

Comparative Evaluation of Remineralization and Antimicrobial Efficacy of Maximum Fluoride Containing Toothpastes Available in India - In-Vivo Study

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ABSTRACT

Background: Early childhood caries (ECC) is a common type of dental caries with an overall prevalence of 49.6% in India. Dentifrices-containing 1000-1500ppm concentration of fluorides and certain remineralizing agents are known to be effective in reversal of early enamel lesions and reduction of dental caries prevalence. However, in India, the mean fluoride content of dentifrices available for children is only 449.5 ± 26.1 ppm. Only very few toothpastes with the recommended concentration of fluoride is available in the Indian market, for children below the age of 6 years.

Aims: To compare and evaluate the remineralization potential and antimicrobial efficacy of toothpaste with highest amount of fluoride, available in India for children below 6years of age, with non-fluoridated remineralizing children's toothpaste. To assess the Total Soluble Fluoride (TSF) concentration of fluoridated toothpastes.

Methodology: 95 extracted or naturally exfoliated human primary teeth were decoronated and sectioned mesiodistally. Specimens were demineralized and randomly

divided into six groups (3 fluoridated and 3 non-fluoridated remineralising toothpaste groups): group 1: Omnident®- 917ppm, group 2: AquaFresh®™ little teeth- 1450ppm, group 3: Colgate kids®- 1000ppm, group 4: Enafix® (Ca. orthophosphate + Ca. Sucrose phosphate), group 5: Aclaim® (Nano HAP) and group 6: Shy-NM® (Novamin). The samples were placed in the demineralizing solution for 48h and subjected to Vickers hardness test (VHT) and SEM to obtain the baseline values. Then the samples were remineralized using the respective toothpastes and were subjected to SEM and VHT at 21days, 3months and 6months intervals. Antimicrobial activity of all the dentifrices was determined by agar well diffusion method and selective ion electrode analysis was used to evaluate the Total Fluoride (TF) and Total Soluble Fluoride (TSF) concentrations among the three fluoridated toothpastes. The data obtained were statistically analysed.

Results: SEM images showed mineral deposition in all the groups obliterating the defects caused due to demineralization at various intervals. Remineralising agents showed significantly higher remineralisation especially SHY-NM when compared to Fluoridated toothpastes. However they did not show any antimicrobial efficacy. Fluoridated toothpastes

showed antimicrobial efficacy against *S. mutans* and the highest antimicrobial efficacy was seen with Aquafresh followed by Omnident and Colgate Kids which was directly proportional to the TSF concentration of the toothpastes.

Conclusions: Highest remineralisation of artificial enamel lesions was shown by non-fluoridated remineralising toothpaste, however, the fluoridated dentifrices displayed both antimicrobial and remineralising properties. The Total Soluble Fluoride (TSF) concentration of fluoride toothpastes were generally slightly less than the Total Fluoride (TF) concentration (TSF/F (%): 97%- AquaFresh®TM, 61.1%- Colgate kids® and 96.5% in Omnident®), which can be correlated to the dose-dependant antimicrobial efficacy as well as remineralisation of the fluoridated toothpastes.

Keywords: remineralization potential, antimicrobial efficacy, toothpaste with highest amount of fluoride, dental caries

INTRODUCTION

Dental caries remains a recognized global health problem despite breakthroughs in scientific knowledge, improved oral hygiene, and the ever-increasing availability of new commercial preventive solutions[1]. A worldwide systematic review revealed that the prevalence of dental caries in children was 46.2% and 53.8% in primary and permanent teeth respectively[2]. In Indian population, the prevalence of dental caries among 3–18 years of age was 52% and the maximum overall prevalence was 58% which was noted in the mixed dentition[3]. Therefore, it is the most common chronic disease of childhood that interferes with normal intake of nutrition, speech, and daily routine activities[4]. Dental caries is a dynamic process with strewn period of demineralization and remineralization, resulting in caries initiation and its progression[1]. When the pH falls below 5, demineralization of the enamel begins, marking the initiation of enamel caries with subsurface enamel demineralisation. This lesion can be remineralized[5,6] before the process continues to form cavitation.

