SUSCEPTIBILITY OF RESTORATIONS AND ADJACENT ENAMEL/DENTIN TO EROSION UNDER DIFFERENT SALIVARY FLOW CONDITIONS

by

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DEDICATION
I dedicate this thesis to my Father, my Mother, my Sisters, and my Brothers. Thank you for your support, prayers, and unconditional love.
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INTRODUCTION
Dental erosion is a multifactorial condition that causes irreversible loss of dental hard tissues without bacterial involvement.\textsuperscript{1} Its prevalence has increased significantly due to frequent exposure of teeth to acids, mostly through the consumption of acidic beverages.\textsuperscript{2,3} Efforts have been made to identify the etiological factors involved in the erosion process to aid in the development of reliable preventive and restorative treatments.

Saliva is considered an important factor modulating dental erosion. It can clear and neutralize erosive acids, form the acquired dental pellicle and remineralize eroded dental hard tissues.\textsuperscript{4-7} These protective mechanisms can be potentially reduced in patients with low salivary flow rate. Studies have shown that erosion is associated with low salivary flow rate and/or low buffering capacity.\textsuperscript{8-10} According to Jarvinen et al. in 1991, patients with unstimulated salivary flow rate of 0.1 ml/min or less were at five-times greater risk of erosion than those with normal flow rates.

Management of dental erosion includes preventive and restorative measures, which are determined by the risk status and complexity of the case. With the recent advancement in adhesive dentistry, erosive tooth loss can be restored conservatively by direct bonded materials, including resin composite, conventional glass ionomer cements (GICs) and their hybrids. However, restorations can also be affected by erosive acids,
potentially decreasing their clinical performance and longevity.\textsuperscript{11-13} Resin composite has
shown the best resistance to acid degradation, but it does not release fluoride.\textsuperscript{14-16}

GICs possess some advantages over resin composites such as good adhesion to
enamel and dentin, coefficient of thermal expansion similar to tooth and long-term
fluoride release. Evidence has shown that the high viscosity GIC presents favorable
outcomes considering anatomical form in patients with radiation-induced caries where
the saliva is critically low;\textsuperscript{17} however, GICs generally exhibit inferior mechanical
properties compared to resin-modified glass ionomer (RMGI) cement and resin
composite especially under erosive challenge.\textsuperscript{14}

Although some studies have shown that fluoride releasing restorations play an
important role reducing dental erosion progression,\textsuperscript{14-18} no consensus has been
established.\textsuperscript{19-21} Furthermore, some clinical studies on xerostomic head and neck
radiation patients have suggested that GIC restorations may be more susceptible to
chemical degradation than RMGI, under acidic conditions.\textsuperscript{22-24} We hypothesize that
restorations that release fluoride and present improved mechanical properties (RMGI and
high viscosity GIC) are more resistant to acid degradation than low-viscosity GIC and
provide better protection to surrounding dental substrates compared to the resin
composite, under highly erosive conditions, such as those observed in hyposalivatory
patients. To date, this has not been studied under standardized \textit{in-vitro} conditions.
OBJECTIVES

The specific aims of this study were to:

1. To evaluate the effect of erosion on direct tooth colored restorations and adjacent human enamel and dentin, under different simulated salivary flow rates.

2. Evaluate the influence of restorative materials on enamel and dentin erosive wear.

HYPOTHESES

The following null-hypotheses were tested:

1. There will be no significant difference in surface loss values among restorative materials.

2. There will be no significant difference in surface loss values between low and high salivary flow rates.

3. There will be no significant difference in surface loss values between any of the associations between restorative materials and salivary flow rates.

The alternative hypotheses were:

1. There will be at least one significant difference in surface loss values among restorative materials.

2. There will be a significant difference in surface loss values between low and high salivary flow rate.
3. There will be at least one difference in surface loss values among any of the associations between restorative material and salivary flow rates.
REVIEW OF LITERATURE
DENTAL EROSION: DEFINITION AND HISTOPATHOLOGY

In dentistry, the term erosion is used to describe the progressive dental tissue loss due to chemical processes without involvement of bacteria.\(^1\) It also can be defined as dissolution of tooth by acids when the surrounding aqueous phase is undersaturated with respect to the tooth.\(^{25}\) Recently, the term “biocorrosion” has been introduced in the dental literature. According to Grippo in 2012, biocorrosion encompasses endogenous and exogenous acidic and proteolytic chemical degradation of enamel and dentin, as well as the piezoelectric electrochemical action on the collagen in dentin. However, this term has not gained popularity yet among researchers in the dental erosion area, being rarely used in publications. In this review, the term ‘erosion’ will be used to refer to the chemical loss of dental hard tissues, as described above.

Human enamel and dentin are composed of organic and inorganic phases including mineral, protein, lipid and water. The mineral part is composed of a calcium-deficient carbonated hydroxyapatite and expressed by the formula \((\text{Ca}_{10-x}\text{Na}_x(\text{PO}_4)_{6-y}\text{(CO}_3)_z\text{(OH)}_{2-u}\text{F}_u)\).\(^{26}\) The substitutions in the mineral crystal lattice, especially carbonate, disturb the mineral structure and create the tooth mineral more acid-soluble than pure hydroxyapatite \(\text{Ca}_{10}(\text{PO}_4)_{6}(\text{OH})_2\). Dental erosion occurs when acids and/or chelating agents interact with the surface of the mineral crystals of the teeth. Upon exposure to
erosive solutions, mineral ions from the hydroxyapatite crystals are released, resulting in
demineralization.\textsuperscript{26}

DENTAL EROSION ETIOLOGY

A series of complex events are necessary for erosion to develop, involving the
interaction of chemical, behavioral and biological factors.\textsuperscript{27}

Chemical Factors

Chemical factors are parameters inherent to the erosive agent, which can be of
intrinsic or extrinsic nature. The buffering capacity, pH and calcium, phosphorus and
fluoride concentration are important properties modulating the erosive attack and mineral
dissolution. These properties determine the degree of saturation with respect to the tooth
mineral, which is the driving force for the mineral loss. Solutions undersaturated with
respect to dental hard tissue can lead to surface demineralization, which causes an
increase in the mineral content on adjacent tooth surfaces. This raises the local pH,
preventing further surface dissolution until a new episode of demineralization (erosion)
takes place.\textsuperscript{28}

Extrinsic acids

The sources of extrinsic acids can be categorized into dietary, occupational,
medications and lifestyle.\textsuperscript{29} In the last decade, the consumption of acidic diet increased
significantly, which may be one of the reasons behind the higher prevalence of dental
erosion among different age groups.\textsuperscript{30} Citric, malic and phosphoric acids are commonly
added as ingredients of beverages and foodstuff and their erosive potential have been proven in several in-vitro, in-situ and in-vivo studies.\textsuperscript{29,31,32} In addition, a series of studies state that the erosive potential of an acidic drink is not solely dependent on its pH, but is also strongly influenced by its buffering capacity and by the chelating properties of the food and beverages.\textsuperscript{7} An acid with greater buffering capacity can retard the action of saliva to neutralize the acid. On the other hand, the calcium, phosphate and fluoride content of foods or beverages can make them supersaturated with respect to tooth mineral, preventing demineralization and favoring remineralization. For example, yogurt with a low pH (~4) has hardly any erosive effect due to its high calcium and phosphate content (Lussi et al., 2004). Moreover, calcium and phosphate added to orange juice (pH 4) did not erode enamel after immersion for 7 days.\textsuperscript{33}

