA-840 JEOL, Tokyo, Japan). The plastic deformation of dentin caused by the triangular indenter on the inhibition layer and outer lesion was recorded photographically.

For analyses of calcium and phosphorous in the inhibition zones and outer lesions, the specimens were again polished with diamond pastes, and then carbon coated. They were line-analyzed with an energy-dispersive x-ray spectrometer (EDS) (SED800, Seiko EG&G Co., Tokyo, Japan) attached to the SEM.

Statistical Analysis. Triangular and Knoop hardness were analyzed by correlation coefficient. Hardness measurements at corresponding depths along the inhibition zone and demineralized lesion were analyzed using one-way analysis of variance (ANOVA) and Fisher's PLSD Test at the 95% level of confidence.

Results

Polarized light microscopy. Demineralized lesions (outer lesions) and inhibition zones along the cavity walls were observed in all samples (Figs. 3.2a and b).

Hardness measurements. The relationship between KHN and HT was analyzed by correlation coefficient. The correlation was highly significant \( (r^2 = 0.81, p < 0.05) \), and a linear relation was found between Knoop and triangular hardness numbers (Fig. 3). The equation was \( y = 0.93x + 2.40 \), indicating that triangular hardness number was almost identical to Knoop hardness number in the case of softened and normal dentin.

Mean values and standard deviations of the triangular hardness of the outer lesions and inhibition layers created by the different materials are depicted in Fig. 3.4. The geometry of both Knoop and triangular indentations on the inhibition zone and on the outer lesion, are presented in Fig. 3.5.

Microhardness of the demineralized lesion was of 28.7 ± 5.9 HT, whereas hardness of the underlying normal dentin 52.4 ± 5.7 HT. The mean triangular hardness of the inhibition zone adjacent to the Fuji II restoration was of 59.2 ± 3.8 HT, values which were not significantly different from those of the sound dentin. The hardness of the inhibition layer created by Fuji II was identical throughout the depth of this layer. Fuji II LC produced an inhibition layer with an average HT number of 48 ± 3.5 and like Fuji II,
the hardness of this layer was constant at all depths. Vitremer produced an inhibition layer with the initial hardness being similar to that of the outer lesion (28.8 ± 8.8 HT and 26.9 ± 3.8 HT, respectively). Nonetheless, this hardness gradually increased as the distance from the top increased, and at 300 μm, it was identical to that of Fuji II LC.

**Calcium and Phosphorous content.** Line-analyses indicated the existence of calcium and phosphorous in the inhibition layer adjacent to Fuji II, as well as Vitremer and Fuji II LC (Fig. 3.6). This correlates well with the triangular hardness findings.

**Discussion**

Bovine teeth were selected as a substitute for human teeth because of the large number of teeth required and human teeth are scarce. The size of bovine teeth simplifies the experimental procedures being beneficial for screening different products.

An acidic buffered solution (pH = 4.5) was used in this study, which does not simulate the oral environment where the pH tends to fluctuate. However, it permitted rapid and consistent formation of outer lesions and inhibition zones that are exemplified in Figs. 3.2a and b. The resulting lesions and inhibition zones were comparable to previous studies which used different acidic solutions and pH (Silverstone, 1967; Swift, 1989; Almqvist and Lagerlof, 1993).

Considerable research has been carried out investigating demineralization, remineralization, and fluoride uptake by the enamel and dentin (Souder and Schoonover, 1944; Swartz and Phillips, 1952; Caldwell et al., 1958; Craig and Peyton, 1958; Hegdahl and Haebö, 1972; Davidson et al., 1974; Arends et al., 1979; Arends et al., 1980; Herksröter et al., 1989). However, microhardness of the inhibition zones created by various fluoride-releasing materials has not been measured. Arends et al., (1980) reported that an empirical linear relationship exists between the lesion depth of artificial enamel carious lesions and Knoop microhardness indentations. Their findings, although for enamel, corresponded with our results for dentin considering the extent of the demineralized lesion.

Because the size of the Knoop hardness indentation on sound bovine dentin was approximately 40 μm at the minimal load (5g) of the apparatus, and it is larger than the width of the inhibition layer (maximum 30 μm), we also utilized a triangular micro indenter with an apex angle of 115° to measure the hardness of the inhibition zone. Indentations were also performed on the outer lesion, adjacent to the Knoop indentations to correlate the two measuring methods. The Knoop hardness results showed a linear correlation with those for triangular hardness ($r^2 = 0.81, p < 0.05$) (fig. 3.3), indicating that triangular hardness testing is a good alternative for measuring narrow surfaces.

The mean values of inhibition zones created around Fuji II LC and Vitremer were 48.3 ± 3.5 HT and 44.0 ± 7.6 HT, respectively (Fig. 3.4). These observations suggest that
within our testing parameters, the resin-modified glass ionomer cements produced a softer inhibition zone compared with that of the conventional glass ionomer cement. Our speculations are that the incorporation of resin components into the glass ionomer cement may reduce uniform release of fluoride and other components to the adjacent cavity wall. This could result in a weaker inhibition zone with areas within this zone being more susceptible to demineralization by secondary caries. Line analysis for calcium and phosphorous content confirmed the microhardness results (Fig. 3.6). A greater content of calcium and phosphorous was found in the inhibition layer, but failed to appear in the outer lesions. Fuji II produced an inhibition zone with a greater microhardness and width compared to those formed adjacent to Fuji II LC and Vitremer (Fig. 3.4). Because of the differences in the formulations of these materials, a difference in their respective capacity to inhibit caries may also exist. Although formation of this layer appears to be associated with the level of fluoride release from the restorative material (Swift, 1989; Donly, 1994), conflicting results regarding the amount of fluoride release and caries inhibition from conventional and resin-modified glass ionomer cements have been presented (Forsten, 1991; Forss, 1993; Takahashi et al., 1993; Dunne et al., 1996; Diaz-Arnold et al., 1995; Forsten, 1995). Takahashi et al., (1993) found no statistically significant differences in fluoride release between Fuji II and Fuji II LC, and Dunne et al., (1996) observed that Fuji II LC provided similar in vitro caries inhibition to a conventional glass-ionomer cement. However, Diaz-Arnold et al., (1995) observed that a conventional glass ionomer cement released greater amounts of fluoride than a resin-modified glass ionomer cement after 24 h storage. Forsten (1995) later observed that fluoride levels released by a resin-modified glass ionomer cements were higher or the same as that for a conventional glass ionomer cement. An explanation for the variation in these results may be the different content of fluoride in the powder and different methods used to determine fluoride release.

Another report has suggested that glass-ionomer cements may promote hypermineralization of carious lesions by possibly depositing minerals, therefore increasing acid resistance (ten Cate and van Dunned, 1995). In the oral environment in addition to pH, saliva proteins and buffer systems may affect fluoride release and dissolution, as well as erosion of non-soluble components of glass ionomer cements (Rezk-Lega et al., 1991; Forss, 1993). Thus, factors such as material composition and release of other elements from the glass ionomers may be more significant and may have a greater influence on formation of the inhibition zone, other than fluoride release alone.

