

# Comparative Evaluation of Longevity of Fluoride Release from Three Different Fluoride Varnishes: An Observational Study

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## ABSTRACT

**Introduction:** The cycles of demineralization and remineralization result in a dynamic process of caries development. Caries are prevented by the shift in the balance from demineralization to remineralization at the tooth-oral fluid interface with the help of salivary fluoride levels (in parts per million). The advantages of fluoride varnish application over other substitutes like dentifrices, mouthrinses, gels, or foams are that varnishes are well tolerated by infants, young children, or children with special healthcare needs and have prolonged therapeutic effects. This study was formulated to evaluate and compare the longevity of fluoride release from different fluoride varnishes, namely Fluor Protector, Enamelast, and Enamel Pro varnish.

**Aim:** To evaluate and compare the longevity of fluoride release from three different fluoride varnishes.

**Materials and methods:** The study samples comprised 72 healthy permanent maxillary anterior teeth. The teeth were divided into four groups, with eighteen teeth in each group. The surfaces of all teeth were then covered by different colored nail varnish according to the respective group, except for a 3 × 3 mm window on the facial (labial) surface of the crown, where the test materials were applied according to their respective group. In group I, Fluor Protector varnish; in group II, Enamelast varnish; and in group III, Enamel Pro varnish was applied. Group IV was the control group; hence, no test material was applied. All specimens were then stored in plastic containers with a pH of 7.2 in artificial saliva at room temperature. The specimens were transferred into new plastic containers after 1 day, containing fresh artificial saliva, and solutions from previous plastic containers were taken for fluoride analysis. This process was repeated sequentially to analyze the amount of fluoride released in ppm from the specimens at the end of 1, 3, and 6 months.

**Statistical analysis:** Analysis was performed on Statistical Package for the Social Sciences (SPSS) software (Windows version 22.0).

**Results:** Conventionally used 1.5% ammonium fluoride varnish, that is, Fluor Protector varnish, showed the least release of fluoride ( $0.03 \pm 0.0$  ppm), while resin carrier-based 5% sodium fluoride varnish, that is, Enamelast varnish showed a good amount of fluoride release for 6 months continuously, that is, ( $0.16 \pm 0.06$  ppm) at last follow-up. Enamel Pro varnish, which has 5% sodium fluoride with amorphous calcium phosphate (ACP) formula, was found to be the best varnish as it released the maximum amount of fluoride in ppm in artificial saliva for up to 3 months ( $0.32 \pm 0.08$ ) but less than Enamelast varnish only at 6 months follow-up that is  $0.09 \pm 0.03$  ppm.

**Conclusion:** The present study concludes that based on the amount of fluoride released for 6 months duration, Enamel Pro varnish, followed by Enamelast varnish and Fluor Protector varnish, are advisable to apply for caries prevention.

**Keywords:** Artificial saliva, Caries, Dentinal hypersensitivity, Enamel Pro varnish, Enamelast varnish, Fluoride varnish, Fluor Protector varnish.

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## INTRODUCTION

The cycles of demineralization and remineralization result in a dynamic process of caries development. Various mineral ions such as calcium and phosphate ions, buffering agents, fluoride, and other substances in saliva reverse the natural demineralization of the tooth in the initial stages.<sup>1</sup> Salivary calcium and phosphate ions are utilized in the process of remineralization, which is aided by fluoride present in saliva, forming a crystalline layer on the teeth's subsurface lesions.<sup>2</sup> Various aspects of the assessment of caries risk and categorization of children into risk groups such as "high," "moderate," and "low" caries risk should be considered. After determining susceptibility to caries, an appropriate prevention regimen for individual patients must be formulated.<sup>3</sup> Since the last century, fluoride has been considered an anticariogenic substance since the introduction of water or salt fluoridation programs.<sup>4</sup>

Due to variations in the concentration of fluoride in water and insufficiency of this ion in many regions, other local means of fluoride exposure should be provided, such as toothpastes, mouthwashes, varnishes, and gels in dental offices or at home.<sup>5</sup> Fluoride varnishes were developed to enhance the time of contact between tooth enamel and fluoride.<sup>6</sup> Fluoride varnish application