Environmental factors involve exposure to acid fumes as in case of workers in factories without proper safeguards. Swimming pools with low pH have also been associated with erosion of an environmental cause. Furthermore, low pH medications and oral hygiene products have also been investigated as potential causes of erosion.\textsuperscript{29}

Lifestyle factors help explain the difference in the dental erosion experience among individuals exposed to the same acid challenge in their diet. Therefore, eating and drinking habits, diets high in acidic fruits and vegetables, excessive consumption of acidic foods and drinks, and oral hygiene practices are of paramount importance and should be thoroughly assessed to determine the erosive potential of a particular acid.\textsuperscript{28}
Intrinsic acids

Another erosive acid equally important is the gastric juice, which reaches the oral cavity as a result of the backflow of gastric contents through the gastroesophageal tract by chronic vomiting or reflux. Gastric juice consists mainly of hydrochloric acid with pH of 1-1.5, which is far lower than the critical pH for enamel and dentin demineralization. Therefore, gastric juice regurgitated into the oral cavity has the potential to induce dental erosion.

Dental erosion induced by intrinsic acids is most likely observed after continuous recurrence of acid exposure over a period of several years. This condition is often associated with chronic vomiting or persistent gastroesophageal reflux as a result of psychological disorders, e.g., in anorexia and bulimia or regurgitation of gastric contents due to some abnormality in the gastrointestinal tract. Individuals with gastric reflux are at higher risk of developing erosion suggesting that careful history taking is important with consideration of other factors of erosion.

Biological Factors

Biological factors of relevance for dental erosion are the saliva (salivary flow, buffering capacity, mineral composition), acquired pellicle, physiological soft tissue movements, dental anatomy and anatomy of oral soft tissues.
Saliva

Saliva has been considered the most important biological factor modulating dental erosion. It has several protective mechanisms, including buffer capacity, acid clearance and neutralization of erosive agents. In addition, it forms the acquired dental pellicle (a protective membrane), reduces demineralization and enhances remineralization. The time required for saliva to neutralize and/or clear the acid from the tooth surface has been measured in vivo and has shown to range between 3 and 5 min. All the above mentioned protective properties of saliva seem to depend on the salivary flow. There are a number of protocols for measuring salivary flow rate, including volumetric test, gravimetric test and calibrated paper test (modified Schirmer’s test). Unstimulated and stimulated whole-salivary flow rates are commonly used to predict the salivary glands hypofunction. The average normal unstimulated flow rate is 0.5 mL/min and unstimulated flow rates of less than 0.1 mL/min are considered an indication of hyposalivation.

Hyposalivation Effects on Erosion

High salivary flow rate provides a favorable environment for the prevention or minimization of initial erosive attack. However, low salivary flow clearly results in insufficient cleaning and buffering of demineralizing acids on tooth surfaces. It is well established that hyposalivation is associated with some conditions that leads to salivary gland impairment. These conditions include diabetes, head and neck radiation therapy, diseases of the salivary gland, alcoholic cirrhosis, cystic fibrosis, hormonal imbalance,
auto-immune diseases, e.g., Sjogren's syndrome, rheumatoid arthritis, systemic lupus erythematosus, among others. Moreover, intake of medications with antisynergic effects is the most common cause of xerostomia. These medications include anticholinergic, antidepressant, antipsychotic, diuretic, antihypertensive, sedative and anxiolytic, antihistamine, opioid analgesic agents and nonsteroidal anti-inflammatory drugs. Clinical studies show association between oral symptoms as erosion and hyposalivation and/or reduced buffering capacity caused by gastroesophageal reflux disease. Thus, assessments of the stimulated and unstimulated flow rates as well as of the buffering capacity of saliva may provide some information about the susceptibility of an individual to dental erosion.

MANAGEMENT OF DENTAL EROSION

Choosing the appropriate preventive and/or restorative intervention is primarily dependent on the patient’s erosion risk level, complexity of the case and extension of the erosive lesions. Thorough case history and assessment of the etiological factors are essential to guide the clinician to the right management approach. Early diagnosis is essential for improving the treatment outcomes.

Prevention

The preventive approach aims to reduce the contact of the erosive acid to the tooth surface and to avoid further mineral loss from enamel and dentin after an erosive attack. When early signs of erosion are noticed clinically or when increased erosion risk
is anticipated, all etiological factors should be considered. The comprehensive case history and diet analysis enable the clinician to identify and highlight the risk factors such as dietary habits and high acid containing foodstufs and beverages as well as possible intrinsic acid exposure. After thorough analysis of the clinical data, a preventive plan is formulated and tailored for every case with the goal of reducing the exposure of enamel and dentin to the erosive agents as much as possible. Preventive recommendations for individuals at high erosion risk include: reducing the frequency of acid exposure, not holding or swishing drinks in the mouth, finishing meal with something rich in Ca/P such as cheese to neutralize acidics, chewing gum after acid intake to stimulate saliva flow, avoiding toothbrushing immediately after acid intake, applying fluoride before the erosive challenge, and using high concentrated topical fluoride periodically.\(^7\)

All of the causative factors must be taken into account in order to provide adequate preventive instructions.\(^50\) When hyposalivation and reduced buffering capacity are identified to be the causes of erosion, recovery of the normal salivary flow should be attempted by mechanical and chemical stimulation, and/or by prescribing oral sialogogues, if applicable.\(^51\)Saliva substitutes may also be considered. Exposure to saliva has been proven to be effective in rehardening eroded enamel;\(^52\) this can be explained by the deposition of salivary calcium and phosphate on softened enamel, after neutralization and clearance of the erosive agent.\(^53\) Anti-erosive agents have also been considered. Although many have been proposed and studied,\(^55\) only few have been
investigated under low salivary flow conditions. There is evidently a need to investigate the efficacy of anti-erosion agents in hyposalivatory conditions.

Restorative Therapy of Dental Erosion

Management of advanced erosive lesions may require restorative therapy. This approach considers several factors, including: the structural integrity of the tooth, hypersensitivity of the exposed dentin, esthetic concerns and increased risk of pulp exposure. The restorative intervention depends on the complexity and extension of the erosive lesions, ranging from direct conservative restorations to indirect fixed or removable prosthesis or even full mouth reconstruction. With the recent improvement in adhesive dentistry, rehabilitation of erosive tooth loss can be achieved in a conservative manner. Resin composite, glass ionomer cement (GIC), resin-modified glass ionomer (RMGI) are restorative material choices.

