IN VITRO STUDY

Potassium Iodide's Effect on Silver Diammine Fluoride Staining Properties as Measured Through Objective Color Analysis Using CIELAB

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Abstract: ***Purpose:*** *To determine the effect of potassium iodide (KI) on the black/gray staining caused by silver diammine fluoride (SDF) when applied to carious lesions.* ***Methods:*** *Extracted caries-free molar surfaces had caries induced to examine the use of SDF and SDF followed by KI (SDF+KI) on extracted permanent molars that had caries induced on their surfaces and were monitored for a period after application. To monitor the color changes, CIELAB color space readings–a color space defined by the International Commission on Illumination–were used. The system is composed of three values, of which the L* measures black to white across a span of zero (black) to 100 (white). Measurements were taken at eight intervals between days zero to 72.* ***Results:*** *L* values were found to be significantly different between SDF and SDF+KI groups and from baseline. On average, the SDF+KI group versus the SDF group was 9.47 units lighter.* ***Conclusion:*** *The findings indicate the application of silver diammine fluoride followed by potassium iodide can reduce the black staining SDF alone causes, potentially making it a viable esthetic option for patients with anterior tooth caries.* *(Pediatr Dent 2024;46(1):52-7) Received May 17, 2023 | Last Revision September 1, 2023 | Accepted September 1, 2023*

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Caries is the most chronic disease of childhood.[1] Treating children for any medical condition or disease, including dental caries, can often be difficult as the child is developing, and behavior may not allow for ideal treatment or favor an alternative treatment method. In these instances, methods of treatment that involve minimal time and require limited cooperation can be the best option. Silver diammine fluoride (**SDF**) has long been used in Japan to treat dental caries. The U.S. Food and Drug Administration approved SDF for the primary use of hypersensitivity in 2014 while also recognizing its off-label for caries arrest and prevention.[2] The 38 percent SDF formulations contain 44,800 ppm fluoride ions and 255,000 ppm silver ions,[3] which has a pH of approximately nine.[4] SDF has been shown to have no significant impact on plaque microbial diversity in cases with successful or unsuccessful caries arrest[5] but was shown to influence the abundance of select bacterial species.[6] It is recommended that SDF should only be applied for carious lesions that do not have pulpal exposure or approach the pulp. Studies show that re-application of SDF on a semiannual or annual basis increases the chances of caries arrest.[7,8]

A prominent side effect of SDF is black staining of the tooth structure and adjacent restorations,[9] which poses an issue for patients with its use in esthetic zones. Staining can show as early as two minutes following SDF application, with an average time of five minutes after application.[10] To help reduce the incidence of black/silver staining, the use of potassium iodide (**KI**) has been proposed. However, studies have shown conflicting evidence regarding the efficacy of KI in the reduction of black staining induced by SDF.[11] There is evidence that the combination of SDF with KI (**SDF+KI**) can enhance antimicrobial effects.[12] Knight et. al demonstrated that SDF+KI showed reduced *Streptococcus mutans* biofilm development over 14 days compared to controls.[11] Studies have been performed where carious teeth were treated with SDF+KI before receiving a restoration and yielded conflicting conclusions about KI's impact on staining.[13,14] Additional studies have examined KI's effect through application to teeth with unprepared carious lesions, which again found conflicting results regarding differences in staining.[10,15,16] SDF+KI has been shown to not have a significant inhibitory effect on bond strength between resin and dentin in an *in vitro* study.[17] A study by Lee et. al showed that SDF and SDF+KI increased surface hardness after application to demineralized bovine incisors.[18]

Teeth are unique to each person. The tooth's shape, growth, and mineralization processes are influenced by various mechanisms, such as one's genetic makeup, the presence or absence of fever, antibiotic experience, fluoride exposure, and genetic disorders such as dentinogenesis imperfecta, amelogenesis imperfecta, and dentin dysplasia.[19] In addition, teeth have various states of demineralization or caries across their surfaces. These factors all influence dental-related experiments. In the previously mentioned studies, a major source of inconsistencies came from the utilization of different teeth for each trial. In the irregularity of the teeth chosen for the studies, the data have been influenced by confounding variables. To minimize or completely resolve the issue with the natural variability of teeth, the current trial utilized a split-tooth design, which consisted of using hemisected teeth, applying treatments to each surface, and then comparing sides. Another issue with previously published studies is the variation of study duration, with some being as limited as 24 hours to seven days post-application for monitoring. The present study mitigated this limitation by monitoring for 12 weeks. By observing for a longer duration, the experiment will be able to determine if the staining changes over time.