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promotes the adsorption of fluoride ions by prolonged contact of fluoride to enamel, which was considered important at that time.<sup>7</sup> To reduce the prevalence of caries or to prevent dental caries in deciduous and permanent dentition in populations at higher risk,

it was concluded that the application of fluoride varnishes semi-annually is necessary by the American Dental Association.<sup>8</sup>

Concentrated forms of topical fluorides, such as fluoride varnishes, interact with saliva, resulting in the production of calcium fluoride (CaF<sub>2</sub>) on enamel by incorporating fluoride ions.<sup>9</sup> CaF<sub>2</sub> is considered to have side effects as it dissolves easily from the tooth surface soon after application.<sup>10</sup> Although other forms of topical fluoride, like fluoride gels, foams, and dentifrices, are available, fluoride varnish has many advantages over other substitutes; it is well tolerated by infants and young children and has prolonged therapeutic effects.<sup>11</sup> Also, its ease of application, with no risk of being swallowed, makes it preferable in younger children or children with special health care needs.

Fluor Protector varnish, that is, 1.5% ammonium fluoride varnish, has been used widely since the 20th century. Since then, many new generations of materials have been developed to enhance the longevity of fluoride release. Enamelast varnish, that is, 5% sodium fluoride xylitol sweetened varnish, occludes dentinal tubules mechanically and chemically to treat tooth hypersensitivity, and by its patent adhesion promoting formula retention is enhanced resulting in superior release and absorption of fluoride.<sup>12</sup> Another varnish is Enamel Pro varnish, which has 5% sodium fluoride along with amorphous calcium phosphate (ACP). ACP leads to the incorporation of calcium and phosphate ions into the tooth surface with an ideal strategy by which the demineralization process can be reverted.<sup>13</sup>

Due to insufficient literature available on these materials, the present study was done to evaluate and compare the longevity of fluoride release from these varnishes, which are Fluor Protector, Enamelast, and Enamel Pro varnish.

## MATERIALS AND METHODS

This study was performed at the Department of Pediatric Dentistry, in the Institute of Dental Sciences, Bareilly, Uttar Pradesh, India, in collaboration with the Indian Institute of Toxicology Research, Lucknow, Uttar Pradesh, aimed to evaluate and compare the longevity of fluoride release from three different fluoride varnishes. The equipment used in the study is shown in Figure 1.

### Methodology

The study samples comprised 72 healthy permanent maxillary anterior teeth. All teeth were made free of debris and calculus with the help of an ultrasonic scaler. All sample teeth were kept in normal saline for use further in the study. These sample teeth were then divided into four groups, with 18 teeth in each ( $N = 18$ ), where group I, group II, and group III were test groups. Group IV was the control group, and all teeth were kept in their respective containers.

All the sample teeth were washed with clean water to remove the normal saline solution and dried with dry sterile gauze. The surfaces of all teeth were covered by different colored nail varnish according to the respective group, leaving a window of 3 × 3 mm on

the facial (labial) surface of the crown, on which the test materials were applied with the help of applicators. In group I, Fluor Protector varnish; in group II, Enamelast varnish; in group III, Enamel Pro varnish was applied; and group IV was the control; hence, no test material was applied on samples. After the test materials were applied, the specimens were stored individually in separate plastic containers containing artificial saliva with pH 7.2 at room temperature. After 1 day, all the specimens were transferred into new separate plastic containers containing fresh artificial saliva. After transferring the specimens from previous containers to new containers, the solutions from previous plastic containers were taken for the fluoride analysis. A thermocol ice box was used to transport samples for testing. This process was repeated sequentially at the end of 1, 3, and 6 months. Fluoride ion concentration was measured by Metrohm 940 Professional IC Vario, which measures released ions of fluoride in parts per million in the solution.

