Staining Outcomes of Silver Diamine Fluoride as an Adjunct in Caries Risk Assessment: A Case Series Evaluation

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Objective: *To investigate the staining outcomes of Silver Diamine Fluoride (SDF) after its application on healthy enamel and its implications on patient's caries risk assessment.* ***Study design:*** *A review of dental and photographic records of 61 caries-free subjects under age 3, who received SDF application as part of a prevention protocol in a private pediatric dental clinic was performed. The age, gender, caries risk assessment and resulting staining were analyzed.* ***Results:*** *Three groups of staining outcomes after SDF application were found: Group 1: no staining (32.8%). Group 2: removable staining (31.1%). Group 3: permanent staining (36.1%). The patterns of the staining were similar to initial lesion distributions in Early Childhood Caries (ECC). The deft scores for all subjects were zero before treatment and remained so at the end of this study except one subject from Group 3 who showed clinical signs of caries at follow-up visits.* ***Conclusions:*** *There are three possible staining outcomes following SDF application on clinically healthy enamel for children under age three without previous caries history; no staining, removable staining and permanent staining. The patterns of the staining were similar to initial lesion distributions in ECC. SDF application may be of use in identifying previously undetected young children with high caries risk. Further research is needed to elucidate the mechanism and cause of staining and to examine the association of the staining outcomes with the future caries incidence of the subject.*

Keywords: *SDF, Caries, Risk assessment*

INTRODUCTION

It is very frustrating for a clinician when toddlers present with caries despite their having an early first dental visit and receiving anticipatory guidance of the dental home[1]. It was suggested that the early and objective identification of children at high caries risk assists in decision-making to appropriately tailor cost-effective interventions, especially in young children[2]. The variables that influence caries risk are especially complex in this age group, which include clinical/biological, environmental and behavior/psychosocial/demographic factors[2,3,4]. In a systemic review of Caries Risk Assessment (CRA), Zero et al, concluded that the best predictor for caries in primary teeth is previous caries experience[5]. However, this predictor is of little use for young children before disease manifestation. There is a need to meet the deficiency of CRA in this regard[6].

Silver diamine fluoride (SDF) has been widely used for treatment of dentin hypersensitivity, caries arrest and prevention[7-12]. Black staining is considered a side effect of SDF application and the primary barrier to its clinical use[13,14]. Although most reports found that SDF selectively stained only carious enamel and dentin but not sound tooth tissue[9], there are a few reports of SDF staining of healthy enamel[15,16]. In a recent study on the staining potential of SDF, application of SDF on clinically sound enamel showed stains in areas of surface irregularities and proximal surfaces[17].

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In histology, silver nitrate has been used to observe mottled enamel and enamel imperfections[18]. It was suggested that silver staining may provide a tool for sensitive and convenient observation of carious lesions in both dentin and enamel[19]. It is understandable that even the earliest clinically detectable carious lesion, such as a white spot lesion, is a late manifestation of the caries disease process[20]. The early lesion is a result of long-term mineral loss under an established cariogenic biofilm. Phosphates are dissolved out of tooth structure due to the result of a skewed caries balance which favors demineralization[20]. Silver ions are commonly used in analytical chemistry to detect phosphates[21].

This study addresses the question of whether or not SDF will cause stain on clinically healthy enamel, and if these stains are associated with the caries process. SDF application is part of the prevention protocol for the first visit in the primary investigator's clinic[22]. Photographic records of infants and young children unable to sit alone in the dental chair are routinely taken at initial and follow-up visits to facilitate diagnosis and parental education. The purpose of the study was to investigate the staining outcomes of SDF application on clinically healthy enamel and its implications on caries risk assessment of the child by performing a chart review of a case series.

MATERIALS AND METHODS

A case series chart review was performed to evaluate the presence and type of enamel staining of healthy primary incisors following SDF preventive application in healthy toddlers.

