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Plaque and Salivary Fluoride Levels in Preschoolers Following Applications of Silver Diamine Fluoride, Sodium Fluoride Varnish, and Their Combination: A Randomized Clinical Trial

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ABSTRACT

Background: The amount of fluoride maintained in the oral cavity aids in the remineralization process.

Hypothesis/Aim: To evaluate and compare plaque and salivary fluoride levels following applications of silver diamine fluoride (SDF), sodium fluoride varnish (NaFV), or both.

Design: Sixty preschoolers randomly received 38% SDF, 5% NaFV, or both (SDF + NaFV). Plaque and saliva were collected at baseline; 5, 30, and 60 min; and 24 and 48 h post-application. Fluoride levels in plaque and saliva were evaluated and statistically compared ($p < 0.05$).

Results: Salivary fluoride levels peaked 5 min post-application in all groups and recovered to baseline within 1 to 24 h. Plaque fluoride levels peaked between 5 and 60 min, then returned to baseline within 1 to 24 h. The SDF group had significantly lower plaque and salivary fluoride levels than the other groups. There were no differences in plaque or salivary fluoride levels between NaFV and SDF + NaFV groups; however, the SDF + NaFV group had the longest salivary fluoride retention.

Conclusions: The application of SDF in combination with NaFV (highest fluoride exposure) resulted in higher fluoride levels in plaque and saliva of preschoolers. Since these levels returned to baseline in less than 24 h, further studies are required to establish the implications for caries arrest and prevention.

1 | Introduction

Early childhood caries (ECC) is defined as the presence of one or more decaying, missing, or filled tooth surfaces in any primary tooth in a child under 6 years of age [1]. ECC affects around 48% of preschool children worldwide, making it a major public health concern [2]. The use of professional topical fluoride treatments, primarily fluoride varnish and silver diamine fluoride (SDF), is one of the simplest and most cost-effective approaches for treating ECC. Varnish containing 5% sodium fluoride (NaF) is widely used and has been found to be effective in preventing and arresting enamel caries in primary teeth [3, 4]. SDF at 38% has demonstrated well-established effectiveness in arresting dentine caries; however, its efficacy in managing lesions in enamel requires further investigation [5]. The major drawback of SDF is black staining of the teeth, which causes esthetic concerns for parents and children [6]. According to the American Academy of Pediatric Dentistry guidelines for the professional use of topical fluoride, sodium fluoride varnish (NaFV) is indicated for preventing dental caries and halting enamel caries progression

Summary

- Why this paper is important to pediatric dentists
  - Both NaFV and SDF + NaFV applications resulted in high fluoride levels in plaque and saliva, with the SDF + NaFV group (higher fluoride exposure) retaining fluoride in saliva for a longer period (returned to baseline within 24 h).
  - Future clinical studies should investigate whether the higher fluoride exposure of the combined therapy of SDF + NaFV affects caries arrest, caries prevention, and if it offers an advantage over spreading the applications across separate visits.

in patients at high risk for caries, while SDF is most frequently indicated for arresting dentinal caries [7, 8]. The combination of both agents may increase the arrest of caries by addressing lesions at various developmental stages. Fluoride acts by enhancing remineralization while inhibiting demineralization. High fluoride concentrations also have antibacterial properties [9]. The longer fluoride lingers in the oral cavity, the more effective it is in preventing and arresting caries [10]. Knowing the kinetics and bioavailability of fluoride in the oral cavity following professional fluoride treatments aids in the understanding of fluoride clearance, as well as how long different types of fluoride agents remain in the oral cavity or are altered over time [11–14].

Dental plaque, oral soft tissues, and tooth surfaces act as fluoride reservoirs in the oral cavity. Fluoride can adhere to these surfaces and gradually release into the saliva. This slow-release mechanism is essential for maintaining adequate fluoride levels in the oral cavity, which can aid in caries prevention and promote enamel remineralization [15]. While fluoride levels in plaque and saliva have been studied in older populations [12, 13, 16, 17], data in preschool children, the age group affected by ECC, are limited. Moreover, the combined application of SDF and NaFV has not been previously investigated in any age group. Measuring and comparing fluoride levels after various professional fluoride applications would enhance our understanding of their bioavailability and could justify the combined use of SDF and NaFV as a viable therapeutic strategy for caries prevention in preschoolers. Therefore, the aims of this study were to evaluate and compare plaque and salivary fluoride levels at different time intervals following professional applications of 38% SDF, 5% NaFV, and a combination of both in preschool children.

2 | Materials and Methods

This three-arm parallel randomized clinical trial was approved by the Institutional Review Board, Faculty of Dentistry and the Faculty of Pharmacy, Mahidol University (COE.No.MU-DT/PY-IRB2023/054.0609) and registered in the Thai Clinical Trials Registry (TCTR20240705002). The study was conducted at child development centers in Nakhon Pathom Province, Thailand, from July 2023 to March 2024. Parents and caregivers were fully informed about the study procedures, including potential tooth staining from the SDF treatment, and they provided written informed consent for eligible children.

