Evaluation of dentinal tubules occlusion using Diode laser, Remin Pro, CPP-ACP (GC Tooth mousse), Fluoridated-Bioactive Glass (Elsenz) and Non-Fluoridated Bioactive Glass (Shy-Nm) as desensitizing methods. An in-vitro Scanning electron microscopic study.

Dr. Srikumar G.P.V. [a], Dr. Nivedita Jaipuriar [b], Dr. Shripriya Rahane [c], Dr. Anjali Gurnani [d], Dr. Megha Ghosh [e], Dr. Vaishnavi Gadbail [f]

[a] M.D.S. Professor and H.O.D. Department of Conservative Dentistry and Endodontics. Triveni Institute of Dental Sciences, Hospital and Research Centre. Bilaspur, Chhattisgarh, India.

[b] PG Student. Department of Conservative Dentistry and Endodontics. Triveni Institute of Dental Sciences, Hospital and Research Centre. Bilaspur, Chhattisgarh, India.

[c] PG Student. Department of Conservative Dentistry and Endodontics. Triveni Institute of Dental Sciences, Hospital and Research Centre. Bilaspur, Chhattisgarh, India.

[d] PG Student. Department of Conservative Dentistry and Endodontics. Triveni Institute of Dental Sciences, Hospital and Research Centre. Bilaspur, Chhattisgarh, India.

[e] PG Student. Department of Conservative Dentistry and Endodontics. Triveni Institute of Dental Sciences, Hospital and Research Centre. Bilaspur, Chhattisgarh, India.

[f] PG Student. Department of Conservative Dentistry and Endodontics. Triveni Institute of Dental Sciences, Hospital and Research Centre. Bilaspur, Chhattisgarh, India.

Corresponding author:
Dr. Srikumar G.P.V.
M.D.S. Professor and H.O.D. Department of Conservative Dentistry and Endodontics. Triveni Institute of Dental Sciences, Hospital and Research Centre. Bodri- 495220. Bilaspur, Chhattisgarh, India.

Abstract- 120 extracted human permanent maxillary first and second premolar teeth were included. All the teeth were then sectioned mesio-distally from the middle-third of crown using a high speed diamond disc to obtain dentin discs with 2mm thickness and any remaining enamel, surface irregularities were eliminated using 1200 fine grit silicon carbide polishing paper to create dentin discs with smooth, uniform surfaces. By using a double-sided tape, each dentin disc of all teeth were fixed on microscopic slides, with polished sides of dentin discs facing up. All the specimens were then randomly divided into 06 groups with 20 specimens per group. Group A; Low output Diode laser; 1W power, Group B; Remin Pro, Group C; CPP-ACP (GC Tooth mousse), Group D; Non- Fluoridated-Bioactive Glass (Shy-NM) and Group E; Fluoridated-Bioactive Glass (Elsenz) were used as desensitizing agents. A pea sized amount of desensitizing agent/paste as per the respective groups was placed on each of the dentin disc (Group F; Control group: 4ml Distilled water was used as desensitizing agent per specimen). By using Oral B Extra soft bristle toothbrush, each dentin disc was brushed in circular motion with light pressure twice a day at an interval of 12 hours for 2 minutes each for 14 days. The specimens were then gently rinsed with deionised water to ensure complete removal of any residual desensitizing agents. The extent of dentinal tubules occlusion in all the specimens were then evaluated using Scanning electron microscope at a magnification of X2000 and a categorical scale of (0-4) was accorded. The data obtained was statistically analysed using Chi-Square test. Low output Diode laser as desensitizing method showed the maximum effectiveness in the occlusion of dentinal tubules followed by Remin Pro and Fluoridated-Bioactive Glass (Elsenz) as desensitizing agent/pastes. Non-Fluoridated-Bioactive Glass (Shy-Nm) was found to be the least effective.
Key words: Desensitizing agent; Dentinal tubules occlusion; Dentinal hypersensitivity; Diode laser; Elsenz; Shy-Nm.

DOI Number: 10.14704/nq.2022.20.11.NQ66184 **NeuroQuantology 2022; 20(11):1891-1906**

Introduction:

Dentinal or Dentin Hypersensitivity (DH) is defined as pain derived from exposed dentin in response to chemical, thermal, tactile or osmotic stimuli which cannot be explained as arising from any other dental defect or disease.[1] The terms DS (Dentin Sensitivity) or DH were used inter-changeably to describe the same clinical condition.[2] It is one of the most commonly encountered clinical problem described as an exaggerated response to application of a stimulus to exposed dentin, regardless of its location.[2] Dentinal hypersensitivity is characterized by short, sharp pain arising from exposed dentine in response to stimuli, typically thermal, evaporative, tactile, osmotic or chemical and which cannot be ascribed to any other dental defect or pathology.[2] Incidence of dentinal hypersensitivity ranges from 4 to 74%.2 A slightly higher incidence of dentinal hypersensitivity is reported in females than in males. While dentinal hypersensitivity can affect individuals of any age, but the most affected patients are in the age group of 20–50 years, with a peak between 30 to 40 years of age. Canines and Premolars of both the arches are the most commonly affected teeth, while the buccal aspect of cervical areas are the most commonly affected sites.[2]

