

Antibacterial, Antiplaque and Anticaries Effect of Silver Diamine Fluoride and Fluoride Varnish in Children

Saloni Verma¹, Suma Sogi^{2*}, Shailja Chatterjee³, Priyanka Sharma⁴, Abhishek Dhindsa⁵, Roopam Kapoor⁶

¹MDS, (Ex- Pg), Department of Pediatric and Preventive Dentistry.

²MDS, Professor and HOD, Department of Pediatric and Preventive Dentistry, Maharishi Markandeshwar College of Dental Sciences and Research (M.M.C.D.S.R.), Mullana, Ambala (Haryana). E-mail: hpsumasogi@gmail.com

³MDS, Professor, Department of Oral and Maxillofacial Pathology, Yamuna Institute of Dental Sciences and Research (YIDSR), Gadholi, Yamunanagar (Haryana).

⁴MDS, Reader, Department of Pediatric and Preventive Dentistry, Maharishi Markandeshwar College of Dental Sciences and Research (M.M.C.D.S.R.), Mullana, Ambala (Haryana).

⁵MDS, Professor, Department of Pediatric and Preventive Dentistry, Swami Devi Dyal Dental College, Panchkula (Haryana).

⁶MDS, Lecturer, Department of Pediatric and Preventive Dentistry, Maharishi Markandeshwar College of Dental Sciences and Research (M.M.C.D.S.R.), Mullana, Ambala (Haryana).

Abstract

Aim: To compare the antibacterial, antiplaque and anticaries effect of silver diamine fluoride (SDF) and fluoride varnish in children.

Materials and Method: A total of 54 children, 12-14 years of age were included in the study. Children were divided into 3 Groups: - Group 1: SDF, Group 2: Fluoride Varnish, Group 3: Normal Saline. All the children were evaluated for *S. mutans* count and Plaque score after 72 hours, 3 and 6 months.

Result: In Group 1, statistically significant reduction in mean *S. mutans* count was seen after 3 days, 3 months and 6 months. However in group 2 and 3 significant reduction in mean *S. mutans* count was observed only after 3 days. A significant reduction in mean plaque score was seen after 3 days and 3 months in group 1, after 3 days in group 2 whereas after 3 days and 3 months in group 3. Intergroup comparison shows non-significant difference in mean *S. mutans* count and plaque score between group 1 and 2, while statistically significant difference in mean *S. mutans* count was found between group 1 and 3 and group 2 and 3 after 3 days, 3 and 6 months. Moreover, a significant difference was found in mean plaque score between group 1 and 3 and group 2 and 3 after 3 days, 3 and 6 months excluding a non-significant value after 3 months between group 2 and 3.

Conclusion: In vivo application of SDF and Fluoride varnish results in significant reduction in *S. mutans* count, plaque score and no carious lesion was detected after the follow-up time intervals.

Keywords: Silver Diamine Fluoride (SDF), Fluoride Varnish, Dental Caries.

DOI: 10.47750/pnr.2022.13.S03.245

INTRODUCTION

Dental caries is a well-recognized multifactorial disease. *Streptococcus mutans* (*S. mutans*) plays a significant role in the progression of dental caries. Several studies have already revealed high number of *S. mutans* in the saliva and plaque in children with active carious lesions as compared to caries free children. So, to reduce dental caries various studies have focused on the prevention of *S. mutans* colonization.^[1,2]

Comprehensive management of dental caries include management of disease and the lesion as well. A more focus on preventive strategies as minimally non-invasive methods are given.^[3] Earlier, Bibby in 1942 presented topical fluoride in the form of 0.1% Na F solution.^[4] Several topical fluorides were introduced and were significantly

proved as an important and effective agent against dental caries.

