The influence of blood and hemostatic agent contamination on bond strength of orthodontic attachments cemented with resin modified glass ionomer cement.

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The Influence of Blood and Hemostatic Agent Contamination on Bond Strength of Orthodontic Attachments Cemented with Resin Modified Glass Ionomer Cement

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Abstract

Effective use of traditional composite resins to attach orthodontic appliances to an impacted tooth requires isolation of the enamel surface during bonding, which can be difficult due to limited access and the presence of contaminates. Resin-modified glass-ionomer cements (RMGIC) have been shown to have the ability to bond to contaminated enamel surfaces. The purpose of this study was to evaluate the effect of two common contaminates experienced in this situation (blood and Hemodent) on the mean shear bond strength of an orthodontic button applied with light-cured RMGIC to an intact enamel surface. The shear-peel forces required to create bond failure were compared to the forces reported during orthodontic treatment to determine suitability for clinical application when bonded under the tested conditions. 125 enamel surfaces from intact human third molars were divided into five Groups of 25 samples. Groups of teeth were exposed to one of five treatments prior to the bonding of a flat button with light-cured RMGIC (Fuji Ortho LC): Group 1-distilled water, Group 2-blood, Group 3-blood then wiped with water, Group 4-blood then wiped with Hemodent, Group 5-Hemodent then wiped with water. Shear-peel debond force was measured with a materials testing instrument. Results were analyzed using a one-way ANOVA and Pearsons and Tukeys tests with p set to ≤ 0.05. Results showed no significant differences between Groups 1, 3 & 5 or Groups 2 & 4. Significant differences were noted between combined Groups 1, 3, & 5 compared to 2 & 4. The tests showed high variability in bond strengths within each Group. Mean debond forces were above the minimum forces required for orthodontic treatment (6-8 MPa) in Groups 1, 3 & 5. The findings suggest when an intact enamel surface is contaminated with either blood or Hemodent, bond strength similar to that of moist enamel can be obtained by wiping the surface clean with water prior to bonding with RMGIC.

Introduction

The need to re-bond orthodontic appliances when the bond to enamel has failed results in extended treatment time, decreased confidence in the care provider, increased cost of the care delivered and potentially additional surgical procedures to re-expose impacted teeth. When bonding an orthodontic button or bracket to an enamel surface in a contaminated environment, such as with a surgically exposed impacted tooth, it is difficult to isolate the enamel from local contaminates such as blood and saliva, and hemostatic agents used during the procedure. Light-cured composite bonding agents are most commonly used to adhere appliances to teeth in order to apply orthodontic forces. When bonding with a composite resin it is important to maintain a contamination free field once the enamel has been etched. This will ensure that the maximum bond strength is obtained between the enamel, the orthodontic bracket and the bonding material (Itoh et al, 1999; Sfondrini et al, 2004; Silverstone et al, 1985). This may be difficult for the operator to accomplish with surgical exposures due to the humidity of the oral cavity, continuous saliva or blood seepage into a surgical area, and the presence of chemical agents used to achieve hemostasis (Cobo & Moro 1994).

The ability to easily achieve a clinically acceptable bond to a contaminated surface would make the procedure of bonding an orthodontic bracket more efficient for the orthodontist, particularly when bonding to a surgically exposed tooth. In addition, tolerance to contamination may simplify many other clinical situations requiring bonding in an environment where isolation of the field is difficult.

A material ideally suited for moist or other such contaminated situations may be the resin modified glass ionomer cement (RMGIC). Fuji Ortho LC (FOLC) is a dual cured (light & self cured) resin-reinforced glass ionomer, based on the widely accepted technology of the hybrid glass ionomer restorative materials. It is composed of a fluoroaluminosilicate glass powder, hydroxyethyl-methacrylate (HEMA) and maleic/acrylic acid copolymer liquids. FOLC is commonly used in orthodontic practices and is commercially available. Similar to self-cure glass-ionomer cements RMGIC is moisture tolerant, releases fluoride over an extended period, bonds without surface etching and the adhesive is easy to remove from the enamel once treatment is completed (Saito et al, 1999; Staley et al 2004). RMGIC also possesses many of the desirable attributes of the light-cured composites such as rapid curing once light activated, a high initial strength and hardness, and clinically proven bond strength (Saito et al, 1999). Thus the bond strength obtained with resin-modified glass-ionomer cement (RMGIC) in a contaminated field may be sufficient to withstand force levels commonly applied during orthodontic treatment.

This experiment was undertaken with the intent of recreating the conditions encountered by a surgeon when exposing a tooth to bond an orthodontic button or bracket. During this type of procedure, it is often very difficult to isolate enamel from blood or saliva contamination in order to bond an orthodontic attachment. The addition of chemical agents such as anesthetics, vasoconstrictors and hemostatic agents add to the potential list of contaminates to the bonding site. The affect of many of theses contaminates on the bond strength attainable with RMGIC has not been studied. Knowing the effect of the chemicals used during surgery may make it more predictable

for the surgeon to attain a clinically acceptable bond, as well as aid other practitioners in bonding under similar situations such as when placing a sub-gingival restoration or cementing a crown in a contaminated environment.

Many different contaminate combinations could potentially be studied to determine their effects on bonding attained with a RMGIC. The addition of Hemodent as a hemostatic agent was chosen as a contaminate because of its use in oral surgery and general dentistry for hemostasis when preparing to bond in a blood contaminated field. The low pH of Hemodent (pH = 1.3) makes it a potential etching solution of the enamel surface (Land et al, 1994). Upon exposure to dentin, Hemodent has shown a mild etching effect in previous studies (Land et al, 1994). Manufacturers of some RMGIC recommend using a mild acid (polyacrylic acid) to remove contaminates and aid in surface wetting prior to bonding. The addition of the acidic Hemodent solution may help to remove contaminates and increase surface wetting of the enamel for the glass ionomer cement, in addition to creating micro-mechanical retentive areas and increased surface area (Glasspoole et al, 2002). It is unknown whether the presence of the aluminum chloride in Hemodent solution will aid, hinder or have no affect on the RMGIC's bond formation.

Background

Development of the acid-etch technique led to the direct bonding of orthodontic brackets to enamel with composite resins (Bishara et al, 1998). This development resulted in improvements in orthodontic treatment including greater patient comfort, elimination of pre-treatment tooth separation and lengthy banding appointments, decreased gingival irritation, easier oral hygiene procedures, improved esthetics, elimination of post-treatment band spaces, and reduced overall chair-side time (Bishara et al, 1998). Clinical improvements related to orthodontic bonding are still needed in three major areas: reduction of white spot lesions, increased tolerance of contamination during bonding to reduce the incidence of bond failures and preservation of enamel during tooth preparation and appliance removal (Gorelick et al, 1982).

Composite Resins

Composite resins now commonly used in the bonding of orthodontic appliances were introduced in the 1960s. The material is composed of silica or quartz filler particles, an organic matrix composed of bisphenol a-glycidyl methacrylate (Bis-GMA) or urethanedimethacrylate (UDMA) and triethylene glycol dimethacrylate (TEGDMA) which have reactive double bonds at each end of the molecules, silane coupling agents, an accelerator such as an amine, and an initiator. The self-cured initiator is often a organic peroxide which reacts with the tertiary amine, producing free-radicals that induce polymerization of the methacrylates. The light-cured systems rely on activation of initiators such as camphoquinone by visible light of an appropriate wavelength producing free radicals that induce polymerization of the methacrylates (O'Brien 1997).

Contamination and Composites

In preparing a tooth surface for bonding with composite resins, the area to be bonded must be isolated. The enamel surface must be etched for 15-20 seconds with a 37% phosphoric acid solution and rinsed with water for 15 seconds before drying with a burst of oil-free air. A thin layer of unfilled resin is placed over the etched surface (this is light cured if using light activated composite). The composite is then placed over the area and cured by either light initiation or chemical initiation (Cacciafesta et al, 2004; O'Brien 1997; Sfondrini et al, 2004; Silverstone et al, 1985). If the surface is contaminated at any step along the way with saliva or blood, re-etching of the surface is advised to remove any organic adhesive coating. Even momentary saliva or blood contamination will plug porosities on the wet enamel and adversely affect the bond and rinsing alone will not remove this coating (Cacciafesta et al, 2004; O'Brien 1997; Sfondrini et al, 2004; Silverstone et al, 1985). Self-etching primers and moisture insensitive primers have been introduced, but these also have contamination sensitive steps during bonding (Cacciafesta et al, 2004; O'Brien 1997; Sfondrini et al, 2004; Silverstone et al, 1985).

Studies have shown that when etched enamel was exposed to salivary contamination for a time period of more than one second, an adherent coating was present that masked the underlying etched enamel and was resistant to washing (Silverstone et al, 1985). If this occurred clinically, Silverstone and associates (1985) recommended re-etching of the enamel surface in order to obtain a satisfactory bond. When bovine enamel that had been treated with either an etchant and a traditional primer (Transbond XT) or a self etching primer (Transbond Plus SEP) was contaminated with

human blood prior to bonding with composite (Transbond), the resulting shear-peel bond force was decreased below clinically acceptable levels (Cacciafesta et al, 2004; Sfondrini et al, 2004).

Composite resins have high tensile and compressive bond strengths and have proven to be effective at minimizing orthodontic bond failures when applied appropriately. Despite the binding strength, there are several side effects of composite resins that could be improved upon. The use of phosphoric acid causes decalcification of enamel. The high bond strengths of the composites although useful during treatment have been associated with fractures and crack formation during debonding procedures, especially when removing ceramic appliances. The difficulty of removing the composite from the enamel at the time of debonding has necessitated grinding away the composite with rotary instruments. This leads to enamel damage even with careful removal. In addition, the challenge of cleaning around the appliances for the patient still allows for significant white spot formation on the enamel (Bishara et al, 1998; Gorelick et al, 1982).

Glass-Ionomer Cements

Glass-ionomer cements, invented by Wilson and Kent in the 1970's, are capable of bonding physio-chemically to non-conditioned tooth structure and metals in the presence of minor water and saliva contamination. The cement uses an acid-base reaction between a basic powder composed of a calcium aluminum fluorosilicate glass, and liquid composed of polycarboxylic acid in the presence of water. The acid component of the glass-ionomer conditions the substrate, while the ionic reaction of the carboxyl Group in the acid enables the cement to adhere to metals and tooth structure (Wilson & Kent 1972; Saito et al, 1999).

