Changes of the Mineral Structure in the Enamel Adjacent to Three Types of Restorative Materials after Immersion in Hydrochloric Acid

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The aim of this study was to assess the changes of the mineral structure in the enamel adjacent to three restorative materials, after the immersion in 0.06 mol/L hydrochloric acid (pH = 1.2) for 30 min and 60 min. The study group included 15 molars, extracted from periodontal reasons, divided in three groups. Class I cavities were prepared and filled with three types of restorative materials: Fuji IX (GC, Japan), Fuji II LC (GC, Japan), Compomer Freedom (SDI limited, Australia). The teeth were sectioned longitudinally in buccal-oral and mesio-distal plans. Three pieces of each tooth were used as samples and were immersed in distilled water, in hydrochloric acid (0.06mol/L, pH = 1.2), for 30 and 60 min. The morphologic structure of dental sections was analyzed in SEM microscopy. The mineral content (calcium, phosphorus, oxygen ions) in the interface tooth/material area, were evaluated using EDX method. The results were analyzed using ANOVA and Bonferroni statistical tests ($p \square 0.05$). The mean calcium, phosphorus and oxygen ions content (wt%) significantly varied between the samples immersed in hydrochloric acid (0.06mol/L, pH = 1.2) after 30 min and 60 min, and samples immersed in distilled water (control sample: 50.4373 for calcium; 23.0260 for phosphorus; 26.2673 for oxygen; 30 min immersion: 26.8333 for calcium, 10.8467 for phosphorus, 58.9587 for oxygen; after 60 min: 2.2420 for calcium, 1.4967 for phosphorus, 76.1913 for oxygen). The mineral structure of enamel at the interface with the tested materials undergoes significant changes, comparing with the control group, after immersion of teeth in hydrochloric acid for 30 and 60 min.

Keywords: GERD, SEM, EDX, erosion, glassionomer cements

Erosion is an irreversible process, characterized by the chemical removal of minerals from the tooth surfaces by acids from either extrinsic or intrinsic sources without bacterial involvement [1].

Gastroesophageal reflux (GERD) is a condition defined as an involuntary passage of gastric fluid against the normal flow of digestive tract [2]. It can be a normal phenomenon in newborns and usually disappears with age; however, in some individuals, its maintenance can be considered a pathological condition [3]. The abnormal exposure of digestive tract to the stomach contents leads to troublesome symptoms and complications caused by the high concentration of H⁺ and action of pepsin, a proteolytic enzyme [4, 5].

One of the most common complications is dental erosion as gastric reflux has a pH of less than 2.0 [6], which is low enough to quickly demineralizes the enamel.

Dental erosion does not only affect enamel. When reaching dentine it cause hypersensitivity, or in severe cases, pulp exposure and even tooth fracture. Clinical performance of restorative materials is affected by erosion as well. Several studies reported that acidic condition degraded glassionomer cements, polyacid modified resin composites, and restorative composite. In the oral environment, saliva modifies the erosive process. Individuals with low or diminished salivary flow are more susceptible to erosive lesions [7].

The aim of study was to assess the changes of mineral structure of the enamel adjacent to restoration in case of

three types of restorative materials, after immersion in 0,06 mol/L hydrochloric acid (pH = 1.2) for 30 min and one hour.

Experimental part

Materials and methods

The study group included 15 molars extracted from periodontal reasons. The teeth were divided in three groups, each group including 5 teeth. After extraction the teeth were stored in 10% formol solution. The organic and inorganic materials were removed using hand instruments and immersion in NaClO 5.25%.

Class I cavities were prepared in the occlusal surface of each tooth, with standard dimensions. These cavities were filled with three types of restorative materials: Fuji IX (GC, Japan), Fuji II LC (GC, Japan), Compomer Freedom (SDI limited, Australia). The samples were sectioned longitudinally in buccal-oral and mesio-distal plans, using active metallic discs. One portion of the crown was immersed in distilled water, and the other two in hydrochloric acid (0.06mol/L, pH = 1.2), for 30 min and respectively for 60 min . Each sample was processed by finishing and polishing with paper discs with decreasing granulation. The samples were fixed on resin plates.