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The purpose of this study was to provide consistent results on whether and how potassium iodide affects silver diammine fluoride staining with an improved experimental design, with the null hypothesis being that KI application would not affect SDF staining.

Methods

The study was approved by the Louisiana State University Health Sciences Center Institutional Review Board (**IRB**) for an exemption (IRB #2320) and performed by one investigator throughout. De-identified permanent teeth were used to qualify for IRB exemption, while molars specifically were chosen due to their larger surface area for evaluation and ease of sectioning. Teeth that were visibly carious and had gross carious lesions or visible demineralization were excluded from the study. Teeth were stored in 10 percent sodium thymol after collection and until sectioning. Twenty teeth were utilized during the experiment. Before sectioning, the teeth were cleaned with pumice and a prophy cup to remove any debris.[20] A three-mm by three-mm piece of tape was placed on each tooth's buccal and lingual surfaces. The teeth were painted with clear nail polish and allowed to dry for 30 minutes before the tape was removed. The Canary System® (Quantum Dental Technologies, Windsor, Ontario, Canada) was used to measure three points across the nine-mm^2 unpainted area to obtain an average baseline caries value. The average Canary value for the buccal sides was 17.8, while the lingual Canary readings averaged 16.8.

The teeth were then placed in a demineralizing solution (pH equals 4.4, 50 mM acetate, 2.2 mM KH_2PO_4, 2.2 mM $CaCl_2$) at 37 degrees Celsius for 72 hours to demineralize each tooth's exposed area. After removal from the acidic solution, a new caries value for each tooth was obtained using the Canary System®. The average Canary value for the buccal sides was 27.8, while the lingual Canary readings averaged 28.0, leading to an increase in the Canary values by an average of 10 and 11.2, respectively. Next, the teeth were sectioned from mesial to distal to yield complete buccal and lingual sides for application. The baseline CIELAB color space reading–a color space defined by the International Commission on Illumination–was recorded for each side using the Nix™ Pro Mini 2 color sensor (Hamilton, Ontario, Canada). The sensor excludes ambient light and uses a calibrated light source with industry-standard 45/zero degree measurement to determine CIELAB values to describe the color. The CIELAB reading consists of three separate units: the L* range equals zero to 100 units; the a* range equals -127 to 128 units; and the b* range equals -127 to 128 units.

Due to the use of the split-tooth design, the section of the tooth, buccal or lingual, was randomly assigned using Microsoft Excel™ (Microsoft Corp., Redmond, Wash., USA) to have 20 subjects in the control and experimental groups. Randomization was performed to control for any potential impact the buccal or lingual surfaces may have had. All sides that received only SDF were marked with a permanent marker to denote this status. The study utilized SDI's Riva Star® (SDI Limited, Bayswater, Victoria, Australia) SDF and followed the manufacturer's instructions for application. One drop of 38 percent SDF was placed on a microbrush and then scrubbed for one minute before being blotted dry with gauze.[20] The control section of each group only received an application of 38 percent SDF. After SDF application in the experimental group, two drops of KI were added with a microbrush for one minute, or until the precipitate became clear, and then were blotted dry with gauze.[20] Once all applications were complete, another CIELAB reading was taken. Between measurements, all control and experimental sections of each tooth were stored in separate, individualized, labeled containers filled with sterile water. Prior to each evaluation, each subject was removed from its container and blotted dry with gauze before the CIELAB reading was obtained. Measurements were taken at intervals of one day, three days, seven days, two weeks, four weeks, eight weeks, and 12 weeks.

Comparisons of values (L*, a*, b*) between treatment groups within each time period were made between treatments using Wilcoxon rank-sum tests. Overall changes from baseline were assessed using linear mixed models with repeated measures for tooth identification and fixed effects for time since treatment (linear in days), the treatment group, and the side of the tooth where the CIELAB reading was obtained. This is analogous to analysis of variance with random effects for each tooth. Deviance-based tests were used to sequentially determine if there was a treatment and time interaction, treatment and tooth side interaction, tooth side and time interaction, and three-way interaction. The estimated average time trends of the two treatment groups for all three values (L*, a*, b*) were plotted.

If a significant difference (overall or by trend) was found, any impact KI had on the discoloration associated with SDF application could be concluded. These values were compared independently. In addition, a repeated measure test was performed with each group's data to determine how the brightness changed over time in a longitudinal analysis.