According to Saito and colleagues (1999), glass-ionomer cement bonds to enamel by the following process. When the two components are mixed, the hydrogen ions of the acid partially dissolve the glass particles in the presence of water, releasing calcium, strontium and aluminum ions. The ions combine with the polycarboxylic acid groups to form a poly-acid salt matrix, which surrounds the glass particles, changing the glass to a silica hydrogel. The acidity of the aqueous cement acts as a mild etchant on the tooth, removing the smear layer. The initial attraction between the enamel and the glass-ionomer is due to a polar attraction of hydrogen bonds. The phosphate ions from the etched hydroxyapatite crystals buffer the weak acid, aided by surrounding water. The bond is further increased by ionic movement of phosphates and calcium ions at the adhesive-enamel interface, creating salt bridges between the adhesive and enamel. Maximum achievable bond strength is only reached after the cement has fully matured, approximately 24 hours (Saito et al, 1999).

The fluoroaluminosilicate glass is able to release excess fluoride from the cement matrix into the surrounding environment over an extended period of time after the material is fully set. The fluoride levels of the material will recharge when the surrounding environment's fluoride levels rise above that of the matrix, as when brushing with a fluoridated tooth paste or using a fluoride mouth rinse. This recharging ability allows the material to act as a fluoride reservoir over the life of the restoration (Saito et al, 1999; Staley et al, 2004).

The ability of the self-cured glass-ionomer cements to bond with contaminated enamel and metal surfaces, and to prevent decalcification of enamel through prolonged fluoride release has led to their implementation in the cementing of orthodontic bands.

Unfortunately the prolonged setting reaction time and late gain of strength makes it difficult to utilize this cement when bonding orthodontic brackets due to bracket drift after placement, sensitivity to moisture contamination or desiccation and an extended wait time until arch-wires may be placed (Compton et al, 1992; Saito et al, 1999).

Resin Modified Glass-Ionomer Cements

In order to produce favorable physical characteristics similar to those of resin composites while retaining the desired features of the glass-ionomer cements, water-soluble resin polymers and polymerizable monomers have been incorporated into the aqueous solution of polyacrylic acid. This enables the material to undergo polymerization while simultaneously utilizing the acid base reaction of the self-cured glass-ionomer (Saito et al, 1999; O'Brien 1997).

In resin modified glass-ionomer cements, the cement powder is composed of a fluoroaluminosilicate glass, similar to the conventional glass-ionomer cements. The liquid is typically composed of a cross-linking material, 2-hydroxyethylmethacrylate, carboxylic acid and water (HEMA). The acid-base setting reaction is initiated upon mixing the powder and liquid, as with conventional glass-ionomers. A photo-polymerizing or oxidation catalyst is used to initiate polymerization of the HEMA and cross-linking material. The catalyst initiates hydrogen bonding between the HEMA polymer and polycarboxylic acid, which rapidly hardens the mixture (Saito et al, 1999).

The addition of the resin monomers decreases setting time and increases the initial strength and hardness of the cement. The rapid setting time also minimizes the cements sensitivity to changes in moisture during maturation. These changes act to ensure more optimal properties as an orthodontic bonding agent (Saito et al, 1999).

Fluoride Release of RMGIC

A significant benefit of the glass-ionomer and resin-modified glass-ionomer cements over traditional composites is that they have been found to release fluoride for a period of three weeks, which is known to reduce the incidence of caries to the adjacent enamel surface (Gao et al, 2000; Staley et al, 2004). Furthermore, RMGIC have been shown to absorb fluoride when it is introduced to the oral cavity, such as while brushing with a fluoridated toothpaste or rinsing with a fluoridated mouthwash, and slowly release it over a three week period (Gao et al, 2000; Staley et al, 2004). Decalcification of the enamel surface around orthodontic appliances is a significant problem during treatment. The ability to continuously release fluoride to the enamel adjacent to the glass-ionomer could be very beneficial if it helps minimize this decalcification and white spot lesions around orthodontic appliances (Gorelick et al, 1982; Staley et al, 2004).

Self-Cured vs. Light-Cured RMGIC

Light cured RMGIC have shown significantly higher shear bond strengths than self-cured RMGIC (Compton et al, 1992, Glasspoole et al, 02). The RMGIC can be light and self cured, and like self cured glass-ionomer cements, it does not require etching of the enamel surface before bonding, it bonds chemically to the enamel, it utilizes ambient moisture during its setting reaction so it is not affected as severely by water and saliva contamination relative to traditional light cured resins (Caciafesta et al, 1998; Chung et al, 1999; Itoh et al, 1999; Itoh et al, 00; Jobalia et al, 1997; Kusy 1994; Lippitz et al, 1998; Silverman et al, 1995; White 1986). Notably, several authors have reported Fuji Ortho LC obtained higher bond strengths if the enamel surface was contaminated with small amounts of water or saliva before application of the RMGIC than if the surface was

dry prior to bonding (Cacciafesta et al, 1998; Chung et al, 1999, Itoh et al, 1999, Itoh et al, 2000; Jobalia et al, 1997; Kirovski & Madzarova 2000).

There is a significant difference in bond strengths between self cured and light cured glass-ionomer cement with one-hour bond strengths of 8.8 MPa vs. 16.7 MPa ($p=0.001$) respectively, when bonded to human premolar enamel conditioned with a weak nitric acid (Compton et al 1992). The 24-hour bond strengths were 16.7 MPa vs. 17.2 MPa respectively. While the self cured cement showed a large increase in bond strength after 24 hours, there was no significant difference between the one-hour and 24 hour bond strengths of the light-cured glass-ionomer cement (Compton et al, 1992). When bonding to conditioned bovine enamel, Glasspoole, Erickson and Davidson reported in 2002, 20% lower bond strengths after 24-hours with a self-cured glass-ionomer (Fuji II) compared to a light-cured glass-ionomer (Fuji LC).

Long Term Success of RMGIC

Glass-ionomer cements have yet to enter mainstream use as the primary agent used in bonding orthodontic brackets. Practitioners are concerned about the lower bond strength obtained when using glass-ionomer cement compared to that of composite. Use of the resin-modified glass ionomer cements (RMGIC) to bond brackets to enamel has become more common in orthodontics and has shown good long-term success compared to traditional composite resins (Fricker 1994, Silverman et al, 1995, White 1986).

Jobalia reported that when bonded to the wet-etched enamel of human molars, Fuji Ortho LC had mean tensile bond strength of 11.4 MPa, comparable to that of a properly applied composite resin (Jobalia et al, 1997). Rix and associates (2001) reported mean shear-peel bond strengths with brackets of 13.57 MPa with wet etched human

premolar enamel using Fuji Ortho LC compared to 20.19 MPa with Transbond XT, a composite resin on dry etched enamel. Both of these values are well above the MPa recommended for orthodontic treatment by Reynolds in 1975.

In a clinical study, Fricker (1996) bonded orthodontic brackets in ten cases using composite resin for one half of each arch and RMGIC for the other. After 12 months of treatment there were two bond failures (3.3%) with the RMGIC and one with the composite (1.6%). The report indicated there was no significant difference in bond failure rates ($p<0.10$). The author pointed out the improvement of RMGIC over self-cured GI when he compared this study to an article he published in 1992, in which he used similar methods and reported a 20% bond failure rate over 12 months when using self-cured glass ionomer cement (Fricker 1994).

In other clinical studies using Fuji Ortho LC (a commonly used orthodontic RMGIC), Silverman and associates (1995) bonded brackets in both arches, including first molars. They reported a 96.8% success rate during eight months of orthodontic treatment when bonding to a tooth surface cleansed with plain pumice and rinsed with water prior to bonding (Silverman et al, 1995). Summers and associates reported a 6.5% bond failure rate with Fuji Ortho LC when used as the adhesive on half the upper and lower arches to treat 22 orthodontic patients. This was compared to a failure rate of 5% of the brackets bonded with a light cured composite, in the same patients (Summers et al, 2004).

Enamel Conditioning

The manufactures recommendations for Fuji Ortho LC suggest the addition of conditioning the enamel surface with 10%-40% polyacrylic acid will increase the bond strength obtained with the RMGIC over the standard non-etched technique (GC Fuji

Ortho LC Instruction Sheet, GC America Inc, Alsip, IL, USA). Use of the polyacrylic acid may increase bond strength by cleaning and wetting the enamel surface and pre-activation of the calcium and phosphate ions on the tooth surface rendering them more available for ion exchange with the cement. This is the same acid used in the cement and any residue left behind by the conditioner should not interfere with the setting reaction (Watson 1999).

As acidic materials, the conditioners may also produce micro-porosity in the enamel surface similar to that created by phosphoric acid, and this could contribute to either increase surface area for chemical bonding or micro-mechanical bonding through polymer penetration, increasing the bond strength (Glasspoole et al, 2002). Summers and associates (2004) showed increased surface roughness when conditioning with 10% polyacrylic acid when compared to non-treated enamel. However, the surface roughness was significantly less than the roughness produced by 37% phosphoric acid (Summers et al, 2004).

In contrast, the benefit of surface conditioning with polyacrylic acid is debatable. Some studies report that the increase in bond strength is statistically significant (Itoh et al, 1999; Kirovski & Madazarova 2000; Lippitz et al, 1998), whereas other researchers report no significant benefits (Cacciafesta, et al,-1998, Chung, et al,-1999, Cook and Youngson 1998). The use of phosphoric acid has also been studied and increased bond strengths have been reported when using it as enamel pretreatment (Cook & Youngson 1998, Glasspoole et al, 2002).

Cortes and associates (1993) reported a significant ($p<0.001$) increase in bond strengths when the enamel surface was etched with 10% phosphoric acid prior to bonding compared to non-etched enamel when using Fuji II LC. However, the mean shear-peel bond strength of the Fiji II LC with non-etched enamel was 11.29 MPa, a strength that is within the range needed for orthodontic treatment (Reynolds 1975). In addition, the latter study found that more non-etched samples failed at the enamel-adhesive interface compared to the etched samples which all failed cohesively (Reynolds 1975). This was attributed to higher enamel-adhesive bond strengths than cohesive bond strengths with the etched samples.

Contamination and RMGIC

To evaluate the potential use of RMGIC in a contaminated field, studies have been conducted to determine the mean bond strengths obtained with RMGIC when the bonding surface is contaminated with water, saliva and blood. These studies, conducted with and without enamel conditioning, reported a significant reduction in bond strength once contaminates were introduced, where others reported an increase in bond strength once the enamel surface was contaminated (mean shear bond strength range of 4-25 MPa; Bishara et al, 1998; Cacciafesta et al, 1998; Chung et al, 1999; Compton et al, 1992; Itoh et al, 1999; Itoh et al, 2000; Kirovski & Madazarova 2000; Lippitz et al, 1998; Reddy et al, 2003).

Saliva

In a 1998 research article evaluating the use of Fuji Ortho LC with and without saliva contamination, Bishara and researchers reported a significant increase in shear-peel bond strength from 3.5 MPa without saliva to 5.8 MPa with saliva, when bonding to non-

etched human molar enamel. Similar results were reported by Itoh and associates (1999) when comparing enamel moistened with water vs. saliva.