The morphologic structure of dental sections was analyzed in SEM microscopy (SEM model VEGA II LSH, TESCAN, connected to an EDX detector, QUANTAX QX2, BRUKER/ROENTEC). The mineral ions content (calcium, phosphor, oxygen) in the enamel adjacent to each

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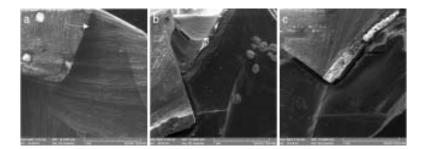


Fig. 1. Images SEM at the interface compomer Freedom-enamel: a – Control, b – after 30 min, c – after 60 min

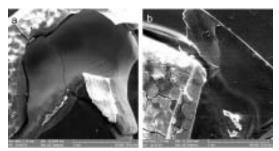


Fig. 2. Images SEM at the interface glassionomer FUJI II-enamel: a - 30 min, b - 60 min

restorative material, were analyzed using ANOVA and post hoc Bonferroni statistical tests.

Results and discussions

SEM aspects showed important changes regarding the morphology of both enamel and restoration surface for the samples immersed in hydrochloric acid for 30 and 60 nin (comparing with control samples). These samples presented fissures and porous structures as well as the widening of space between the restoration and the cavity walls. The alterations were more severe for the samples immersed for 60 min in acid solution, comparing with the samples immersed in acid solution for 30 min.

For the compomer material, the SEM images have revealed the development of micro-fissures and micropores in the structure of enamel adjacent to the restorative material. A gap was observed between the restorative material and the dental tissue (fig. 1b and c) compared with the reference sample (fig. 1a).

The resin-modified glassionomer had a porous appearance and presented numerous fissure. Also the enamel area presented some fissures, a porous structure and the widening of the spaces between the enamel prisms (fig. 2a and b).

The chemically cured glassionomer also presented a porous structure, roughness and fissures. A gap was also

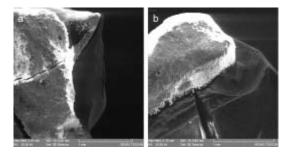


Fig. 3. Images SEM at the interface glassionomer FUJI IX-enamel: $a - 30 \min b - 60 \min$

present at the interface between filling and cavity walls (fig. 3a and b).

In table 1 are presented the mean values for calcium, phosphorus and oxygen for control groups.

In order to find out if there are significant statistical differences, it was performed an ANOVA test. The null hypothes was: between the three study groups of compomers there are no significant statistical differences. The research hypothesis was: between the three study groups of compomers there are significant statistical differences.

The mean concentration of calcium, phosphorus and oxygen ions significantly varies for all three groups (p = 0.0001). Post hoc Bonfereroni statistical test showed significant statistical differences between the mean concentrations of calcium and phosphate ions concentrations from all the three study groups control (table 2, p \land 0.05). No significant statistically differences were obtained when compared the mean oxygen ion concentration from compomer samples (Freedom) and Fuji II glassionomer cement samples (p = 0.251 > 0.05) (table 2).

In table 3 the mean ions content for calcium, phosphorus and oxygen are different between the three groups immersed for 30 min in hydrochloride acid.

Element	Restorative	No	Mean	Std. Dev.
	material	Samples	(wt %)	
	Freedom control	5	50.9040	0.05459
Ca	Fuji II control	5	53.5480	0.08438
Ca	Fuji IX control	5	46.8600	0.07280
	Total	15	50.4373	2.84753
	Freedom control	5	23.8480	0.02280
р	Fuji II control	5	20.1400	0.03742
1	Fuji IX control	5	53.5480 0. 46.8600 0. 50.4373 2. 23.8480 0. 20.1400 0. 25.0900 0. 23.0260 2. 25.2460 0. 25.5100 0.	0.05612
	Total	15	23.0260	2.17689
	Freedom control	5	25.2460	0.35501
0	Fuji II control	5	25.5100	0.12884
	Fuji IX control	5	28.0460	0.06656
	Total	15	26.2673	1.32260