Fluoride is the gold standard for preventing dental caries, with multiple systematic reviews confirming its role. The maximum permissible fluoride concentration for a toothpaste varies according to location and age. The EAPD recommends the use of fluoride toothpaste of concentration 500ppm from eruption of first tooth to less than 2years of age, 1000ppm for children 2-6years (1000+ fluoride concentrations may be considered based on the individual caries risk) and 1450ppm for children over 6years of age[18]. On similar lines, IAPD Bangkok Declaration also recommends to perform twice daily toothbrushing with fluoridated toothpaste with at least 1000 ppm in all children, using an age- appropriate amount of paste to effectively reduce the burden of ECC[19]. In India, the Drugs and Cosmetics Act, 1940, gives the labelling guidelines for fluoridated toothpaste, but the mean fluoride content of dentifrices is only 449.5 ± 26.1 ppm[20]. This may be due to fear of inadvertent swallowing of injudicious amount of fluoridated dentifrice while tooth brushing which may lead to chronic fluoride toxicity in the form of dental fluorosis[20]. However, a child-sized toothbrush covered with a full strip of toothpaste holds approximately 0.75g to 1.0g of toothpaste, and each gram of fluoride toothpaste, contains approximately 1.0mg of fluoride and a child may inadvertently swallow as much as 0.3-0.8mg [22] which is under the recommendation of daily intake of F. In children, only a 'rice-grain size' or 'pea-sized' amount of toothpaste is recommended, which further reduces the concentration of fluoride in case of ingestion.

Considering the narrow gap between caries reduction benefit and fear of fluoride toxicity, regulatory authorities have limited the fluoride concentration in non-prescription toothpastes to within 1,000–1,500 ppm[11]. This has led to the development of fluoride alternatives such as effective remineralizing agents such as casein phosphopeptides (CPPs) and amorphous calcium phosphate (ACP)- filled

methacrylate composites. Bioactive glass, Nano- HAP, Calcium sucrose orthophosphate have also been introduced and incorporated into toothpaste. Recently, newer remineralising agents such as Bioactive glass, Nano-HAP, Calcium sucrose orthophosphate have been introduced and incorporated into toothpaste and are among the commonly available toothpastes in India. Bioactive glass has several unique characteristics, such as the ability to biomimetically mineralize tissues in a way similar to the body's mineralization characteristics [12]. Nano-HA is a promising material that is biocompatible, bioactive and similar to the bone and mineral structure of teeth [13]. CaSP is another remineralising agent that supplies both calcium and phosphate in a soluble form [14]. Additional to reversal of incipient caries, prevention of dental caries is also utmost priority. This can be achieved by mechanical plaque control and oral hygiene products containing antimicrobial agents and toothpastes are a widely used agents for dental biofilm control [14,15]. Therefore this study was planned to evaluate and compare paediatric toothpaste with highest amount of fluoride available in India for their remineralizing potential of initial caries and antimicrobial effectiveness over the non-fluoride remineralising toothpastes. Promotion of the use of fluoride toothpaste should go hand in hand with quality control measures on its efficacy. Moreover, fluoride content should be accurately labelled on the tube and the carton [16]. For fluorides to be effective in the oral environment, toothpastes must contain adequate concentration of TSF to interfere with the development of caries, thereby reducing enamel demineralization and promoting remineralization. Hence, we also determined the Total Soluble Fluoride of fluoridated toothpastes which is responsible for the anti-caries and anti-bacterial actions of fluoride.

MATERIALS AND METHODS

The study followed an *in vitro* experimental design and was carried out

after obtaining Institutional Ethics Committee clearance. To assess and compare remineralization potential 95 extracted or naturally exfoliated human primary teeth were selected.

Inclusion criteria: Extracted or exfoliated human primary teeth that showed no evidence of white spot lesions, enamel cracks, caries or developmental defects were selected and the exfoliated teeth had at least 1/4th root structure remaining.

Exclusion criteria: Primary teeth having caries, restorations or having any developmental defects were excluded and teeth having less than 1/4th of root structure were also excluded.

