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DÖRT FARKLI FLUORİD SALINIMI YAPAN RESTORATİF MATERYALDEN FLUORİD EMİLİMİNİN SIĞIR MİNESİNDE DEĞERLENDİRİLMESİ: İN VİTRO ÇALIŞMA

EVALUATION OF FLUORIDE UPTAKE FROM FOUR DIFFERENT FLUORIDE-RELEASING RESTORATIVE MATERIALS BY BOVINE ENAMEL: IN VITRO STUDY

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ABSTRACT

Aim: The aim of this study was to compare the fluoride uptake from four different of fluoride releasing restorative materials by bovine enamel and to determine the effect of time on this uptake.
Material and Method: A glass ionomer cement (Fuji II), resin-modified glass ionomer cement(Fuji II LC), giomer (Beautifil) and compomer (Dyract AP) were elected. A total of 120 bovine enamel slabs were prepared from the buccal surfaces of the bovine teeth. The slabs were divided into 5 subgroups (n=6) representing 5 time intervals (1, 7, 14, 21 and 35 days). Fluoride-ion selective electrode method was used to determine the enamel fluoride uptake was determined.
Results: Glass ionomers and giomer had significantly higher fluoride uptake than compomer ($p<0.05$). Glass ionomers and giomer had similar fluoride uptake in all test intervals.
Conclusion: Considering the enhanced physical properties and long-term fluoride release similar to glass ionomers, giomers are found to be promising.
Keywords: Giomer, glass ionomer, fluoride uptake, fluoride-ion selective electrode

ÖZET

Amaç: Bu çalışmanın amacı fluorid salınımı yapan 4 farklı tip restoratif materyalin sığır dişlerinin minesinde flourid emilimini karşılaştırmak ve bu emilime zamanın etkisini belirlemektir.
Gereç ve Yöntem: Çalışmada cam iyonomer siman (Fuji II), rezin modifiye cam iyonomer siman (Fuji II LC), giomer (Beautifil) ve kompomer (Dyract AP) materyalleri kullanıldı. Sığır dişinin minesinin bukkal yüzeylerinden toplam120 adet örnek hazırlandı. Örnekler her bir materyal grubu için 5 zaman aralığını (1, 7, 14, 21 ve 35 gün) temsil eden 5 alt gruba (n =6) ayrıldı. Fluorid emilimini belirlemek icin florür iyonu seçici elektrot yöntemi kullanıldı.
Bulgular: Cam iyonomerler ve giomer kompomere göre ($p <0.05$) anlamlı olarak daha yüksek fluorid emilimi gösterdi. Cam iyonomerler ve giomerin tüm test aralıklarında benzer fluorid emilimi gösterdiği belirlendi.
Sonuç: Giomerlerin gelişmiş fiziksel özellikleri ve cam iyonomerlere benzer uzun süreli fluorid salınımı göz önüne alındığında çocuk diş hekimliği için uygun bir materyal olduğu bulgulanmıştır.
Anahtar Kelimeler: Giomer, cam iyonomer, fluorid alımı, florür iyonu seçiçi elektrot

INTRODUCTION

Fluoride has been well-documented as an anticariogenic agent, and with the observation of inhibiting secondary caries formation, particular attention has been focused on the development of various fluoride-releasing restorative materials[1-6].

A variety of mechanisms are involved in the anticariogenic effects of fluoride, including the reduction of demineralization, the enhancement of remineralization, inhibition of acid production in plaque and the inhibition of microbial growth and metabolism[5,7,8]. Therefore, fluoride released from dental restorative materials is assumed to affect caries formation through all these mechanisms; thus reduce or prevent demineralization, promote remineralization of dental hard tissues and inhibit secondary caries[6,8].

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Conventional glass ionomer cements (GIC) possess certain unique properties that make them useful as restorative and adhesive materials, including adhesion to tooth structure, anticariogenic properties due to release of fluoride, thermal compatibility with tooth enamel, and biocompatibility[1,2]. On the other hand, to overcome the problems such as moisture sensitivity and low initial mechanical strengths typical for GIC, resin modified glass ionomer (RMGIC) and polyacid-modified composite resin (compomer) were developed. RMGICs have a true acid-base reaction on mixing[9], free radical polimerization and similar fluoride release and rechargeability to GICs. In compomers the acid-base reaction is quite limited, it only occurs after contacting with water. It should be noted that compomers behave primarily like composite resins and do not release fluoride to the same extent as do GICs or RMGICs[4-6,10].

Recently, a new category of hybrid material (giomers) was introduced. Giomers include pre-reacted glass-ionomers to form a stable phase of glass-ionomer fillers in the restorative. Unlike compomers, fluoro-alumino-silicate glass particles react with polyacrylic acid prior to inclusion into the resin matrix. Unlike GICs and RMGICs, both giomers and compomers require a resin bonding system after acid etching[4,10-14].