2.1 | Sample Size Estimation

The sample size was calculated using a one-way ANOVA to compare means among the three groups. Based on a previous study by Jabin et al. (2022), which reported salivary fluoride increases of 3.5 ppm for NaFV and 4.5 ppm for SDF at 5 min post-application [17], we estimated a change of 5 ppm for the SDF + NaFV group. Using a standard deviation of 1, a two-sided significance level of 0.01, and 90% power, 17 children per group were required. To account for a 10% dropout rate, the final sample size was increased to 20 children per group, totaling 60 children.

2.2 | Inclusion and Exclusion Criteria

Healthy and cooperative children aged 3 to 6 years with a full primary dentition of 20 teeth and at least one tooth with dentinal caries were included in this study. The exclusion criteria were children with a known allergy to fluoride, silver, or colophony agents and children who had received professional fluoride treatment in the past 3 months.

2.3 | Clinical Procedures

Before the beginning of the intervention, each child was given a new toothbrush and fluoride-free toothpaste (Pureen kids strawberry flavor, Summit Corporation Overseas Ltd., Bangkok). Parents were instructed to maintain regular brushing routines using these supplies starting 2 days before the intervention and continuing through the 3-day study period. At the intervention and follow-up visits, participants were instructed to eat breakfast early so that a 1-h interval could elapse before plaque and salivary samples were collected.

At baseline, demographic data (sex and age) and oral health status were noted. One calibrated pediatric dentist performed oral examinations using the decayed, missing, filled teeth (dmft) index according to the WHO criteria [18] and plaque index (PI) following the Greene and Vermillion criteria [19]. The baseline plaque and salivary samples were collected from all participants.

Participants were randomly allocated using block randomization (block size of six) into three groups:

1. Group I (SDF): One drop (25–50 μL) of 38% SDF (Topamine, PharmaDesign Co. Ltd., Thailand) applied to all dentinal caries for 1 min
2. Group II (NaFV): 0.25 mL of 5% NaF varnish (Duraphat, Colgate-Palmolive, USA) applied to all tooth surfaces for 1 min
3. Group III (SDF + NaFV): Sequential application of SDF and NaFV using the same volumes and methods as above

For all groups, teeth were isolated and cleaned with gauze before fluoride application. In the SDF group, SDF was applied solely to teeth with dentinal caries lesions, leaving the remaining teeth in the oral cavity untreated. In the NaFV group, NaFV was applied to all teeth in the oral cavity. Additionally, for the SDF + NaFV group, SDF was applied to teeth with dentinal caries, while NaFV was applied to the rest of the teeth in the mouth. Topical fluoride was not applied to teeth with suspected or evident pulpal exposure. Post-treatment instructions included avoiding hard foods for 2 h and maintaining a 1-h fasting period before subsequent sample collections.

2.4 | Saliva Collection

Each subject was instructed to provide 3 mL of unstimulated whole saliva using the drooling technique for 5 min, collecting it in a re-sealable plastic bottle. Subjects were instructed to let saliva collect at the base of their mouths without sucking or stimulating flow.

2.5 | Plaque Collection

To minimize salivary contamination, the subjects were instructed to swallow any remaining saliva before collecting the plaque. A spoon excavator was used with a gentle scraping motion to ensure that there was no direct contact with the enamel surface and to avoid contamination with food debris and calculus. Pooled plaque samples weighing at least 2 mg were obtained from the labial, buccal, palatal, and lingual surfaces of teeth in each quadrant. Furthermore, after the application of professional fluoride, pooled plaque samples were collected from tooth surfaces that were covered with visibly and clearly identified dental plaque in order to prevent the collection of fluoride from plaque-free tooth surface areas that had been coated with SDF or NaFV. The collected plaque samples were then carefully placed on plastic strips and stored in re-sealable plastic tubes that were pre-weighed before the plaque collection process. At each time point of collecting saliva and plaque samples, this method was used.

Saliva and plaque samples were collected at baseline; at 5, 30, and 60 min; and at 24 and 48 h after professional fluoride application. Within an hour of collection, the samples were placed on ice and stored at −20°C. At the end of the day, all samples were transferred and stored at −80°C for future fluoride analysis.

