Caspian Journal of Dental Research
e-ISSN: 2322-2395 p-ISSN: 2251-9890
Babol University Of Medical Sciences

Comparison Of Fluoride Release And Enamel Remineralization Among Three Fluoride-Containing Materials In Primary Teeth

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Article type
Research Paper

Received: 28 Dec 2025
Revised: 20 Feb 2026
Accepted: 17 Oct 2026
Pub. online: 22 Nov 2026

ABSTRACT

Introduction: Fluoride is a key agent in preventive dentistry. This study aimed to compare fluoride release and enamel remineralization capacity among three materials, including Silver Nitrate-Fluoride (SNF) (25%), Silver Diamine Fluoride (SDF) (30%), and Sodium Fluoride (SF) (5% varnish), in primary teeth.

Materials & Methods: In this in vitro study, 105 sound primary teeth extracted for orthodontic purposes were sectioned, prepared, and mounted in acrylic resin. Samples were randomly assigned to three groups (n = 35 per group), and artificial caries were induced using a cariogenic biofilm. The groups were treated with the respective materials. Fluoride release was measured using a calibrated pH/ion meter equipped with a fluoride ion-selective electrode, and enamel remineralization was assessed using DIAGNOdent. Data were analyzed using one-way ANOVA and Tukey post hoc tests.

Results: A significant difference in fluoride release was observed among the three materials ($P < 0.001$). SNF released significantly more fluoride than both SDF and SF, and SDF released significantly more fluoride than SF ($P < 0.001$ for all pairwise comparisons). No significant differences were observed among the groups at baseline or during demineralization. However, after two weeks of remineralization, a significant difference was detected ($P = 0.006$). The remineralization percentages in the SNF and SDF groups were significantly higher than in the SF group ($P = 0.050$ for SDF vs. SF and $P < 0.001$ for SNF vs. SF); however, the difference between the two silver-containing groups was not statistically significant.

Conclusion: Silver-containing fluoride compounds (SNF and SDF) appear to be more effective options for preventing and managing caries in primary teeth, particularly in high-risk pediatric populations.

Keywords: Dental Caries; Fluorides; Silver Compounds; Tooth Remineralization

Cite this article: Shojaei N, Shahveghar N, Rikhtegaran S, Halimi Z, Sohrabi A. Comparison Of Fluoride Release And Enamel Remineralization Among Three Fluoride-Containing Materials In Primary Teeth. Caspian J Dent Res. 2026; 15: 59-67.

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Publisher: Babol University of Medical Sciences

Introduction

Dental caries is a prevalent chronic disease in children that can cause pain, infection, eating and speaking difficulties, and complications affecting permanent dentition [1, 2].

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Therefore, effective and accessible preventive strategies for early childhood caries are essential, particularly in underserved populations [2]. Fluoride is a cornerstone of caries prevention, enhancing enamel remineralization and resistance to acid dissolution through fluorapatite formation [3, 4]. Although community water fluoridation is effective, variations in fluoride concentrations and inconsistent monitoring highlight the need for additional topical fluoride applications, including toothpastes, mouth rinses, gels, and varnishes [5, 6].

Among topical fluoride formulations, varnishes have gained particular acceptance due to their ease of application, prolonged enamel contact time, and suitability for young children [6, 7]. More recently, silver-containing fluoride compounds have attracted considerable attention. Silver Diamine Fluoride (SDF), available as 30% and 38% solutions, has been extensively studied for its ability to arrest dentin caries in primary teeth and promote remineralization through fluoride release [8, 9]. Commercially available products such as Cariestop (Biodinâmica, Ibiporã, Brazil) provide 30% SDF formulations that have demonstrated effectiveness in caries management. Similarly, the sequential application of silver nitrate solution (typically 25% silver nitrate) followed by SF varnish (5% sodium fluoride) has shown efficacy in arresting simulated dentin caries in vitro [10, 11]. SF varnish (5%) remains the standard topical fluoride preparation in many clinical settings [7, 12].