Dentinal hypersensitivity develops in two phases: Lesion localization and Lesion initiation. Lesion localization occurs by loss of protective covering over the dentin, thereby exposing it to external environment. It includes loss of enamel by attrition, abrasion, erosion or abfraction. Another cause for lesion localization is gingival recession which can be due to tooth brush abrasion, pocket reduction surgery, tooth preparation for crown, excessive flossing or secondary to periodontal diseases. Not all exposed dentin is sensitive and for dentin to become sensitive, Lesion initiation has to happen and it occurs only after the protective covering of smear layer is removed, leading to exposure and opening of dentinal tubules.[3] There are two basic approaches for the treatment of dentinal hypersensitivity. The first is to treat the tooth with a nerve depolarizing agent which penetrates into the dentin tubules and depolarizes the nerve synapse, thereby reducing sensitivity by preventing the conduction of pain impulses. The second approach is to treat the tooth with chemical occluding agents or pastes/dentrifices, which prevents the flow of dentinal fluid and thereby reducing tooth sensitivity.[4]

In our study, we evaluated the dentinal tubules occlusion using Diode laser, Remin Pro, CPP-ACP(Casein Phospho Peptide- Amorphous Calcium Phosphate), Fluoridated-Bioactive Glass (Elsenz) and Non-Fluoridated Bioactive Glass (Shy-Nm) as desensitizing methods byScanning Electron Microscope (SEM) analysis.

LASER (Light Amplification by Stimulated Emission of Radiation) used for the treatment of dentinal hypersensitivity can be divided into two categories: Low output power lasers and Middle output power lasers. Laser works by two mechanisms on sensitive teeth.[5] First; by melting effect with the crystallization of dentin's inorganic component and coagulation of fluids contained in the dentinal tubules. Second; by reducing the pulpal nerve pain threshold. Among the Lasers, Diode lasers showed the best results even in the treatment of high-grade dentinal hypersensitivity.6 Low output power Diode laser for the treatment of dentinal hypersensitivity is an appropriate strategy to promote bio-modulatory effects, minimizes pain and reduces inflammatory processes. Diode lasers showed reduction in dentinal hypersensitivity equal to or superior to the other conventionally used desensitizing agents or dentrifices.[1]

Remin Pro is a water based paste containing fluoride, hydroxyapapite and xylitol, used as desensitizing agent. Fluoride and hydroxyapatite reinforces remineralisation and strengthen the enamel surface.7 When Remin Pro is applied on the tooth surface, fluoride is converted to fluorapatite after coming in contact with saliva and once the fluorapatite is formed, the tooth surface becomes more resistant to acid attacks of cariogenic bacteria. Xylitol helps in preventing it from being converted into harmful lactic acid by cariogenic bacteria thus allowing the teeth to naturally remineralize with less interruption.[8]

CPP–ACP(GC Tooth mousse) is a desensitizing agent/paste preventing the dissolution of calcium and phosphate ions in tooth structure and maintains a super-saturated solution of bioavailable calcium and phosphate ions.[2] GC tooth mousse also contains a high concentration of neutral sodium fluoride, stannous fluoride and strontium fluoride; which forms mineral precipitates and these precipitates physically seal dentinal tubules with a plug that resists normal pulpal pressures, acid challenges and reduces flow of dentinal fluid.[9]

Fluoridated-Bioactive Glass(BaG) (Elsenz) desensitizing paste contains Bioglass in the form of Nova Min for the treatment of dentinal hypersensitivity and repair of sensitive teeth. It is based upon 5% w/w Calcium-Sodium Phospho Silicate (CSPS) technology. The CSPS technology delivers a hydroxyapatite like reparative layer to the surface of dentin, making it resistant to acid challenge.10 Fluoride containing bioactive glasses forms Fluorapatite, considered to be more acid resistant than hydroxy-carbonate apatite. An increase in phosphate content in the glass allows the formation of fluorapatite rather than fluorite, even at a lower pH. Furthermore, flourapatite is less soluble compared to hydroxyapatite and hydroxyl-carbonate apatite, thus making it more resistant to acid attacks.[11]

Non-Fluoridated Bioactive Glass (Shy-Nm) desensitizing paste also contains Bioglass in the form of NovaMin and is based on Calcium-Sodium Phospho Silicate. Sodium ions of N-FBaG begins to exchange with hydrogen ions in the tooth structure followed by rapid release of calcium and phosphate ions from the N-FBaG in the tooth structure. These reactions occur within seconds of exposure of particles of the desensitizing agent to saliva and the release of the calcium and phosphate ions continues as long as the particles are exposed to the aqueous environment.[12]

Materials and Method:

