Fluoride Release and Recharge of Different Fissure Sealants with Various Fluoride Preventive Treatments

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Abstract

Objective: Pit and fissure sealants have the ability to release fluoride into the oral environment and reuptake fluoride from the fluoridated products. Therefore, the purpose of this study was to investigate the fluoride release and the rechargibilty potential of different fissure sealants after exposure to various fluoride preventive treatments with different fluoride concentrations.

Materials and Methods: Sixty caries free, extracted teeth were sealed with four types of sealants: Helioseal, Ultraseal XT plus, Fuji II LC, and Fuji Triage. The teeth were stored in test tubes containing 5ml artificial saliva, and the fluoride release was measured daily on the first week, on day 21, and after 28 days using a fluoride specific electrode. On day 29, the fissure sealants were divided into three subgroups, and then subjected to three different fluoride treatments (APF gel, APFgel-Colgate toothpaste, and Prevident toothpaste). The fluoride release was measured daily in the first week, and then weekly for two weeks.

Results: The glass ionomer sealant (Fuji Triage) released significantly more fluoride than the other tested sealants ($p<0.0001$). After the fluoride treatments, all fissure sealants showed high fluoride release, but significantly more with APF gel and APFgel-Colgate tooth paste ($p<0.0001$).

Conclusions: The glass ionomer based sealant (Fuji Triage) released significantly more fluoride than the other tested fissure sealants (resin modified glass ionomer, resin sealants).The use of APF gel and APFgel-regular toothpaste (Colgate) treatments significantly recharged the sealants more than highly fluoridated toothpaste (Prevident). The high fluoride toothpaste (Prevident) provided significantly more fluoride only for glass ionmer sealants (Fuji Triage) and resin modified glass ionomer sealants (Fuji II LC).

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Introduction:

The prevalence of dental caries declined after the introduction of fluoride in dental practice. Fluoride inhibits demineralization and enhances remineralization, thus aiding in the biological prevention of caries and the formation of Fluorhydroxyapatite, which is more resistant to bacterial acid attacks [1].

Different fluoride preventive measures are available to control caries as a disease like systemic and topical fluorides. However, it was found that fluoride treatments are less effective on occlusal surfaces [2]. Therefore, development of pit and fissure sealants opened a new effective measure in caries prevention.

As pit and fissure sealants proved their abilities in caries protection by occluding these highly susceptible areas[3], the incorporation of fluoride into sealants maintained a slow and continuous release of fluoride into oral cavity. Although the amount of fluoride released from most fluoride releasing materials is less than other fluoride preventive treatments like toothpastes, mouth washes, and fluoride gels, fluoride from the material is constantly released so the exposure time of enamel to fluoride is increased, and therefore this maximizes remineralization to the adjacent tooth surface. This was observed by an in vitro study, which showed that resin-modified glass ionomer cements in constant contact with an adjacent incipient carious lesion had the same remineralization ability as twice-daily brushing with a fluoridated dentifrice (1,100 parts per million fluoride) [4] .

Most of fluoride releasing materials can compensate the fluoride that is released from them early by reuptake of fluoride from exogenous sources [5, 6].

Several studies were conducted in order to find an effective fluoride provider that boosts fluoride release from fissure sealants without affecting their properties.

In the oral cavity, the patient is exposed to different fluoride sources such as fluoride gels, fluoridated dentifrices, and mouth washes that have different concentrations of fluoride.

Therefore, the purpose of this study was to investigate the fluoride release and the rechargibilty potential of different fissure sealants after the exposure to various fluoride preventive treatments with different fluoride concentrations.

Literature Review

Dental Caries as a Disease and Its Prevention:

Dental caries is an ecological and multifactorial disease that results from a complex interaction between cariogenic food, bacteria, and tooth structure in such a way as to encourage demineralization of the tooth enamel with resultant caries formation.

The demineralization of enamel results from destructive action of the organic acids (lactic, pyruvic, acetic, propionic, formic, butyric) that are produced after glycolysis of food by the cariogenic bacteria such as *mutans Streptococci* and possibly *lactobacilli,* which are present in plaque [7].

Dental caries start after tooth eruption. Its incidence increases with age and is considered as one of the most common diseases in children. According to The Center for Disease Control and Prevention1(CDC), from 1999 through 2004, 42 percent of children aged 2 to 11 years have dental caries in their primary teeth, and 59 percent of adolescents aged12 to 19 years have dental caries in their permanent teeth[8].

In the United States, 45.3 percent of children and adolescents from 5 to 17years had carious teeth [9]. In adults, 93.8 percent had evidence of past and present coronal caries [10]. However, prevalence of dental caries has declined in industrialized countries over the past three decades. From 1986-87, 50 percent of 5-17 year old children in the U.S. were completely free of decay and of restorations in their permanent teeth [11]. Prevalence of dental caries also varies according to tooth surface; for example, occlusal surfaces of posterior teeth are more susceptible to dental caries than smooth surfaces due to the presence of more pits and fissures.

Demineralization and Remineralization:

Caries formation is a dynamic process that takes place in a microbial bio-film called a pellicle, which plays a role in the ongoing physical-chemical equilibrium with the tooth surface and oral environment [12].

After ingestion of cariogenic food containing sugar, the pH of plaque declines due to acids resulted from fermentation of food by bacteria. This drop in pH leads the calcium and phosphate ions to diffuse from hydroxyapatite crystals in the enamel through the pellicle into the oral cavity. This mechanism is called demineralization. After the neutralization of plaque acids by buffers in saliva, calcium and phosphate ions in the saliva are transmitted through the pellicle into the enamel, and this is known as remineralization. Remineralization occurs between periods of demineralization. Thus, demineralization and remineralization can be considered as a dynamic process characterized by the flow of calcium and phosphate out of and back into tooth enamel. Since this concept has been fully understood, dental researchers, who are interested in Cariology, are investigating ways to slow down or eliminate enamel demineralization and at the same time enhance or initiate enamel remineralization.

Fluoride in Caries Prevention:

Fluorides, dietary habits, oral hygiene, and sealing of pits and fissures have a high significance in prevention and control of dental caries. However, the exposure of dentition to fluoride is considered as the most effective measure in caries prevention, and this could explain the decrease of dental caries observed in the late decades in developed/ industrialized countries [13].

The primary role of fluoride in caries prevention is to inhibit demineralization and enhance remineralization. When the pH is low, the fluoride releases from dental plaque and combines to calcium and phosphate to form a more enamel-resistant structure known as Fluorhydroxyapatite. Fluorhydroxyapatite is less soluble in acidic environments [1].

Fluoride also inhibits the manner in which cariogenic bacteria metabolize carbohydrates by blocking enzymes involved in the bacterial glycolytic pathway and affects the production of adhesive polysaccharides [14].

Fluoridation Methods:

A. Systemic Fluoride:

1. Water Fluoridation:

Water fluoridation is considered as the most widely used public health intervention for the prevention of dental caries. In 1962, the U.S. Public Health Service established that fluoride ranges from 0.7-1.2 ppm constitute the optimal concentration in dental caries prevention[15]. However, in areas where water fluoride is lower than optimal fluoride, additional fluoride supplements are recommended[16]. Several epidemiological studies have found that water fluoridation is effective in reducing dental caries in both children and adults [17-19].

2. Fluoride Supplements:

During the 1940s, another source of fluoride was introduced. Fluoride tablets, or sometimes lozenges, employ sodium fluoride in order to enhance the topical effect of fluoride. Tablets and lozenges are expected to be chewed or sucked for one to two minutes before being swallowed. Supplements for infants are available as a liquid and used with a dropper. However, the American Dental Association (ADA) Council on Scientific Affairs (CSA) recommended that dietary fluoride supplements should be prescribed only if children are at high risk of developing caries or if the optimal fluoride level in drinking water is insufficient [8].

Studies suggest that the systemic effect of fluoride is when it is embedded in calcified structures like enamel in pre-eruptive stage [20]. However, fluoride can serve as a topical preventive when it is available in saliva [21].

B. Topically Applied Fluoride:

Fluoride can be applied topically as fluoridated dentifrices, mouthwashes, varnishes, and fluoride gels. Toothpastes are the most recommended form of self applied fluoride as they are easy to use and inexpensive [22].

In the United States, three sources of fluoride are approved by the Food and Drug Administration to be used in toothpastes: sodium fluoride, stannous fluoride and sodiummonofluorophosphate [23]. However, studies showed that sodium fluoride results in more caries prevention than the others[24].

Most of the dentifrices used in the United States contain 1,100 parts per million (ppm) F, which provides approximately 1 mg of fluoride ion (F) per gram of dentifrice [7]. However, in order to prevent the possibility of systemic ingestion of fluoride from tooth pastes, the American Academy of Pediatric Dentistry recommended that a pea-sized amount of dentifrice should be applied by the child's caregiver to the brush the teeth [25].

Mouth rinses can be used in home use caries prevention. The FDA has approved neutral pH mouth rinses containing 0.02 percent sodium fluoride (90ppm fluoride ion) or 0.05 percent sodium fluoride (226 ppm fluoride ion) and acidulated phosphate mouth rinses containing 0.22 percent sodium fluoride (100 ppm fluoride ion) for OTC use [23]. However, fluoride mouth rinses should be used only for children at moderate or high risk of developing caries.

Although the home use of fluoride mouth rinses and dentifrices proved their ability in caries prevention, they require patient compliance and cooperation. Therefore, for patients with a high caries risk, it is advisable to use professionally applied fluoride, like fluoride varnishes and gels. Fluoride varnishes contain natural resins, which provide the adhesive behavior that extends the contact of fluoride to enamel. In the United States, two forms and concentrations of fluoride varnishes are available: sodium fluoride (2.26% F) or di-fluoride silan (0.1% F) [13]. Previous studies concluded that fluoride varnishes can provide a significant caries-inhibiting effect in both primary and permanent dentitions [26].

Another form of professionally applied fluoride is Fluoride gels like APF(acidulated phosphate fluoride) gel .Fluoride gels are recommended for persons with active decay and at high risk, for those undergoing head and neck radiation therapy, and for older adults experiencing root caries [22]. APF gel contains a high concentration of fluoride 1.23 percent (12,300 ppm) in the form of sodium fluoride and an acidity of pH=3, where, at this pH, the APF gel encourages formation of fluoroapatite crystals. APF gel is recommended to be applied by tray for four minutes every six months [27].

Although topical fluorides can provide caries protection, they are more effective on smooth surfaces than occlusal surfaces [2]. Therefore, mechanically occluding these areas by pits and fissure sealants was developed as a different preventive strategy than systemic or topical fluoride.

Pits and Fissure Sealants/Literature Review:

Sealants were developed in 1960s to protect the occlusal surfaces of the teeth. They have been known as the materials that apply to the pits and fissures of occlusal surfaces of the teeth to protect these highly susceptible areas from caries by mechanically blocking them and thus, prevent the bacteria from reaching food that is present in the oral cavity. Consequently, the processes of fermentation and caries formation will be ceased [3].

Types of Sealants:

The sealants can be classified according to their composition into two types: resin-based sealants and glass ionomers. The resin-based (Bis-GMA resin) sealants were first introduced by Bowen; they are used with acid etching of the enamel surface to get a good retention [28]. The resin-based sealants are further classified into filled and unfilled resin. In the filled type of the resin, a filler material, usually consisting of quartz and silica particles, is added to the resin to increase both bond strength and resistance to abrasion and wear. The amount of filler can range from small percent to up to 75 percent. From a clinical point of view, it is important to know the filler content of the sealant, because sealants with a high percentage of filler need more occlusal adjustments and curing time during light polymerization [29]. Clinicians often notice a slight increase in viscosity with filled resins as compared to unfilled resins. This explains the high marginal adaptation and micro tensile bond strength to the enamel by the unfilled resin sealants [30]. On the other hand, according to the way of polymerization, the resin-based sealants are classified as chemically or self polymerized sealants (in which the free radical reactions take place by tertiary amines) and light polymerized sealants (in which the induction of free radical reaction is accomplished by light curing device). However, clinically, both self and light polymerized sealants have equivalent efficacy when applied to an etched enamel surface [31, 32]. Moreover, resins can be either clear or opaque. Although there is no difference in the clinical effectiveness between them, the opaque resins are easier during the application and evaluation at the follow up visits [33].