In conclusion, a linear correlation between triangular and Knoop hardness numbers was obtained. The triangular hardness test was demonstrated to be excellent for examining very narrow areas. Hardness of the inhibition zone adjacent to the conventional glass ionomer cement was significantly higher than that of the inhibition zones adjacent to the resin-modified glass ionomer cements.
Table 3.1. **Restorative materials employed**

<table>
<thead>
<tr>
<th>Material</th>
<th>Brand name</th>
<th>Content</th>
<th>Batch</th>
</tr>
</thead>
<tbody>
<tr>
<td>Conventional Glass-ionomer Cement</td>
<td>Fuji Ionomer Type II</td>
<td>Conditioner: 10% polyacrylic acid, Powder: Fluoro-aluminosilicate glass, Liquid: Acrylic-maleic acid copolymer, Polybasic carboxylic acid, Water, P/L ratio = 2.7(g/g)</td>
<td>071141</td>
</tr>
<tr>
<td>Resin-modified Glass-ionomer Cement</td>
<td>Fuji Ionomer Type II LC</td>
<td>Conditioner: 10% polyacrylic acid, Powder: Fluoro-aluminosilicate glass, Liquid: Acrylic-maleic acid copolymer, HEMA, Water, CQ, P/L ratio = 3.0(g/g)</td>
<td>071241</td>
</tr>
<tr>
<td>Resin-modified Glass-ionomer Cement</td>
<td>Vitremer Primer 46% HEMA, 39% Ethyl Alcohol, Powder: Fluoro-aluminosilicate glass, potassium persulphate, ascorbic acid, Liquid: 50% Polycarboxylic acid copolymer, 20% HEMA, Water, 13% carboxylic acid copolymer</td>
<td>3303P, 3303A3, 3303L</td>
<td></td>
</tr>
</tbody>
</table>

Chemical Names for Abbreviations: HEMA: 2-hydroxy ethyl methacrylate; CQ: Camphorquinone
Figure 3.1. Schematic diagram of specimens subjected to acid demineralization. It represents a specimen with an inhibition layer adjacent to the restoration. Large indentations represent KHN measured on the outer lesion and intact dentin. Small triangular indentations represent HT measured both on outer lesion and inhibition zone.

Figure 3.2. Polarized light microscope photographs of artificial caries lesions and inhibition layers adjacent to (A) Conventional GIC and (B) Resin-modified GIC. The conventional GIC was lost during slicing procedure. R= Restorative Material, OL= Outer Lesion, IZ= Inhibition Layer. Magnification x 20.
Figure 3.3. Relationship between Knoop and triangular hardness numbers.

Figure 3.4. Mean values of HT measured along the depth of the inhibition layers and outer lesions. Note that differences between hardness of IZ for the different materials and outer lesion became insignificant beyond 400 µm.
Figure 3.5. SEM photograph of outer lesion an inhibition layer. The arrows indicate triangular hardness indentations. Asterisks indicate Knoop hardness indentations. IZ = Inhibition zone, OL = Outer lesion. Magnification x 300.
Figure 3.6. SEM photograph and line-analysis of calcium and phosphorous contents in the inhibition layers produced by Fuji II. Note existence of calcium and phosphorous on the inhibition layer and absence in the outer lesion. IZ = Inhibition layer, OL = Outer lesion. Magnification x 300.
Chapter 4

Bond strength and interface micromorphology of an improved resin-modified glass ionomer cement.

Introduction

Glass ionomer cements possess unique properties as a restorative material, leaching fluoride and bonding dynamically to tooth structure through ion exchange (McLean, 1988). However, they are very technique sensitive, due to water uptake and loss, they lack toughness, and they exhibit a high degree of porosity (McLean and Wilson, 1977).

Recently, by mixing the glass ionomer cement constituents with resinous components, a new generation of restorative cements has been developed, namely resin-modified glass ionomer cements (RmGIC). The RmGIC undergoes a two-type setting reaction. First, an acid/base reaction occurs by mixing the glass powder and the polyalkenoic liquid. Then, with exposure to visible light, polymerization takes place, due to the incorporation of methacrylate groups and light activators (Watson, 1990; Mitra, 1991). The RmGIC retains the beneficial properties of the conventional glass-ionomer cements, such as releasing fluoride and physicochemical bonding to dental tissue, but with improved mechanical properties, working time, esthetics, and decreased sensitivity to water (Mitra, 1991; Walls, 1986; Mount, 1993). Therefore, these materials have become popular as an esthetic filling material to restore carious lesions or for patients with a high caries risk.

Since the conventional glass ionomer cements can adhere physicochemically to the tooth structure, they were initially placed directly into the cut cavity, without any prior surface treatment. However, the smear layer which covers the cut dental surfaces (Pashley, 1984) may break cohesively and fail during polymerization shrinkage (Tao and Pashley, 1988). It was reported that bond strength to dental tissue without pre-treatment was inconsistent (Hewlett et al., 1991; Ngo et al., 1986) and that clinical retention (Ngo H et al., 1986) and shear bond strength to dentin could be improved by removing the smear layer (Powis et al., 1982). Since then, pre-treating the cavity walls with acid solutions became routine before restoring with a glass ionomer or a resin-modified glass ionomer cement.

Polyacrylic acid is an effective pre-treatment agent, however, to date there is no consensus as to the optimal concentration and application time. Fuji II LC requires dentin conditioning with a 10% polyacrylic acid solution (DC) for 20 seconds to achieve greater bond strengths. With the attempt to shorten operating time and create prompt bond
strengths to both enamel and dentin, a 20% polyacrylic acid conditioner containing 3% \( \text{AlCl}_3 \) to be applied for 10 s was developed, along with an improved version of Fuji II LC which is superior in polishability by minimizing powder particle size. The purposes of this study were to evaluate the effect of the new cavity conditioner (CC) and improved Fuji II LC (Fuji II LC-I), on shear bond strength to dentin and enamel, and to analyze the micromorphology appearance of the cement/tooth interface.

Materials and Methods.

Ninety-six crowns of stored frozen bovine incisors were cut from their roots at the cementum-enamel junction, taking care not to remove the pulp tissue. The teeth were divided into enamel and dentin substrates for bonding groups, and then each group subdivided into three subgroups: a non-treatment group (control) \((n = 12)\), a group treated with 10% polyacrylic acid (DC+Fuji II LC) \((n = 36)\), and a group treated with 20% polyacrylic acid containing 3% \( \text{AlCl}_3 \) (CC+Fuji II LC-I) \((n = 36)\). The enamel and dentin surfaces were ground flat, and finished with # 600-, 800- and 1000- grit silicon carbide paper in order, under running water. To prevent the embedding medium from entering the pulp chamber, the entrance was covered with wax. The teeth were then embedded in plaster taking care not to contaminate the prepared surface which stood several millimeters above the plaster. A double-sided adhesive vinyl tape with a hole of 3 mm diameter to demarcate the area for bonding was placed on the prepared surface. No surface treatment was performed for the control group. The surfaces were treated either with DC for 20 seconds or CC for 10 seconds, rinsed and air dried. Acrylic tubes of 4 mm diameter x 2 mm height were placed over the demarcated area. The RmGICs and conditioners used in this study, manufacturer, and compositions are listed in Table 4.1. Fuji II LC and Fuji II LC-I were mixed following the manufacturer's instructions and while the surface was still glossy, it was transferred to a C-R syringe tip (Centrix™, Connecticut, USA) and bulk filled into the respective tubes. Excess material was pressed into the tubes with a glass slide and light cured from the top for 60 seconds. Except for the specimens (dentin: \( n = 12 \); enamel: \( n = 12 \)), which were tested 5 minutes after light curing to test early bond strength, the samples were stored 24 hours or 1 week in tap water at 37°C. Before testing the specimens, the vinyl tape was carefully removed so that the shear blade could be positioned closer to the junction of the tooth and cement interface. The jig used to for the shear bond test (ISO standard TR 110405) was mounted in an universal testing machine (Autograph AG-500B, Shimadzu Co., Kyoto, Japan), and the specimens were loaded at a crosshead speed of 1 mm/min. The debonded surfaces were examined visually, and with a stereomicroscope at 20x magnification. The results were analyzed by two-way analysis of variance (ANOVA) and Fisher's PLSD Test.