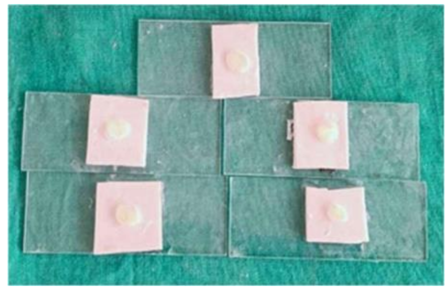
One-hundred twenty freshly extracted human permanent maxillary first and second premolar teeth were collected in Triveni Institute of Dental Sciences, Hospital and Research centre, Bilaspur. India. Inclusion Criteria: Teeth without any previous; Crown or root surface caries, restorations, clinically detectable tooth cracks or fractures, stains, wear facets, attrition, abrasion, erosion, abfraction lesions, white spot lesions, hypoplasia, dentin sclerosis and teeth extracted only for orthodontic purposes and periodontally compromised teeth. Exclusion criteria: Teeth with previous; Caries, restorations, clinically detectable tooth cracks or fractures, stains, wear facets, attrition, abrasion, erosion, abfraction lesions, white spot lesions, hypoplasia, dentin sclerosis and also teeth which were subjected to pre-treatment with any chemical agents. All teeth were cleaned of superficial debris, calculus, residual tissue tags with ultrasonic instruments and were stored in deionized water (Stanbio Reagents Pvt. Ltd, Kolkata, India) at room temperature until used.

All teeth were then sectioned mesio-distally from the middle-third of crown perpendicular to its long axis using high speed diamond disc (DFS, Germany) attached to straight hand piece (NSK, Japan) to obtain dentin disc specimens with dimensions measuring 8mm mesio-distally, 6mm bucco-palatal and thickness or height of 2mm (Figure 1). Any remaining enamel and surface irregularities over dentin discs were removed using 1200 fine grit silicon carbide polishing paper (Desai Agency, Surat, Gujarat, India) to obtain dentin discs with smooth and uniform surfaces. All dentin disc specimens were then placed in glass beakers (Borosil, Mumbai, India) filled with deionised water and sonicated in ultrasonic bath (Confident Dental Equipments Pvt Ltd, Bangalore, India) for 10 minutes to ensure complete removal of polishing abrasives. By using double-sided tape (Nippon Industries, New Delhi, India), dentin discs of each tooth were fixed over microscopic slides (Plaza Industrial Corporation, Delhi, India), with the polished side of each dentin disc facing up (Figure 2). All the specimens were then randomly divided into six groups with 20 specimens per group.

Figure 1
Dentin disc

Figure 2

Dentin discs fixed on microscopic slides



Group A: Low Output Diode laser (Epic X, Biolace Inc, California, USA) was used for dentinal tubules occlusion. The specimens received irradiation with Low output Diode laser of 980 nm wavelength, 320 micrometer core diameter optic fiber working at a continuous power mode with 1W (Watt) output power, frequency 60 Hz, 1.5 Ampere current at 100 KV (KiloVoltage) directed perpendicular to the exposed tooth surface. Each dentin disc specimen was irradiated for 30 seconds once a day for three consecutive days.

Group B: Remin Pro (Voco Gmbh, Cuxhaven, Germany), Group C: Casein PhosphoPeptide - Amorphous Calcium Phosphate (CPP-ACP) (GC Recaldent Tooth Mousse, GC America Inc., Alsip, USA), Group D: Non-Fluoridated Bioactive Glass (SHY- NM) (Group Pharmaceuticals Ltd, Karnataka, India), Group E: Fluoridated Bioactive Glass (Elsenz) (Group Pharmaceuticals Ltd, Malur, Karnataka, India) were used as desensitizing agents/pastes (Figure 3). In all the specimens of Group B, C, D and

E; A pea sized amount of desensitizing paste/agent as per the respective groups was placed on each of the dentin discs. Whereas in Group F(Control group): Distilled water was used as desensitizing agent. 4ml of distilled water was used per specimen. By using Oral B Extra soft bristle toothbrush (Oral B Sensitive) (Rialto Enterprises Pvt. Ltd., Chennai, India) (Figure 4), with the brush bristles placed at 900 angulation in contact with the dentin disc, each dentin disc was brushed in circular motion with light pressure twice a day at an interval of 12 hours for 2 minutes each (counted using a stop watch) for 14 consecutive days. In each group, one tooth brush was used on 10 specimens, after which it was replaced with a new tooth brush for the remaining 10 specimens. All specimens were then gently rinsed with deionised water to ensure complete removal of any residual desensitizing paste/agent from the tooth surface.

The extent of dentinal tubules occlusion was evaluated using Scanning Electron Microscope (SEM) (Sigma 300 VP, Carl Zeiss, Oberkochen, Germany) (Figure 5). All the specimens (dentin discs) were dried in a dessicator for 2 days, sputter coated with a thin gold layer and each dentin disc was then examined at an operating voltage of 5kV(KiloVoltage) at a working distance of 8.6mm, at a magnification of X2000 under Scanning electron microscope and photomicrographs were taken of each specimen. A categorical scale of (0-4 Scores) was accorded to all the specimens following the criteria of Davies M et al,13 and CL Chen et al,14 to evaluate the photomicrographs obtained from Scanning electron microscope for the extent of dentinal tubules occlusion (Figure 6) and the data was recorded.

Score 0: Totally Un-occluded (0% dentinal tubules occlusion).

Score 1: Mostly Un-occluded (<25% of dentinal tubules occluded)

Score 2: Partially Occluded (>25% to <50% of dentinal tubules occluded).

Score 3: Mostly Occluded (>50% to <100% of dentinal tubules occluded).