Cacciafesta and associates (1998) reported mean bond strength of 9.8 MPa when bonding an orthodontic button to wet unconditioned but polished bovine enamel with Fuji Ortho LC. When the enamel was conditioned with 10% polyacrylic acid for 10 seconds and then rinsed with water prior to bonding, the mean strength increased to 15.7 MPa. When saliva contamination was added as the final step before bonding, the mean debond force increased to 23.8 MPa. Considering only 6-8 MPa are required for treatment, the authors suggest that the conditioning step may be eliminated when bonding with Fuji Ortho LC (Cacciafesta et al, 1998, Reynolds 1975).

In study by Jobalia and collegues (1997) involving human molars and Fuji Ortho LC, non-etched enamel moistened with water produced mean bond strength of 8.7 MPa, whereas moistening the teeth with human saliva produced mean bond strength of 9.3 MPa (Jobalia et al, 1997). Thus several studies show that the presence of saliva contamination does not significantly reduce the bond strength and may significantly increase the bond strength.

Blood

Kirovski and Madazarova (2000) contaminated human enamel with water, saliva and human plasma after etching the surface with 10% polyacrylic acid and then bonded with RMGIC. The authors reported an increase in bond strength over wet etched enamel when the surface was first contaminated with saliva. Even higher bond strengths were found when the surfaces were contaminated with human plasma. The authors were

unable to explain the bond strengths found when the enamel was contaminated with plasma (Kirovski & Madazarova, 2000).

Reddy, Marker and Ellis (2003) evaluated the effects of blood contamination on composite and RMGIC before and after bonding to conditioned enamel. They reported a decrease of nearly 50% in bond strength when the enamel was contaminated with human blood prior to bonding with either material. There was no significant difference if the contamination occurred immediately after the bracket was bonded compared to no contamination at all (Reddy et al, 2003).

In another study, the enamel surface was contaminated with water, saliva or blood, and the bracket was placed on the enamel with the contaminate layer intact on the enamel surface. They reported a 70-80% decrease in bond strength with blood contamination present compared to when saliva or water was present. This result was found regardless of whether or not the enamel had been etched prior to contamination. The bond strengths with water were similar to those obtained with saliva contamination or dry enamel, and all three Groups exceeded 13 MPa of mean shear bond strength (Itoh et al, 2000).

With regard to the blood contamination results, the authors also reported that 85% of transmitted light used for curing was attenuated by as little as 0.015 mm of blood. With 0.5 mm of blood present, 100% of transmitted light from a light-curing unit was attenuated, compared to 14% if 0.5mm of saliva was present. This light attenuation can drastically affect the curing of RMGIC by inhibiting activation of the photo-polymerizing agent, negating the light cured properties of the cement. The authors suggest this may have caused the dramatic reduction in bond strength (Itoh et al, 2000).

Despite the presence of contamination, average bond strengths reported for RMGIC are often greater than the forces used during orthodontic treatment (between 6-8 MPa) (Cacciafesta et al, 1998; Chung et al, 1999; Compton et al, 1992; Itoh et al, 1999; Itoh et al, 2000; Kirovski & Madzarova 2000; Lippitz et al, 1998; Reynolds 1975; Rix et al, 2001). However, studies on the effect of hemostatic agents, or a mixture of hemostatic agent and blood contamination during bonding with RMGIC such as during a surgical exposure and bracket placement procedures, have not been found in the literature.

Hemostatic Agents

Hemostatic agents are often implemented to aid in isolating a tooth's surface during use of a bonding agent. Reports on the effects of hemostatic agents on dentin are available in the literature. No published studies were found on the effects of hemostatic agents on enamel or bond strengths obtained in their presence.

Studies evaluating the effect of hemostatic agents on dentinal smear layers have reported an etching effect. The etching results in opening of the dentinal tubules and removal of the smear layer (Land et al, 1994, Land et al, 1996). The combination of these factors has been associated with enhance mechanical retention when bonding (Garcia-Godoy 1992). The etching reported is not surprising when considering the pH of the solutions commonly associated with hemostasis. For example, the pH of 10-21% AlCl and 15-21% $FeSO_4$ ranged from 0.8-2.0 pH (Land et al, 1994, Woody et al, 1993).

Land and associates (1994) have studied the effect of hemostatic agents on the dentin. Exposure to 15.5% ferric sulfate (Astringedent, pH=0.8) hemostatic solution for 30 seconds after crown preparation effectively removed the dentinal smear layer but the

dentinal tubules remained largely occluded. Two minutes of exposure completely removed the smear layer and exposed the dentinal tubules. Noticeable dentinal etching was noted after five minutes of 21.3% $AlCl_3$ (Hemodent, pH=1.3) exposure as well as with 8% epinephrine (Orostat, pH=2.0) exposure (Land et al, 1994).

It has been suggested that cleansing of the tooth surface may enhance mechanical retention and the ability to obtain a stronger bond with composites and RMGIC (Garcia-Godoy 1992, Glasspoole et al, 2002). This etching effect of the hemostatic agents relative to non-etched enamel needs to be evaluated for potential increase in the mean shear-peel bond strengths obtained with RMGIC to enamel as a byproduct of providing hemostasis.

Bond Failure Site

Many current studies have evaluated the site of bond failure when comparing composites to RMGIC and the effects of enamel conditioning and contamination on the bond failure site. Failure of a bonding material to withstand either the tensile or shearing forces placed on an appliance would depend on two main factors: 1) bond strength including the enamel-adhesive bond, the cohesive strength of the bonding material and the adhesive-attachment bond; and 2) the surface area of the attachment. A cohesive failure of the material would indicate that both the bond strength attained with the tooth and with the attachment is higher than the cohesive strength of the material.

In order for a material to be suited for orthodontic use it must produce a reliable bond that withstands forces typically applied during treatment. Reynolds reported that the maximum orthodontic forces are unlikely to exceed 5 Kg (48N) and that maximum

mean tensile bond strengths of 60-80 Kg/cm^2 (~6-8 MPa) would be sufficient for orthodontic treatment (Reynolds-1975).

Using a stereoscopic light microscope, Compton and associates (1992) evaluated the site of bond failure when using light and self cured glass ionomer cements on conditioned enamel surfaces. Adhesive-enamel failures only occurred in self-cured GI samples (23%). Cohesive failure occurred 77% of the time with self-cured and 50% of the time with light cured RMGIC. Failure at the adhesive-bracket interface occurred in 50% of the light cured, but was not seen in the self cured GI. This study suggests that with light cured materials, cohesive strength and adhesive bond strength to the bracket is less than its adhesive bond to the enamel. Also the light cured RMGIC appeared to have a stronger bond to enamel than the self cured GI (Compton et al, 1992).

Cortes and associates (1993) reported 100% cohesive failure of RMGIC (Fuji II LC) when the enamel was etched with 10% phosphoric acid. All non-etched samples failed at the adhesive-enamel bond (Cortes et al, 1993). Similar results were published by Shammaa and colleagues (1999). In this study 21 of 30 samples left no adhesive on the unconditioned human enamel with either a wet or dry surface when bonded with Fuji Ortho LC. The bond failure sites were evaluated at 10x magnification (Shammaa et al, 1999).

Cacciafesta reported bond failure analysis that revealed failure of the RMGIC (Fuji Ortho LC) bond predominantly at the enamel-adhesive interface. This finding was found regardless of conditioning or contamination with saliva. Only when using a mechanically retentive ceramic bracket did the bonds fail predominantly at the bracket-

adhesive interface. Analysis was performed under light stereomicroscope at 20x magnification for this study (Cacciafesta et al, 1998).

Comparing light-cured composites and RMGIC in 1998, Cook and Youngson (1998) used a projection microscope to determine the percent of adhesive remaining on the enamel after bond failure. They found 89% of the composite samples had adhesive remaining on the enamel surface, while 57% of the RMGIC samples had adhesive remaining on the enamel surface.

Chung, Cuozzo and Mante (1999) reported 100% failure of the RMGIC bond at the adhesive-enamel interface when the enamel was not conditioned. When the enamel was conditioned with 37% phosphoric acid, 100% failure at the adhesive-bracket interface occurred, indicating greater enamel-adhesive bond strength. While this evaluation was done by visual interpretation of the samples, the report does not clarify if magnification was used (Chung et al, 1999).

In a study by Glasspoole and associates (2002), when the enamel was pretreated with 10% polyacrylic acid or 35% phosphoric acid, bond failures were found to be cohesive or partially cohesive/partially at the enamel-adhesive interface. Without any surface treatment, failure was primarily at the enamel-adhesive interface with few cohesive failures. Their analysis was done with 12x magnification under a light microscope (Glasspoole et al, 2002). These results show that a conditioned enamel surface often produces a stronger enamel-adhesive bond than a non-conditioned enamel surface, resulting in cohesive bond failure.

Enamel Damage During Bonding and Debonding

Data presented by Hobson and associates in 2001 reported a 40% enamel fracture rate when removing brackets bonded to mandibular incisors with a light cured composite, at a mean force of 9.5 MPa. The authors suggested that for mandibular incisors, bond strength greater than 8-9 MPa more than exceeds clinical requirements. Any greater bond strength may lead to excessive enamel damage during debonding (Hobson et al, 2001).

The decreased bond strength of the RMGIC compared to composites may help minimize enamel fracture during debonding procedures, and was recommended by Kusy in his 1994 letter to the editor, "When is stronger better?" published in the American Journal of Orthodontics and Dentofacial Orthopedics, August, 1994. In his letter he states "if that additional strength (obtained with a composite over resin modified glass ionomer cement) did not improve or optimize the clinical treatment of patients, and especially if it had a negative impact on the treatment of patients then the strength criterion would be, by itself grossly misleading." In this letter he is referring to research supporting the use of composite cements based on their higher bond strength. Dr. Kusy is questioning the need for the higher bond strengths in light of the tendency of composite to cause enamel damage during debonding due to its high bond strength.

Finally, enamel is less affected by the use of RMGIC bonding procedures when compared to traditional composites because phosphoric acid is not required to prepare the teeth and the RMGIC can be scaled off the enamel using hand instruments during debonding, minimizing the damage often caused by the rotary instruments needed to remove traditional composite from the enamel surface (Jobalia et al, 1997, White 1986).

Enamel Type Used

Current studies have utilized a variety of sources for the enamel substrate in bonding. The majority of the studies use bovine incisors or human premolars although incisors, canines and molars have been used as the enamel source. It is important to consider the source of the enamel when comparing the bonding strengths reported.