Resin Composite

Resin composite exhibits successful long-term clinical performance in small to moderate-sized restorations evaluated for more than 10 years. Several studies have investigated the clinical performance of resin composite restorative materials when being subjected to persistent erosive insults, which may adversely affect their physical properties and influence their durability. It has been shown that exposure of resin composite to acidic products such as acidulated fluoride may cause filler loss of resin composite restoration. Only few studies have examined the effect of dietary erosive
agents on the performance of resin composites. They have concluded that resin composite had the best resistance to acid degradation compared to GIC or RMGI.\textsuperscript{14-16,74,75} Unlike glass ionomer materials, resin composite does not provide protection for the surrounding enamel and dentin substrates, due to the lack of fluoride release.\textsuperscript{14,20,76}

Glass Ionomer Cement (GIC) and Resin-Modified Glass Ionomer (RMGI)

In 1960s the first GIC was introduced by Wilson and Kent at a government laboratory in London.\textsuperscript{77} It was produced by mixing a fluoroaluminosilicate glass powder with a polyacid dissolved in water to form hard cement.\textsuperscript{78} GIC presents several characteristics that make it a favorable option: biocompatibility, adhesion to calcified substrates, elastic modulus similar to the dentin and fluoride release. On the other hand, it possesses characteristics that make its use infrequent: technical difficulties related to the material’s stickiness, poor esthetics and solubility, particularly in acidic oral environments.\textsuperscript{66} Due to the shortcomings in the performance of GIC in high stress bearing areas, high viscosity GIC have been introduced to enhance the wear resistance by reducing filler size and increasing the powder-to-liquid mixing ratio.\textsuperscript{79}

Advancement in dental materials has led to production of RMGIs in the 1980s in an attempt to improve the mechanical properties of GIC. Like GIC, RMGI are produced by mixing a fluoroaluminosilicate glass powder with a polyacid dissolved in water except that the liquid phase contain hydrophilic monomers and polymers like HEMA, which make the material polymerizable. This modification in the material’s composition make
the material less susceptible to acidic degradation compared to the former GIC.\textsuperscript{16,80} In a long-term clinical study, the authors had concluded that the clinical performance of RMGI restorations was superior to that of the composite restorations placed in non-curious cervical lesions after seven years of service.\textsuperscript{81} In this study, the failure criteria were the partial or total loss of the restoration; however, the surface loss and roughness of the restorative materials were not assessed.

Fluoride-Releasing Restorative Materials
Protection against Erosion

Fluoride release of GIC and RMGI has exhibited protective effect on the surrounding enamel and dentin.\textsuperscript{14,18,59,75} It has been reported that RMGI can release fluoride at a rate comparable to that by GIC. The daily fluoride release is 8 ppm to 15 ppm on the first day of material’s application, and it decreases to 1 ppm to 2 ppm on the seventh day.\textsuperscript{82} According to Wan Bakar and McIntyre,\textsuperscript{75} GIC materials were found to be vulnerable to severe damage, while they provided protection in the surrounding mineralized tooth structure in patients experiencing strong citric- or gastric-acid induced erosion. Domiciano et al.\textsuperscript{18} tested the effect of GIC and composite resin restorations on root dentin after an erosive challenge \textit{in situ}, using microhardness testing. The authors found that dentin adjacent to the GIC showed significantly higher microhardness values than the other tested materials, concluding that GIC can reduce the progression of root dentin erosion at restoration margins. Furthermore, Soares et al.\textsuperscript{14} evaluated the erosive effect on restorative materials and on surrounding dentin using micro energy-dispersive
X-ray fluorescence spectrometry and scanning electron microscopy. GIC surrounded dentin was found to be less susceptible to acid erosion than dentin adjacent to other restorative materials due to fluoride release. Similarly, Zhou et al.\textsuperscript{59} in 2012 have studied the remineralization effects of different dental materials including GIC-based dental materials using quantitative light-induced fluorescence, microhardness, surface 3D topography and scanning electron microscopy. The authors reported that remineralization of the artificial enamel lesions was better for GIC-based materials (Fuji III LC light-cured glass ionomer pit and fissure sealant and BC glass polyalkenoate base cement) in comparison to NaF-based dental materials (Clinpro XT varnish, F-varnish) and CPP-ACP-based material (Tooth Mousse), which was explained by their potential for more controlled and sustained release of remineralization agents. On the other hand, few studies have found no protective effect of fluoride releasing materials on the surrounding enamel and dentin against erosion.\textsuperscript{19-21}

Fluoride-Releasing Restoration and Hyposalivation

Hu et al.\textsuperscript{17} compared the performance of two high viscosity GIC applied in patients after radiation therapy. The authors concluded that after two years, the restorations were able to prevent secondary caries and no restoration had failed from surface erosion. Another study targeting the xerostomic head and neck cancer patients,\textsuperscript{24} reported that there was a rapid and severe dissolution of GIC restorations subjected to mild acidic agent. McComb et al.\textsuperscript{23} studied three types of restorative materials (resin composite, GIC and RMGI) in post-radiation xerostomic patients, associated or not with
use of neutral pH sodium fluoride gel in custom trays. They found that among fluoride gel users, there were no differences in the performance of the restorative materials, while in the fluoride non-user group, the fluoride releasing materials (GIC and RMGI) provided greater than 80-percent protection against recurrent caries compared to resin composite. Moreover, RMGI had less failure related to the marginal adaptation and/or anatomical form than that of GIC after 12 months while the composite exhibit no changes on the marginal adaptation and/or anatomical form. Similar findings were reported in a study by De Moor et al.\textsuperscript{83} in 2011, the authors evaluated the clinical performance of GIC, RMGI and resin composite restorative materials in class V cavities in xerostomic head- and neck-irradiated cancer patients over two years, in terms of marginal adaptation, anatomical form and recurrent caries and the patients were instructed to use neutral 1.0-percent sodium fluoride gel in custom trays daily. The results showed that among the low fluoride compliance participants, GIC was associated with better protection against recurrent caries compared with the RMGI and resin composite but exhibited a compromised marginal adaptation.

Similarly, Haveman et al.\textsuperscript{22} have studied GIC, RMGI and amalgam restorations in xerostomic patients over two years in terms of marginal adaptation, anatomical form and caries in adjacent tooth structure in relation to the daily use a neutral topical sodium fluoride gel. The study results showed no significant differences between materials in regard to caries at the cavosurface margin in high fluoride compliance group. Whereas among fluoride non-users, the results revealed that teeth restored with amalgam
restorations had a significantly higher incidence of caries at the cavosurface margins than teeth restored with either GIC or RMGI restorations and thus, the authors concluded that the fluoride releasing materials may reduce the incidence of caries around the restorations in high-risk patients who do not routinely use topical fluoride. However, the authors found no statistically significant difference between restorations with regard to marginal integrity or anatomical form.

Therefore, there is evidence that fluoride release and improved mechanical properties are determinant factors for restoration longevity under hyposalivatory conditions. Nonetheless, there is not enough information regarding the recommendation of restorative materials for hyposalivatory patients suffering from dental erosion.

EROSION EXPERIMENTAL MODEL

In-vivo studies are ranked at the top in the hierarchy level of evidence. However, they present some limitations such as lack of a sensitive and reproducible quantitative method for erosive tooth wear measurement, high cost and long duration of the study.84 For those reasons, in-situ and in-vitro models can be adopted to overcome some of the limitations of the in-vivo studies, by simulating the oral environment under well-controlled conditions in the oral cavity.84 Data from laboratory studies along with clinical findings are necessary to guide clinicians in their clinical judgment and practice.

