Comparative Evaluation of fluoride releasing and recharging ability for various pit and fissure sealants

(An in vitro study)

Vian O. Majeed

Background and Objective: This study was conducted to evaluate the releasing and recharging ability of fluoride for four different types of pit and fissure sealants.

Methods: Four different pit and fissure sealants: GIC Ketac® Cem radiopaque, 3M ESPE, Photac Fill, 3M ESPE (light-curing, GIC), Palfique (Universal flow, Tokuyama) and Charisma (composite) were used in this study, ten cylindrical specimens were prepared of each material (10 mm in diameter and 1 mm in thickness). Specimens were dipped in 5 ml deionized water and fluoride release was detected using a fluoride-specific ion electrode every day from 1,2,3,4,5,6,7,14,21 and 28 days. On day 28, specimens were exposed to a fluoride varnish and then dipped in water, and then the fluoride concentration was measured after 24 hours and one week.

Results: All sealants revealed the highest fluoride release on the first day after dipping and the fluoride release sharply decreased after 24 hs and slowly decreased after 3 days. On the first day, Ketac presented the highest fluoride release (37.8 µg/cm^2 ±0.12) followed by Photac (36.4 µg/cm^2 ±0.45), Palfique (15.5 µg/cm^2 ±1.16) and Charisma (8.2 µg/cm^2 ±0.026). Both Ketac and Photac were statistically significant with Palfique and Charisma in all experimental fluoride release periods. After fluoride uptake, Ketac and Photac released the highest fluoride (7.8 µg/cm^2 ±0.52, 7.3 µg/cm^2 ±0.53) respectively after 24 hours followed by Palfique (2.4 µg/cm^2 ±0.11) and Charisma showed the lowest fluoride release (0.37 µg/cm^2 ±0.025).

Conclusions: Within the limitations of this short term study, two glass ionomers were shown to have highest capacity in fluoride release and uptake, followed by Palfique and fluoride releasing composite.

Key words: Fluoride, Fissure sealant, Fluoride release and rechargability.

Department of P.O.P Dentistry, College of Dentistry, Hawler Medical University, Erbil, Iraq.
Corresponding author: Vian Omar
Email: vian.omer@hmu.edu.krd

Introduction

The most common chronic dis-ease of the childhood is dental caries, despite the fact that it is largely preventable.[1] Occlusal surfaces with deep pits and fis-sures have been distinguished as susceptible areas for the initiation of dental caries.[2]

Fissure sealants have been shown to be an effective method for preventing carious defects on occlusal surfaces, because they create a physical barrier against food and bacteria.[3] This preventive effect may be increased by the fluoride-releasing properties that some materials have, because fluorides reduces demineralization and support remineralization of dental hard tissues.[4,5]

Glass ionomer as a sealant has the property of fluoride release which is responsible for anticariogenic action. Glass ionomer based sealants reveals a higher fluoride release than resin based sealants.[6] However, Glass ionomer has weak mechanical properties which are lower to resin based sealants. Resin-based sealants have excellent retention rates but are hard to use in wet environment.[7]

In order to overcome these problems, resin-modified GICs were introduced that also differ from their precursors for their photo-polymerization ability.[8]

In vitro studies have revealed that pit and fissure sealants with fluoride-containing can be recharged by fluoridated products.[9-11] This may be attributed to their long-term effectiveness in caries inhibition.[12] Although interest in fluoride-releasing sealants and their possible anticariogenity appears to have increased, the majority of studies concerning with fluo-ride release and uptake have focused on restor-ative materials with relatively few studies looking at fluoride release and uptake of pit-and-fissure sealants.[10,11]

The fissure sealants have a significant role in prevention of pit and fissure caries, and studies on the fluoride release from different types of fissure sealant materials are required, therefore, this study was done to compare the ability of fluoride release before and after recharging from different types of pit and fissure sealants with different compositions like conventional glass ionomer sealant, modified resin-based glass ionomer sealants, flowable resin sealants and conventional composite resin and their recharge after exposure to 5% fluoride varnish.

