

Evaluate the tubule occluding ability of four desensitizing agents amine fluoride, calcium sodium phospho silicate, pro-arginin and CPP-ACP (casein phosphopeptide-amorphous calcium phosphate) using scanning electron microscope.

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Abstract

Background: Dentin hypersensitivity is characterized by short, sharp pain arising from exposed dentin in response to stimuli, typically thermal, evaporative, tactile, osmotic, or chemical, and which cannot be ascribed to any other form of dental defect or disease. Traditionally, the term dentine hypersensitivity was used to describe this distinct clinical condition; however,

several authors have also used the terms cervical dentine sensitivity (CDS) or cervical dentine hypersensitivity (CDH) or dentine sensitivity (DS), and root dentine sensitivity (RDS)/root dentine hypersensitivity (RDH). Several studies showed that casein phosphopeptide- amorphous calcium phosphate (cpp-acp) and arginine are effective in occluding dentinal tubules but information regarding dentinal tubule

occluding ability of Novamin (Calcium sodium phospho silicate) and Amine fluoride is less. Therefore the present study aim to evaluate the tubule occluding ability of four desensitizing agents.

Material and Method: An in vitro study comparing Occluding ability of Amine fluoride, Calcium sodium phospho silicate, Pro-arginin & CPP-ACP (Casein Phosphopeptide - Amorphous Calcium Phosphate) using Scanning Electron Microscope.

Result: Among the treated groups the specimens brushed with CPP-ACP (Casein Phosphopeptide - Amorphous Calcium Phosphate) containing agent Recaldent GC tooth mousse dentifrice showed the highest percentage of tubule occlusion, as it precipitated the hydroxycarbonate apatite over the entire dentin surface followed by Amine fluoride containing agent (Amflor), Pro-Argin containing agent(Colgate sensitive) Oral-B sensitive, Calcium sodium phospho silicate containing agent (Shy-NM).

Keyword: Tubule Occluding Ability, Dentine Hypersensitivity, Desensitizing Agents.

Introduction

Dentin hypersensitivity is characterized by short, sharp pain arising from exposed dentin in response to stimuli, typically thermal, evaporative, tactile, osmotic, or chemical, and which cannot be ascribed to any other form of dental defect or disease^(1,2) Traditionally, the term dentine hypersensitivity was used to describe this distinct clinical condition; however, several authors have also used the terms cervical dentine sensitivity (CDS) or cervical dentine hypersensitivity (CDH) or dentine sensitivity (DS), and root dentine sensitivity (RDS)/root dentine hypersensitivity (RDH) ^(1, 3-8).

Dentin hypersensitivity is a common oral condition that affects as many as 4-69% ^(1,2) of patients. Females appear to suffer more than males presumably due to their

overall health care and better oral hygiene awareness⁸. It has been described as the “common cold of dentistry”⁹ by some and “toothbrush disease” by others, when it occurs in the presence of gingival recession.¹⁰

Gysi attempted to explain “the sensitiveness of dentin,” and described the phenomenon of fluid movement in dentin tubules.¹¹ More than sixty years later, Brännström proposed the “hydrodynamic theory” as a mechanism to explain the transmission of pain-producing stimuli of the dentin.^{12,13} Other theories have been proposed as potential mechanisms by which pain transmission can occur, but these have largely been discounted.^{1,14}

The hydrodynamic theory suggests that dentin hypersensitivity is a result of inward or outward movement of fluid within the dentin tubules. This results in a pressure change across the dentin which activates intra-dental nerve fibers, via a mechanoreceptor response, to cause pain.¹⁵

Most pain-producing stimulus requires that dentin tubules are open at the dentin surface and patent to the pulp. Several studies of exfoliated teeth, with clinically characterized “sensitive” and “non-sensitive” areas of exposed dentin, have shown that tubules are greater in number (eight times), larger in diameter (two times), and are open in “sensitive” teeth, whereas tubules are fewer in number, smaller in diameter, and are usually blocked in their “non-sensitive” counterparts.^{16,17}

As the rate of fluid flow through dentin tubules is proportional to the fourth power of the tubule radius, it is highly likely that the difference in tubule diameter between “sensitive” and “non-sensitive” teeth is of clinical relevance to the treatment of dentin hypersensitivity.¹

Today, dozens of in-office sensitivity treatments and mass market sensitivity relief toothpastes are available worldwide.¹⁵ Various chemical have been used for the

occlusion of open dentinal tubules like strontium chloride, silver nitrate, sodium fluoride, potassium oxalate, stannous fluoride, calcium hydroxide, ferrous oxide etc.¹⁸

Recently calcium sodium phosphosilicate, casein phosphopeptide- amorphous calcium phosphate (cpp-acp) arginine and amine fluoride are available as an desensitizing agent.

