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Evaluation and Comparison of the Antimicrobial Efficacy of Silver Diamine Fluoride, Stannous Fluoride, and Sodium Fluoride Using Tube Dilution Method: An in-vitro Study

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Abstract

Aim: The study aimed to evaluate and compare the antimicrobial efficacy of three fluoride materials using the tube dilution method.

Materials and Method: The materials evaluated for antimicrobial efficacy were Silver diamine fluoride (Kids-e-Dental, e-SDF, Mumbai), Stannous fluoride, and Sodium fluoride (Madras Fluorine Private Ltd, Chennai). The antimicrobial testing was done by tube dilution method. The materials were prepared in two-fold dilutions and streptococcus mutans was inoculated into each dilution. The results were read at 24 hours and 48 hours. There was a statistical difference between the materials at both the time intervals (24h and 48h) with P-values 0.0010 and 0.0010 respectively.

Conclusion: SDF proved to be a good antimicrobial activity with a minimal inhibitory concentration of0.156mg/ml.

Keywords: Silver Diamine Fluoride, Stannous Fluoride, Sodium Fluoride, Tube Dilution Method.

Introduction

Dental caries refers to the localized destruction of susceptible dental hard tissues by acidic by- products from the bacterial fermentation of dietary carbohydrates. The main pathogen responsible for dental caries is Streptococcus mutans. The progression of dental caries and prevention of the occurrence of dental caries is possible by several approaches. Most commonly employed are effective oral hygiene measures, fluoride application, pit-and-fissure sealants, use of xylitol, and the development of a dental caries vaccine[1]. Fluorides play an important role in the control of dental caries lesions, by reducing demineralization and promoting remineralization.[2]

Bibby began era of topical fluorides with the use of a solution of 0.1% sodium fluoride (NaF).Subsequently, over the years, various other topical fluoride agents such as stannous fluoride(SnF2), acidulated phosphate fluoride (APF), varnish-containing fluoride, and amine fluoride have evolved[3]. Fluorides such as sodium

fluoride, and stannous fluoride varnishes are used as preventive reagents because of their remineralization and antimicrobial abilities[3]. Silver diamine fluoride known as SDF, is used as a cavity cleanser, which has many beneficial effects such as inhibition of demineralization, conservation of collagen from degradation, increasing microhardness of dentine post-application, and inhibiting the active growth of cariogenicbacteria[4]

Silver diamine fluoride (SDF) has an anti-cariogenic effect because it facilitates remineralization by inhibiting biofilm formation and caries prevention[5].The product was cleared for sale in the United States in August 2014 by the Food and Drug Administration as a Class II medical device[6].The combined effects of silver and fluorides in SDF have been hypothesized to have the ability to prevent caries progression and the development of new caries simultaneously[7].Hence, this study estimates the minimal concentration at which these fluorides are antibacterial against streptococcus mutans. Thus, the study aims to evaluate and compare the antimicrobial efficacy of silver diamine fluoride, stannous fluoride, and sodium fluoride against streptococcus mutans.

Materials and Methodology

The materials evaluated for antimicrobial efficacy were Silver diamine fluoride (Kids-e- Dental, e-SDF, Mumbai), Stannous fluoride, and Sodium fluoride (Madras Fluorine Private Ltd, Chennai). The antimicrobial testing was done by tube dilution method. All the procedures were carried out under sterile precautions in a bio safety cabinet.

The master dilution (10mg/ml) was prepared by dissolving 200 milligrams of the material in20ml of sterile Brain Heart Infusion (BHI) Broth. Further dilutions of 5mg/ml, 2.5mg/ml,1.25mg/ml, 0.0625 mg/ml, 0.0312mg/ml and 0.0156mg/ml were prepared. Ten microfibers of standardized streptococcus mutans were added to all the dilutions prepared. One group was positive control in which 10 microliter of s mutans suspension was mixed with 1ml of brain heart infusion (BHI) broth without any materials. A negative control group had 1 ml of brain heart infusion heart (BHI) broth without s mutans. One last group was used to check for the sterility of the material. The tubes were then incubated at 35°C for 24 hours. Subcultures from each of the test tubes were made on Brain Heart Infusion (BHI) agar and read at the end of 24and 48 hours of incubation. This experimental procedure was repeated 3 times.