2.6 | Estimation of Fluoride in Plaque and Saliva

Prior to fluoride measurement, plaque and saliva samples were removed from the freezer and left to thaw at room temperature for 1 h. The collected saliva samples were transferred to a new tube and centrifuged at 10,000 g for 10 min to remove cellular debris, bacteria, and the majority of proteins from the saliva [14, 20–22]. Then the supernatant was collected, and the free salivary fluoride concentrations in the supernatant were measured directly using a fluoride-ion selective electrode (F-ISE) (model 96-09BN, Orion, Cambridge, MA, USA), which was connected to an ion selective meter (model EA 940, Orion). The fluoride standard solutions (Orion) at a concentration of 0.1, 1, 10, and 100 ppm were freshly prepared and used for obtaining the calibration plots of the F-ISE.

A total ionic strength adjustment buffer (TISAB III; Orion), 200 μL, was mixed with 2 mL of the supernatant using a magnetic stirrer. Then the F-ISE was inserted into the solution, and the fluoride concentration was determined.

For dental plaque, the fluoride concentration was evaluated using a modified microdiffusion technique [23]. In short, approximately 2 mg of plaque samples were dissolved in 1 mL of deionized water and placed in a 10-cm plastic dish. Then 1 mL of 5 M perchloric acid ($HClO_4$; Loba Chemie Pvt. Ltd., Mumbai, India) saturated with hexamethyldisiloxane (HMDS; Sigma-Aldrich, St. Louis, MO, USA) was added to another side of the dish. In a separate 3-cm plastic dish, a trapping solution consisting of 2 mL of 0.1 M sodium hydroxide (NaOH; Ajax Finechem, New South Wales, Australia) was placed inside the 10-cm dish, which was immediately sealed with petroleum jelly and paraffin paper. Next, all the samples were incubated at 45°C with continuous rotary motion shaking at 100 rpm for 24 h. After that, each trapping solution was mixed with TISAB III in a ratio of 10:1 and the concentration of fluoride was measured using the F-ISE. The fluoride concentration in plaque was reported in ppm.

The total amount of fluoride in plaque and saliva over the whole measured time period (area under the curve; AUC) was calculated using the trapezoidal method.

2.7 | Statistical Analysis

All data were analyzed using IBM SPSS version 28.0 (SPSS, Chicago, IL, USA) with a significance level of 0.05 for all tests. Demographic data were presented using descriptive statistics. The differences among the three groups were tested using the Pearson chi-square test for qualitative variables and the one-way ANOVA for quantitative variables (i.e., age, weight, dmft scores, and PI). Due to the skewness of the plaque and salivary fluoride levels, the Kruskal-Wallis test was applied to test the differences among the three groups at each time point, whereas Friedman's test was used to compare the six different time points in each group. The Bonferroni correction was applied for multiple comparisons. The Kruskal-Wallis test was used to compare the AUC of plaque and salivary fluoride among the three groups, followed by Mann–Whitney U tests.

Intra-observer reliability for the PI was analyzed using the weighted kappa (Kw), and a Kw of at least 0.8 was obtained at the beginning of the study. Regarding the intra-observer reliability of the fluoride measurements, 10% of the plaque and saliva samples was used to determine intra-class correlation coefficients (ICCs).

3 | Results

Of 79 children, 60 who met the inclusion criteria were randomly placed into three groups, each receiving a different type of professional fluoride application. The participants were 29 males (48.3%) and 31 females (51.7%), with a mean age of 44.6 ± 6.2 months and mean weight of 16.5 ± 3.4 kg. The overall oral health status of the participants indicated

TABLE 1 | Demographic characteristics of the study participants.

Variables	Group			p
	SDF $n = 20$	**NaFV $n = 20$**	**SDF + NaFV $n = 20$**	
Sex n (%)				0.627
Male	11 (55)	8 (40)	10 (50)	
Female	9 (45)	12 (60)	10 (50)	
		Mean (SD)		
Age (months)	45.2 (5.5)	42.9 (6.2)	45.9 (6.7)	0.277
Oral health status				
PI	2.1 (0.5)	1.8 (0.5)	2.1 (0.5)	0.149
dmft	7.1 (6.2)	5.8 (6.3)	10.0 (6.0)	0.095
Number of fluoride treated teeth/Total[a]				
SDF	5/20	—	5/20	N/A
NaFV	—	20/20	15/20	N/A

Abbreviations: dmft, decayed, missing, and filled teeth; N/A, not applicable; NaFV, sodium fluoride vanish; PI, Plaque Index; SDF, silver diamine fluoride.
[a]Total number of teeth used for plaque collection.