The inclusion criteria of this survey were:

- Healthy patient, 36 months of age and under
- Oral examination showed no clinical signs of decalcification or visible enamel defects of erupted teeth (deft=0)
- Clinical photos of pre- and post- SDF applications clearly show anterior teeth of the child
- The participants returned for at least one 3 or 6-month recall

The protocol of the clinic for SDF application was as follows:

1. Caries risk assessment was completed according to AAPD Caries-Risk Assessment Form for 0-5 years old for dental providers[3] and charting (clinical and/or radiographic findings).
2. Routine clinical photographic examination of the dentition (Nikon D610 camera, Nikkor Micro 60 mm lens and Meike MK-14EXT Macro Ring Flash). The camera settings were as follows: shutter speed 1/200 second; aperture value f/29.
3. Parental explanation and consent were obtained for the use of SDF. Parental explanation included a short educational review of the caries process. The formation of cariogenic plaque sustained mineral loss, early white spot lesion and finally visible cavitation were all explained to the parent. Photographs depicting various degrees of staining of the tooth after SDF application were also shown to the parent.
4. The child's teeth were cleaned by a dentist with a toothbrush using a prophylaxis paste (Young Dental, Earth City, MO, USA). Tooth surfaces were dried with gauze before SDF (Advantage Arrest, Elevate Oral Care, FL, USA) application. A micro sponge-tip applicator was used for application. One applicator was found to be adequate for a full primary dentition. A gloved finger was used to smear the whitish saliva, which appears when SDF is in contact with saliva, over the entire tooth surface. This significantly reduced the amount of SDF needed. SDF was applied to all the erupted teeth. Following SDF application, fluoride varnish (3M™ ESPE™ Clinpro™ White varnish, MN, USA) was applied to all tooth surfaces.
5. Parent(s) were advised to watch for staining during the succeeding days following treatment. If staining was noted, they were requested to send photographs taken with a mobile phone via a social media application (LINE, Line Corporation) of their child's anterior teeth.
6. Clinical photographs of the dentition were taken at all subsequent recall visits.

A chart review was performed for all patients receiving SDF application as part of the above protocol in the principal investigator's private pediatric dental clinic in Taipei, Taiwan.

Statistical analysis

Collected data were entered into a computer and analyzed using SPSS 20.0 software for Windows (SPSS Inc., Chicago, USA). Chi-square test and ANOVA were performed. The level of statistical significance for all tests was set at $p=0.05$.

RESULTS

The survey identified a total of 128 subjects who had received SDF application as part of the prevention protocol between March 2018 and March 2019. A total of 61 subjects met the inclusion criteria. The age range of the subjects was from 9 to 33 months old. There were 37 males and 24 females with 21 subjects assessed to have moderate caries risk and 40 assessed to have low caries risk.

The staining outcomes after SDF application were categorized into three groups (table 1). In Group 1: No staining (32.8%), children's parents reported no staining and follow-up examination also showed no stains after the initial SDF application (Figure 1a-d). In Group 2: Removable staining (31.1%), cases showed stains, usually within the same day of the application as observed at the application visit (Figure 2b) or as reported by parent(s) (Figure 2e). The stains were removable by tooth brushing or rubber cup with a prophylaxis paste in slow speed handpiece (Figure 2c, 2f). In Group 3: Permanent staining (36.1%), cases showed stains, usually within the same visit or the same day of the application per parental report (Figure 3a-d). Some stains were not removable even with dental prophylaxis as described above. No other side effects were noted. The photos from the parents were obtained usually within a few days after the SDF application, if stains were noted. The exact date of staining occurrence was obtained from the parents.

The location and distribution patterns of stains (Figure 2b, 2e, 3b, 3d and Figure 4a-b), both removable and non-removable, were found to approximate initial lesions as described in ECC. They appeared mostly on the buccal surfaces of primary maxillary incisors or molars. The stains appeared parallel to the gingival margin, in the contact areas, or on lingual surfaces of primary maxillary incisors. Stains were initially brown in color and changed later to black. The deft scores for all subjects were zero before treatment and remained

Table 1 Three staining groups after receiving SDF application and corresponding representative case number.