Methods

In the present study, forty samples of four different types of fissure sealants were prepared (10 samples for testing fluoride release and recharging), the used materials were: GIC Ketac® Cem radiopaque, 3M ESPE; Photac Fill, 3M ESPE (light-curing, GIC); Palfique (Universal flow, Tokuyama) and Charisma (composite) as shown the following diagram (Figure 1).

Sample preparation for glass ionomer cement (Group 1):

Glass ionomer cement was prepared according to the manufacturer instruction, by using sterile cement spatula and cement slab, one spoonful of powder mixed with two drop of liquid until colloidal gel was resulted, then by using the spatula applied to the metal mold and filled, the material was covered with glass microscopic slide with (1mm) in thickness; then a light pressure was applied by finger to expel excess material from the mold (Figure 2).

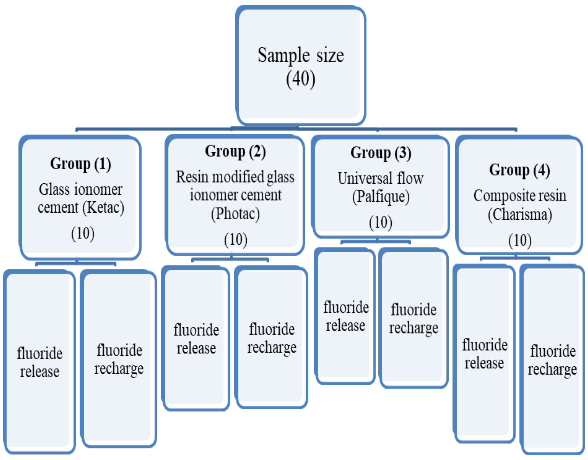

Figure 1. Flowchart illustration of study design

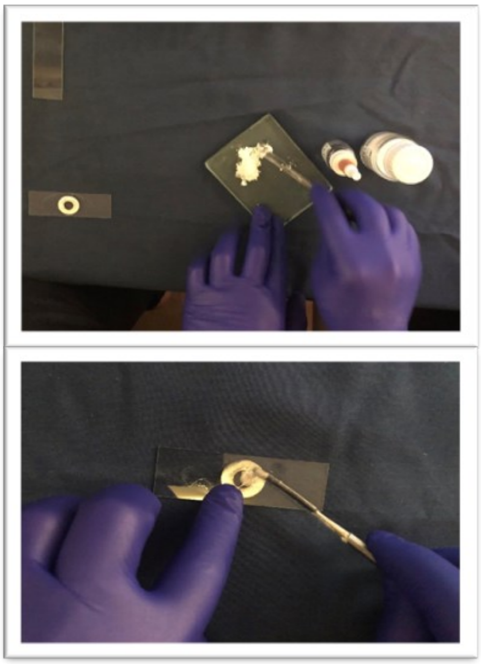

Figure 2. Preparation of conventional glass ionomer cement

Sample preparation for resin modfied glass ionomer cement, flowable sealant and composite resin (Groups 2, 3 and 4):

A stainless steel mold (10 mm in diameter and 1 mm in thickness) was used; it isolated with a thin layer of Vaseline before the application of sealants; a myler matrix was first secure on a glass slide to form the base of the mold, then filled with the materials which were used in this study, the material was covered with another myler matrix strip and glass microscopic slide with (1mm) in thickness; then the light pressure was ap-

plied by finger to expel excess material from the mold. Each specimen was light cured for 40 seconds using light emitting diode (LED) light curing unit (DTE Woodpecker LED, China) with an intensity of 1600-1800 MW/cm² measured with an intensity meter (DTE LED, China),[13] as shown in Figure 3.
Immediately after hardening of the specimen they were removed by hand and cleaned from excess material, the diameter and thickness of the samples were measured by using Vernier caliper.