The teeth selected were rinsed under tap water to get rid of debris. Calculus and soft tissues were cleaned from the surfaces of the teeth using ultrasonic scaler and stored in formalin solution until use. Enamel specimen preparation was done by decoronating teeth at CEJ and sectioned mesiodistally using diamond disc at slow speed with water as coolant. Then windows were created of 4mm x 4mm in size for each sample and the rest of the area on the sample were made completely resistant to acid by carefully coating it with nail varnish. Then custom made plastic cylindrical moulds were prepared and self-cure acrylic resin were sprinkled into them. Each enamel specimen was then embedded onto the top of partially set acrylic resin and were allowed to set completely. These specimens were then stored in artificial saliva. The specimens were immersed in the solution at 37°C for 2-4 days to create artificial lesions. The post-demineralisation evaluation was done by visual method using international caries detection and assessment system II (ICDAS II) [17]. If no lesions were found, the sample was reimmersed in demineralization solution. After demineralization, 5 samples were washed in running water, damp dried and subjected to SEM and VHT for baseline values. The demineralising solution were

changed after every 5 days. The remaining 90 specimens were then divided into 6 groups and subjected to remineralization treatment.

90 teeth were divided into 6 groups:-

- 1) group 1: treated with Omnident®- 917ppm.
- 2) group 2: treated with AquaFresh®™ little teeth- 1450ppm.
- 3) group 3: treated with Colgate kids®- 1000ppm.
- 4) Group 4: treated with Enafix® (Ca. orthophosphate + Ca. Sucrose phosphate).
- 5) group 5: treated with Aclaim® (Nano HAP).
- 6) group 6: treated with Shy- NM® (Novamin).

Specimens were brushed with a cotton swab dipped with pea-size amount of toothpaste for 2 mins, then rinsed with tap water for 5 seconds, and reimmersed into the artificial saliva. Each group were assigned its own container of artificial saliva to avoid cross-contamination. These procedures were conducted twice a day, once in the morning at 7:30 and in the evening at 21:00. Artificial saliva was changed every 24 hours. In the oral environment, after food is taken, oral bacteria decompose sugar and produces acid, causing pH decline. To simulate this in-vitro, all the specimens were immersed in the demineralization solution for 3 hours and in artificial saliva for the remaining 21 hours every day. The 3-hour demineralization time compensates for the total pH decrease time after 3 meals.

Intervals- 21 days, 3 month and 6 months.

After the completion of each treatment duration, specimens were subjected to VHT and SEM.

The antimicrobial activity of dentifrices was assessed with agar well diffusion method using De Man, Rogosa and Sharpe (MSR) Agar was used for *Lactobacillus acidophilus* and *Mitis Salivarius* (MS) Agar for *Streptococcus mutans*. *Streptococcus mutans* and *Lactobacillus* strains were streaked respectively onto the separate

plates and a lawn culture was prepared. Once the organisms were inoculated and spread on the surface of the medium, wells of 25 mm diameter each was punctured on each plate with the help of cork borer and agar plugs. The wells were then filled with 6 different toothpastes respectively on each plate using micro pipettes. The plates were incubated at 37°C in the incubator for 3 days. The zone of inhibition was then measured for each disc after 3 days of inoculation to the nearest millimetre using a digital vernier calliper.

Total Fluoride and Total Soluble Fluoride concentration was analysed for 3 Fluoridated toothpastes were also carried out using Fluoride ion electrode test. Three toothpaste tubes, with their caps closed, was carefully squeezed in order to homogenise the contents. The tubes were then opened and the first 3g of toothpaste to emerge from the tube was discarded and analysis was done in duplicates according to a protocol modified from Pearce.[4] 90-110mg of toothpaste was weighed out then homogenised in 10.0mL of distilled water. The suspension was prepared and proper mixing was ensured using a stirrer. Duplicates of 0.25mL of the suspension was then transferred to sterilised containers for Total Fluoride (TF) analysis. The remaining suspension was transferred to a test tube and centrifuged (3,000g, 10min, r.t.) to remove Ionic Fluoride (IF) bound to the abrasive. Duplicates of 0.25mL of the supernatant was transferred to a sterilised plastic container to determine Total Soluble Fluoride (TSF) concentrations. For all TF and TSF containers, 0.25mL of 2.0M HCl was added, and after 1 h at 45°C, the samples were neutralised with 0.5mL 1.0M NaOH and buffered with 1.0mL of TISAB II (1.0M acetate buffer, pH 5.0, containing 1.0 M NaCl and 0.4% CDTA). Analysis was carried out using an ion specific electrode coupled to an ion analyser calibrated with fluoride standards.