Several studies showed that casein phosphopeptide-amorphous calcium phosphate (cpp-acp) and arginine are effective in occluding dentinal tubules^{15,19}. But information regarding dentinal tubule occluding ability of Novamin (Calcium sodium phospho silicate) and Amine fluoride is less.

Hence the aim of the study is to evaluate the tubule occluding ability of four desensitizing agents Amine fluoride, Calcium sodium phospho silicate, Pro-arginin & CPP-ACP (Casein Phosphopeptide - Amorphous Calcium Phosphate) using Scanning Electron Microscope.

Aims and objectives

The objective of the present study was to Evaluate the tubule occluding ability of four desensitizing agents Amine fluoride, Calcium sodium phospho silicate, Pro-arginin & CPP-ACP (Casein Phosphopeptide - Amorphous Calcium Phosphate) using Scanning Electron Microscope.

Methodology:

Specimens were obtained from 60 sound premolars extracted for orthodontic purpose from the patients of age group 15 to 25 years. All the teeth were stored in 10% neutral buffered formaline NBF (freshly prepared) at room temp for no longer than one month prior to the use.

Inclusion Criteria

Intact human permanent premolar teeth (maxillary or mandibular), Freshly extracted teeth, Non carious teeth and Teeth with no sign of fracture teeth.

Exclusion Criteria

Carious teeth, Grossly destructed teeth, Teeth with any anomalies and Fractured teeth.

Dentine Specimen Preparation

Dentine specimens of size 3x3x2mm were prepared from the cervical 1/3rd each premolar using double-sided diamond disk attached to water-cooled air motor (NSK TH88625) and straight handpiece (NSK. ONX90715). The specimens were randomly divided into 6 groups of 10 dentin blocks. These dentin blocks were mounted on 2mm thick plastic plate by using Cyanoacrylate adhesive (super glue). Each dentine block was polished with of lex polishing discs of coarse, medium, fine, extra fine disks and final polishing was done with prismagloss fine and extra fine polishing pastes containing 1 and 0.2 micron alumina abrasives. All the specimens were ultrasonicated (at 42000 Hz, 5 cycles) for 12 min in distilled water to remove residual smear layer and to open the dentine tubule to simulate hypersensitive cervical dentin.

Experimental groups and treatments

Specimens were randomly divided into six groups each with ten dentine specimens

Group-1 control: Specimens were immersed in artificial saliva for 7 days. The artificial saliva contained (mmoles/L): distilled water 700ml, caoH2 1.56mM, Kcl 150.00mM, Hcl 36.00mM, H3po4 0.088mM, buffer 99.7mM, pH 7.2.

Group-2 control: Specimens were brushed with distilled water, for 2 min twice per day for 7 days.

Group-3: Specimens were brushed with CPP-ACP (Casein Phosphopeptide - Amorphous Calcium

Phosphate) containing agent (Recaldent GC tooth mousse) for 2 min twice per day for 7 days.

Group-4: specimens were brushed with Pro-Argin containing agent (Colgate sensitive) for 2 min twice per day for 7 days.

Group-5: specimens were brushed with Amine fluoride containing agent (Amflor) for 2 min twice per day for 7 days.

Group-6: specimens were brushed with Calcium sodium phospho silicate containing agent (Shy-NM) for 2 min twice per day for 7 days.

Specimens of groups 3, 4, 5, and 6 were brushed with dentifrices slurries prepared by diluting 2gm of dentifrice with 6 ml of distilled water. Brushing was performed with Oral B powered brush which is a battery powered toothbrush with soft round bristle at an oscillation speed of 8000/min. Brushing was done for 2min for each dentine block from equidistance by attaching bristle protector to the toothbrush shaft. After each brushing session specimens were washed under running tap water and then kept in artificial saliva filled plastic container.

SEM analysis

After the last brushing session, dentine specimens were washed with distilled water, after drying thin layer of gold sputter coating was done (QUORUM Q 150R ES, UK) Photomicrographs were taken using Scanning Electron Microscope (ZEISS GEMINI SUPRA 55, Germany) from the centre of 3x3x2mm dentine block at 1000X magnification.