Results

The sample size of 10 teeth per treatment group was determined based on a Wilcoxon rank-sum test and preliminary data collected on 10 teeth (five for each treatment). L* was assessed prior to treatment and after 14 days. The average increase in the change from baseline for SDF versus SDF+KI was 20.80 (P=0.063), and the estimated Kernal density was used to simulate these differences in 10 prospective subjects. Explicitly, the authors employed the density function, using R statistical software (R Foundation for Statistical Computing, Vienna, Austria), with the default Kernal choice (a Gaussian Kernal) to estimate the distribution of the change in distances. The authors then used the estimated Kernal density to randomly generate prospective values of the difference using the sample function in R. Collectively, this approach used preliminary data to perform a power analysis, in part because no preliminary data existed in the literature but also because this better mimicked experimental settings of the study. The Wilcoxon rank-sum test had a power of 99 percent across 1,000 randomly generated datasets based on this preliminary data. Since the planned analysis had two teeth sides measured across eight time points compared to the baseline, it was suspected that the repeated measures linear mixed modeling approach would have sufficient power to detect a difference in treatment effects.

Table 1 displays the unadjusted comparison of values within each period. SDF had significantly higher L* values than SDF+KI for all periods following treatment. From day one onward, this difference held for a* and b* comparisons as well. Table 2 displays the linear mixed model regression results for the change in L*, a*, and b* from baseline. Compared to SDF+KI, SDF only had a larger decrease in L* (coefficient equals -9.40; 95 percent confidence interval [**95% CI**] equals -11.75 to -7.04; P<0.001) and b* (coefficient equals -2.47; 95% CI equals -3.37 to -1.57; P<0.001) across all periods. This trend was the opposite for a*, where SDF only had a more positive increase from baseline (coefficient equals 1.43; 95% CI equals 0.58 to 2.28; P=0.002). For L*, there was a treatment-time interaction, further decreasing SDF's L* measure versus baseline compared to SDF+KI (coefficient equals -0.08; 95% CI equals -0.15 to -0.01; P=0.02). For a*, there was a reverse interaction effect, with the baseline change increasing for SDF relative to SDF+KI (coefficient equals 0.02; 95% CI equals 0.01 to 0.04; P=0.001).

Figures 1, 2, and 3 display the average estimated change from baseline for the L*, a*, and b* measures, respectively, for teeth with SDF and SDF+KI. These regression lines were averaged over other tooth characteristics, including their random effects and tooth side, to better display the comparative change over time of these measures. The raw average changes are also plotted in the dotted lines for each measure. For L* (Figure 1), the estimated SDF change from baseline relative to SDF+KI was 9.40 lower at zero days, ending at about 15.31 lower at day 72. For a* (Figure 2), the estimated SDF change from baseline was 0.72 higher at day zero and 2.73 higher at day 72 than SDF+KI. For b* (Figure 1), the estimated SDF change from baseline relative to SDF+KI was 2.47 lower at all time points.

Results for buccal versus lingual side differences from baseline were also noted (Table 2). There was a significant interaction between the side of the tooth and time, with the buccal side a* measures decreasing over time relative to baseline (coefficient equals -1.42; 95% CI equals -2.73 to -0.10; P=0.05). Buccal side measures decreased from baseline relative to the lingual side for L* (-2.47 difference; 95% CI equals -4.24 to -0.71; P=0.007) and for b* (-1.05 difference; 95% CI equals -1.95 to -0.16; P=0.022).

Discussion

The principal negative effect of utilizing SDF is the black staining of tooth structure.[15] The application of KI to the tooth structure following SDF application has been proposed to help reduce the amount of staining.[11,12] The KI is speculated to react with the superfluous silver ions to form white silver iodide.[13,21,22] Recent systemic reviews called for more evidence to confirm these assumptions.[2,23] To provide more evidence, this *in vitro* study utilized a longer period of observation (72 days) and a split-tooth design to control for the irregularity between teeth subjects. The color change in this study was assessed using the CIELAB system, which had been used in one previous study.[13] The L* value,

Table 2. MIXED MODEL REGRESSION RESULTS SHOWING THE EFFECT OF SDF* VS SDF+KI*ON EACH CIELAB**, INCLUDING INTERACTIONS WITH TIME AND SIDE OF APPLICATION