Staining Group	No. of Children	%	Age [a] in Month	Gender [b] (M/F)	Caries Risk Assessment [c] (M/L)	Representative Cases
Group 1: No Staining	20	32.8	19.4 (6.9)	10 /10	5/15	Case No. 1 & 2
Group 2: Removable Staining	19	31.1	22.4 (6.3)	9 /10	7/12	Case No. 3 & 4
Group 3: Permanent Staining	22	36.1	15.7 (6.4)	17/5	9/13	Case No. 5 & 6
Total/Average	61	100	19.0 (7.0)	37/24	21/40	
Significance			P=0.007[d]	P=0.134	P=0.535	

a Standard Deviation in parentheses

b M: Male, F: Female

c M: Moderate, L: Low

d Statistically Significant

Figure 1, Group 1: No staining, no stain noted after SDF application.

Clinical photos of cases before and after SDF application. Case No. 1, a 12-month-old boy, (a) before and (b) at 6 months visit. Case No. 2, a 17-month-old girl, (c) before SDF application and (d) at the 6-month follow-up visit.

Figure 2, Group 2: Removable staining, stains noted but were removable.

Clinical photos of cases before and after SDF application. Case No. 3, a 17-month-old girl, (a) before and (b) within a few minutes after SDF application. Yellowish stains were noted on the buccal surfaces of primary maxillary incisors and canines. All stains were removable by brushing. (c) At a 4-month follow-up visit. Case No. 4, a 30-month-old boy, (d) before and (e) at the same day of SDF application. Black stains were noted on the buccal surfaces of primary maxillary incisors. The outlines of the stains were parallel to the gingival margins. The photo was taken by his mother. (f) At the 2-month follow-up visit, the stains were removable by toothbrushing, as shown in the photo.

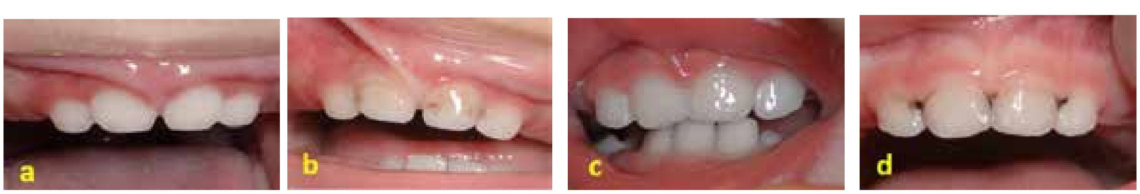

Figure 3, Group 3: Permanent staining, stains noted and some were non-removable either by toothbrushing or dental prophylaxis.

Clinical photos of cases before and after SDF application. Case No. 5, an 11-month-old girl, (a) before and (b) two weeks after SDF application. The non-removable stains were noted in the interproximal areas and parallel to the gingival margin. Case No. 6, a 12-month-old girl, (c) before and (d) 3 days after SDF application. The stained areas were at interproximal areas and parallel to the gingival margin.

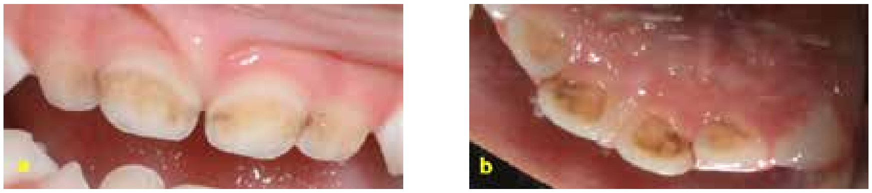

Figure 4: Clinical photos of Case No. 5 (Group 3), at 17 months of age (6 months after SDF application) showing breaks in the enamel surface. (a) Stains on the buccal surfaces of primary maxillary incisors became darker over time. (b) On the lingual surfaces of the primary maxillary central incisors, enamel lesions are detected. The enamel of all other stained surfaces remained intact.

so at the end of this study excluding one subject (Figure 4) from Group 3 who showed signs of caries in enamel at 6-month follow-up visit. In this subject, breaks in the enamel surface were able to be clearly detected due to the staining. For all other subjects in Group 3, all the stained surfaces remained intact.