Statistical analysis

Percentage of occluded tubules was obtained by dividing the total number of occluded tubules by total number of tubules in each photomicrography. This result was then multiplied by 100 to obtain the percentage of occluded tubules for each photomicrography. Results were

statistically analyzed by one-way Analysis of Variance (ANOVA) and Tukey test.

Results:

The analysis of the study were evaluated and tabulated as follows

SEM photomicrograph

Fig-1, shows the SEM photomicrograph (at 1000x) of Group-1 dentin specimen where specimens were kept in artificial saliva for 7 days without brushing.

Fig-2, shows the SEM photomicrograph (at 1000x) where dentin specimens were brushed with distilled water.

Fig-3, shows the SEM photomicrograph (at 1000x) where dentin specimens were brushed with CPP-ACP (Casein Phosphopeptide - Amorphous Calcium Phosphate) containing agent (Recaldent GC tooth mousse) for 2 min twice per day for 7 days.

Fig-4, shows the SEM photomicrograph (at 1000x) where dentin specimens were brushed with Pro-Argin containing agent (Colgate sensitive) for 2 min twice per day for 7 days.

Fig-5, shows the SEM photomicrograph (at 1000x) where dentin specimens were brushed with Amine fluoride containing agent (Amflor) for 2 min twice per day for 7 days.

Fig-6, shows the SEM photomicrograph (at 1000x) where dentin specimen were brushed with Calcium sodium phospho silicate containing agent (Shy-NM) for 2 min twice per day for 7 days.

Tables

Table 1: Table depicting the percentage of dentinal tubule occlusion of each dentin specimen in each group.

Table 2: Mean, Standard Deviation, minimum and maximum numbers of occluded tubules per 100 dentinal tubules on the dentin surface in each experimental group.

Table 3: Comparison of occluded tubules per 100 dentinal tubules on the dentin surface between experimental group using one way ANOVA.

Table 4: Comparison of Artificial saliva and Distilled water for percentage of occluded tubules using Tukey Post Hoc test.

Table 5 :Comparison of Artificial saliva and Test groups for percentage of occluded tubules using tukey post hoc test.

Table 6: Comparison of Distilled water and Test groups for percentage of occluded tubules using tukey post hoc test.

Table 6: Comparison of Test groups for percentage of occluded tubules using tukey post hoc test

Graphs

Graph 1: Mean numbers of occluded tubules per 100 dentinal tubules on the dentin surface in each experimental group.

Graph 2: Mean numbers of occluded tubules per 100 dentinal tubules on the dentin surface in Artificial saliva and Distilled water.

Statistical analysis

Data collected by experiments were computerized and analyzed using the Statistical Package for Social Sciences (SPSS version 16.0).

Since the data were continuous type, parametric tests were used for analysis. Mean, Standard Deviation, minimum and maximum numbers of occluded tubules per 100 dentinal tubules on the dentin surface in each experimental group were calculated. One way Analysis of Variance (ANOVA) test was used for multiple group comparisons followed by Tukey post hoc for group wise comparisons. p-value <0.05 was considered statistically significant. The null hypothesis (H0) stated for present study was as follows:

There is no significant difference between the groups in percentage occlusion of dentinal tubules on the dentin surface.

The alternate hypothesis (H1) stated was: The experimental groups differs significantly in percentage occlusion of dentinal tubules on the dentin surface.

Fig. 1: shows the SEM photomicrograph (at 1000x) of Group-1 dentin specimen where specimens were kept in artificial saliva for 7 days without brushing.

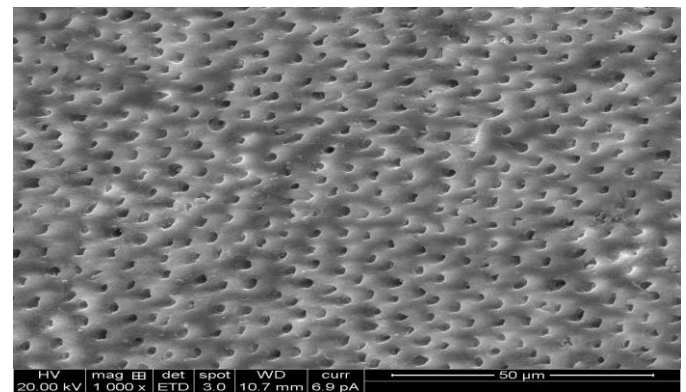


Fig. 2: Shows the SEM photomicrograph (at 1000x) where dentin specimens were brushed with distilled water.