In-vitro models can be performed over a shorter period of time, require fewer staff, do not involve participant compliance issues, are relatively inexpensive and correlate well with in-situ results. However, it should be kept in mind that they cannot
simulate the oral environment with all of the biological variations that influence erosion process.\textsuperscript{84}

Dental Testing Substrate for Erosion Studies

Human or bovine (lower incisors) teeth have been used in dental erosion experiments. Although human enamel and dentin are the substrates of choice, bovine teeth are considered appropriate for most of \textit{in-situ} and \textit{in-vitro} studies as they are easier to obtain, and behave similarly to the human dental substrate.\textsuperscript{85,86}

Artificial substrates, such as hydroxyapatite discs, have been used and are considered suitable for exploratory \textit{in-vitro} studies.\textsuperscript{87}

To prepare the study specimens, samples are embedded in materials such as epoxy resin to ensure retention, then flattened and polished for measurement. Specimens can be masked with tape or nail varnish to expose the test area. The samples should be kept in moist conditions between cycles and during overnight storage.\textsuperscript{87}

Erosive Agent

\textit{In-vitro} studies have reported the erosive potential of different types of dietary acids e.g: citric acid, soft drinks and sodas, sports beverages,\textsuperscript{32} acidic candies,\textsuperscript{88} and fruit juices.\textsuperscript{89,90} The selection of an erosive agent for a study depends on its objectives. For modeling of extrinsic agents, there are important parameters that should be considered including acid concentration (with respect to buffer capacity and concentration of undissociated acid), degree of saturation, calcium and phosphate concentrations,
inhibitors of erosion and pH. The duration of acid contact is also relevant and usually set at 2 min/cycle.

Remineralization Solution

The investigation of remineralization in vitro has been performed using manual or computer assisted pH cycling models. Artificial mouth models have also been used in an attempt to replicate the oral environment, standardizing the contact and flow of erosive and remineralizing agents across the substrate in a controlled manner. In-vitro models usually require large volume of remineralization solution as saliva. Thus, using natural human saliva may become impractical as the collection procedure is time consuming, the saliva samples show high intra- or inter-sample variability, and some of its components can be rapidly degraded. Additionally, the study should comply with cross-infection procedures and regulations concerning the use of human samples. Alternatively, artificial saliva provides the advantage of being easily prepared in large amounts, with a consistent chemical composition.

Simulation of Hyposalivatory Conditions In Vitro

The ethical issues of in-vivo studies and the complexity of the human oral environment have led to development of laboratory models mimicking intra-oral conditions. Different artificial mouth model designs have been successfully reported. Wiegand et al. and Attin et al. used a model consisting of 12-specimen chambers. Each chamber was connected to two multichannel pumps which allow for alternating
rinsing of the samples with different liquids. For rinsing the samples, the liquids were pumped from a reservoir into a channel located between the surface of the enamel specimens and the top of the chambers. Temperature and pumps were controlled by a computer and software. A similar design was reported by Scaramucci et al.\textsuperscript{63} and Borges et al.\textsuperscript{62} to simulate different artificial salivary flow rates. The model involves the use a multichannel peristaltic pump connected to a custom-made acrylic device in which the specimens are loaded and exposed to the test solutions through the chambers. The flow-rate and duration for both the demineralization and remineralization solutions were automatically controlled by the pumps. These \textit{in-vitro} models successfully tested some variables that could not be easily investigated in \textit{in-vivo} conditions.
METHODS AND MATERIALS
STUDY DESIGN

An in-vitro erosion cycling model based on a previous study was used. Two experimental factors were investigated in this study: restorative dental materials at four levels, and simulated salivary flow rate at two levels.

Restorative dental materials used in this experiment were:
1. Resin composite (Filtek Z250, 3M-ESPE, St. Paul, MN).
2. Low viscosity GIC (Fuji II, GC America Inc., Alsip, IL).
3. High viscosity GIC (Fuji IX, GC America Inc., Alsip, IL).
4. Resin-modified glass ionomer (Fuji II LC, GC America, Inc., Alsip, IL).

Simulated salivary flow rates at two levels were:
1. Normal (0.5 ml/min).
2. Low (0.05 ml/min).

Bovine enamel and dentin specimens were prepared (n = 16/group) and submitted to the testing protocols, to be repeated for 5 days. This study was conducted according to the complete block design with 4 repetitions per block. A total of four statistical blocks were performed, completing a total of 16 specimens per group. Surface loss of the restorative material and surrounding enamel and dentin surfaces was the study outcome measure.
Sample Size Calculation

Based on a previous study using a similar dental erosion model, the standard deviation of the enamel surface loss was anticipated to be approximately 4.25 µm. With a sample size of 16 specimens per restorative material-flow rate combination, the study had 80-percent power to detect a 6.0-µm difference in enamel surface loss between any two restorative materials for each flow rate and a 5.3-µm difference between flow rates for each restorative material, assuming two-sided tests for each factor conducted at an overall 5-percent significance level.

Specimen Preparation

Two hundred and thirty enamel and dentin slabs from bovine incisors were cut (4 mm width x 4 length mm x 2 mm thickness) using a microtome (Isomet, Buehler, Lake Bluff, IL) and ground using 1200 grit paper (MDFuga, Struers Inc., Cleveland, Ohio). One enamel and one dentin specimen were positioned 0.5 mm to 0.8 mm apart from each other in the center of one compartment of the rubber mold. Acrylic resin (Varidur, Buehler) mixture was prepared by mixing 6 parts by volume of polymer with 1 part of monomer for 20 seconds using metal spatula. Then, the mixture was poured into the space of the rubber mold until it covered the enamel and dentin slabs completely and was left to set for 10 minutes. Then, the resin blocks were positioned on a cylindrical specimen holder (Struers, Inc.) with the resin surface of the specimens opposing the exposed teeth slabs facing up as shown in Figure 1. They were ground with #500 grit paper on an automated grinding/polishing machine at 300 rpm (Rotoforce-4, Struers Inc.)
under water irrigation (Figure 2). The specimens were removed from the specimen holder and remounted with the surface exposing the dental slabs facing up (Figure 3). Then, they were ground and polished with #1200, #2400 and #4000 grit papers (MDFuga, Struers Inc.) sequentially. Following the polishing procedure, the specimens were placed under running DI water for 3 minutes. The specimens were kept in a relative humidity environment and 128 specimens were chosen based on the quality of enamel and dentin (Figure 4) then randomized into the 8 experimental groups according to the restorative materials used (Table 1) with low or normal salivary flow rate (n = 16/group).

Preparation for Restorative Materials

A box-shaped cavity (1.2 x 4 x 2 mm) was prepared manually in each specimen between the enamel and dentin slabs to receive its corresponding restorative material using a high-speed handpiece with air-water coolant and diamond fissure burs (No. 835KR.31.008, Brasseler; Savannah, GA) (Figure 5 and 6).

Restorative Materials Application

Irradiance of the light curing unit (Demetron Optilux VCL 401, Kerr) was monitored before and during restorative materials application using a curing radiometer (Cure Rite; Dentsply, USA) to ensure an acceptable irradiance output (minimum irradiance of 400 mW/cm²). Then, the cavities were filled according to manufactures instructions as follows:
Filtek Z250 Group

Enamel and dentin surfaces were etched with 37-percent phosphoric acid gel (Scotchbond Etchant, 3M ESPE Dental Products, St Paul, MN) for 15 seconds, followed by rinsing and blotting the excess water. After that, two consecutive coats of Adper Single Bond Plus adhesive (3M ESPE, St. Paul, MN) were applied to enamel, and dentin surfaces followed by gentle air drying for 2 seconds then light cured for 10 seconds. Resin composite material (Filtek Z250) was applied as one increment, and then covered with polyester strip. A glass slab was placed over the top of polyester strip and held under hand pressure to remove excess material. Then, the material was light-cured through the polyester strip and glass slab for 20 seconds.