In 1964, Schmidt introduced Fluoride Varnish to dentistry.^[5] Since then, it became the most widely used professionally applied topical fluoride agent in Europe. Fluoride Varnish applied professionally two to four times a year would substantially reduce tooth decay in children. However, more vigorous research is needed to show significant results in the treatment.^[6]

Even though there are multiple topical fluoride agents, still none of them have proved satisfactory. A relatively new fluoridated agent SDF with its hypothesized ability to halt the carious process and simultaneously prevent the formation of new caries has been used in Japan since 1970's. SDF proves to be safe, effective, competent and could become a vital component for comprehensive and

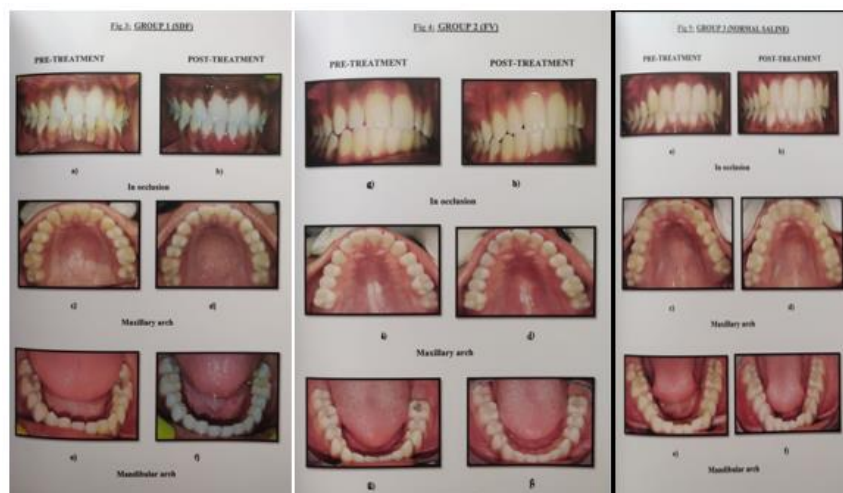
effective preventive programs that fulfill the WHO Millennium Goals. [7,8] Topical application of SDF has received much attention in the past years and this could be due to its ease of application and low cost.[9] Hence, this present study was conducted with an aim to evaluate antibacterial, antiplaque and anticaries effects of SDF and Fluoride varnish in Children.

MATERIALS AND METHOD

The present prospective randomized controlled study was conducted in the Department of Pediatric and Preventive Dentistry, M.M.C.D.S.R, Mullana. Total 144 children (both male and females) were screened from primary and secondary government funded schools in Ludhiana and Yamuna Nagar district, Haryana. Out of them, 54 children with an age group of 12-14 yrs and decayed missing filled teeth score >2 and <5 were included in the study. Children with special health care needs, ulcerative gingivitis/stomatitis, with known silver allergy and whose parents were not willing to participate were excluded from the study. Informed written consent was taken from parents of all the participants and patient record form was filled. Ethical approval for study design and protocol was taken from Institutional Ethical Committee of MMCD SR, MMDU, Mullana.*

MAHARISHI MARKANDESHWAR UNIVERSITY, MULLANA, AMBALA			
Communication of Decision of the Ethics Committee (IEC)			
			Project No: 908
Project title: Clinical study of antibacterial, antiplaque and anticaries effects of silver diamine fluoride and fluoride varnish in children			
Principal Investigator: Saloni Verma			
Name of Institution: MMCD SR, MMU, Mullana			
<input checked="" type="checkbox"/>	New review	<input type="checkbox"/>	Revised review
<input type="checkbox"/>		<input type="checkbox"/>	Expedited review
Date of review (D/M/Y): 30-06-2016 17.12.16			
Date of previous review, if revised application: [Signature]			
Decision of the Ethics Committee:			
<input checked="" type="checkbox"/>	Recommended	<input type="checkbox"/>	Recommended with suggestions
<input type="checkbox"/>	Revision	<input type="checkbox"/>	Rejected
Suggestions/ Reasons/ Remarks: To follow guidelines issued by DCGI Min of Health and family welfare from time to time			
Recommended for a period of : Two years			
Please note *			
- Inform EC immediately in case of any Adverse events and Serious adverse events.			
- Inform EC in case of any change of study procedure, site and investigator			
- This permission is only for period mentioned above. Annual report to be submitted to EC.			
- Members of EC have right to monitor the trial with prior intimation.			
			[Signature]
			Dr. B.K. Agrawal Member Secretary Ethics Committee M.M University

