Antibacterial Activity of Surface Pre-reacted Glass (S-PRG) Resin Composite Versus Conventional Glass Ionomer Cement: An In Vitro Study

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Abstract

Aim: The aim of the present in vitro study was to evaluate the antibacterial activity of S-PRG resin composite versus conventional glass ionomer cement.

Materials and Methods: A total number of Forty cylindrical disc formed specimens for antibacterial activity test were made in a standardized sterile teflon mold with an internal diameter of 6mm and a thickness of 2 mm were assigned into two equal groups: S-PRG resin composite and conventional glass ionomer cement. For S-PRG resin composite the sterile teflon mold were put on the highest point of a sterile glass slide (1.2mm thickness) and a celluloid strip (0.05mmthickness), the restorative material samples were packed into the mold using gold plated composite applicator, the second celluloid strip was used to overlay the top side of the mold to inhibit formation of air inhibited surface layer. Additional glass slide and the weight 500gm were used to give a consistent, good packing of the specimens and to extrude the excess material. Employed weight and glass slide were eliminated, light cured for 10 seconds (wave length range 440-490 nm)from the top, bottom and sides. The tip of the light curing unit was kept focused in direct contact with the celluloid strips on the top surface of the mold perpendicular to it.

Results: The following results would be: The highest inhibition zone was for S-PRG resin composite at 72 hours (T_2) against lactobacillus acidophilus, followed by those at 48 hours ($T_{1,}$), then those at 7 days in comparison to conventional glass ionomer cement. The highest inhibition zone was for S-PRG resin composite at 72 hours (T_2) against streptococcus mutans, followed by those at 48 hours ($T_{1,}$), then those at 7 days in comparison to conventional glass ionomer cement.

Conclusions: S-PRG resin composite (Giomer) application has excellent antibacterial activity and caries inhibitory effect, S-PRG resin composite (Giomer) makes good candidates for use in dental applications.

Keywords: Antibacterial Activity - Glass Ionomer Cement - *Lactobacillus Acidophilus* - *Streptococcus Mutans*

Introduction

Dental caries is one of the most significant public oral microbiological diseases worldwide, it occurs as localized destruction of dental hard tissues by acidic byproducts produced by bacteria **(1, 2).** The most important cariogenic organism are *Streptococcus mutans* in initial enamel carious lesions and *Lactobacillus acidophilus* in advanced carious lesions **(1, 3).**

Secondary caries is the main reason for the restoration failure that may occur at the interface between the restoration and the tooth structures as a result of microleakage and demineralization due to invasion and adhesion of bacteria to the tooth surface and restorative materials **(4)**. Prevention of secondary caries has been attempted to allow for more durable and successful restoration **(5, 6).**

There are numerous available dental restorative materials that contain fluoride have caries inhibitory effect in the market including glass ionomer cements, resin modified glass ionomers, polyacid modified resins (compomers), S-PRG resin composites (giomers) and resin composites **(7).** Glass ionomers Cements are bioactive restorative materials that characterized by their chemical adhesion to the tooth, excellent biocompatibility, cariostatic and antibacterial property due to fluoride content **(4, 8).** Fluoride plays a significant role in dentistry in the treatment of incipient dental caries by reversal the demineralization process and enhancement the remineralization by replacing the hydroxyl groups in the upper layers of the hydroxyapatite crystals to be Fluor apatite which results in a hard dental tissues with less solubility as well as prevention for future dental caries **(4, 9).** Fluoride can be used as a reservoir

releasing small amounts of fluoride in restorative materials especially in patients with high caries risk increasing the tooth resistance to caries, prevent bacterial growth **(10).**

There are disadvantages of glass ionomers cements such as they are esthetically poor, sensitive to moisture contamination, prolonged setting reaction, compromised mechanical properties and the brittle nature which necessitates support of the surrounding tooth structure affecting its performance as result it is better in single-surface restorations compared to multi-surface restorations **(7).** As the low mechanical properties of conventional glass ionomer cements, some modification were made to increase it **(11).**