Bovine teeth have been compared to human teeth in order to determine their applicability for bonding studies due to their easy availability. One report stated that with bovine teeth, critical surface tension of the enamel was lower, the crystal grains were larger and more lattice defects were present due to their rapid formation in development compared to human teeth (Nakamichi et al, 1983). The mean values reported in this study were all slightly lower than with human enamel, but were not statistically significant. Despite the differences, the authors suggest that bovine teeth are an acceptable alternative for bonding studies in place of human enamel (Nakamichi et al, 1983).

Hobson and associates (2001) reported that with human teeth, the type of tooth had a significant effect ($p=0.05$) on bond strength and bond failure site when using light cured composite. Significant differences were noted in bond strengths between incisors, canines, premolars and molars in this study as well as between first and second premolars and premolars from different arches. The bond strength of the incisors was generally the highest, and the bond strengths decreased progressively on more posterior teeth. The authors believe these findings agree with the theory that the increase in aprismatic enamel on posterior teeth may decrease bond strength (Hobson et al, 2001).

Enamel Preparation Prior to Bonding

Current studies have prepared the tooth surface for bonding by using many different techniques. Some of the common techniques in the literature involve the use of: silicate carbide (SiC) abrasives, fluoride free pumice, polishing pastes or no abrasive treatment at all prior to bonding (Bishara et al, 1998; Cacciafest et al, 1998; Compton et al, 1992; Cortez et al, 1993; Glasspoole et al, 2002; Kanemura et al, 1999;). These treatments may result in a flat enamel surface or leave the convex surface found naturally and may alter the characteristics and fluoride saturation of the enamel rods exposed for bonding (Cortez et al, 1993; Glasspoole et al, 2002; Kanemura et al, 1999). Kanemura, Sano and Tagami (1999) studied the effect of grinding the enamel surface compared to cleansing with toothpaste prior to bonding. The authors do not specify if the toothpaste was fluoride free. They reported no significant difference between the two preparation methods when the enamel was etched with phosphoric acid prior to bonding. However, when treating intact enamel, higher pH self-etch primers produced significantly lower bond strengths compared to the bond strengths obtained with phosphoric acids. SEM evaluation revealed that the greater etching with phosphoric acid promoted deeper resin penetration into the intact enamel layer than with self-etching primers (Kanemura et al, 1999).

Purpose of Study

The purpose of this study was to determine the mean shear-peel bond strength of a light cured orthodontic RMGIC bonding agent (Fuji Ortho LC) when applied to intact enamel with and without contamination of blood, hemostatic agent, or blood-hemostatic agent mixtures. The values obtained were to be compared to the bond strengths reported in the literature as necessary during orthodontic treatment and tooth extrusion. Thus the study will determine whether a bond made under the contaminated conditions is sufficient to resist failure from the shear forces applied by the orthodontist during tooth extrusion.

The null hypotheses in this study are:

1. Contamination with blood or hemostatic agent will not alter the mean bond strength of the light-cured glass-ionomer cement compared to the non-contaminated enamel surface.
2. Contamination with blood and hemostatic agent will not alter the mean shear bond strength of the light cured glass-ionomer cement compared to the non-contaminated enamel surface.
3. The mean shear bond strengths obtained with the resin modified glass-ionomer cement will be sufficient to withstand the forces applied during orthodontic treatment.

Materials and Methods

Institutional Review Board Approval

Approval for use of human tissue (teeth and blood) for this project was obtained from Oregon Health and Sciences University (OHSU) Institutional Review Board (IRB) on December 9th, 2003 (IRB approval #8064). The OHSU IRB uses rules and policies from the Office for Human Research Protections (OHRP) to determine the appropriate review level of the project. OHSU IRB determined the tissues utilized in this project were exempt from review according to the Federal Policy for the Protection of Human Subjects Code of Federal Regulations Title 45, Part 46, Subpart A, section 46.101 paragraph b; which allows exemption for research involving the collection or study of existing specimens if the subjects supplying the specimens cannot be identified directly or through identifiers linked to the subjects.

Samples

Seventy unerupted human third molar teeth collected from local area oral surgeons were used as the enamel substrate. The teeth were cleaned of tissue debris and stored at room temperature in a 1% Chloramine T solution. Teeth were used within ninety days of extraction. This allowed for simulation of the enamel conditions encountered when surgically uncovering an impacted tooth and bonding an orthodontic appliance to its enamel surface. Teeth with demineralization or other signs of lengthy exposure to the oral cavity were excluded. Using a high speed hand-piece and #556 crosscut fissure bur (Brassler, USA), the roots of the teeth were removed and the crowns were sectioned vertically into halves along the buccal groove and middle of the lingual surface, taking care not to damage the flat mesial or distal surfaces. This sectioning

allowed the teeth to fit within the mounting blocks utilized (described below). 15 tooth sections had no flat surface available for bonding and were discarded, leaving 125 samples for this study.

The sectioned teeth were mounted in a plexiglass tube of 13 mm internal diameter and 17mm in length using light-activated acrylic (Triad Custom Tray Material, Dentsply International, York, PA) and positioned such that a flat mesial or distal surface protruded for bonding (Figure 1). The acrylic was pressed into the tooth section's empty pulp chamber for retention of the sample. Samples were exposed to light for 5 minutes (Triad Visible Light Curing System, Model 2000; Dentsply International, York, PA 17405-0872). Following the curing, the samples were submerged in distilled water at room temperature (~37°C) for storage prior to treatment and bonding.

Human blood (5 ml) was collected by a phlebotomist on staff at the OHSU Veterans Hospital from the *Venae mediana antebrachii* of a healthy 29-yr-old male human subject to be used as a surface contaminate. The blood was collected into a disposable vacuum tube lined with 0.5 mg heparin (Vacutainer, lined w/ sodium heparin, 5ml; Becton Dickinson Vacutainer Systems, Franklin Lakes, NJ) to prevent clotting. The blood sample was used within two hours of collection to minimize alterations to its constituents.

Hemodent (Premiere Dental Products Co., PA) 21.3% Aluminum Chloride (pH = 1.3) without epinephrine, was used as the hemostatic agent for this study because it is a commonly used, commercially available, hemostatic agent. It was applied to the tooth samples following the manufacturer's recommendations.

Treatments

The 125 samples were randomly divided into five Groups, for a total of 25 samples per Group. Each tooth was rinsed with distilled water and dried for two seconds with an oil-free air syringe to remove excess moisture and avoid desiccation prior to surface treatment. Each sample is wiped with an applicator sponge dampened with distilled water for 2 seconds prior to undergoing one of the five procedures described below (Figure 2):

- *Group 1*: No additional enamel surface treatment.
- *Group 2*: 20 μL of blood placed over the enamel surface.
- *Group 3*: 20 μL of blood placed over the enamel surface, surface wiped with an applicator sponge soaked in distilled water for 5 seconds, removing the visible blood contamination from the bonding surface.
- *Group 4*: 20 μL of blood placed over the enamel surface, surface wiped with an applicator sponge soaked in Hemodent astringent, for 5 seconds, removing the visible blood contamination from the bonding surface.
- *Group 5*: Hemodent soaked applicator sponge wiped over the bonding surface for 5 seconds, surface wiped with an applicator sponge soaked in distilled water for 5 seconds, removing Hemodent from the bonding surface.

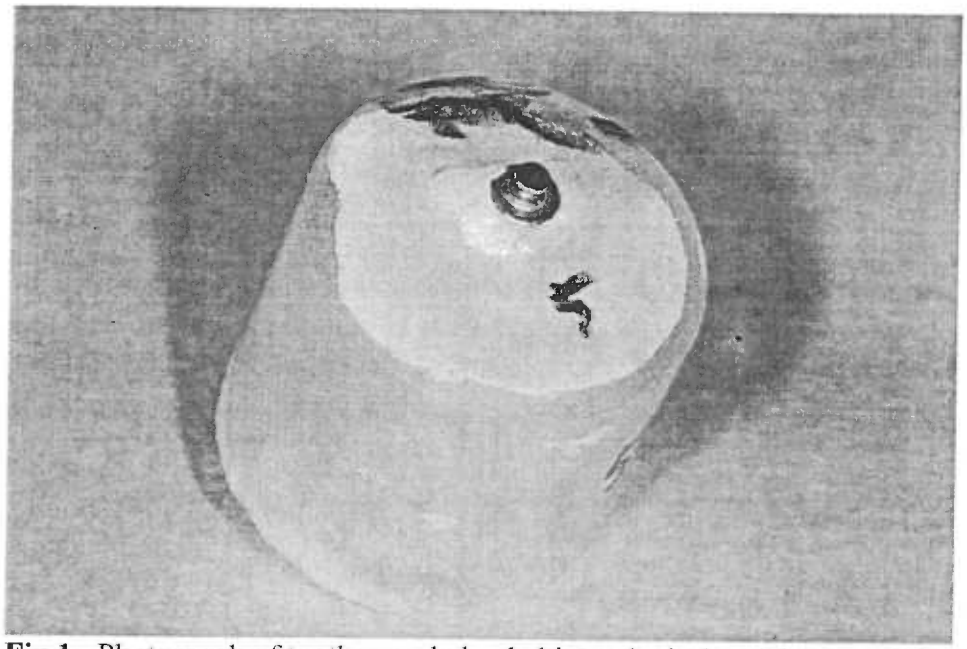

Fig 1. Photograph of tooth sample loaded into plexi-glass tube containing Triad acrylic and an orthodontic button was bonded onto the tooth with glass-ionomer cement.

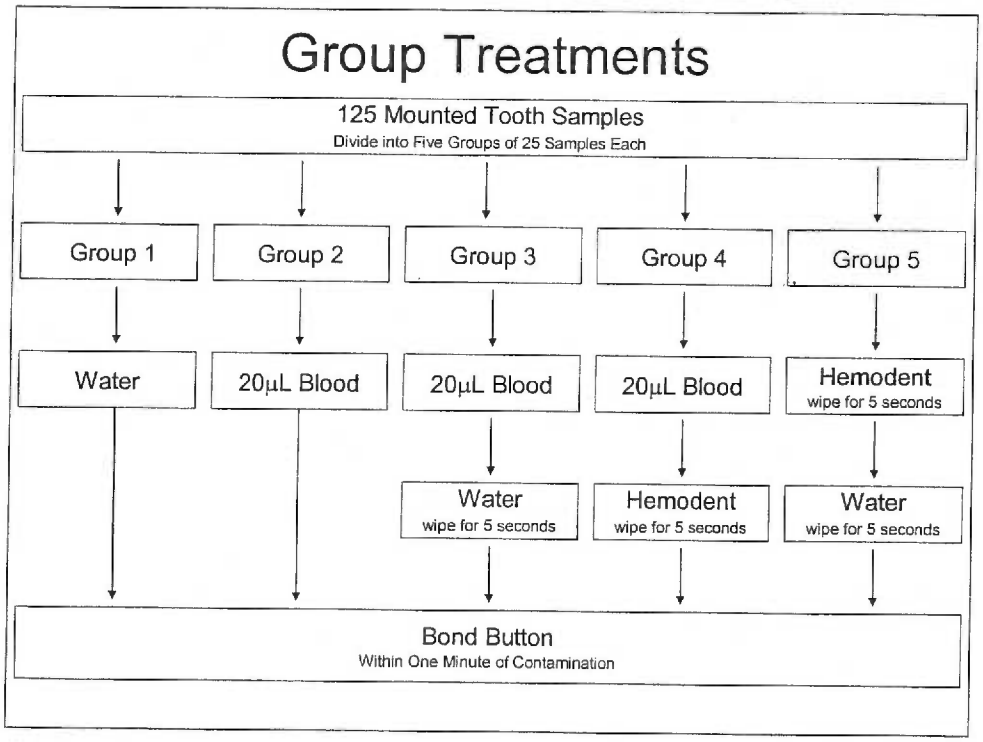

Fig 2. Flow Chart of Group Treatments.