Children were divided into 3 groups on a random basis: Group 1- Study Group: SDF (38%w/v), Group 2- Control group: Fluoride varnish FV (6% NaF, 6% CaF₂) and Group 3- Placebo group: Normal saline was applied in the study group. (Fig 3,4,5)



Method of Saliva Collection

Baseline saliva samples were collected for study/control group. The resting whole saliva sample of the patient was collected using a sterile cotton swab, from all surfaces of the teeth and immediately transferred to a screw capped plastic tube containing Todd-Hewitt transport media. All tubes were immediately sealed. It was then transferred in

bacteriological laboratory for culture. S.mutans growth was studied on Mitis Salivarius agar culture plates. [10]

Following appropriate dental treatment for the decayed teeth, patients were evaluated for plaque score and S. mutans counts for all the three groups at baseline, 72 hrs, 3 months and 6 months time interval. Plaque index (Silness and Loe) and Mitis Salivarius agar media was used for

determining plaque score and *S. mutans* count respectively. After 6 months time interval, any broken restoration and new carious lesion was verified.

Clinical Procedure

Fluoride varnish was applied on the teeth with a thin brush. After Fluoride varnish has set, children were allowed to rinse with water. For SDF application, whole mucosal surface was covered with the vaseline, to protect it from mild burning sensation due to SDF. Isolation of the teeth was done with the help of cotton rolls & low volume suction. Application of SDF was done with small cotton pellet held with tweezer and applied for 3-4 minutes on all tooth surfaces in single quadrant at one time. The application was done in lower arch first (as saliva accumulates rapidly) starting from the most distal to mesial tooth followed by the upper arch. This procedure was repeated on all quadrants in a similar manner. As per manufacturer's instructions, after 4-5 minutes of application patient was allowed to rinse his/her mouth by gargling with normal saline. For Normal saline group, following the treatment, patient was allowed to rinse his/her mouth by gargling with normal saline.

Microbiological Procedure

The isolation of *S. mutans* was done in the Department of Oral and Maxillofacial Pathology, MMCDSR, Mullana. Mitis Salivarius agar media was prepared and autoclaved. 0.5% potassium tellurite was added in media and was poured into petri-dishes. Samples were streaked onto the agar plates which were inoculated at 37°C for 72 hours in an incubator. The colonies were identified by the morphological characteristics i.e., round, elevated, blue in colour ranging from pin-point to pin-head size with glossy surface and can be detached from the agar surface. (Fig 1) The *S. mutans* counts for all the three groups in CFU/ml were counted using the magnifying glass.

