Research Article

Evaluate Antimicrobial Properties of Fluoride Release Dental Resin Composite

Abdullah Amjad Alrwaili[1*], Radhi Ibrahim Albalawi[1], Abdullah Saad Alasiri[1], Hajar Mutwkil Othman[1], Abrar Hatab Alqahtani[1], Heba Mohammed Almughwi[1] and Abdallah F. Elsadany[2]

[1]Dentist graduated from Vision Colleges of Dentistry and Nursing; [2]Assistance Professor of Restorative Dental Science at Vision Colleges of Dentistry and Nursing

Corresponding author:
Abdullah Amjad Alrwaili,
Vision Colleges of Dentistry and Nursing, Riyadh, Saudi Arabia,
Al Yarmuk District-Wathilan Street, 13251
E-mail: alrwailio09@gmail.com

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Abstract

Aim of study: Aim of this study to evaluate the antimicrobial properties of fluoride release dental resin composite (SureFill SDR, DENTSPLY Caulk, Milford, Delaware, USA) compared to conventional Glass ionomer (Ketac™ Molar Quick, 3M/ESPE, St. Paul, USA). **Materials and methods:** Ten specimens of fluoride release dental resin composite (SureFill SDR) and conventional Glass ionomer (Ketac™ Molar Quick) were prepared with 8 mm diameter and 2 mm thickness. Then specimens finished and polished. Then each specimen was placed in a tube containing 5 mL of deionized water. The tubes were incubated at the controlled temperature of 37°C for up to 30 days. The storage solution was collected and collected solution was mixed with TISAB III solution. Fluoride ion concentration in the solution was then measured using fluoride specific ion electrode. An adapted agar diffusion test, used for the assessment of antimicrobial activity, was applied in the microbiological studies. **Results:** there was fluoride release of all composite specimens (SureFill SDR) were below effective threshold (0.1 ppm-1 ppm) mean ± standard deviation (0.001 ppm ± 0.0005 ppm), while fluoride release of Ketac™ Molar Quick was (6.1 ppm ± 0.2 ppm). Antimicrobial activity: after 1-day storage there was no inhibition zone detected in both blood agar and agar-agar plates in composite specimens while there was inhibition zone in glass ionomer specimens, after 15 days' bacterial growth was observed in both incubation media of composite specimens. **Conclusion:** SureFill One show little fluoride release (below effective threshold) and there is no antimicrobial activity especially cariogenic bacterial other than Ketac™ Molar Quick.

Keywords: Cariogenic bacterial; Glass ionomer; Fluoride ion; Antimicrobial activity; Bacterial Growth.

Introduction

Dental resin composites have been frequently used in the past three decades for restoring hard tissue such as enamel and dentine in both posterior and anterior teeth, owing to the good esthetic properties and strength. [1,2] A dispersed phase which comprise of glass filler particles are disseminated in order to fortify a polymerizable resin matrix and silane coupling agents are generally found in dental composite resins. Zirconium/silica based inorganic glass filler particles are normally distributed in an organic matrix of resin components such as bisphenol, other components which are cured during application include urethane dimethacrylate (UDMA), triethylene glycol dimethacrylate (TEGDMA), and glycidyl methacrylate (BIS-GMA). [3]

Secondary tooth decay has been a major factor in the failures of resin composites shown by various studies. [4] The antibacterial properties of several composite resins were investigated by various studies, the studies reveled a failure to exhibit any inhibition of bacterial growth after polymerization of the resin restorative materials. [5,6]

Furthermore when compared with enamel and other types of restorations composite resins have displayed more dental biofilm accumulation in the long term. The dearth of inhibitory effect against cariogenic bacteria such as *streptococcus mutans* is an example shown from hard evidence that degradation of composites is a result of the formation of biofilm.[7,8] Moreover, recurrent carious lesions progress around these restorations as a consequence of adhered bacteria which infect the nearby hard and soft tissues such as gingiva, enamel, and dentine, which necessitates replacement of the restoration, requiring further loss of tissue. [9] Hence, focusing on antimicrobial treatment regarding resin composites is considered as one of the approaches in order to prolong the survival time of these materials. [10]

By modifying the resin matrix or the filler particles of resin composites an addition of an antibacterial element can be attained. A released soluble antimicrobial activity and stationary non-released antibacterial agents are two approaches used to provide resin composites that possess antibacterial activity. The bulk of the restoration releases an antibacterial agent by a soluble agent which is steadily released after a while. [11,12] Even though