Results

The broths from all the tubes were plated on Brain Heart Infusion agar plates and incubated for 24 hours and 48 hours before reading. Silver diamine fluoride was inhibitory to streptococcus mutans at the lowest concentration (0.156mg/ml). Stannous fluoride and sodium fluoride were not inhibitory to streptococcus mutans at any dilutions [Table 1]. The experimental results were repeated 3 times and results were consistent in all the periods.

Discussion

Fluorides such as sodium fluoride, and stannous fluoride varnishes are used as preventive reagents because of their remineralization and antimicrobial abilities[2].A newly available fluoride i.e., Silver Diamine Fluoride (SDF) has been evaluated for antimicrobial efficacy against streptococcus mutans in this study. The minimum concentration at which the fluorides inhibit the growth of bacteria was evaluated by using the tube dilution method and is mentioned as Minimal Inhibitory Concentration (MIC). It is an efficient method for evaluating as there is a direct contact between the microorganisms and experimental materials which allows a more realistic interaction[8].

The results of this study showed that the MIC of Silver diamine fluoride against streptococcus mutans was found to be 0.156mg/ml. stannous fluoride and sodium fluoride were not inhibitory to streptococcus mutans at any dilutions.38% SDF is a colourless solution containing 44,800 ppm fluoride ions[9]. Its fluoride concentration is the highest among the fluoride agents available for dental use. SDF contains silver and fluoride, which form a complex with ammonia. Ammonia can keep the solution at a constant concentration for a certain period. It has been suggested that silver ions at a concentration of 20 ppm can inhibit the growth of S. mutans. It concluded that silver ions have three antimicrobial effects: first, silver ions can destroy the cell wall structure of bacteria second, they can inhibit enzyme activities and influence metabolic processes; and, third, they can inhibit the replication of bacterial DNA. Fluoride is used to prevent caries. Hence, the combined effects of silver and fluorides have been hypothesized to have the ability to prevent caries progression and the development of new caries simultaneously[7]. SDF has been used to manage caries in particular in patients with high caries prevalence. Silver Nanoparticles have shown a great inhibitory effect on the growth of cariogenic bacteria, which might be an important reason why SDF can arrest caries even without the removal of carious tooth structure. However, the main disadvantage of its use is discoloration of carious teeth, which can cause patient dissatisfaction. Some researchers have proposed using potassium iodide after topical application of SDF to reduce the staining effect by generating silver iodide. However, this white product, silver iodide, is considered to be photosensitive and can turn dark with exposure to light[7]. Thus, the advantage of SDF over other fluorides is that in addition to having similar anti-bacterial properties, the interaction of SDF to teeth results in the synergistic formation of fluoroapatite. The amount of fluoride released is double that seen with other fluoridating agents. This adds to the anti-bacterial activity and may result in increased longevity and help to prevent caries.

Conclusion

In conclusion, within the limitations of the present study, silver diamine fluoride showed that the MIC against streptococcus mutans was found to be 0.156mg/ml. stannous fluoride and sodium fluoride were not inhibitory to streptococcus mutans at any dilutions. The antibacterial activity of stannous fluoride and sodium fluoride will probably be inhibitory at higher concentrations of material.

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Legend Tables and Figures

SL. No	Dilution	Silver diamine fluoride	Stannous Fluoride	Sodium Fluoride
Tube No. 1	10 mg/ml	NG	Growth	Growth
Tube No. 2	5 mg/Ml	NG	Growth	Growth
Tube No. 3	2.5 mg/Ml	NG	Growth	Growth
Tube No. 4	1.25 mg/mL	NG	Growth	Growth
Tube No. 5	0.625 mg/mL	NG	Growth	Growth
Tube No. 6	0.313 mg/mL	NG	Growth	Growth
Tube No. 7	0.156 mg/mL	NG	Growth	Growth
Positive Control	-	Growth	Growth	Growth
Negative Control	-	NG	NG	NG
Sterility Control	-	NG	NG	NG

Table 1

Figure 1