The second main type of sealants are glass ionomer cements, which were introduced in 1974 by McLean and Wilson [34]. Due to their fluoride release properties, GICs are effective in caries prevention, even if part of it is lost[35]. Another advantage of this cement is its hydrophilic property, which allows its application in the clinical situation where the control of moisture is difficult, such as with partially erupted teeth. However, the mechanical properties of GIC sealants like wear resistance are lower than the resins; therefore, the introduction of resin modified GICs were introduced as a preferable solution [36, 37]. This material is introduced by adding Hydroxyethylmethacrylate or HEMA to the conventional glass ionomer material to enhance the mechanical properties, and to be esthetically acceptable. Therefore, in view of these differences in the properties between resin-based and glass ionomer sealants, the American Dental Association recommended that resin-based sealants are the first choice of material for dental sealants, where as glass ionomer cement may be used as an interim preventive agent when resin-based sealant is indicated to be placed, but concerns about moisture control may compromise such placement [38].

Clinical Indications of Fissure Sealants:

According to the guidelines of the American Dental Association, fissure sealants are recommended to be used in the prevention of dental caries in children's primary and permanent teeth and adults' permanent teeth when the tooth or the patient is considered to be at a high risk of developing dental caries. Also, fissure sealants are recommended to be placed over early (non-cavitated) carious lesions to arrest caries progression [39]. Other factors that should be considered when deciding to place fissure sealant are the oral hygiene control, fluoride exposure, and potential restrictions of the patient's ability to perform self care [40].

The Effectiveness of Fissure Sealants in Caries Prevention:

Pits and fissures are considered as the most susceptible area due to their anatomical shape which leads to sticking of food and microorganisms and therefore caries formation, but after introduction of fissure sealants to the dental field, occlusal caries were reduced by about 71 percent [41]. However, the long term success of fissure sealants in caries prevention depends on the caries risk of the individual [42], the prevalence of decay in the country [43], the marginal integrity, and the length that the sealant stays intact (mainly because it is found that Caries rates are equal between the teeth with fractured or lost sealant and the teeth without sealant [44]). Therefore, to overcome this problem, fluoridated fissure sealants were introduced to lower the rate of caries around the margins of fractured or worn sealant.

Systematic reviews of randomized controlled trials have proved the ability of fissure sealants in arresting caries in incipient carious lesions in children, adolescents, and young adults after five years of placement of the sealant [45, 46].

Another in vitro study concluded a high ability of fluoridated sealants in inhibition of demineralization on enamel surface [47]. However, in contrast to glass ionomer based sealants, resin based sealants, either fluoridated or non fluoridated, they cannot interfere with enamel mineral loss [48]. Kevien et al and others investigated the ability of fluoride containing composite, fluoride containing glass ionomers, and non fluoride containing resin composites in inhibiting demineralization and enhancing remineralization of incipient carious lesions on the interproximal enamel of teeth adjacent to those restored with the materials. They found that resin modified glass ionomers inhibit demineralization of carious lesions more than the other materials, and this is could be due to its fluoride contents [49].

Fluoride and Fissure Sealants:

Fluoride has been considered as a cariostatic agent as it enhances the formation of fluoroapatite crystals and remineralization of tooth structure. Fluoride also interferes with pellicle and plaque formation and inhibits microbial growth and metabolism [50]. However, some studies state that anticariogenic effects of fluoride that are released from fluoridated restorations are most likely from remineralization [51, 52]. On the other hand, the topical application of fluoride may lead to reduction of fluoride of the enamel [53]. Additionally, treatment of tooth surface with topical fluoride (APF gel) before application of the fissure sealant resulted in reduction of bond strength of the fissure sealant [22]. Therefore, in order to increase the exposure time of fluoride to enamel surface and provide long term caries protection, the fluoride has been incorporated into the restorative materials to maintain a slow and continuous release of fluoride into oral cavity.

The fluoride was incorporated into the sealant by two different techniques. In the first, the fluoride was incorporated into the unpolymerized resin as soluble salt, so after the placement of the fissure sealant into enamel surface, the salt dissolves and fluoride ions are released. The other technique is by chemical bond between an organic fluoride and the resin, so the fluoride will be released by ionic exchange [54, 55]. However, in order to view the effect of these two techniques on strength of the fissure sealants, the National Institute of Dental Research stated that the first technique may results in reduction of strength of the sealant, as the fluoride is released by dissolution of salt. Where as in the second technique, the fluoride was replaced by another ion rather than lost, so this did not lead to a measurable decrease in strength of the sealant [56].

Kadoma et al acknowledged that adding fluoride to fissure sealants gave them higher properties than their predecessors in terms of retention, low-grad, and continuous release of fluoride, and the formation of fluoroapatite crystals in the enamel as a process of remineralization [57].

The first fluoride releasing sealant (fluoroshield, LD Caulk/Dentsply) was available at 1980, and its fluoride release was evaluated over a period of seven days, and found to release about 3.5µg/ml on the first day and reduced sharply to 0.4µg/ml in the last days [58]. After induction of fluoridated fissure sealants into the dental field, several in vitro studies were conducted to evaluate and compare the clinical effectiveness of fluoridated sealants with non fluoridated sealants.

There are some in vitro studies that had proved the ability of fluoridated resins in providing more protection against caries than conventional sealants [59-61]. The addition of fluoride to fissure sealants may change some properties of this material, but there was no difference in micro leakage [62], bond strength [63], and the retention rate between fluoridated and non fluoridated sealants.

Many materials that can release fluoride are now available. For example, glass-ionomers, resin modified glass-ionomer cements, polyacid-modified composites (compomers), and composites. However, a good caries protection from these materials depends on the amount of fluoride that release from them [64]. To look at the fluoride release in terms of the type of the material, the GICs and resin-modified GICs exhibit high rates of fluoride release during the first days, and after that, the amount of fluoride that is released from the material reduces rapidly. This is known as the burst effect of the GICs, and then the fluoride is released at lower and continued levels for an extended period of time [65].

This is in agreement with a study that was conducted to compare the amount and pattern of fluoride release from various fluoride releasing materials (*FluroShield, Helioseal-F, Ultraseal XT, Baritone L3,and Teethmate-F),* and concluded that all the fluoridated sealants tested released measurable fluoride throughout the test period (30 days) in a similar pattern: the greatest amount of fluoride was released in the first 24 hours after mixing, and then fell sharply on the second day and decreased slowly for the last days [66]. Additionally, according to Creanor et al, the amount of fluoride that was released from different kinds of glass ionomers over a period of 60 days dropped sharply from 15.3-155.2 ppm F on day one to range 6.3-44.3 ppm F on day two, and the range of fluoride on day 60 was 0.9-3.99 ppm [67]. Another evaluation of fluoride release from glass ionomers over one year was 0.5-7ppm F [68]. However; some authors have advocated that the high amount of fluoride that is released from the GICs because of their contents of fluoride, as the glass ionomer cements contain 10-23 percent fluoride [69].

Resin modified GICs were found to release fluoride that is considerably similar in amount to conventional GICs [70]. Attar et al found that the manner of fluoride release is like to conventional GIC as the amount of fluoride dropped from 8-15ppm F on the first day to1-2 ppm F on the seventh day[5].

Resin composites release much lower levels of fluoride when compared to conventional and resin modified glass ionomer cements [71, 72]. The amount of fluoride that is released from resin composites depends on several factors, including their fluoride content, type and particle size of filler, and type of resin and porosity [65, 73].

On the other hand, there are several factors that may affect the ability of fluoride-containing dental materials to release fluoride, such as the chemical composition of the material, time of curing, type of media, pH of the saliva, plaque formation, and how often the storage media is changed [74-76].

Dejan et al studied the fluoride release from five glass ionomer materials in different storage medias and different pHs (I- saline, II- acidic solution pH = 2.5, III- acid solution pH = 5.5, IV- NaF solution) and compared them with resin based fluoride releasing materials (Helioseal F). This study, which resulted in low concentrations of fluoride, was released in low pH, and glass-ionomers showed significantly higher fluoride concentrations when compared to the HSF ($p < 0.001$) [77]. However, considerable amounts of fluoride should be released at a low pH where there is a cariogenic challenge, and this was concluded by Jennifer et al who found that fluoride releasing materials have an earlier release of more fluoride at low pH 4 than at pH 7 [78].

The Rechargibilty of Fissure Sealants and Other Fluoride Containing Materials:

As mentioned before, fissure sealants provide good protection against caries by releasing fluoride into the oral cavity; however, the amount of this released fluoride reduces with passage of time, and this could affect the ability of sealants in providing long term cariostatic action.

Most fluoride releasing materials can compensate the loss of fluoride by re uptaking of fluoride from exogenous sources, and thus act as a rechargeable system [5, 6, 79]. Although, the extent to which these materials can be recharged depends on type and permeability of filling material, as fully permeable material can keep the ions obtained from outside deeply in material while semi permeable can just keep them superficially ,on the frequency of fluoride exposure ,and on the kind and concentration of the fluoridating agent [80, 81].

Most studies on this property (rechargibilty) found that glass ionomer based materials are better in re uptaking fluoride than resin based materials [72, 82], and this could be contributed to the nature of the glass ionomer as it is hydrophilic, porous, and a permeable material that can easily allow up taking and re releasing of fluoride [81]. A small amount of fluoride that is released from the resin materials may be from the few ions that are present in superficial surfaces [72].

The rechargibilty of fluoride releasing materials was tested by exposing these materials to different fluoride sources. Some in vitro studies investigated the fluoride release, and recharge from different pit and fissure sealants by subjecting the sealant to APF gel as a recharging agent, and most of these studies proved that the GIC based sealants have a higher initial fluoride release and high rechargibilty by APF gel than the other sealants (polyacid-modified resin composites and resin-based sealants) [83-85].

Delbem et al compared the effects of two fluoride gels (APF gel, neutral gel) in recharging different materials for various times of application (two minutes, four minutes). APF gel was better in recharging materials than neutral gel, especially when used for four minutes [86].

Despite this combination of fissure sealants with APF gel leading to more caries protection, APF gel was founded to affect the properties of sealants; for example, to increase surface roughness of glass ionomer sealants without causing roughness in resin based sealants [87]. Additionally, Moslime et al concluded that “APF gel alters surface hardness of unfilled sealant materials and in respect to micro hardness; it is preferable to use filled sealants if patients are to receive oral care including APF gel application to reduce any adverse effect” [88]. On the other hand, a long term (three months) evaluation of fluoride release from glass ionomers and compomers after application of APF gel resulted in lower fluoride release from the material than before exposing it to APF gel [89]. This could be attributed to acidic action of APF gel that may change the surface structure of the materials. However, as patients are exposed daily to different fluoride sources like fluoridated dentifrices and mouth washes, researchers started to evaluate the ability of these materials in providing fluoride.

Cildir et al examined one month refluoridation by mouthwash (0.05% NaF) and fluoridated toothpaste on fluoride release of glass ionomer cements and polyacid-modified composite resins. All tested materials showed high level of fluoride after application of mouthwash and tooth paste, but it was higher with glass ionomer cements ($p<0.0001$) [79].

There was another study which investigated the fluoride release from resin modified glass ionomer cement (Vitremer) after recharging it with NaF toothpastes (Acta and Pepsodent) and NaF solutions with different fluoride concentrations (0.05 percent,0.2 percent, 2 percent). All the treatments recharged the cement but the 2 percent NaF solution had higher recharging potential than the other treatments [90]. Olsson et al evaluated application of toothpaste slurries either fluoridated (0.05 percent fluoride) or non fluoridated at different pH (2.6, 5.7, 8.3) on fluoride release of modified glass ionomer cements over a period of 30 days. After application, fluoride release was high with fluoridated slurries with an acidic pH (2.6) [91].

Recently, Rao et al evaluated the effect of fluoridated tooth paste (1000 ppm) on recharging non resin auto- cured glass ionomer cement (Fuji VII). After exposure of Fuji VII to fluoridated tooth paste for two minutes twice daily, it did not show any rechargibilty potential by toothpaste [92].