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the effect of the antibacterial agent is accomplished, the agent released has quite a few disadvantages including the generation of a porous structure; time limited efficacy and the risk of toxicity of nearby tissues due to the difficulty in monitoring the rate of diffusion. Chlorhexidine, antibiotics, fluoride, silver ions, iodine and quaternary ammonium compounds are all a low molecular weight soluble antibacterial agents that have been introduced. [13]

Fluoride has multiple antibacterial mechanisms, such as, hindering the development of pellicle and biofilm, inhibition of growth and metabolism of microbes, reducing the process of demineralization and enhancing the remineralization process. So, few fluoride releasing systems were reported to produce antibacterial effects such as, ytterbium trifluoride (YbF3), strontium fluoride (SrF2) or leachable glass fillers. [14,15] The formation of carious lesions is presumed to be disturbed through the released fluoride from restorative materials which will reduce the process of demineralization and promote remineralization of enamel and dentine. [16]

Glass ionomers, resin modified glass ionomer cements, polyacid-modified composites (compomers), composites, and amalgams are among the fluoride-containing dental restoratives available nowadays. [17] The ability of the products to release fluoride varies due to their varied matrices and setting processes. Restoratives' antibacterial and cariostatic activities, on the other hand, are thought to be linked to the degree of fluoride released. [18]

Bacteria or microbes that are difficult to grow are called fastidious‖ due to their requirement of special nutrition environment, thus; a concentrated medium called blood agar is used to culture these bacteria. [19] Utilization of blood agar helps to culture a wide variety of pathogens, especially those which do not grow easily such as, *Haemophilus influenzae, Streptococcus pneumoniae* and *Neisseria species*. [20] It is also essential to identify and distinguish between hemolytic bacteria, particularly *Streptococcus* species. Also certain bacteria certain bacteria such as *Bacillus, Streptococcus, Enterococcus, Staphylococcus* and certain strains of aerococcus secrete cytolytic toxins causing hemolysis (destruction of red blood cells) which is detected by a differential medium. [21]

Adding antibiotics, chemicals or dyes can make the blood agar selective for specific pathogens. Examples include purple crystal blood agar to collect *Streptococcus pyogens* from pharyngeal swabs and kanamycin or neomycin blood agar to collect anaerobic bacteria from pus. [22,23]

A commonly used technique for the evaluation of the antibacterial activity of dental restorations and medications is an agar diffusion technique. A direct comparison of the efficacy of different filling materials against the target pathogens is one of the main advantages of this technique, demonstrating the type of restorative material that has a possibility of eliminating bacteria from the oral cavity. [24] Having a wide antibacterial property is of great importance for the restorative materials and an investigation of the antibacterial field of these restorative materials should be carried out. [25]

Aim of this study to evaluate the antimicrobial properties and fluoride release of dental resin composite (SureFill SDR, DENTSPLY Caulk, Milford, Delaware, USA) compared to conventional Glass ionomer (Ketac™ Molar Quick, 3M/ESPE, St. Paul, USA).

Materials and Methods

Ten specimens of fluoride release dental resin composite (SureFill one SDR, DENTSPLY Caulk, Milford, Delaware, USA) were prepared in a half-split stainless-steel round mold of 8 mm diameter and 2 mm thickness. Mold was put on a glass slide covered by Mylar strip and separating medium was applied to mold wall with a brush, then composite materials was applied to the mold cavity with a special gun. After that, glass slide covered with Mylar strip was put the top of the mold. Curing was carried out on the top then the bottom of the specimens before removal from the mold. Curing was achieved by light-emitting-diode LED curing unit for 20 secs with four overlapping light exposures to cure the entire length of specimen. Wavelength range of LED curing unit was between 430 nm-485 nm and output intensity was at 1200 mW/cm^2 [Figure 1]. In the same manner 10 specimens of conventional glass ionomer (Ketac™ Molar Quick, 3M/ESPE, St. Paul, USA) were prepared in a half-split stainless-steel round mold of 8 mm diameter and 2 mm thickness. Mold was put on a glass slide covered by Mylar strip and separating medium was applied to mold wall with a brush, then glass ionomer materials was applied to the mold cavity with a special gun. After that, glass slide covered with Mylar strip was put the top of the mold. Finally, the glass ionomer specimens were left to set [Figure 1].