A clear understanding of the mechanisms involved in the anticariogenic effects of fluoride and determining which materials have optimal fluoride release for caries resistance would ensure appropriate restorative material selection at the clinical setting[1,7,8]. With this aim, the relative concentrations and the duration of fluoride release in addition to fluoride uptake have been the interest of research. In general, it has been suggested that decreased physical properties are associated with increased fluoride release[4-6]. Therefore, research into the development of fluoride-releasing materials is ongoing with the hope of maintaining the physical properties of these materials and providing long-term fluoride release and subsequently uptake by dental tissues.

The aim of this in vitro study was to compare the fluoride uptake from four different fluoride releasing restorative materials by bovine enamel and to determine the effect of time on this uptake.

MATERIALS AND METHODS

A GIC (Fuji II), RMGIC (Fuji II LC), giomer (Beautifil) and compomer (Dyract AP) were evaluated. 30 cylindrical discs were prepared from each tested material utilizing a teflon split mold (2 mm in diameter and 1 mm in thickness). The mold was slightly over-filled with material, covered with Mylar matrix strip (Yates and Bird/Motloid, Chicago) and pressed flat between two glass slides. Then the specimens were polymerized with a light-curing unit (LED LCU, Elipar Freelight, 3M ESPE, Germany) for the time suggested by the manufacturer except for Fuji II. The light output was tested using a radiometer.

Bovine permanent incisor teeth obtained from 45- to 48-month-old cattle were obtained and stored frozen until use. All cattle were born and lived in the same area. Only specimens without carious lesions or other defects on the buccal surface were used in the present study. Prior to specimen preparation, teeth were left at room temperature for approximately 3 h. Then, the teeth were rinsed thoroughly with water, and the buccal surfaces were lightly cleaned with a rubber cup and pumice, washed, dried, and swabbed with a cotton pellet soaked in acetone to remove residual organic debris.

A total of 120 bovine enamel slabs were prepared and cut with a diamond saw (IsoMet, Buehler, Lake Bluff, IL, USA) under water spray from the buccal surfaces of the bovine teeth, and assigned to one of four tested materials. Each of these groups contained 30 enamel slabs that belonged to one bovine jaw in order to ensure the same initial fluoride enamel content. Five slabs of each group were used as controls to determine the baseline fluoride concentration. The remaining slabs were divided into 5 subgroups (n=6) representing 5 time intervals (1, 7, 14, 21 and 35 days).

The slabs were attached with sticky wax to a plastic post with the enamel surfaces facing upwards. The enamel surface was covered with a sticky teflon paper (0.08 mm thick, Unigasket, Sarnico, Italy) with a circular hole (1.5 mm diameter) in the middle in order to limit the area for enamel uptake. The remaining slab surfaces were covered with nail varnish. For each slab, a disc of the tested material was transferred on the teflon paper and attached with sticky wax without sealing off the whole circumference. The tooth slabs with the attached disc materials were then suspended in polystyrene tubes of 10 mL (300900, Eurotubo Deltalab, Barcelona, Spain) containing 2 mL of synthetic saliva (3 m*I* calcium, 1.8 m*I* phosphorus, 150 m*I* sodium chloride, and 1% carboxymethylcellulose (CMcellulose)) adjusted to pH 7.0 with sodium hydroxide according to the time period. The tubes were covered with laboratory film (Parafilm M, American National Can, Chicago, IL, USA). The solutions were kept unstirred and renewed every 2 days. At the tested time intervals, the specimens were removed from the synthetic saliva and dried with compressed air. The specimens were only used once to avoid contamination. The disc of the material was removed, and the integrity of the seal was evaluated by depositing 0.4 µ L of distilled water on the demarcated enamel with a 1-µL microsyringe (Hamilton,Basel, Switzerland). The disappearance of the drop of water from the demarcated biopsy site indicated a defective marginal seal. Those specimens were discarded.

After removal, specimens were etched with perchloric acid and the dissolved enamel was analyzed in terms of fluoride and calcium. The acid etch biopsy technique was used to determine the enamel fluoride uptake and calcium was determined via atomic absorption spectrophotometry[1]. The amount of fluoride in the samples was determined by direct potensiometric analyses with the use of a combination fluoride-ion selective electrode (Orion combination fluoride ionalyser 96-09-00, Orion research, Cambridge, USA). The results were statistically analyzed by ANOVA, Scheffe's and Dunnett's T3 tests.