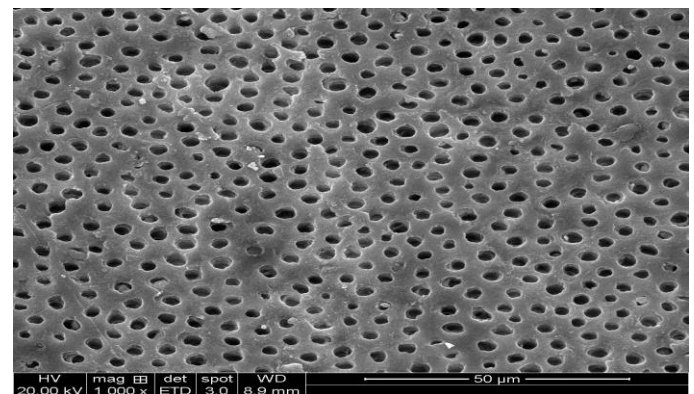


Fig.3: Shows the SEM photomicrograph (at 1000x) where dentin specimens were brushed with with CPP-ACP (Casein Phosphopeptide - Amorphous Calcium Phosphate) containing agent (Recaldent GC tooth mousse) for 2 min twice per day for 7 days.

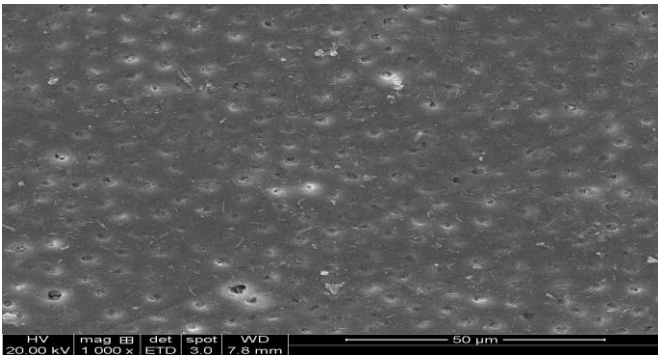


Fig. 4: Shows the SEM photomicrograph (at 1000x) where dentin specimens were brushed with Pro-Argin containing agent (Colgate sensitive) for 2 min twice per day for 7 days.

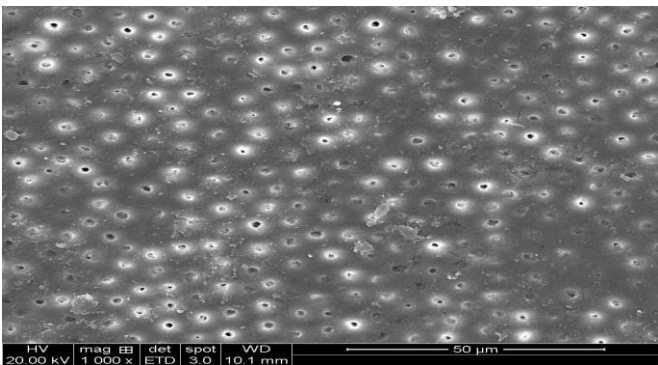


Fig. 5: Shows the SEM photomicrograph (at 1000x) where dentin specimens were brushed with Amine fluoride containing agent (Amflor) for 2 min twice per day for 7 days.

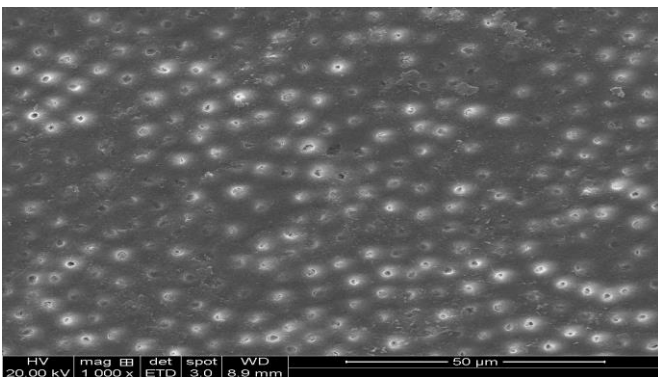


Fig. 6: Shows the SEM photomicrograph (at 1000x) where dentin specimen were brushed with Calcium sodium phospho silicate containing agent (Shy-NM) for 2 min twice per day for 7 days.