 Table 1

 COMPOSITION OF THE ENAMEL MINERALS ADJACENT TO RESTORATIVE MATERIAL (CONTROL)

Table 2
BONFERRONI STATISTICAL TEST RESULTS (CONTROL GROUPS)

			Freedom			Fuji II			Fuji IX		
		Ca	Р	0	Ca	Р	0	Ca	P	0	
Freedom	Ca	-	-	-	0.000	-	-	0.000	-	-	
	P	-	-	-	-	0.000	-	-	0.000	-	
	0.	-	-	-	-	-	0.251	-	-	0.000	
Fuji II	Ca	0.000	-	-	-	-	-	0.000	-	-	
	Р	-	0.000	-	-	-	-	-	0.000	-	
	0	-	-	0.251	-	-	-	-	-	0.000	
Fuji IX	Ca	0.000	-	-	0.000	-	-	-	-	-	
	Р	-	0.000	-	-	0.000	-	-	-	-	
	0	-	-	0.000	-	-	0.000	-	-	-	

Table	3

COMPOSITION OF THE ENAMEL MINERALS ADJACENT TO RESTORATIVE MATERIAL (IMMERSION FOR 30 MINUTES)

Element	Restorative	No.	Mean	Std. Dev.	
	material	Samples	(wt %)		
Ca	Compomer 30min	5	3.6860	0.11971	
	Fuji II 30min	5	21.3460	0.06731	
	Fuji IX 30min	5	55.4680	0.13461	
	Total	15	26.8333	22.24766	
Р	Compomer 30min	5	1.0200	0.04359	
	Fuji II 30min	5	3.9200	0.08746	
	Fuji IX 30min	5	27.6000	0.15652	
	Total	15	10.8467	12.32366	
0	Compomer 30min	5	86.4700	0.23409	
	Fuji II 30min	5	74.4480	0.13103	
	Fuji IX 30min	5	15.9580	0.09524	
	Total	15	58.9587	31.88100	

The mean concentration of calcium, phosphorus and oxygen ions significantly varies for all three groups (p = 0.0001). Post hoc Bonferroni statistical test showed significant statistical differences between the mean concentrations of calcium and phosphate ions concentrations from all the three study groups, after 30 min immersed in hydrochloride acid (table 4, $p \square 0.05$).

In table 5 it can be noticed that the mean ions content from calcium, phosphorus and oxygen were different between the three groups after storing the samples for an hour in erosive solution.

The mean concentration of calcium, phosphorus and oxygen ions significantly varies for all three groups (p = 0.0001). Post hoc Bonferroni statistical test showed significant statistical differences between the mean concentrations of calcium and phosphate ions

concentrations from all the three study groups, after 60 min immersed in hydrochloride acid (table 6, p \Box 0.05). No significant statistical differences were obtained when compared the mean phosphate ion concentration from compomer samples (Freedom) and Fuji IX glassionomer cement samples (p = 0.623 > 0.05)

The glass-ionomer cements consist of a mixture of liquid and powder. The powder consists of particles of aluminum silicate glass, containing calcium CaF, NaF, AlF and P. The liquid contains poliacrylic acid copolymerized with maleic acid and itaconic acid. The setting reaction is an acid-base reaction. The polycarboxylic acid attacks the superficial layer of aluminum silicate glass, releasing F, Ca, Al, Si and ions. The Ca and F ions participate to the formation of soluble calcium polycarboxylate. Later, Al and F ions will replace Ca ions, allowing the formation of aluminum