Bonding Procedure

The RMGIC bonding agent Fuji Ortho LC (GC Corp., Tokyo, Japan), was applied following the manufacturers non-etch technique recommendations at room temperature (~37°C). Each capsule was mixed utilizing an amalgamator (Vari-mix III - Model VM-D; Caulk Dentsply, Milford, DE) for 10 seconds.

Within one minute of initial contaminate contact, a FOLC capsule was mixed and dispensed onto the bonding pads of five flat lingual buttons (part #30-000-00, GAC International, Bohemia, NY 11716) using the syringe provided by the manufacturer with the system. The same operator placed all buttons in this experiment. Each button was immediately placed onto the enamel surface of a tooth sample and pressed with sufficient pressure to express excess adhesive until the base was in contact with the tooth. Excess cement was then removed with a dental explorer being careful not to move the button before polymerization. Each capsule had a working time of 3 minutes, which provided adequate time and material for more than 5 buttons per capsule.

Each button was light cured from two opposite sides for 15 seconds each (total of 30 seconds) using a halogen curing light with a minimum of 400 mW/cm^2 emitted radiance (Ortholux XT 704-084, 3M Unitek, Monrovia, CA, USA). Verification of output intensity was made via the radiometer provided on the Ortholux light-curing unit, before and after bonding each Group of 25 samples. The surface was then rinsed with distilled water and the specimens were immersed in 37°C distilled water for 24 hours prior to testing (Thelco Thermal Regulating Unit Model 2, GCA Precision Scientific, Winchester, VA, USA).

Debonding Procedure

For bond strength testing, the acrylic block was secured in the upper frame of a universal testing machine (Instron, Model 1125, Canton, MA, USA) with a 5000 pound load cell (Baldwin SR-4, Model DI 430, Baldwin-Lima-Hamilton Corp., Philadelphia, PA, USA) so that the button base of the sample paralleled the direction in which the shear force would be applied. A steel hook was connected to the button as close to the enamel surface as possible and attached to the Instron machine (See Figure 3). The specimens were displaced at a crosshead speed of 0.01 inch/minute at room temperature until failure. This displacement speed was found to be the most common used in the current published dental literature. Each test was continued until the bracket debonded from the enamel surface.

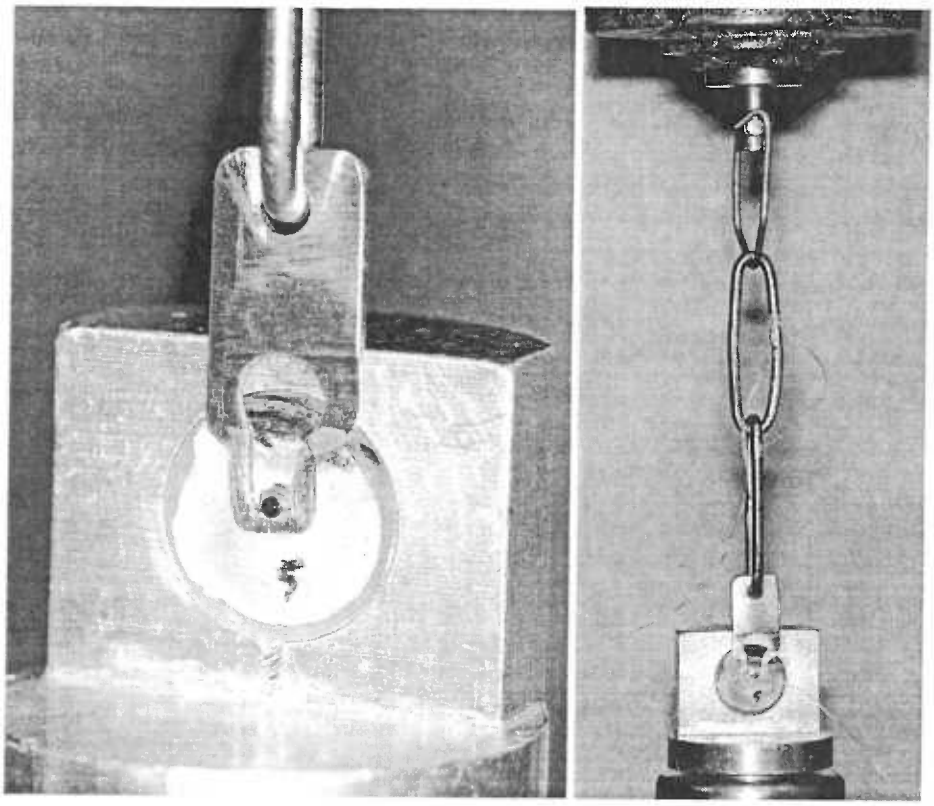

Fig 3. Photographs of mounted sample and Instron apparatus ready for shear-peel bond strength testing.

Conversions

The mean shear-peel bond strengths for the samples were determined from the strip chart recording in pounds force when moving at 0.01 inch/minute. Each measurement was converted to mega-pascals (MPa) as a ratio of force to surface area of the bracket. The bonding surface area of the flat lingual buttons was reported by the engineering department of GAC, the manufacturer, to be 9.072 mm^2.

Analysis of Bond Failure Site

After debonding, all samples were evaluated using the Adhesive Remnant Index (ARI) as described by Artun and Bergland (1984) (Table I). Following this index, each sample was placed into one of five categories based on the percentage of the enamel surface covered by the bonding pad of the button that had RMGIC remaining when viewed under 10x magnification with a stereomicroscope. In addition, five samples from each Group were chosen at random and carbon coated for evaluation under a scanning electron microscope (SEM) at x20 magnification and the percent of material remaining on the sample was calculated using Scion Image Beta 4.02 for Windows (Scion Corporation, Frederick, MD) (Figure 4). The SEM results were compared to the ARI for accuracy.

Table I. Adhesive remnant index categories and descriptions

Category	Description
0	No adhesive on the bonding area
1	< 1/3 of bonding surface with adhesive
2	1/3 - 2/3 of bonding surface with adhesive
3	> 2/3 of bonding surface with adhesive
4	Whole surface covered

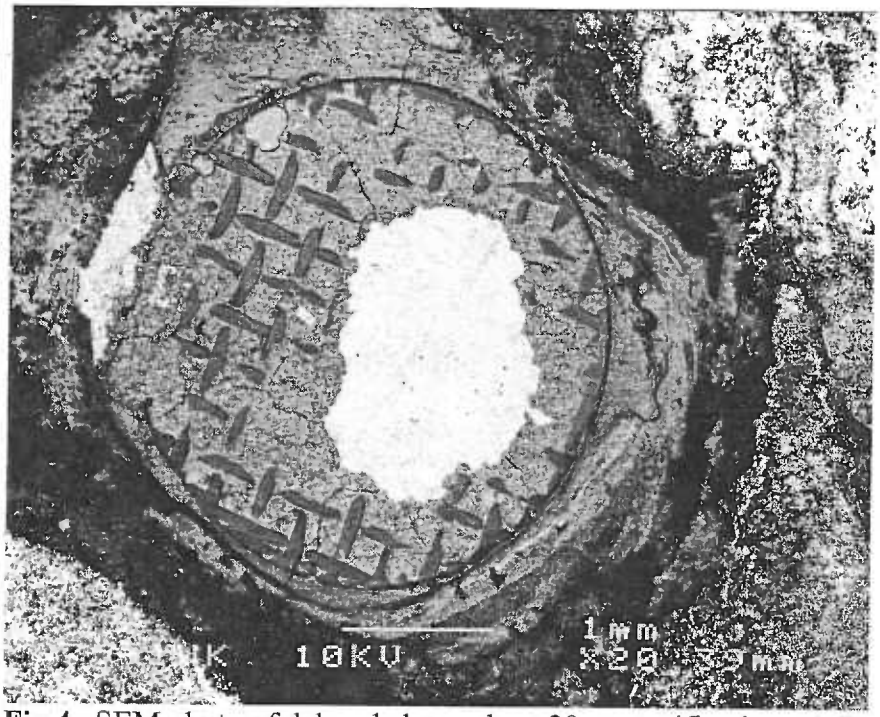

Fig 4. SEM photo of debonded sample at 20x magnification. Adhesive remnant index categorized as a "3" (> 66 % adhesive on the bonding surface), measurement of adhesive remaining on the bonding surface was calculated to be 72.5%.

Statistics

Statistical analysis was performed with the program SPSS (Version 12.0, Leadtools, Lead Technologies, Inc., Chicago, IL, USA). The one-way ANOVA test was used to evaluate for significant differences between the mean shear bond strengths of all five Groups. Tukeys test was incorporated to determine which specific Groups mean shear bond strengths were significantly different from each other. The ARI data is categorical data and requires non-parametric tests for its statistical evaluation. The ARI scores of all groups were ranked prior to testing. The Kruskal-Wallis test was used to evaluate for any significant differences between the mean ranked ARI of all Groups. The Mann-Whitney test and Bonferroni correction was incorporated to determine which specific Groups ranked ARI scores were significantly different from each other. Correlations between shear bond strength and the Adhesive Remnant Index based on surface treatment was evaluated with the Pearson coefficient test. The level of significance for all tests was set to $p=0.05$ (See Appendix B). The statistical tests utilized and the levels of significance are those used for similar research published in the current dental literature.

Results

Table II and Figure 5 summarize the mean shear-peel bond strengths and standard deviations of each Group. All Groups wiped with water prior to bonding, Groups 1, 3 and 5, were not statistically different in their mean shear-peel bond strengths ($p > 0.99$). These Groups had mean shear-peel bond strengths that were greater than the Groups exposed to blood or hemodent and not wiped with water prior to bonding, Groups 2 and 4 ($p < 0.01$). The mean shear-peel bond strengths of Groups 2 and 4 were not statistically different from each other ($p > 0.99$).