	L	a	b
SDF vs SDF+KI	-9.40 (-11.75, -7.04) P† <0.001	1.43 (0.58, 2.28) P=0.002	-2.47 (-3.37, -1.57) P<0.001
Cheek side vs tongue side	-2.47 (-4.24, -0.71) P=0.007	0.55 (-0.22, 1.32) P=0.177	-1.05 (-1.95, -0.16) P=0.022
Time since application	-0.06 (-0.11, -0.01) P=0.024	(0.00, 0.02) P=0.09	-0.01 (-0.03, 0.01) P=0.193
SDF time interaction	-0.08 (-0.15, -0.01) P=0.02	(0.01, 0.04) P=0.001	Non-significant P=0.352
SDF cheek side	Non-significant P=0.22	-1.42 (-2.73, -0.10) P=0.05	
Time cheek side interaction		Non-significant P=0.183	

* SDF=silver diammine fluoride; KI=potassium iodide.
** Forward variable selection was used for each CIELAB. The CIELAB is a color space defined by the International Commission on Illumination and consists of three separate units: L* range=0 to 100 units; a* range=-127 to 128 units; and b* range=-127 to 128 units. L* measures black to white, a* measures green to red, and b* measures blue to yellow.
† P-values from Mixed model regression results for L*, a*, and b* displaying estimated coefficients, 95% confidence intervals, which used a level of significance of P<0.05. Random effects were used for tooth IDs.

Table 1. AVERAGE CIELAB READINGS* FOR SDF** AND SDF+KI** GROUPS BEFORE APPLICATION AND ACROSS THE EXPERIMENT'S DURATION

Time (days)	L			a			b		
	SDF only†	SDF+KI	P‡	SDF only†	SDF+KI	P‡	SDF only†	SDF+KI	P‡
Before	58.78 (5.82)	61.78 (5)	0.076	3.3 (1.53)	3.08 (1.47)	0.744	20.61 (2.97)	20.43 (2.96)	0.946
0	52.79 (3.66)	56.26 (4.89)	0.007	3.38 (1.3)	2.74 (1.07)	0.127	22.72 (3.44)	21.39 (2.21)	0.18
1	40.71 (9.38)	50.61 (4.55)	<0.001	3.73 (1.77)	2.57 (1.18)	0.039	20.02 (4.6)	22.76 (3.5)	0.032
3	39.88 (10.61)	51.42 (4.58)	<0.001	3.73 (1.69)	2.82 (2.11)	0.01	18.28 (4.67)	21.38 (2.85)	0.018
7	39.19 (10.35)	51.35 (4.16)	<0.001	3.9 (1.67)	2.59 (1.07)	0.002	18.02 (3.49)	20.4 (2.22)	0.021
14	40.25 (12.08)	51.35 (5.87)	<0.001	3.57 (1.52)	2 (1.01)	0.001	18.26 (3.99)	20.8 (3.23)	0.048
28	37.16 (9.96)	50.88 (4.5)	<0.001	3.6 (1.69)	2.57 (1.01)	0.016	17.44 (3.99)	21.25 (3.11)	0.003
56	36.61 (9.5)	49.64 (4.84)	<0.001	4.05 (2.36)	2.39 (1.49)	0.009	18.41 (4.93)	21.75 (2.83)	0.005
72	36.82 (9.85)	50.14 (5.61)	<0.001	4.12 (2.25)	2.23 (1.29)	0.003	18.4 (4.86)	21.45 (2.75)	0.007

* The CIELAB is a color space defined by the International Commission on Illumination and consists of three separate units: L* range=0 to 100 units; a* range=-127 to 128 units; and b* range=-127 to 128 units; L* measures black to white, a* measures green to red, and b* measures blue to yellow.
** SDF=silver diammine fluoride; KI=potassium iodide.
† Average values (± standard deviations) at each time-point for the CIELAB components L*, a*, and b* for each treatment group. Averages are computed for the combined sample of tooth and tongue-side measurements.
‡ P-values are reported using Wilcoxon rank-sum tests.