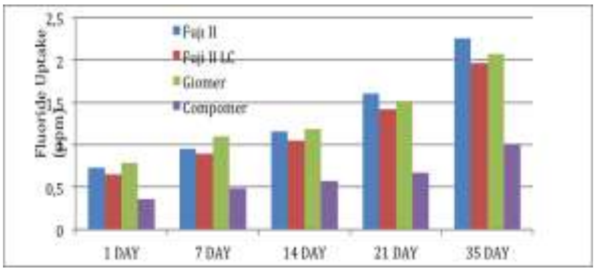
RESULTS

In GIC group, the F uptake was 0.73 mgF/cm^3 at day 1, 0.95 at day 7 and reached 2.26 at day 35 whereas that of RMGIC group was 0.65, 0.89 and 1.96 mgF/cm^3 respectively. For giomer, the level of F uptake was 0.79 (day 1), 1.05 (day 7) and reached 2.07 mgF/cm^3 (day 35) while compomer was 0.38, 0.49 and 1.02 mgF/cm^3 respectively in Table 1 and Graph 1. Significantly higher levels of fluoride uptake were observed with both glass ionomers and giomer whereas the least amount of fluoride uptake was found with compomer in all test intervals ($p<0.05$) (Table 1). No significant difference was found in the amount of fluoride uptake between two glass ionomers and giomer ($p>0.05$)(Table 1).

Table 1: Fluoride-ion selective electrode test results (mgF\cm^3) mean and standard deviation (SD) .

Day	Fuji II	Fuji II LC	Beautifil	Dyract AP
1	0.73 (0.38)	0.65 (0.44)	0.79 (0.22)	0.36 (0.09)*
7	0.95 (0.26)	0.89 (0.15)	1.10 (0.16)	0.48 (0.06)*
14	1.16 (0.30)	1.05 (0.07)	1.19 (0.12)	0.57 (0.06)*
21	1.60 (0.25)	1.42 (0.08)	1.52 (0.15)	0.67 (0.05)*
35	2.26 (0.33)	1.96 (0.08)	2.07 (0.18)	1.01 (0.08)*

**Statistically significant difference p <0.05 was determined*



Graph 1: Fluoride uptake according to the distribution of material group to days.

DISCUSSION

Fluoride release is biologically important, because it has cariostatic effect. It is known that fluoride is taken up by adjacent tooth substrate, presumably by the ion exchange of F^- for OH^- in hydroxyapatite to form fluoroapatite, therefore strengthens dentin[6,8,11]. For this reason, manufacturers investigated the possibility of adding fluoride to various restorative materials in order to increase their cariostatic properties[2]. In an effort to develop fluoride-releasing restorative materials GIC, RMGI and compomers were introduced and all of them have different setting mechanisms. The RMGICs are set by an acid-base reaction and free radical polymerization mechanism[9]. The compomers set by free radical polymerization only with limited acid-base reaction occurring later as the material absorbs water from the oral environment. Fluoride release from RMGICs is known to be similar to that of conventional GICs, whereas compomers produce a low and relatively constant fluoride release[1]. The pattern of fluoride release from both GIC and compomers is characterized by an initial rapid release followed by a rapid reduction in the amount of release after a short immersion period[6,8].

Considering the fact that the fluoride-releasing mechanism of GIC was derived from its acid-base reaction phase, Roberts et al.[12] developed a newly revolutionary pre-reacted glass-ionomer (PRG) filler technology. The PRG fillers were prepared by the acid-base reaction between fluoroaluminosilicate glass and polyacrylic acid in the presence of water to form a wet siliceous hydrogel. This new technology has proposed a new category coined as "Giomer" and described as fluoride-releasing, resin based dental materials that comprise PRG fillers[12,13]. Since this technology has been developed the authors have suggested that the use of PRG fillers promoted rapid fluoride release through a ligand exchange within the prereacted hydrogel. For giomer the hydrogel layer of the glass filler is more extensive than that in compomers. Therefore a more extensive acid-base reaction has been carried out before blending with resin. As a result of this information, it is expected that giomer will demonstrate more effective fluoride releasing characteristics than the other resin-matrix materials. Itota et al.[14] reported that fluoride release occurs subsequent to water uptake, either as a result of dissolution of the glass filler particles or via the later generation of ionic reaction on the surface of the glass particles. Hence, in addition to the good fluoride-release function, another potential benefit is their fluoride recharging capability[4,12]. Relevantly, it is known that the amount of fluoride release from restorative materials provides a potential fluoride reserve and therefore a source of fluoride recharge in oral environment. Therefore, the aim of this study was to determine amount of fluoride uptake by bovine enamel from Giomer, GIC, Compomer and RMGIC during the first day and at days 7, 14, 21 and 35.