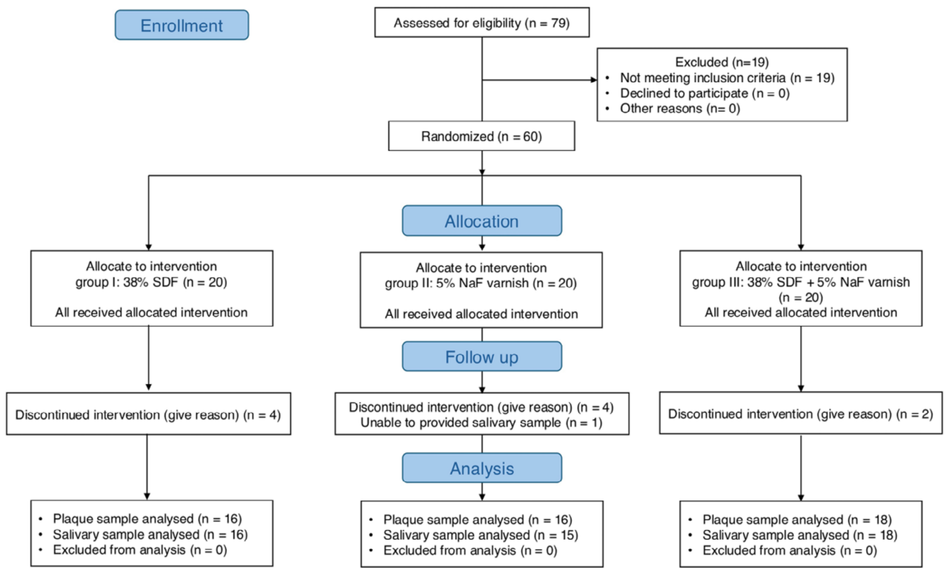

FIGURE 1 | CONSORT flow diagram of the randomized controlled trial.

poor oral hygiene, with a mean PI of 2.0 ± 0.5, and a very high prevalence of caries was indicated by a mean dmft score of 7.6 ± 6.3. The baseline demographic characteristics of each group are presented in Table 1. No statistically significant differences were observed between sex, age, PI, and dmft among the groups ($p > 0.05$). The intra-examiner reliability was acceptable, with a Kw value of 0.87 for the baseline PI assessment. Within 3 days of the sample collection, 10 children had not fully participated and one child was unable to provide a saliva sample, so the resulting total was 50 participants. A CONSORT flow diagram is shown in Figure 1.

The repeatability of the plaque and salivary fluoride analyses was acceptable for intra-examiner fluoride measurements (ICC: 0.7 and 1, respectively). The fluoride levels in saliva in groups at different time points are shown in Figure S1; Table S1. Salivary fluoride levels peaked at 5 min post-application in all groups, and these levels returned to baseline within 60 min in the SDF and NaFV groups, while in the SDF + NaFV group, they returned to baseline after 24 h. In contrast, the plaque fluoride levels peaked at 30 min in the SDF and SDF + NaFV groups, while two peaks were observed at 5 and 60 min in the NaFV group. Additionally, plaque fluoride levels returned to baseline at 60 min for the SDF group and at 24 h for both the NaFV and SDF + NaFV groups (Figure S2; Table S2).

A comparison of the salivary fluoride levels among groups found that at 5, 30, and 60 min post-application, the fluoride levels in the NaFV and SDF + NaFV groups were significantly higher than in the SDF group. No significant difference was observed between the NaFV and SDF + NaFV groups at any time points (Figure 2). The dental plaque fluoride levels in the NaFV group were significantly higher than in the SDF group at 5 and 60 min post-application. At 30 and 60 min post-application, the fluoride levels in the SDF + NaFV group were significantly higher than in the SDF group. In contrast, no significant difference was observed between the NaFV and SDF + NaFV groups at any time point (Figure 3).

The AUC, which represents the total amount of fluoride in saliva and plaque over the entire measurement period (Figure 4), showed no significant difference in salivary fluoride AUC