			Freedom			Fuji II			Fuji IX		
		Ca	Р	0	Ca	Р	0	Ca	Р	0	
Freedom	Ca	-	-	-	0.000	-	-	0.000	-	-	
	P	-	-		•	0.000	-		0.000	•	
	0	-	-	-	-	-	0.000		-	0.000	
Fuji II	Ca	0.000	-	-	-	-	-	0.000	-	-	
	Р	-	0.000	-	-	-	-	-	0.000	-	
	0	-	-	0.000	-	-	•	-	-	0.000	
Fuji IX	Ca	0.000	-	-	0.000	-	-	-	-	-	
	Р	-	0.000		-	0.000	-		-	•	
	0	-	-	0.000	-	-	0.000	-	-	-	

	Table 4
BONFERRONI STATISTICAL TEST RESULTS ((GROUPS AFTER 30 MINUTES IMMERSED IN HYDROCHLORIDE ACID)

Table 5

COMPOSITION OF THE ENAMEL MINERALS ADJACENT TO RESTORATIVE MATERIAL (IMMERSION FOR 1 HOUR)

Element	Restorative	No.	Mean	Std. Dev.
	material	Samples	(wt %)	
Ca	Compomer 1 hour	. 5	2.1500	0.06124
	Fuji II 1 hour	5	1.8620	0.04970
	Fuji IX 1 hour	5	2.7140	0.14876
	Total	15	2.2420	0.37717
P	Compomer 1 hour	5	1.8620	0.12518
	Fuji II 1 hour	5	0.8500	0.04743
	Fuji IX 1 hour	5	1.7780	0.10918
	Total	15	1.4967	0.48354
0	Compomer 1 hour	5	76.6000	0.47534
	Fuji II 1 hour	5	77.8320	0.09680
	Fuji IX 1 hour	5	74.1420	0.11278
	Total	15	76.1913	1.60990

Table 6

BONFERRONI STATISTICAL TEST RESULTS (GROUPS AFTER 60 MINUTES IMMERSED IN HYDROCHLORIDE ACID)

		Freedom			Fuji II			Fuji IX		
		Ca	Р	0	Ca	Р	0	Ca	Р	0
Freedom	Ca	-	-	-	0.002	-	-	0.000	-	-
	Р	-	-	-	-	0.000	-	-	0.623	
	0	-	-	-	-	-	0.000	-	-	0.000
Fuji II	Ca	0.002	-	-	-	-	-	0.000	-	-
	Р	-	0.000	-	-	-	-	-	0.000	-
	0	-	-	0.000	-	•	-	-	-	0.000
Fuji IX	Ca	0.000	-	-	0.000	-	-		-	-
	Р	-	0.623	-	-	0.000	-	-	-	-
	0	-	-	0.000	-	-	0.000	-	-	-

polycarboxylate. The release of fluoride ions is due to the reaction of alumina-fluoride-silicate glass interacting with polyalkenoic acid. The glassionomer cements are vulnerable to acid attack, because of their inhomogeneous composition and porous structure [8, 9].

Resin-modified glassionomers were developed in order to compensate some deficiencies of traditional glassionomers cements. These materials contain also an additional hydrophilic resine as well as a photoactivating component.

The compomers represent a new class of hybrid restorative materials that combine the qualities of composite resins with those of glassionomer cements. The composition and setting reaction resemble to composite resins, providing an increased stability and lower fluoriderelease comparing to glassionomer cement.

Our study aimed to assess the resistance to acid corrosion of enamel at the interface cavity/restorative materials in the clinical situations of erosive lesions treated with three types of restorative treatment: a chemical-cured glassionomer, a resin-modified glassionomer and a compomer.

Dental erosion is partly a natural process, which occurs over a lifetime as a consequence of chewing and food consumption. Based on changes in consumer habits, especially due to the increased availability of new acidic food products and intensive marketing, intensified oral hygiene measures as well as chewing habits (with antagonistic teeth) and bruxism, the processes of dental erosion can be considerably enhanced [10-12]. For this process, the dissolution of the dental hard tissue by means of acidic substances is primarily held responsible, which can be caused on the one hand by extrinsic factors like acidic food and drinks, mouth rinses, but also by certain types of medication, and on the other hand it can also be caused by intrinsic processes like frequent vomiting leading to contact with gastric acid [13-15].