Score 4: Completely Occluded (100% of dentinal tubules occluded)

Figure 3

Desensitizing methods used in the study

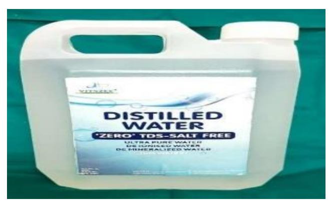
Group A

Group B

Group C

Group D

Group E



Group F

Group A: Low output Diode laser
Group B: Remin Pro

Group C: CPP-ACP(GC Tooth Mousse)

Group D: Non-F-BaG (Shy-Nm)

Group E: F-BaG (Elsenz)
Group F: Distilled water

Figure 4

Oral B Extra soft bristle toothbrush (Sensitive)

Figure 5

Scanning Electron Microscope (SEM)

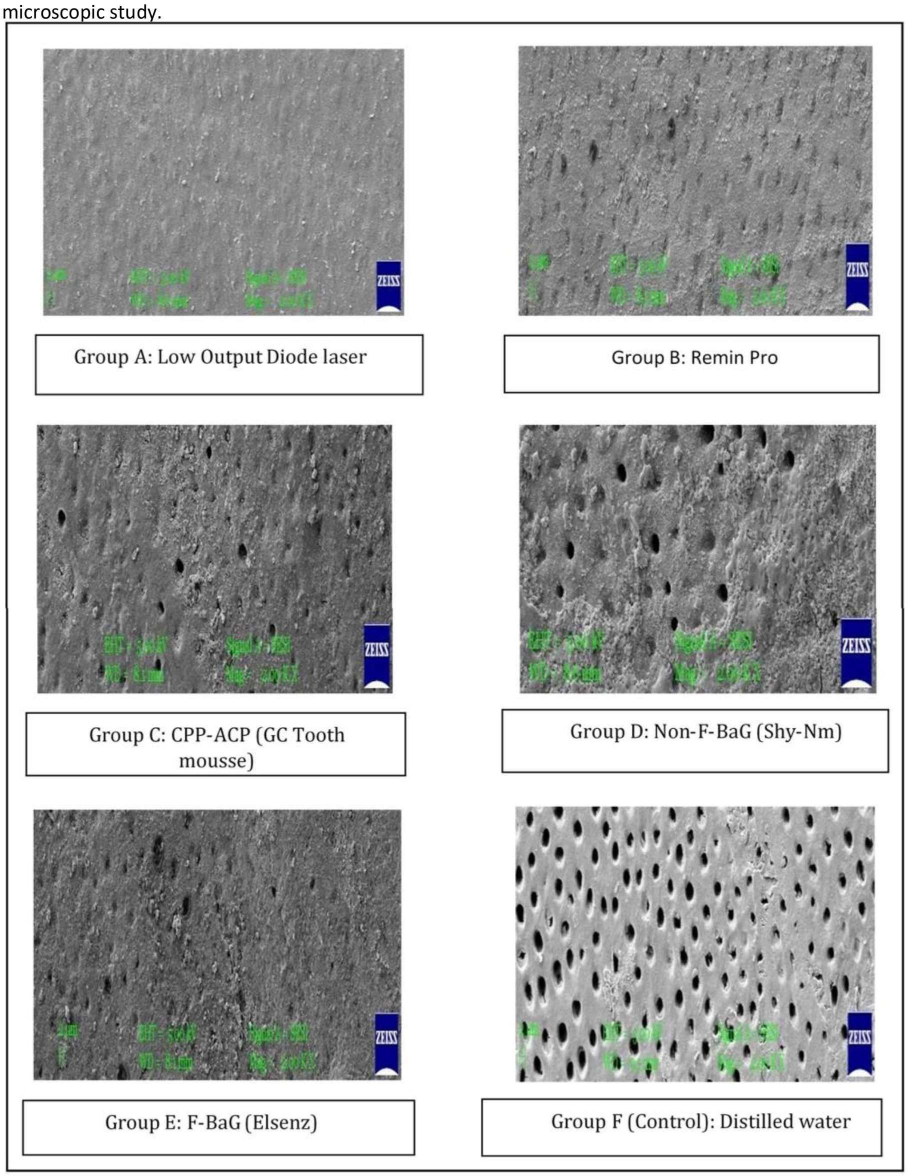


Figure 6

Scanning electron microscopic photomicrographs of Group A, Group B, Group C, Group D, Group E, Group F Specimens.

Results:

The obtained data of all specimens was tabulated in Microsoft excel and statistically analysed with Computer software; Statistical Package for Social Sciences (SPSS) Version 24. Chi-Square test helps to find the difference in the distortion of attributes in different groups was due to sampling variation or not. It was used to test the significance of difference between two properties when there are more than two groups to be measured (Table 1).