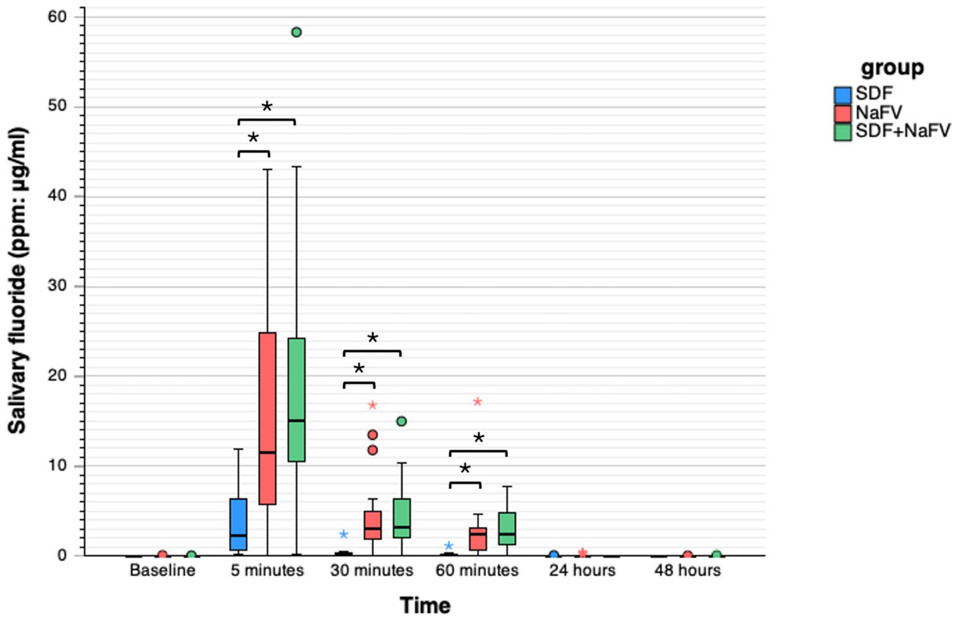

FIGURE 2 | Salivary fluoride levels at different time points in groups. *Statistical significance $p < 0.01$.

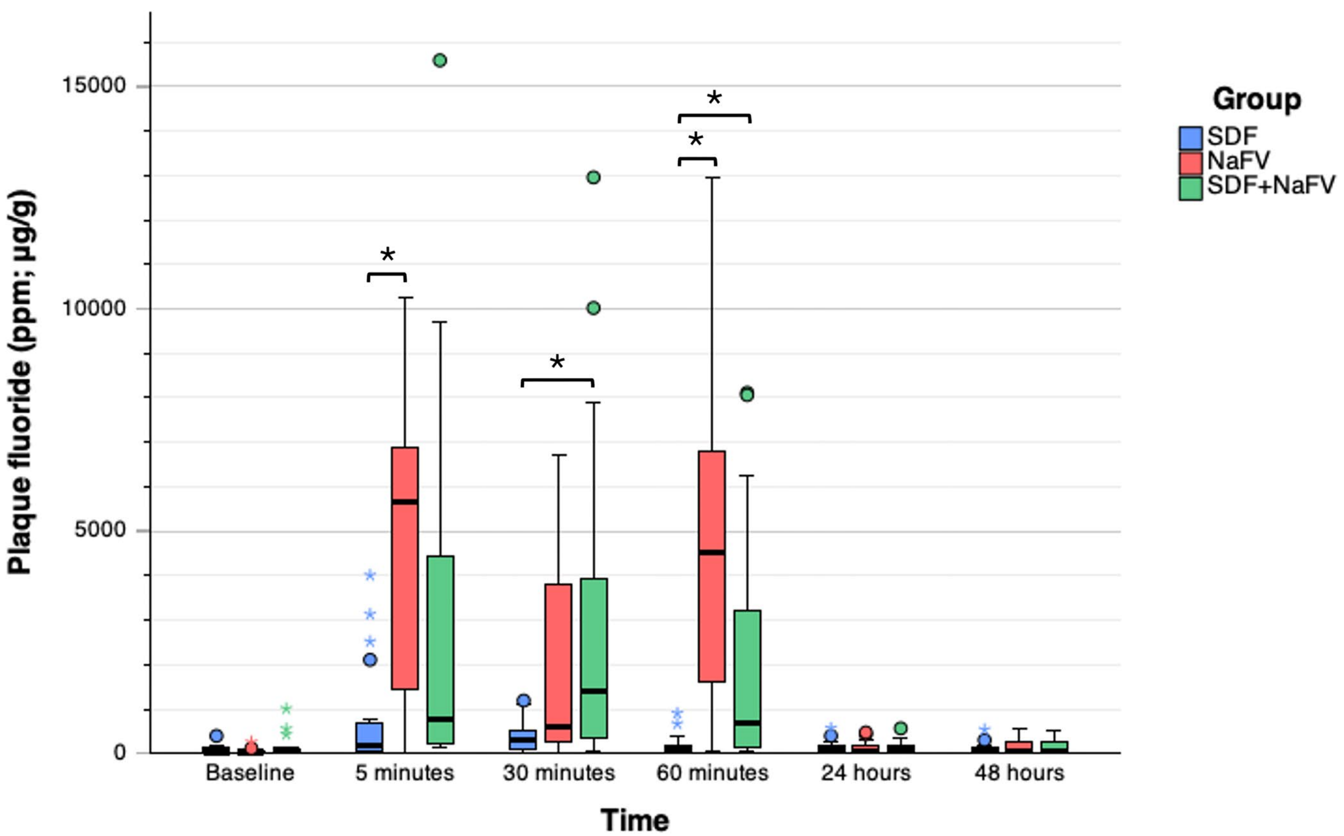

FIGURE 3 | Plaque fluoride levels at different time points in groups. *Statistical significance $p < 0.05$.