Introduction of a new category of hybrid aesthetic restorative material "bioactive smart dental materials" has increased in recent decades that combines fluoride releasing capability of conventional glass ionomer and the durability of composites has been known as S-PRG resin composite (Giomers) by incorporating particles of pre-reacted glass filler in the matrix of composite material to improve their long-term marginal stability with greater safety and efficacy against secondary caries, better biocompatibility, smooth surface finish and prevention of bacterial access to dentinal tubules and ultimately the pulp **(12, 13).** Surface pre-reacted glass-ionomer (S-PRG) filler is a bioactive functional glass that releases six ions: borate (BO_3^{3-}), aluminum (Al^{3+}), silicate (SiO_3^{2-}), strontium (Sr^{2+}), sodium (Na^+) and fluoride (F^-)·S-PRG filler that can strengthen tooth structure, inhibit bacterial growth and adhesion to resin surface, suppress tooth demineralization, enamel remineralization **(14, 15).** Beautifil is a tooth colored bioactive restorative material that uses resin base and surface reaction type SPRG filler technology where only the surface of the glass filler was reacted while the glass core remains un reacted while Beautifil II is a second-generation Giomer introduced into market claiming better optical properties plus fluoride release & recharge capacity **(12).**

The universal hybrid S-PRG resin composite (Giomer) called Beautiful II LS (Shofu Inc., Kyoto, Japan) promises dentists minimum shrinking and maximum aesthetics. The S-PRG technology (Surface Pre-reacted Glass reaction) used by Beautifil II LS makes it smart and bioactive with sustained fluoride release and recharging, antiplaque impact that reduces plaque adherence and prevents bacterial colonisation and plaque buildup. Therefore, determining Beautifil II LS's antibacterial activity was crucial.

Materials and Methods

1. Materials:

Light cured S-PRG resin composite (Giomer): Beautiful II LS, chemically cured conventional glass ionomer cement: Ionostar molar.

2. Methods:

Ethical approval:

The protocol of this study was approved by the Council of Conservative Dentistry – Faculty of Dentistry – October 6 University and the ethical issues were reviewed and revised by the Research Ethics Committee - Faculty of Dentistry– October 6 University on March 2022 (Approval No. RECO6U/13-2022).

A power analysis was designed to have adequate power to apply a two-sided statistical test of the null hypothesis that there is no difference would be found in the antibacterial activity between different groups. By adopting an alpha level of (0.05) a beta of (0.2) i.e. power=80% and an effect size (d) of (0.909) calculated based on the results of **Alsamolly et al. (10)**; the predicted sample size (n) was a total of (40) samples (i.e.20 samples per group). Sample size calculation was performed using G*Power version 3.1.9.7.

A total of forty samples were used in the study for antibacterial activity test and divided into two groups (each group 20 specimen). In Group I the discs were made of S-PRG resin composite. In Group II the discs were made of conventional glass ionomer cement(GIC) as control group. The antibacterial effect of each group were assessed after three-time intervals: 48 hours, 72 hours and 7days. Each group was further divided into two subgroups (A,B) according to placement into *Streptococcus mutans* and *Lactobacillus acidophilus* (N=10).

A standardized teflon mold (2mm thickness and 6mm internal diameter) were used for specimen construction. **Group Ⅰ** twenty cylindrical disc formed specimens S-PRG resin composite (Beautiful II LS; SHOFU Inc., Kyoto, Japan). The sterile teflon mold was put on the highest point of a sterile glass slide (1.2mmthickness) and a celluloid strip(0.05mmthickness) (stripmat, polydentia, CH-6805 Mezzovico, Switzerland, the restorative material samples were packed into the mold using sterile gold Plated composite applicator (Medesey, Italy), the second celluloid strip was used to overlay the top side of the mold to inhibit formation of air inhibited surface layer.

Additional glass slide and weight 500gm were used to give a consistent, good packing of the specimens and to extrude the excess material**.** Employed weight and glass slide were eliminated. Light cured for 10 seconds from the top, bottom and sides utilizing light-curing device(X-cure light cure, Guilin Wood pecker; Germany, wave length 440-490 nm**.** The tip of the light curing unit was kept focused in direct contact with the celluloid strips on the top surface of the mold perpendicular to it. Following photo polymerization the cylinder-shaped specimens were detached from their molds **(10, 16, 17).**

Group II twenty cylindrical disc formed specimens conventional glass ionomer cement capsule (Ionostar molar; VOCO GmbH, Cuxhaven, Germany). It was activiated directly before use by pressing down the end of the capsule onto a hard surface and mixed using high frequency mixer (GIC mixer; VOCO GmbH, Cuxhaven, Germany, for 10 seconds and was inserted into the teflon mold using the holder of applicator (Detrey applicator; Dentsply, America,. Its final setting after 3-5 minutes. Following complete setting the cylinder-shaped specimens were detached from their molds.