For the ionomer-tooth interface SEM observation, erupted non-curious human
third molars which had been stored frozen were used. The bonded specimens were prepared using the sandwich technique, described by Inokoshi et al., (1990). The specimens were stored in water for 1 day at 37°C. The bonded 'sandwiches' were then sectioned perpendicular to the flat dentin surface, into approximately equal halves. The two-halves were embedded in a self-curing epoxy resin. After overnight cure, the surfaces of the cut specimens were polished to high gloss with abrasive discs and diamond pastes successively up to 0.25 μm grit size. The specimens were gold-sputter-coated and the polished surfaces observed under an SEM (JXA-840, JEOL, Tokyo, Japan).

After SEM observation, the polishing steps were repeated and the specimens were etched with argon-ion-beam for 270 seconds (Inokoshi et al., 1990) (E1S-1E, Elionix Ltd., Tokyo, Japan). The samples were then gold-sputter-coated and observed again under an SEM.

The specimens were then re-polished with diamond pastes and were subjected to 10% phosphoric acid treatment for 3 to 5 seconds (Gwinett and Kanca, 1992; Sano et al., 1995) followed by 5% sodium hypochlorite immersion for 5 minutes (Wang and Nakabayashi, 1991) for dissolution of the mineral content and removal of exposed collagenous material of the dentin. After being thoroughly rinsed in water, the treated specimens were air-dried, gold-sputter-coated, and observed with the SEM.

For analysis of the effect of dentin pre-treatment with both acid solutions, human dentin disks of approximately 2 mm thick were cut, and finished with 1000-grit silicon carbide paper. The control specimen was observed without any pre-treatment. The remaining samples were treated actively with DC (10% polyacrylic acid) for 20 sec or CC (20% polyacrylic acid containing AlCl3 ) for 10 seconds. The dentin disks were immersed in 2-Methyl-2-Propanol for one hour, and freeze-dried at -20°C. The specimens were then immediately gold sputter-coated and observed with the SEM.

Results

The resulting shear bond strength values and standard deviations are shown in Tables 4.2 and 4.3. The control group results, which were measured at one day are shown in Table 4.4. The results of two-way ANOVA disclosed no statistically significant interaction between differences of conditioners and storage time for enamel and dentin. For the enamel specimens, no significant difference existed between bond strengths of the two RmGICs systems at 5 minutes. As expected, significant higher bond strengths were obtained for both materials at 1 day, and remained stable after 1 week. However, significantly higher bond strengths to enamel were obtained at 1 day and 1 week for CC+ Fuji II LC-I, compared to those obtained with DC+ Fuji II LC.

Interestingly, for the dentin specimens, the 5 minutes bond strength of CC+ Fuji II LC-I was significantly higher than that of DC+Fuji II LC. However, the bond
strengths of both materials were similar at 1 day and 1 week. What was initially thought by visual inspection to be adhesive failure, turned out being a mixture of adhesive and cohesive material failure, very close to the bonded area for most of the specimens, when observed with the stereomicroscope.

SEM observations of the polished and argon-ion etched interfaces of DC+Fuji II LC/dentin and CC+ Fuji II LC-I/dentin are shown in Figs. 4.1 and 4.2. In these cross-sections, a resin-rich layer and an indistinct zone (approximately 1μm thick) were noted between the RmGICs and the underlying dentin. Tags which were formed by the resinous part of the RmGICs and which included small glass particles were observed (Fig. 4.3). An indistinct zone beneath the CC+Fuji II LC restoration was also observed following argon-ion etching (Fig.4.4). When the specimens were exposed to phosphoric acid followed by sodium hypochlorite, the existence of a resin-rich layer overlying the demineralized dentin was confirmed (Figs. 4.5 and 4.6). All cross-sections, including the control group, showed resinous tag formation when subjected to phosphoric acid sodium hypochlorite treatment, suggesting that the polyacrylic acid incorporated in the cement, can easily penetrate the tubules (Figs. 4.5 and 4.6).

The morphological characteristics of the dentin surface followed by DC and CC pre-treatment are presented in Figs. 4.7 - 4.10. Both treatments succeeded to remove the smear layer, but varied slightly in the amount of opening of tubule orifices. A clear difference was not observed between the degree of demineralization between both conditioners.

**Discussion**

Because of the large number of teeth required, and the lack of plentiful human teeth, bovine teeth were selected as a substitute for human teeth for the bond strength portion of this study. It has been previously shown that the size of bovine teeth may eliminate some factors that may influence testing for bond strengths (Suzuki and Finger, 1988; Tagami et al., 1993) and simplifies the experimental procedures. In addition, little or no difference was observed for bond strength tests when comparing human and bovine teeth (Nakamichi et al., 1983; Fowler et al., 1992). Therefore, the use of bovine incisors is beneficial for screening the experimental products.

The smear layer (Fig. 4.11) may cohesively break or fail during shear bond tests or during polymerization shrinkage of the restorative material (Hewlett et al., 1991). As shown in Fig. 4.5, although resinous tags could be formed, the underlying dentin was susceptible to sequential acid/base treatment which removed the smear layer and some of the underlying dentin (Gwinnett and Kanca, 1992). The polyacrylic acid of the cement failed to penetrate into the intertubular dentin or to improve bond strengths. By modifying or removing the smear layer, Hinoura et al. (1991) showed that this problem
could be overcome. The results of this study correspond with previous reports, and confirm the fact that RmGICs provide considerable bond strengths, when they followed an acidic pre-treatment of dentin or enamel (Holtan et al., 1990; Bell and Barkmeier, 1994; Triana et al., 1994). Pre-treatment of dentin with acidic conditioner removes the smear layer and demineralizes the superficial dentin layer, allowing the HEMA in the RmGICs to penetrate the exposed collagen network (Friedl et al., 1995; Titley et al., 1996).

The shear bond strengths to dentin found in this study for DC+Fuji II LC at 1day and 1week (14.4 and 13.2 MPa, respectively) correspond to values previously reported ranging between 11 and 15 MPa (Bell and Barkmeier, 1994; Triana et al., 1994; Friedl et al., 1995; Charlton and Havemann, 1994; Kato et al., 1995; Fritz et al., 1996). Our enamel bond strengths for 1day and 1week were 15.9 and 16.3 MPa respectively, compared to previously reported values (Charlton and Havemann, 1994; Kato et al., 1995; Burgess and Burkett, 1993; Swift et al., 1995) ranging from 9 to 14 MPa. On the other hand, the new system (CC+ Fuji II LC-I), showed greater bond strength to dentin 5 minutes after light curing, remaining statistically unchanged to 1day and 1week periods. This is possibly the greatest clinically-related advantage of this new system over DC/Fuji II LC, yielding higher early bond strengths after light curing, and shortening operating time.