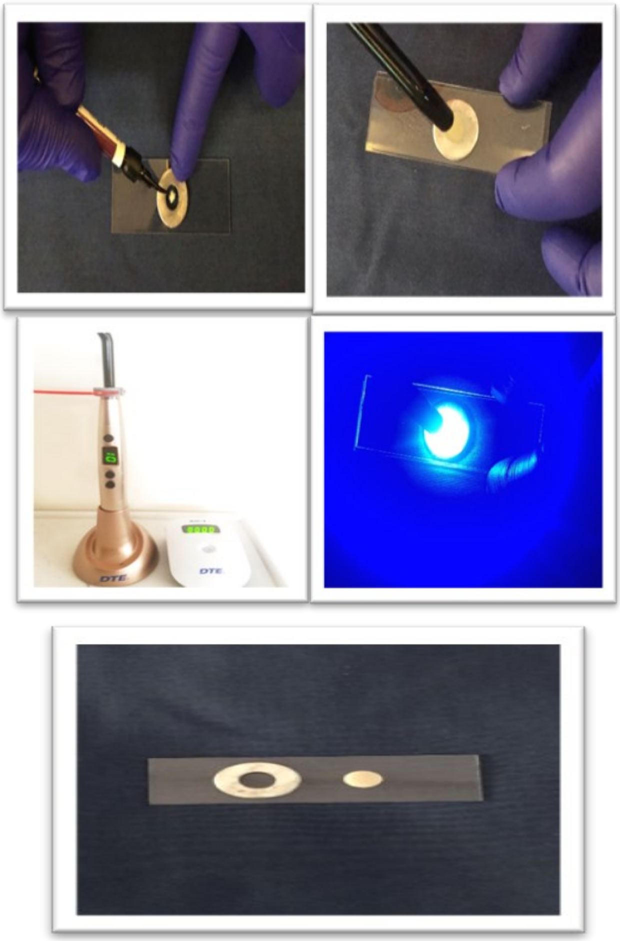

Figure 3. Preparation of the sample

Fluoride release measurement

After the sample were set, they were removed directly from the mold. Before measurement of fluoride release, each sample was placed into plastic polyethylene vial (50 ml) which contain 5 ml deionized water and stored in an incubator at 37°C for 24 hours. The containers were placed obliquely to allow full immersion of the samples in the storage water to minimize contact with the walls as mentioned by Yassin[14] and Ismail.[15] The water in the containers was changed daily for the first week, then storage water was changed one day before measurement at 14 days, 21 days and 28 days to prevent saturation.
After removal of the samples from incubator fluoride measurement was started. At the time of fluoride measurement, each sample was removed from its container by tweezer and the storage water emptied for analysis. The specimens then washed with 1ml of distilled deionized water and dried in a paper towel then they were placed into fresh containers containing 5 ml of DDW for the next measurement.
Reading of each storage sample solution was recorded after adding 0.5 ml of total ionic strength adjusting buffer (TISAB II-Romania) is an acetic buffer solution to provide a background of constant ionic strength for fluoride according to Ghajari[16].
Fluoride release was measured by using a calibrated ion selective electrode attached to an ion meter (Precision ion meter 931, China), as shown in Figure 4. The instrument was calibrated daily and every two hours by a standard fluoride solution, with different concentrations (0.01,0.1,1,10,100 ppm F) were prepared from a 1000 ppm standard solution. Then the electrode was immersed into the storage solution and was gently stirred manually during the analysis for 3 minutes prior to measurement as described by Ismail.[15] After that time record the measurement of fluoride in ppm by ion selective electrode. Between measurements of samples, the electrode was washed by using wash bottle with DDW and blotted dry by paper towel. When the instrument not in use, the electrode will immerse in a standard fluoride solution as instructed by manufacturer. All values of fluoride release ions were calculated in a ppm the readings record from the ppm converted to micrograms per unit surface area (µg/cm^2) by dividing it by the surface area of the sample. Therefore, the results were presented as the rate of fluoride released per unit surface area of the sample per day (µg/cm^2/day). All con-