A one-way analysis of variance (ANOVA) was performed to assess the differences found among the three staining groups with respect to the subjects' mean age. A statistically significant difference was noted; younger children were more likely to present with permanent staining ($p=0.007$). However, the Chi-square test showed no significant differences among the three groups by gender, deft and level of caries risk assessment at baseline.

DISCUSSION

This study's findings showed that there are three possible staining outcomes following SDF application on clinically healthy enamel for children under age three without previous caries history; no staining, removable staining and permanent staining.

The majority of the cases in this series (63.9%) did not present with any permanent SDF staining at follow-up visits. This finding is contrary to previous reports which have suggested that application[15,16] of SDF on newly erupted teeth will result in permanent staining due to the immature and porous enamel of these newly erupted teeth. The stain distributions demonstrated in these reports are similar to those of Group 3 (Figures 3, 4) of the present study. Although in our study younger children were more likely to present with permanent staining (Group 3), even children as young as age 9 months of age receiving application of SDF did not necessarily result in any permanent staining of the enamel.

The main components of SDF consist of 25% silver, 8% ammonia and 5% fluoride. Fluoride and ammonia alone or in combination do not stain tooth structure. Silver compounds, however, are known to cause staining when in contact with components present in oral environment due to three possible chemical formations[15,19,23]: 1) silver/protein reactions 2) silver phosphate layers and 3) silver sulfide precipitates.

1. Silver/protein reactions

SDF staining of carious dentin has widely been accepted to be caused by silver/protein reactions. Upon application of SDF to a decayed tooth surface, a squamous layer of silver-protein conjugates immediately begins to form. The formation of metallic silver compounds is the cause of black staining[23]. However, since enamel contains only 4% of organic materials by weight, it is unlikely to be the main cause of the staining on clinically healthy enamel.

2. Silver phosphate

Brown/black stains may appear when silver ions react with phosphates present in the plaque and tooth structure. Calcium and phosphates are dissolved out of tooth structure into the plaque during the caries processes[20,24]. Studies have shown that the major products of the reaction of SDF with tooth tissue are calcium fluoride and silver phosphate which has been confirmed by *in vitro* studies[25,26]. The possible chemical reaction mechanism of SDF with hydroxyapatite has been further proposed by Owais *et al*[26]. At acidic pH, the main silver-containing reaction product of SDF and hydroxyapatite is silver phosphate (Ag_3PO_4), which is initially yellowish in color turning black with time.

The removable brown/black stains found in group 2 indicate that the reaction was within the plaque and not in the tooth structure. The formation of silver phosphate appears to be the best possible explanation for the brown/black stain. Although saliva is super-saturated with calcium and phosphate, the observation of the staining pattern, which occurred only on plaque-retentive areas and not on all saliva-covered areas, clearly rules out the possibility that the phosphate from saliva alone may cause stain.

3. Silver sulfide

Black staining may appear due to silver sulfide formation either on the external tooth surface or deeper within the enamel[19]. Following SDF application, silver ions may be retained due to enamel defects, roughened surfaces or penetration into porous enamel. Precipitation of silver sulfide takes place when silver ions react with sulfur compounds[8,23]. Sulfur compounds from oral microbes, food particles and air are available to react with the silver ions. It is postulated that the non-removable stain in Group 3 is caused by silver ions that have penetrated the enamel due to the dynamic process of mineral dissolution and redesposition which occurs at the enamel-plaque interface after eruption[20]. It has been suggested that these subtle undetected changes within the enamel surface correspond to those observed after 1 week of exposure to a cariogenic challenge of dental plaque in a clinical controlled experiment,[24] representing active and inactive enamel lesions at the subclinical level[20]. The appearance of non-removable stain in Group 3 may thus be an early indicator that the stained areas are either actively undergoing cariogenic attacks or have done so in the past. Eventually, after sustained mineral loss, these early microscopic porosities will become clinically visible incipient lesions[20]. On the other hand, if the unfavorable environmental conditions are improved by regular plaque removal or use of topical fluorides, the active lesion may become inactive and fail to progress into a visible cavitation[24].