Fuji II, Fuji IX and Fuji II LC Groups

A Cavity conditioner (GC America, Alsip, IL) was applied to the bonding surfaces using a micro-tip applicator for 10 seconds followed by rinsing and drying the excess water leaving surfaces moist. Before the restorative material’s capsule activation, the capsule side was tapped on a hard surface to loosen the powder then the capsule plunger was pushed against the tabletop until it flushed with the main body. Then, the capsule was activated by clicking it once in a metal GC capsule applier.

Following capsule activation, the capsule was immediately placed into a high speed amalgamator (Vari-Mix III VM-D, Caulk Dentsply) and mixed for at high speed (4200 RPM) for 10 seconds. After that, the mixed capsule was loaded into the GC Capsule Applier and the mixture was extruded directly into the preparation as one
increment then covered with polyester strip. A glass slab was placed over the top of polyester strip and held under hand pressure to remove excess material. After six minutes from start of mixing, GC Fuji COAT LC (GC America, Alsip, IL) was immediately applied to the surface Fuji II and Fuji IX restorations and light cured for 10 seconds. For Fuji II LC group, the material was light cured through the polyester strip and glass slab for 20 seconds, followed by application of GC Fuji COAT LC and light cured for 10 seconds.

The specimens were kept in a humid environment at 37°C for one week before testing to allow post-irradiation hardening of composite restorations and stabilization of the setting reaction of GIC restorations. Then, they were individually finished and polished using the same grinding and polishing procedure described in the specimen preparation section (Figure 7 and 8). Each group was treated independently to avoid contamination. Following polishing procedures, the resin blocks were removed from cylindrical specimen holders, cleaned from the attached sticky wax and labeled (n = 16 per group) (Figure 9).

Surface Area Delimitation

Unplastized Polyvinyl Chloride tape (UPVC, 1.5x8 mm) was placed on two sides of the specimens, leaving an area of 1x8 mm exposed in the center of each of the enamel, dentin and restorative material surfaces (Figure 10 and Figure 11).
Solutions Preparation

A solution of 0.3-percent citric acid anhydrous (Sigma C1857) in DI water (natural pH approx. 2.6) was prepared and used as an erosive agent. The artificial saliva formulation shown in Table II (pH adjusted to 7.0 with HCl) was prepared and used as a remineralization medium (Figure 12).

Erosion and Remineralization Cycling

The daily treatment regimen was comprised of four demineralization challenges, each followed by a remineralization treatment. After the last cycle each day, specimens were kept in a closed container with a humid environment at 37°C, until the next test day. The cycle was continued for 5 days. The daily treatment schedule is summarized in Table III.

Demineralization

A multichannel peristaltic pump (Masterflex LS, Cole Palmer, IL) was used (Figure 13). Each specimen was placed in the assigned closed acrylic chamber of a custom made acrylic device (4 specimens/device) (Figure 14) with the polished surface of the tested substrates facing a closed chamber (approx. 7.1 mm in diameter and 1 mm in height) (Figure 15), which was connected to the pump through plastic tubes (Figure 16). Before starting the erosive challenge each day, the inlet tubes and acrylic chambers were filled with DI water to eliminate air bubbles then the valves of the inlet tubes were closed immediately to avoid the entrance of air into the chamber. Tubes of the demineralization
pump were then connected to the acrylic device and citric acid solution was flushed to
eliminate air entrapments through the free-end tube then the inlet tube valve was opened
and the citric acid solution was run into the tubes at 0.6 ml/min flow rate until it reaches
the specimens inside the chambers. The specimens were exposed to the demineralizing
solutions for 2 min at room temperature. For the whole study, a total of 6 L of citric acid
were used.

Remineralization

After erosive challenge, the inlet tube was closed and the acrylic device was
connected to the remineralization pump through plastic tubes. Artificial saliva solution
was flushed to eliminate air entrapments through the free-end tube then the inlet tube
valve was opened and the artificial saliva solution was allowed to run into plastic tubes
until it reached the specimens at a flow rate of either 0.5 ml/min, simulating a normal
flow rate, or 0.05 ml/min, simulating a low flow rate for 60 minutes (Figure 17). Saliva
was used at room temperature. A volume of 60 ml and 600 ml of artificial saliva were
used per specimen for the low and normal salivary flow rates groups, respectively. For
the whole study, a total of 42,240 ml was used.

Assessment of Erosive Lesions

After completing 5 days of cycling, the UPVC tapes were removed and three
surface areas (restoration: 2 mm long (X) x 1 mm wide (Y); enamel and dentin: 2 mm
long (X) x 3 mm wide) were scanned with an optical profilometer (Proscan 2000,
Scantron, Venture Way, Tauton, UK) (Figure 18). The length of each scan covered both the treated area and reference surfaces. The step size was set at 0.01 mm and the number of steps at 200 in the (X) axis; and at 0.1 mm and 10, respectively, in the (Y) axis.

Surface loss measurements were performed with the use of dedicated software (Proscan Application software v. 2.0.17). For the calculation of the restoration surface loss, the height of the treated area was subtracted from the average height of the two reference areas. A 3-point height tool was applied with auto and manual leveling. For enamel and dentin, similar approach was used; however, each substrate was divided into 3 sub-areas based on the proximity to the restoration: Enamel/Dentin 1 (right next to the restoration), Enamel/Dentin 2 (1 mm apart), Enamel/Dentin 3 (2 mm apart) (Figure 19). The measured surface loss for enamel, dentin and restoration are illustrated in Table VII to Table IX, respectively.

Statistical Analysis

Using mixed-model ANOVAs evaluated surface loss in terms of the effects of restorative material, salivary flow rate, and their interaction; the ANOVAs also included a random effect to account for the block design. A Sidak adjustment was used to control the overall significance level for the pair-wise comparisons within each factor. Separate analyses were performed for the measurements of the restorative material, enamel, and dentin. Data were tested for normal distribution and homocesdasticity. The statistical analysis was accomplished using Statistical Analysis System (2002-2010 SAS Institute Inc., Cary, NC). The significance level was set at 5 percent.
RESULTS
ENAMEL

Prior to analysis, a square root transformation was used to normalize the surface loss data. Enamel 2 and Enamel 3 areas were excluded from the analysis due to lack of significant results. Thus, only Enamel 1 area was used for the analysis, as that location was identified as the most relevant for this study.

The analysis revealed significant differences between salivary flows (p-value < 0.001) and significant differences between restorative materials (p-value < 0.001). The interaction of flow and material is not significant (p-value > 0.05); (Table IV).

Comparing the salivary flow rates, surface loss is lower in flow 0.5ml/min than for flow 0.05 ml/min (p-value < 0.001). Surface loss is significantly higher for enamel adjacent to Filtek Z250 than for enamel adjacent to Fuji II LC (p-value < 0.05) and Fuji IX (p-value < 0.001). Also, enamel surface loss was significantly higher when it was adjacent to Fuji II than the one adjacent to Fuji IX (p-value < 0.05); (Table IV).