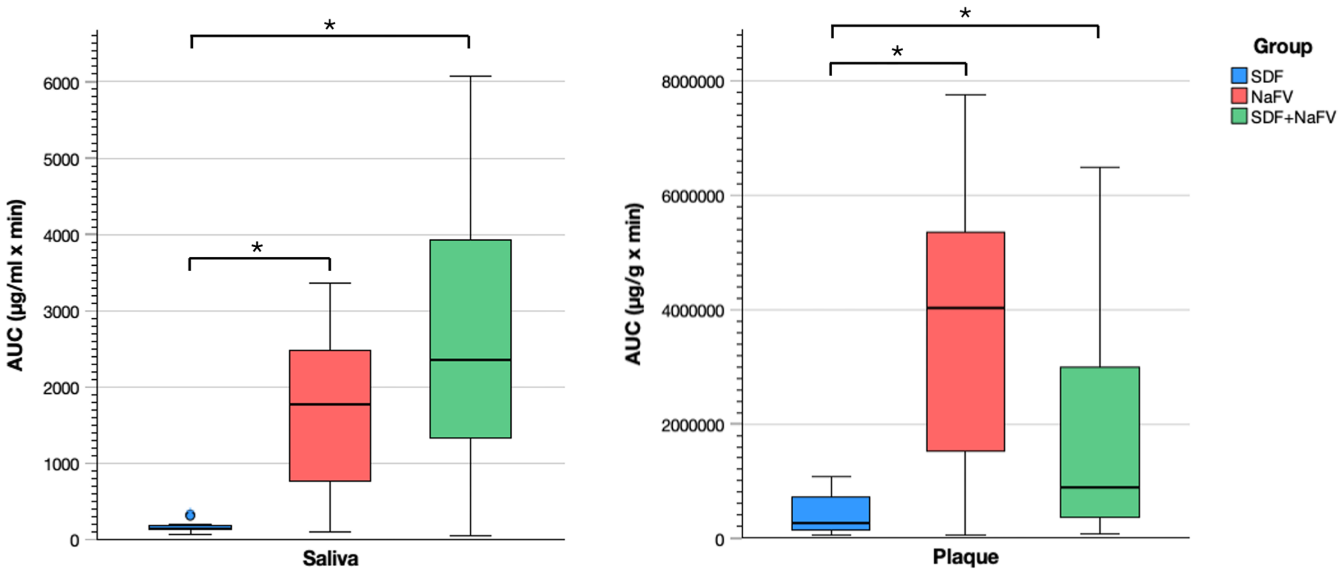

FIGURE 4 | Area under curve (AUC) of fluoride in saliva and plaque following SDF, NaFV, and SDF + NaFV application. *Statistical significance $p < 0.01$.

between the NaFV and SDF + NaFV groups, whereas the SDF group had a significantly lower salivary fluoride AUC than the NaFV and SDF + NaFV groups. For dental plaque, there was no significant difference in plaque fluoride AUC between the NaFV and SDF + NaFV groups; however, the SDF group had a significantly lower plaque fluoride AUC than the NaFV and SDF + NaFV groups.

4 | Discussion

Professional topical fluoride is the effective treatment for preventing and arresting dental caries [24]. The present study evaluated and compared the in vivo retention and clearance of plaque and salivary fluoride after different topical fluoride treatment techniques had been used. The demographic data, as well as the baseline plaque and salivary fluoride concentrations, revealed no significant differences across groups, indicating that the participants in each group shared common characteristics.

Topical fluoride significantly elevates and maintains fluoride levels in both saliva and dental biofilm [25]. The total amount of fluoride ion delivered by topical fluoride application influences how much fluoride is elevated and retained in the oral cavity. In this study, approximately 1.64 to 1.76 mg of fluoride ion was applied in the SDF group [26], while the NaFV group received about 5.65 mg [27]. Consequently, the highest amount of fluoride ion was delivered in the SDF + NaFV group, which totaled about 7.25 mg.

Salivary fluoride serves as a reservoir, delivering fluoride to dental plaque through diffusion, with higher concentrations potentially providing a greater reservoir capacity [14]. In our study, salivary fluoride levels peaked at 5 min after application in all groups that received different types of professional fluoride. This rapid increase indicated fluoride ion dissolution, elevating salivary fluoride concentrations shortly after administration, and this corresponded with many previous studies in which fluoride levels peaked within 1 to 15 min [11, 12, 14, 17, 28, 29]. However, Dehailan et al. reported a peak of salivary fluoride at 30 min after NaFV application because the first evaluation was set at that time point [13]. The results showed that, for the NaFV group, salivary fluoride levels returned to normal after 60 min had elapsed from application, whereas a study of school-aged children reported a 24-h return to baseline [13]. This could be attributed to the fact that the salivary flow, fluoride clearance rate, and saliva composition varied among age groups [30]. For the SDF group, the results showed that the salivary fluoride returned to baseline within 60 min post-application. In contrast, Jabin et al. observed a slightly elevated salivary fluoride concentration compared with the baseline level at 2 h after SDF application [16]. This may have been because, in their research, SDF was applied to all the teeth of school-aged children, resulting in a higher amount of SDF being used than in this study. Consequently, the fluoride was retained in the oral cavity for a longer period.