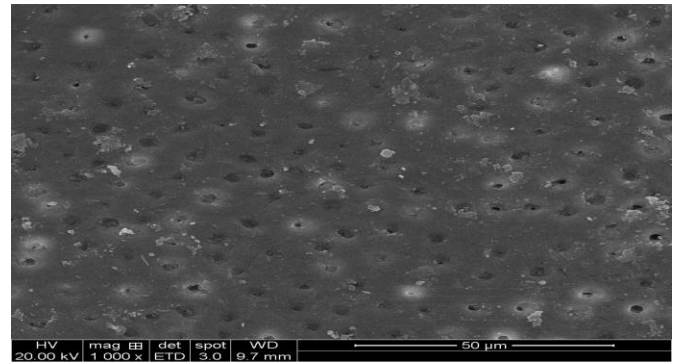


Table 1: Table depicting the percentage of dentinal tubule occlusion of each dentin specimen in each group.

Sn.	Group 1 Control	Group 2 Control	Group 3	Group 4	Group 5	Group 6
1	11	4.46	86.2	41.16	71.12	24.01
2	10.75	5.61	90.12	46.81	68.12	28.11
3	12.23	4.98	91.86	41.8	69.11	31.76
4	11.18	7.12	89.21	39.01	70.26	24.17
5	14.1	4.19	92.2	46.45	88.12	29.01
6	10.21	4.86	96.12	48.16	86.1	20.04
7	11.21	5.87	91.82	44.18	61.97	44.18
8	18.96	9.12	94.81	40.24	71.28	26.48
9	10.54	6.18	91.26	41.25	77.82	21.16
10	12.08	4.02	93.62	32.12	71.12	32.33

Table 2: Mean, Standard Deviation, minimum and Maximum numbers of occluded tubules per 100 dentinal tubules on the dentin surface in each experimental group.

Experimental Groups	N	Mean	SD	Minimum	Maximum
Artificial saliva	10	12.23	2.62	10.21	18.96
Distilled water	10	5.64	1.56	4.02	9.12
CPP-ACP containing agent	10	91.72	2.83	86.20	96.12
Pro-Argin containing agent	10	42.12	4.67	32.12	48.16
Amine fluoride containing agent	10	73.50	8.16	61.97	88.12
Calcium sodium phospho silicate containing agent	10	28.12	6.98	20.04	44.18

Table 3: Comparison of occluded tubules per 100 dentinal tubules on the dentin surface between experimental group using one way ANOVA.

Sum of Squares	df	Mean Square	F value	P value
58653.542	5	11730.708	455.770	0.000 (<0.001) Significant Difference

Table 4: Comparison of Artificial saliva and Distilled water for percentage of occluded tubules using Tukey Post Hoc test.

Experimental Groups	N	Mean	SD	Minimum	Maximum
Artificial saliva	10	12.23	2.62	10.21	18.96
Distilled water	10	5.64	1.56	4.02	9.12
Mean Difference	6.58				
P value	0.057 (>0.05), Not Significant				

Table 5: Comparison of Artificial saliva and test groups for percentage of occluded tubules using Tukey Post Hoc test.

	N	Mean difference	P value	Inference
Group 1- group 3	20	-79.50	0.000	Significant
Group 1- group 4	20	-2989	0.000	Significant
Group 1- group 5	20	-61.28	0.000	Significant
Group 1- group 6	20	-15.90	0.000	Significant

Table 6: Comparison of Distilled water and test groups for percentage of occluded tubules using Tukey Post Hoc test.

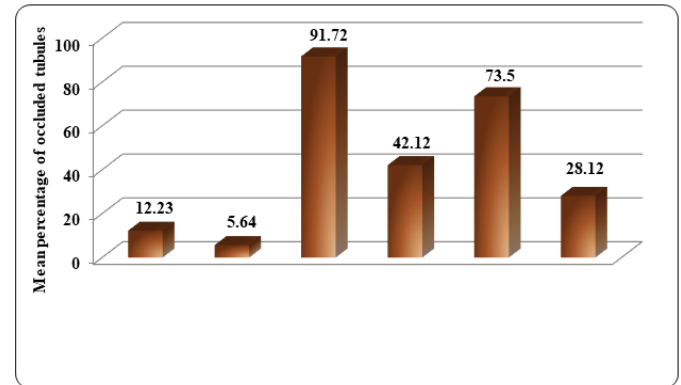
	N	Mean difference	P value	Inference
Group2- group 3	20	-67.86	0.000	significant
Group2- group 4	20	-22.48	0.000	significant
Group2- group 5	20	49.6	0.000	significant
Group2- group 6	20	18.22	0.000	significant

Table 7: Comparison of test groups for percentage of occluded tubules using Tukey Post hoc test.

