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Comparison of antibacterial efficacy of different restorative materials – An *in vitro* study

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Abstract

Aim of the Study: The purpose of this *in vitro* study was to evaluate and compare the antibacterial efficacy of different restorative materials against *S. mutans* which is an important cariogenic bacteria present in dental plaque.

Methodology: A Direct Contact Test was performed to test the antibacterial efficacy of various commercially available restorative materials. They were divided into the following groups:

Group I – Saline (Control)

Group II – Composite

Group III – GIC

Group IV – DPI Amalgam Alloy

Group V – Dispersalloy

Results: GIC and Amalgam displayed superior antibacterial properties when compared to Composite and in amalgam high copper amalgam displayed better antibacterial properties than low copper amalgam.

Statistical Analysis: Statistical analysis was done using one way analysis of Variance - ANOVA test.

Conclusion: So within the limitations of the present study it can be concluded that GIC showed highest antibacterial efficacy followed by DPI Amalgam alloy and Dispersalloy for the first and second day and after which all the test materials showed equal antibacterial efficacy, whereas Composite showed least antibacterial efficacy for all the seven days.

Keywords: Comparison, antibacterial efficacy, restorative materials

1. Introduction

The development of secondary caries, pulpal inflammation and gingivitis resulting from bacterial invasion under and/or around dental restorations is a major clinical problem in operative dentistry [1]. Dental plaque deposited on and around fillings is the source of the recurrence of human dental decay. The existence of a microspace (at the interface between the restorative material and the cavity wall) along which micro-organisms can penetrate. This promotes the recurrence of caries [2].

Acidogenic bacteria play the main role in the development of dental caries. *Streptococcus mutans* is the single most important organism in the initiation of dental caries. *Streptococcus mutans* (*S. mutans*) is the bacteria which is involved in the transition from nonpathogenic to cariogenic biofilm, although many other microorganisms also play a role in the pathogenesis of the dental caries [3]. Therefore, restorative material which is effective against *S. mutans* might be considered as an anticariogenic agent. The interaction between bacteria and solid surfaces often results in the attachment and subsequent proliferation of the microorganisms, culminating in a well-defined biofilm [4]. Restorative materials with longlasting antibacterial surface properties may reduce the biofilm and thus reduce disease recurrence. Due to this potentiality for leakage, any restorative/ base/liner material used in the mouth should have anti-bacterial properties in defense against bacterial migration [5].

So the purpose of this *in vitro* study was to evaluate and compare the *in vitro* antibacterial activity of different restorative materials against *S. mutans* which is an important cariogenic bacteria present in dental plaque.

2. Materials and Methodology

Streptococcus mutans was isolated from human saliva from a patient with carious lesion and were used in this study. The sample was inoculated in Mitis Salivarius Bacitracin agar (MSB Agar – Hi Media, Mumbai) and incubated at 37 °C in Candle Extinction Jar for 24 hours. Colonies resembling *Streptococcus Mutans* were identified by biochemical tests. The identified *Streptococcus mutans* bacteria strains were cultured aerobically overnight at 37 °C in 5 ml of brain–heart infusion broth (BHI) (Hi Media, Mumbai). *Streptococcus mutans* culture was adjusted to 0.5 McFarland standard (bacterial count 1.5×10^8) from overnight growth of *S. mutans* on brain–heart infusion broth (Himedia, Mumbai). Four test materials namely Composite - Ivoclar Vivadent Inc., GIC - Gold Label Type 2 Universal Restorative GIC - GC, DPI Amalgam – Mumbai, Dispersalloy - Dentsply Caulk and one control group Saline were used in the study. They were divided into five groups
 Group I – Saline (Control)
 Group II – Composite
 Group III – GIC
 Group IV – DPI Amalgam Alloy
 Group V – Dispersalloy
 Sterile durhams tube (Hi Media, Mumbai) were obtained and a 3mm marking from the base was done on it and all the test materials were placed until the marking. 100µl of adjusted culture was added over test materials placed in the durhams tube and they were incubated at 37 °C in moist chamber. Each day 250µl of sterile BHI broth added to each durhams tube and mixed and 10µl transferred to 990µl of BHI broth and vortexed. 100µl of the dilution is transferred to BHI agar and spread using sterile L spreader. The incubation of all the plates

was carried out in Candle Extinction jar for 24 h at 37 °C. 240µl of culture is discarded from the durhams tube containing the test materials and 250µl of fresh BHI broth was added and incubated at 37 °C in Candle extinction jar. After incubation, the bacterial colonies were counted from the plates using digital colony counter and the CFU/ml was calculated based on the dilutions performed. Statistical analysis was done using ANOVA test.

3. Results

Table 1: Bacterial count of each group for seven days

	Group I	Group II	Group III	Group IV	Group V
Day 1	3342	1424	48	636	386
Day 2	3320	1398	14	142	66
Day 3	3450	1467	0	0	0
Day 4	3370	1480	0	0	0
Day 5	3360	1437	0	0	0
Day 6	3472	1388	0	0	0
Day 7	3380	1420	0	0	0

Table 2: Mean bacterial count and statistical analysis

Groups	Mean Bacterial Colony Count over seven days	Standard Deviation	F Value†	P value
I	3384.9	55.891	913.1	<0.001*
II	1430.6	33.763		
III	8.9	18.032		
IV	111.1	237.413		
V	64.6	143.855		