Another in vitro study was conducted to examine fluoride reuptake by giomer and compomer after daily exposure to different fluoride treatments with low concentrations (fluoridated dentifrice (500 ppm) once-daily, fluoridated dentifrice (500 ppm) twice-daily, and fluoridated dentifrice (500 ppm), once-daily plus fluoridated mouthwash (225 ppm) with different deminerlizing and reminerlizing solutions. Both materials released more fluoride at acidic solution, and the amount of fluoride became higher as the material is exposed to more fluoride (twice application of fluoridated dentifrices daily) [93]. Therefore, according to previously mentioned studies, after a short period of time, the fluoride level in the oral cavity declines to its baseline value. Therefore, maintaining high concentrations of fluoride in the oral environment by higher frequency exposure to the fluoridated products like mouthwashes, toothpastes, and fluoride gels may recharge fluoride containing materials and maintain fluoride release for longer periods. However, during daily life, the patient is exposed to different types of fluoridated products in different combinations, either by clinical application of APF gel every six months, daily home use of fluoridated toothpastes, or a combination of both.

Therefore, the purpose of this study was to investigate the fluoride release and the rechargibilty potential of different fissure sealants after exposure to various fluoride preventive treatments with different fluoride concentrations.

The Objectives:

The purpose of this study was to investigate the fluoride release and the rechargibilty potential of different fissure sealants after exposure to various fluoride preventive treatments with different fluoride concentrations.

Hypothesis:

1. The mean of the fluoride release will be high after recharging the fissure sealants with topical fluoride preventive treatments containing the highest fluoride concentration.
2. GIC based materials will have a higher fluoride release and rechargibilty potential than resin based sealants.

Clinical Significance of the Study:

Providing an effective and continuous source of fluoride is necessary for fissure sealants to compensate fluoride that leached constantly from the material in oral cavity, and therefore, fissure sealants can provide prolonged caries protection.

Materials and Methods:

1. Sample Size Calculation:

A power calculation was performed using nQuery Advisor (version 7.0). Assuming a variance in means of $\sigma^2 = 0.266$ for the different fluoride preventive treatment, a variance in means $\sigma^2 = 0.710$ for fissure sealants and within group standard deviation of $\sigma w=1.18$, a sample size of N=5 per subgroup is adequate to achieve a significance level of $\alpha= 0.05$, a power of 84 percent to detect a difference in fluoride preventive treatments, and a power of 99 percent to detect a difference in fissure sealants.

2. Teeth Selection and Preparation for Fissure Sealants:

Sixty caries free extracted permanent molars were used. All the teeth were stored in disinfected solution after the extraction. Soft tissue, calculus, and other debris were mechanically removed by hand scalers. The pits and fissures on the occlusal surface of the teeth cleaned by a water and pumice, then roots of teeth polished by protective varnish (protective nail polish). After that, teeth were numbered from one to sixty and then randomly divided into four groups by using a randomization program on website (www.random.org).

3. Application of the Fissure Sealants to the Teeth:

All the fissure sealants were applied to teeth according to the manufacturer's directions.

Group 1: Helioseal (Ivoclar vivadent AG, 558520,Schaan, Liechtenstein):

After proper isolation of the working field, the enamel surface of the teeth was etched by etching gel Email preparator (Ivoclar Vivadent, Schaan, Liechtenstein). Etching was performed for 30 seconds. The etched enamel surface was then rinsed and dried with water and oil free air. Helioseal was applied by disposable cannula to entire pits and fissures. After 15 seconds, the Helioseal was light cured for 20 seconds using a standard light curing unit (Demintron, Kerr, Germany).

Group 2: Fuji Triage (GC America Inc., 001946,Tokyo, Japan):

The sealant was first mixed by triturating it in a high-speed mixer for 10 seconds, then the mixed capsule was loaded into applicator to deliver the material by thin nozzle to the pits and fissures of the teeth without etching as it is a chemically setting sealant; it took approximately 2 1⁄2 minutes for the Fuji Triage to set.

Group 3: Ultraseal XT Plus (Ultradent, 84095,Utah, USA):

Prior the placement of the sealant, proper moisture control was insured, because Moisture contamination either in liquid or vapor form just prior to resin placement would compromise bond and seal.

Etching of teeth was done by 35 percent phosphoric acid (Ultra-Etch) by using the Inspiral brush tip, etching with Ultra-Etch for 20 seconds, and then teeth were rinsed with full air/water spray and dried.

After that, a drying and priming agent 99 percent Ethyl Alcohol (PrimaDry) was applied with a black micro brush tip for five seconds, and dried by gently blowing with a moisture-free and oil-free air. Subsequently, using an inspiral brush tip a very thin layer of a low-viscosity pit and fissure sealant (Ultraseal XT plus syringe, Ultradent products, Utah, USA) was applied and left for one minute to penetrate. Finally, the sealant was light cured for 20 seconds using a standard light curing unit.

Group 4: Fuji II LC (GC America Inc., 000139,Tokyo, Japan)

The capsule was taped on flat surface to fluff the powder, and then the capsule was activated by depressing the button on the bottom. The capsule was placed in a high speed amalgamator, triturated for 10 seconds, and then placed in applier for delivery. Mixed Fuji II LC was applied to occlusal surface of the teeth and light cured for 20 seconds using a standard light curing unit.

4. Measurement of Initial Fluoride Release:

After application of the fissure sealants, each tooth was immersed in a test tube containing 5 ml of artificial saliva and stored in incubator (Precision water bath, Thermosientific, MA, USA) at a temperature of 37˚C for 24 hours. Artificial saliva was prepared weekly and stored in refrigerator until it needed.

The ingredients of artificial saliva:

-0.077g of Calcium chloride ($CaCl_2$).

-0.049g of Magnesium Chloride Hexahydrate (MgCl2 6H2O).

- 0.544g of Potassium dihydrogen phosphate (KH2PO4).

- 2.23g of potassium chloride (KCl).

- 0.019g of Sodium azide (NaN_3).

- 4.76g HEPES (2-hydroxyethyl)-1-piperazineethanesulfonic acid)

All these materials were dissolved in one liter of distilled water, and the pH of saliva was adjusted by buffers to be 6.2, and measured by pH electrode (ROSS Ultra® 8102 BN, Thermo Scientific, MA, USA).

The initial fluoride release were measured from Fuji Triage, Fuji II LC, Ultraseal XT plus, and Helioseal (control) daily in the first week, after 21 days, and then after 28 days from the placement of the teeth in the saliva. The saliva solution was exchanged 24 hours before each measurement.

A fluoride specific ion electrode (ORION 9609 BN, Thermo Fisher Scientific, MA, USA) was used to measure concentrations of fluoride ions in saliva. The electrode was connected to digital ion analyzer (ORION 2115101.Thermo Fisher Scientific, MA, USA). The electrode was used according to the manufacturer's directions, and calibrated before starting measurements .The concentrations of the standard solutions were 0.001 ppm, 1ppm, and 10ppm.The electrode was rinsed with distilled water and dried with tissue after each measurement in order to prevent overlap in readings.

The measurement of fluoride was carried out by adding 3 ml of each specimen solution with 3 ml of TISAB II (Total ionic strength adjustment buffer, 940906, Orrion Research, Inc, Beverly MA, USA), and then the two solutions were mixed using a magnetic stirrer for one minute before placement of electrode in the mixed specimen. The TISAB II was added in order to adjust the pH, and to decomplex the fluoride ions that bound to H and other ions.

5. Rechaging of Fissure Sealants with the Various Fluoride Preventive Treatments:

On day 29, each group was divided into three subgroups so that each group of the fissure sealants was subjected to three fluoride preventive treatments.

-Subgroup 1: APF gel. (12,300ppm, Acclean, Henry Schein Inc, NY, USA)

-Subgroup 2: APFgel and regular toothpaste (Colgate). (1450ppm, Colgate.Oralphamaceuticals Inc, NY, USA)

- Subgroup 3: High fluoride containing (Prevident) toothpaste (5000ppm, Colgate-Palmolive, NY, USA)

Subgroup 1: the APF gel 1.23 percent was applied to the teeth by immersing them in 10ml of the solution for four minutes, after which the teeth were rinsed with distilled water for 30 seconds and returned back to the saliva tell the time of fluoride measurement.

Subgroup2: the APF gel was applied for four minutes, then two hours later teeth were brushed with slurry of Colgate Total toothpaste. Teeth were brushed daily for three weeks; Slurry was prepared daily by mixing 7g of Colgate Total and 70 ml of distilled water. Each tooth was brushed in 2ml of the slurry for 60 seconds using electric brush (Oral B, Braun, Kornberg, Germany). The teeth then were rinsed thoroughly with distilled water and returned back to the tube containing new saliva.

Subgroup 3: Slurry from Prevident toothpaste was prepared daily by mixing 7g from tooth paste and 70 ml distilled water. Each tooth daily was brushed with 2ml of Prevident slurry for 60 seconds, and then rinsed with distilled water and stored back in fresh saliva.

The amount of the fluoride ions that were released from the sealants after the recharging process were measured daily in the first week, and then weekly for two weeks using the same fluoride electrode(ORION 9609 BN, Thermo Fisher Scientific, MA, USA) and the same procedure .

Statistical analysis:

Repeated measures ANOVA (Analysis Of Variance) by SAS version 9.2 with post hoc tests (Tukey's HSD) were used to analyze the initial fluoride release among fissure sealants at different days, and to compare the fluoride release from fissure sealants among the three fluoride treatments. The critical level of alpha was set at 0.05. For each material, differences in means before and after application of topical fluoride treatments were evaluated using paired t-tests.

Results:

1. Initial Fluoride Release:

Means and standard deviations of all tested fissure sealants at 1-7, 21, and 28 days are presented in Table 2.

All fissure sealants showed high concentrations of fluoride release on the first day especially Glass ionomer sealant (Fuji Triage), and then started to decrease to lower values until the day of recharge.

Throughout the study, Fuji Triage released more fluoride than the others, followed by Fuji II LC, Ultraseal XT plus and then the lowest one Helioseal.

The statistical analysis by Repeated measures ANOVA showed significant differences in initial fluoride release between Fuji Triage and each of the other sealants ($p<0.0001$) while there was no statistical significance between Fuji LL LC –Ultraseal ($p=0.0627$), Fuji LL LC – Helioseal ($p=0.0632$), and Helioseal -Ultraseal ($p=0.9964$).

Figure2 shows the pattern and fluoride release from Helioseal, Ultraseal XT plus, Fuji II LC, and Fuji Triage.

2. Fluoride Release after Application of Various Fluoride Treatments:

In the second part of the study, all fissure sealants released more fluoride than on day 28 (before recharging). However, there was an increase in fluoride release after immersing the teeth in APF gel (sub group1and 2) than after brushing the teeth with high fluoride containing toothpaste (prevident) group (sub group 3).

The fluoride release from fissure sealants at APF gel groups, sub group 1 (as shown in figure3) and sub group2 (as shown in figure4), lasted only for two days, then declined sharply to lower concentrations. Conversely, after brushing with fluoridated toothpaste (Prevident), despite the fact that its fluoride was lower than other groups, it was increasingly released, especially from Fuji Triage (Figure 5).

Table 3 shows means and standard deviations of all fissure sealants after fluoride treatments.

Repeated measures ANOVA showed that APFgel, and APF–toothpaste (Colgate) groups released significantly more fluoride than highly fluoridated toothpaste (Prevident) group ($p<0.0001$). However, there was no significant difference between the APF gel group and APF-toothpaste (Colgate) ($p=0.18$).

Moreover, Fuji Triage released significantly more fluoride than Fuji II LC ($p=0.0057$) and Helioseal ($p=0.0008$), and there was no significant difference between Fuji Triage and Ultraseal XT plus ($p=0.1658$).

There was no statistical significance between Fuji II LC, Helioseal ($p=0.55$) and Fuji II LC and Ultraseal ($p=0.1655$), while there was statistically significant difference between Helioseal and Ultraseal ($p=0.047$)

For each material, differences in means before and after application of topical fluoride treatments are shown in table4.