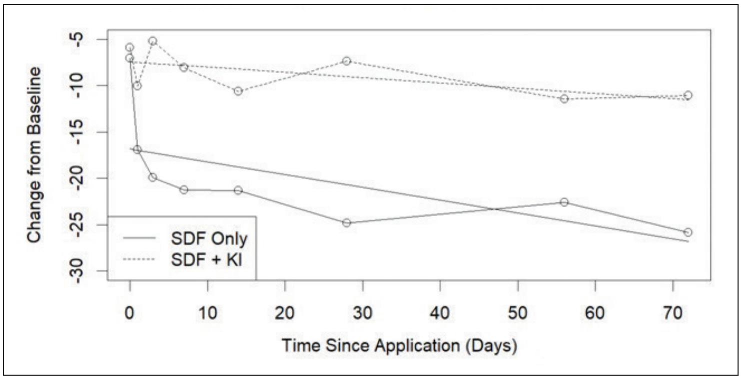


Figure 1. L* estimated change from baseline. L* is a component of the CIELAB color space, which ranges from zero (white) to 100 units (black). Average estimated L* change from pre-application by treatment group. "SDF only" refers to the treatment of the tooth surfaces (buccal or lingual) with silver diammine fluoride alone (SDF), while "SDF + KI" refers to the treatment of the tooth with SDF followed by potassium iodide (KI). The connected line segments are the average changes from baseline (i.e., the mean of all day one minus day zero measures, or the mean of all day seven minus day zero measures) for SDF and SDF+KI separately. The solid lines are the estimated linear regression lines for SDF and SDF+KI, averaged over tooth side effects, if present. These are derived from the estimated repeated measures regressions shown in Table 2. Both lines are statistically significantly diferent in terms of their intercepts and time trends.

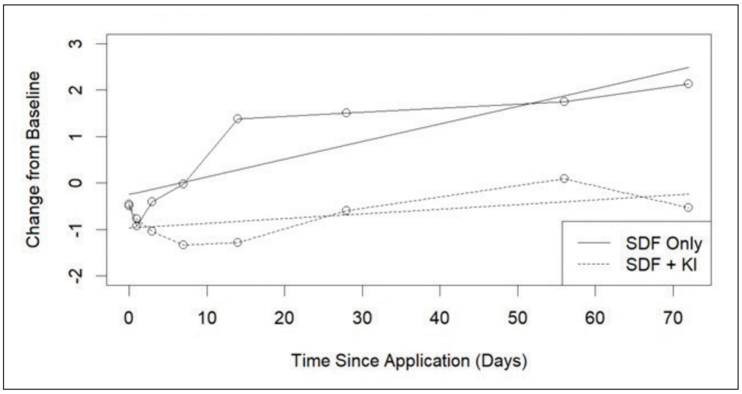
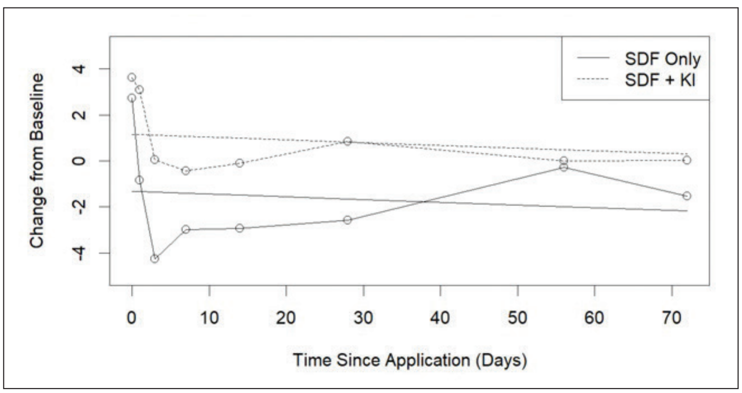


Figure 2. a* estimated change from baseline. a* is a component of the CIELAB color space, which ranges from -127 (green) to 128 (red). Average estimated a* change from pre-application by treatment group. "SDF only" refers to the treatment of the tooth surfaces (buccal or lingual) with silver diammine fluoride alone (SDF), while "SDF + KI" refers to the treatment of the tooth with SDF followed by potassium iodide (KI). The connected line segments are the average changes from baseline (i.e., the mean of all day one minus day zero measures, or the mean of all day seven minus day zero measures) for SDF and SDF+KI separately. The solid lines are the estimated linear regression lines for SDF and SDF+KI, averaged over tooth side effects, if present. These are derived from the estimated repeated measures regressions shown in Table 2. Both lines are statistically significantly different in terms of their intercepts and time trends.



which is correlated with lightness, ranges between zero (black) and 100 (white), while a* and b* values range from -127 (green) to 128 (red) and -127 (blue) to 128 (yellow), respectively.