*Statistically Significant
 †ANOVA

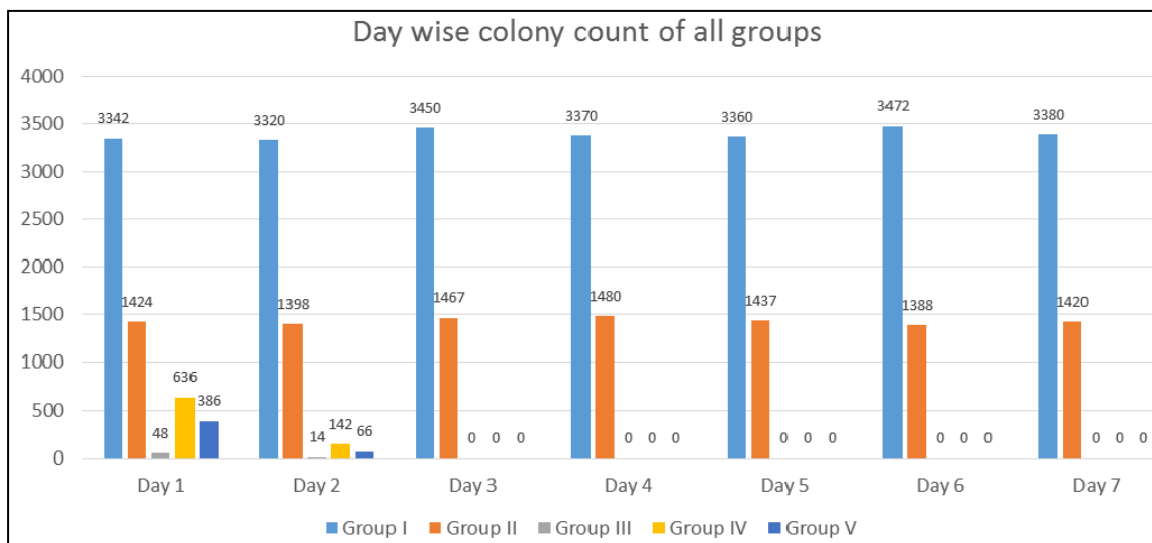


Fig 1: Day wise count

4. Discussion

The present study was conducted to evaluate the antimicrobial effectiveness of various commercially available restorative materials. The most frequently employed methods for testing the antimicrobial effect of dental materials are those based on direct contact test (DCT) [6]. 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. Composite restorative materials represent one of the many

successes of modern biomaterials research, since they replace biological tissue in both appearance and function. In the present study the composite material has no antibacterial effect after being cured, which may explain why composites accumulate more plaque than other filling materials. Sven-Ake Lundin *et al* in their investigative studies relating to the antibacterial properties of various restorative materials *in vitro* have revealed that the composite resins fail to produce zone of inhibition around them under laboratory conditions [7]. This is in agreement with the present study where the antibacterial efficacy of the composite resins is low when compared with other restorative materials.

Glass ionomer cements (GIC) have been widely used as a caries preventive aid. It is a biocompatible material, and it promotes both inhibition of demineralization and additional remineralization of tooth structures adjacent to fillings, as well as interferes with bacterial growth, stabilizing the microbiota despite the presence of fermentable carbohydrates^[8]. In the present study GIC has a fast and good antibacterial efficacy. GIC's fluoride releasing property has been documented in the literature^[9, 10]. After being released from GIC, the fluoride ions take part in the de- and remineralization phenomena, and may act directly on the carious process^[11, 12]. The different antibacterial properties of Glass Ionomer cements might be related both to varied compositions existent in these materials (presence or absence of oxides, type of acids present in the composition) and to fluoride releasing property. According to Vermeersch *et al.*, the low pH of GICs, during setting may contribute more to their antibacterial properties than their fluoride-leaching capabilities^[13]. Studies on the activity of different glass ionomer cements on cariogenic bacteria showed that, they all promoted growth inhibition of cariogenic bacteria assayed.

Dental amalgams are widely used as filling materials in restorative dentistry. In recent years, an increasing number of new amalgams, with different copper content, have become commercially available. These variations probably arise from differing bacterial sensitivity to the materials, although the precise source of the antibacterial effect is difficult to ascertain because of the complexity of amalgam structure and composition. In the present study two different amalgam alloys namely – DPI Amalgam Alloy - Mumbai and Dispersalloy - Dentsply Caulk were taken based on their copper content i.e, copper content in DPI Amalgam alloy is 26% and Dispersalloy is 11% to assess the effect of copper on antibacterial efficacy of amalgam. When the elements in amalgam alloy were compared an investigation suggests that the order of antimicrobial potential of elements in amalgams would be Hg, Cu and Zn^[14]. The lack of antibacterial activity of pure silver and tin, together with the absence of bacterial inhibition indicate that these metals contribute minimally, if at all, towards the antibacterial properties of amalgams. Opperman and Johnson have reported that copper accumulation in dental plaque can reduce plaque acidogenicity and inhibit the growth of strep. mutans and modern non gamma - 2 amalgam alloys release more copper than conventional low copper amalgam alloys at low pH^[15]. However, based upon these results, when the two amalgam alloys were compared i.e, DPI Amalgam Alloy - Mumbai and Dispersalloy - Dentsply Caulk - DPI Amalgam alloy which has higher copper content than Dispersalloy has more bacterial inhibition in the first two days when compared to Dispersalloy and it can be concluded that copper played an important role in antimicrobial activity of dental amalgams but this activity may probably depend on the release of these components from the amalgam and the presence of these components at the surface of the sample. This strong antibacterial effect confirms the observations of some previous workers such as Orstavik^[16].

5. Conclusion

So within the limitations of the present study it can be concluded that GIC showed highest antibacterial efficacy followed by DPI Amalgam alloy and Dispersalloy for the first and second day and after which all the test materials showed equal antibacterial efficacy, whereas Composite showed least antibacterial efficacy for all the seven days.

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