T-tests indicated statistically significant differences after application of APF gel and APF gel-toothpaste (Colgate) in all tested fissure sealants. While in the high fluoride containing tooth paste (prevident) group, statistical significances were found in GIC based sealant Fuji Triage with p value <0.0001, and resin modified GIC Fuji II LC with P value=0.002.

Discussion:

Initial Fluoride Release from Fissure Sealants:

Introduction of fluoride to restorative materials enhanced their antibacterial and cariostatic properties due to the fact that fluoride that is released from them strengthens the adjacent enamel and reduces secondary caries formation [94]. However, their ability in caries prevention depends on their fluoride contents and release.

In this study the cumulative fluoride release from the tested fissure sealants is similar to previous laboratory studies [79, 95] in which the glass ionomer based sealants have higher initial fluoride than resin based sealants [83]. However, all the fissure sealants showed that the fluoride leached out at a decreasing rate over time, with high amounts at the first week, then drops to a lower concentration in the last week. The amount of fluoride release was from the highest to the lowest as following: Fuji Triage, Fuji II LC, then Ultraseal XTplus and Helioseal.

Fuji Triage, which is a glass ionomer sealant with higher rates of fluoride release, contributed to its chemical composition as Glass-ionomer cements are composed of fluoride-containing silicate glass and polyalkenoic acids which are set by an acid–base reaction between the components. During the setting reaction, different ions are released from the glass, including fluoride.

Previous studies suggested two ways by which fluoride is released from the glass ionomer. The first is a short-term reaction, which involves rapid dissolution from outer surface into solution, whereas the second is more gradual and resulted in the continued diffusion of ions through the bulk cement [96-98]. So accordingly, an initial high release from glass-ionomers over the first 24 hours seems to be due to the burst of fluoride released from the glass particles when reacting with the polyalkenoic acid during the setting reaction [5],where as the sustained, low, and prolonged fluoride release is contributed to dissolving of glass particles in the acidified water of the hydrogel matrix [99, 100]. The initial burst effect of fluoride release in glass ionomers based materials may clinically have some anticariogenic benefits as it forms calcium fluoride on the enamel surface immediately after application of the material [101], and this in turn can enhance remineralization of enamel surface [102]. Additionally it could have some biological effects, such as bactericidal action [65].

The resin modified glass ionomer sealant (Fuji II LC), which is considered as a modification from conventional glass ionomer, is formed by adding a resin component such as hydroxyethyl methacrylate (HEMA) or Bis-GMA in the polyacrylic acid, and liquid component in conventional glass ionomer in order to overcome the problems of moisture sensitivity and low early mechanical strength in Glass-ionomer cements. However, in terms of fluoride release some studies showed that both conventional and resin modified glass ionomers release fluoride in equal amounts[70], others found that conventional release more fluoride than resin modified [103, 104], and this is as in this study in which Fuji Triage (conventional) released more fluoride than Fuji II LC(resin modified glass ionomer). However, the fluoride release could be affected not only by the formation of complex fluoride compounds and their interaction with polyacrylic acid, but also by the type and amount of resin used for the photochemical polymerization reaction [105, 106].

Resin-based sealants like Ultraseal XTplus have patterns in fluoride release that is different from that in glass ionomer based ones. They lacked the initial burst fluoride release, and continued to be released in sustained amounts for a period of time [71, 107].

Most of the previous studies showed that the fluoride that is released from resin based materials is much lower than glass ionomers .This is could be due to high permeability and hydrophilicity of glass ionomers [71, 72, 100].Takahashi et al investigated fluoride release from various glass ionomer materials and composite resins over five weeks. They found that fluoride release from Fuji II LC was similar to that from the glass ionomer Fuji II but significantly greater than that from KS and composite resins [6].

Additionally, in a systemic review, all in vitro studies reported that resin modified glass ionomers have higher fluoride efficacy than composites, except a single in vivo study which showed no difference in fluoride content of plaque adjacent to class III restoration with either material [108].

In this study, unexpectedly we found no statistical difference in fluoride release between Fuji II LC (resin modified) and Ultraseal XTplus (resin-based). This could be because of the contributing factors that affect fluoride release like the media that was used in this study such as artificial saliva, which was not used in most previous studies as they used distilled water as a media. Although the use of artificial saliva as a media simulates the oral environment, it retards the release of fluoride ions. This could be related to high ionic strength of the saliva which may result in lower diffusion of ions in saliva than in distilled water. Additionally, the formation of pellicle over the surface of material can impede ion release [75, 109]. Moreover, artificial saliva contained organic components which may have interfered with the sensitivity of the lanthanum fluoride membrane of the fluoride electrode [110].

Fluoride release from the materials was also affected by pH of immersing media. Glass ionomers, resin-modified glass ionomers, and composites released high amounts of fluoride at an acidic pH. This may be related to increase dissolution of the material at low pH leading to more fluoride release [71, 82, 111, 112]. Conversely, in this study, the pH was around neutral (pH=6.2). This is can be considered in future studies as the pH in the oral cavity is fluctuant and not fixed around neutral.

Recharging of Fissure Sealants with Fluoride Preventive Treatments:

The evidence about synergistic anticariogenic effects of fissure sealants when used together with various topical fluoride treatments is strongly supported by the literature [113, 114]. The findings of these previous studies could support the concept of the ability of fissure sealants in re uptaking fluoride from exogenous sources because anticariogenic effects of fluoride depend mainly on the amount and for how long it is available within the oral cavity.

In this study the rechargibilty of four types of fissure sealants were examined by application of three different fluoride treatments.

All fissure sealants showed high amount of fluoride after immersing them in APF gel and even when the teeth were brushed with regular (Colgate) toothpaste after the APF gel, and this is in line with most previous studies [83, 86].This is could be explained by two facts: first, the concentration of fluoride in APF gel is high, as it was reported before that the fluoride release from the material depends on concentration of recharging agent [115, 116]. Secondly, phosphoric acid that was present in it can etch the surface of the material and enhance its fluoride release[117].

Fissure sealants exhibited high initial bursts of fluoride after immersion in APF gel, and then they experienced a sharp drop in fluoride rate. This is may be associated with washout of fluoride ions that are retained on the surface pores and cracks [89] that may have resulted from erosive action of acidic APF gel. Therefore, thoroughly cleaning of the APF gel is necessary for proper evaluation whether the detected fluoride is from superficial one, or if the fluoride was embedded deeply in the structure of the material. This is was investigated by Kula et al who found that Ultrasonic cleaning is better in removing APF gel than the use of spray of air and water [118].

Interestingly, resin based sealants (Ultraseal XT plus) and non-fluoride containing sealants (Helioseal) showed measurable fluoride re release after gel application. This is in agreement of previous studies which suggested the reason of that release from surface fluoride which embedded in pores and cracks [83, 119]. However, glass ionomer sealants (Fuji triage) significantly released more fluoride than Helioseal (control) and resin modified glass ionomer (Fuji II LC). This is due to its properties as it is a hydrophilic material which enhances the exchange of ions by passive diffusion [65] and a permeable. Therefore, it can absorb the ions deep into its bulk [88].

Since the refluoridation of the material results in re filling the pores and spaces that was initially occupied by fluoride, increasing the concentration of the recharging agents could result in saturation of the material, which limits the extent of re uptaking the external fluoride [120]. This is can be noticed in this study where the brushing of teeth with regularly fluoridated toothpaste after immersing them in fluoride gel didn’t significantly increase the fluoride release.

Brushing of teeth with highly fluoridated toothpaste (Prevident) for one minute resulted in lower uptaking of fluoride by sealants than fluoride gel. This is consistent with Ahn et al and Rao et al who found low potential of tooth paste in providing fluoride to the materials [92, 119], and a study which found that glass ionomers have lower fluoride release and antibacterial effect when used with toothpaste (0.1 percent F) than with fluoride gel (1.25 percent F) [121]. This is may be related to high concentrations of fluoride gel and its acidic pH. As mentioned before, this material releases more fluoride in acidic environment. Olsson et al found that resin modified glass ionomer samples, when treated with fluoridated and non fluoridated tooth paste slurries at pH 2.6, 5.7 and 8.3, the highest fluoride release was with the acidic slurry regardless whether it was fluoridated or not [91].

Therefore, as the pH of Prevident that was used in this study was 8.1, this is can explain the low fluoride release from fissure sealants, and this can be considered in future studies to investigate the effect of pH of the tooth paste slurry in fluoride release. Another factor that affects this study is the time. Teeth were brushed only for one minute, which could lower the exposure of sealants to fluoridated tooth paste. This is could be explains the ability of toothpaste as a recharging material in the study of Attin et al [82] who exposed the specimens to toothpaste for five minutes and Olsson et al [91] who applied the toothpaste for 30 minutes over 10 days.

Moreover, (Dhull et al) proved high rechargibilty can be obtained by increasing brushing time. Brushing twice a day provides more fluoride than once [93], but clinically it is impractical to increase exposure time of high fluoridated toothpastes.

In this study glass ionomer (Fuji Triage) and resin modified glass ionomer (Fuji II LC) showed an increase in fluoride release after brushing with high fluoride containing toothpaste (Prevident) than resin based sealants (Ultraseal and Helioseal). The higher ability of glass ionomer materials in uptaking fluoride than resinous material was documented by previous studies [82, 91, 122]. This can be related to porosity of glass ionomer, so can easily up take fluoride, and as suggested previously that the material with high initial fluoride release has high rechargibilty [65].

Finally,this study was an initial investigation of the fluoride behavior of fissure sealants limited by several laboratory factors that my affect the accuracy of the results. Therefore, more in vivo studies are needed for proper evaluation of the fluoride property in fissure sealants and to draw more clinically applicable conclusions under conditions of oral environment.

Conclusions:

1. Glass ionomer based sealants (Fuji Triage) released significantly more fluoride than the other tested fissure sealants (resin modified glass ionomer, resin sealants).

2. All tested sealants, especially glass ionomer (Fuji Traige), showed high fluoride re uptake after application of APF gel, which has the highest concentration of fluoride.

3. The use of APF gel and APFgel- regular toothpaste (Colgate) treatments significantly recharged the sealants more than high fluoride containing (Prevident) toothpaste.

4.The high fluoride containing toothpaste (Prevident) provided significantly more fluoride only for glass ionmer sealant (Fuji Triage) and resin modified glass ionomer sealant (Fuji II LC).

5. Within the limitations of this study, it can be concluded that glass ionomer based sealants provide higher initial fluoride and high rechargibilty than the other sealants. Therefore they can be used in high risk patients together with fluoride treatments in order to achieve more caries prevention.