Scoring of Dentinal tubules occlusion	Group A Low Output Diode laser (n=20)		Group B Remin Pro (n=20)		Group C CPP-ACP (GC Tooth mousse) (n=20)		Group D Non-Fluoridated Bioactive Glass (SHY-NM) (n=20)		Group E Fluoridated-Bioactive Glass (ELSENZ) (n=20)		Group F Control (Distilled water) (n=20)	
	n*	%*	n	%	n	%	n	%	n	%	n	%
Score 0	0	0.0	0	0.0	0	0.0	0	0.0	0	0.0	20	100.0
Score 1	0	0.0	0	0.0	16	80.0	18	90.0	0	0.0	0	0.0
Score 2	0	0.0	4	20.0	4	20.0	2	10.0	4	20.0	0	0.0
Score 3	4	20.0	14	70.0	0	0.0	0	0.0	15	75.0	0	0.0
Score 4	16	80.0	2	10.0	0	0.0	0	0.0	1	5.0	0	0.0

Table 1 chi square test

n*: number of specimens

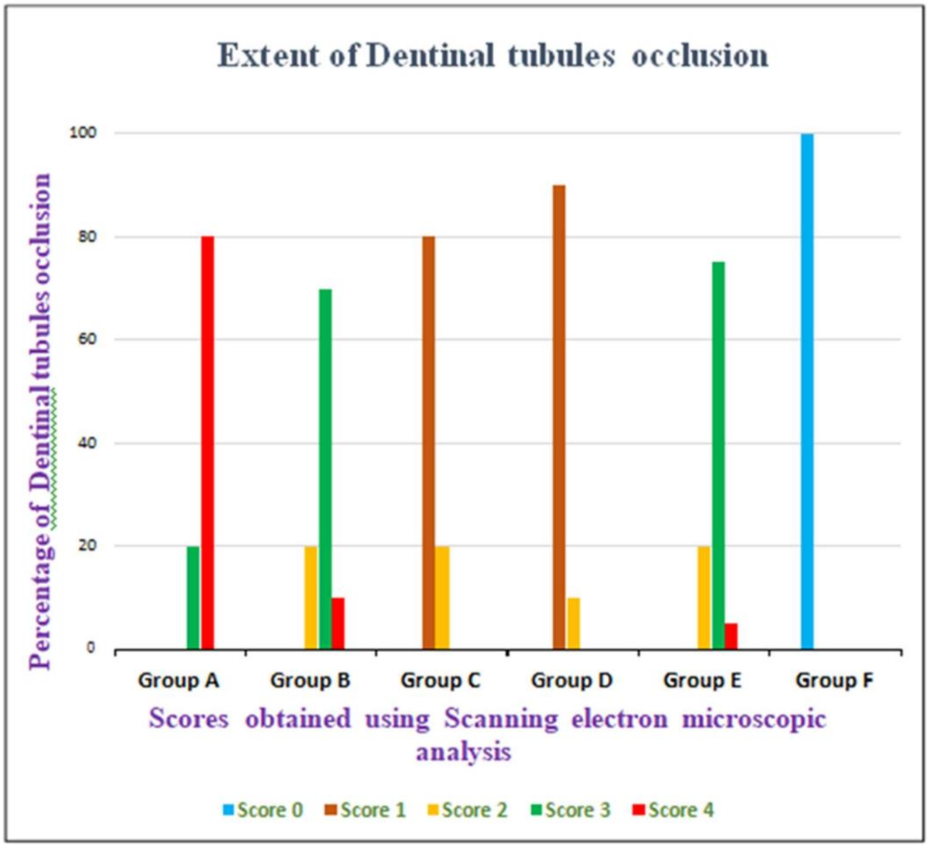
%*: percentage of specimens

Group A (Low output Diode Laser) showed the maximum occlusion of dentinal tubules with 80% of specimens with Score 4 (Completely occluded: 100% Dentinal tubules occlusion) and 20% of the specimens with Score 3 (Mostly occluded: >75% Dentinal tubules occlusion) followed by Group B (Remin Pro); 70% of specimens with Score 3 (Mostly occluded: >75% Dentinal tubules occlusion), 20% specimens with Score 2 (Partially Occluded: >25% to <50% of Dentinal tubules occlusion) and 10% specimens with Score 4 (Completely occluded: 100% Dentinal tubules occlusion) followed by Group E (Fluoridated Bioactive Glass [Elsenz]); 75% of the specimens with Score 3 (Mostly occluded: >75% Dentinal tubules occlusion), 20% of specimens with Score 2 (Partially Occluded : >25% to <50% of Dentinal tubules occlusion) and 5% of specimens with Score 4 (Completely occluded: 100% Dentinal tubules occlusion) followed by Group C [CPP-ACP](GC Tooth Mousse); 80% of the specimens with Score 1 (Mostly Unoccluded : <25% of Dentinal tubules occlusion) and 20% of specimens with Score 2 (Partially occluded : >25% to <50% of Dentinal tubules occlusion) followed by Group D (Non-Fluoridated Bioactive Glass [SHY-NM]) showed 90% of specimens with Score 1 (Mostly Unoccluded: <25% of Dentinal tubules occlusion) and 10 % specimens with Score 2 (Partially occluded : >25% to <50% of Dentinal tubules occlusion) (Graph 1).

In our study, Completely occluded dentinal tubules or Maximum occlusion of the dentinal tubules was seen in the specimens of Group A; Low output Diode laser used as desensitizing method, followed by Mostly occluded dentinal tubules seen in specimens of Group B; Remin Pro used as desensitizing agent/paste. Mostly Un-occluded dentinal tubules were seen in specimens of Group D; Non-Fluoridated Bioactive Glass(Shy-Nm) used as desensitizing agent/paste. Whereas, Group F; Control group (Distilled water) No dentinal tubules occlusion was seen in any of the specimens.