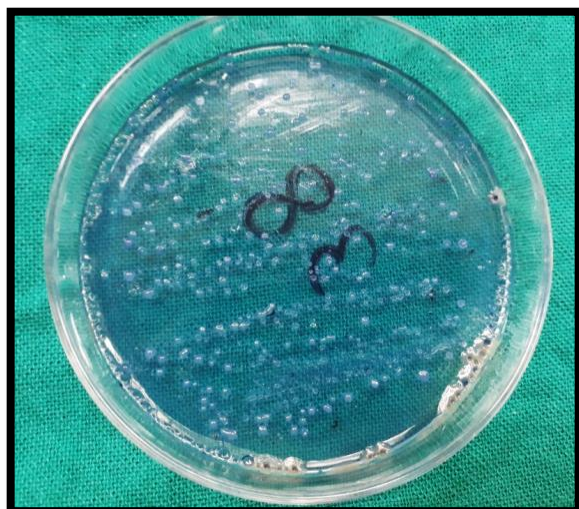


Fig. 1: *S. mutans* colony growth on mitis salivarius agar

RESULTS

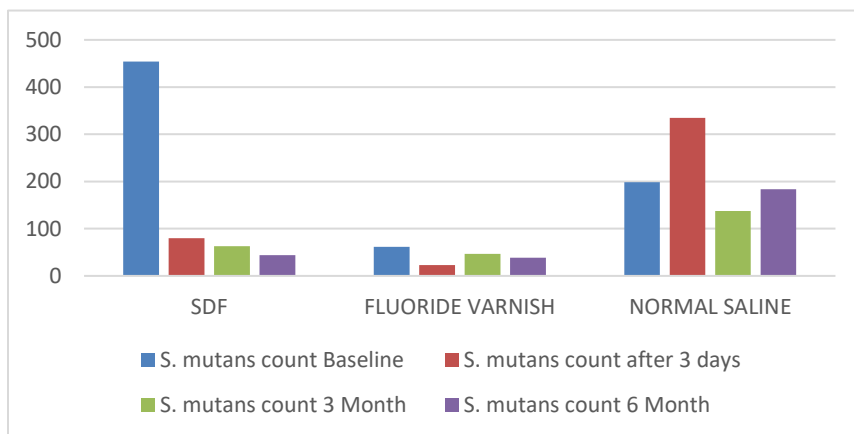
The present study included 54 children (32 were males and 22 females) age group of 12-14 years with the mean age of 13.5 years.

In group 1 (SDF), statistically significant difference was observed between the mean value of baseline *S. mutans* count and count after 3 days, 3 months and 6 months with *p* value (<0.001) and also between the mean value of baseline plaque score and score after 3 days, 3 months with *p* value (<0.001). However, no statistically significant difference was seen in mean plaque score after 6 months of SDF application with the *p* value of 0.135.

In group 2 (fluoride varnish), for both *S. mutans* and plaque score, statistically significant difference was found between the mean value of baseline *S. mutans* count and counts after 3 days (0.016), however no significant difference was seen after 3 and 6 months.

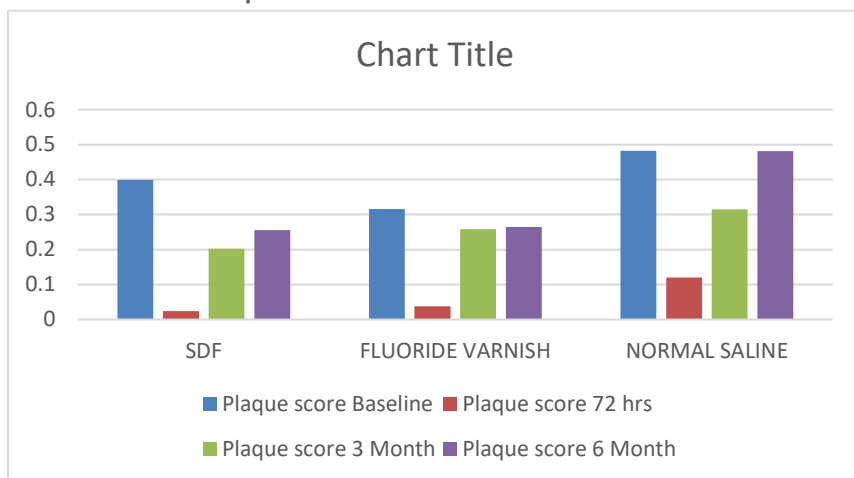
In group 3 (normal saline), statistically significant difference was found between the mean baseline *S. mutans* count and count after 3 days and a significant difference was observed between mean baseline plaque score and score after 3 days and 3 months.