### Statistical Analysis

Data were calculated in mean ± standard error. All the groups were compared using two factors: repeated measure (RM) and analysis of variance (ANOVA). The significance of the mean difference within (intra) and between (inter) the groups was calculated using Tukey's honestly significant difference *post hoc* test. A two-tailed ( $\alpha = 2$ )  $p < 0.05$  was considered statistically significant. Analysis was performed on Statistical Package for the Social Sciences (SPSS) software (Windows version 22.0).

## RESULTS

The outcome of this study was that the amount of fluoride released in artificial saliva was measured using parts per million.

The amount of fluoride released among these three groups (groups I, II, and III) over the postperiods (day 1, 1 month, 3 months, and 6 months) are summarized in Table 1 and also shown



Fig. 1: Metrohm 940 Professional IC Vario

Table 1: Fluoride release (ppm) of four groups over the periods

Period	Group I (n = 18)	Group II (n = 18)	Group III (n = 18)
Day 1	0.56 ± 0.09	1.38 ± 0.21	3.47 ± 0.19
1 month	0.36 ± 0.06	1.24 ± 0.13	1.57 ± 0.12
3 months	0.11 ± 0.02	0.26 ± 0.06	0.32 ± 0.08
6 months	0.03 ± 0.01	0.16 ± 0.06	0.09 ± 0.03

The fluoride release of three groups over the periods was summarized in mean ± SE

in Figure 2. For the first three postperiods, it was highest in group III as compared to groups II and I, respectively. However, at the end of 6 months, that is, the fourth follow-up, the maximum release of fluoride was reported as (group II > III > I). No fluoride release was seen from group IV at any postperiod. Comparing the effect of both group and period together on fluoride release, RM ANOVA showed that the effect was ( $F = 137.20, p < 0.001$ ) and period ( $F = 156.17, p < 0.001$ ) on fluoride release (Table 2).

Further, for each group, comparing the difference in mean fluoride release between the periods (i.e., intragroup), the Tukey test showed ( $p < 0.001$ ) different and higher fluoride release at day 1 as compared to 1, 3, and 6 months in group III (Table 3 and Fig. 3).

Similarly, for each period, comparing the difference in mean fluoride release between the groups (i.e., intergroup), the Tukey test presented ( $p < 0.001$ ) different and higher fluoride release in both groups II and III as compared to group I at both day 1 and 1 month

(Table 4 and Fig. 4). Moreover, at day 1, it also showed significantly ( $p < 0.001$ ) different and higher fluoride release in group III as compared to group II.

## DISCUSSION

The most common childhood disease is dental caries. Caries widely affect the economy and the quality of life, making it a serious health issue of public interest.<sup>14</sup> For many children, it is still challenging and time-consuming to prevent dental caries.<sup>15</sup> In many developing countries, it has been found that dental and periodontal diseases are consistently increasing the economic burden on people.<sup>16</sup> This increases the need for caries preventive programs. The application of fluoride is one such preventive strategy. Remineralization occurs more readily with surface-softened lesions than with a subsurface lesion due to the easy penetration of calcium, phosphate, and fluoride ions to the outer surface.<sup>17</sup> Saliva at an early stage can