Fig1. Groups and Subgroups

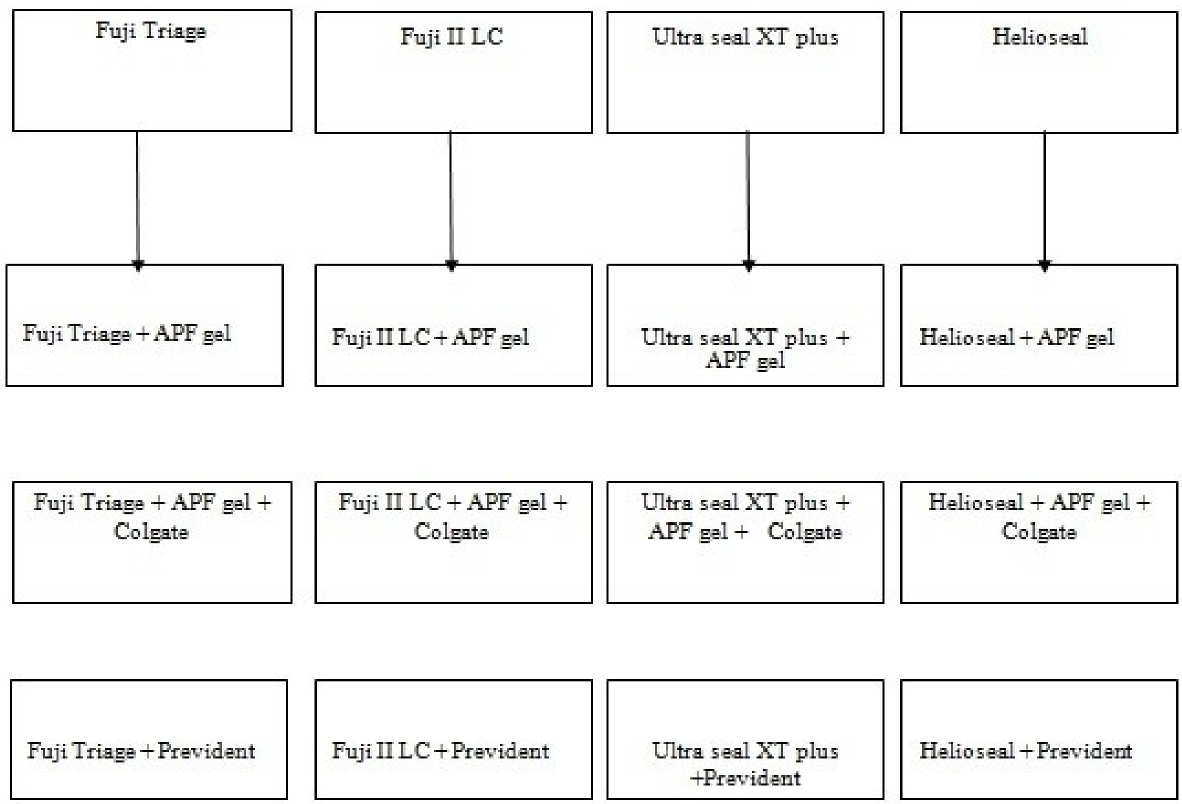

Fig 2. Comparision of Fluoride Release from Four Different Fissure Sealants

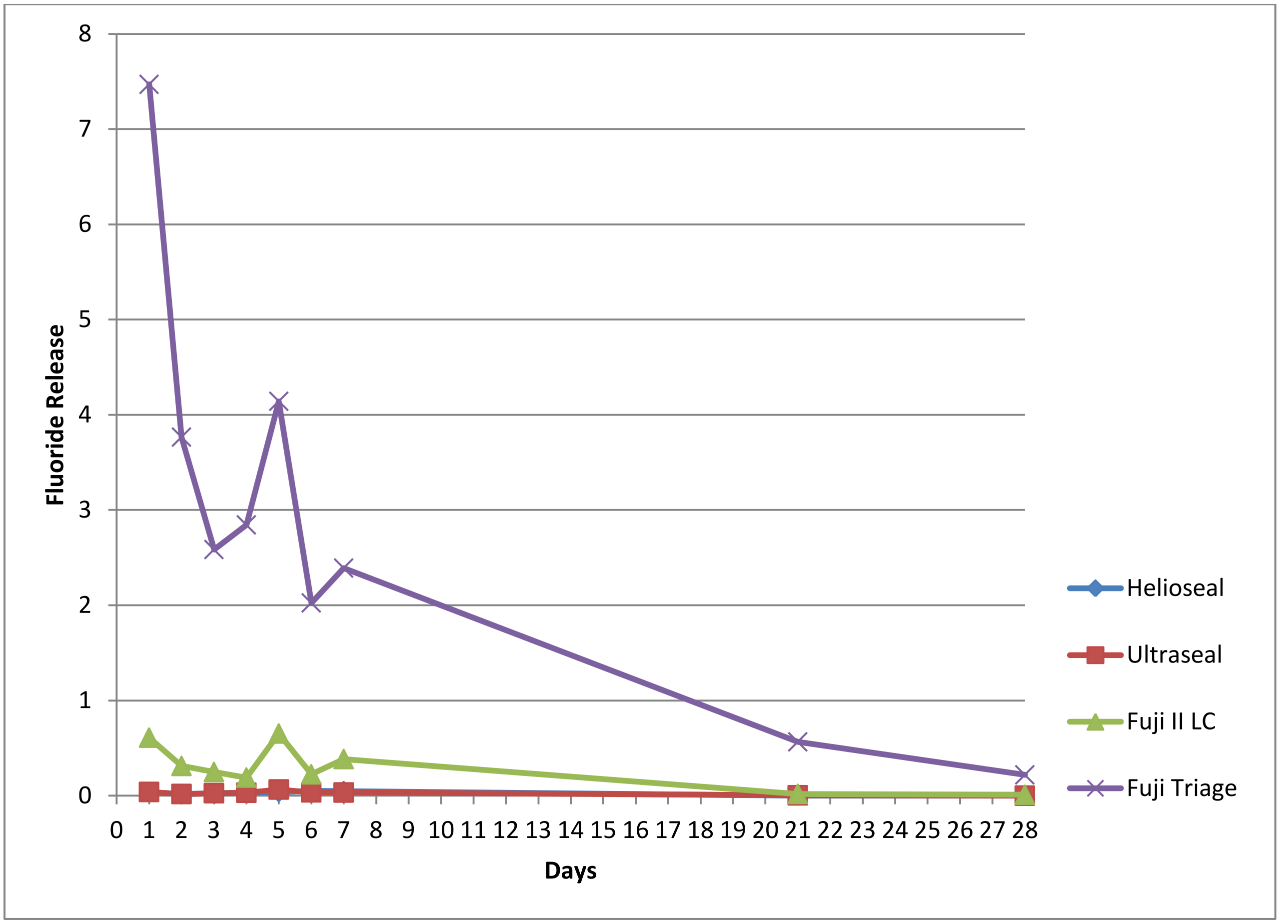

Fig 3. Fluoride Release From Fissure Sealants after APF Gel Application

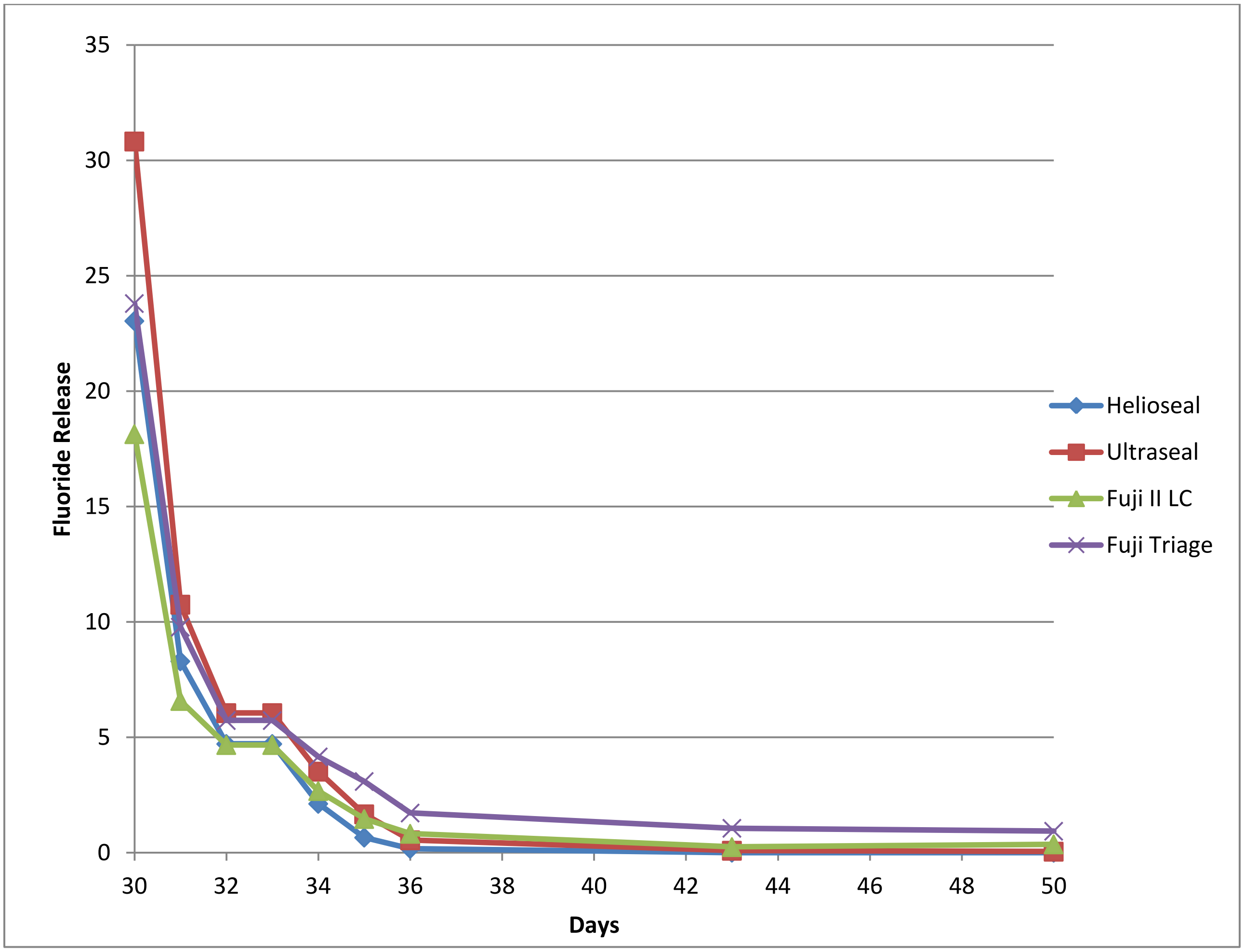

Fig 4. Fluoride Release from Fissure Sealants after APF Gel-Colgate ToothPaste Application

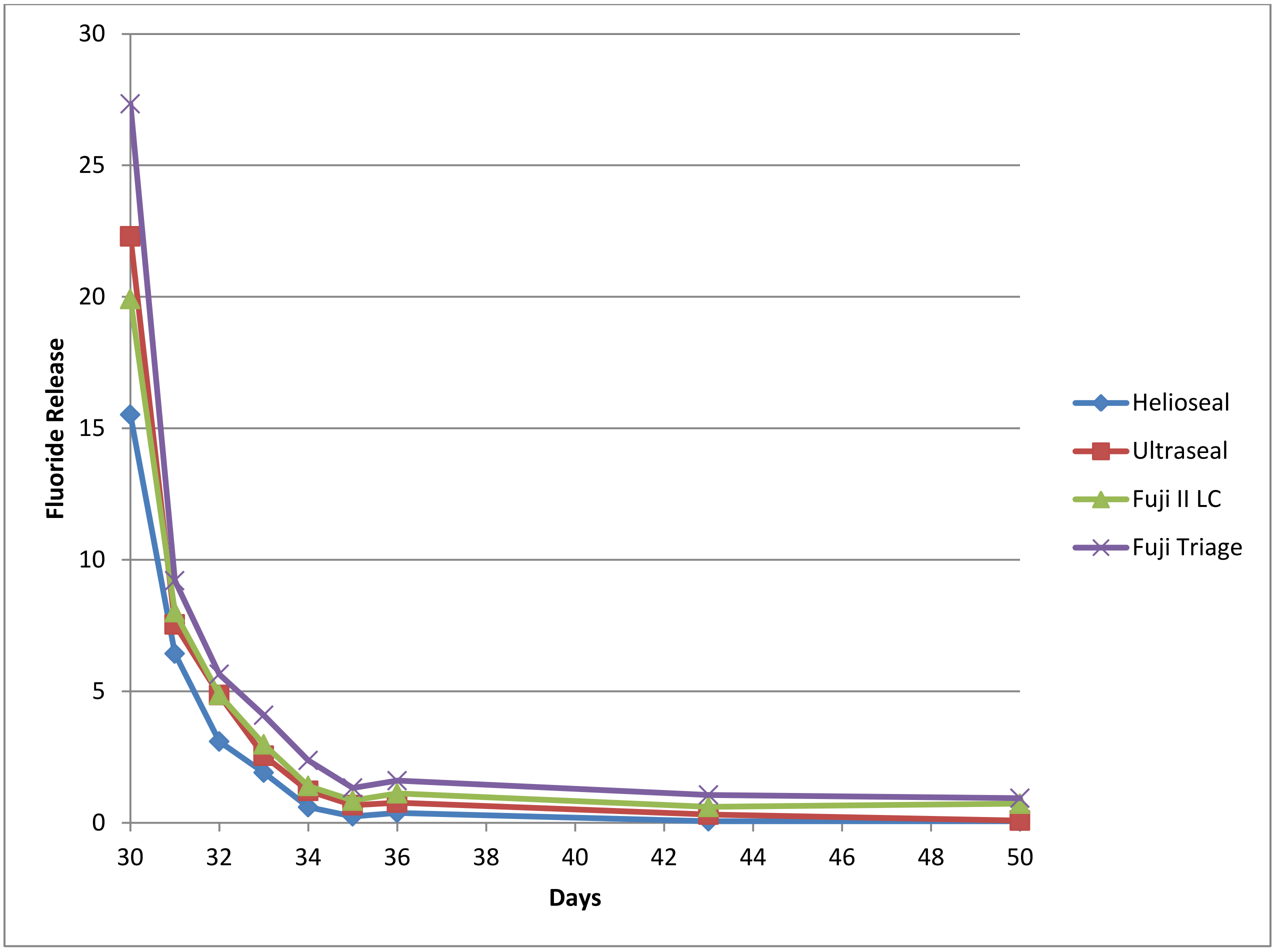

Fig 5. Fluoride Release From Fissure Sealants after Prevident ToothPaste Application