Intergroup comparison done by Posthoc Turkey test, fluoride varnish has the least mean value for *S. mutans* count after 3 days, 3 and 6 months, no significant difference was seen in mean *S. mutans* count between group 1 and group 2 (*p* value 0.68), while a statistically significant difference after 3 days, 3 months and 6 months in the mean *S. mutans* count was noted between group 1 and 3 (*p* value 0.002, 0.021 and < 0.001) and group 2 and 3 (*p* value < 0.001, 0.004 and < 0.001). (Table 1 and Graph 1) For plaque score, SDF has the least mean value after 3 days, 3 and 6 months, similarly no significant difference was seen in mean plaque score between group 1 and group 2 (0.855), while a significant difference was observed in between group 1 and 3 and between group 2 and 3 after 3 days, 3 months and 6 months excluding a non-significant value after 3 months between group 2 and 3 (*p* value 0.219). (Table 1 and Graph 2).



Graph 1: Intergroup Comparison - Posthoc Test for S. Mutans

Anova Test for Plaque Score



Graph 2: Intergroup Comparison - Posthoc Test for Plaque Score

Anova Test for Plaque Score

Table 1: Intergroup comparison - Posthoc Tukey Test

Dependent Variable	(I) group	(J) group	Mean Difference (I-J)	Std. Error	P VALUE
S. mutans count Baseline	SDF	Fluoride Varnish	392.222*	91.195	<0.001
		Normal Saline	255.500*	91.195	0.019
	Fluoride Varnish	Normal Saline	-136.722	91.195	0.3
S. mutans count after 3 days	SDF	Fluoride Varnish	57.333	69.262	0.688
		Normal Saline	-255.000*	69.262	0.002
	Fluoride Varnish	Normal Saline	-312.333*	69.262	<0.001
S. mutans count 3 Month	SDF	Fluoride Varnish	15.889	27.148	0.829
		Normal Saline	-75.000*	27.148	0.021
	Fluoride Varnish	Normal Saline	-90.889*	27.148	0.004
S. mutans count 6 Month	SDF	Fluoride Varnish	5.278	29.899	0.983
		Normal Saline	-139.167*	29.899	<0.001
	Fluoride Varnish	Normal Saline	-144.444*	29.899	<0.001
Plaque score Baseline	SDF	Fluoride Varnish	0.083333	0.067269	0.436
		Normal Saline	-0.08278	0.067269	0.441
	Fluoride Varnish	Normal Saline	-1.661111*	0.067269	0.044
Plaque Score 72 Hrs	SDF	Fluoride Varnish	-0.01389	0.025994	0.855
		Normal Saline	-0.0961111*	0.025994	0.002
	Fluoride Varnish	Normal Saline	-0.0822222*	0.025994	0.007
Plaque Score 3 Month	SDF	Fluoride Varnish	-0.05611	0.033555	0.226
		Normal Saline	-1.127778*	0.033555	0.004
	Fluoride Varnish	Normal Saline	-0.05667	0.033555	0.219
Plaque Score 6 Month	SDF	Fluoride Varnish	-0.00889	0.058885	0.988
		Normal Saline	-0.2261111*	0.058885	0.001
	Fluoride Varnish	Normal Saline			

DISCUSSION

Dental caries is still one of the most common chronic diseases affecting various age groups in all the countries and on their populations with varying degree of severity. One of the most powerful weapons that have been proved to control and prevent dental caries in last 50 years is Fluoride. Loesche & Marsh mentioned that dental caries is associated with increase in the proportions of bacteria in the plaque, especially *S. mutans* and lactobacilli, which are capable of demineralizing the enamel. [11-14]

SDF is a cost-effective agent used for caries control and is available in 10%, 30%, 38% silver containing solution but 38% solution appears to be most effective. In vivo trials by Chu et al and Llodra et al, 38% SDF solution was commonly used to arrest dental caries. [9,15]