Storage of specimens:

The specimens of S-PRG resin composite and conventional glass ionomer cement were stored in sterile pouches in a dry condition for 24 hours at room temperature 25°C till testing **Şirinoğlu-Çapan et al. (2020).**

Preparation of Culture Media:

ATCC 25175 Type strains *Streptococcusmutans (*16 rRNA gene, c serotype carious dentin) and ATCC4356 Type strains *Lactobacillus acidophilus* obtained from (Microbiological Resources Centre, Cairo, Egypt) (Cairo MIRCEN**)** were cultured onto the Brain Heart Infusion broth (BHI, Oxoid, and Basingstoke, England**)** at 37°C in an anaerobic Co2 incubator (Binder, German**)** and used as inoculums. The growth of both microorganisms was confirmed by turbidity of the suspension and then adjusted to 0.5 McFarland's turbidity standard (Densimat, BioMerieux, France**)** (bacterial count 1.5 x 10^8**).** Then the inoculum's suspension that was 10 µL of adjusted brain heart infusion broth culture of each *Streptococcus mutans* and *Lactobacillus acidophilus* was spread using two glass sterile triangular spreader for each bacteria over Trypticase soy yeast extract Agar (Oxoid, and Basingstoke, England) for the growth of Streptococcus mutans and De Man, Rogosa and Sharpe (MRS) agar (Oxoid, and Basingstoke, England) for the growth of Lactobacillus acidophilus to get a lawn culture of both the bacteria and approved to dry for 10 minutes at room temperature for uniform dispersion.

Ten petridishes (90mm x 15mm) (Poland) were used for each bacteria **Revathi et al. (2), Alsamolly et al. (10) and Vimala et al. (17).**

Incubation of Specimens:

For the agar diffusion test, the prepared disc-shaped specimens were placed on culture agar plates and then the agar plates were incubated at 37° C in an Co2 anaerobic incubator(Binder, Germany). Each Petri dish was filled with two specimen from each material**, Revathi et al. (2), Şirinoğlu-Çapan et al. (2020), Alsamolly et al. (10) and Vimala et al. (17).**

Measurement of Inhibition Zones:

The antibacterial activity of **Group Ⅰ** and **Group Ⅱ** for each strains was evaluated by measuring the diameter of bacterial growth inhibition zones in millimeters by digital caliper (Steco, Germany) at 48 hours, 72 hours and 7 days. The measurement was repeated three times for each specimen and the mean was calculated. The reading of inhibition zones (mm) was performed with a precision of 0.05 cm using digital caliper, **Revathi et al. (2), Alsamolly et al. (10) and Vimala et al. (17)**

Statistical analysis:

Numerical data of inhibition zones were presented as mean and standard deviation (SD) values. Data were analyzed using one-way analysis of variance (ANOVA) followed by post-hoc test. The significance level was set at $p \leq 0.05$. Statistical analysis was done using statistical package for social sciences (SPSS) version 22 for windows.

Results

Intergroup comparisons mean and standrard deviation values of inhibition zone (mm) for S-PRG resin composite and Conventional glass ionomer cement. When groups Ⅰ and Ⅱ were compared, it was found that there was statistically significant difference between the S-PRG resin composite and conventional glass ionomer cement against *S. mutans* and *L. acidophilus* ($p \leq 0.05$) at all time interval at 48 hours, at 72hours and after one week. S-PRG resin composite had statistically significant largest mean values of inhibition zone in comparison to conventional glass ionomer cement against *S. mutans* and *L. acidophilus* ($p \leq 0.05$) at all time interval at 48 hours, at 72hours and after one week were presented in **Table (2)**.