Statistically significant difference was seen between the specimens of Group A Vs Group B, Group A Vs Group C, Group A Vs Group D, Group A Vs Group E and Group A Vs Group F. Statistically significant difference was also seen between Group D (Non-Fluoridated Bioactive Glass) [Shy-Nm] Vs Group E (Fluoridated-Bioactive Glass) [Elsenz] (P[Probability] value < 0.05).

However, No statistically significant difference was seen between Group B (Remin Pro) and Group E (Fluoridated-Bioactive Glass [Elsenz]) and also between Group C [Casein Phospho Peptide - Amorphous Calcium Phosphate](CPP-ACP) and Group D (Non-Fluoridated Bioactive Glass)[Shy-Nm] (P value > 0.05).



Graph 1: Vertical Bar Graph–Dentinal tubules occlusion with various Desensitizing methods

Discussion:

In our study, Low out-put power Diode laser showed the maximum efficiency in completely occluding the dentinal tubules compared to other desensitizing agents used. This corroborates with previous studies of Corneli R et al,[1] Asnaashari M et al,[15] El Mobadder M et al,[16] Patil AR et al[17] reported better efficiency of Diode laser in the occlusion of dentinal tubules compared to Remin Pro, CPP-ACP, Non-Fluoridated Bio-active Glass desensitizing pastes and stated that Diode laser produces mild, random, irregular melting of peri-tubular and inter-tubular dentin causing obliteration and occlusion of the dentinal tubules. According to Asnaashari et al[15], Low output power Diode laser shows bio- modulatory effect, minimizes pain and inflammatory response. Diode laser acts on dentinal hypersensitivity by two mechanisms. Firstly; By melting effect, with crystallization of inorganic component of dentin and coagulation of fluids in the dentinal tubules. Secondly; By reducing the pulpal nerve's pain threshold. In our study, the power setting of Diode laser was 1W (Low out-put power) and this was postulated to provoke a sufficient increase in temperature to melt and occlude dentinal tubules.[15] Rimaneepong V et al[18] showed Diode laser used at a power setting of 0.8W, 1W, 1.6W and 2W caused partial occlusion of dentinal tubules by melting and contraction of dentin without causing any adverse effect on pulp tissue and concluded that the main advantages of diode laser in the treatment of dentinal hypersensitivity also includes; its small size, affordability,portability, ease of use and time saving. In our study, No cracks or destruction of any specimens exposed to low output diode laser were noticed under SEM analysis.

Saliva has shown to naturally occlude dentinal tubules by forming a protective glycoprotein layer with the help of salivary calcium, phosphate ions and this process of natural dentinal tubules occlusion is reported to be extremely slow and tubule plugging gets easily dislodged by dietary acids, thereby rendering it in-effective in providing lasting relief. Hence, various chemical formulations in the form of desensitizing pastes or dentrifices are developed to occlude the dentinal tubules reducing dentinal hypersensitivity.[10] Dentrifices with mineralization promoting agents contain calcium and phosphate ions causing effective occlusion of dentinal tubules of teeth. Desensitizing dentrifices/pastes like CPP-ACP (GC Tooth mousse), Remin Pro, Fluoridated-Bioactive Glass (Elsenz) and Non-Fluoridated Bioactive Glass (Shy-Nm) showed the ability to occlude dentinal tubules preventing dentinal hypersensitivity.[19]

In our study, Remin Pro as desensitizing agent was highly effective in occluding dentinal tubules compared to CPP-ACP and this corroborates with Mebin George Matthew et al[20] in their research concluded that percentage of dentinal tubules occlusion was significantly higher with Remin Pro(71.%) compared to CPP-ACP(65%) under SEM analysis.

NovaMin present within the Bioactive Glass(BaG) is an important constitiuent in both Fluoridated-Bioactive Glass (Elsenz) and Non-fluoridated Bioactive Glass (Shy-NM) desensitizing dentrifices/pastes. NovaMin binds to the exposed dentinal surface and reacts with it to form a mineralized layer. Calcium Sodium PhosphoSilicate (CSPS) in NovaMin has a strong attraction to collagen and physically occludes the dentinal tubules. Initial reactivity of the NovaMin particles is associated with formation of surface negative charge which facilitates binding to side groups on Type-1 collagen fibers of dentin. NovaMin can quickly occlude the dentinal tubules in forming a protective layer on the dentin surface and this layer is not only mechanically strong but also resistant to acid attack. Further, the continuous release of calcium ions from novamin over-time has shown to maintain the protective effects on dentin and maintain the occlusion of the dentinal tubules.[21,22] Halenur Altan et al[23] showed that the dentrifices containing bioactive glass appears to be more effective in its penetration into the prismatic and inter-prismatic structure of deminerlised enamel.