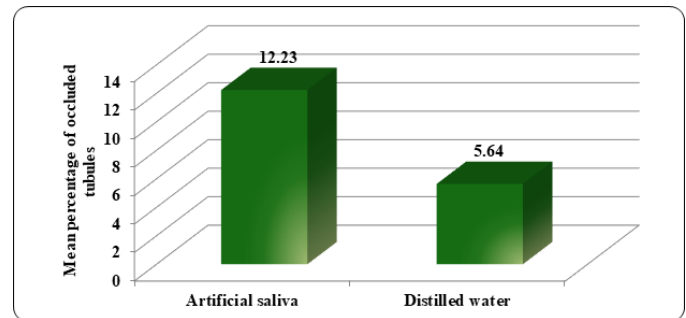
	N	Mean difference	P value	Inference
Group3- group 4	20	49.6	0.000	significant
Group3- group 5	20	18.22	0.000	significant
Group3- group 6	20	63.60	0.000	significant
Group4- group 5	20	-31.38	0.000	significant

Group4- group 6	20	14.00	0.000	significant
Group5- group 6	20	45.38	0.000	significant

Graph 1: Mean numbers of occluded tubules per 100 dentinal tubules on the dentin surface in each experimental group.



Graph 2: Mean numbers of occluded tubules per 100 dentinal tubules on the dentin surface in Artificial saliva and Distilled water.



Discussion

The oral cavity presents an assorted variety of diseases, among which dentinal hypersensitivity are one of the most common. Dentinal hypersensitivity has been recorded for over two millennia, and the earliest documented treatment method was opium therapy, which dates back to 400 BC, being advocated as recently as 1000 AD.²⁰

Studies of the prevalence of dentin hypersensitivity have reported widely differing levels, ranging from 4–57% in individuals within general dental practice settings.^{1, 21, 22, 23} These wide variations have been attributed to a number of factors, including the method of assessment or diagnosis, the population base and setting, and

behavioral factors, such as oral hygiene habits and intake of acidic foods and drinks. There is a slightly higher incidence of dentin hypersensitivity in females than in males, which may reflect oral hygiene and dietary practices.¹

Dentin hypersensitivity is typically experienced when the root of the tooth has been exposed to the oral environment as a result of gingival recession. Gingival recession may occur naturally, compounded by poor oral hygiene habits, especially overzealous tooth brushing, or it may result from surgical or non-surgical periodontal treatment. Dentin hypersensitivity occurs more frequently in the cervical area of the roots, where the cementum is very thin. Periodontal procedures such as scaling and root planing, may entirely remove this thin cementum layer and induce root hypersensitivity. As people live longer, healthier lives and maintain their dentitions, there will be an increased demand on dental professionals to manage the sensitivity of cervical exposed dentin, as well as any secondary issues that may arise from the discomfort associated with dentin hypersensitivity. In particular, dentin hypersensitivity may render tooth brushing more difficult in some individuals, with the result that persistent and continued accumulation of dental plaque may increase the incidence of caries, gingivitis, and more serious periodontal problems.²⁴

Several theories have been proposed to explain the mechanism of dentin hypersensitivity, including the odontoblast transducer theory, the dentin receptor theory, and the hydrodynamic theory.^{25,26} Scientific evidence supports the hydrodynamic theory and, for this reason, it is generally favored by the dental community. The hydrodynamic theory¹ (modified by Brännström⁶ in 1963) assumes that fluid movement within the dentin tubules is the basis for the transmission of painful

sensations. Specifically, it proposes that non-noxious stimuli at the tooth surface cause fluid movement within the dentin tubules, affecting the pulpal mechanoreceptors and resulting in the sensation of pain. In 1994, Nöhri, et al.²⁷ provided an addendum to the hydrodynamic theory, suggesting that the perception and sensation of pain was directly related to the stimulation of the nerves within the pulp via electrical current.

Regardless of the Etiology, the problem needs to be addressed in order to provide patients with improved oral comfort and quality of life. To this end, a number of agents have been proposed to help control dentin hypersensitivity and relieve discomfort; some can be used by the patient at home, others must be applied in the dental office by a dental professional. One approach by which this can be achieved is to reduce the diameter of open dentin tubules in order to limit the displacement of fluids within them (decreased hydrodynamic flow), thereby blocking neurotransmission and decreasing the response to painful stimuli. According to Trowbridge and Silver,²⁸ this could be achieved by forming a smear layer on the exposed dentin, using topical agents that form insoluble precipitates within the tubules, or by blocking the entrance to tubules with plastic resins. This approach has been most extensively applied in professionally administered products.