Intragroup comparisons total mean and standard deviation values of inhibition zone (mm) for S-PRG resin composite and conventional glass ionomer cement in two different bacteria streptococcus mutans and lactobacillus acidophilus at all time interval at 48 hours, at 72hours and after one week. At all times, the S-PRG resin composite were shown to have a greater mean zone of bacterial inhibition indicating a higher antibacterial activity. The antibacterial activity of S-PRG resin composite showed a total mean of zone of inhibition of (8.78 ± 0.29)mm against *streptococcus mutans* and against *lactobacillus acidophilus* showed a total mean of zone of inhibition of (9.66 ± 0.29) mm. The antibacterial activity of conventional glass ionomer cement showed a total mean of zone of inhibition of (7.52 ± 0.22)mm against streptococcus mutans and against lactobacillus acidophilus showed a total mean of zone of inhibition of (7.58 ± 0.19) mm. The maximal inhibition zone diameter was visible after 72 hours with both S-PRG resin composite and conventional glass ionomer cement. However, this inhibitory activity showed a progressive reduction over a period of 7 days with both S-PRG resin composite and conventional glass ionomer cement as shown in **Table (3).**

Discussion

Dental caries is the most widespread multifactorial oral disease which lowers a patient's quality of life. Bacteria, the type of diet, the host's immune response, and disruption of the micro-ecological balance of dental plaque are the main causes of dental caries. *Streptococcus mutans* is the primary cariogenic bacteria that plays a vital role and responsible for the initiation of dental caries. *Lactobacillus acidophilus* is the pioneering microorganisms and responsible for the caries progression, especially in dentin. The development of secondary caries is one of the main reasons why dental restorative materials fail. In order to minimize bacterial growth and colonization, restorative materials should ideally be chosen to have antibacterial properties or enhancements to acid resistance **(2,3, 18, 19, 20).**

Fluoride release over time and interaction with tooth structure at the apatite crystal surface creates fluoride salts like calcium fluoride (fluoroapatite). This results in a caries-inhibiting effect. Fluoride has an anticariogenic action and can reduce acid formation.

Additionally, it inhibits demineralization and promotes remineralization. Fluoride's impact on the prevention of dental caries may result from the disruption of the ecological balance in the mouth, from preventing salivary glycoproteins from adhering to hydroxyapatite, or possibly from directly limiting the growth of S. Mutans. S. Mutans' growth rate was inhibited by fluoride, even though glucose served as the main carbon and energy source. Enolase is necessary for the metabolism of glucose and lactose. It is the glycolytic pathway enzyme that is most fluoride sensitive. Therefore, restorative materials should have the ability to release fluoride in order to prevent the development and advancement of caries **(7, 10, 17, 20).**

The modern-day GIC is a biomimetic, tooth-coloured and fluoride-releasing material. It is the only dental restorative material which can bond chemically to the enamel and dentin. As result it is ideal for peripheral seal of deep cavitated lesions. Children and patients with dental fear or learning disabilities can

benefit greatly from the atraumatic restorative approach (ART). Antimicrobial property of GIC because of their fluoride release and/or their acidity (low pH)while setting. Also, the reduction in bacterial counts is not reliable obtained by placing conventional GICs in cavities; therefore, innovation was required to potentiate the antibacterial effect of GIC **(3, 21).**

S-PRG resin composite (Giomer) is a new hybrid, biocompatible, fluoride-releasing, resin-based dental material that improves the physical, mechanical, esthetic, and biological properties of glass ionomers. The acid-reactive pre-reacted glass (PRG) fluoroaluminosilicate particles(FASG) are reacted with polyalkenoic acid (PAA) in the presence of water forming a glass ionomer matrix construct, freeze-dried, milled, silanized, ground, used as fillers and then mixed with a dimethacrylate resin matrix can be considered as PRG technology. The reaction is detected in surface and are called surface reaction (surface reaction type, S-PRG fillers) **(2, 22).** Giomer materials of the third generation can be found in both conventional and flowable resin-based materials. The conventional form, Beautifil II LS (Shofu Inc.), combines the characteristics of resin composites and glass ionomers **(23).**

Giomer (PRG) filler is found to be a reservoir for fluoride ions play an important role for fluoride releasing and recharging abilities of the resin based materials **(24).**

Therefore, the present study was conducted to compare the antibacterial activity of S-PRG resin composite and conventional glass ionomer cement by observing the zone of inhibition around the samples in the culture plates by using an agar diffusion test against two different bacteria responsible for initiation and progression of caries *S. mutans* and *Lactobacillus acidophilus* respectively evaluated at three different time intervals.