The distribution and location patterns of the stains are similar to the lesion patterns of ECC. The clinical timing of appearance, coloration changes, removable or non-removable of staining following SDF application on clinically healthy tooth enamel can be correlated to the three plaque related stages of the caries process as proposed by Takahashi and Nyvad[27] in their Extended Caries Ecological Hypothesis. They suggested a direct relationship between acidogenic and aciduric shifts in the composition of the dental biofilm and changes in the mineral balance of the dental hard tissues. In the dynamic stability stage, the mineral balance between biofilm and enamel is a net mineral gain (Group 1 no staining). In the acidogenic and aciduric stages, the mineral balances are tilted towards mineral loss (Group 2 and/or 3). The ability to detect the presence of large amounts of phosphate in the plaque may help indicate the stage of the plaque. The appearance of brown/black stains on tooth surfaces following SDF application may indicate the presence of large amounts of phosphate in the plaque and therefore may serve as a marker of the stage of the caries process over the tooth surface. This may imply that the caries process stages of Groups 2 and 3 can be diagnosed as being acidogenic and/or aciduric. In the concept of microbial homeostasis, for a given mature biofilm, the composition of the biofilm remains stable even after eating or brushing[28]. This may provide a possible explanation as to why stains appear over newly cleansed tooth surfaces (Figure 2, Case No. 3).

The finding of enamel caries in one subject from Group 3 at later follow-up visits suggests that the staining outcomes of SDF on clinically healthy enamel may have clinical value in supplementing the current practice of CRA. Traditional disclosing agents consist of a selective dye used to visualize and identify dental biofilm on the surfaces of teeth. Their use is limited to determining the presence of plaque. However, SDF may be used as both a disclosing agent and a diagnostic tool. In one in vitro study[29], it was shown that the depth of enamel removed during prophylaxis is approximately 0.08 µm thus allowing stain removal within the outer layers of enamel with prophylaxis. The fact that some of the stains caused by SDF application may be removed by a toothbrush, whereas others are removable only by prophylaxis may indicate that the former stains are within the plaque and the latter on the most superficial surface of the enamel. As explained above, the possibility of detecting the presence of large amounts of phosphate in the plaque may help indicate the stages of the plaque. Determination of the stage of plaque combined with the detection of micro-porosities at subclinical level may be of use in identifying high risk subjects, especially in very young children without previous history of caries[30,31]. This staining approach in principle provides a direct evaluation of the combined results of the caries complex at the enamel-plaque interface. However, beyond the esthetic concerns of the stain, unlike traditional disclosing agents, the action of SDF is not reversible and cannot be regarded as a non-invasive evaluation tool.

Early detection of high-risk individuals would be cost-effective and could greatly enhance the efficacy of prevention by intervening at the earliest stage of caries development[31,32]In addition to detection, the visualization of the stain is beneficial in at least two aspects. One is to quantify the amount of plaque and its clinical characteristics, like plaque score, for research purposes. The other is educational for parent. The presence of SDF staining may motivate a parent already in the early removal of dental plaque by showing the presence and quantity of their child's plaque. It can be a strong motivating factor to encourage parents to adopt the needed preventive measures at home which requires much compliance. Indeed, in this study parents reported that the appearance of staining motivated their brushing practices at home despite initial resistance of the child, as well as increasing the duration of cleaning especially when stains were difficult to remove by brushing.

Further research is needed to elucidate the mechanism and cause of staining and should include various characteristics such as timing, degree of color change, removable or non-removable, the staining distribution patterns and its association with the future caries incidence of the subject.

CONCLUSIONS

There are three possible staining outcomes following SDF application on clinically healthy enamel for children under age three without previous caries history: no staining, removable staining and permanent staining. SDF application may be of use in identifying previously undetected young children with high caries risk. Further research is needed to elucidate the mechanism and cause of staining and to examine the association of the staining outcomes with the future caries incidence of the subject.

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