After composite and GIC set, excess was removed. All specimens were sequentially finished with a 600# and 1,200# silicon carbide paper, then finished sequentially with a complete series of Soflex discs (3M ESPE, St Paul, MN, USA). For standardization a single operator, using a low-speed handpiece at approximately 4,000 rpm-5,000 rpm, performed the finishing procedure [Figure 2]. After that, the specimens were polished by

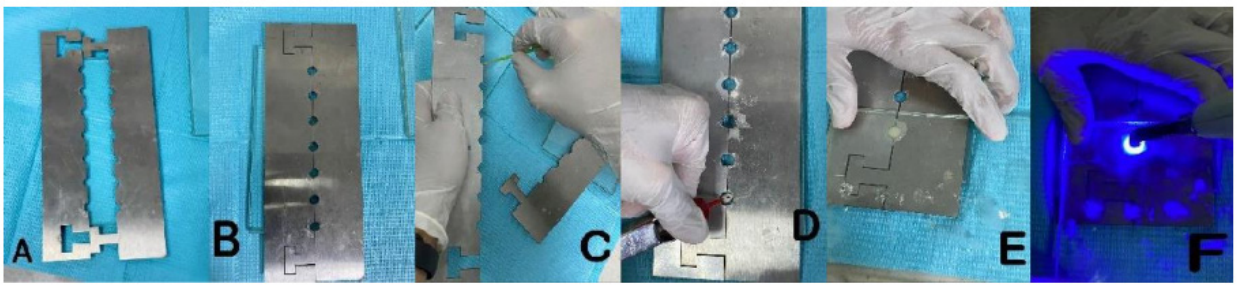

Figure 1: Preparation of composite and GIC specimens; A,B): Half split mold closed and open; C): Application of separating medium; D): Application of composites and GIC with a special gun; E): glass slide covered with Mylar strip was put the top of the mold; F): light curing of specimens.

rubber cup and polishing paste, polished surfaces were water-rinsed with an air-water syringe for 60 secs, to remove any surface debris left and then were air-dried for 30 sec.

Then each specimen of composite and GIC was placed in a tube containing 5 mL of deionized water. The tubes were incubated at the controlled temperature of 37°C for 1day, 7 days, 14 days and up to 30 days. The storage solution was collected and collected solution was mixed with Total ionic strength adjustment buffer III solution (TISAB III solution, Orion Ionplus, Thermo Fisher Scientific) with a ratio of 1:10 (volume ratio). Fluoride calibration standards (0.1 Fppm, 1 Fppm, 10 Fppm, 100 Fppm) were prepared using a standard fluoride solution (Orion Ionplus, Thermo Fisher Scientific). Fluoride ion concentration in the solution was then measured using fluoride specific ion electrode (Orion Versastar Pro, Thermo Fisher Scientific)

An adapted agar diffusion test, used for the assessment of antimicrobial activity, was applied in the microbiological studies. The following standard strains/lines were used for the evaluation: *Streptococcus mutans* ATCC 25175, *Streptococcus sanguis* ATCC 10556, and *Lactobacillus casei subsp. casei* ATCC393. The strains were grown on BHI medium (Oxoid) for 18 hours at 37°C and afterwards suspensions with densities of 0.5 on the McFarland scale was prepared. Agar ISO-SENSITEST (Oxoid) with 5% ram blood was inoculated with 0.1 ml of the suspension using a cotton swab and then the prepared composite specimens were put on the plates. The plates with *streptococci* were incubated in incubator. After 24 hours, and 30 days the zones of bacterial growth inhibition were measured in mm. the same procedures also repeated with agar-agar plate [Figure 3].

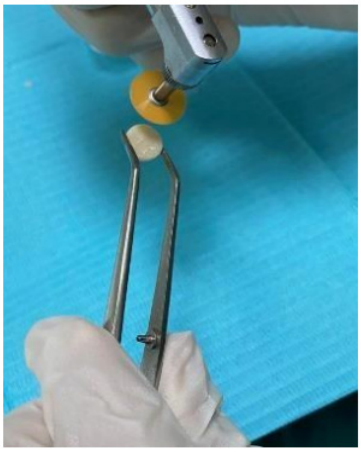

Figure 2: Specimen finishing and polishing.

Data will be collected and tabulated and statistically analyzed by an IBM compatible personal computer with SPSS Statistical Package of Social Science version 20 (SPSS Inc. Realesed 2011. IBM SPSS statistics for windows, version 20.0, Armnok, NY: IBM Corp.).