The ARI score modes ranged between 1 (Groups 1, 2 and 5) and 2 (Groups 3 and 4) with individual samples showing a range of 0-3 and no sample had an index of 4 (Tables III and IV). The ranked ARI results for each Group and the frequency distribution of each score is listed in Table IV and Figure 6. There was a significant difference in the ARI scores between Groups 1, 2, 4 and 5 compared to Group 3 ($p<0.001$).

There was a significant positive correlation of 0.278 (Pearsons two-tailed test) found between the MPa and ARI when all samples were combined ($p \leq 0.01$). There was no significant correlation between the MPa and ARI within each individual Group (Appendix B).

Table II. Groups, number of samples, conditions applied prior to bonding and descriptive statistics of each Group.

Group	N	Contaminate #1	Contaminate #2	Mean	SD	SE	Range	95% Confidence Interval	
								Lower Bound	Upper Bound
1	25	Water	-	7.08 [a]	2.27	0.45	7.74	6.14	8.01
2	25	Blood	-	2.98 [b]	2.86	0.57	9.71	1.80	4.16
3	25	Blood	Water	7.08 [a]	2.50	0.50	10.32	6.04	8.10
4	25	Blood	Hemodent	2.82 [b]	1.51	0.30	6.51	2.19	3.43
5	25	Hemodent	Water	6.77 [a]	2.18	0.43	8.35	5.86	7.67

Means designated by superscript a are significantly different from those with superscript b using ANOVA and Tukeys tests ($p<0.01$). Same letter superscripts (a and b) designate means with no significant differences using ANOVA and Tukeys tests ($p>0.99$).

Table III. Frequency of distribution of Adhesive Remnant Index scores (%).

Group	ARI = 0	ARI = 1	ARI = 2	ARI = 3	ARI = 4
1	1 (4%)	11 (44%)	10 (40%)	3 (12%)	0 (0%)
2	6 (24%)	12 (48%)	6 (24%)	1 (4%)	0 (0%)
3	0 (0%)	3 (12%)	12 (48%)	10 (40%)	0 (0%)
4	1 (4%)	10 (40%)	13 (52%)	1 (4%)	0 (0%)
5	4 (16%)	13 (52%)	7 (28%)	1 (4%)	0 (0%)

Score 0 = no adhesive remains on the enamel surface bonding area; 1 = less than 1/3 of the area was covered with adhesive; 2 = 1/3 - 2/3 of the area was covered with adhesive; 3 = greater than 2/3, but not all of the area was covered with adhesive; 4 = all of the area was covered with adhesive.

Table IV. Adhesive Remnant Index mode, ranked mean and standard deviation.

Group	Mode	Ranked Mean	Ranked SD
1	1	64.84 [a]	31.96
2	1	44.54 [a]	32.11
3	2	92.48 [b]	25.88
4	2	64.22 [a]	28.47
5	1	48.92 [a]	31.03

Means designated by superscript a are significantly different from those with superscript b using Kruskal-Wallis and Mann-Whitney tests ($p<0.01$). Same letter superscripts (a and b) designate means with no significant differences using Kruskal-Wallis and Mann-Whitney tests ($p>0.99$).

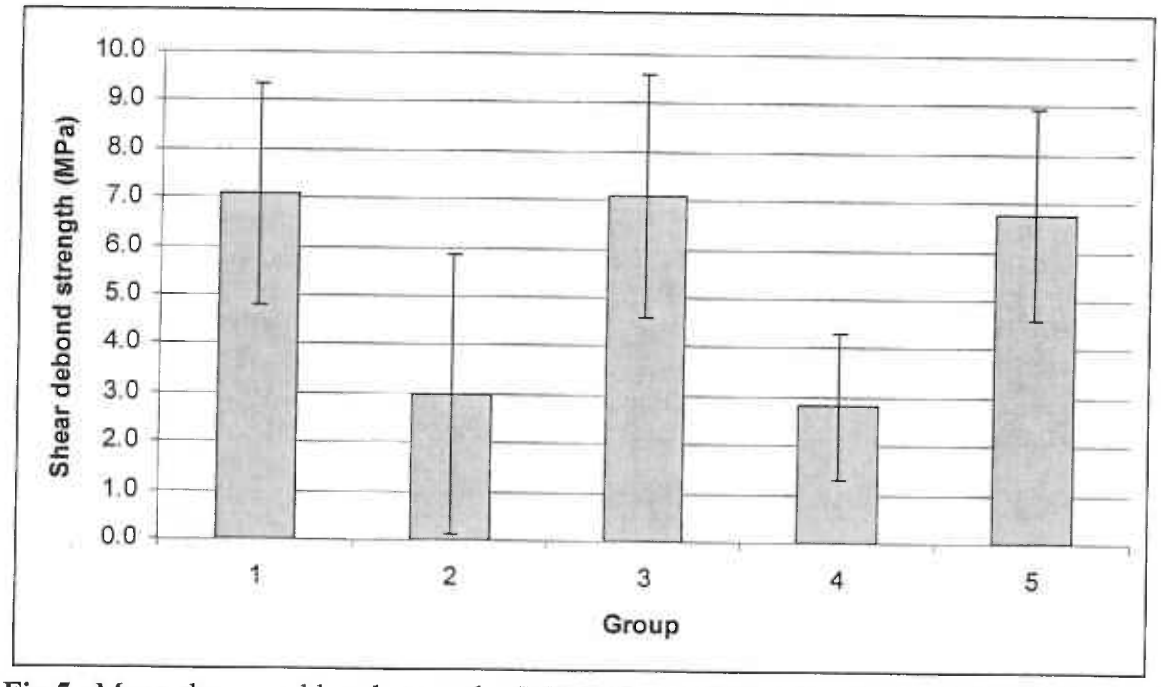

Fig 5. Mean shear-peel bond strengths (MPa) of each Group (1, water only; 2, blood only; 3, blood then water; 4, blood then Hemodent; 5, Hemodent then blood). Error bars represent the standard deviations of each Group.

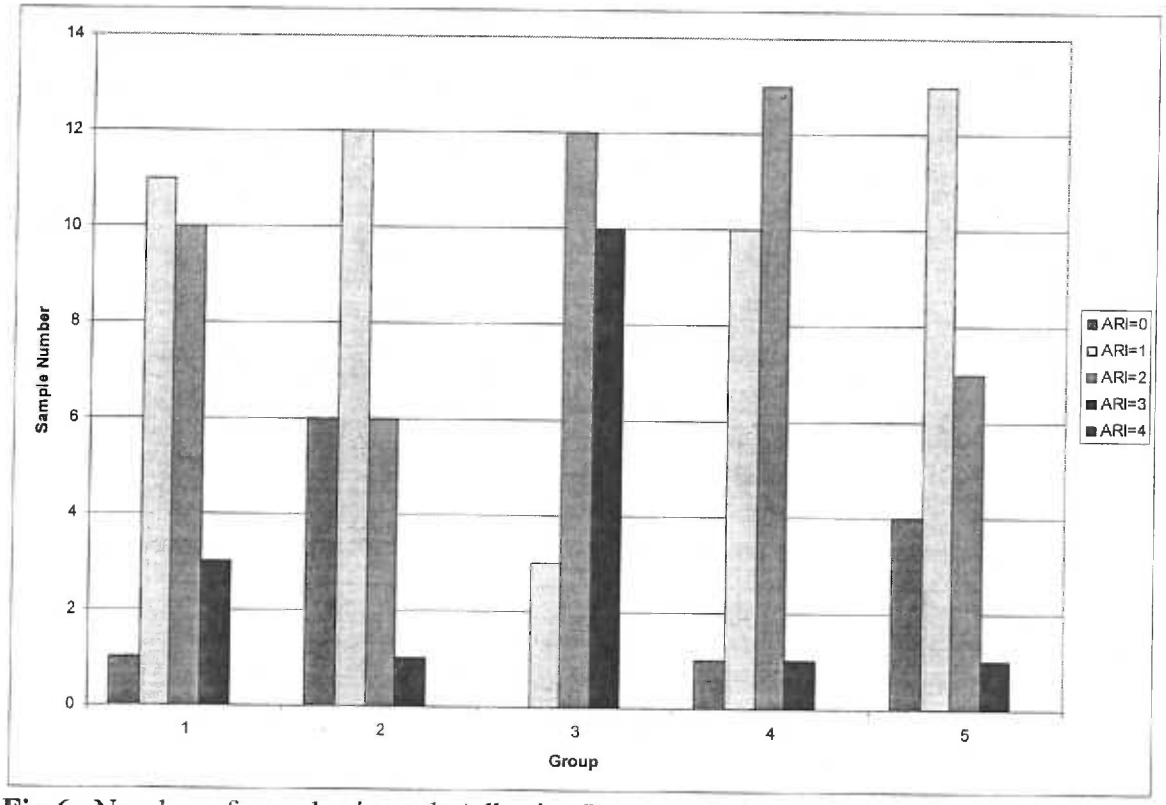

Fig 6. Number of samples in each Adhesive Remnant Index category within each Group.

Discussion

This experiment was undertaken with the intent of replicating the conditions encountered by a surgeon when exposing a tooth in order to bond an orthodontic button or bracket. The objective was to determine whether a clinically acceptable bond could be obtained using RMGIC on a blood and/or Hemodent contaminated enamel surface with no other pretreatment by the practitioner.

The unique property of enhanced polymerization when small amounts of water are present on the bonding surface makes RMGIC potentially well suited for use in bonding brackets to un-erupted teeth. During this type of procedure, it is often very difficult to isolate all areas of enamel from blood or saliva contamination and to keep the surface dry during the bonding procedure. The addition of agents such as anesthetics, vasoconstrictors and hemostatic agents add to the potential list of contaminates to the bonding site and yet studies on the effect of many of these on the bond strength attainable with RMGIC has not been to be published. While there are many substances that could be studied to determine their effects on a bond attained with a RMGIC, Hemodent was selected as the hemostatic agent in this study because of its common use in oral surgery and general dentistry for hemostasis. Knowing the affect on bond strength of chemicals that may contaminate the tooth surface during bonding procedures will make the production of a clinically acceptable bond more predictable and aid practitioners in bonding under similar situations such as when placing a sub-gingival composite restoration or cementing a crown in a contaminated environment.

As reported by Katona in his 1994 article, pure shear force testing is not currently possible. The term 'shear-peel' is more appropriate due to the fact that the debond force is being applied at a distance from the adhesive producing tensile stresses in one location and compressive forces in another, all in the same system. This method of application cannot produce a true shear force on the system. For this experiment the point of force application was placed as close to the adhesive-enamel interface as possible in order to simulate a sheer debond situation as closely as possible.