Regarding bonding to enamel, both bonding systems produced low early bond strengths (i.e. 5 minutes), which increased after 1 day. However, bond strengths for CC+ Fuji II LC-I were significantly greater at 1 day and 1week. The reason for the different results between early bond strengths at 5 minutes to enamel and dentin is still unclear, but may be due to the difference in nature and water content of the substrates.

Through visual inspection, most of the specimens appeared to show an adhesive fracture pattern. However, when examined under 20x magnification, the surfaces seemed to be covered with a fine film, suggesting a cohesive fracture close to the bonded surface within the cement. This film has been previously observed and classified by Berry and Powers (1994).

As an additional study (unpublished data), the RmGICs were exchanged, so that Fuji II LC was applied to the CC-treated surface, and Fuji II LC-I was applied to the DC-treated surface. These results indicated similar data as the ones obtained in this study, indicating that slight differences in bond strengths were due to the change in pre-treatment conditioner. The CC is composed of 20% polyacrylic acid containing 3% AlCl₃. It is thought that AlCl₃ strengthens the collagen fibers after demineralization (information provided by the manufacturer).

The interface micromorphology between Fuji II LC/dentin and Fuji II LC-I/dentin showed tags within in the dentinal tubules, formed by the resinous part of the material, which were also previously described (Kato et al., 1995). Although it may be assumed that CC would produce a slightly greater depth of demineralization of the dentin surface (Fig. 4.2), the bond strengths of both materials to dentin were not significantly
different. Other than resin tags, a resin-rich layer and an indistinct zone were observed between the RmGICs and the underlying dentin. The addition of AlCl₃ may have avoided the complete collapse of demineralized collagen, enabling perhaps, an optimal permeability of HEMA, which is necessary for maximal bond strength (Van Dijken, 1992).

Microphotographs of Fuji II LC or Fuji II LC-I bonded to enamel showed similar interface appearance, although significantly different bond strengths were obtained at 1 day and 1 week. Carvalho et al. (1995) have suggested that micromechanical interlocking may exist at the interface of a RmGIC. Our SEM observations did not clarify if other mechanisms, besides micromechanical interlocking, are responsible for the higher bond strengths.

The new version of RmGIC with a cavity conditioner reduces operating time, and showed significant improvement in early bond strength both to enamel and dentin. Being by nature a hybrid material, which combines the beneficial properties of conventional ionomers and resin composites, release fluoride, and bonds efficiently to enamel and dentin, this new system may be an excellent restorative material for caries-prone patients.

Table 4.1. RmGICs, conditioners, compositions, batch numbers, and manufacturer.

<table>
<thead>
<tr>
<th>Brand name</th>
<th>Abbreviation</th>
<th>Content</th>
<th>Batch</th>
<th>Manufacturer</th>
</tr>
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<tbody>
<tr>
<td>Fuji Ionomer</td>
<td>Fuji II LC</td>
<td>P: Fluoro-aluminosilicate glass L: Acrylic-maleic acid copolymer, HEMA, Water, CQ P/L ratio = 3.0g/1.0g conditioner: 10% polyacrylic acid</td>
<td>#071241</td>
<td>GC Corp.</td>
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<tr>
<td>Type II LC</td>
<td></td>
<td></td>
<td>#291141</td>
<td>Hasunuma, Tokyo, Japan</td>
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<tr>
<td>Fuji Ionomer</td>
<td>Fuji II LC-I</td>
<td>P: Fluoro-aluminosilicate glass L: Acrylic-maleic acid copolymer, HEMA, Water, CQ P/L ratio = 3.2g/1.0g conditioner: 20% polyacrylic acid containing 3% AlCl₃</td>
<td>#120161</td>
<td>GC Corp.</td>
</tr>
<tr>
<td>Type II (Improved)</td>
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<td></td>
<td>#170161</td>
<td>Hasunuma, Tokyo, Japan</td>
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### Table 4.2. Shear bond strength (MPa ± S.D.) of Fuji II LC and Fuji II LC-I to enamel.

<table>
<thead>
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<th></th>
<th>5 min</th>
<th>1 day</th>
<th>1 week</th>
</tr>
</thead>
<tbody>
<tr>
<td>DC+Fuji II LC</td>
<td>4.0 (1.7)</td>
<td>15.9 (1.8)</td>
<td>16.2 (2.6)</td>
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<tr>
<td>CC+Fuji II LC-I</td>
<td>6.7 (3.1)</td>
<td>20.1 (6.6)</td>
<td>19.7 (1.5)</td>
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Bars indicate no statistically significant difference among figures.

### Table 4.3. Shear bond strength (MPa ± S.D.) of Fuji II LC and I-Fuji II LC to dentin.

<table>
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<th>5 min</th>
<th>1 day</th>
<th>1 week</th>
</tr>
</thead>
<tbody>
<tr>
<td>DC+Fuji II LC</td>
<td>9.5 (4.0)</td>
<td>14.4 (3.0)</td>
<td>13.2 (1.3)</td>
</tr>
<tr>
<td>CC+Fuji II LC-I</td>
<td>13.4 (2.2)</td>
<td>14.4 (5.7)</td>
<td>16.4 (2.0)</td>
</tr>
</tbody>
</table>

Bars indicate no statistically significant difference among figures.

### Table 4.4. Shear bond strength (MPa ± S.D.) of Fuji II LC and Fuji II LC-I to untreated enamel and dentin (1 day).

<table>
<thead>
<tr>
<th></th>
<th>Enamel</th>
<th>Dentin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fuji II LC</td>
<td>8.7 (5.1)</td>
<td>5.8 (4.0)</td>
</tr>
<tr>
<td>Fuji II LC-I</td>
<td>3.5 (1.0)</td>
<td>5.1 (3.0)</td>
</tr>
</tbody>
</table>

Bar indicates no statistically significant difference among figures.
Figure 4.1. Polished surface of DC+ Fuji II LC/dentin interface. A resin rich layer (L) was observed between the RmGIC (R) and underlying dentin (D). SEM. Bar = 1μm.

Figure 4.2. Polished surface of CC+ Fuji II LC-I/dentin interface. Arrows indicate an indistinct zone between the RmGIC (R) and dentin (D). SEM. Bar = 1μm.
Figure 4.3. DC+ Fuji II LC/dentin interface after argon-ion etching. RmGIC (R) penetrated the dentin tubules (T). Arrows indicate glass particle within the tag. SEM. Bar = 1μm.

Figure 4.4. CC+ Fuji II LC-I/dentin interface after argon-ion etching. Arrows indicate an indistinct zone between the RmGIC (R) and the underlying intact dentin (D). SEM. Bar = 1μm.
Figure 4.5. RmGIC/dentin interface of the control group (without surface pre-treatment) after treatment with 10% phosphoric acid and 5% sodium hypochlorite. Resinous tag formation (T) is observed, however, separation between the RmGIC (R) and the underlying dentin (D) has occurred. SEM. Bar = 1μm.

