

## Glass Ionomer Cement Release of the Fluoride as Anti-Cariogenic Properties among Four Different Types. Comparative Evaluation

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### Abstract

**Purpose:** The aim of this study was to evaluate the anti-cariogenic property (Fluoride ion release and its antibacterial properties) among four types of glass ionomer cements used as a permanent filling of caries cavities.

**Materials and Methods:** Four types of glass ionomer cements were used in this study.

[1] Fuji IX (GC, Japan); Conventional glass-ionomer cement,

[2] Fuji II LC (GC, Japan); Resin-modified glass ionomer cement (RMGIC),

[3] Dyract AP (Dentsply, Konstanz, Germany); Polyacid-modified composite resin (compomer) and

[4] Ketac N100 (3M ESPE St. Paul, MN, USA); Nano-filled resin-modified GIC.

The antibacterial activity of each material was evaluated against the *Streptococcus mutans* after day 2,7,14 and 30.

**Results:** All types of GIC restorative materials developed antibacterial activity but decreased depending increased the tested time.

**Conclusion:** The best one developed antibacterial activity was Fuji IX (GC, Japan); Conventional glass-ionomer cement followed by Ketac N100 (3M ESPE St. Paul, MN, USA); Nano-filled resin-modified GIC while the least one was Dyract AP (Dentsply, Konstanz, Germany); Polyacid-modified composite resin (compomer).

**Keywords:** Glass Ionomer; Fluoride Release; Resin Modified Glass Ionomer; Compomer; Nanoionomer; Streptococcus Mutans

### Abbreviations

GIC: Glass Ionomer Cement; RMGIC: Resin Modified Glass Ionomer Cement; DCT: Direct Contact Test; PBS: Phosphate-Buffered Saline

### Introduction

Dental caries process mostly started with infection by cariogenic bacteria, leading to acid production as a result of the bacterial carbohydrates metabolism within the oral biofilm. Lactic acid which is the main acid of metabolism, attack the mineral components of the tooth, leading to a process of demineralization, with subsequent degeneration of the organic component and finally a cavity is formed within the tooth [1]. Before the development of the cavity, a carious lesion looks like a white spot with a relatively intact, mineral-rich, but porous surface. It covers a subsurface area with a reduced mineral content [2]. Although it is known that acido-genic bacteria play a key role in the development of dental caries, [3-4]. However, various therapeutic procedures for the treatment of dental cavities do not

always eliminate all microorganisms from the caries focus [5-8]. The presence of bacteria in dental tissue left behind or bacterial invasion through a micro-leakage developing between the tooth and the filling may result in secondary caries [9]. Thus, antibacterial action is a desired feature of materials used for dental filling.

Modern materials are typically designed to be resistant to secondary caries and to micro-leakage at the edges, properties they possess on account of their ability to release fluoride and to be bonded to the prepared tooth surface. Margins of restorations are of particular importance, and lack of integrity of these may significantly increase the risk of secondary caries [10-13]. Secondary caries is, in fact, the most frequent indication for replacement of all types of restoration [10,14] and the limited durability of dental restorations means that some patients are in continuous restorative cycles that result in larger and larger restorations and more complex therapeutic measures.

Many new filling materials, characterized by the release of fluoride (F) ions, were developed in the last decade as a means of protection against recurrent caries. Of these, the most important are the glass-ionomer cements and their hybrids (resin-modified glass-ionomer cements and polyacid-modified composite resins; so-called "compomers" in addition to newly nano-ionomer derivatives). By releasing fluoride, these materials offer protection to the hard dental tissues [15] and the surrounding micro-environment [16,17].

There are a number of mechanisms by which release of fluoride protects the teeth. First, the presence of small amounts of fluoride in the saliva reduces the solubility of the mineral phase of the tooth mineral. Second, fluoride incorporated into the mineral phase leads to the formation of a thin layer of fluor-apatite, which is less soluble even at low values of pH than hydroxyapatite. Third, fluoride may interfere with the metabolism of cariogenic bacteria by inhibiting essential enzyme-mediated processes. All of these mechanisms shift the demineralization/remineralization equilibrium back in favour of remineralization [18,19].

Glass-ionomers, for example, have been reported to contribute to the remineralization on incipient enamel lesions *in vitro* [20]. Such studies on the effects of fluoride on dentine reveal that low fluoride concentrations may lead to hypermineralization of dentine [21,22]. In fact, the choice of the restorative material can be crucial in determining whether demineralization or remineralization occurs in the dentine tissue surrounding a restoration. Incipient caries like lesions under glass-ionomer restorations have been found to remineralize and even to hypermineralize, whereas amalgam and composite restorations have been shown to be predominantly associated with further remineralization of the specimens [19]. The distinct zone of interaction found between the glassionomer cement and hard dental tissues contributes to the adhesion and high resistance to microleakage of glass-ionomer cements restorations. According to the opinion of many authors, F ions may be responsible for the anti-microorganism action of these materials [23-27].