DENTIN

Prior to analysis, a log transformation was used to normalize the surface loss data. Dentin 2 and Dentin 3 areas were excluded from the analysis due to lack of significant results. Thus, only Dentin 1 area was used for the analysis, as that location was identified as the source of the most relevant results.
The analysis revealed significant differences between salivary flows (p-value < 0.001), and significant differences between materials (p-value < 0.05). The interaction of flow and material is not significant (p-value > 0.05); (Table V). As with enamel surface loss, dentin surface loss is significantly lower in Flow 0.5 ml/min than for Flow 0.05 ml/min (p-value < 0.001) and the surface loss is significantly higher for dentin adjacent to Filtek Z250 than for dentin adjacent to Fuji II LC (p-value < 0.05) and Fuji IX (p-value < 0.05).

RESTORATIONS

Prior to analysis, a constant was added to the surface loss data to remove negative values. Then, a log transformation was used to normalize the surface loss data.

The analysis revealed significant differences between salivary flows (p-value < 0.001), and significant differences between materials (p-value < 0.001). The interaction of flow and material is also significant (p-value < 0.001); (Table VI). Comparing the salivary flow rates, surface loss of Fuji II, Fuji II LC, and Fuji IX was lower in salivary flow 0.5 ml/min than for salivary flow 0.05 ml/min (p-values < 0.001, < 0.05 and < 0.001, respectively), but there was no difference in Filtek Z250 surface loss in both salivary flow rates (p-value > 0.05); (Table VI). Regarding the restorative materials’ surface loss, the analysis revealed surface loss was significantly lower for Fuji II LC than for Fuji II (p-value < 0.001) and Fuji IX (p-value < 0.001) for 0.5 ml/min and 0.05 ml/min flow rates and the surface loss was the lowest among Filtek Z250 for both flow rates (Table VI).
TABLES AND FIGURES
**TABLE I**

Study groups according to restorative dental materials*

<table>
<thead>
<tr>
<th>Material Group</th>
<th>Manufacturer</th>
<th>Classification</th>
<th>Shade</th>
<th>Lot No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Filtek Z250</td>
<td>3M-ESPE, St. Paul, MN, USA</td>
<td>Microhybrid Resin composite</td>
<td>A2</td>
<td>N546786</td>
</tr>
<tr>
<td>Fuji II</td>
<td>GC Corporation, 76-1Hasunuma-Cho, Itabashi-Ku, Tokyo, Japan</td>
<td>Low viscosity GIC</td>
<td>A2</td>
<td>1305011</td>
</tr>
<tr>
<td>Fuji IX</td>
<td>GC Corporation, 76-1Hasunuma-Cho, Itabashi-Ku, Tokyo, Japan</td>
<td>High viscosity GIC</td>
<td>A2</td>
<td>1311051</td>
</tr>
<tr>
<td>Fuji II LC</td>
<td>GC Corporation, 76-1Hasunuma-Cho, Itabashi-Ku, Tokyo, Japan</td>
<td>RMGI</td>
<td>A2</td>
<td>1311081</td>
</tr>
</tbody>
</table>

*GIC, glass ionomer cement; RMGI, resin-modified glass ionomer cement.
### TABLE II
Artificial saliva formulation

<table>
<thead>
<tr>
<th>Chemicals</th>
<th>Quantity (g/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CaCl$_2$$\cdot$2H$_2$O</td>
<td>0.213</td>
</tr>
<tr>
<td>KH$_2$PO$_4$</td>
<td>0.738</td>
</tr>
<tr>
<td>KCl</td>
<td>1.114</td>
</tr>
<tr>
<td>NaCl</td>
<td>0.381</td>
</tr>
<tr>
<td>Tris buffer</td>
<td>12</td>
</tr>
</tbody>
</table>
TABLE III

Daily treatment schedule

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Treatments*</th>
</tr>
</thead>
</table>
| Challenge 1 | CA: 2 min  
Remin: 60 min |
| Challenge 2 | CA: 2 min  
Remin: 60 min |
| Challenge 3 | CA: 2 min  
Remin: 60 min |
| Challenge 4 | CA: 2 min  
Remin: 60 min  
Overnight: humid environment, 4°C |

*CA, citric acid; remin; saliva remineralization.
### TABLE IV

Surface loss of enamel adjacent to studied materials in microns: mean (standard deviation)*

<table>
<thead>
<tr>
<th>Flow</th>
<th>Filtek Z250</th>
<th>Fuji II</th>
<th>Fuji II LC</th>
<th>Fuji IX</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.05</td>
<td>19.75 (4.27)</td>
<td>17.06 (2.61)</td>
<td>16.33 (2.30)</td>
<td>15.79 (2.41)</td>
</tr>
<tr>
<td>0.50</td>
<td>11.57 (2.51)</td>
<td>11.40 (1.98)</td>
<td>10.70 (2.39)</td>
<td>9.12 (1.66)</td>
</tr>
</tbody>
</table>

*Different capital letters in rows and lower case letters in columns represent statistically different means (p < 0.05).

### TABLE V

Surface loss of dentin adjacent to studied materials in microns: mean (standard deviation)*

<table>
<thead>
<tr>
<th>Flow</th>
<th>Filtek Z250</th>
<th>Fuji II</th>
<th>Fuji II LC</th>
<th>Fuji IX</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.05</td>
<td>23.08 (3.48)</td>
<td>22.30 (2.70)</td>
<td>20.47 (2.58)</td>
<td>20.63 (2.34)</td>
</tr>
<tr>
<td>0.50</td>
<td>14.83 (2.05)</td>
<td>14.00 (2.48)</td>
<td>13.90 (1.68)</td>
<td>13.76 (1.89)</td>
</tr>
</tbody>
</table>

*Different capital letters in rows and lower case letters in columns represent statistically different means (p < 0.05).

### TABLE VI

Surface loss of studied materials in microns: mean (standard deviation)*

<table>
<thead>
<tr>
<th>Flow</th>
<th>Filtek Z250</th>
<th>Fuji II LC</th>
<th>Fuji II</th>
<th>Fuji IX</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.05</td>
<td>-0.35* (0.19) A,a</td>
<td>2.17 (0.73) A,b</td>
<td>13.03 (6.79) A,c</td>
<td>16.74 (7.72) A,c</td>
</tr>
<tr>
<td>0.50</td>
<td>-0.37* (0.23) A,a</td>
<td>0.90 (0.34) B,b</td>
<td>6.40 (3.46) B,c</td>
<td>7.30 (2.94) B,c</td>
</tr>
</tbody>
</table>

*Means followed by different capital letter in columns and lower case letters in rows are statistically different (p < 0.05). Negative values represent that the experimental area is above the reference area, possibly because of the convexity of specimens (Steiner-Oliveira et al., 2010).
TABLE VII
Examples of profilometric analysis images of enamel surface loss

<table>
<thead>
<tr>
<th>Enamel adjacent to/ salivary flow rate</th>
<th>0.05 ml/min</th>
<th>0.5 ml/min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Filtek Z250</td>
<td>![Image]</td>
<td>![Image]</td>
</tr>
<tr>
<td>Fuji II LC</td>
<td>![Image]</td>
<td>![Image]</td>
</tr>
<tr>
<td>Fuji IX</td>
<td>![Image]</td>
<td>![Image]</td>
</tr>
<tr>
<td>Fuji II</td>
<td>![Image]</td>
<td>![Image]</td>
</tr>
</tbody>
</table>
### TABLE VIII
Examples of profilometric analysis images of dentin surface loss adjacent to different restorative materials