Several in vitro studies have evaluated fluoride release and the caries-arresting potential of topical agents, showing that fluoride varnishes, SDF, silver nanoparticle - SF combinations, and silver nitrate followed by SF can promote remineralization or inhibit caries progression [11, 13-16]. However, differences in methodologies and outcome measures limit direct comparisons, and few studies have simultaneously compared SDF, SNF, and SF under identical experimental conditions in primary teeth for both fluoride release and enamel remineralization [8, 9, 13, 14].

Despite the growing body of evidence supporting the individual efficacy of each agent, comparative in vitro studies directly evaluating both fluoride release and enamel remineralization among SDF (30%), SNF (25%), and SF (5%) in primary enamel are limited. Specifically, the relative performance of these three materials - in terms of fluoride ion release kinetics and subsequent enamel microhardness recovery following artificial caries formation - has not been systematically compared under standardized conditions. Furthermore, most previous studies have employed variable concentrations, different caries induction protocols, and heterogeneous outcome assessments, limiting the generalizability of their findings [11, 14, 15].

Therefore, this in vitro study was designed to evaluate and compare fluoride release and enamel remineralization capacity of three commercially available formulations - SNF (Silver Nitrate-Fluoride 25%), SDF (Silver Diamine Fluoride 30%, Cariestop, Biodinâmica, Ibiporã, Brazil), and SF (Sodium Fluoride 5% varnish, Arya Dent, Tehran, Iran) - in primary teeth. By maintaining consistent experimental conditions across all groups, this study aims to provide direct comparative evidence to inform clinical decision-making regarding the optimal topical fluoride agent for caries prevention in pediatric populations.

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Materials & Methods

Study Design

This laboratory-based in vitro study compared fluoride release and changes in enamel status following treatment with three fluoride-containing materials in primary teeth. Enamel status was assessed using DIAGNOdent. The study protocol was approved by the Ethics Committee of Tabriz University of Medical Sciences (Ethical code: IR.TBZMED.REC.1401.784).

Sample Size Calculation

Sample size estimation was performed based on the study by Mighani and Shafagh [17]. Considering a mean fluoride release difference of 163 units between two fluoride varnish groups, a standard deviation of 230, a significance level of $\alpha = 0.05$, and a statistical power of 80%, the minimum required sample size was calculated as 35 specimens per group. Accordingly, a total of 105 samples were included in the study, with 35 specimens allocated to each of the three experimental groups.

Specimen Selection and Preparation

A total of 105 sound primary teeth (a mix of incisors and molars) extracted for orthodontic purposes were collected. Teeth with caries, cracks, restorations, or discoloration were excluded from the study. Immediately after extraction, specimens were stored in 0.9% normal saline solution. For disinfection, the teeth were immersed in 5% sodium hypochlorite solution for 24 hours, followed by thorough rinsing with distilled water [18].

The roots were sectioned 1 mm below the cementoenamel junction (CEJ) using a CNC cutting device (Fanavaran Nanopars, Shiraz, Iran) equipped with a double-sided diamond disk (200 µm thickness). Each crown was sectioned mesiodistally into buccal and lingual halves. The exposed enamel surfaces were sequentially polished using silicon carbide abrasive papers (800, 1000, 1200, and 2000 grit) under continuous water irrigation with manual circular motion for one minute per grit [19]. Each specimen was embedded in a self-curing acrylic resin block (15 × 15 × 10 mm), leaving the enamel surface exposed. A standardized window (2 × 2 mm) was created in the middle third of the crown, while the remaining surfaces were coated with nail varnish to restrict the area of study [20].

Experimental Groups and Artificial Caries Formation

Specimens were randomly allocated tothree experimental groups (n = 35 per group): Group 1: SF - 5% Sodium Fluoride varnish (Arya Dent, Tehran, Iran; 0.5 mL). Group 2: SDF - 30% Silver Diamine Fluoride solution (Biodinâmica Química e Farmacêutica Ltda., Ibiporã, Paraná, Brazil; 5 mL bottle). Group 3: SNF - 25% Silver Nitrate solution mixed with Fluoride solution.

Artificial carious lesions were induced on enamel surfaces using a cariogenic biofilm model, as described previously [11, 16]. Briefly, specimens were inoculated with a cariogenic bacterial biofilm under appropriate culture conditions to produce enamel demineralization. Baseline enamel status was assessed using DIAGNOdent (KaVo Dental GmbH, Biberach,

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Germany) before demineralization. The depth and severity of demineralization were standardized by maintaining consistent biofilm exposure times and culture conditions across all specimens [11].