A. Descriptive statistics will be expressed in mean (x) and standard deviation (SD).

B. Student t-test of fluoride release between composite and glass ionomer in every day

Results

Fluoride release of all tested composite and GIC specimens are listed in [Table 1].

As table shown there is fluoride release of all composite specimens were below effective threshold (0.1 ppm-1 ppm).

While glass ionomer specimens were demonstrated higher fluoride release on the first day (8.5 ppm ± 0.5 ppm). Over the third day (7.4 ppm ± 0.6 ppm), the fluoride release decreased until it reached to lest level at thirtieth day (6.1 ppm ± 0.2 ppm), but still higher than effective threshold (0.1 ppm-1 ppm).

Student t-test revealed that there was highly significant difference between composite and glass ionomer in all days.

Antimicrobial activity of composite specimens; after 1-day storage there was no inhibition zone detected in both blood agar and agar-agar plates [Figure 4], after 30 days' bacterial growth was observed in both incubation media [Figure 5].

Antimicrobial activity of glass ionomer specimens: after 1-day storage there was inhibition zone detected in both blood agar and agar-agar plates [Figure 6].

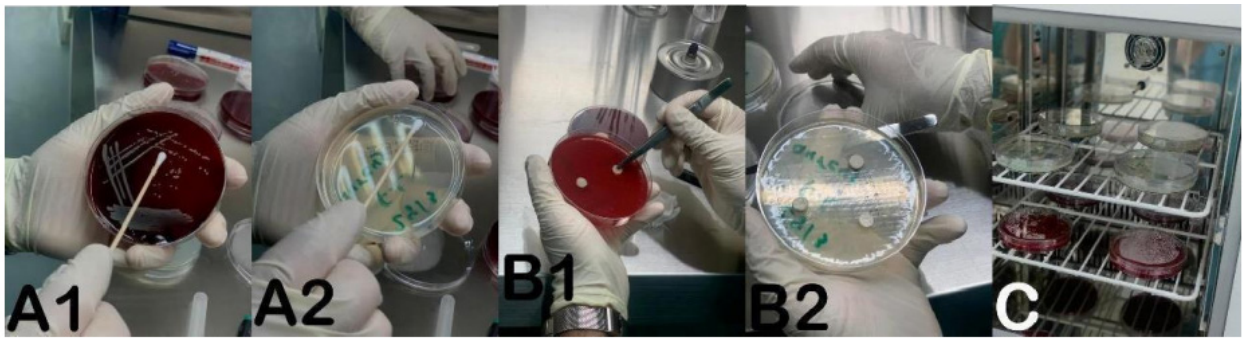

Figure 3: Antimicrobial test; A1, A2): Cotton swap of bacteria in storage media blood agar and agar-agar respectively; B1, B2): Specimens placed in storage media blood agar & agar-agar respectively; C): Specimens incubation.

Table 1: Fluoride release (ppm) of all composite and glass ionomer Specimens.

Materials	Fluoride release (ppm) after 1 day	Fluoride release (ppm) after 7 days	Fluoride release (ppm) after 14 days	Fluoride release (ppm) after 30 days
Glass ionomer	8.5 ± 0.5	7.4 ± 0.6	6.9 ± 0.4	6.1 ± 0.2
Composite	0.008 ± 0.0006	0.006 ± 0.0005	0.002 ± 0.0001	0.001 ± 0.0005
P value	0.000	0.000	0.000	0.000

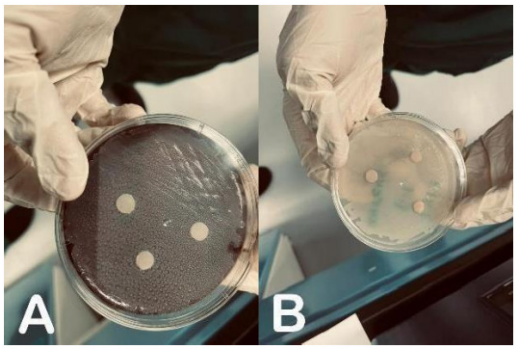

Figure 4: Antimicrobial activity of Composite specimens after 1-day incubation (no inhibition zone).