Results of this study generally showed two levels of mean shear-peel bond strengths. The first level consisted of samples wiped with water prior to bonding regardless of contaminate used on the enamel (Groups 1, 3 and 5). These samples had mean shear-peel bond strengths of 6.77 MPa to 7.08 MPa, values that are within the 6-8 MPa recommended bond strength for orthodontic treatment. Our results are similar to the mean shear-peel bond strengths reported in previous *in vitro* research with RMGIC (Cortez et al, 1993, Fricker 1994, Itoh et al, 1999, Jobalia et al, 1997, Kirovski & Madazarova 2000, Lippitz et al, 2000, Reynolds 1975, Rix et al, 2001, Silverman et al, 1995).

An interesting finding in this study was the similarity in the effect of blood contamination and Hemodent contamination on bond strength when either was left on the enamel prior to bonding. These Groups (2 & 4), had the lowest mean shear-peel bond strengths in this study (2.98 MPa and 2.81 MPa respectively), significantly lower than when bonding was preceded with a water rinse (Groups 1, 3 & 5; $p < 0.001$). The mean shear-peel bond strengths of Groups 2 and 4 were substantially less than the recommended strengths for orthodontic purposes (Reynolds et al, 1975). The data in

Group 2 is not surprising when compared to other studies in the literature where the effect of blood contamination on the shear-peel bond strength has been determined to be unreliable due to its low sheer bond strength. Itoh and associates (2000) reported a mean shear-peel bond strength of 4.2 MPa (SD=1.8) when the enamel surface was contaminated with human blood, the shear-peel bond strength obtained was 70-80% less than that of moist un-etched enamel when using RMGIC. Reddy and his colleagues (2003) reported a mean shear-peel bond strength of 2.5 MPa (SD=2.1) with RMGIC when the bonding surface was contaminated with blood, 50% lower than the mean bond strength obtained with moist non-etched enamel. Results of the latter study were most similar to results reported here, where the addition of blood contamination decreased the mean shear-peel bond force from 7.08 MPa to 2.98 MPa, or by 42%. The high standard deviation compared to the mean sheer-peel bond strength obtained in the presence of these contaminates shows that the reliability of the bond obtained has a high variability.

Itoh and associates (2000) also evaluated the light attenuation properties of blood and found that 0.015mm of blood will attenuate 85% of the transmitted light due to filtering. This is likely due to a combination of filtering of the wavelengths necessary to activate the camphoquinone (around the 400nm) by the blood components and by scattering of the light by particles in the blood suspension. These results suggest that even a small amount of blood contamination around the button base may inhibit light curing of the RMGIC. This was the situation in Group 2, where bonding of the sample occurred in a pool of blood. The sample did undergo self-curing to obtain a minimal bond to the enamel surface.

Blood sample was stored in a heparin lined vial after collection to prevent coagulating prior to its use. The lining prevents coagulation imitation by acting as a barrier between the blood and the glass vial. The use of a lining was chosen rather than adding heparin pellets to a vial, in order to minimize incorporation of the anticoagulant into the sample. Contact with the heparin lined collection tube may have altered the effect of the blood on RMGIC bonding and curing when compared to an *in-vitro* situation.

Of particular interest was whether the effect of blood contamination on the bond strength could be reduced by cleansing the enamel with Hemodent prior to bonding. Normally, once an enamel surface has been contaminated, a strong acidic solution such as phosphoric acid is required to clean the enamel surface of organic debris prior to bonding with composites (Silverstone et al, 1985). The removal of this organic layer has also been recommended for RMGIC (Itoh et al, 2000, Reddy et al, 2003). Hemodent, with a pH of 1.3, has been shown to have an etching effect when placed onto exposed dentin, removing the smear layer and opening tubules (Land et al, 1994, Land et al, 1996). It has been postulated that the acidity of the Hemodent may act similar to polyacrylic acid, a mild acid which is used occasionally as an enamel conditioner with RMGIC to remove contaminates and aid in surface wetting prior to bonding (Land et al, 1994). There is also the potential for the Hemodent to condition the enamel surface similar to that achieved by phosphoric acid, creating micro-mechanical retentive areas and increased surface area for bonding (Glasspoole et al, 2002).

Our data shows that the addition of the Hemodent did not increase the bond strength after the enamel surface has been contaminated with human blood (Group 4).

The bond strengths obtained were statistically similar to Group 2, contaminated with blood and no Hemodent rinse. The lack of a significant difference between Groups 2 & 4 suggests that following blood contamination there is no benefit to conditioning the surface for one minute with an acidic Hemodent solution. No group evaluating the effects of Hemodent only or Blood, Hemodent and water treatments prior to bonding were used. The addition of these may help to evaluate other treatment scenarios encountered in a clinical setting. The results indicate that bonding with RMGIC in a clinical situation where Hemodent or blood contamination is present would not be advised without cleansing the enamel surface first with water.

Pretreatment with Hemodent prior to bonding to moistened enamel (with no other contaminates; Group 5) was not found to be statistically different from bonding to moistened enamel alone (Group 1). This is additional evidence against the theory that Hemodent may act as a conditioner on the enamel to increase the bond strength, as is the case with polyacrylic acid (Itoh et al, 1999, Kirovski and Madazarova 2000, Lippitz et al, 1998). These results also show Hemodent does not increase bond strength similar to phosphoric acid, which produces micro-porosities in the enamel resulting in more bondable surface area (Cook & Youngson 1998, Glasspoole et al, 2002).

The lack of an etching effect may be due to the weak acidic nature and/or the short time the Hemodent remained on the surface of the enamel prior to bonding (less than one minute). The intrinsic buffering effects of bicarbonate in the blood may have negated the acidity of the Hemodent, decreasing any etching effect. The exposure time of the Hemodent to the enamel surface was limited to one minute in this study. Land and associates in 1994 and 1996 found that two minutes of Hemodent exposure was required

to etch dentin which was shown to etch more rapidly than enamel. It was not surprising that there was no etching effect considering that in this study the exposure time was no more than fifteen seconds, significantly less than that required to etch dentin. In addition, by wiping the surface with the applicator sponge, any increased surface roughness may have been smoothed, possibly negating any potential increase in bond strength due to increased surface area from etching. However, the data does show that the presence of the Hemodent solution prior to wiping the enamel with water does not hinder the bond formed by the RMGIC over that of the moist enamel (Group 1).

To evaluate whether greater shear-peel bond strengths are the result of increased bond formation to the tooth, the presence of adhesive on the enamel bonding surface was evaluated and compared with the bond strength values required to debond the bracket. Among the Groups there was a positive correlation (Pearsons coefficient = 0.278) between the MPa and ARI ($p \leq 0.001$). However, no significant correlations were noted within each individual Group. The difference is likely due to the high sample size when comparing all Groups (n=125) compared to when evaluating each Group (n=25). The low coefficient number and dependence on a high sample size for significance indicates that the correlation is weak. We have been unable to find other literature that evaluated this potential correlation between the mean ARI and mean shear bond strength.

Interestingly, there was a significant difference between the ranked ARI means for Groups 1, 2, 4 & 5 compared to Group 3 (blood wiped with water), the latter having significantly more material adhering to its enamel surfaces than in the other Groups ($p \leq 0.001$; see Appendix B). However, the mean shear-peel bond strengths of Group 1, the bonding environment recommended by the manufacturer, and Group 3 were identical

(see Table II). It was unexpected that wiping the surface clear of visible blood contamination with water was sufficient to remove the detrimental effects of blood contamination on the bond strength and amount of adhesive adhering strongly to the enamel surface. No explanation was found for the higher ARI scores in Group 3.

Only the presence of adhesive remaining on the enamel surface was considered in the ARI. Cohesive failure was not evaluated in this study and it is unknown whether the adhesive remaining on the enamel surface had failed cohesively or at the bracket-adhesive interface.

Areas of excess RMGIC outside of the bonding area were noted in multiple samples. This material was missed when removing excess cement next to the button prior to light curing. The presence of additional adhesive may increase the force required to cause bond failure and result in higher mean shear-peel bond strength. However, the incidence of excess adhesive was random throughout the study and should not have affected one group more than another.

Third molars were chosen for this project due to the ease of collection of un-erupted samples. Hobson and associated (2000) reported that molars tend to have lower mean bond strength due to the presence of an aprismatic enamel surface compared to anterior teeth. Also, the mesial and distal surfaces were used in this study rather than the buccal or lingual which are more frequently used for bonding. This needs to be considered when comparing the sheer-peel bond strengths reported here to other studies. It is possible that the shear-peel bond strengths obtained to a canine or incisor would be higher than our results to a third molar.

Evaluation of the literature shows that high standard deviations and ranges of shear-peel bond strengths are common in other studies. Bishara and associates (1998) reported a range of 0.1-6.5 MPa (SD=1.9) when bonding to wet non-etched enamel and 3.3-11.7 MPa (SD = 2.8) when bonding to wet etched enamel using FOLC. Chung and associates (1999) reported a range of 0.81-7.26 MPa (SD = 1.89) when bonding FOLC to wet non-etched enamel and a range of 1.88-12.91 MPa (SD = 2.46) when bonding to wet etched enamel. According to Reddy and colleagues (2003), mean bond strength of FOLC bonded to non-etched incisors was 3.8 MPa (SD = 2.7) when moist and 2.1 MPa (SD = 2.1) when contaminated with blood prior to bonding.

When evaluating this study, several additional limitations were noted in the methods used. The slow application of force used with the Instron machine may not accurately mimic the rapid force application typically responsible for bond failures seen *in-vivo*. Samples from each Group were not randomized during the preparation of each sample, so there may be a difference in efficiency or slight changes in procedure from the first Group to the last that may have affected the outcome. This may include the actual force used to seat each button. The actual force applied by the operator was not measured and may have changed during bonding. Finally, the curing light was tested for a minimum output level, not for actual output value during the experiment; so it is not known if there was any change in output intensity during the project.

Composite resins have been shown to have higher tensile and compressive bond strengths than RMGIC and have proven to be effective at minimizing orthodontic bond failures when applied appropriately. The drawback to their use in environments such as the one studied is the bonding surface must remain contamination free during the entire

bonding procedure and when etched enamel is exposed to contamination, re-etching of the enamel surface is required in order to obtain a satisfactory bond (Silverstone et al, 1985). If adequate isolation can be maintained during the bonding procedure, use of resin composites will provide reliable tensile and shear bond strengths for orthodontic extrusion. If isolation cannot be maintained, the operator may consider wiping the surface with water prior to bonding and attaching the appliance with a RMGIC.