SDF solution comprises silver, fluoride and ammonia. When SDF is applied to teeth, it penetrates into the enamel and dentin, 2 to 3 times more subsurface, fluoride is deposited in the teeth than with other fluoride solutions. The teeth then act as a fluoride reservoir and may increase its long-term efficiency. SDF reduces caries in 2 ways: Primarily it acts as an antimicrobial agent, as the silver ions react with thiol groups of amino and nucleic acids, disrupting metabolic and reproductive pathways of the bacteria resulting in cell death. At the same time, SDF react with hydroxyapatite forming fluorapatite, reducing or preventing further caries. SDF will turn soft carious dentin and enamel into dark brown harder or black sclerotic surface. [7, 16]

Fluoride Varnish is effectively used in young children, patient with gag reflex and physically or mentally disabled persons. Following Fluoride Varnish an application, the tooth surface is coated with a sticky film and thereby enhancing the penetration and absorption of fluoride ions into the tooth surface. [17]

In the present study, SDF and Fluoride Varnish were applied once after restorative treatment, according to manufacturer's instruction. Chu et al found that annual application of 38% SDF was substantially more effective than 5% NaF varnish at 3 months interval in arresting and preventing dental caries in maxillary anterior teeth. [15]

In present study, in SDF group there was a significant difference observed between the mean value of baseline *S. mutans* counts and count after 3 days, 3 and 6 months as well. Likewise, Llodra et al found 38% SDF as an effective anticariogenic agent for both primary and permanent teeth while Chu et al found it effective in primary anterior teeth. A significant difference was found between the mean values of baseline plaque score and score after 3 days, 3 months; however no significant difference was seen after 6 months of SDF application. After restorative treatment and SDF application, no new carious lesion was seen after 3 and 6 months follow-up in the present study which is in accordance with an in vitro study done by Suzuki et al. [10, 15, 18]

In Fluoride Varnish group, a significant difference was seen between the mean value of baseline *S. mutans* count and count after 3 days however no significant difference was seen after 3 months and 6 months. These findings were similar to the study done by Klimek et al. Correspondingly, a significant difference was found between the mean values of baseline plaque score and score after 3 days, however no significant difference was seen after 3 months and 6 months. Loesche et al and Klimek et al found reduction in plaque score after application of FV. [13, 19]

On intergroup comparisons in the present study, no significant difference was observed in the mean *S. mutans* count between SDF and Fluoride Varnish after 3 days, 3 and 6 months. However comparing SDF and Normal saline group and Fluoride varnish and Normal saline group, significant difference was seen after 3 days, 3 months and even after 6 months. Shah S et al found significant reduction in the *S. mutans* counts in saliva by SDF as compared to Fluoride Varnish and APF gel in their study. [20]

A non-significant mean plaque values were seen after same time periods between SDF and Fluoride Varnish group. Whereas, a significant difference was observed in mean plaque value between SDF and Normal saline groups after same time intervals provided which was in agreement with the study done by Suzuki et al, where silver ions in combination with fluoride offer a possible antiplaque action of the agent. Likewise, a significant difference was found between Fluoride Varnish and Normal Saline groups after 3 days and after 6 months, but a non-significant difference was seen after 3 months which was in contrast to the study done by Klimek et al, where significant reduction in plaque score was found after application of fluoride varnish at all follow up time intervals. On the other hand, Shah S et al found decrease in Plaque score in all the subjects, but no significant difference was seen between SDF, Fluoride Varnish and APF gel group. [18-20]

CONCLUSION

In vivo application of SDF and Fluoride Varnish was efficient in reducing both *S. mutans* count and plaque score and no carious lesions were detected after follow-up time intervals as well. SDF and Fluoride Varnish can be efficaciously used as an antimicrobial, antiplaque and anticariogenic agent in young permanent teeth.

LIMITATIONS OF THE STUDY

The time period included in the study was not long enough to check the efficacy of SDF and Fluoride varnish for the reduction of caries susceptibility.

REFERENCES

- Randal Eckert, Jian He, Daniel K, Yarbrough, Fengxia QI. Targeted killing of streptococcus mutans by a pheromone –guided smart antimicrobial peptide. *Antimicrob Agents Chemother.*, 2006; 50(11):

3651-57.