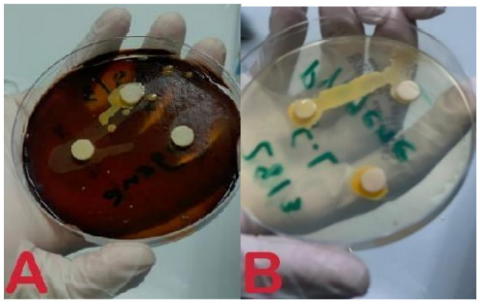

Figure 5: Antimicrobial activity of Composite specimens after 30-days incubation (bacterial growth).

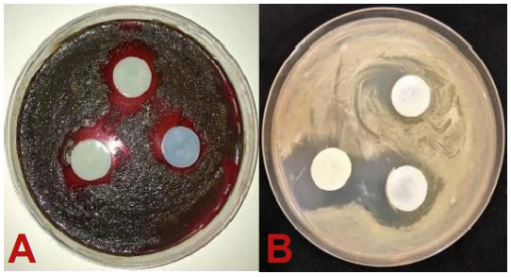

Figure 6: Antimicrobial activity of Glass ionomer specimens after 1-day incubation (inhibition zone).

Table 2: Composition of Surefill one, Ketac™ Molar Quick general function of components.

Material	Component	General function
Composite SureFill SDR, DENTSPLY Caulk, Milford, Delaware, USA	Modified polyacid (MOPOS)	Etchant, adhesion promoter, crosslinker between covalent and ionic network
	Bifunctional acrylate (BADEP)	Crosslinker in the covalent network
	Acrylic acid	Reactive diluent, Primer, crosslinker between covalent and ionic network
	Water	Solvent for polyacid and resins, etching aid
	Reactive glass filler	Filler supporting wear resistance and mechanical strength, Fluoride release.
	Non-reactive glass filler	Radiopacifier, rheology modifier
	Initiator	Photo- and redox initiator system
	Stabilizer	Stabilize monomers upon storage
Glass ionomer Ketac™ Molar Quick, 3M/ESPE, St. Paul, USA	Powder	Aluminum-calcium-lanthanum fluorosilicate glass, 5% polycarbonate acid
	Liquid	Polycarbonic acid and tartaric acid

Discussion

Presumably, released fluoride ions promote low soluble fluorohydroxyapatite formation which may aid in reducing the risk of tooth demineralization. [26] Furthermore, reduction and inhibition of bacterial growth and their metabolism can be achieved from the release of fluoride. Commonly observed in materials which are polymer-based is the diffusion-controlled release of active materials. [27]

Glass ionomers are the most cement/filling materials that release fluoride, this is due to their powder composition (Aluminum-calcium-lanthanum fluorosilicate glass), so that, they have highly antimicrobial activity. SureFill one uses reactive glass fillers as described in [Table 2]. [28] Therefore, an inherent acid-base reaction of fluoride release is to be expected. The significance of fluoride release for clinical success has been argued over the years. [29] Glass ionomers are well known to provide an initial discharge of fluoride that substantially decreases in the long run. [30] In comparison to traditional glass ionomers, compomers (i.e. Dyract) provide similar fluoride release rates in the long term were shown in vitro measurements. [31] The Dyract filling materials did not show any signs of recurrent decay after two decades, this was revealed by a recently published clinical study in non-caries cervical lesions. [32] Additionally, in an in-situ study, non- fluoride releasing composite (i.e. Spectrum TPH) was compared with Dyract extra in the proximal contact of class II restorations resulted in a reduction of demineralization compared to the enamel control along with resin composites. [33] And although a fluoridated tooth paste was used twice daily, a substantial difference in the development of early caries was still found. [34]

An initial high boost followed by an immediate sharp drop in release rate was shown in glass ionomers as anticipated. As previously stated, from long-term standpoint, the release rates of fluoride of different restorative materials approximate one another. [35,36]

Regarding the above, a comparison was made between Surfil one to compomers, resin modified glass ionemers, and glass ionomers. [37] Water and a storage media that was exchanged subsequent to every measurement point were used to store three specimens per material. [38]

Stationary non-released reactive glass filler leads to very little fluoride release (below effective threshold) in all specimens in deionized water and no evidence of antimicrobial activity particularly cariogenic bacteria as shown in the current study. [39]

Conclusion

SureFill one show little fluoride release (below effective threshold) and there is no antimicrobial activity especially cariogenic bacterial other than Ketac™ Molar Quick which has highfluoride release and high antimicrobial activity.

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