The SDF versus SDF+KI-treated sides showed significant changes in L*, a*, and b* from their baseline values. SDF had a larger decrease in L* and b* than SDF+KI when compared to baseline, while SDF had a more positive increase in a* than SDF+KI from baseline. The average L* difference between the SDF and SDF+KI groups was -9.47, which was found to be significant. This unit difference on the L* spectrum indicates a greater tendency toward a darker shade for the SDF compared to SDF+KI. The significant difference in the three values that comprise CIELAB, particularly the L* value, between groups rejects the null hypothesis in favor of the alternative hypothesis that SDF+KI reduces staining and color change when compared to SDF application alone.

For the CIELAB value L*, there was a significant time treatment interaction, while tooth-side treatment and tooth-side time interactions were insignificant. There were no significant interactions found for b* readings with tooth side treatment, tooth side time, and time treatment interactions. The treatment time and tooth-side treatment interactions were significant for the CIELAB a* values, while the time tooth side was not significant. The significant difference noticed between the buccal and lingual sides with response to both SDF and SDF+KI may have been an anomaly as the L* and b* values were not significant for this discrepancy or possibly caused by silver's tarnishing. It is worth noting that CIELAB's three components did not have significant interaction for the tooth side time interaction, indicating that the buccal and lingual sides did not have a different change in CIELAB values over the experiment's duration, regardless of the treatment. This lack of differences could have been brought about because of them both being smooth surfaces. The lack of any significant interactions with the b* value shows that there was minimal influence by SDF or SDF+KI on the blue-yellow spectrum of the tooth's color.

This study had limitations, including the use of artificial demineralized teeth, which produce a "clean," unstained surface, unlike real caries which have rough surfaces and may influence a degree of penetration of SDF and SDF+KI. In addition, this study was performed by one individual throughout without any double-checking. Sterile water, the storage medium used, may

Figure 3. b* estimated change from baseline. b* is a component of the CIELAB color space, which ranges from -127 (blue) to 128 (yellow). Average estimated b* change from pre-application by treatment group. "SDF only" refers to the treatment of the tooth surfaces (buccal or lingual) with silver diammine fluoride alone (SDF), while "SDF + KI" refers to the treatment of the tooth with SDF followed by potassium iodide (KI). The connected line segments are the average changes from baseline (i.e., the mean of all day one minus day zero measures, or the mean of all day seven minus day zero measures) for SDF and SDF+KI separately. The solid lines are the estimated linear regression lines for SDF and SDF+KI, averaged over tooth side effects, if present. These are derived from the estimated repeated measures regressions shown in Table 2. Both lines are statistically significantly different in terms of their intercepts but not time trend.

have impacted the experimental groups as it does not simulate the oral environment because it lacks enzymes, microflora, and electrolytes, which may influence staining. The experiment did not utilize thermocycling in its methodology, which could have served to replicate the oral environment's natural fluctuation in temperature. There may be a benefit, too, from sectioning the teeth further to determine the penetration of stain into demineralized dentin. It is important to note, however, that there is doubt over the effectiveness of caries prevention in teeth treated with SDF+KI. An earlier study showed evidence that SDF alone was more effective than SDF+KI at secondary caries prevention.[15] As most of the previous studies utilized a digital spectrophotometer,[17,24] it would be beneficial to repeat the study utilizing a spectrophotometer. In addition, storing the teeth in a solution that replicates the oral environment could help better compare the two treatments because salivary enzymes could influence the results. More studies are necessary to evaluate the caries arrest potential of SDF+KI relative to SDF alone. While this study showed that SDF combined with KI can limit staining, additional studies utilizing teeth with real caries are needed to support these findings.

SDF+KI's potential may impact dentistry through greater acceptance of SDF as a minimally invasive method of caries treatment that can be used in the anterior esthetic zone and for more lesions overall. This could prove beneficial for those unable to tolerate a definitive dental procedure because it is a non-invasive treatment that could have an esthetic result. Another potential benefit from SDF's increased use is reduced financial strain on parents of low socio-economic status as well as the Medicaid system because SDF is more affordable than other treatments for carious lesions such as composite restorations and full-coverage restorations.[25]

Conclusions

Based on this study's results, the following conclusions can be made:

1. There is greater caregiver acceptance of silver diammine fluoride followed by potassium iodide because this method can reduce the degree of staining when compared to SDF alone.
2. Buccal or lingual application of SDF or SDF+KI does not affect the degree of staining of smooth surfaces; further studies about smooth surface staining in the esthetic zone are warranted to confirm these findings.

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