Some methods have been suggested for testing the antimicrobial effect of dental materials. The most frequently employed methods are those based on direct contact test (DCT) [28,29]. The direct contact test is a relatively new method that provides the information on the bacterial viability and growth rate and quantitatively measures the effect of direct and close contact between the microorganisms and the tested materials, regardless of the solubility and diffusibility of their components. The growth inhibitory effect of GIC is considered beneficial in preventing bacterial colonization. In addition, the antibacterial activity, during the time, assumes clinical relevance [30].

The aim of this study was to evaluate the anti-cariogenic property (Fluoride ion release and its antibacterial properties) among four types of glass ionomer cements used as a permanent filling of caries cavities.

### Materials and Methods

Four types of glass ionomer cements were used in this study.

[1] Fuji IX (GC, Japan); Conventional glass-ionomer cement,

[2] Fuji II LC (GC, Japan); Resin-modified glassionomer cement (RMGIC),

[3] Dyract AP (Dentsply, Konstanz, Germany); Polyacid-modified composite resin (compomer)

[4] Ketac N100 (3M ESPE St. Paul, MN, USA); Nano-filled resin-modified GIC

Restorative materials used in this study are shown in table 1. The antibacterial activity of each material was evaluated against the *Streptococcus mutans*, this study followed the methodology of the study of Elaheh VD., *et al* [31].

Material	Type	Manufacturer
Fuji IX	Conventional glass-ionomer cement	GC, Japan
Fuji II LC	Resin-modified glass-ionomer cement (RMGIC)	GC, Japan
Dyract AP	Polyacid-modified composite resin (compomer)	Dentsply, Konstanz, Germany
Ketac N100	Nano-filled resin-modified GIC	3M ESPE St. Paul, MN, USA

**Table 1:** Restorative materials used in this study.

### Preparation of glass ionomer samples

Six wells (7 mm diameter and 3 mm thickness) were punched in the Muller-Hilton agar plates and filled with the four types of GIC restorative materials. A uniform surface was achieved by using a small flat-ended dental instrument, such as a dental spatula. The material was allowed to set in accordance with the manufacturer’s recommendation either by chemical cure (acid base reaction) as in conventional GIC (Fuji IX) or by light cured or both methods as in other three types.

### Antibacterial activity test

Bacterial strain from stock cultures was cultivated in Brain Heart Infusion broth (Difco, Detroit, USA) at 37°C, for 24h. The top 4 mL of the resulting undisturbed bacterial cultures were transferred to new test tubes and centrifuged for 10 min at 3, 2 gravity. The resulting supernatant was discarded and the bacteria was resuspended in 5 mL of phosphate-buffered saline (PBS) with a pH of 7.5 (Sigma-Aldrich, St. Louis) and mixed gently by vortexing for 10 sec. We used DCT to test the antibacterial properties and anti-cariogenic effect of the different types of GIC. The antimicrobial susceptibility profiles were determined by disk diffusion agar method according to CLSI M100-S12 protocols (2005). In each sterilized Petri dish (20100 mm), a base layer containing 15 mL of blood agar mixed with 100 µl of inoculum was prepared. After the solidification of culture medium, wells measuring 7 mm in diameter were made in each plate and the testing materials were transferred to wells. Two wells were served as the positive control without the four tested GIC restorative materials. Plates were incubated at 37°C for 48h and after that, diameters of zones of inhibition produced around the specimens were measured at three different points. The size of inhibition zones was calculated through subtracting the diameter of specimen (7 mm) from the average of three measurements of the halo. All measurements were performed twice by the same blinded operator. Antibacterial tests were repeated 5 times to confirm the homogeneity of the results. Moreover, diameters of zones of inhibition produced around specimens were measured after the re-incubation of plates at 37°C for 5 days.

### Statistical Analysis

The mean diameter of inhibition zone values for each material was used for statistical analysis by using General Linear Models to compare the inhibition zones of bacteria around each GIC restorative material. Tukey’s studentized post-hoc tests were performed to identify the differences among the GIC restorative materials, the level of significance set at  $p < 0.001$ . A Tukey’s complementary test was also used to determine if there was a significant difference between the inhibitory effects of 2,7,14 and 30 days specimens ( $p < 0.001$ ).

### Results

The mean values and standard deviations of the inhibition zones for each material according to the bacteria strain, at different days, are shown in (Table 2). There was a significant difference between the types of GIC restorative materials according to time of evaluation

2,7,14 and 30 days and the antibacterial activity decreased by time to reach the lowest level at day 30 as seen in (Figure 1). The antibacterial property of Fuji IX (conventional glass ionomer) was the highest among all other types in all tested days with a statistical significant difference ( $p < 0.001$ ). A reduction in the measured inhibition zones was observed in all samples after the day 7 towards day 14 and day 30 that was not significant in both Fuji IX (Conventional Glass Ionomer) and Ketac N100 (Nano-Filled Resin-Modified GIC), but significant with Dyract AP (Compomer) and Fuji II LC (Resin-Modified Glass-Ionomer Cement (RMGIC)).