Figure 4. Fluoride ion meter

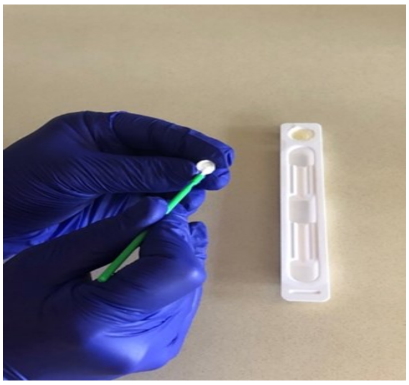

Figure 5. Fluoride recharging of the sample by fluoride varnish

versions were undertaken using Microsoft Excel software 2016

Fluoride recharging

After completion of first part of the study, the specimens were individually rinsed with 1 ml deionized water and allowed to air dry for 1 min. 5% sodium fluoride varnish was applied to the tested sealants using a disposable brush and allowed to dry for 5 minutes according to the manufacturer's instructions (Figure 5). Each specimen was then rinsed three times with 3 ml deionized water, air dried for 1 min and placed in a tube containing 3 ml of deionized water, at 37 °C. Fluoride measurements were carried out at 24hours and 7days The mean and standard deviation values of each experimental group were calculated.

One-way analysis of variance (ANOVA) was used to compare the mean values between materials on each day, multiple comparisons were carried out using LSD test. Level of significance was set at 0.05.

Results

For all the sealants, the greatest amount of fluoride released was on day one. After 24 hours, fluoride released declined rapidly until recharge but continued until the entire test period. At day 1, fluoride release was highest from Ketac followed by Photac, Palfique and Charisma. GIC based sealant released the higher amount of fluoride compared to resin-based sealant.

Table 1 illustrates a statistically significant difference between all mean groups. Mean and standard deviations indicating the fluoride ion release of the sealant materials were determined on the days 1,2,3,4,5,6,7,14,21, and 28. The comparisons of the mean values for measurement days are provided in Table 2.

Higher concentration of fluoride ions release was observed on day 1 for all materials. The Ketac was the sealant material with the highest fluoride ion release on day one (37.8 µg/cm^2), followed by a gradual decrease continued to a constant level of fluoride ion release. Statistically there was no significant difference between Ketac and Photac which recorded second higher fluoride ion release (36.4 µg/cm^2) and both sealants were statistically not significant with Palfique and Charisma which recorded lower fluoride ion release (15.5 µg/cm^2) and (8.2 µg/cm^2) respectively for all measurements.

Mean (±SD) concentration of fluoride release after recharging with fluoride varnish are shown in Table 2. For both one and 7 days again Ketac (15.3 µg/cm^2, 7.8 µg/cm^2) and Photac (14.2 µg/cm^2, 7.3 µg/cm^2) released a higher amount of fluoride than Palfique (6.1 µg/cm^2, 2.4 µg/cm^2) and Charisma (3.2 µg/cm^2, 0.37 µg/cm^2). Furthermore, no significant difference was detected between Palfique and Charisma, while both sealants were statistically differing than Ketac and Photac in the measurements performed on the first and 7th day. In other words, after the first week, a constant fluoride ion release level has been reached, as seen in Table 3.

Table 1. ANOVA test for all mean groups

	Sum of square	df	Mean square	F	Sig.
Between	1307.2	3	435.7	11.3	0.00
Within	1382.2	36	38.3		
Total	2689.5	39			

Table 2. Fluoride release parameters of sealant materials the measures are in µg/cm^2

