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Comparison of Anti-microbial Efficacy of 3.8% Silver Diamine Fluoride and 3% Sodium Hypochlorite as Irrigant in Primary Molars-A Combined in Vitro and in Vivo Study.

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Abstract

Introduction: Due to different anatomy of root canals in primary molars, complete disinfection is impossible so newer irrigants like Silver diamine fluoride (SDF), are required as adjunct during the endodontic procedures.

Objective: Comparison of the anti-bacterial action of 3.8% silver diamine fluoride (1:10 dilution) and 3% sodium hypochlorite as an irrigant against Enterococcus faecalis.

Method: Study was planned in 2 parts. In the first part, root canal opening was done on extracted teeth and were inoculated with ATCC E. faecalis. Canals were prepared till universal hand K files 40 no. and were irrigated with 3.8% silver diamine fluoride and 3% sodium hypochlorite. In second part pulpectomy was done in 30 primary carious molars. Root canals were irrigated with 3.8% SDF and 3% Hypochlorite and samples were taken with paper points before and after irrigation & given for the culture of E-faecalis. The CFU was compared and the efficiency of irrigants was tested.

Result: There was marked reduction in the colony counts in the post-procedure samples with both of the anti-microbial solutions, the silver diamine fluoride and sodium hypochlorite.

Conclusion: Newer better and effective irrigating agents can help disinfecting the root canal of primary molars making pediatric dentistry less cumbersome.

Keywords: Antimicrobial efficacy, Silver Diamine Fluoride, Sodium Hypochlorite, Enterococcus faecalis.

Introduction

The elimination of intracanal microorganisms is essential for the long-term success of root canal

treatment[1] which can be achieved through mechanical cleaning and shaping in conjunction with irrigation through antibacterial agents.[2,3] The use of an antimicrobial inter appointment dressing is also significant for bacterial elimination within the root canal space.[4] Calcium hydroxide is the most widely used material for reduction of bacterial activity followed by chlorhexidine, phenolic derivatives or formocresol.[5] Calcium hydroxide has high alkaline nature which is responsible for altering the biologic properties of lipopolysaccharides in the cell walls of gram-negative bacteria.[6] However, Enterococcus faecalis has been reported to be resistant to alkaline stress. This is due to its ability to penetrate the dentinal tubules and possible interspecies communication. [7,8] Various materials have been used for elimination of Enterococcus faecalis from root canals since many years. Recently, antimicrobial silver compounds had been advocated for this purpose.[9]

Silver fluoride regimens were found to be effective in inhibiting the growth of cariogenic bacteria because it has antimicrobial effect and can cause deposition of silver compound in the root canals.[10,11] A 3.8 w/v% silver diamine fluoride (Ag[NH3]2F) solution (Saforide RC; Bee Brand Medico Dental Co Ltd, Osaka, Japan) has been developed for intracanal irrigation which represents 1:10 dilution of the original 38% Ag(NH3)2F solution used for root canal infection.[12]

In the present study, the effect of antibacterial efficacy of 3.8% Ag(NH3)2F solution was evaluated by using a single-species E. faecalis biofilm against 3% sodium hypochlorite.

Materials and Method

Study was planned in 2 parts, in vivo and in vitro.

In vitro study:

Root canal opening was done in extracted molars. Canals were prepared upto universal hand K files 40 no. with proper cleaning and shaping. Teeth were given for inoculation with E-faecalis in microbiology lab. A standardized suspension of E. faecalis (Mc Farland's Standard) was prepared by sub-culturing American Type Cell Culture (ATCC) strain 29212 in Brain Heat Infusion (BHI) broth. All samples except 2 were used as negative control and they were individually immersed in 5 milliliters BHI broth containing 1 milliliter of bacterial suspension. The inoculated teeth were stored at 37°C and 100% humidity for 72 hours to ensure the growth of E-faecalis. According to protocols, canals were irrigated with 3% sodium hypochlorite (H group) and 3.8% SDF (S group) and control (C group) (SDF used was diluted in the distilled water in 1:10 ratio.) as seen in figure 1. Samples were collected in the transport medium i.e. peptone water and given for culture. The E-faecalis bacteria was isolated and CFU was counted from the petridish.[13]

In vivo study

30 primary carious molars were selected.

Inclusion criteria

- Carious primary molars indicated for pulpectomy.

Exclusion criteria

- Medically compromised patients.
- Children undergoing orthodontic therapy.
- Any signs of root resorption (less than 2/3rd root length)
- Intraradicular bone loss
- Patient allergic to silver ions

Root canal opening was done and canals were prepared up to universal hand K files 40 no. Canals were irrigated using both the solutions namely Silver Diamine Fluoride and Sodium Hypochlorite. Samples were taken with paper points pre and post irrigation (A

& B group) as seen in the figure 2 and collected in suitable transport media (Peptone water). Samples were sent for culture of E-faecalis to microbiology lab. The CFU was compared and the efficacy of irrigant was tested using statistical analysis using Independent and Paired t-test.[13]

Results

The scores obtained were stored in an excel spread sheet (Microsoft, Inc., Redmond, Wash) and statistical analysis was carried out using Statistical Package for Social Sciences (SPSS) 20. Independent t-test and Paired-t test were used with significant p value <0.05.