Figure 4.6. DC+ Fuji II LC/dentin interface after treatment with 10% phosphoric acid and 5% sodium hypochlorite. Resinous rich layer (L) and tag formation (T) are observed, without separation at the interface. SEM. Bar = 1μm.
Figures 4.7 and 4.8. Following conditioning with 10% polyacrylic acid for 20 s, and 20% polyacrylic acid for 10 s respectively, the collagenous intertubular network became evident. The tubules are superficially widened due to slight loss of peritubular dentin, however, no significant difference in demineralization degree was noted between both conditioners. SEM Bar = 1μm.
Figures 4.9 and 4.10. Following conditioning with 10% or 20% polyacrylic acid for 20 s and 10 s respectively, the dentin disks were freeze-dried and fractured. No significant difference of degree of demineralization was noted. SEM. Bar = 10μm.
Figure 4.11. Dentin smear layer produced by grounding with sequentially finer abrasives up to 1000-grit silicon carbide paper. SEM. Bar = 1μm.
Chapter 5

Adhesion of resin-modified glass ionomer cements using resin bonding systems

Introduction

Since the development of glass ionomer cement by Wilson & Kent (1972), these materials have undergone many modifications and improvements. The incorporation of methacrylate groups to conventional glass ionomers improved handling and working characteristics (Antonucci et al., 1988; Mitra, 1989), as well as reducing their brittleness, moisture sensitivity, and improving bond strengths to the tooth (Mathis and Ferracane, 1989; Holtan et al., 1990; Wilson, 1990; Mitra, 1991). Resin-modified glass ionomer cement (RmGIC) is considered a good substitute for dentine in deep cavities, due to their physical properties, while resin composite is a good substitute for enamel because of their superior strength, surface integrity, and esthetics (McLean, 1996). Adhesion of these materials to enamel has been successfully achieved; however, with regard to dentine, forces of contraction of the RmGIC during light irradiation may lead to failure of the bond between the RmGIC and dentine.

Volumetric shrinkage during polymerization of the RmGIC (Feilzer et al., 1988) can affect bond strength of this material to dentine. While initial set is rapid by light activation, the chemical setting reaction continues for up to 24 hours, and is responsible for the final physical properties of the cements (Bourke et al., 1992). Similar to resin composites, RmGICs shrink approximately 3% in volume during setting (Feilzer et al., 1988). Although this intrinsic stress can be relieved by later water sorption (McLean, 1996, Davidson and De Gee, 1984) the initial polymerization shrinkage of the RmGICs may lead to adhesive failure at the tooth-filling interface compromising the longevity of the restoration (Ciucchi et al., 1997).

Bond strength of RmGICs to dentine is inconsistent when the smear layer is not removed (Helwett et al., 1991), probably because the smear layer can break cohesively and fail as a result of the polymerization shrinkage (Tao and Pashley, 1988). Pre-treatment with polyacrylic acid removes the smear layer and improves the bond strengths of the RmGICs to dental tissue (Triana et al., 1994; Carvalho et al., 1995; Swift et al., 1995). Other conditioners that demineralize the dentine surface have also shown improved bond strengths, suggesting the importance of micromechanical-bonding (Carvalho et al., 1995; Smith, 1992; Titley et al., 1996).

Increase of the bond strength of RmGIC to dentin and consequent prevention of immediate separation at the interface may be altered by first bonding the dentin with an adhesive resin, which has demonstrably greater bond strengths. This study aimed to
evaluate the effect of three adhesive resin systems on shear bond strength of RmGIC to dentine, and to analyze the interfacial micromorphology of the cement/adhesive/tooth interface.

Materials and methods

*Specimen Preparation for Shear Bond Strength Test*

The resin-modified glass ionomer cements, adhesive systems used in this study, and their manufacturers are listed in Table 5.1. The adhesive systems used in this study were from the same manufacturer and selected because Clearfil Photo Bond is an acid-etching system that does not require priming, Clearfil Liner Bond is an acid-etching system that requires priming, and Clearfil Liner Bond II is a self-etching primer system. The crowns of 96 bovine incisors, stored frozen before use, were used in this study. The crowns were cut from their roots at the cementum-enamel junction, taking care not to remove the pulp tissue to avoid dehydration of the specimen. Superficial flat dentine surfaces were prepared, and finished with 1000-grit silicon carbide paper under running water. The teeth were embedded in dental stone taking care not to contaminate the prepared surface, which protruded several millimeters above the stone. To prevent the embedding medium from entering the pulp chamber, the entrance was covered with dental wax. After the stone had set, a double-sided adhesive vinyl tape, in which a hole of 3 mm in diameter had been punched, was placed on the prepared surface to demarcate the bonding area. The teeth were then randomly divided into 4 groups for bonding:

**Control group:** Surface treatment was performed following each manufacturer's instructions. For the specimens to be bonded with Fuji II LC, the dentin substrate was treated with 10% polyacrylic acid for 20 seconds, rinsed with water and gently air-dried. For the specimens to be bonded with Vitremer, the primer was applied to the surface for 30 seconds, gently air-thinned, and light-cured (Optilux 400, Demetron/Kerr, Danbury, CT) for 20 seconds. Twelve specimens were prepared for each material.

**Clearfil Photo Bond group:** The surfaces were etched with 37% phosphoric acid gel for 30 seconds, rinsed with water and gently air-dried. One drop of each Photo Bond adhesive resin base and catalyst were mixed, applied to the etched surface, gently air-blow to remove the alcohol solvent, and light-cured for 10 seconds.

**Clearfil Liner Bond group:** The surfaces were treated with a gel containing 10% citric acid with 20% calcium chloride for 40 seconds, rinsed with water and gently air-dried. SA Primer (3% N-methacryloyl-5-aminosalicylic acid in alcohol) was applied to the etched surface and gently air-dried until complete evaporation of the alcohol. A thin layer of Photo Bond adhesive resin was then applied to the surface, gently air blown, and light-cured for 10 seconds.
Clearfil Liner Bond II group: One drop each of Liner Bond II self-etching primers A and B were mixed, applied to the surface for 30 seconds and gently air-dried to remove excess of water and alcohol. A thin layer of LB Bond was painted on the surface and light-cured for 20 seconds.

Following the bonding procedures, acrylic tubes 4 mm diameter x 2 mm high were positioned over the bonded area. Fuji II LC or Vitremer were mixed following each manufacturer's instructions, respectively, transferred to a C-R syringe tip (Centrix™, Shelton, Connecticut, USA), and bulk-filled into the tubes. Excess material was pressed into the tubes by placing a glass slide over the tube, and then light cured from the top for 60 seconds. All specimens were then stored for 24 hours in tap water at 37°C prior to bond testing. Twelve specimens were prepared for each bonding system with Fuji II LC and Vitremer.

Before testing the specimens, the vinyl tape was carefully removed so that the shear blade could be positioned at the junction of the tooth and bonded interface. The samples were mounted on the jig for the shear bond test described in ISO standard TR110405 (ISO) and were stressed at a crosshead speed of 1 mm/minute in an universal testing machine (Autograph AG-500B, Shimadzu Co., Kyoto, Japan). The debonded surfaces were examined with a stereomicroscope at 20X magnification. The means and standard deviations were calculated from the bond test data. The results were analyzed by two-way analysis of variance (ANOVA) and Fisher's PLSD Test (p<0.05).

