COMPARATIVE EVALUATION OF 940 NM DIODE LASER IRRADIATION ON DENTINAL TUBULES OCCLUSION WITH OR WITHOUT SMEAR OF GRAPHITE – A SCANNING ELECTRON MICROSCOPIC STUDY.

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ABSTRACT

Aim : The present study aimed to compare the number of un occluded dentinal tubules and the diameter of un-occluded dentinal tubules with 940 nm diode laser with, or without graphite and desensitizing agent.

Materials and methods: This in-vitro study consisted of three groups. Cross sectional slice of single rooted teeth (premolar) extracted for orthodontic purpose was used for the study. Group 1 (n=5) teeth coated with graphite and irradiated with 940 nm diode laser. Group 2 (n=5) teeth irradiated with 940 nm diode laser. Group 3 (n=5) teeth treated with desensitizing agent. Then SEM analysis using 2500 X magnification was done to analyse number of un occluded dentinal tubules and diameter of remaining dentinal tubules .Statistical analysis was done using Mann- Whitney test and Kruskal-Wallis test.

Results: Results concluded that there was no statistical difference between the three groups - diode laser with graphite group, diode laser and desensitizing agent in the remaining number of un occluded dentinal tubule occlusion. Diode laser with Graphite group (1.40±1.949) showed reduced number of un occluded dentinal tubules. There was statistical difference between three groups in diameter of remaining open dentinal tubules after the usage of Diode laser with graphite (0.2760±0.39068) compared to diode laser group and desensitizing agent group.

Conclusion: The results of this invitro SEM study showed test group (graphite with diode laser), when compared to control group (desensitizing paste), is more effective in the occlusion of dentinal tubules

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thereby reducing the dentinal hypersensitivity. Hence it can be used as a therapeutic strategy for treating dentinal hypersensitivity.

Key words – Diode laser, Graphite, Dentinal hypersensitivity

INTRODUCTION

Dentin hypersensitivity (DH) is a term for an aberrant response of stimuli, most commonly chemical, thermal, tactile, or osmotic. It is indicated by acute, localised, brief pain from denuded dentin, which will be absent in a healthy tooth under normal circumstances.(1) The potential reasons of dentin hypersensitivity come in a wide variety, but a key contributing element is the loss of enamel and removal of cementum from the root, exposing dentin.(2)

Various theories have been proposed for biological process of DH, three major theories of dentinal sensitivity are direct innervation theory, odontoblast receptor, fluid movement/hydrodynamic theory.(3) Brannstrom's hydrodynamic theory is the one that is most currently accepted which states that the movement of fluid in the dentinal tubules results in displacement of odontoblasts located in the external layers of the pulp or in the internal terminals of tubules .(4)

The methods of treatment for DH are centred on lowering dentinal permeability. For this reason, earlier restorative materials, desensitising agents have been used to occlude dentinal tubules. An ideal desensitising product shouldn't cause tooth discoloration, should be relatively painless, simple to apply, quick, and permanently effective, and should permit occlusion of dentinal tubules without damaging the pulp.(5)

The He-Ne laser is the first laser to be used to treat dentine hypersensitivity, according to Senda et al. (1985). Since then, various lasers, such as CO_2, Er: YAG, etc., have been utilised for the treatment of dentinal hypersensitivity.

These hard tissue lasers works by increasing the temperature of the surface they are irradiated at, which can cause the dentinal surface to melt and re-crystallize resulting in closure of tubule orifices.(6)

In early 2000's diode laser have been used for the treatment of dentinal hypersensitivity. Nevertheless, dentin has a low absorption coefficient for diode lasers. This limited absorption in dentine results in the propagation of the laser beam to the pulp tissue which will generate heat and perhaps cause unfavourable side effects including hyperaemia or irreversible pulpitis.(7)

So various desensitizing agents like sodium fluoride, potassium nitrate, varnish were combined with diode laser for treating dentinal hypersensitivity.

Because diode lasers are more readily absorbed by dark pigments, graphite was used in conjunction with a diode laser to close dentinal tubules in an in vitro investigation by Ummana et al. in 2013. Hence, the graphite paste will prevent the laser beam from reaching the pulp, which will limit any potential adverse effects on the pulpal tissue. Through a melting process, the graphite paste will also seal off the dentinal tubules.

This study was aimed to compare the number of un occluded dentinal tubules and the diameter of un-occluded dentinal tubules with 940nm diode laser with, or without graphite and desensitizing agent.