<table>
<thead>
<tr>
<th>Dentin adjacent to/ salivary flow rate</th>
<th>0.05 ml/min</th>
<th>0.5 ml/min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Filtek Z250</td>
<td><img src="image1.png" alt="Image" /></td>
<td><img src="image2.png" alt="Image" /></td>
</tr>
<tr>
<td>Fuji II LC</td>
<td><img src="image3.png" alt="Image" /></td>
<td><img src="image4.png" alt="Image" /></td>
</tr>
<tr>
<td>Fuji IX</td>
<td><img src="image5.png" alt="Image" /></td>
<td><img src="image6.png" alt="Image" /></td>
</tr>
<tr>
<td>Fuji II</td>
<td><img src="image7.png" alt="Image" /></td>
<td><img src="image8.png" alt="Image" /></td>
</tr>
</tbody>
</table>
### TABLE IX
Profilometric analysis images of restorative materials’ surface loss

<table>
<thead>
<tr>
<th>Material/salivary flow rate</th>
<th>0.05 ml/min</th>
<th>0.5 ml/min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Filtek Z250</td>
<td><img src="filtek_z250_0.05.png" alt="Image" /></td>
<td><img src="filtek_z250_0.5.png" alt="Image" /></td>
</tr>
<tr>
<td>Fuji II LC</td>
<td><img src="fuii_lc_0.05.png" alt="Image" /></td>
<td><img src="fuii_lc_0.5.png" alt="Image" /></td>
</tr>
<tr>
<td>Fuji IX</td>
<td><img src="fuii_0.05.png" alt="Image" /></td>
<td><img src="fuii_0.5.png" alt="Image" /></td>
</tr>
<tr>
<td>Fuji II</td>
<td><img src="fuii_0.05.png" alt="Image" /></td>
<td><img src="fuii_0.5.png" alt="Image" /></td>
</tr>
</tbody>
</table>
FIGURE 1. Three resin block mounted on a cylindrical specimen holder by sticky wax with the resin surface facing up.
FIGURE 2. Struers Rotoforce-4.

FIGURE 3. Resin blocks mounted on cylindrical specimen holders with enamel and dentin specimens facing up.
FIGURE 5. Mounted resin blocks with prepared cavities between enamel and dentin slabs.

FIGURE 6. Enlarged picture of the resin block showing the dimensions of the cavity preparation.
FIGURE 7. Restorative dental materials applied in the mounted resin blocks.

FIGURE 8. Resin block with polished enamel, dentin and restoration substrates.
FIGURE 9. Labeled specimens.

FIGURE 10. Specimens' reference surfaces covered with UPVC tape.
FIGURE 11. Specimen block showing the dimensions of the testing surface after surface area delimitation with upvc tape.
FIGURE 12. Artificial saliva solution with final pH measured with pH meter.

FIGURE 14. Custom-made acrylic device with four specimens.
FIGURE 15. Specimens’ surfaces facing the closed chambers of the acrylic device.
FIGURE 16. Acrylic devices connected to the multichannel peristaltic pump by plastic tubes.
FIGURE 17. Remineralization of the specimens by artificial saliva solution at low salivary flow rate (0.05 ml/min) for 60 minutes.

FIGURE 19. Locations of profilometric scans on enamel, dentin and restoration surfaces.
DISCUSSION
EXPERIMENTAL MODEL CONSIDERATIONS

The in-vitro test used in this study was based on an artificial mouth model,\textsuperscript{93,94} which can properly isolate and control most of the study variables. It allows for very sensitive and reproducible measurements, while simulating some of the relevant conditions present in the oral cavity. This model showed to be appropriate for testing the performance of restorative materials under different salivary flow rates, which would not be possible with experimental set ups previously published in this research area.\textsuperscript{14,76,97-99}

In addition to the test of different salivary flow rates, our experimental model simulated erosive challenges (acid exposure) and allowed for testing different substrates simultaneously. Artificial saliva was used, instead of natural saliva, due to the large volume needed and to maintain a standard mineral composition of the solution throughout the study phases. The artificial saliva was pumped at two different flow rates, 0.5 and 0.05 ml/min, for one hour in each cycle to represent normal and low salivary flow conditions,\textsuperscript{40,42} respectively. A solution of 0.3-percent citric acid anhydrous (Sigma C1857) in deionized water (pH approx. 2.6) was chosen to mimic a dietary erosive attack in the oral cavity.\textsuperscript{63} It was injected in the flow cell at 0.6 ml/min for two minutes. A statistical complete randomized block design was adopted to allow for a better control of the testing procedures, minimizing the chance for errors during the experiment.
The effects of erosive challenges on dental hard tissues and restorative dental materials have been assessed using various laboratory assessment techniques including surface profilometry, microradiography, chemical analysis for mineral ion concentrations, surface hardness, and scanning electron microscopy (SEM).\textsuperscript{100-102} Profilometry has been reported as the most commonly applied quantitative method to determine both dentin and enamel loss in \textit{in-vitro} and \textit{in-situ} studies,\textsuperscript{100} and was therefore used in this study.

**ENAMEL**

The enamel surface loss was influenced by the type of adjacent restoration and salivary flow condition. As shown in Table IV, surface loss of enamel was significantly higher (p-value < 0.05) in low (0.05 ml/min) compared to normal (0.5 ml/min) salivary flow. This can be explained by insufficient clearing and buffering of the acid on enamel surface at low salivary flow.\textsuperscript{103} This is supported by previous studies showing that reduced saliva output decreases its acid clearing and neutralizing capacities, contributing to the development of erosive lesions.\textsuperscript{104-106}

Our hypothesis stated that erosion protection would be observed around fluoride-containing restorations, more so under highly erosive conditions. This was partially confirmed, as our results showed that enamel surface loss was less evident among fluoride-containing restoration groups; with Filtek Z250 exhibiting the highest mean values in low compared to normal flow rates (19.75\textmu m and 11.57 \textmu m, respectively) followed by Fuji II (17.06 \textmu m and 11.40 \textmu m, respectively), Fuji II LC (16.33 \textmu m and
10.70 µm, respectively) and Fuji IX (15.79 µm and 9.12 µm, respectively). This is in agreement with previously reported clinical data, where the protective effect of the fluoride releasing materials was evident under low salivary flow conditions.22-24,83

There is compelling evidence suggesting that the benefits of topical fluoride are more profound in situations of extremely high risk for dental demineralization, as observed with rampant caries development in patients suffering from hyposalivation.107 These circumstances allow for the prolonged fluoride retention in the mouth, due to the absence of salivary clearance and dilution action.108 However, the results of this study did not show greater impact of fluoride against erosion under low salivary flow conditions. Perhaps, this result may be attributed to the more aggressive conditions simulated in the current study, as imposed by the erosive acid compared to the weak cariogenic acids. Also, fluoride retention at the surface may have been reduced by the lack of salivary pellicle and dental plaque, in the experimental model used.