SEM Observation

Two erupted non-carious human third molars stored frozen were used for each experimental group and control, within two weeks following extraction. A pair of dentine disks, approximately 1-1.5 mm thick, was cut from each tooth using a low-speed diamond saw microtome (Leitz 1600, Wetzler, Germany) under running water. Disks with enamel remnants and exposure of pulp horns were discarded. One surface of each disk was ground with 600-grit silicon-carbide paper under running water to create a standardized smear layer. The dentine surfaces were bonded in the same manner as for the bond strength test. The specimens were stored in tap water for 24 hours at 37°C. The bonded assemblies were then sectioned perpendicularly to the flat dentin surface, into approximately equal halves disclosing 4 bonded surfaces for SEM observation, and were embedded in a self-curing epoxy resin (Epon 815, Thonide 245, Nisshin EM, Tokyo, Japan). The surfaces of the cut embedded specimens were polished to a high gloss with abrasive discs and diamond pastes successively, down to 0.25 μm particle size. The samples were gold-sputter-coated and observed under the SEM (JXA-840, JEOL, Tokyo, Japan).

After SEM observation, the coated surfaces of the specimens were again polished
and then etched with an argon-ion-beam for 270 seconds (E1S-1E, Elionix Ltd., Tokyo, Japan) to better determine the structures at the bonded interface. The samples were again gold-sputter-coated and observed under the SEM.

The specimens were re-polished a third time. After this, they were subjected to 10% phosphoric acid treatment for 3-5 seconds (Gwinnett and Kanca, 1992), followed by 5% sodium hypochlorite immersion for 5 min (Wang and Nakabayashi, 1991), so as to dissolve mineralized tissue and remove any collagen fibers which had not been enveloped by the adhesive resin (Gwinnett and Kanca, 1992). After thoroughly rinsing in water, the treated specimens were air-dried, gold-sputter-coated, and observed with the SEM.

Results

The mean shear bond strengths and standard deviations for the four groups are presented in Table 5.2. No significant interaction was found between the adhesive systems and the RmGIC when the data was analyzed by two-way Anova.

The shear bond strength of Fuji II LC was significantly higher than that of Vitremer ($p<0.001$) in the control group. When the adhesive systems were applied, a significant increase of the shear bond strengths of Vitremer to dentine occurred, and no significant difference between the two RmGICs could be detected. Although bond strengths increased significantly when Clearfil Liner Bond II was used with Vitremer, this difference was less so for Fuji II LC. Bond strengths for Vitremer seemed to be more affected by the adhesive systems than those for Fuji II LC. The bond strength of Vitremer was low (7.4 MPa) when bonded directly to the dentine, but significantly increased when used in conjunction with the bonding systems (2 of which included dentine etch and one-self-etching) (Photo Bond = 12.0 MPa, $p<0.005$; Liner Bond = 12.6 MPa, $p<0.01$; and Liner Bond II = 17.3 MPa, $p<0.001$). On the other hand, the bond strength of Fuji II LC with Photo Bond (14.5 MPa) or Liner Bond (15.0 MPa) did not differ significantly from the control (13.4 MPa, $p>0.05$). The bond strengths of Vitremer or Fuji II LC applied in conjunction with Liner Bond II were identical (17.3 MPa, $p>0.05$). The failure modes for all materials were classified as mixed with cohesive material failure and adhesive failure between the bonding resin and dentine; however, three LB II specimens showed cohesive failure in dentine.

The RmGIC/dentine control interfaces are shown in Figs. 5.1 and 5.2. SEM photographs of the RmGIC/adhesive/dentine interfaces are shown in Figures 5.3 – 5.8. Even after desiccation and exposure to the high vacuum for SEM observation, no separation between the RmGIC/adhesive interface, and adhesive/dentine interface was observed. The micromorphology of the interfaces exhibited interaction between the adhesive systems and the underlying dentin, demonstrating the formation of hybrid layers. Photo Bond strongly demineralized the underlying dentine, producing a 4 - 5 μm thick
hybrid layer. For Photo Bond specimens only, the layer of bonding resin was not observed between the RmGICs and underlying hybrid layer. Ca-agent created less demineralization of the surface, with a hybrid layer approximately 2 – 3 µm thick being observed. Compared with the other adhesive systems, Liner Bond II produced the thinnest hybrid layer, about 1 – 2 µm thick. However, the bond strength for this system was the greatest and identical for both Vitremer and Fuji II LC. All adhesive systems demonstrated a continuous link between the bonding resins and RmGICs yielding a continuous interface.

Figures 5.7 and 5.8 show Fuji II LC/Liner Bond II/ dentine and Vitremer/Liner Bond II/ dentine interfaces, respectively. Long resin tags penetrating dentinal tubules and a thin hybrid layer were clearly observed. Little effect of the RmGIC on dentin bonding over the adhesive bonding resins was recognized, and particles of the RmGIC were not observed within the resin tags.

Discussion

Because of the large number of teeth required for these types of study, and a lack of human teeth, bovine teeth were selected as a substitute for human teeth in the bond strength testing. The size of bovine teeth may eliminate some factors that can influence bond strength testing, as well as simplify the experimental procedure (Suzuki and Finger, 1988; Tagami et al., 1993). Furthermore, similarities concerning bond strength tests have been reported when comparing human and bovine teeth (Nakamichi et al., 1983; Fowler et al., 1992). Therefore, bovine incisors are believed to be suitable substrates for evaluating bonding systems. On the other hand, because it is also important to evaluate and predict how dental restorative systems affect human dentine, human teeth were used for the SEM observation part of the study.

To date, various attempts have been made to improve mechanical and adhesive properties of RmGICs to enamel and dentine. In order to improve bond strengths of RmGICs to dentine, various acidic conditioners and primers have been used to pre-treat the dentine (Powis et al., 1982; Berry and Powers, 1994). Pre-treatment of the dentine with acidic conditioners removes the smear layer and demineralizes the superficial dentine layer, allowing the HEMA (2-hydroxy ethyl methacrylate) incorporated in the RmGICs to penetrate the exposed collagen fiber network (Titley et al., 1996; Friedl et al., 1995).

The shear bond strengths for Vitremer obtained in this study are comparable to previously reported results ranging between 5.9 to 9.7 MPa (Triana et al., 1994; Bell and Barkmeier, 1994; Berry et al., 1994; Erickson and Glasspoole 1994; Pawlas et al., 1994). The bond strength of Vitremer was low (7.4 MPa) when bonded directly to the dentine, but increased significantly when used in conjunction with the bonding systems. The results for Fuji II LC were similar to results previously published, which ranged from 11.0 to 15.4
MPa (Triana et al., 1994; Friedl et al., 1995; Berry et al., 1994; Charlton and Havemann 1994; Kato et al., 1995; Fritz and Finger, 1996; Garcia-Godoy, 1992). Interestingly, when the adhesive system Liner Bond II was used, bond strengths for both Vitremer (17.3 MPa) and Fuji II LC (17.3 MPa) increased, being significantly greater than the other groups tested. In previous work, Liner Bond II has been demonstrated to produce greater bond strengths to bovine dentine compared with the other two bonding systems (Burrow et al., 1994) when using a resin composite. This same situation also occurred in this study, therefore, the results in this study demonstrate that RmGICs can bond very well to bonding resins, producing similar results to bonding of resin composites.