In our study, F-BaG (Elsenz) as desensitizing agent/paste showed maximum efficiency in dentinal tubules occlusion compared to Non-F-BaG (Shy-Nm). Fluoro-calcium phosphosilicate (F-BaG) is a fluoride releasing bioactive glass recently incorporated in desensitizing dentrifices or tooth pastes. It differs from the conventional calcium-sodium phosphosilicate(Non-F-BaG) by the presence of CaF2(Calcium Fluoride) in the glass, higher phosphate content and smaller average particle size of 6μm enabling it for deeper penetration into the dentinal tubules causing occlusion of tubules.[24,25] Moreover, the presence of CaF2 increases the glass dissolution and higher phosphate content also enhances the ability to form fluoro-apatite crystals even at low pH. Fluoro-Calcium PhosphoSilicate dentifrices have shown to instantly occlude the dentinal tubules.[26,27]

When NovaMin particles come into contact with saliva, an immediate release of sodium ions occur, which increases the local environmental pH and this combination of sodium ion release with rise in pH facilitates the rapid precipitation of calcium phosphate HCA (Hydroxy Carbonate Apatite) layer and release of calcium and phosphate ions. The CSPS particles acts as reservoirs to release calcium and phosphate ions continuously into the local environment.[28]

Our study also corroborates with a previous study of Desai A et al[10] concluded specimens treated with Fluoridated-Bioactive Glass (F-BaG)(Elsenz) demonstrated higher dentinal tubules occlusion compared to Non-fluoridated bioactive glass(Shy-Nm) as desensitizing agent/paste. Shaikh K et al[29] in their comparative study of desensitizing pastes, observed the percentage of dentinal tubules occlusion for Fluoridated-Bioactive Glass (Elsenz) as 88% compared to Non-Fluoridated Bioactive Glass (Shy-Nm) with 78%. Our study showed; no significant difference in the extent of dentinal tubules occlusion between F-BaG (Elsenz) and Remin Pro as desensitizing agents.

CPP-ACP is the acronym for a complex of Casein Phospho Peptide-Amorphous Calcium Phosphate. When CPP-ACP was used as desensitizing agent on tooth surface, it showed substantial crystal-like deposits within the lumen of dentinal tubules and as peptide-complexes binds to the tooth surface, it is said to deliver bioavailable calcium and phosphate for remineralization resulting in the occlusion of dentinal tubules.[30] However in our study, CPP-ACP showed minimal dentinal tubules occlusion compared to Diode laser, Remin Pro and Fluoridated-Bioactive Glass (Elsenz).

In our study, all the dentin disc specimens used were sectioned from the middle-third crown of teeth, so as to obtain dentinal tubules in circular cross sections as the mean dentinal tubules diameter is a reliable index in the SEM analysis of specimens among in-vitro studies.[10,19] Scanning Electron Microscope(SEM) is one of the main testing instrument for in-situ analysis of micro-areas. It has the advantages of higher resolution, larger depth of field, higher magnification and strong stereoscopic vision. Photo micrographs of SEM shows clearer definitions than those by optical microscopy. Early SEM's were applied only to study the morphology, location and distribution of sub-microscopic areas in the surface layer of fluid inclusions. However, newer SEM's show greater potential for identifying the capture minerals and daughter minerals in fluid inclusions. Even, at the magnification of 2000X - 20,000X, extremely clear photos can be obtained with SEM.31

Conclusion:

Within the limitations of this study it was found that; Maximum or completely occluded dentinal tubules were seen with Low-output Diode laser used as the desensitizing method. Among the desensitizing agents/pastes used, Remin Pro was found to be the most effective in dentinal tubules occlusion followed by Fluoridated-Bioactive Glass (Elsenz). Non-Fluoridated Bioactive Glass (Shy-NM) was found to be the least effective in dentinal tubules occlusion.However, further in-vivo studies are recommended to confirm and correlate the findings of this in-vitro study to a clinical scenario.

Acknowledgments

We are grateful to two anonymous reviewers for their valuable comments on the earlier version of this paper.

References:

1. Corneli R, Kolakemar A, Damda A, et al. An in-vitro evaluation of dentinal tubule occlusion using three desensitizing methods: A scanning electron microscopic study. J Conserv Dent; 2020:23:86-90.

2. Sanjay Miglani, Vivek Aggarwal, Bhoomika Ahuja, et al. Dentin hypersensitivity: Recent trends in management. J Conserv Dent; 2010:13:4:218-224.

3. Dababneh R, Khouri A, Addy M, et al. Dentine hypersensitivity: An enigma? A review of terminology, epidemiology, mechanisms, aetiology and management. Br Dent J; 1999;187:606-611.

4. Addy M. Etiology and clinical implications of dentine hypersensitivity. Dent Clin North Amer; 1990:34:503-514.

5. Al-Saud LM, Al-Nahedh HN. Occluding effect of Nd:YAG laser and different dentin desensitizing agents on human dentinal tubules invitro : A scanning electron microscopy investigation. J. Oper Dent; 2012:37:340-355.

6. Umberto R, Claudia R, Gaspare P, et al. Treatment of dentine

hypersensitivity by diode laser : A clinical study. Int J Dent; 2012:1-8.

7. Kiana Salehzadeh Esfahani, Romina Mazaheri, Leila Pishevar, et al. Effects of treatment with various remineralizing agents on the microhardness of demineralized enamel surface. J dent Res Dent Clin Dent Prospect; 2015:9:4:239-245.