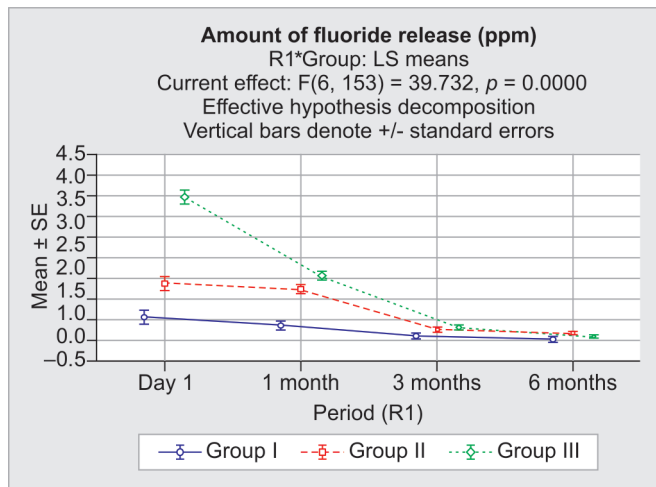


Fig. 2: Mean fluoride release of three groups over the periods

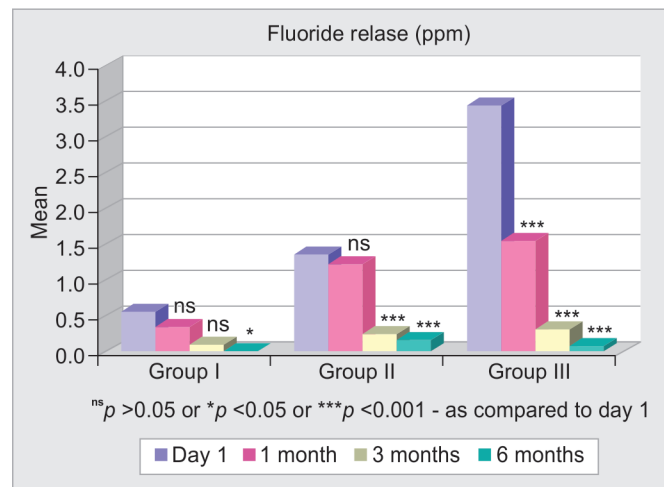


Fig. 3: For each group, comparisons of difference in mean fluoride release between the periods

Table 2: Comparisons ( $p$ -value) of mean fluoride release of three groups over the periods using RM ANOVA

Source of variation	Sum of square	Degree of freedom	Mean square	F-value	p-value
Group	43.45	2	21.72	137.20	<0.001
Error	8.08	51	0.16		
Period	102.34	3	34.11	156.17	<0.001
Period × group	52.08	6	8.68	39.73	<0.001
Error	33.42	153	0.22		

RM ANOVA, repeated measures analysis of variance; F-value, ANOVA F-value

Table 3: For each group, comparison ( $p$ -value) of difference in mean fluoride release (ppm) between the periods by Tukey test

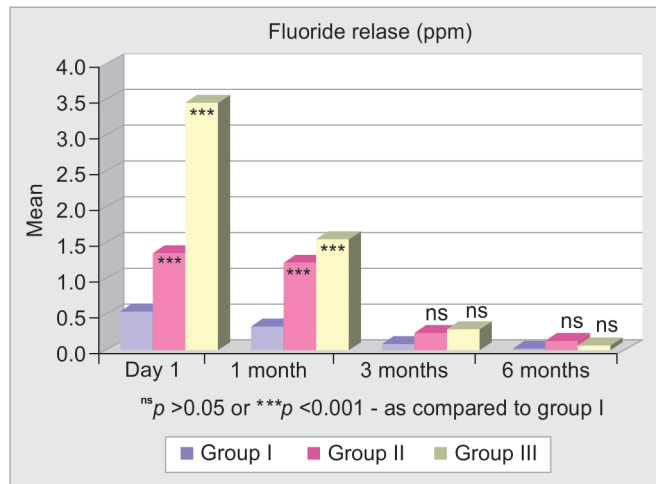
Comparison	Group I		Group II		Group III	
	Mean difference	p-value	Mean difference	p-value	Mean difference	p-value
1 day vs 1 month	0.20	0.981	0.14	0.999	1.90	<0.001
1 day vs 3 months	0.45	0.138	1.12	<0.001	3.15	<0.001
1 day vs 6 months	0.53	0.030	1.22	<0.001	3.38	<0.001
1 vs 3 months	0.25	0.903	0.98	<0.001	1.25	<0.001
1 vs 6 months	0.33	0.597	1.07	<0.001	1.48	<0.001
3 vs 6 months	0.08	1.000	0.10	1.000	0.23	0.952

Diff, difference

**Table 4:** For each period, comparison (*p*-value) of difference in mean fluoride release between the groups by Tukey test