Fluoride Release Measurement

Fluoride ion release from each fluoride solution was measured using a calibrated pH/ion meter (Metrohm AG, Herisau, Switzerland) equipped with a fluoride ion-selective electrode [21]. Calibration was performed with standard fluoride solutions of known concentrations. During measurements, solutions were gently agitated to ensure uniform ion distribution. Fluoride concentration was recorded in parts per million (ppm). The electrode was rinsed with distilled water and dried between measurements to prevent cross-contamination [21].

Remineralization Protocol

Following demineralization, a remineralization protocol was applied over five consecutive days [13]. Specimens in each group were immersed daily in their respective fluoride solutions for 15 minutes, followed by immersion in artificial saliva for one hour. Later the same day, specimens were immersed for 15 minutes in the corresponding toothpaste slurry. For the remainder of the day, samples were stored in artificial saliva. This protocol was repeated for five consecutive days. The control condition consisted of specimens stored only in artificial saliva after demineralization, without any fluoride treatment.

Follow-up Measurements

All specimens were evaluated over a 14-day period. DIAGNOdent readings were recorded at three time points: baseline (intact enamel), after demineralization, and following remineralization [22]. These measurements were recorded for subsequent statistical analysis.

Statistical Analysis

Statistical analyses were performed using SPSS software (version 20.0; IBM Corp., Armonk, NY, USA). Descriptive statistics were calculated, and data were presented as mean ± standard deviation. The normality of the data distribution was assessed using the Shapiro-Wilktest. One-way analysis of variance (ANOVA) was applied to compare fluoride release, DIAGNOdent readings, and remineralization percentages among the three experimental groups. When significant differences were identified, post hoc pairwise comparisons were conducted using the Tukey test. A value of $P \leq 0.05$ was considered significant.

Results

Fluoride Release

The mean fluoride release values (µS/cm) were 4.48 ± 0.16 for SNF (Silver Nitrate-Fluoride 25%), 4.32 ± 0.14 for SDF (Silver Diamine Fluoride 30%), and 4.01 ± 0.10 for SF (Sodium Fluoride 5% varnish). One-way analysis of variance demonstrated a statistically significant difference among the three materials ($p < 0.001$). Pairwise comparisons revealed that SNF released significantly more fluoride than both SDF (mean difference = 0.15 µS/cm, 95% CI: 0.07 to 0.23, $p < 0.001$) and SF (mean difference = 0.46 µS/cm, 95% CI: 0.38 to

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0.54, p < 0.001). SDF also showed significantly higher fluoride release compared to SF (mean difference = 0.31 µS/cm, 95% CI: 0.23 to 0.38, p < 0.001).

Enamel Progression (DIAGNOdent)

Table 1 presents the DIAGNOdent readings at baseline, after demineralization, and after remineralization. At baseline and after demineralization, no significant differences were observed among the three groups. However, after two weeks of remineralization, significant differences were observed among the three materials for DIAGNOdent readings (p = 0.006).

Table 1. Comparison of DIAGNOdent readings (AU) during enamel demineralization and remineralization across three groups.

Materials	Baseline Enamel Mean ± SD (AU)	Demineralized Enamel Mean ± SD (AU)	Remineralized Enamel Mean ± SD (AU)
SNF (25%)	3.89 ± 1.25	20.66 ± 2.44	7.86 ± 1.68
SDF (30%)	3.43 ± 1.04	20.97 ± 3.09	8.49 ± 3.01
SF (5% varnish)	3.97 ± 1.48	19.77 ± 2.58	9.66 ± 2.11

Remineralization Percentage

According to the data shown in Table 2 a statistically significant difference was observed among the three materials (p < 0.001). Pairwise comparisons demonstrated that SNF and SDF showed comparable remineralization (mean difference = 3.19%). Both SNF (mean difference = 10.95%, 95% CI: 3.30 to 18.61, p < 0.001) and SDF (mean difference = 7.76%, 95% CI: 0.11 to 15.42, p = 0.050) showed significantly higher remineralization compared to SF.