The data obtained in this study and in previous literature shows that when water is present on the bonding surface, RMGIC is capable of producing bond strength above the minimum reported necessary for orthodontic treatment. The 95% confidence interval for bond strengths produced bond strengths of 5.86 MPA to 8.10 MPa, values within those recommended for orthodontic treatment (see Tables II and III; Reynolds 1975). In addition, bonding can be reliably produced even when the surface is contaminated with blood or Hemodent as long as the surface is wiped clean with water before bonding.

Conclusions

In this study the effects of blood and Hemodent contamination of enamel before bonding with RMGIC on the mean shear-peel bond strengths and ARI was evaluated.

The findings of this study were:

- Clinically acceptable shear-peel bond strengths of 6-8 MPa can be produced when bonding to an untreated, water moistened enamel surface with RMGIC.
- Bonding with RMGIC to enamel in the presence of Hemodent or blood contamination will result in significantly decreased bond strength.
- Conditioning the enamel with Hemodent, a mild acid, for one minute prior to water rinsing and then bonding with RMGIC will not significantly increase shear-peel bond strength over that of water moistened enamel.
- Hemodent or blood contamination may be removed by wiping the enamel surface with a water soaked applicator sponge prior to bonding with RMGIC. The resulting shear-peel bond strengths will not be significantly different from those obtained with moist enamel without contamination exposure.
- A weak positive correlation exists between the mean bond strength obtained and ARI.

Future studies aimed at the effects of other agents such as anesthetics, vasoconstrictors and hemostatic agents on the ability of RMGIC to produce a clinically acceptable bond to enamel and dentin should be useful to clinicians. The effect of theses agents on bonding with either light cured composites and RMGIC may have wide spread clinical applicability. In addition, controlled *in vivo* studies directed toward increasing the reliability of the bond strength obtained with RMGIC would be beneficial in developing a clinically acceptable protocol.

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Appendix A

Excel Data Spreadsheet

Appendix B

SPSS Statistical Output

ANOVA: Test MPa Between All Groups

Oneway ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
MPa	Between Groups	500.235	4	125.059	23.484	.000
	Within Groups	639.044	120	5.325		
	Total	1139.279	124			

MPa

Tukey HSD

Group	N	Subset for alpha = .05	
		1	2
Blood then Hemodent	25	2.81640	
Blood Only	25	2.98280	
Hemodent then Water	25		6.77040
Blood then Water	25		7.07520
Water Only	25		7.08000
Sig.		.999	.990

Means for Groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 25.000.

Kruskal-Wallis: Test ARI Ranking Between All Groups

All Groups ARI Ranks

	Group	N	Mode	Mean Rank	SD
ARI RANK	1-Water Only	25	1	64.84	31.96
	2-Blood Only	25	1	44.54	32.11
	3-Blood then Water	25	2	92.48	25.88
	4-Blood then Hemodent	25	2	64.22	28.47
	5-Hemodent then Water	25	1	48.92	31.03
	Total	125			

Test Statistics: All Groups

	RANK of ARI
Chi-Square	30.576
df	4
Asymp. Sig.	.000

a Kruskal Wallis Test
b Grouping Variable: Group

Tukey: Test MPa Between Two Groups

Multiple Comparisons

Tukey HSD

Dependent Variable	(I) Group	(J) Group	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
MPa	Water Only	Blood Only	4.097200(*)	.652709	.000	2.28940	5.90500
		Blood then Water	.004800	.652709	1.000	-1.80300	1.81260
		Blood then Hemodent	4.263600(*)	.652709	.000	2.45580	6.07140
		Hemodent then Water	.309600	.652709	.990	-1.49820	2.11740
	Blood Only	Water Only	-4.097200(*)	.652709	.000	-5.90500	-2.28940
		Blood then Water	-4.092400(*)	.652709	.000	-5.90020	-2.28460
		Blood then Hemodent	.166400	.652709	.999	-1.64140	1.97420
		Hemodent then Water	-3.787600(*)	.652709	.000	-5.59540	-1.97980
	Blood then Water	Water Only	-.004800	.652709	1.000	-1.81260	1.80300
		Blood Only	4.092400(*)	.652709	.000	2.28460	5.90020
		Blood then Hemodent	4.258800(*)	.652709	.000	2.45100	6.06660
		Hemodent then Water	.304800	.652709	.990	-1.50300	2.11260
	Blood then Hemodent	Water Only	-4.263600(*)	.652709	.000	-6.07140	-2.45580
		Blood Only	-.166400	.652709	.999	-1.97420	1.64140
		Blood then Water	-4.258800(*)	.652709	.000	-6.06660	-2.45100
		Hemodent then Water	-3.954000(*)	.652709	.000	-5.76180	-2.14620
	Hemodent then Water	Water Only	-.309600	.652709	.990	-2.11740	1.49820
		Blood Only	3.787600(*)	.652709	.000	1.97980	5.59540
		Blood then Water	-.304800	.652709	.990	-2.11260	1.50300
		Blood then Hemodent	3.954000(*	.652709	.000	2.14620	5.76180

Mann-Whitney: Test ARI Ranking Between Two Groups

Group 1 vs 2 Ranked ARI

	Group	N	Mean Rank	Sum of Ranks
RARI	1	25	29.74	743.50
	2	25	21.26	531.50
	Total	50		

Test Statistic: Groups 1 vs 2

	RARI
Mann-Whitney U	206.500
Wilcoxon W	531.500
Z	-2.209
Asymp. Sig. (2-tailed)	.027

a Grouping Variable: Group

Group 1 vs 3 Ranked ARI

	Group	N	Mean Rank	Sum of Ranks
RARI	1	25	19.66	491.50
	3	25	31.34	783.50
	Total	50		

Test Statistic: Groups 1 vs 3

	RARI
Mann-Whitney U	166.500
Wilcoxon W	491.500
Z	-3.027
Asymp. Sig. (2-tailed)	.002

a Grouping Variable: Group

Group 1 vs 4 Ranked ARI

	Group	N	Mean Rank	Sum of Ranks
RARI	1	25	25.60	640.00
	4	25	25.40	635.00
	Total	50		

Test Statistic: Groups 1 vs 4

	RARI
Mann-Whitney U	310.000
Wilcoxon W	635.000
Z	-.053
Asymp. Sig. (2-tailed)	.957

a Grouping Variable: Group

Group 1 vs 5 Ranked ARI

	Group	N	Mean Rank	Sum of Ranks
RARI	1	25	28.84	721.00
	5	25	22.16	554.00
	Total	50		

Test Statistics: Groups 1 vs 5

	RARI
Mann-Whitney U	229.000
Wilcoxon W	554.000
Z	-1.758
Asymp. Sig. (2-tailed)	.079

a Grouping Variable: Group

Groups 2 vs 3 Ranked ARI

	Group	N	Mean Rank	Sum of Ranks
RARI	2	25	16.68	417.00
	3	25	34.32	858.00
	Total	50		

Test Statistics: Groups 2 vs 3

	RARI
Mann-Whitney U	92.000
Wilcoxon W	417.000
Z	-4.474
Asymp. Sig. (2-tailed)	.000

a Grouping Variable: Group

Groups 2 vs 4 Ranked ARI

	Group	N	Mean Rank	Sum of Ranks
RARI	2	25	21.18	529.50
	4	25	29.82	745.50
	Total	50		

Test Statistics: Groups 2 vs 4

	RARI
Mann-Whitney U	204.500
Wilcoxon W	529.500
Z	-2.263
Asymp. Sig. (2-tailed)	.024

a Grouping Variable: Group

Groups 2 vs 5 Ranked ARI

	Group	N	Mean Rank	Sum of Ranks
RARI	2	25	24.42	610.50
	5	25	26.58	664.50
	Total	50		

Test Statistics: Groups 2 vs 5

	RARI
Mann-Whitney U	285.500
Wilcoxon W	610.500
Z	-.568
Asymp. Sig. (2-tailed)	.570

a Grouping Variable: Group

Groups 3 vs 4 Ranked ARI

	Group	N	Mean Rank	Sum of Ranks
RARI	3	25	31.92	798.00
	4	25	19.08	477.00
	Total	50		

Test Statistics: Groups 3 vs 4

	RARI
Mann-Whitney U	152.000
Wilcoxon W	477.000
Z	-3.384
Asymp. Sig. (2-tailed)	.001

a Grouping Variable: Group

Groups 3 vs 5 Ranked ARI

	Group	N	Mean Rank	Sum of Ranks
RARI	3	25	33.90	847.50
	5	25	17.10	427.50
	Total	50		

Test Statistics: Groups 3 vs 5

	RARI
Mann-Whitney U	102.500
Wilcoxon W	427.500
Z	-4.291
Asymp. Sig. (2-tailed)	.000

a Grouping Variable: Group

Groups 4 vs 5 Ranked ARI

	Group	N	Mean Rank	Sum of Ranks
RARI	4	25	28.92	723.00
	5	25	22.08	552.00
	Total	50		

Test Statistics: Groups 4 vs 5

	RARI
Mann-Whitney U	227.000
Wilcoxon W	552.000
Z	-1.812
Asymp. Sig. (2-tailed)	.070

a Grouping Variable: Group

Pearson's Correlation: MPa to ARI

Group 1-5 Correlations

		MPa	ARI
MPa	Pearson Correlation	1	.278(*)
	Sig. (2-tailed)	.	.002
	N	125	125
ARI	Pearson Correlation	.278(*)	1
	Sig. (2-tailed)	.002	.
	N	125	125

Correlation is significant at the 0.01 level (2-tailed).

Group 1 Correlations

		MPa	ARI
MPa	Pearson Correlation	1	.256
	Sig. (2-tailed)	.	.218
	N	25	25
ARI	Pearson Correlation	.256	1
	Sig. (2-tailed)	.218	.
	N	25	25

Group 2 Correlations

		MPa	ARI
MPa	Pearson Correlation	1	.316
	Sig. (2-tailed)	.	.124
	N	25	25
ARI	Pearson Correlation	.316	1
	Sig. (2-tailed)	.124	.
	N	25	25

Group 3 Correlations

		MPa	ARI
MPa	Pearson Correlation	1	-.112
	Sig. (2-tailed)	.	.592
	N	25	25
ARI	Pearson Correlation	-.112	1
	Sig. (2-tailed)	.592	.
	N	25	25

Group 4 Correlations

		MPa	ARI
MPa	Pearson Correlation	1	-.354
	Sig. (2-tailed)	.	.083
	N	25	25
ARI	Pearson Correlation	-.354	1
	Sig. (2-tailed)	.083	.
	N	25	25

Group 5 Correlations

		MPa	ARI
MPa	Pearson Correlation	1	.323
	Sig. (2-tailed)	.	.116
	N	25	25
ARI	Pearson Correlation	.323	1
	Sig. (2-tailed)	.116	.
	N	25	25