One-way ANOVA test was used to compare the mean values for both antimicrobial efficacy and fluoride concentration analysis.

One-way ANOVA test followed by Tukey's post hoc analysis was used to compare the mean values of different minerals by SEM and Hardness values between different study groups at each time interval. Repeated measures of ANOVA test followed by Bonferroni's post hoc test was used to compare the mean values of different minerals by SEM and Hardness values between different time intervals in each study group. The level of significance was set at $P < 0.05$.

RESULTS

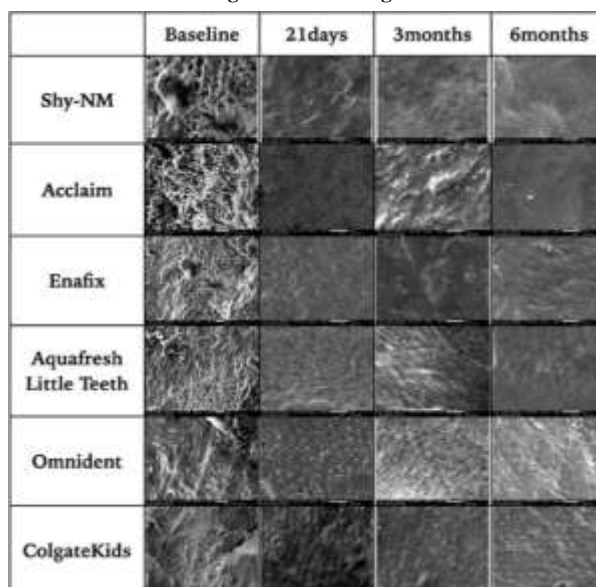
SEM results:

Normal enamel shows a characteristic fish scale appearance with a smooth and intact

surface. Following demineralization, the rods appeared collapsed due to lack of proper orientation of the hydroxyapatite crystals and the fish scale appearance of normal enamel

disappeared. The enamel surface appeared rough and uneven, and increased porosities were observed. A minor honeycomb pattern of demineralized enamel was observed. After remineralization, all the test groups revealed a layer of surface deposition of minerals obliterating the defects, filling up the rods and interrod region, showing an uneven yet more homogeneous surface at each interval compared to the baseline (Figure - 1).

Figure 1. SEM images



Vickers Hardness Test Results:

To quantitatively analyse the remineralisation of the tested samples, VHT was performed. Table-1(Graph-1) summarises the mean microhardness b/w different time intervals using One-way ANOVA Test.

All the groups showed significant increase in remineralisation with time, Non-Fluoridated Remineralising toothpastes had

better remineralisation, maximum remineralisation was seen in, ShyNM among the Non- Fluoridated Remineralising toothpastes and Aquafresh among the fluoridated toothpastes and it can also be seen that, remineralisation at 21days, by non-fluoride remineralising toothpastes, was more, than remineralisation achieved, at 6 months by Fluoridated toothpastes.