Plaque fluoride levels are influenced by fluoride availability in the oral cavity, including salivary fluoride, and the time required for fluoride diffusion into the plaque biofilm, which is at least 5 to 30 min [11, 14]. This may explain the elevated plaque fluoride concentration observed in this study, with its peak occurring 30 min after application in the SDF and SDF + NaFV groups. Interestingly, the plaque fluoride levels in the NaFV group displayed two distinct peaks at 5 and 60 min after application, or a biphasic pattern. This biphasic pattern showed an initial rapid peak within 30 min, followed by a slower, sustained peak lasting several hours [11, 31]. The pattern of this study could be explained by early fluoride uptake into the plaque during short-term exposures, which resulted in elevated plaque fluoride concentrations near the saliva interface but low concentrations near the enamel surface, leading to an initial fluoride peak within the first 5 min. After 60 min of fluoride exposure, fluoride gradually penetrated into deeper plaque layers, so that higher fluoride concentrations were detected in the second peak [32]. The biphasic pattern was also observed in dental plaque following the use of dentifrices containing high fluoride concentrations [11]. Additionally, a biphasic pattern of fluoride was found in the plaque biofilm following the application of 5% NaFV [13].

When the groups were compared, it was found that the group receiving SDF alone had a shorter interval of fluoride retention in both the saliva and dental plaque, whereas the groups receiving NaFV, which included groups II (NaFV) and III (SDF + NaFV), had a longer interval of fluoride retention, with a maximum of 24 h of fluoride retention in plaque and saliva following SDF + NaFV application. This may be due to differences in the total amount of fluoride exposure, fluoride formulation, and application method, which all influenced fluoride retention in saliva and plaque [12–14, 33]. In the case of fluoride varnish, NaF dissociated in saliva, gradually releasing fluoride into the saliva and dental biofilm. The sticky properties of the varnish allowed for the slow release of fluoride, enabling prolonged exposure to the teeth [12, 13, 34]. The shorter oral retention of fluoride in the SDF group compared to NaFV could be attributed to several factors. First, the total amount of fluoride ion applied in the SDF group was significantly lower than in the NaFV and SDF + NaFV groups, resulting in fewer available fluoride ions in the oral cavity. In addition, fluoride from SDF predominantly diffused into the lesion, which reduced its surface availability [35]. Finally, even though the fluoride in SDF is stabilized by ammonia, it quickly dissolved into fluoride ions upon application, resulting in a brief period of oral retention [16, 17].

The study found that using SDF alone resulted in significantly lower fluoride levels in both plaque and saliva than using NaFV or SDF + NaFV. This could be related to the fact that, while 38% SDF had a higher concentration of fluoride than 5% NaFV, its application delivered less fluoride because lower quantities were used, particularly in this case, where only a drop of SDF was utilized. Furthermore, the amount of NaFV required was nearly double that of the amount of SDF [36]. As a result, when SDF was disseminated into the oral cavity, it yielded a relatively low fluoride concentration.

Comparison between the NaFV and SDF + NaFV groups revealed that fluoride levels in dental plaque returned to baseline within 24 h in both groups, with no significant difference in plaque fluoride levels at any time point between the two groups. However, at 5 to 60 min after fluoride treatment, only the NaFV group displayed a biphasic pattern with two separate peaks at 5 and 60 min, whereas the SDF + NaFV group had a continuous and steady level of plaque fluoride. As for salivary fluoride, there were no significant differences in fluoride levels between the two groups. However, fluoride levels in saliva returned to baseline 24 h post-application in the SDF + NaFV group, whereas it took 60 min for the NaFV group's levels to return to baseline. This could have been because the combined use of SDF and NaFV resulted in a higher total amount of fluoride being delivered into the oral cavity, necessitating a longer duration of salivary clearance, resulting in prolonged salivary fluoride retention in the SDF + NaFV group.

Regarding concerns about fluoride toxicity, the probable toxic dose of fluoride ingestion is 5 mg/kg. When the total amount of fluoride (6.77–7.89 mg) used in the SDF + NaFV group was compared with the weight of the children in our study, the range of fluoride given per kilogram of body weight fell to 0.26 to 0.65 mg/kg, which was significantly lower than the threshold of concern. As a result, it was considered safe to apply both forms of fluoride concurrently. However, changes in the bioavailability of the fluoride and its dynamics in plaque and saliva when the combination SDF + NaFV was given have yet to be investigated and compared. This was the first study to address this knowledge gap.