Materials	Streptococcus mutans (inhibition zones)			
	Day 2	Day 7	Day 14	Day 30
Fuji IX	22.80 ± 1.30	21.20 ± 1.30	20.10 ± 1.09	19.88 ± 0.09
Fuji II LC	14.0 ± 0.70	13.40 ± 0.54	11.30 ± 0.79	9.17 ± 0.71
Dyract AP	12.40 ± 0.54	11.0 ± 0.70	9.20 ± 0.34	5.11 ± 0.21
Ketac N100	15.33 ± 1.11	14.32 ± 0.75	12.67 ± 0.88	11.39 ± 0.44

Table 2: The mean values and standard deviations of the inhibition zones for each material.

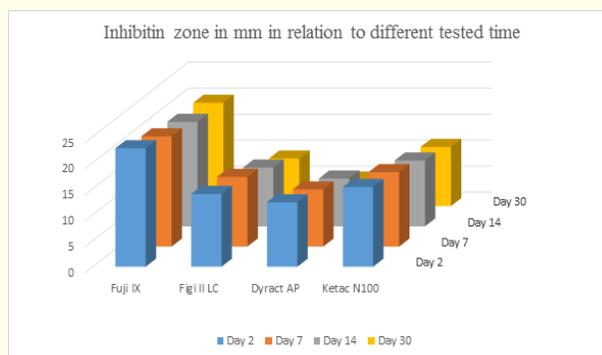


Figure 1: Showing the mean values and standard deviations of the inhibition zones for each material.

## Discussion

Prevention of secondary caries around existence restorative material is reliant on the patient control of dental plaque. However, many patients do not take care of their oral hygiene perfectly. Consequently, antibacterial properties of permanent restorative materials are desirable. Conventional glass ionomer presents approving and essential properties such as biocompatibility to dental pulp, ability of chemical bonding to enamel and dentin and fluoride releasing, which can play an important role in the inhibition of bacteria growth and caries progression [33-36]. Different *in-vitro* methods have been used to study the antibacterial activity of dental materials. Boeckh., *et al.* throughout their experiments using strains of *S. mutans*, showed the important role of this microorganism in caries etiology [28]. The methodology applied in this research was based on DCT to verify the inhibition zone of materials evaluated and focused on the standardization of the experimental conditions, in particular in relation to the specimens' dimensions and microorganism suspension. According to the results, all glass ionomer restorative materials evaluated the inhibited bacterial growth, but with differences according to the material. The differences in growth inhibition between these restorative materials may be related to their inherent potency and to different compositions [32]. Yap and others reported that there was no antibacterial activity despite the presence of fluoride in the agar around the set materials [37]. We found that all four GICs almost inhibited the growth of *S. mutans* in the day 2. This effect lasted for at least one week. The most credible cause of the reduced bacterial growth after direct contact with the GIC is fluoride release from this material combined

with a pH fall around the material as described elsewhere [38-39]. The concentration of fluoride in a specific dental material does not reflect its rate of release. In consequence, the antibacterial properties of glass ionomer restorative materials are different from one material to another. From a clinical point of view, the fluoride release of the GICs may drop significantly with long-term usage as reported in other studies [40-41]. Our information recommends that further studies are required to examine the levels of fluoride release and the effects of GICs on complex biofilms. In the present study, we observed that the conventional glass ionomer cements (Fuji IX) had significantly more antibacterial effect in comparison to other types of GIC restorative materials. Other studies have also reported the most antibacterial properties of GICs between different restorative materials [43,44]. This could be explained by the combined effect of low pH of GICs and their fluoride-leaching capabilities [42]. In all experiments, the antibacterial activity measured with Fuji IX (conventional glass ionomer) and Ketac N100 (Nano-filled resin-modified GIC) greater than that of Dyract AP (compomer) and Fuji II LC (Resin-modified glass-ionomer cement (RMGIC)), which is estimated to be correlated with the higher fluoride release rates observed with the formers.

Results revealed that Dyract AP (compomer) was the least type in antimicrobial activity. Dyract AP is a polyacid-modified composite (compomer), which has strontium aluminium fluoride silicate glass and strontium fluoride embedded in 1,6 bis (methacryloxy-2-ethoxycarbonylamino)-2,4,4-trimethylhexane (UDMA) [26,45,47]. As far as compomers are concerned, light polymerization results in resin binding, and, despite acid-base reactions proceeding inside the material, F release is slower [25,26,28,45,47]. According to some authors [23,25,26,47], the level of F release depends on possibilities for its diffusion outside and not on its concentration in the material. This property is related to the hydrophilicity of the material because F ions are released from that part of matrix into which water diffuses. Many researchers therefore explain differences between different groups of materials by water-sorption mechanisms [28,45,46,47].

### Conclusion

Based on the results of the present study, it can be concluded that all the evaluated GIC restorative materials displayed some antimicrobial activity. This antimicrobial activity was time dependent. Fuji IX (Conventional Glass Ionomer) and Ketac N100 (Nano-Filled Resin-Modified GIC) were the most active antibacterial cements. Combined with the mechanical and biological properties, these differences should be taken into account when one is choosing the type of GIC as permanent restoration in clinical use.

### Conflict of Interest

All authors declare there is no competing interest related to the study, authors, other individuals or organizations, and no financial support for this study.

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