8. Blaes J. Tooth protection, freedom, whitening, and better gums. Dental economics. Pennwell corporation; 2011.

9. Walsh LJ. The effects of GC tooth mousse on cervical dentinal sensitivity: A controlled clinical trial. S Afr dent J; 2010; 12:4-12.

10. Dessai A, Shetty N, Srikant N, et al. Evaluation of the effectiveness of fluoridated and non- fluoridated desensitizing agents in dentinal tubule occlusion using scanning electron microscopy. An in-vitro study. Dent Res J; 2020:17:193-199.

11. Bakri MM, Hossain MZ, Razak FA, et al. Dentinal tubules occluded by bioactive glass-containing toothpaste exhibit high resistance toward acidic soft drink challenge. Aust Dent J; 2017:62:186-191.

12. Shah S, Shivakumar AT, Khot O, et al. Efficacy of NovaMin- and Pro-Argin-containing desensitizing dentifrices on occlusion of dentinal tubules. Dent Hypotheses; 2017:8:104- 109.

13. Davies M, Paice EM, Jones SB, et al. Efficacy of desensitizing dentifrices to occlude dentinal tubules. Eur J Oral Sci; 2011:119:497-503

14. Chen CL, Parolia A, Pau A, et al. Comparative evaluation of the effectiveness of desensitizing agents in dentine tubule occlusion using scanning electron microscopy. Aust Dent J; 2015:60:65-72.

15. Asnaashari M, Moeini M. Effectiveness of lasers in the treatment of dentin hypersensitivity. J. Lasers Med Sci; 2013:4:1-7.

16. El Mobadder M, Namour A, Namour M, et al. Dentinal hypersensitivity treatment using diode laser 980 nm: In-vivo study. Dent J Basel; 2019:7:5:1-6.

17. Patil AR, Varma S, Suragimath G, et al. Comparative evaluation of efficacy of iontophoresis with 0.33% sodium fluoride gel and diode laser alone on occlusion of dentinal tubules. J Clin Diagn Res; 2017:11:123-126.

18. Rimaneepong V, Palamara JE, Wilson PR, et al. Pulpal space pressure and temperature changes from Nd:YAG laser irradiation of dentin. J Dent; 2002:30:291-296.

19. Ghafournia M, Tehrani MH, Nekouei A, et al. In vitro evaluation of dentin tubule occlusion by three bioactive materials: A scanning electron microscopic study. Dent Res J; 2019:16:166- 171.

20. Mathew Mebin George, Ashu Jagdish, Khan Md Muzammil, et al. Efficacy of remineralizing agents to occlude dentinal tubules in primary teeth subjected to dentin hypersensitivity in vitro SEM study. Journal of Family Medicine and Primary Care; 2020:9:1:354-358.

21. Burwell A, Jennings D, Muscle D, et al. NovaMin and dentin hypersensitivity – In-vitro evidence of efficacy. J Clin Dent; 2010:21:66-71.

22. Litkowski LJ, Hack GD, Sheaffer HB, et al. Occlusion of dentin tubules by 45S5 bioglass. Ceramics in Medicine; 1997:10:411-414.

23. Halenur Altan, Zehnep Gotzap, Kamil Kahraman, et al. Inhibition effect of different toothpastes on demineralization of incipient enamel lesions. Oral Health Prev Dent; 2019:17:179-185.

24. Lynch E, Brauer DS, Karpukhina N, et al. Multi-component bioactive glasses of varying fluoride content for treating dentin hypersensitivity. Dent Mater; 2012:28:168-178.

25. Jones JR, Brauer DS, Hupa L, et al. Bioglass and bioactive glasses and their impact on health care. Int J Appl Glass Sci; 2016:7:423-434.

26. Mneimne M, Hill RG, Bushby AJ, et al. High phosphate content significantly increases apatite formation of fluoride-containing bioactive glasses. Acta Biomater; 2011:7:1827-1834

27. Brauer DS, Karpukhina N, O'Donnell MD, et al. Fluoride-containing bioactive glasses: Effect of glass design and structure on degradation, pH and apatite formation in simulated body fluid. Acta Biomater; 2010:6:3275-3282.

28. Layer TM. Development of a fluoridated, daily-use toothpaste containing NovaMin technology for the treatment of dentin hypersensitivity. J Clin Dent; 2011:22:59-61.

29. Shaikh K, Pereira R, Gillam DG, et al. Comparative evaluation of desensitizing dentifrices containing biomin, novamin and fluoride on dentinal tubule occlusion before and after a citric acid challenge. A Scanning electron microscopic in-vitro study. J Odontol; 2018:2: 1-6.

30. Jitendra Saraf , Ramesh Amirisetty , RM Zade, et al. Evaluation of effectiveness of CPP- ACP combination in treating dentinal hypersensitivity following non-surgical periodontal therapy – A randomized clinical trial. Chhattisgarh Journal of Health Sciences; 2013:1:1:32-34.

31. McMullan D. An improved scanning electron microscope for opaque specimens. Procedings of the IEE - Part II: Power Engineering; 1953:75:245–256.