In 1982, dentin hypersensitivity was described as an enigma, because it was frequently encountered and poorly understood. The past twenty-five years have witnessed an evolution in the scientific understanding of this condition. Largely based on a suggestion in 1983²⁶, the term “dentin hypersensitivity” was formally defined in 1997 in the guidelines for clinical trials.³⁰ This definition was officially accepted in 2003, with one minor change, by the Canadian Advisory Board on Dentin Hypersensitivity in their consensus-based

recommendations for the diagnosis and management of dentin hypersensitivity.² Clearly, the condition is no longer the enigma that was once described. Nonetheless, there is a need for continued basic and clinical research that will lead to improved prevention and management of dentin hypersensitivity.¹

Today, dozens of in-office sensitivity treatments and mass market sensitivity relief toothpastes are available worldwide. Clinical studies demonstrate the effectiveness of some in-office products, e.g., 5% sodium fluoride varnish;³¹ nonetheless, there is a paucity of data on the majority of these products.^{1,21} There appears to be a significant body of clinical data to support the efficacy of potassium-based dentifrices, and these products have been acknowledged to provide relief to their users.⁵⁹ However, some authors have concluded that the data are equivocal.^{29,33} Despite the reported prevalence of dentin hypersensitivity, it is noteworthy that a relatively small percentage of sufferers seek professional treatment to alleviate their condition and/or use an everyday sensitivity relief toothpaste.²

Hence the aim of the study is to evaluate the tubule occluding ability of four desensitizing agents Amine fluoride, Calcium sodium phospho silicate, Pro-arginin & CPP-ACP (Casein Phosphopeptide - Amorphous Calcium Phosphate) using Scanning Electron Microscope.

To determine the dentinal tubule occluding ability of each of dentifrice dentine specimen of size 3x3x2mm were prepared from the cervical 1/3rd each premolar using double-sided diamond disk attached to water-cooled air motor (NSK TH88625) and straight handpiece (NSK. ONX90715). The specimens were randomly divided into 6 groups of 10 dentin blocks. These dentin blocks were mounted on 2mm thick plastic plate by using Cyanoacrylate adhesive (super glue). Each

dentine block was polished with of lex polishing discs of coarse, medium, fine, extra fine disks and final polishing was done with prismagloss fine and extra fine polishing pastes containing 1 and 0.2 micron alumina abrasives. All the specimens were ultrasonicated (at 42000 Hz, 5 cycles) for 12 min in distilled water to remove residual smear layer and to open the dentine tubule to simulate hypersensitive cervical dentin.

Specimens of groups 3, 4, 5, and 6 were brushed with dentifrices slurries prepared by diluting 2gm of dentifrice with 6 ml of distilled water. Brushing was performed with Oral B powered brush which is a battery powered toothbrush with soft round bristle at an oscillation speed of 8000/min to as to maintain the constant speed. Brushing was done for 2min for each dentine block from equidistance by attaching bristle protector to the toothbrush shaft so as to apply equal pressure on all specimens. After each brushing session specimens were washed under running tap water and then kept in artificial saliva filled plastic container to create the condition like of oral environment.

After the last brushing session, dentine specimens were washed with distilled water, after drying thin layer of gold sputter coating was done Photomicrographs were taken using Scanning Electron Microscope from the center of 3x3x2mm dentine block at 1000X magnification.

Percentage of occluded tubules was obtained. Results were statistically analyzed by one-way Analysis of Variance (ANOVA) and Tukey test.

The specimens brushed with dentifrices exhibited higher percentage of tubule occlusion than G-1 and G-2 control groups. The specimens immersed in artificial saliva (G-1) without brushing and the specimens brushed with distilled water without dentifrice (G-2) were not significantly different (Fig-11 & 12). These specimens

showed the mean tubule occlusion percentage of 12.23 % and 5.64 %. Saliva plays a role in naturally reducing dentin hypersensitivity; first, by transporting calcium and phosphate into dentin tubules to induce tubule plugging; and second, by forming a surface protective layer of salivary glycoprotein with calcium and phosphate.³² The tubule occlusion in these groups can be due to the calcium phosphate precipitate on the dentin surface since artificial saliva is supersaturated with respect to the dentin hydroxyapatite. The results obtained are similar to the results of the study in which dentin specimens immersed in artificial saliva initially there will be decrease in the concentration of calcium and phosphate ions then the increase in their concentration indicates the precipitation followed by the partial dissolution of hydroxyapatite leading to the subsequent tubule occlusion, and the lower percentage in the G-2 specimens may be due to the smear layer formed by the brushing process.^{28,34,35} Despite a reduction in the tubule lumen and complete occlusion of some tubules (Figs. 11 and 12), specimens from control groups still had most of the tubular orifices opened.