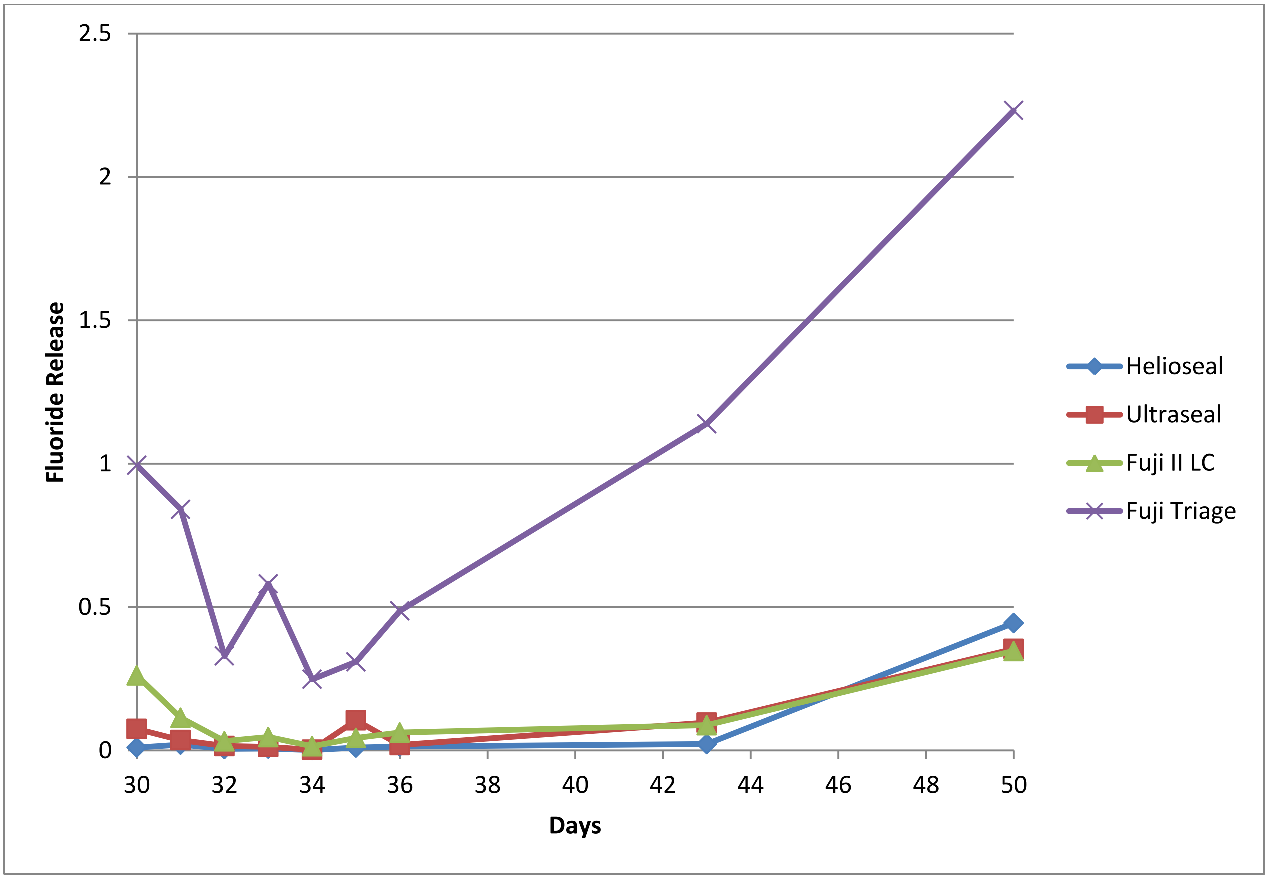

Table1. Materials Used in the study

Product	Type of material	Composition	Manufacturer
Fuji Triage	Glass ionomer	Powder: Alumino-silicate glass Liquid: polyacrylic acid.	GC AMERICA INC.
Fuji II LC	Resin-modified glass ionomer cement	Powder:fluoraluminosilicate glass Liquid:polycarboxylicacid,TEGDMA and HEMA	GC AMERICA INC.
Ultraseal xt plus	Flowable composite	Diurethane Dimethacrylate,Bis GMA,SodiumMonofluorophosphate	UltradentProducts,Inc. south Japan
Helioseal	Conventional resin	Mixture of Bis-GMA, dimethacrylate, titanium dioxide, initiators astabilizers	Ivoclar Vivadent AG,
APF gel	Acidulated phosphate fluoride gel(12300 ppm fluoride)	Sodium fluoride, orthophosphric acid.	Preventive Technologies,Inc
Colgate total	Tooth paste(1450 ppm F)	Sodium Fluoride (1450ppmF) Aqua, Hydrated Silica, Glycerin, Sorbitol, PVM/MA Copolymer, Sodium Lauryl Sulphate, Aroma, CI 77891, Cellulose Gum, Sodium Hydroxide, Sodium Fluoride, Carrageenan, Triclosan, Sodium Saccharin, Limonene.	Colgate
Prevident	Tooth paste(5000 ppm F)	Sodium fluoride 1.1% (w/w) Purified water, sorbitol, hydrated silica, PEG-12, tetrapotassium pyrophosphate, sodium lauryl sulfate, mint flavor , xanthan gum, sodium benzoate.	Colgate

Table. 2: Means (Standard Deviations) of Initial Fluoride Release from Fissure Sealants

Fissure sealants	Day1	Day2	Day3	Day4	Day5	Day6	Day7	Day21	Day28
Helioseal	0.03 (0.04)	0.01 (0.005)	0.025 (0.02)	0.03 (0.01)	0.03 (0.011)	0.05(0.01)	0.04 (0.011)	0.002 (0.006)	0.0008 (0.0002)
Ultraseal	0.03(0.04)	0.01(0.004)	0.02(0.01)	0.03(0.02)	0.06(0.08)	0.03(0.004)	0.03 (0.002)	0.003 (0.008)	0 (0)
Fuji II	0.60(0.35)	0.31(0.23)	0.24(0.16)	0.19(0.11)	0.65(0.37)	0.22(0.1)	0.38 (0.27)	0.01 (0.03)	0.009 (0.01)
Fuji *Triage	7.46(2.18)	3.76(1.98)	2.58(1.02)	2.84(2.21)	4.14(2.17)	2.02(1.19)	2.38 (1.4)	0.56 (0.51)	0.28 (0.26)

* Fuji Triage significantly ($p<0.0001$) released more fluoride than each of the other sealants.

Table. 3: Means (Standard Deviations) of Fluoride Released from Sealants after Fluoride Treatments

Fluoride treatments	Fissure sealants	Day30	Day31	Day32	Day33	Day34	Day35	Day36	Day43	Day50
APF gel[A]	Helioseal	23.04 (6.46)	8.29 (2.32)	4.71 (2.02)	2.12 (1.46)	0.65 (0.71)	0.17 (0.29)	0.05 (0.06)	0.005 (0.006)	0.004 (0.005)
	Ultraseal	30.82 (6.95)	10.75 (2.82)	6.05 (1.71)	3.51 (1.17)	1.66 (0.49)	0.54 (0.44)	0.47 (0.41)	0.08 (0.09)	0.04 (0.07)
	Fuji ILC	18.14 (5.72)	6.58 (3.08)	4.67 (1.79)	2.67 (0.57)	1.46 (0.54)	0.82 (0.43)	0.7 (0.44)	0.24 (0.27)	0.36 (0.33)
	*Fuji Triage	23.8 (4.77)	9.77 (2.82)	5.73 (1.33)	4.15 (0.55)	3.09 (1.25)	1.72 (1.01)	1.64 (0.38)	1.06 (0.58)	0.93 (0.89)
APFgel[B] + Colgate	Helioseal	1.52 (3.83)	6.43 (2.84)	3.09 (1.57)	1.91 (1.72)	0.59 (0.69)	0.25 (0.43)	0.38 (0.58)	0.06 (0.061)	0.06 (0.09)
	Ultraseal	22.3 (5.23)	7.54 (2.51)	4.86 (1.71)	2.55 (0.64)	1.21 (0.65)	0.67 (0.46)	0.77 (0.54)	0.31 (0.29)	0.08 (0.007)
	Fuji ILC	19.92 (6.26)	8.01 (3.07)	4.87 (1.85)	2.99 (1.36)	1.4 (0.9)	0.85 (0.67)	1.12 (0.72)	0.6 (0.43)	0.7 (0.5)
	*Fuji Triage	27.34 (7.42)	9.21 (1.28)	5.65 (1.16)	4.09 (0.92)	2.37 (0.81)	1.32 (0.59)	1.6 (0.63)	1.06 (0.62)	0.93 (0.64)
Prevident	Helioseal	0.01 (0.011)	0.02 (0.01)	0.005 (0.007)	0.006 (0.005)	0.001 (0.002)	0.01 (0.01)	0.014 (0.013)	0.022 (0.02)	0.4 (0.5)
	Ultraseal	0.07 (0.08)	0.036 (0.034)	0.015 (0.017)	0.012 (0.011)	0.002 (0.003)	0.1 (0.2)	0.019 (0.014)	0.09 (0.12)	0.3 (0.5)
	Fuji ILC	0.26 (0.28)	0.11 (0.13)	0.03 (0.031)	0.05 (0.04)	0.014 (0.015)	0.04 (0.05)	0.06 (0.05)	0.08 (0.07)	0.34 (0.19)
	*Fuji Triage	0.99 (0.69)	0.84 (0.44)	0.32 (0.33)	0.5 (0.6)	0.2 (0.1)	0.31 (0.33)	0.5 (0.4)	1.13 (0.49)	2.23 (0.73)

A: Fluoride release from fissure sealants in APF gel group is statistically significant than in prevident group ($p<0.0001$).

B: Fluoride release from sealants in APF gel +Colgate group is statistically significant than in prevident group ($p<0.0001$).

* Fuji Triage significantly ($p=0.0057$) released more fluoride than Fuji IILC, and Helioseal (0.0008).

Table.4: Comparisons of Means (Standard Deviations) of Fluoride Release from Fissure Sealants before and after Each Fluoride Treatment

Fluoride treatments	Fissure sealants	Before treatment	After treatment	P value
APFgel[A]	Helioseal	0.04(0.01)	4.5(0.8)	<0.0001
	Ultraseal	0.03(0.02)	6.06(1.45)	0.001
	Fuji II	0.29(0.08)	3.97(0.44)	<0.0001
	Fuji Triage	2.72(0.53)	5.76(0.56)	0.001
APF gel + Colgate [B]	Helioseal	0.002(0.003)	3.14(0.57)	<0.0001
	Ultraseal	0.024(0.004)	3.15(0.57)	<0.0001
	Fuji II	0.26(0.06)	4.5(0.95)	<0.0001
	Fuji Triage	3.04(0.17)	5.95(0.68)	0.001
Prevident	Helioseal	0.02(0.001)	0.051(0.052)	0.16
	Ultraseal	0.02(0.001)	0.051(0.052)	0.16
	Fuji II [1]	0.31(0.05)	0.11(0.03)	0.002
	Fuji Triage [2]	2.9(0.28)	0.79(0.07)	<0.0001

A, B: All fissure sealants showed statistically significant recharge after application of APF gel, and APF gel Colgate tooth paste.