The materials used in this in vitro study were S-PRG resin composite and conventional glass ionomer cement.

Beautifil® II LS (Low Shrink), a new generation of bioactive composite that is recommended for all restorations (Class I to Class V), incorporates bioactive Giomer chemistry, used in all Shofu restorative materials, which has been to sustain release and recharge fluoride and other beneficial ions, gives general dentists the ability to lessen polymerization volumetric shrinkage (0.85%) and shrinkage stress (2.72 MPa), greater strength, increased wear resistance, predictable and useful tooth-like aesthetics with natural fluorescence, as well as **Burtea et al. (25).**

IonoStar Molar is a new VOCO capsule design bulk glass ionomer providing improved access to a difficult-to-reach area in the mouth better than conventional application capsules for easy and clean automatic application. The material performs exceptionally well due to its non-sticky nature and marginal adaptation and no longer needs an activator. Simply pressing the capsule against a hard surface causes the coloured piston to move, which is then mixed as usual in a high-frequency mixer. It cures in five minutes. Due to its long-lasting high fluoride release, it reduces postoperative sensitivity. As well as its high abrasion resistance and compressive strength.

The agar plate diffusion is an accepted, effective and accurate method. The agar plate diffusion method enables bacteria to be examined in a standard, reasonable, and straightforward approach for identifying the resistance **(2, 10).**

The results of the current study showed that group Ⅰ; S-PRG resin composite (Giomer) showed statistically significant higher mean inhibition zone diameter values after 72 hours in comparison to group Ⅱ; conventional glass ionomer cement. This direct correlation between the S-PRG resin composite and the increase mean inhibition zone diameter values may be attributed to presence of S-PRG filler that has inhibitory effect on streptococcus mutans and the increase in the fluoride release.

The results of the current study are consistent with the findings **Hotwani et al. (26)** who evaluated the antibacterial activity of resin modified glass ionomer cement (GC Fuji II™ LC) and giomer (Beautifil-II) against *Streptococcus mutans* after 24 h, 48 h, and 7 days for each group in triplicates. They found at all times, the giomer specimens were shown to have a greater mean zone of bacterial inhibition indicating a higher antibacterial activity. The maximal inhibition was visible after 24 hours with both giomer and RMGIC. However, this inhibitory activity showed a progressive reduction over a period of 7 days with both giomer and RMGIC. The rationale for the difference in fluoride release between the RMGIC

and the giomer could be related to the materials' porosity, which may have a big impact on how much fluoride is released. The higher resin content of RMGIC might serve as a barrier to the diffusion of fluoride and water. The amount of fluoride produced and the material's physical characteristics may affect the capacity of fluoride-containing restorative materials to prevent the growth of bacteria that cause cavities.

Antibacterial properties of restorative cements have been attributed to their fluoride release **(7).**

Results of the present study are in accordance with **Tiwari et al. (7)** who evaluated in vitro antibacterial activity for two conventional glass ionomer cements (GC II and GC IX), and a zirconia reinforced glass ionomer cement (Zirconomer) against Streptococcus mutans after 48 hours using the agar inhibition test. They demonstrated that statistically significant largest zone of inhibition was observed with reinforced GIC (Zirconomer) which had maximum antibacterial activity against Streptococcus mutans after 48 hours followed by two conventional glass ionomer cements (GC II and GC IX).

The antibacterial activity seems to be influenced by factors including the materials' chemical composition, low pH, and the release of F- and other ions **(27).**

Results of the this study are in accordance with **El-Dosoky et al. (28)** who evaluated the antibacterial activity in vivo study of the conventional glass ionomer and glass ionomer containing different concentrations of chlorhexidine 1% and 2% against streptococcus mutans and lactobacillus at baseline and after 7 days. They found that conventional glass ionomers were outperformed by chlorhexidine-containing glass ionomers in terms of antibacterial activity while maintaining the glass ionomer's mechanical properties.

The findings of the present study are in agreement with **Burtea et al. (25)** showed that the mix of the pre-reacted glass and the resin matrix has a significant impact on the physico-chemical characteristics (residual monomer, fluoride release) of the giomers. The pre-reacted glass produced by using an improved hydrophilic resin matrix and polyalkenoic acid containing L-leucine residue releases fluoride more quickly and for a longer period of time.