Comparison	Day 1		1 month		3 months		6 months	
	Mean difference	<i>p</i> -value	Mean difference	<i>p</i> -value	Mean difference	<i>p</i> -value	Mean difference	<i>p</i> -value
Group I vs II	0.82	<0.001	0.88	<0.001	0.15	0.998	0.14	0.999
Group I vs III	2.91	<0.001	1.21	<0.001	0.21	0.965	0.06	1.000
Group II vs III	2.09	<0.001	0.33	0.545	0.06	1.000	0.07	1.000

Diff, difference

**Fig. 4:** For each period, comparisons of difference in mean fluoride between the groups

reverse the natural demineralization of the tooth due to various minerals, buffering agents, fluoride, and other substances.

Topical fluorides have been effectively used for a long time to prevent dental caries, especially in children.<sup>18</sup> Fluoride is found to enhance the saturation of calcium and phosphate ions in plaque, lowering the solubility constant of calcium and fluoride, ultimately preventing their loss from the tooth structure.<sup>19</sup> Fluor Protector is a fluoride varnish with 1.5% ammonium fluoride. It was developed in 1975 by Arends and Schuthof. It primarily has 0.9% difluoro silane in solvents like polyurethane varnish base with ethyl acetate and isoamyl propionate. The fluoride content in this composition is equivalent to 1000 ppm in solution.<sup>20</sup>

The present study showed that the least fluoride was released from Fluor Protector varnish (group I), where the maximum amount of fluoride was released on day 1, and the minimum release was observed at the end of 6 months. The reason for the least release of fluoride is that saliva is usually saturated with calcium, and topical application of fluoride leads to the formation of an insoluble CaF<sub>2</sub> layer, which adheres strongly to the porous tooth surfaces. The adsorption of hydrogen phosphate ions additionally stabilizes the CaF<sub>2</sub> layer, preventing the further loss of fluoride into the saliva.<sup>21</sup> The results of this study were found in line with the results observed by Virupaxi et al.,<sup>1</sup> to compare and evaluate the duration of fluoride release from three compositions of fluoride varnishes—Clinpro TM XT, Fluoritop SR, and Fluor Protector varnish, where least fluoride was released from Fluor Protector varnish. According to the study conducted by Arends et al. in 1997, Fluor Protector more readily penetrated the dentinal tubules than the resinous varnish Duraphat.<sup>22</sup>

Enamelast varnish is a xylitol-sweetened varnish containing 5% sodium fluoride solute in resin solvent. Flavors make it more

acceptable to children. It has a patent formula of an adhesion-promoting agent, advancing retention and providing superior fluoride release and uptake.<sup>12</sup> In a study conducted by Godoi et al.<sup>12</sup> to evaluate the remineralization effect on artificial carious lesions, the result showed that Enamelast varnish promoted surface remineralization and higher fluoride concentration than the other varnishes. Contrary to their finding, the present study reported comparatively lesser fluoride release from Enamelast varnish (group II), consecutively for 3 months, with significantly ( $p < 0.001$ ) different and higher fluoride release from Enamel Pro varnish (group III). However, at the end of 6 months, a higher amount of fluoride release was seen from Enamelast varnish than from Enamel Pro varnish and Fluor Protector varnish.

Enamel Pro varnish contains ACP to provide additional restorative power. Four times higher fluoride absorption into enamel is aided by ACP.<sup>2</sup> Nalbantgil et al.<sup>23</sup> evaluated the effect of two sealers—Durafluor and Enamel Pro varnish, adjacent to orthodontic brackets. Results revealed that minimum demineralization was seen in Enamel Pro varnish. The present study showed the maximum fluoride release from the Enamel Pro varnish group on day 1. Fluoride-releasing rates and their concentration were found to be gradually decreasing in follow-up assessments at 1, 3, and 6 months from all the groups. However, at all the follow-up periods, maximum fluoride was released in group III, followed by groups II and I (Enamel Pro varnish > Enamelast varnish > Fluor Protector varnish).