Silent GERD exists when the typical symptoms of belching, sour taste, or heartburn do not exist [16]. Dentists may be the first to consider the possibility of GERD based on the patterns of erosion [17]. Up to 20% of individuals with GERD may suffer from dental erosion. Patients with low unstimulated salivary flow are estimated to be at five times greater risk of having erosion than those with a normal flow rate [18].

There are many unfavorable consequences to dental erosion, including sensitivity and compromised esthetics. Restoration of lesions can be effective in solving many of these problems.

SEM analysis appears to be an efficient and acceptable method of examining such features as surface topography, filler size and distribution, interface adhesion and porosity [18, 19].

In our study, SEM analysis showed evidence of differences in surface morphology of both restorative material and adjacent enamel. EDX analyze showed that significant changes mineral composition are associated to surface alterations.

The values of calcium, phosphorus and oxygen ions content (wt%) after immersion in hydrochloric acid (0.06 mol/L, pH = 1,2) were higher in samples immersed 30 min comparing with control (control: 50.4373 for calcium; 23.0260 for phosphorus; 26.2673 for oxygen; samples after immersion 30 min: 26.8333 for calcium, 10.8467 for

fosforus, 58.9587 for oxygen). The values of calcium, phosphorus and oxygen after immersion in hydrochloric acid (0.06 mol/L, pH = 1,2) were lower in samples immersed 1 hour comparing with control (control: 50.4373 for calcium; 23.0260 for phosphorus; 26.2673 for oxygen; samples after immersion 60 min: 2.2420 for calcium, 1.4967 for phosphorus, 76.1913 for oxygen). The immersion for 30 min is associated with the release of calcium, phosphorus and oxygen ions, both from dental structures and glassionomer cements. After the immersion time increases, the mineral level decreases significantly.

All the tested materials release fluoride ions, but the quantity and pattern is different. The use of glassionomer cements is recommended for the treatment of carious lesions due to the fluoride ions release [20].

P.N. Pereira et al. (1998) showed the presence of inhibition areas in dental caries both in carious lesions treated with traditional and resin-modified glassionomer cements [21-28]. Their study concluded that traditional glassionomer cements have a superior ability to prevent artificial carious lesions at the interface tooth/restorative materials, comparing with resin-modified glassionomer Fuji II LC.

Despite the ability of counteracting demineralization in dental caries, this effect of glass-ionomer cements is questionable in erosive lesions [28]. The mechanism of fluoride release supposes a chemical structure with less resistance to corrosion. When low *p*H is maintained for long time (erosive lesions produced by *gastroesophageal reflux disease*), the corrosion of restorative materials would be significant and could conduct to porous structure, adhesion loss and low mechanical resistance [29, 30]. These deficiencies are higher for traditional glassionomer cements comparing with resin-modified glassionomer cements. Thus is doubtful if these disadvantages could be compensating by the remineralising effect on adjacent hard dental tissues [31].

Many studies suggest that glassionomer cements are the most adequate materials for the restoration of these types of lesions [32, 33]. Our results show as unlikely the effect to stop or slow down the demineralization of adjacent enamel for all three tested materials. Despite the initial release of calcium and phosphorus ions from glassionomer cements, after the increasing of immersion time, the values of minerals decrease significantly both at the level of dental tissues and materials.

Further studies are requested to assess the long term effects of the fluoride-releasing materials used for the restorative treatment of erosive lesions.

However, it seems most likely that without eliminating the cause of the erosion, the destructive process will continue, no matter what material would be used.

Conclusions

The mineral structure of the enamel adjacent to the tested restorative materials significantly changed, comparing with the control group, after the immersion of teeth in hydrochloric acid (0.06mol/L, pH = 1.2), for 30 min and respectively for 1 h.

After the acid attack, in the initial stages, the levels of Ca, P and O increased because of the releasing of these ions both from dental structures and restorative material. After 1 h of immersion, the lower levels of minerals were associated with structure alterations of both enamel and glassionomer cements.

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