1, 2: After prevident application only Fuji II LC ($p=0.002$), and Fuji Triage ($p<0.0001$) significantly recharged.

References:

1. Ten Cate, J.M., Current concepts on the theories of the mechanism of action of fluoride. Acta odontologica Scandinavica, 1999. 57(6): p. 325-9.
2. Batchelor, P.A. and A. Sheiham, Grouping of tooth surfaces by susceptibility to caries: a study in 5-16 year-old children. BMC oral health, 2004. 4(1)p.2.
3. Simonsen, R.J., Pit and fissure sealant: review of the literature. Pediatric dentistry, 2002. 24(5): p. 393-414.
4. Marinelli, C.B., et al., An in vitro comparison of three fluoride regimens on enamel remineralization. Caries research, 1997. 31(6): p. 418-22.
5. Attar, N. and M.D. Turgut, Fluoride release and uptake capacities of fluoride-releasing restorative materials. Operative dentistry, 2003. 28(4): p. 395-402.
6. Takahashi, K., Emilson, C. G. & Birkhed, D, . Fluoride release in vitro from various glass ionomer cements and resin composites after exposure to NaF solutions. . Dent. Mater. 9, 350-354 1993.
7. Winston, A.E. and S.N. Bhaskar, Caries prevention in the 21st century. Journal of the American Dental Association, 1998. 129(11): p. 1579-87.
8. Rozier, R.G., et al., Evidence-based clinical recommendations on the prescription of dietary fluoride supplements for caries prevention: a report of the American Dental Association Council on Scientific Affairs. Journal of the American Dental Association, 2010. 141(12): p. 1480-9.
9. Kaste, L.M., et al., Coronal caries in the primary and permanent dentition of children and adolescents 1-17 years of age: United States, 1988-1991. Journal of dental research, 1996. 75 Spec No: p. 631-41.
10. Winn, D.M., et al., Coronal and root caries in the dentition of adults in the United States, 1988-1991. Journal of dental research, 1996. 75 Spec No: p. 642-51.
11. Brunelle, J.A. and J.P. Carlos, Recent trends in dental caries in U.S. children and the effect of water fluoridation. Journal of dental research, 1990. 69 Spec No: p. 723-7; discussion 820-3.

12. Zero, D.T., Dental caries process. Dental clinics of North America, 1999. 43(4): p. 635-64.
13. Ioan Danila, M.P. and R. Iasi, Fluoride in caries prevention – methods and techniques. Oral health and dental mangment - 2002 - 1.
14. Hamilton, I.R., Biochemical effects of fluoride on oral bacteria. Journal of dental research, 1990. 69 Spec No: p. 660-7; discussion 682-3.
15. U.S. Public Health Service. Public Health Service Drinking Water Standards. Washington: U.S. Department of Health, Education, and Welfare, Public Health Service;.
16. Driscoll WS,What we know and don't know about dietary fluoride supplements – the research basis. ASDC J Dent Child1985; 52: 259-264.
17. Parnell, C., H. Whelton, and D. O'Mullane, Water fluoridation. European archives of paediatric dentistry : official journal of the European Academy of Paediatric Dentistry, 2009. 10(3): p. 141-8.
18. Treasure, E.T., et al., The York review--a systematic review of public water fluoridation: a commentary. British dental journal, 2002. 192(9): p. 495-7.
19. Truman, B.I., et al., Reviews of evidence on interventions to prevent dental caries, oral and pharyngeal cancers, and sports-related craniofacial injuries. American journal of preventive medicine, 2002. 23(1 Suppl): p. 21-54.
20. Murray JJ., Efficacy of preventive agents for dental caries: systemic fluorides—water fluoridation. 1993. Caries Res 1993;27(suppl 1):2-8.
21. Centers for Disease Control and Prevention. Recommendations for using fluoride to prevent and control dental caries in the United States. MMWR Recomm Rep 2001;50(RR-14):1-42.
22. Ripa LW, A critique of topical fluoride methods (dentifrices, mouthrinses, operator- and self-applied gels) in an era of decreased caries and increased fluorosis prevalence. J Public Health Dent. 1991; 51: 23-41.
23. U.S. Food and Drug Administration. Anticaries drug products for over-the-counter human use. Final monograph. Fed Regist.1995; 60 (194): 52474-510.

24. Bowen WH, The role of fluoride toothpastes in the prevention of dental caries. J RSoc Med 1995;88:50-7.

25. American academy of pediatric dentistry reference manual 2011-2012. . Pediatr Dent. . 2011;33:1-349.

26. Marinho VC, H.J., Sheiham A, Logan S, One topical fluoride (toothpastes, or mouthrinses, or gels, or varnishes) versus another for preventing dental caries in children and adolescents. . Cochrane Database Syst Rev. 2004;(1):CD002780.

27. Ernest Newbrun, D.M.D., Ph.D., Topical Fluorides in Caries Prevention and Management: A North American Perspective. Journal of Dental Education 2011.65(10):p.1078-83 .

28. Rafael L,Bowen, in dental filling material compromising vinyl silane-treated fused silica and binder consisting of the reaction product of bisphenol and glycidylacrylate. U.S. Patent 3,006, 112. 1962.

29. Tilliss, T.S., et al., Occlusal discrepancies after sealant therapy. The Journal of prosthetic dentistry, 1992. 68(2): p. 223-8.

30. Papacchini, F., et al., Noninvasive pit and fissure sealing: microtensile bond strength to intact bovine enamel of different pit and fissure sealants in a simplified fissure model. The journal of adhesive dentistry, 2006. 8(6): p. 375-80.

31. Shapira, J., et al., Comparative clinical study of autopolymerized and light-polymerized fissure sealants: five-year results. Pediatric dentistry, 1990. 12(3): p. 168-9.

32. Muller-Bolla, M., et al., Retention of resin-based pit and fissure sealants: A systematic review. Community dentistry and oral epidemiology, 2006. 34(5): p. 321-36.

33. Simonsen, R.J., Retention and effectiveness of dental sealant after 15 years. Journal of the American Dental Association, 1991. 122(10): p. 34-42.

34. McLean, J.W. and A.D. Wilson, Fissure sealing and filling with an adhesive glass-ionomer cement. British dental journal, 1974. 136(7): p. 269-76.

35. Seppa, L. and H. Forss, Resistance of occlusal fissures to demineralization after loss of glass ionomer sealants in vitro. Pediatric dentistry, 1991. 13(1): p. 39-42.

36. Aranda, M. and F. Garcia-Godoy, Clinical evaluation of the retention and wear of a light-cured pit and fissure glass ionomer sealant. The Journal of clinical pediatric dentistry, 1995. 19(4): p. 273-7.
37. Futatsuki, M., et al., Wear of resin-modified glass ionomers: an in vitro study. The Journal of clinical pediatric dentistry, 2001. 25(4): p. 297-301.
38. Deery, C., Pits and fissure sealant guidelines. Summary guideline. Evidence-based dentistry, 2008. 9(3): p. 68-70.
39. Beauchamp, J.e.a., Evidence-based clinical recommendations for the use of pit-and-fissure sealants: a report of the American Dental Association Council on Scientific Affairs. 2009. . Dent. Clin. North Am. 53, 131-147
40. Sasa, I. and K.J. Donly, Sealants: a review of the materials and utilization. Journal of the California Dental Association, 2010. 38(10): p. 730-4.
41. Locker, D., Jokovic, A. & Kay, E. J. Prevention., The use of pit and fissure sealants in preventing caries in the permanent dentition of children. 2003. Br. Dent. J. 195, 375-378
42. Oulis C, T.M., Mastrogiannakis T, Mamai-Chomata E, Polychronopoulou A, Athanasouli T., Oral health status and treatment needs of the Hellenic population-proposals for improvement. Hellenic Stomatological Review 2009, 53(2):97-120.
43. Leskinen, K., et al., Comparison of the effectiveness of fissure sealants in Finland, Sweden, and Greece. Acta odontologica Scandinavica, 2008. 66(2): p. 65-72.
44. Mertz-Fairhurst, E.J., G.S. Schuster, and C.W. Fairhurst, Arresting caries by sealants: results of a clinical study. Journal of the American Dental Association, 1986. 112(2): p. 194-7.
45. Beauchamp, J.e.a., Evidence-based clinical recommendations for the use of pit-and-fissure sealants: a report of the American Dental Association Council on Scientific Affairs. . [Review] [99 refs][Reprint in Dent Clin North Am. 2009 Jan;53(1):131-47, x; PMID: 19215748]. J. Am. Dent. Assoc. 139, 257-268 (2008).
46. Griffin, S.O., et al., The effectiveness of sealants in managing caries lesions. Journal of dental research, 2008. 87(2): p. 169-74.
47. Salar, D.V., et al., Potential inhibition of demineralization in vitro by fluoride-releasing sealants. Journal of the American Dental Association, 2007. 138(4): p. 502-6.

48. Kantovitz, K.R.e.a., Inhibition of mineral loss at the enamel/sealant interface of fissures sealed with fluoride- and non-fluoride containing dental materials in vitro.. Acta Odontol. Scand 2006. 64, p:376-383

49. Donly, K.J., et al., Evaluating the effects of fluoride-releasing dental materials on adjacent interproximal caries. Journal of the American Dental Association, 1999. 130(6): p. 817-25.

50. Fejerskov O, C.B., Dynamics of caries lesion formation. In: . 1996. Fejerskov O, Ekstrand J, Burt BA, editors. Fluoride in dentistry. Copenhagen: Munksgaard; 1996. p.187–213.

51. Svanberg, M., I.A. Mjor, and D. Orstavik, Mutans streptococci in plaque from margins of amalgam, composite, and glass-ionomer restorations. Journal of dental research, 1990. 69(3): p. 861-4.

52. Feierskov O,Ekstrand J, in Physicochemical aspects of fluoride-enamel interactions. In: Fejerskov O, Ekstrand J, Burt BA, editors. Fluoride in Dentistry, 2nd Edition. Copenhagen: Munksgaard, 1996. p. 252–72 [Chapter 14].

53. Stookey, G.K., et al., The effect of rinsing with water immediately after a professional fluoride gel application on fluoride uptake in demineralized enamel: an in vivo study. Pediatric dentistry, 1986. 8(3): p. 153-7.

54. Ripa, L.W., Sealants revisted: an update of the effectiveness of pit-and-fissure sealants. Caries research, 1993. 27 Suppl 1: p. 77-82.

55. Jensen, O.E., R.J. Billings, and J.D. Featherstone, Clinical evaluation of Fluroshield pit and fissure sealant. Clinical preventive dentistry, 1990. 12(4): p. 24-7.

56. Fluoride-releasing sealants. Journal of the American Dental Association, 1985. 110(1): p. 90.

57. Kadoma, Y., Kojima, K. & Masuhara, E, . Studies on dental fluoride-releasing polymers. IV: Fluoridation of human enamel by fluoride-containing sealant.. 1983. Biomaterials 4, 89-93

58. Cooley, R.L., et al., Evaluation of a fluoride-containing sealant by SEM, microleakage, and fluoride release. Pediatric dentistry, 1990. 12(1): p. 38-42.

59. Hicks, M.J. and C.M. Flaitz, Caries formation in vitro around a fluoride-releasing pit and fissure sealant in primary teeth. ASDC journal of dentistry for children, 1998. 65(3): p. 161-8.

60. Hicks, M.J. and C.M. Flaitz, Caries-like lesion formation around fluoride-releasing sealant and glass ionomer. American journal of dentistry, 1992. 5(6): p. 329-34.

61. Jensen, M.E., et al., Effects of a fluoride-releasing fissure sealant on artificial enamel caries. American journal of dentistry, 1990. 3(2): p. 75-8.

62. Park, K., Georgescu, M., Scherer, W. & Schulman, A, . Comparison of shear strength, fracture patterns, and microleakage among unfilled, filled, and fluoride-releasing sealants. . Pediatr. Dent. 15, 418-421 1993.