The SEM observations disclosed distinct interactions between the bonding resins and the underlying dentine. As expected, the different conditioners and adhesive resins produced different degrees of demineralization and thickness of hybrid layers. It was also noted that the RmGICs exerted little effect on the bonding capacity of the different adhesive systems, and consequently in bond strengths. Since the RmGICs contain HEMA and other resinous components, they were able to interact with the bonding resin forming a chemical union (Figs. 5.3 - 5.8). Interestingly, the Photo Bond group specimens did not display a layer of bonding resin (Figs. 5.3 and 5.4). This may be probably a result of an incompletely polymerized bonding resin and a very thick hybrid layer that may have acted as a sponge absorbing the uncured resin.

Resin-modified glass-ionomer materials have been widely used and are recommended for restoring cervical lesions that include enamel and dentine margins, and for the caries-prone patient because of the possible cariostatic effect of fluoride. Adhesion of recently developed RmGICs to dentine has been improved; however, comparable bond strengths to dentine bonding adhesives have not yet been achieved. Therefore, we improved bond strengths by applying adhesive resins prior to placement of a RmGIC. Our results demonstrated that the shear bond strengths of Vitremer was significantly less when applied directly to the dentine. Since Fuji II LC requires conditioning with 10% polyacrylic acid, and Vitremer requires a primer, the difference in dentine conditioners and material compositions may be the reason for the different bond strength values (Mitra and Kedrowski, 1994; Triana et al., 1994). Current studies are being carried out to analyze what factor was responsible for the increased bond strength.

It has also been previously reported that fluoride ions could diffuse through adhesive resins when RmGICs were coated, although in smaller quantities (Castro et al., 1994). Thus, should it be that fluoride ions could diffuse through adhesive resins, the benefit of using RmGICs over dentine bonding materials may not be lost. Further studies are necessary in order to determine this possibility.
Conclusions

Shear bond strength of Vitremer to dentine was significantly increased when recent adhesive resin systems were used. Shear bond strength of Fuji II LC to dentine significantly increased when the self-etching primer system was used. A technique of bonding dentine with a resin and then applying RmGIC may be a good solution for those materials that produce weak bonding to dentine.

Table 5.1. Restorative materials employed

<table>
<thead>
<tr>
<th>Material</th>
<th>Brand name</th>
<th>Components</th>
<th>Batch</th>
<th>Manufacturer</th>
</tr>
</thead>
<tbody>
<tr>
<td>Resin-modified glass-ionomer</td>
<td>Fuji Ionomer Type II LC</td>
<td>Conditioner:</td>
<td>071141</td>
<td>GC Corp., Tokyo, Japan</td>
</tr>
<tr>
<td>cement</td>
<td></td>
<td>Powder:</td>
<td>071241</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Liquid:</td>
<td>291141</td>
<td></td>
</tr>
<tr>
<td>Resin-modified glass-ionomer</td>
<td>Vitremer Primer</td>
<td>Primer:</td>
<td>3303P</td>
<td>3M Dental Products, MN, USA</td>
</tr>
<tr>
<td>cement</td>
<td></td>
<td>Powder:</td>
<td>3303A3</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Liquid:</td>
<td>3303L</td>
<td></td>
</tr>
<tr>
<td>Adhesive Resins</td>
<td>Clearfil Photo Bond</td>
<td>K-etchant:</td>
<td>K-145</td>
<td>Kuraray Co., Osaka, Japan</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Photo Bond:</td>
<td>Catalyst: 236 Universal: 339</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Clearfil Liner Bond</td>
<td>CA-agent:</td>
<td>1134</td>
<td>Kuraray Co., Osaka, Japan</td>
</tr>
<tr>
<td></td>
<td></td>
<td>SA Primer:</td>
<td>021</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Photo Bond:</td>
<td>Catalyst: 191 Universal: 296</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Clearfil Liner Bond II</td>
<td>Primer:</td>
<td>001</td>
<td>Kuraray Co., Osaka, Japan</td>
</tr>
<tr>
<td></td>
<td></td>
<td>LB Bond:</td>
<td>0002</td>
<td></td>
</tr>
</tbody>
</table>

K-etchant: 37% H₃PO₄; CA-agent: 10% citric acid in 20% calcium chloride; SA Primer: 3% n-methacryloyl 3 aminosalicylic acid in ethanol

Table 5.2. Shear bond strength of Fuji II LC and Vitremer using different adhesive systems and statistical analysis

<table>
<thead>
<tr>
<th>Material</th>
<th>Control</th>
<th>Photo Bond</th>
<th>Liner Bond</th>
<th>Liner Bond II</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fuji II LC</td>
<td>13.4 (1.6)</td>
<td>14.5 (2.6)</td>
<td>15.0 (2.9)</td>
<td>17.3 (6.3)</td>
</tr>
<tr>
<td>Vitremer</td>
<td>7.4 (2.6)</td>
<td>12.0 (2.0)</td>
<td>12.6 (1.8)</td>
<td>17.3 (4.9)</td>
</tr>
</tbody>
</table>

Bars indicate no statistically difference among figures by the Fisher’s PLSD Test (p>0.05).
Figure 5.1. Polished Fuji II LC (F) / dentine (D) interface after surface treatment with 10% polyacrylic acid. Note a resin-rich layer (R) that was slightly worn off during polishing procedures.

Figure 5.2. Vitremer (V)/dentine (D) interface after surface treatment with Vitremer primer. A resin rich layer can not be clearly observed.
Figure 5.3.  Fuji II LC/Clearfil Photo Bond/dentine (D) interface after argon ion beam etching. The specimen displays a 4 – 5 µm thick hybrid layer (H). The bonding resin is imperceptible between the RmGIC (F) and hybrid layer. Note the good interaction between the RmGIC and hybrid layer, and formation of resin tags (T) although bonding resin is imperceptible.

Figure 5.4.  Vitremer (V)/Clearfil Photo Bond/dentine (D) interface after argon ion beam etching. This specimen displays a hybrid layer (H) of approximately 8 µm thick, and the layer of bonding resin is imperceptible. Resin tags are apparent (T).
Figure 5.5. Fuji II LC/Clearfil Liner Bond/dentine (D) interface after argon ion beam etching. A 2 – 3 µm thick hybrid layer, resin tags (T), and a distinct border between the bonding resin (B) and the RmGIC (F) are observed.

Figure 5.6. Vitremer (V)/Clearfil Liner Bond/dentine (D) interface after argon ion beam etching. Note a 2 – 3 µm thick hybrid layer (H) and a non-distinct border between the bonding resin (B) and the RmGIC.
Figure 5.7. Fuji II LC/Clearfil Liner Bond II interface after exposure to 10% phosphoric acid for 3 – 5 seconds followed by immersion in 5% sodium hypochlorite for 5 minutes. A hybrid layer of 1 – 2 μm thick (black arrowheads), and long resin tags can be observed. A good interaction between the bonding resin (B) and the RmGIC (F) can be seen.