Table 2. Comparison of remineralization percentage (%)* among the three materials.

Materials	N	Mean ± SD (%)	P-value**
SNF (25%)	35	61.26 ± 9.98^{A}	
SDF (30%)	35	58.07 ± 16.65^{A}	< 0.001
SF (5% varnish)	35	50.31 ± 12.94^{B}	

-Different capital letters indicate significant differences between remineralization percentages for each material.
*Remineralization percentage calculated from DIAGNOdent readings: [(Demineralized - Remineralized) / (Demineralized - Baseline)] × 100
**One-way ANOV

Discussion

In this in vitro study, SNF showed the highest fluoride release and remineralization percentage, followed by SDF and SF. Although remineralization did not differ significantly between SNF and SDF, the statistical comparisons with SF require verification before

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definitive conclusions can be drawn. These findings suggest that fluoride release and remineralization may vary depending on the formulation used.

SNF exhibited significantly higher fluoride release than SDF and SF, potentially due to differences in chemical composition and formulation, while SF varnish is designed for prolonged surface contact rather than rapid ion release [3, 21]. Previous in vitro studies reported substantial fluoride release from SDF, with findings varying across fluoride varnishes and experimental conditions, including fluoride concentration, sampling intervals, and testing media [21, 23, 24].

The higher fluoride release from silver-containing agents may reflect their chemical properties, including interactions with enamel that facilitate gradual fluoride release and reservoir formation [25, 26]. In contrast, SF may provide less sustained fluoride availability over time [25, 26]. Remineralization followed a similar pattern, with SNF and SDF outperforming SF, consistent with fluoride's role in promoting fluorapatite formation and increasing enamel resistance to acid dissolution [3, 27].

Our findings align with previous in vitro studies showing that SDF reduces dentine lesion depth compared with SF and that silver nitrate combined with SF inhibits demineralization and promotes remineralization [11, 14]. Although Yoshikawa et al. reported strong remineralization with SF combined with CPP-ACP, their study did not evaluate silver-containing agents, limiting direct comparisons [13]. The comparable remineralization observed with SNF and SDF suggests that both formulations may effectively support mineral recovery [11, 28].

The superior remineralization observed with silver-containing agents may be attributed to fluoride-mediated mineral deposition and fluorapatite formation, while silver ions' antimicrobial effects were not directly assessed in this study [3, 16, 27, 29]. The observed association between higher fluoride release and greater remineralization, with SNF showing the highest values, supports the importance of fluoride availability and release kinetics in enamel repair [3, 25, 27, 30].

However, the comparable remineralization observed between SNF and SDF, despite numerical differences in fluoride release that were statistically significant, suggests that factors beyond fluoride ion concentration may influence remineralization outcomes. These may include the formulation of the material, its interaction with the enamel surface, and the presence of other ions such as silver [25]. The similar performance of both silver-containing agents indicates that the presence of silver, in combination with fluoride, may provide additional benefits for remineralization beyond fluoride alone [11, 28].

Limitations of the Study

Several limitations should be considered, including the in vitro design and use of artificial saliva, which cannot fully replicate the oral environment and microbiome [8]. The 14-day observation period also limits conclusions regarding long-term effectiveness, while differences in material concentrations and application protocols may affect comparisons with previous studies. Furthermore, although DIAGNOdent is a validated, non-invasive method for monitoring mineral changes, it provides an indirect assessment of remineralization compared with direct methods such as microhardness testing.

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Conclusion

Silver-containing fluoride formulations warrant further investigation as potential approaches to managing early enamel lesions in primary teeth. Before clinical recommendations can be made, well-designed clinical studies should establish their effectiveness, safety, acceptability, and long-term outcomes.

Funding

This study was derived from the doctoral thesis of Nastaran Shojaei and was financially supported by Tabriz University of Medical Sciences, Tabriz, Iran.

Acknowledgments

The authors would like to thank the Dental Research Center and the Faculty of Dentistry at Tabriz University of Medical Sciences for their support and for providing laboratory facilities during the experimental phase of this study.

Conflict of interest

The authors declare that they have no competing interests.

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