Our results indicated that using NaFV alone or a combination of SDF and NaFV led to no significant differences in salivary and plaque fluoride levels in preschool children. Even though the levels of plaque and salivary fluoride were not significantly different, the combined use of SDF and NaFV resulted in the longest salivary fluoride retention. The combination of SDF and NaFV, with its different objectives of using SDF to arrest dentinal caries and NaFV to promote remineralization and the arrest of enamel caries, is still necessary and offers twice the beneficial outcomes of using NaFV alone. Moreover, the effectiveness of preventing and arresting caries is believed to be equal to or higher in the group receiving both SDF and NaFV [5, 37, 38]. Additionally, previous studies have primarily involved only school-age children or older individuals [12, 13, 16, 17]. This was the first study to focus on preschool children given these two types of fluoride, which are frequently employed to prevent and arrest caries progression, which could lead to ECC [38].

Regular brushing routines with fluoride-free toothpaste were permitted in this study because most participants exhibited moderate to heavy plaque accumulation and poor oral hygiene. This indicates that, despite regular brushing, the brushing technique was ineffective for removing plaque. As a result, a sufficient amount of plaque can be obtained without the need to refrain from regular brushing procedures.

The limitations of this study were that participant enrollment was slightly lower than expected because some children were ill with common colds and fevers and could not take part in the study. Moreover, for the salivary fluoride analysis, this study used supernatant from centrifuged saliva rather than whole saliva or salivary sediment to evaluate salivary fluoride because analysis of free fluoride in the saliva supernatant is more relevant to fluoride's cariostatic effect and may be used to predict the efficacy of topical fluorides [39]. The disadvantage is that the concentration of fluoride in the supernatant is much lower than in whole saliva or salivary sediment, making it harder to detect in those who have low fluoride levels in their saliva's inorganic components [12, 40]. Furthermore, the participants clearly demonstrated individual variations. For example, in contrast to the overall group's average and median, the results showed that 4 of the participants in the SDF group and 3 of the individuals in the NaFV and SDF + NaFV groups had nearly undetectable fluoride levels in their saliva at baseline and during their recovery periods. These individual variations may have contributed to the group's high standard deviation. Regarding the time when fluoride levels in plaque and saliva returned to baseline, the lack of additional time points between 1 and 24 h may have restricted the precision in determining a certain return-to-baseline time. To better capture the return-to-baseline time, additional time points between 1 and 24 h should be included in future research. Lastly, SDF is delivered in an aqueous solution, which allows it to penetrate deep into the dentinal tubules. The fluoride ions also migrate largely into the lesion, which limits their availability at the surface [35, 41]. As a result, detecting fluoride in plaque after SDF application may not accurately reflect the total fluoride exposure or the extent to which fluoride exerts its therapeutic effect on the tooth structure.

In conclusion, professional fluoride applications significantly elevated salivary and plaque fluoride levels, hence improving fluoride bioavailability. However, when SDF alone was applied, plaque and salivary fluoride levels were lower than when NaFV was applied. Fluoride treatment with NaFV and a combination of SDF and NaFV increased fluoride levels in saliva and plaque, with no significant differences between the groups. Additionally, SDF + NaFV application resulted in prolonged salivary fluoride retention, which returned to normal within 24 h. These findings contribute to the existing evidence on the bioavailability of fluoride in plaque and saliva of preschoolers following professional topical fluoride applications. Future clinical studies should investigate how this bioavailability affects the caries arrest of treated lesions and the prevention of new lesions. It is also important to examine whether higher fluoride exposure in a single visit is preferable to distributing applications over multiple visits to limit exposure in younger children.

Author Contributions

S.H. and S.N. conception and study design; S.N., V.J., and C.A. methodology; C.A. investigation; S.H. and C.A. data acquisition and analysis; C.A. visualization; S.H. and S.N. supervision; S.H. and C.A. writing – original draft preparation; S.H., S.N., V.J., and C.A. writing – reviewing and editing. All authors have read and agreed to the published version of the manuscript.

Acknowledgements

The authors have nothing to report.

Ethics Statement

Approval was obtained from the Ethical Institutional Review Board, Faculty of Dentistry and the Faculty of Pharmacy, Mahidol University (COE.No.MU-DT/PY-IRB2023/054.0609) and registered in the Thai Clinical Trials Registry (TCTR20240705002).

Consent

Informed consent was obtained from all participants and their legal guardians.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

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Supporting Information

Additional supporting information can be found online in the Supporting Information section. **Figure S1:** Salivary fluoride levels in groups at different time points. Different letters above the bars indicate statistically significant differences between time points ($p < 0.05$). **Figure S2:** Plaque fluoride levels in groups at different time points. Different letters above the bars indicate statistically significant differences between time points ($p < 0.05$). **Table S1:** Salivary fluoride levels (ppm; μg/mL) at different time points. **Table S2:** Plaque fluoride levels (ppm; μg/g) at different time points.