DENTIN

Eroded dentin exhibited relatively more surface loss compared to enamel. This may be explained by the differences in their mineral and organic contents and relatively higher critical pH for demineralization.34,100

As with the enamel findings, a similar trend was observed in the mean surface loss of dentin adjacent to each restorative material under both low and normal salivary flow conditions. The highest dentin surface loss was observed adjacent to Filtek Z250 at low salivary flow, when compared to the normal salivary flow (23.08 µm and 14.83 µm,
respectively), followed by Fuji II (22.30 μm and 14.00 μm, respectively), Fuji II LC (20.47μm and 13.90 μm, respectively) and Fuji IX (20.63 μm and 13.76 μm, respectively). Some studies have shown that the fluoride release of GICs is enhanced under acidic conditions,\textsuperscript{109-111} which leads to the hypothesis that the increased level of leached fluoride may enhance the anti-erosive effect on the surrounding tooth. In this study, higher material surface degradation was observed for the GICs; however, no additional protection due to the increased fluoride availability from material degradation was observed on adjacent dentin surfaces. In fact, the low viscosity GIC did not even offered significant protection compared to the negative control group (Filtek Z250). This contradictory result may be explained by the severity of the erosive challenge, reducing the protective action of fluoride.

RESTORATIVE MATERIALS

Biodegradation of restorative dental materials under function is a complex mechanism and may be attributed to various factors in the oral environment.\textsuperscript{112} Chemical degradation is one relevant aspect, which has been studied \textit{in vivo} for direct tooth-colored restorative dental materials,\textsuperscript{22-24,83} \textit{in situ} \textsuperscript{20,113} and \textit{in vitro}.\textsuperscript{11,13,75,76} However, there is limited knowledge about the influence of dietary erosive acids on the surface of restorative dental materials under reduced salivary flow conditions such as in xerostomic head and neck cancer patients.

In this study, the degree of restorative materials’ degradation by erosive acid was affected by the materials’ type and the salivary flow condition. The greatest surface
degradation was found for the high and low viscosity GIC restorations (Fuji II or Fuji IX) under normal and low salivary flow conditions. Significantly, less prominent erosive effect was observed for the RMGI under both salivary flow rates. The pronounced erosion of the GICs is explained by the dissolution silicate-glass hydrogel network peripheral to the glass particles.\textsuperscript{23,114} Wan Bakar and McIntyre\textsuperscript{75} have examined the depth of bulk loss of tooth-colored restorative materials by gastric acids, lemon juice and cola drinks in vitro, by measuring restorations’ marginal and body degradation. The results showed that degradation of GIC was usually greater at the margins than in the body. Furthermore, McComb and others\textsuperscript{23} studied three types of restorative materials (Resin composite, GICs and RMGI) in post-radiation xerostomic patients. They found that regardless of the use of neutral pH sodium fluoride gel, the failure related to the marginal adaptation and/or anatomical form after 24 months was greatest among GIC group (89 percent) compared to the RMGI (67 percent) and resin composite (41 percent) groups. Lower surface alteration among RMGI restorations may be attributed to the presence of reinforcing resin within the matrix that made the material less susceptible to acidic degradation compared to the GICs.\textsuperscript{16,80}

Unlike the other materials, resin composite (Filtek Z250) presented no measurable surface degradation under both the normal and low salivary flow rates as shown in table 6, showing its ability to withstand the acidic challenge at high erosive risk conditions. This is consistent with \textit{in-vitro} and \textit{in-vivo} studies.\textsuperscript{14-16,23,75}
In light of the results of this *in-vitro* study, the model used was able to reproduce the highly erosive conditions observed in the hyposalivatory population. The influence of other important factors that have been proven to play major role in dental erosion development such as toothbrushing abrasion, acquired dental pellicle and dental plaque should be thoroughly further assessed in future studies. In addition, adhesion and microleakage of restorations placed eroded dental substrate should be tested to determine the restorative materials with more favorable clinical outcomes.
SUMMARY AND CONCLUSIONS
The objective of this study was to measure the surface loss of four different restorative materials and the surrounding enamel and dentin after erosive challenge under normal and low salivary flow conditions. From the data collected, the results can be summarized as follows:

1. The experimental model used could reproduce the highly erosive conditions expected in patients suffering from hyposalivation.

2. Resin composite restorations did not suffer any measurable surface loss under any of the erosive testing conditions. However, they were incapable of providing adjacent enamel and dentin protection against erosion.

3. The high viscosity GIC was able to provide protection against erosion for the surrounding enamel and dentin but was vulnerable to surface degradation under erosion.

4. The low viscosity GIC was unable to provide enough protection against erosion for the surrounding enamel and dentin and was vulnerable to surface degradation under erosion.

5. Resin modified glass ionomer restorations showed intermediate surface degradation and protection against erosion for the surrounding enamel and dentin.
Based on these findings, patients at higher risk for erosion with low exposure to fluoride and in need for restorations may benefit from the use of fluoride-containing restorative materials that are to resist erosive degradation. RMGI showed the ability to provide protection for the surrounding tooth against erosion, and to resist to surface changes due to the erosive challenge. The potential use of RMGIs as the restorative material of choice for the restorative procedures in patients at higher risk for erosion should be further confirmed in more clinically relevant conditions.
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ABSTRACT
SUSCEPTIBILITY OF RESTORATIONS AND ADJACENT ENAMEL/DENTIN TO EROSION UNDER DIFFERENT SALIVARY FLOW CONDITIONS

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BACKGROUND: Dental erosion is a multifactorial condition that causes irreversible loss of dental hard tissues. Its development is highly influenced by saliva, with higher prevalence in hyposalivatory patients. There is no scientific consensus
regarding the restorative treatment of choice for eroded teeth in such highly erosive conditions and to date, this has not been studied under *in-vitro* conditions. **OBJECTIVES:** To investigate the effect of erosion on direct tooth-colored restorations and adjacent enamel/dentin under different simulated salivary flow rates. **METHODS:** Bovine enamel and dentin specimens were prepared \( n = 16 \) and restored with the testing restorative materials, resin-composite (Filtek Z250), resin-modified glass ionomer (Fuji II LC), high viscosity glass ionomer cement (Fuji IX), and low viscosity glass ionomer cement (Fuji II). Then, submitted to an *in-vitro* erosion cycling model simulating different salivary flow rates (normal 0.5 ml/min and low 0.05 ml/min) and dental erosion protocols for 5 days. Surface loss of the restorative material and surrounding enamel/dentin surfaces were analyzed. A mixed-model ANOVAs and Sidak adjustment were used for statistical comparisons \( p < 0.05 \). **RESULTS:** The surface loss was lower at 0.5 than at 0.05 ml/min, for all tested restorative materials except resin composite. Surface loss was higher in enamel and dentin adjacent to Filtek Z250 compared to Fuji II LC and Fuji IX, with no significant difference in enamel and dentin surface loss adjacent to Filtek Z250 and Fuji II. The restorations surface degradation was significantly lower for Filtek Z250 than for Fuji II, Fuji II LC, and Fuji IX, at both 0.5 and 0.05 ml/min; moreover, the surface loss was significantly lower for Fuji II LC than for Fuji II and Fuji IX, which did not differ from each other. **CONCLUSION:** Within the limitations of this study, it can be concluded that low salivary flow promoted higher erosive conditions. The use of Fuji II LC and Fuji IX may reduce erosive effects on enamel and dentin adjacent to restoration.
Of the materials evaluated, resin-modified glass ionomer restoration may be the most suitable for restoration for patients at higher risk of erosion with low exposure to fluoride.
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