The findings of the present study are in agreement with **Khere et al. (20)** who reported that all the GIC (Micron bioactive, GC Fuji IX GP Extra, Bioglass r) evaluated demonstrated antibacterial activity against *S. Mutans.* The improved antibacterial activity of GC Fuji IX GP Extra was proven. Therefore, it might be helpful for people with a high caries risk.

The data of the current study coincide with **Tartici et al. (29)** that showed the antibacterial activity was highest for Resin modified glass ionomer cement (Riva Light Cure) and Giomer (Beautifil II) in comparison to conventional glass ionomer cement (Riva chemical cure).

The results of the present study are complementary to the findings of **Nahar et al. (30)** who compared the effectiveness of Giomer with glass Ionomer cement in treating cervical caries clinically. They found that 2.5% had history of secondary caries formation of the study population with glass ionomer cement and no history of Secondary caries formation of the study population with giomer. Giomer restoration was more acceptable to patients than Glass ionomer for the treatment of cervical caries during a 12-month period.

There results of the present study are in disgreement with **Revathi et al. (2)** who reported that 24 hours, 48 hours and 7 days' time interval Giomer does not show any antibacterial properties against *S. mutans* and *L. acidophilus*. This may be explained firstly by the fact that using of giomer (Beautifil flow plus) while we use Beautifil® II LS (Low Shrink), a new generation of bioactive Giomer which has been to sustain release and recharge fluoride. Secondly, *L. acidophilus* and *S. mutans* were incubated microaerobically at 37 °C for 24 hours in a carbon dioxide jar while we use Co_2 anaerobic incubator (Binder, Germany) which is more accurate and efficient. Thirdly, while the chemical reaction of the cement continues the antibacterial capacity also increased throughout this time. Additionally, freshly mixed cement had greater antibacterial activity than set cement.

Conclusions

The following conclusions could be drawn from the present in vitro study's findings S-PRG resin composite (Giomer) application has excellent antibacterial activity and caries inhibitory effect, S-PRG resin composite (Giomer) makes good candidates for use in dental applications.

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Table (1): Materials Specifications, Composition, Manufacturer and lot number

Material	Specification-S	Composition	Manufacturer	Lot number
Beautiful II LS	Lightcured (S-PRG) resincomposite (Giomer) (Shade A2)	Mixture of glass powder (The pre-reacted glass(PRG) fluoro-aluminosilicate particles are added to poly acids forming a glass ionomer matrix construct), Urethane diarylate, Bis-MPEPP, BisGMA,TEGDMA, Polymerization initiator, Pigments	SHOFU Inc., Kyoto, Japan www.shofu.com	032145
Ionostar molar	Chemically cured conventional glassionomer cement(Shade A2)	Fluoroaluminosilicate glass, polyacrylic acid, tartaric acid	VOCO GmbH, Cuxhaven, Germany. www.voco.com	2104600

Table (2): Intergroup comparisons mean and standrand deviation values of inhibition zones

	S-PRG resin composite (GI)		Conventional Glass Ionomer Cement (GII)		P –value
	Mean ± S.D.		Mean ± S.D.		
	Str. Mutans	*Lb. acidophilus*	*Str. Mutans*	*Lb. acidophilus*	
48 hr	8.79 ± 0.2804	9.55 ± 0.3396	7.47 ± 0.2401	7.46 ± 0.1629	≤ 0.05*
72 hr	8.97 ± 0.3064	9.97 ± 0.2146	7.73 ± 0.1825	7.98 ± 0.2136	
7 days	8.58 ± 0.2707	9.44 ± 0.3182	7.37 ± 0.2254	7.31 ± 0.1966	

*S.D.: Standrand deviation P:Propability value; * significant (**P** ≤ 0.05)*

Table (3): Intragroup comparisons of total mean and standrand deviation values of inhibition zones