Figure 5.8. Vitremer – Clearfil Liner Bond II/dentine interface after exposure to 10% phosphoric acid for 3 – 5 seconds followed by immersion in 5% sodium hypochlorite for 5 minutes. A hybrid layer of 1 – 2 μm thick (black arrowheads), and long resin tags can be observed. Note a very good interaction between the bonding resin (B) and the RmGIC (V).
Chapter 6

General conclusions and future perspectives

Secondary caries and dentin bonding has become two of the most challenging and studied topics in the restorative dentistry. As patients live longer the probability of cervical lesions and/or secondary caries around cervical restorations to occur increases, lead by periodontal problems, decrease in coordination movements and consequent plaque accumulation. However, root surface caries present specific operative difficulties, which are reduced retention capacity and difficult access of the restoration. Adhesive restorative materials, especially the resin-modified glass ionomer cements (RmGICs) have been indicated as first choice restoratives for root surface caries because of their potential anticariogenic effect. Fluoride has been also incorporated to recent adhesive restoratives and bonding systems as to protect the restoration against secondary caries. These recent bonding systems produce high bond strengths however, fail to inhibit secondary caries. In order to increase the adhesive capacity of the resin-modified glass ionomer cements, preserving the potential anticariogenic effect of these materials, improved conditioners and use of adhesive resins should be considered.

Chapter 2

The aims of this investigation were to determine through polarized light microscopy the capacity of three glass ionomeric restoratives and a fluoride releasing resin composite system to inhibit in vitro secondary caries in root dentin, as well as to measure the width and height of the inhibition zones, and the depth of the outer lesions. The results indicated that although the three glass ionomeric materials used in this study produced an inhibition zone adjacent to the restorations, the height and width of this zone was material dependent. The conventional glass ionomer cement (Fuji Ionomer Type II; GIC) produced an inhibition zone with greatest height and thickness compared to the resin-modified glass ionomers (Fuji II LC and Vitremer; RmGIC). The outer lesions that were produced were similar in depth for all materials, indicating that they were little influenced by the restorative materials. Since previous studies have suggested that fluoride released by GIC and RmGIC are similar or greater for the latter, it can be speculated that other factors, e.g. other ions that are released together with fluoride, may be responsible for a in vitro caries inhibition. The resin composite containing fluoride failed to produce an inhibition zone, even though visual separation of the restoration at the cavity margin was not evident. Presumably, the demineralizing solution may have leaked through the base of the hybrid layer creating a wall lesion. Therefore, even recent adhesive resin composite systems that produce high bond strengths to dentin do not protect the restoration against secondary caries when the environment is prone to caries formation.
Chapter 3

In chapter 2, the presence of inhibition zones was determined and the dimensions of these zones measured. However, polarized microscopy by itself does not reveal the quality of the inhibition zone, for example, the degree of mineralization. Therefore, the purpose of the investigation of this chapter was to measure the microhardness of the inhibition zones and outer lesions produced adjacent to the glass ionomeric restorations when subjected to *in vitro* acid challenge. Knoop hardness number (KHN) of all outer lesions were initially measured along the lesion and parallel to the cavity wall. However, because the size of the knoop indenter is greater than the width of the inhibition zone, an alternative microhardness tester that measures the triangular hardness (HT) was used to determine the hardness of the inhibition zone. The HT of the outer lesions and inhibition zones were determined and correlated with KHN values, and a linear correlation obtained. Therefore, the triangular hardness test showed to be efficient to measure the hardness of narrow areas. When comparing hardness of the different inhibition zones, the zone created adjacent to the GIC was significantly greater than those created by the RmGICs. These results confirmed the previous polarized microscopy data (chapter 2), and demonstrated that the mineral content of the inhibition zone around the GIC was greatest. Similar to chapter 2, it can be speculated that other factors, rather than fluoride alone, may influence the formation of mineralized inhibition zones.

Chapter 4

This study focused in testing the effect of an improved resin-modified glass ionomer cement (Fuji II LC-I) on shear bond strengths to enamel and dentin that were conditioned with an agent composed of 20% polyacrylic acid and 3%A1Cl₃ (cavity conditioner), and analyzing the micromorphology appearance of the cement/tooth interface. Since bonding of glass ionomeric restoratives can be inconsistent (Helwett et al., 1991; Ngo et al., 1986) without removal of the smear layer, and clinical retention and bond strengths to dental substance are improved when the smear layer is removed (Powis et al., 1982), pre-treating the cavity walls with 10% polyacrylic acid solution (dentin conditioner) became routine prior to restoring with ionomeric materials. The Fuji II LC-I posses superior polishability to the former version, and the cavity conditioner requires a 10 second application rather than a 20 second application for the dentin conditioner.

The results indicated that bond strengths of both versions to enamel were similar at 5 minutes after light-curing, and that they significantly increased at 1 day remaining stable to 1 week, being the improved version significantly higher than the former version. Regarding bond strengths to dentin, the improved version showed significant higher bond strengths than the former version at 5 minutes after light-curing, remaining stable to a period of 1 week. The increase of early bond strengths after light-curing of Fuji II LC-
I/cavity conditioner system is possibly the greatest clinically-related advantage over the former system, because it permits the clinical operator to finish and polish the restoration safely after light-curing.

Chapter 5
Bonds of the restorative materials to dentin or enamel may fail losing its retention to the cavity, promoting leakage of fluids from the oral environment into the cavity, being detrimental to the longevity of the restoration. Since bond strengths of resin-modified glass ionomer cements are low and volumetric shrinkage during light-curing is similar to the resin composite systems, the study in chapter 5 aimed to increase shear bond strengths of RmGICs to dentin by first bonding the dentin with an adhesive resin. Three adhesive resin systems (Clearfil Photo Bond, PB; Clearfil Liner Bond, LB; and Clearfil Liner Bond II, LBII) and two resin-modified glass ionomer cements (Fuji II LC and Vitremer,) were used in the present investigation. The results indicated that shear bond strengths of both Fuji II LC and Vitremer were quite low when applied directly to the dentin surface, however significantly increased when a recent self etching primer system (LBII) was used. Among the three systems used in this study, bond strengths of resin composites have been reported to be highest for the LBII (Burrow et al., 1994). These previous results demonstrate that RmGICs can bond very well to bonding resins, producing relatively high bond strengths to dentin. The micromorphology appearances disclosed a good interaction between the bonding systems and the RmGICs, as well as good interaction between the bonding systems and the underlying dentin. Moreover, because fluoride ions have been reported to diffuse through adhesive resins (Castro et al., 1994), the anticariogenic benefit of the ionomeric materials may not be lost. Therefore, the technique of bonding dentin with an adhesive resin and then applying a RmGIC may be a good restorative solution for those materials that produce weak bonding to dentin.

Unfortunately, to date no restorative material posses all the properties desired in a perfect filling material (Watts, 1996). Successful and appropriate application of glass ionomeric restoratives will depend upon conscientious indication and careful handling skills, as much as the inherent restorative formulation. The ideal restorative material should be able to provide short- and long-term sealing of the cavity by bonding to the tooth tissue, undergo minimal contraction during light-curing, and possess anticariogenic properties by releasing and uptaking fluoride into and from the adjacent restoration or teeth, and oral environment.
References

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