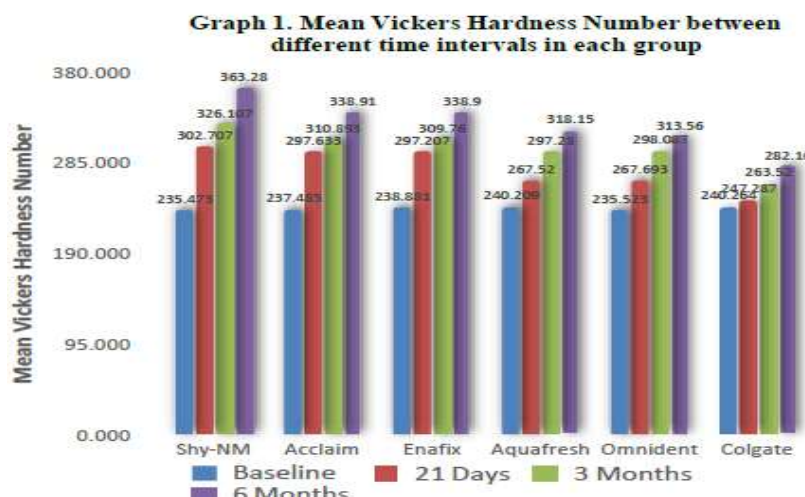


Table-2 shows multiple comparison of mean difference in VHN b/w time intervals in each group using Bonferroni's post hoc Test. In all the study groups, the mean VHN significantly increased between different time intervals ($p < 0.001$), except for Colgate

Kids group. Multiple comparison between time intervals demonstrated that the mean VHN for all the study groups except Colgate Kids at T3 was significantly highest as compared to T0, T1 & T2.

Table 1. Comparison of mean Vickers Hardness Number b/w different time intervals using One-way ANOVA

Groups	Baseline		21 Days		3 Months		6 Months		p-value
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	
Shy-NM	235.473	4.141	302.707	2.336	326.107	4.629	363.280	2.518	<0.001*
Omnident	235.523	1.584	267.693	2.206	298.087	3.326	313.560	3.349	<0.001*
Acclaim	237.485	2.889	297.633	3.013	310.893	0.999	338.913	5.372	<0.001*
Enafix	238.881	6.950	297.207	2.942	309.760	1.996	338.900	3.452	<0.001*
Aquafresh	240.209	3.818	267.520	2.535	297.280	2.449	318.153	1.676	<0.001*
Colgate	240.264	7.925	247.287	5.715	263.520	6.171	282.160	5.854	<0.001*

* - Statistically Significant

Table 2. Information labelled on the toothpaste samples and results of fluoride analysis.

Groups	Fluoride Compound	Claimed Fluoride Concentration (F*)	Total Fluoride (TF) Concentration	Total Soluble Fluoride (TSF) Concentration	TSF/TF(%)
Aquafresh	NaF	1450ppm	1440ppm	1397ppm	97%
Colgate	SMFP	1000ppm	826ppm	505ppm	61.1%
Omnident	SMFP	917ppm	923ppm	891ppm	96.5%

Fluoride concentration analysis:

The three toothpaste brands analysed in this study had declared fluoride concentration of 1450ppm (Aquafresh), 1,000ppm (Colgate kids) and 917ppm (Omnident). Analysis revealed mean Total Fluoride concentrations (TF) to be 1,440ppm, 826ppm and 923ppm respectively. The TF concentration in one toothpaste (Omnident) was found to be slightly more than declared on their labelling.

The Total Soluble Fluoride (TSF) concentration was generally slightly less

than the TF concentration in the toothpastes analysed. The percentage of TSF concentration in the toothpastes was 97%, 61.1% and 96.5% in Aquafresh, Colgate and Omnident respectively. (Table-3)

Antimicrobial Efficacy Test:

Non- Fluoridated remineralising toothpastes did not show any antimicrobial efficacy. On the contrary, Fluoridated toothpastes showed good antimicrobial potential against *S. mutans* when 100µg of toothpaste was used but there was no effect on *L.*

acidophilus. It also showed dose-dependant antimicrobial activity i.e. AqF showed significantly higher zone of inhibition, followed by Omnident and then Colgate kids. (Table-3)

Toothpastes	Table 3. Inhibition zone of Bacteria in millimeter			
	<i>L. acidophilus</i>		<i>S.mutans</i>	
	50 µg	100 µg	50 µg	100 µg
Omni dent	-	-	-	16
Aqua fresh	-	-	17	28
Colgate kids	-	-	-	11
Enafix	-	-	-	-
Acclaim	-	-	-	-
Shy-NM	-	-	-	-

DISCUSSION

Dental caries remains a major health problem despite advances in caries prevention methods. The caries process is a continuum from the first molecular changes in the apatite crystals of the tooth, to a visible white spot lesion, to dentine involvement and eventual cavitation. These white spot lesions can be intact for 6 to 7 years, which either become arrested or revert to sound enamel in 75% of cases. In about 25% cases, it proceed to cavitation.[18] Scientific advances in cariology, dental products, and diagnostic systems have altered dentistry's approach to diagnosis and management of dental caries, over the last few decades[19].