Bacteria	S-PRG Resin Composite Group I	
	Time	Mean ± SD
Streptococcus mutans	48 hr	8.79 ± 0.28
	72 hr	8.97 ± 0.31
	7 days	8.58 ± 0.27
	Total	8.78 ± 0.29
Latobacillus acidophilus	48 hr	9.55 ± 0.34
	72 hr	9.97 ± 0.21
	7 days	9.44 ± 0.32
	Total	9.66 ± 0.29
Bacteria	**(CGIC) Group II**	
	Time	Mean ± SD
Streptococcus mutans	48 hr	7.47 ± 0.24
	72 hr	7.73 ± 0.18
	7 days	7.37 ± 0.23
	Total	7.52 ± 0.22
Latobacillus acidophilus	48 hr	7.46 ± 0.16
	72 hr	7.98 ± 0.21
	7 days	7.31 ± 0.20
	Total	7.58 ± 0.19

Figures

After 48 hours:

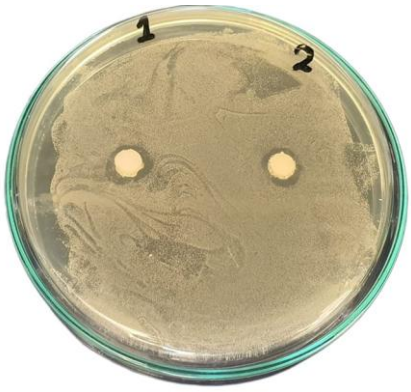

Figure 1: Zone of inhibition against *Streptococcus mutans*

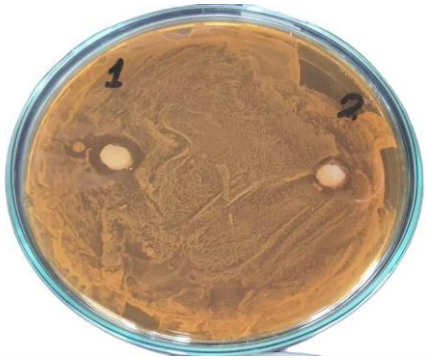

Figure 2: Zone of inhibition against *lactobacillus acidophilus*

After 72 hours:

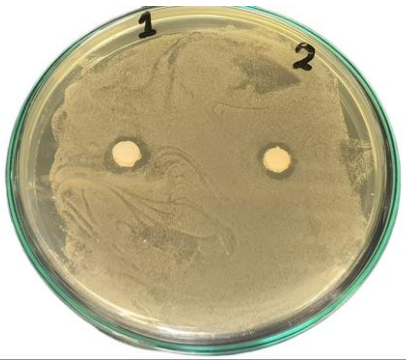

Figure 3: Zone of inhibition against *Streptococcus mutans*

Figure 4: Zone of inhibition against lactobacillus acidophilus

After one week:

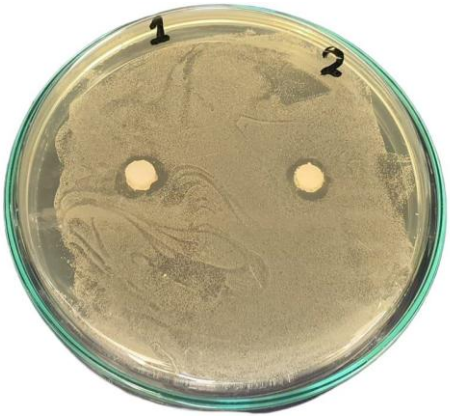

Figure 5: Zone of inhibition against *Streptococcus mutans*

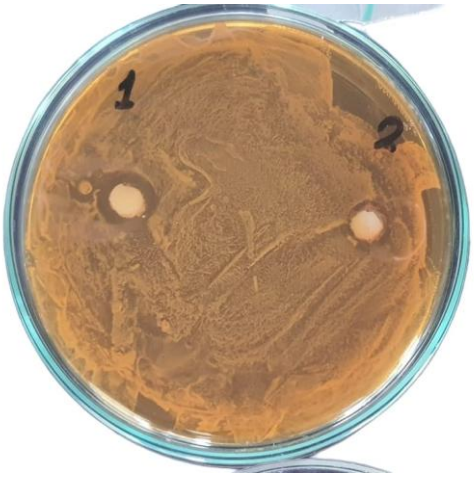

Figure 6: Zone of inhibition against *lactobacillus acidophilus*

Against *Streptococcus mutans:*

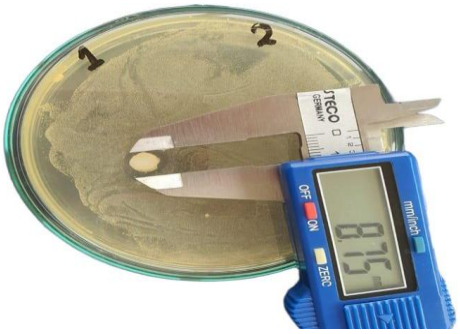

Figure 7: Measurement of inhibition zone around S-PRG resin composite (Group Ⅰ)