Other treatment groups G-3, G-4, G-5 & G-6 showed the significantly greater percentage of tubule occlusion when compared to the control groups G-1 and G-2.

In Group-3 the samples were brushed with CPP-ACP containing agent (Fig-13). The mean percentage of tubule occlusion seen in these groups is 91.72%. Tukey post hoc test for pairwise comparison of occluded tubules per 100 dentinal tubules on the dentin surface showed Significant Differences in which group 3 > 1, 2, 4, 5, 6.

Reynolds EC showed that the casein phosphopeptide (CPP) contains phosphoserine sequences which get attached and stabilized with amorphous calcium phosphate (ACP). This stabilized CPP-ACP prevents the

dissolution of calcium and phosphate ions and maintains a supersaturated solution of bioavailable calcium and phosphates.³⁶

The peptides of CPP-ACP bind to the dentine surface, then promote the deposition of mineral deposits within dentine tubules. This process has been shown to significantly decrease dentine permeability by creating precipitates on the dentine surface that reduce the diameter of dentinal tubules.³⁶⁻³⁹

Lauren J walsh⁴⁰ and Shalini gugnani¹⁵ in their controlled study also showed decrease dentinal sensitivity however in their study they had not mentioned about their apparent mechanism of action.

In Group-4 the samples were brushed with Pro-Argin containing agent (Fig-13). The mean percentage of tubule occlusion seen in these groups is 42.12%. Tukey post hoc test for pairwise comparison of occluded tubules per 100 dentinal tubules on the dentin surface showed Significant Differences in which group 4 > 1, 2, 6.

Diane Cummins in his study confirmed with high resolution Scanning Electron Microscopy (SEM) images that the desensitizing prophylaxis paste provides complete occlusion of open dentin tubules, and freeze fracture images have shown that the plug reaches a depth of two microns into the tubule.¹⁵

In addition, in vitro study by Kleinberg I. on the mechanism of action of arginine have demonstrated tubule occlusion.³²

In Group-5 the samples were brushed with Amine fluoride containing agent (Fig-14). The mean percentage of tubule occlusion seen in these groups is 73.50%. Tukey post hoc test for pairwise comparison of occluded tubules per 100 dentinal tubules on the dentin surface showed Significant Differences in which group 5 > 1, 2, 4, 6.

The principal mechanism of fluoride to relieve DHS is its chemical ability to reduce and block fluid movements in the dentin tubules through formation of calcium-phosphorous precipitates as well as calcium fluoride (CaF₂) and fluorapatite (FAP).⁴¹⁻⁴³

T. BACHANEK in his study concludes that Amine fluoride dental rinse and dentifrice effectively occluded dentin tubules (partially or totally) and therefore may reduce patients hypersensitivity symptoms in-vivo.⁴⁴

In Group-6 the samples were brushed with Calcium sodium phospho silicate containing agent (Fig-14). The mean percentage of tubule occlusion seen in these groups is 28.12%. Tukey post hoc test for pairwise comparison of occluded tubules per 100 dentinal tubules on the dentin surface showed Significant Differences in which group 6 > 1, 2.

In the studies done by **Ananthakrishna.S et al (2012)**⁴⁵ and **Tirapelli C et al (2010)**⁴⁶ showed that calcium sodium phospho silicate is an effective agent in dentinal tubule occlusion however results showed that the percentage of tubule occlusion is less than the other treatment groups.

Conclusion

The following conclusions were drawn from the study:

- 1) All the treatment groups exhibited significantly higher percentage of tubule occlusion compared to control groups.
- 2) Among the treated groups the specimens brushed with CPP-ACP (Casein Phosphopeptide - Amorphous Calcium Phosphate) containing agent Recaldent GC tooth mousse dentifrice showed the highest percentage of tubule occlusion, as it precipitated the hydroxycarbonate apatite over the entire dentin surface followed by Amine fluoride containing agent (Amflor), Pro-Argin containing agent (Colgate sensitive) Oral-B

sensitive, Calcium sodium phospho silicate containing agent (Shy-NM) .

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