In this study, an acid etch method was used to determine the bovine enamel fluoride uptake from four fluoride-containing materials. Duckworth and Lynch[15] revealed that the acid etch method is in good agreement with the abrasion technique. On the other hand, Dijkman and Arends[16] demonstrated that an underestimate of the results could occur by acid etching method if the fluoride in the fluid phase is transferred into deeper enamel layers, as a consequence of the acid penetrating between the enamel prisms faster than the prisms themselves get dissolved. However, selective dissolution at prism boundaries is more likely to occur during the acid etching process.

Bovine enamel is more porous than human enamel and in agreement with Ahiropolous et al.[1] this might lead to greater fluoride uptake from the former compared with the latter; this possibly represents a limitation of our study. Otherwise, human tooth would vary extensively according to either the age of the individual or the formation of the enamel. Although the bovine teeth used in our study are not applicable for a specific human cavity, they offer a standardized model by providing large, flat test surfaces with low fluoride content and comparable anatomy and calcification[17]. In accordance with the other studies, Ahiropoulos et al.[1] revealed that human teeth provide small surfaces with differences in fluoride content among all groups of teeth. In the light of above knowledge, in the present study, to have relatively large enamel surfaces all from one jaw and reassure the baseline of fluoride content we chose bovine enamel.

Regarding materials, there were no significant differences between fluoride uptake from Giomer, GIC and RMGIC at all tested days. When the amount of initial fluoride uptake is compared among the groups, the order was Giomer > GIC >RMGIC >Compomer. This finding is different from the findings of Yap et al.[18] who evaluated the same materials for the release of fluoride and revealed that the glass-ionomers released significantly more fluoride than the giomer and compomer at day one. Furthermore, researchers reported that giomer is surrounded with resin matrix and had a lower porosity. Accordingly, the porosity of this kind of material is lower than GICs, fluoride release was not expected to be as much as the GIC[18,19]. In the present study, the order of initial fluoride uptake can be explained by the nature of the fluoridated glass incorporated into each material and the level of glass ionomer matrix layer surrounds the glass filler in the set material. According to Tay et al.[20] Giomer contains a fluoridated glass filler within glass-ionomer glass matrix layer. But giomer has thicker hydrogel layer due to the prior reaction of asid and glass filler before incorporation in the resin matrix.

Furthermore, Ikemura et al.[21] revealed that this glass-ionomer matrix contains much complexed fluoride and is easily penetrated by water resulting in a significant greater fluoride release from the material. In this study, the highest fluoride uptake by bovine enamel was observed from Giomer at the first day evaluation as well as at days 7 and 14. Similarly, this study confirms the previous observations made by Itota et al.[14] and Dhull and Nandlal[10] that the extent of hydrogel matrix of glass filler incorporated into the materials affected the amount of fluoride release at the initial period.

The present study revealed significant differences in the mean daily fluoride uptake from compomer and the other materials. Furthermore, compomer which gave the lowest fluoride uptake levels among all days, contains strontium-fluoro-silicate glass filler in which a thin layer of glass-ionomer matrix has been formed on the surfaces of the glass particles by reaction of glass with acid which is present in the matrix. This result in accordance with Gao et al.[22] indicates that the extent of glass-ionomer matrix layer in compomer was insufficient to promote enhanced fluoride release. Similarly, many researchers explained that GIC showed greater fluoride release as compared to resin composite or compomer[5,6,23,24]. On the other hand, it has been shown that fluoride uptake from GIC can prevent demineralization of enamel.[1] The considerable amount of fluoride uptake from both GIC and RMGIC was in agreement with the results of previous investigations[1,23,24]. In the present study, the degree of fluoride uptake by bovine enamel was similar with conventional, RMGIC and Giomer. Although there were no significant differences among glass-ionomers for all of the evaluated days, the fluoride uptake from giomer and conventional GIC had the highest values. This is probably because these materials have well-established glass-ionomer matrix around the glass filler particles. Additionally, Xu and Burgess[25] suggested that the material with higher initial fluoride release has a higher fluoride-recharging capability. Also the fluoride release from material over several days might be dependent on the initial fluoride release. Although the fluoride uptake was initially higher from Giomer than the other materials, it declined after the 14th day, and the amount of fluoride uptake from giomer was lower than that from GIC at days 21 and 35. Eliades et al.[26] found that net peak absorbance area ratio of the carboxylate salts formed to the unionized carboxyls (which reflects the extent of the acid-base reaction) and fluoride release of materials reached a plateau after 14 days. They suggested that salt formation stabilized the structure from where fluoride ions were initially released due to dissolution. This may explain the higher fluoride uptake from giomer till day 14 compared to days 21 and 35.

CONCLUSIONS

It is concluded that fluoride-releasing restorative materials have influence on enamel fluoride uptake with during different time intervals. Considering the enhanced physical properties and long-term fluoride release similar to GICs, giomers are found to be promising in promoting caries resistance. Further in vivo and in vitro studies are required to verify these preferable effects, particularly regarding secondary caries inhibition.

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