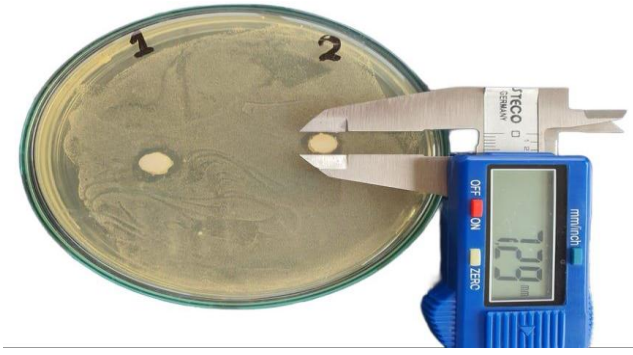

Figure 8: Measurement of inhibition zone around conventional glass ionomer cement (Group Ⅱ)

Against *lactobacillus acidophilus*:

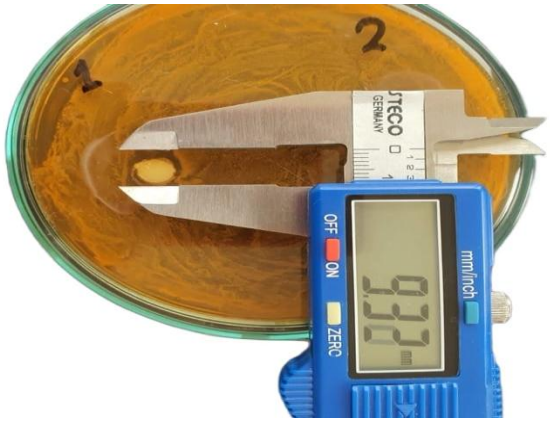

Figure 9: Measurement of inhibition zone around S-PRG resin composite (Group Ⅰ)

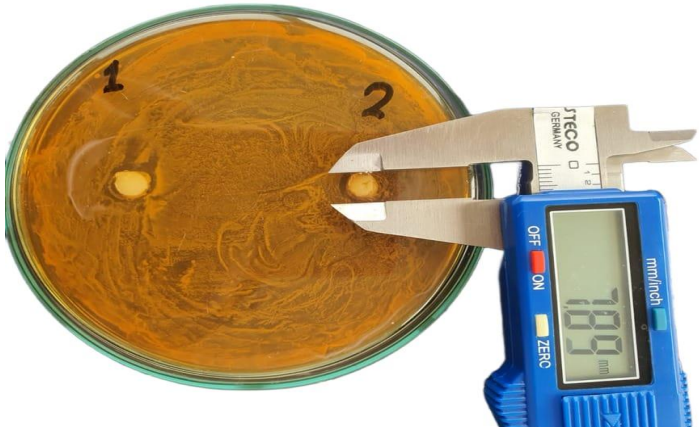

Figure 10: Measurement of inhibition zone around conventional glass ionomer cement (Group Ⅱ)

Figure legends:

Figure 1: Zone of inhibition against *Streptococcus mutans*

Figure 2: Zone of inhibition against *lactobacillus acidophilus*

Figure 3: Zone of inhibition against *Streptococcus mutans*

Figure 4: Zone of inhibition against lactobacillus acidophilus

Figure 5: Zone of inhibition against *Streptococcus mutans*

Figure 6: Zone of inhibition against *lactobacillus acidophilus*

Figure 7: Measurement of inhibition zone around S-PRG resin composite (Group Ⅰ)

Figure 8: Measurement of inhibition zone around conventional glass ionomer cement (Group Ⅱ)

Figure 9: Measurement of inhibition zone around S-PRG resin composite (Group Ⅰ)

Figure 10: Measurement of inhibition zone around conventional glass ionomer cement (Group Ⅱ)