In in-vitro study except for control group post-procedure cultures from the test samples showed no growth with either of the irrigants as seen in table I. Thus, the initial pre sampling showed the E-faecalis count of >105 which was reduced to NIL when irrigated with Silver Diamine Fluoride.

In in-vivo study 30 samples were collected after preparing the canal till 40 K file using only saline as an irrigant. 30 samples were collected using Sodium Hypochlorite and SDF as a irrigating agent with 15 samples in each group i.e. 15 in group A and 15 in group B. By statistical analysis, it was found that there was no significant difference between the two groups i.e. A group and B group for present samples, but there was significant difference within the groups i.e. in between the samples of the same group. There was marked reduction in the colony counts in the post-procedure samples with both of the anti-microbial solutions, the silver diamine fluoride and sodium hypochlorite as seen in table II.

Discussion

In our study we have evaluated that antimicrobial efficacy of silver diamine fluoride (3.8%) is equivalent to that of the sodium hypochlorite. We have used 3.8% SDF in 1:10 dilution with distilled water as to sterilize the root canals which is similar to the study done by Hiraishi et al in 2010. They reported that there was no change in its antibacterial efficacy even after dilution.

Also we have found a significant reduction of E-faecalis count by using SDF as an irrigant, the reason might be compared to the same study by Hiraishi et al in 2010 in which they stated that the presence of silver deposits in dentinal tubules suggested the possibility for the Ag(NH3)2F to penetrate and reduce/eliminate biofilms formed in dentinal tubules.[13,14]

We have done an in vitro study on extracted molars so as to confirm the efficacy of 3.8 % SDF as an irrigant in root canal so that later it can be used in patients which was similar to the study done by Mathew et al in 2012. They did a study which concluded that both 3.8% silver diamine fluoride and 2% chlorhexidine showed a superior capacity to sterilize the root canals than control groups. Thus, the use of SDF as an endodontic irrigant is feasible as it can effectively remove the microbe present in the canal and circumpulpal dentin. [14,15]

Noriko et al in 2010 studied the effect of 3.8% SDF and sodium hypochlorite on E. faecalis biofilm. They reported 100% efficiency of 3.8% SDF against E. faecalis after a direct 60-minute exposure.[13]

It has been shown by Tanaka et al in 1970, that an aqueous solution of AgF has powerful disinfectant and protein coagulating actions, and also has a considerably potent action, which occludes the dentinal tubules of root canal wall in terms of the electric resistance.[16] Silver in SDF interacts with the sulphydryl and thiol groups present in the bacterial amino acids and nucleic acids. [17,18] This inhibits cell division, cellular respiration, metabolism, and biofilm formation. Several investigators have studied the cellular mechanisms that

are affected by silver, thus proving its effectiveness as an anti-bacterial agent. [19,20,21] Silver Diamine fluoride has antibacterial properties and is as efficient as other irrigants in cleaning the root canals. With several benefits silver Diamine fluoride comes with a drawback of discoloration of teeth and soft tissue. In this study as it was used in the canals there was less concern of discoloration of teeth. Also as a precautionary measure we have irrigated the root canals thoroughly with 0.9% saline after irrigation with SDF. But to overcome this limitation Knight et al in 2005 proposed the use of Potassium iodide after application of SDF to the tooth structure. The free silver ions in solution will react with Potassium Iodide to precipitate as a creamy white silver iodide crystals. Hence, free silver ions will be no longer available to react with sulfur and other reagents in the mouth to form black precipitates into the teeth, thereby removing the discoloration.[22] Further research in this direction is still required.

Conclusion

Thus, silver diamine fluoride can be considered as an effective solution having equivalent anti-microbial efficacy to sodium hypochlorite. The mechanical advantages of the use of SDF as an intra-canal medicament and its further action to prevent re-infection of the root canal by preventing biofilm formation and proliferation has to be the focus of future research in this field.

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Legends Tables And Figures

Test id	Pre-procedure counts (CFU/mL)	Post-procedure counts (CFU/mL)
C1	$>10^5$	$>10^5$
C2	$>10^5$	$>10^5$
S1	$>10^5$	NIL
S2	$>10^5$	NIL
H1	$>10^5$	NIL
H2	$>10^5$	NIL

Table 1: Pre and Post procedural count of E-faecalis in control group

Samples	Mean	SD	*p* value
Pre procedure A group	78000	27307.7	0.00015
Post procedure A group	34666.67	30,675	
Pre procedure B group	74666.67	27996.5	0.00001
Post procedure B group	15333.33	21336.3	
Post procedure difference between A and B group			0.57

Table 2: Significant reduction in CFU of *E-faecalis* in pre and post sampling of A group and B group.

Figure 1: Study done extracted molars.

Figure 2: Sample taken from patient in paper Point

Figure 3: Samples stored in peptone water.

Figure 4: inoculation of E-Faecalis in chocolate agar