Virupaxi et al.<sup>1</sup> measured fluoride ion concentration in artificial saliva using ion-selective electrodes. The disadvantages noted while using ion-selective electrodes are that the precision is very low, and organic solutes can foul the electrodes. Therefore, in the present study, the fluoride content released in specimens was calculated by Metrohm 940 Professional IC Vario, which is an ion chromatograph with high precision. It is used in liquid chemistry analysis where it can detect the concentration of major low molecular weight organic and inorganic anions such as fluoride, chloride, nitrate, and sulfate as well as major cations such as lithium, sodium, potassium, calcium, and magnesium in parts per million.<sup>24</sup>

Fluoride varnishes can be an effective preventive strategy for caries development or progression and an effective treatment modality for hypersensitivity due to exposed dentinal tubules. Hence, the use of fluoride varnishes is not limited to children and adolescents to prevent caries; it is also advisable to apply varnish in elderly patients as a cure for hypersensitivity.

## CONCLUSION

Systemic consumption, as well as topical application of fluoride, has been proven to be the gold standard in the prevention of dental caries. Fluoride varnishes, due to their ease of application and least patient cooperation, are the best fluoride therapy for younger children. The present study compared three different formulations of fluoride varnishes for their longevity in the release of fluoride



in artificial saliva. Results showed that conventionally used 1.5% ammonium fluoride varnish, that is, Fluor Protector varnish, showed the least release of fluoride. Resin carrier-based 5% sodium fluoride varnish, that is, Enamelast varnish, showed a good amount of fluoride release for 6 months. Enamel Pro varnish was found to be the best varnish as it released the maximum amount of fluoride in ppm in artificial saliva for up to 3 months. In the 6th month, Enamelast varnish showed a better amount of fluoride released than Enamel Pro varnish and Fluor Protector varnish. These results suggest that from different fluoride varnishes, the longevity of fluoride release decreases at sequential intervals. So, fluoride varnish application should be done, depending upon the amount and duration of fluoride release from that varnish.

### Limitations

The only limitation of the present study is that it does not mimic the diverse conditions present in the oral cavity, such as saliva, antimicrobial proteins, enzymes, and the absence of the demineralization process, all of which may affect dental caries development.

### Recommendations

The use of fluoride varnishes is a noninvasive technique that requires the least cooperation from uncooperative patients or children with special health care needs. Application of topical fluoride, as suggested by the results of this study, is recommended to be done four times per year in patients with high caries risk. It also minimizes dentinal hypersensitivity, proving its importance in individuals of all age-groups.

### Future Scope of the Study

Fluoride is a double-edged sword, and the present study was conducted in *in vitro* conditions, so further studies in *in vivo* settings are required to generalize and confirm the results and safety of various fluoride varnishes used in this study.