"Prevention of Extension" has given way to the new paradigm of minimally invasive dentistry[20]. The minimally invasive approach to treat dental caries incorporates the dental science of detecting, diagnosing, intercepting and treating dental caries at microscopic level[21]. One of the four core principles of MID[22] is - regeneration i.e., to arrest and reverse incipient lesions, using appropriate topical agents including fluorides and biomimetic agents; Hence, shifting of the dynamic balance of the demineralization- remineralization cycle to net mineral gain within the lesion, through bioavailable mineralizing ions,[23] is the key to prevention of dental caries.

In the present in-vitro study, artificial caries was created using the demineralizing solution and then was subjected to

remineralization using Fluoridated (AquaFresh little teeth, Colgate kids, Omnident) and non-fluoridated remineralizing toothpastes (Shy-NM®, Enafix®, Aclaim®) to compare their remineralising potential over 6months. The remineralisation was tested using both Vickers hardness test and Scanning electron microscopy to assess the surface microhardness and changes on the enamel surface respectively.

The average hardness value of enamel is in the range of 316.0-328.4 VHN [24]. In the present study, the mean enamel surface microhardness of primary molars between the group at baseline range from 235.474 - 240.264 (VHN), which clearly shows no significant difference and the SEM image of demineralized samples(baseline) showed deep porosities giving a typical honeycomb appearance and/or single or groups of focal porosities without a protective layer. After remineralisation treatment, improvement in the mean surface hardness (VHN) of demineralized enamel, was observed in all the groups at various time periods tested. All the groups showed decrease in the porosities and smooth homogenous flat surface with slightly discernible prism shadows by the end of 6months (Figure - 11). SEM images of remineralized groups revealed that enamel surfaces were covered by a superficial layer of precipitated crystals to repair the erosive enamel, suggesting the remineralizing ability of all the tested dentifrices. The increase in enamel surface microhardness observed was highest with toothpastes containing remineralising agents when compared to fluoridated toothpastes at all time intervals (Table/Graph-1). On observation of these results, it can be suggested that the non-fluoridated remineralising toothpastes demonstrated significantly greater increase in the surface microhardness and displayed homogenous remineralisation when compared to Fluoridated toothpastes. This could be attributed to fluoride-mediated salivary remineralization which is restricted

to the outer 30µm of the enamel surface [25]. This surface only remineralization improves neither the aesthetics nor the structural properties of the subsurface lesion [26] and it prevents penetration of ions into the depth of the lesion (remineralises the carious lesion superficially only) [27]. Whereas, the newer biomimetic remineralizing agents have the capability to create apatite crystals within completely demineralized collagen fibers [28]. Grewal N et al also had similar findings where nano-HAP exhibited highest remineralization potential in terms of mineral gain followed by amine fluoride and sodium monofluorophosphate dentifrice.[29] Gangwar A et al, reported that bioactive glass (novamin) exhibited superior remineralization potential on artificially induced carious lesion in primary teeth [30].

Maintenance of good oral hygiene is the key to the prevention of dental diseases [31]. The activities of oral microflora are responsible for most oral diseases, including dental caries. So, any agent which can decrease the load of these bacteria in oral cavity, is capable of preventing caries. Therefore, inclusion of antimicrobial agent dentifrices has been stressed in order to limit these oral microorganisms [32]. In this study, in-vitro antimicrobial efficacy was done for six commercially available toothpastes for children. Three toothpastes tested in this study contained Fluoride as an active ingredient, which theoretically could be useful in controlling the oral microbial population and the other three toothpastes contained remineralising agents (NanoHAP, Novamin and Ca. orthophosphate). *Streptococcus mutans* and *Lactobacillus acidophilus* were the microorganisms chosen in this study since as *S.mutans* has been strongly associated with the initiation of caries, while there is a correlation between *Lactobacilli* is involved in the further development of carious lesions[33]. Data from the present study (Table-3) showed that toothpastes containing remineralising agents did not show any

antimicrobial potential. The lack of efficacy of remineralising toothpastes suggests that its usefulness is limited only to remineralisation of carious teeth, and not in reducing microbial load.