63. Marcushamer, M., Neuman, E. & Garcia-Godoy, F, . . Fluoridated and nonfluoridated unfilled sealants show similar shear strength. . Pediatr Dent. 19, 289-290 1997.

64. Wiegand, A., Buchalla, W. & Attin, T, . Review on fluoride-releasing restorative materials--fluoride release and uptake characteristics, antibacterial activity and influence on caries formation. . Dent. Mater. 23, 343-362 2007.

65. Xu, X. and J.O. Burgess, Compressive strength, fluoride release and recharge of fluoride-releasing materials. Biomaterials, 2003. 24(14): p. 2451-61.

66. Garcia-Godoy, F., et al., Fluoride release from fissure sealants. The Journal of clinical pediatric dentistry, 1997. 22(1): p. 45-9.

67. Creanor, S.L., et al., Fluoride uptake and release characteristics of glass ionomer cements. Caries research, 1994. 28(5): p. 322-8.

68. Perrin, C., M. Persin, and J. Sarrazin, A comparison of fluoride release from four glass-ionomer cements. Quintessence international, 1994. 25(9): p. 603-8.

69. Forsten, L., Fluoride release and uptake by glass ionomers. Scandinavian journal of dental research, 1991. 99(3): p. 241-5.

70. Robertello, F.J., et al., Fluoride release of glass ionomer-based luting cements in vitro. The Journal of prosthetic dentistry, 1999. 82(2): p. 172-6.

71. Karantakis, P., et al., Fluoride release from three glass ionomers, a compomer, and a composite resin in water, artificial saliva, and lactic acid. Operative dentistry, 2000. 25(1): p. 20-5.

72. Attar, N. and A. Onen, Fluoride release and uptake characteristics of aesthetic restorative materials. Journal of oral rehabilitation, 2002. 29(8): p. 791-8.

73. Dijkman, G.E., et al., Long-term fluoride release of visible light-activated composites in vitro: a correlation with in situ demineralisation data. Caries research, 1993. 27(2): p. 117-23.

74. Dhondt, C.L., E.A. De Maeyer, and R.M. Verbeeck, Fluoride release from glass ionomer activated with fluoride solutions. Journal of dental research, 2001. 80(5): p. 1402-6.

75. Bell, A., et al., The effect of saliva on fluoride release by a glass-ionomer filling material. Journal of oral rehabilitation, 1999. 26(5): p. 407-12.

76. Hayacibara, M.F., Ambrozano, G. M. & Cury, J. A, . Simultaneous release of fluoride and aluminum from dental materials in various immersion media. . Oper. Dent. 29, 16-22 2004.

77. Markovic, D., B.B. Petrovic, and T.O. Peric, Fluoride content and recharge ability of five glassionomer dental materials. BMC oral health, 2008. 8: p. 21.

78. Moreau, J.L. and H.H. Xu, Fluoride releasing restorative materials: Effects of pH on mechanical properties and ion release. Dental materials : official publication of the Academy of Dental Materials, 2010. 26(11): p. e227-35.

79. Cildir, S.K. and N. Sandalli, Fluoride release/uptake of glass-ionomer cements and polyacid-modified composite resins. Dental materials journal, 2005. 24(1): p. 92-7.

80. Han, L., et al., Effect of fluoride mouth rinse on fluoride releasing and recharging from aesthetic dental materials. Dental materials journal, 2002. 21(4): p. 285-95.

81. Preston, A.J., et al., Fluoride recharge of aesthetic dental materials. Journal of oral rehabilitation, 1999. 26(12): p. 936-40.

82. Attin, T., et al., Fluoride release/uptake of polyacid-modified resin composites (compomers) in neutral and acidic buffer solutions. Journal of oral rehabilitation, 1999. 26(5): p. 388-93.

83. Bayrak, S.e.a., Fluoride release and recharge from different materials used as fissure sealants. . Eur. J. Dent. 4, 245-250, (2010).

84. Kavaloglu Cildir, S. and N. Sandalli, Compressive strength, surface roughness, fluoride release and recharge of four new fluoride-releasing fissure sealants. Dental materials journal, 2007. 26(3): p. 335-41.

85. Koga, H., et al., Comparison of short-term in vitro fluoride release and recharge from four different types of pit-and-fissure sealants. The Bulletin of Tokyo Dental College, 2004. 45(3): p. 173-9.

86. Delbem, A.C., et al., Fluoride release/recharge from restorative materials--effect of fluoride gels and time. Operative dentistry, 2005. 30(6): p. 690-5.

87. Shafiei F, M.M., . . In-vitro Study of the Surface Roughness of Two Fissure Sealants after Repeated Topical Acidulated Phosphated Fluoride Application. . Shiraz Univ Dent J 2010; 11(2): 117-123, 2010.

88. Moslemi M, K.S., Shadkar M, Ghasemi A, Tadayon N., Effect of APF gel on the micro hardness of sealant materials. Res. J. Biol. Sci 4 (6): 724-727,, 2009.

89. Gao, W. and R.J. Smales, Fluoride release/uptake of conventional and resin-modified glass ionomers, and compomers. Journal of dentistry, 2001. 29(4): p. 301-6.

90. Posada, A., C.G. Emilson, and D. Birkhed, Fluoride release in vitro from a resin-modified glass ionomer after exposure to NaF solutions and toothpastes. Swedish dental journal, 2000. 24(3): p. 117-25.

91. Olsson, C., C.G. Emilson, and D. Birkhed, An in vitro study of fluoride release from a resin-modified glass ionomer cement after exposure to toothpaste slurries of different pH. Clinical oral investigations, 2000. 4(4): p. 233-7.

92. Rao, A. and P. Sudha, Fluoride rechargability of a non-resin auto-cured glass ionomer cement from a fluoridated dentifrice: an in vitro study. Journal of the Indian Society of Pedodontics and Preventive Dentistry, 2011. 29(3): p. 202-4.

93. Dhull, K.S. and B. Nandlal, Effect of low-concentration daily topical fluoride application on fluoride release of giomer and compomer: an in vitro study. Journal of the Indian Society of Pedodontics and Preventive Dentistry, 2011. 29(1): p. 39-45.

94. Tyas, M.J., Cariostatic effect of glass ionomer cement: a five-year clinical study. Australian dental journal, 1991. 36(3): p. 236-9.

95. Adem Ku¸sg, Mustafa Ülkerb, Barı ¸s Kemerc, Onur Sarayd, Conversion degree, microhardness, microleakage and fluoride release of different fissure sealants. Journal of the mechanical behaviour of medical materoals (2010)5 9 4 ± 5 9 9.

96. Verbeeck RM, D.M.E., Marks LA, De Moor RJ, De and T.L. Witte AM, . Fluoride release process of(resin-modified) glass-ionomer cements versus (polyacid-modified) composite resins. Biomaterials1998;19:509–19.

97. Lee SY, D.D., Huang HM, Shih YH, . Fluoride iondiffusion from a glass-ionomer cement. J Oral Rehabil 2000;27:576–86.

98. Tay WM, B.M., . Fluoride ion diffusion from polyalkenoate (glass-ionomer) cements. Biomaterials1988;9:454–6.

99. De Moor RJ, V.R., De Maeyer EA., Fluoride release profiles of restorative glass ionomer formulations. . Dent Mater 1996;12:88–95.

100. Vermeersch G, L.G., Vreven J, . Fluoride release from glass-ionomer cements, compomers and resin composites. J Oral Rehabil 2001;28:26–32.

101. Rolla, G. and E. Saxegaard, Critical evaluation of the composition and use of topical fluorides, with emphasis on the role of calcium fluoride in caries inhibition. Journal of dental research, 1990. 69 Spec No: p. 780-5; discussion 820-3.

102. Wiltshire, W.A., Determination of fluoride from fluoride-releasing elastomeric ligature ties. American journal of orthodontics and dentofacial orthopedics : official publication of the American Association of Orthodontists, its constituent societies, and the American Board of Orthodontics, 1996. 110(4): p. 383-7.

103. Bertacchini, S.M., et al., Solubility and fluoride release in ionomers and compomers. Quintessence international, 1999. 30(3): p. 193-7.

104. Carvalho, A.S. and J.A. Cury, Fluoride release from some dental materials in different solutions. Operative dentistry, 1999. 24(1): p. 14-9.

105. Momoi, Y. and J.F. McCabe, Fluoride release from light-activated glass ionomer restorative cements. Dental materials : official publication of the Academy of Dental Materials, 1993. 9(3): p. 151-4.

106. Tjandrawinata, R., M. Irie, and K. Suzuki, Marginal gap formation and fluoride release of resin-modified glass-ionomer cement: effect of silanized spherical silica filler addition. Dental materials journal, 2004. 23(3): p. 305-13.

107. Eichmiller FC, Mrrjenhoff W.A, Fluoride-releasing dental restorative materials,Oper. Dent. 23 (1998) 213–218.

108. Group, T.M.R., fluoride release of resin-modified GIC versus fluoride containing composite resin-a quantitatve systematic review. J Minim Interv Dent 2010 ; 3 (2), 2010.

109. Williams, J.A., R.W. Billington, and G.J. Pearson, A long term study of fluoride release from metal-containing conventional and resin-modified glass-ionomer cements. Journal of oral rehabilitation, 2001. 28(1): p. 41-7.

110. Wiltshire, W.A. and S.D. Janse van Rensburg, Fluoride release from four visible light-cured orthodontic adhesive resins. American journal of orthodontics and dentofacial orthopedics : official publication of the American Association of Orthodontists, its constituent societies, and the American Board of Orthodontics, 1995. 108(3): p. 278-83.

111. Abu-Bakr, N.H., et al., Effect of alcoholic and low-pH soft drinks on fluoride release from compomer. Journal of esthetic dentistry, 2000. 12(2): p. 97-104.

112. Nicholson, J.W. and B. Czarnecka, The release of ions by compomers under neutral and acidic conditions. Journal of oral rehabilitation, 2004. 31(7): p. 665-70.

113. Ripa, L.W., G.S. Leske, and F. Forte, The combined use of pit and fissure sealants and fluoride mouthrinsing in second and third grade children: final clinical results after two years. Pediatric dentistry, 1987. 9(2): p. 118-20.

114. Rantala EV, Caries incidence in 7-9-year-old children after fissuresealing and topical fluoride therapy in Finland. . Community Dent Oral Epidemiol 1979;7:213-7.

115. Diaz-Arnold, A.M., et al., Short-term fluoride release/uptake of glass ionomer restoratives. Dental materials : official publication of the Academy of Dental Materials, 1995. 11(2): p. 96-101.

116. Hatibovic-Kofman, S., G. Koch, and J. Ekstrand, Glass ionomer materials as a rechargeable fluoride-release system. International journal of paediatric dentistry / the British Paedodontic Society [and] the International Association of Dentistry for Children, 1997. 7(2): p. 65-73.

117. Garcia-Godoy, F. and S. Leon de Perez, Effect of fluoridated gels on a light-cured glass ionomer cement: an SEM study. The Journal of clinical pediatric dentistry, 1993. 17(2): p. 83-7.

118. Kula, K., S. Nelson, and V. Thompson, In vitro effect of APF gel on three composite resins. Journal of dental research, 1983. 62(7): p. 846-9.

119. Ahn, S.J., Lee, S. J., Lee, D. Y. & Lim, B. S., Effects of different fluoride recharging protocols on fluoride ion release from various orthodontic adhesives. J. Dent. 39, 196-201 (2011).

120. Itota, T., et al., Fluoride release and recharge in giomer, compomer and resin composite. Dental materials : official publication of the Academy of Dental Materials, 2004. 20(9): p. 789-95.

121. Seppa, L., H. Forss, and B. Ogaard, The effect of fluoride application on fluoride release and the antibacterial action of glass ionomers. Journal of dental research, 1993. 72(9): p. 1310-4.

122. Buchalla W, A.T., Kalb K, Hellwig E., Buchalla W, Attin T, Kalb K, Hellwig E. Fluoride release anduptake of a new experimental composite in vitro and insitu. . Deutsche Zahnarztiche Zeitschrift 1998;53:707–12.