	Ketac		Photac		Tokoyama		Charisma	
Days	**Mean**	**(±SD)**	**Mean**	**(±SD)**	**Mean**	**(±SD)**	**Mean**	**(±SD)**
1	37.8^A	(±0.12)	36.4^A	(±0.45)	15.5^B	(±1.16)	8.2^B	(±0.026)
2	15.2^A	(±0.26)	14.0^A	(±0.27)	7.1^B	(±0.29)	0.4^C	(±0.02)
3	14.4^A	(±0.25)	13.3^A	(±0.2)	6.5^B	(±0.29)	0.4^B	(±0.023)
4	13.5^A	(±0.22)	12.4^A	(±0.29)	6.1^B	(±0.26)	0.38^B	(±0.017)
5	13.2^A	(±0.24)	12.1^A	(±0.37)	5.8^B	(±0.22)	0.38^B	(±0.019)
6	11.6^A	(±0.45)	10.4^A	(±0.5)	4.9^B	(±0.22)	0.34^B	(±0.018)
7	10.8^A	(±0.42)	10.3^A	(±0.4)	4.5^B	(±0.19)	0.32^B	(±0.016)
14	9.7^A	(±0.45)	9.1^A	(±0.53)	4.0^B	(±0.18)	0.31^B	(±0.017)
21	9.1^A	(±0.43)	8.6^A	(±0.38)	3.8^B	(±0.17)	0.29^B	(±0.017)
28	8.7^A	(±0.46)	8.3^A	(±0.49)	3.5^B	(±0.15)	0.3^B	(±0.017)

The same superscript letter indicates no significant differences in fluoride release in time. ANOVA test indicate significant differences in fluoride release among different materials ($P < 0.05$).

Table 3. Means and mean ranks of Fluoride recharging by type of material after application of fluoride varnish, the measurements are in µg/cm^2

	Ketac		Photac		Tokoyama		Charisma	
Days	**Mean**	**(±SD)**	**Mean**	**(±SD)**	**Mean**	**(±SD)**	**Mean**	**(±SD)**
1	15.3^A	(±1.17)	14.2^A	(+1.42)	6.1^B	(±0.42)	3.2^B	(±0.46)
7	7.8^A	(±0.52)	7.3^A	(±0.53)	2.4^B	(±0.11)	0.37^B	(±0.025)

The same superscript letter indicates no significant differences in fluoride release in time. ANOVA test indicate significant differences in fluoride release among different materials ($P < 0.05$).

Discussion

The fluoride ion release is a complicated mechanism affected by intrinsic and extrinsic factors, such as material solubility, composition, powder-liquid ratio, surface area Studies have shown that the fluoride release occurs rapidly and most of the release occurs in the first two days. In particular, this initial release occurs in the first 24- hour period and called as "burst effect". Subsequently, a high reduction in the amount of fluoride release takes place. Especially after the second week, the daily fluoride release reaches a plateau and no change is observed day by day.[17,18]

Several investigations have been performed on F release from dental materials, as this property is associated to their cariostatic effect.[19] The release of F- from dental materials is controlled by various intrinsic and extrinsic factors. The intrinsic factors include composition, powder/liquid ratio, mixing time, temperature, specimen geometry, permeability, surface treatment and finishing.[20] Temperature, specimen geometry, permeability, surface treatment and finishing were standardized for all materials. However, the composition, powder/ liquid ratio and mixing time vary in according to the studied materials. Extrinsic factors include type of storage medium, experimental design and analytical methods.[21] Fluoride release from glass ionomer cements occurs by means of three mechanisms: surface loss, diffusion through pores and cracks, and bulk diffusion.[22] The highest F- release from the restorative materials studied was seen at the first day and decreased thereafter up to the third day. The high level of F- release on the first day might have been caused by the initial surface loss; while the relatively constant F- release during the following days might be due to the F- ability to diffuse through cement pores and fractures. Bulk F- diffusion occurs during the maturation period as a consequence of the contact of the material with the storage medium.[20]

The present study established the fact that all materials released fluoride. It was likewise established that the amount of released fluoride was greater during the first 24 hours (burst effect), to then decline on the second day, and then gradually decrease with the passing of time.

The initial high level of fluoride release seen in the case of KF and PF is called the "burst effect" of fluoride and is because of rapid release of fluoride from the glass particles as they set. The initial superficial rinsing effect also may be responsible for the initial high level of fluoride release.[23] Later fluoride releases become slower and is because of the gradual dissolution of glass into the hydrogel matrix.[24]

All the fissure sealants evaluated in this study released measurable amounts of fluoride. This observation is consistent with the findings of many other authors.[8] From day one to 28, both Ketac and Photac showed significant higher fluoride release if compared to Palfique and Charisma.