- Patidar D, Sogi S, Singh V, P Shinu, Loomba A, Patidar DC. Salivary levels of *Streptococcus mutans* and *Streptococcus sanguinis* in early childhood caries: An in vivo study. *JISPPD* 2018; 36: 386-90.
- Chu CH. Treatment of Early childhood Caries: A review and case report. *Gen Den.*, 2000; 48(2): 142-48.
- Basil G. Bibby. A Test of the Effect of Fluoride-Containing Dentifrices on Dental Caries. *J Dent Res.*, 1945, 24: 297-303.
- Schmidt HFM. A new immersion agent with a particularly long-lasting intensive fluoridation effect. *Stoma.* (In German). Translate into English. A new varnish with extremely long lasting fluoridation effect. 1964; 17: 14-20.
- Azarpazhooh A, Main PA. Fluoride varnish in the prevention of dental caries in children and adolescents: a systematic review. *J Can Dent Assoc.*, 2008; 74(1): 73-79.
- Rosenblatt A, Stamford TC, Niederman R. Silver Diamine Fluoride: A Caries "Silver Fluoride Bullet". *J Dent Res.*, 2009; 88(2): 116-125.
- Yamaga R, Yokomizo I. Arrestment of caries of deciduous teeth with diamine silver fluoride. *Dental Outlook.* 1969; 33: 1007-13.
- Llodra JC, Rodriguez A, Ferrer B, Menardia V, Ramos T, Morato M. Efficacy of silver diamine fluoride for caries reduction in primary teeth and first permanent molars of schoolchildren: 36-month clinical trial. *Journal of Dental Research* 2005; 84: 721–24.
- Priya YK, Prathibha KM. Methods of collection of saliva - A Review. *International Journal of Oral Health Dentistry*; July-September 2017; 3(3): 149-153.
- Featherstone JDB. The science and practice of caries prevention. *J Am Dent Assoc.*, 2000; 131(7): 887-99.
- Ten Cate JM. Fluorides in caries prevention and control: Empiricism or science. *Caries Res.*, 2004; 38(3): 254-57.
- Loesche WJ. Role of *Streptococcus mutans* in human dental decay. *Microbiol Rev.*, 1986; 50(4): 353-80.
- Marsh PD: Microbiologic aspects of dental plaque and dental caries. *Dent Clin North Amer.*, 1999, 43: 599-614.
- Chu CH, Lo EC, Lin HC. Effectiveness of silver diamine fluoride and sodium fluoride varnish in arresting dentin caries in Chinese pre-school children. *Journal of Dental Research* 2002; 81: 767–70.
- Burgess JO, Vaghela PM. Silver Diamine Fluoride. A Successful Anticariogenic Solution with Limits. *Advances in Dental Research* 2018; 29(1) 131-34.
- Tafaraji R, Biria M, Ameri F, Torabzadeh H, Qahari P, Baghban A. Effect of two fluoride varnishes on the color stability of three resin-based restorative materials: an in vitro study: *Journal of Investigative and Clinical Dentistry* 2016; 7: 355–60.
- Suzuki T, Nishida M, Sobue S, Moriwaki Y. Effects of diamine silver fluoride on tooth enamel. *J Osaka Univ Dent Sch.*, 1974; 14: 61-72.
- Klimek J, Prinz H, Hellwig E, Ahrens G. Effect of a preventive program based on professional tooth cleaning and fluoride application on caries and gingivitis. *Community Dent Oral Epidemiol.*, 1985; 13(6): 295-98.
- Shah S, Bhasker V, Venkartarghavan K, Choudhary P, Ganesh M. Efficacy of silver diamine fluoride as an antibacterial as well as antiplaque agent compared to fluoride varnish and acidulated phosphate fluoride gel: An in vivo study. *Indian Journal of Dental Research* 2013; 24(5): 575-81.