Contrary to our results, Mohamed et al. [34] reported novamin-containing toothpaste showed to be an effective antimicrobial agent in *S.mutans* count reduction. P. Stoor et al concluded that the bioactive glass paste appears to possess a broad antimicrobial effect on microorganisms of both supra- and sub- gingival plaque[35]. The antibacterial effect of Novamin is attributed to the increased pH in the aqueous(saliva) oral environment due to the release of alkaline ions and the constant release of sodium ions that increases the osmotic pressure and positive pressure on the bacterial cell membrane[34]. Our findings of negative antibacterial effect of novamin, however, could be related to the invitro conditions in our investigation, as the osmotic pressure shift could not occur due to the absence of aqueous media.

The three Fluoridated toothpastes showed good antimicrobial activity against one of the tested microorganisms - *S. mutans*. Among them, AquaFresh®TM little teeth and Omnident® showed better antimicrobial efficacy as well as remineralisation potential when compared to Colgate kids®. The overall better potential of Fluorides can be considered dose-dependent, as AquaFresh®TM little teeth toothpaste which contains 1397ppm of total soluble fluoride(TSF) concentration showed significantly higher zone of inhibition when compared to Omnident® (891ppm) followed by Colgate kids® (505ppm). Hence the antimicrobial and remineralising potential of fluorides is directly proportional to TSF concentration available in the toothpastes.

There can be discrepancies observed at local manufacturing units of global toothpastes brands. All the three fluoridated toothpastes analysed in this study had mentioned total fluoride levels on the tube or carton and their actual total fluoride content levels

differed from that of labelling. The TF concentration in Omnident® toothpaste was 1% higher than their stated level while other toothpastes (AquaFresh®TM little teeth and Colgate kids®) were lesser than the mentioned concentration. These differences existing between the declared and actual TF concentrations in toothpastes could lead to under- or over-exposure of fluoride, both being detrimental for the consumer. The soluble fluoride concentration in AquaFresh®TM and Omnident® toothpastes shows concentration closer to that of the claimed values. Whereas, Colgate kids® toothpaste contained only half the concentration (505ppm) mentioned on the toothpaste tube or carton (Table-3). Sebastian ST et al also reported that the measurement of total and free fluoride concentrations of toothpastes available in India showed inhomogenities[36]. Another research by Thakkar VP et al showed that the available fluoride content of one of the ten toothpastes in the study was substantially less than the total fluoride content[16] The probable reason for this is the incompatibility between the abrasive agent and the type of fluoride compound present in the composition of toothpaste, making the percentage of soluble fluoride less than TF concentration[37]. In general, NaF toothpastes have better compatibility with silica abrasives [38], whereas, SMFP toothpastes are more compatible with calcium containing abrasives[39], although calcium carbonate abrasive can still inactivate fluoride in a MFP toothpaste[36]. The mean TSF concentration was slightly greater in NaF/Si based toothpastes than the others as observed previously by various studies [40,41]. Supporting this finding, AquaFresh®TM little teeth toothpaste in our study, contains a similar composition (NaF/Si) and showed good results. Whereas, Colgate kids® toothpaste contains silica as an abrasive which is considered not compatible with the form of fluoride source it contains (SMFP), which could be the reason for decrease in the percentage of TSF in our findings. Hence, the TSF

concentration remains inadequate to achieve optimal remineralisation and antimicrobial efficacy.

CONCLUSION

From the observations of the present study, it can be inferred that among the tested toothpastes, remineralising agents is the most effective means of enhancing remineralisation when compared to fluoridated toothpastes. However, fluoridated toothpastes possessed antimicrobial efficacy in addition to remineralisation, which helps to control plaque and cariogenic microflora. Since our study was carried out in an invitro environment, further in vivo studies are necessary to confirm our findings & discern the true clinical efficacy of these agents as saliva, plaque and many other confounding factors which may affect the efficacy of the studied agents. Further research could be directed towards alternative use of both fluoridated and non- fluoridated remineralising toothpastes, simultaneously, to examine both remineralization and antimicrobial efficacy.

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