These results are explained by the diffe-rent composition of the tested materials. Ketac fill consists of a Glass-Ionomer Cements with fluoroaminosilicate glass and Photac is fluo-ride-releasing composite sealants with F content.

In GlCs, there is an acid base reaction resulting in the leaching of Ca2+, Al3+ and F- ions to form a polysalt matrix. This may be responsible for the short term elu-tion process. In composites, there is no acid-base reac-tion; the only source of fluoride would come from glass filler particles, resulting in a slow diffusive release. The reason of the rapid decrease of fluoride release during subsequent weeks is likely due to the initial burst of fluoride released from the glass particles as they dissolve in the polyalkeonate acid during the setting reaction. The later slow release occurs as the glass dissol-ves in the acidified water of the hydro gel matrix.[12]

The combination of sealant and topical fluoride application has shown synergistic anti-car-iogenic properties stemming from the recharge ability of fluoride-releasing fissure sealants.[16]

This study analyzed fluoride release and recharge of four different sealants exposed to 5% fluoride varnish. According to Preston et al.[25] the exact mecha-nism of fluoride recharge is unknown.

Material composition, the diffusion of fluoride through the material and differences in surface energy may influence fluoride recharge and subsequent re-lease. The results of the present study are in line with the observation by Xu and Burgess[26] that materials with higher initial fluoride release have higher recharge capacity.

Previous studies have shown that conventional and resin-modified GICs are capable of recharge, whereas resin-based materials are not.[27,28] In the present study, fluoride release from Ketac and Photac was found to increase fol-lowing exposure to fluoride varnish, but to a much lesser extent the other two materi-als.

The ability of a fissure sealants to act as a fluoride reservoir is mainly dependent on the type and permeabi-lity of filling material, on the frequency of fluoride exposure and on the kind and concentration of the fluori-dating agent.[29] Glass-ionomers are mostly found to have significantly better capability to act as a fluoride re-servoir than composite resin-based materials.[9] This fact can be explained by the loosely bound water and the solutes in the porosities in the glass-ionomer, which may be exchanged with an external medium by passi-ve diffusion. The absorption and re-release of fluoride might be determined by the permeability of the material. Thus, a completely permeable substance could absorb the ions deep into its bulk, while a relatively impermea-ble material can only absorb fluoride into the immediate subsurface. In general, materials with higher initial fluoride release have higher recharge capability. Profluorid Varnish is a colophony-based varnish con-taining 5% sodium fluoride (22,600 ppm fluoride). The fluoride ion, together with the calcium ions, causes a pre-cipitation of calcium fluoride.

The recharge agent used was 5% fluoride varnish . Following recharge, there was an increase in fluoride release from all the materials. However, there was rapid fall in fluoride release in subsequent days. After recharge Ketac and Photac released the greatest amount of fluoride compared to other materials. This shows that material with greater release capacity has higher recharge capacity.[26]

Koga et al.[30] showed that conventional resin based sealant lacked the property of recharge. In this study, resin based sealants released fluoride after recharge though in smaller amount compared to glass ionomer based sealant. Shimazu et al.[31] concluded that resin-based sealant containing S-PRG filler had greater recharge capacity than conventional sealants which is in accordance with our study. The exact mechanism of recharge is not known. Many factors influence the recharge capacity like the permeability of the material, the surface energy of the material and composition of the material. Greater the permeability of the material greater is the ability of the material to absorb and re-release fluoride.[25]

There are certain limitations of this study. The use of one type of fluoride with certain concentration and this study was carried out for a shorter duration of time. These limitations should be considered in future studies.

Conclusion

Keeping in mind the limitations of the study certain conclusions can be drawn; fluoride containing pit and fissure sealants released fluoride over a considerable duration. Glass ionomer based sealant with a higher release and recharge capacity can be used in children with high susceptibility to caries. Ketac had the greatest ability for fluoride release and recharge while Charisma being the least.

Conflicts of interest:

The author reports no conflicts of interest.

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