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### REFERENCES

- Virupaxi SG, Roshan NM, Poornima P, et al. Comparative evaluation of longevity of fluoride release from three different fluoride varnishes - an *in vitro* study. *J Clin Diagn Res* 2016;10(8):ZC33-ZC36. DOI: 10.7860/JCDR/2016/19209.8242
- Schemahorn BR, Wood GD, Mc Hale W, et al. Comparison of fluoride uptake into tooth enamel from two fluoride varnishes containing different calcium phosphate sources. *J Clin Dent* 2011;22(2):51-54.
- Vaikuntam J. Fluoride varnishes: should we be using them? *Pediatr Dent J* 2000;22(6):513-516.
- Bansal A, Ingle NA, Kaur N, et al. Recent advancements in fluoride: a systematic review. *J Int Soc Prev Community Dent* 2015;5(5):341-346. DOI: 10.4103/2231-0762.165927
- Abdoli E, Javadinejad S, Khosravanifard B. Comparison of fluoride uptake into dental enamel from two types of sodium fluoride varnishes (in vitro). *J Res Dent Sci* 2015;11(4):215-220.
- Bonetti D, Clarkson JE. Fluoride varnish for caries prevention: efficacy and implementation. *Caries Res* 2016;50(Suppl 1):45-49. DOI: 10.1159/000444268
- Comar LP, Souza BM, Grizzo LT, et al. Evaluation of fluoride release from experimental TiF4 and NaF varnishes in vitro. *J Appl Oral Sci* 2014;22(2):138-143. DOI: 10.1590/1678-775720130574
- Pańczyzyn DP, Kaczmarek U. Fluoride release from fluoride varnish under *in vitro* and *in vivo* conditions. *Dent Med Probl* 2017;54(4):327-331.
- Epple M, Enax J, Meyer F. Prevention of caries and dental erosion by fluorides-a critical discussion based on physico-chemical data and principles. *Dent J (Basel)* 2022;10(1). DOI: 10.3390/dj10010006
- Seppä L. Fluoride varnishes in caries prevention. *Med Princ Pract* 2004;13(6):307-311. DOI: 10.1159/000080466
- Attiguppe P, Malik N, Ballal S, et al. CPP-ACP and fluoride: a synergism to combat caries. *Int J Clin Pediatr Dent* 2019;12(2):120-125. DOI: 10.5005/jp-journals-10005-1608
- Godoi FA, Carlos NR, Bridi EC, et al. Remineralizing effect of commercial fluoride varnishes on artificial enamel lesions. *Braz Oral Res* 2019;33:e044. DOI: 10.1590/1807-3107bor-2019.vol33.0044
- Zabokova-Bilbilova E, Sotirovska-Ivkovska A, Evrosimovska B, et al. Effect of fluoride varnish on demineralization adjacent to orthodontic brackets. *Balk J Stom* 2012;16:157-160.
- Omitola O, Arigbede A. Prevalence of dental caries among adult patients attending a tertiary dental institution in the South-South region of Nigeria. *Port Harcourt Med J* 2011;6(1):52-58.
- Urzuai I, Mendoza C, Arteaga O, et al. Dental caries prevalence and tooth loss in Chilean adult population: first national dental examination survey. *Int J Dent* 2012;2012:810170. DOI: 10.1155/2012/810170
- Agbelusi GA, Jeboda SO. Oral health status of 12-year-old Nigerian children. *West Afr J Med* 2006;25(3):195-198. DOI: 10.4314/wajm.v25i3.28277
- Roopa KB, Pathak S, Poornima P, et al. White spot lesion: a literature review. *J Pediatr Dent* 2015;3(1):1. DOI: 10.4103/2321-6646.151839
- Newbrun E. Topical fluorides in caries prevention and management: a North American perspective. *J Dent Edu* 2001;65(10):1078-1083.
- Corona SA, Nascimento TN, Catirse AB, et al. Clinical evaluation of low-level laser therapy and fluoride varnish for treating cervical dentinal hypersensitivity. *J Oral Rehabil* 2003;30(12):1183-1189. DOI: 10.1111/j.1365-2842.2003.01185.x
- Zero DT, Raubertas RF, Fu J, et al. Fluoride concentrations in plaque, whole saliva, and ductal saliva after application of home-use topical fluorides [published erratum appears in *J Dent Res* 1993 Jan;72(1):87]. *J Dent Res* 1992;71(11):1768-1775. DOI: 10.1177/00220345920710110201
- Nelson DG, Jongebloed WL, Arends J. Morphology of enamel surfaces treated with topical fluoride agents: SEM considerations. *J Dent Res* 1983;62(12):1201-1208. DOI: 10.1177/00220345830620120501
- de Bruyn H, Arends J. Fluoride varnishes: a review. *J Biol Buccale* 1987;15(2):71-82.
- Nalbantgil D, Oztoprak MO, Cakan DG, et al. Prevention of demineralization around orthodontic brackets using two different fluoride varnishes. *Eur J Dent* 2013;7(1):41-47.
- Seubert A, Frenzel W, Schäfer GH, et al. Sample preparation techniques for ion chromatography. *Metrohm* 2021;2:1-12.