STUDY OBJECTIVE

To compare the number of un occluded dentinal tubules irradiated with 940nm Diode laser with, or without graphite.

To compare the diameter of un occluded dentinal tubules with 940nm Diode laser with, or without graphite.

RATIONALE

To assess if diode laser with graphite is an effective method to reduce dentinal hypersensitivity by comparing the dentinal tubule closure with desensitizing paste.

MATERIALS AND METHOD

The study was conducted on the single rooted premolars which was extracted for orthodontic purpose from patients attending outpatient department of Amrita School of Dentistry. Teeth which were periodontally compromised, fractured, malformed, with dental caries and those with restorations were excluded from the study.

Sample size calculation – Sample size was calculated from a previous study done by Maleki-Pour et al in 2015(8). The minimum sample size for the present study comes to 5 in each group totalling to 15.

Preparation of dentinal disc: The selected teeth were sound and devoid of carious lesions, cracks, or fractures. The enamel was removed with plain cut tungsten carbide fissure burs at high speed under a continuous water spray, and crown dentin disc cross sectional slices with a thickness of 2mm were made. The dentin discs were polished using 600-grit carborundum paper and then washed with distilled water. Tetracycline hydrochloride was used to etch each specimen for 1 minute, exposing the dentinal tubules, and then it was thoroughly cleaned with distilled water. To prevent microbial growth all of the discs were stored in 4 ^{o}C distilled water containing 0.2% thymol.

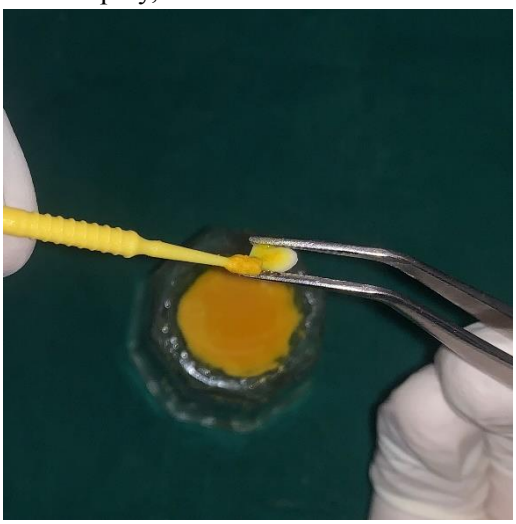

Fig 1 Dentinal discs were etched with tetracycline hydrochloride for 1 min

Treatment

Experimental group 1

All the five teeth in the group received a graphite coating and a radiation treatment with 980 nm diode laser. The graphite powder - ARTGRAF which is soluble in water, was used as an enhancer. To coat the dentinal surface the graphite was mixed with distilled water. After that,940 nm diode laser (200 µm laser fibre tip with a 1 W, continuous mode) was used for irradiation. As the particle size of the residual graphite was larger than the average diameter of the dentinal tubules, it was possible to easily remove residual graphite from these samples by gently rinsing them with distilled water.

Fig 2 a **Fig 2 b**

Fig 2 a,b Application of graphite paste and irradiating with 940 nm diode laser

Experimental group 2

All the five teeth in the group were irradiated with 940 nm. The dentinal disc was irradiated

with by using 940 nm diode laser on it with 1 W, continuous mode, irradiation speed: 1 mm/sec for 10 sec, laser fibre diameter: 200 µm**.**

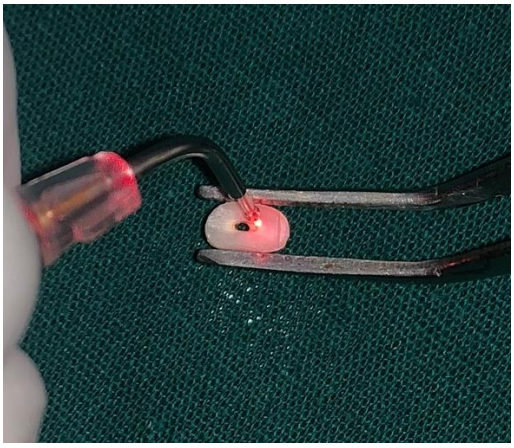

Figure – 3 Irradiation of 940 nm diode laser

Experimental group3(control group)

All the five teeth in the group were treated with desensitizing paste. The dentinal disc was treated with thermoseal RA.

Figure 4 Application of desensitizing agent on tooth surface

STUDY DESIGN

Invitro study - Comparative study.

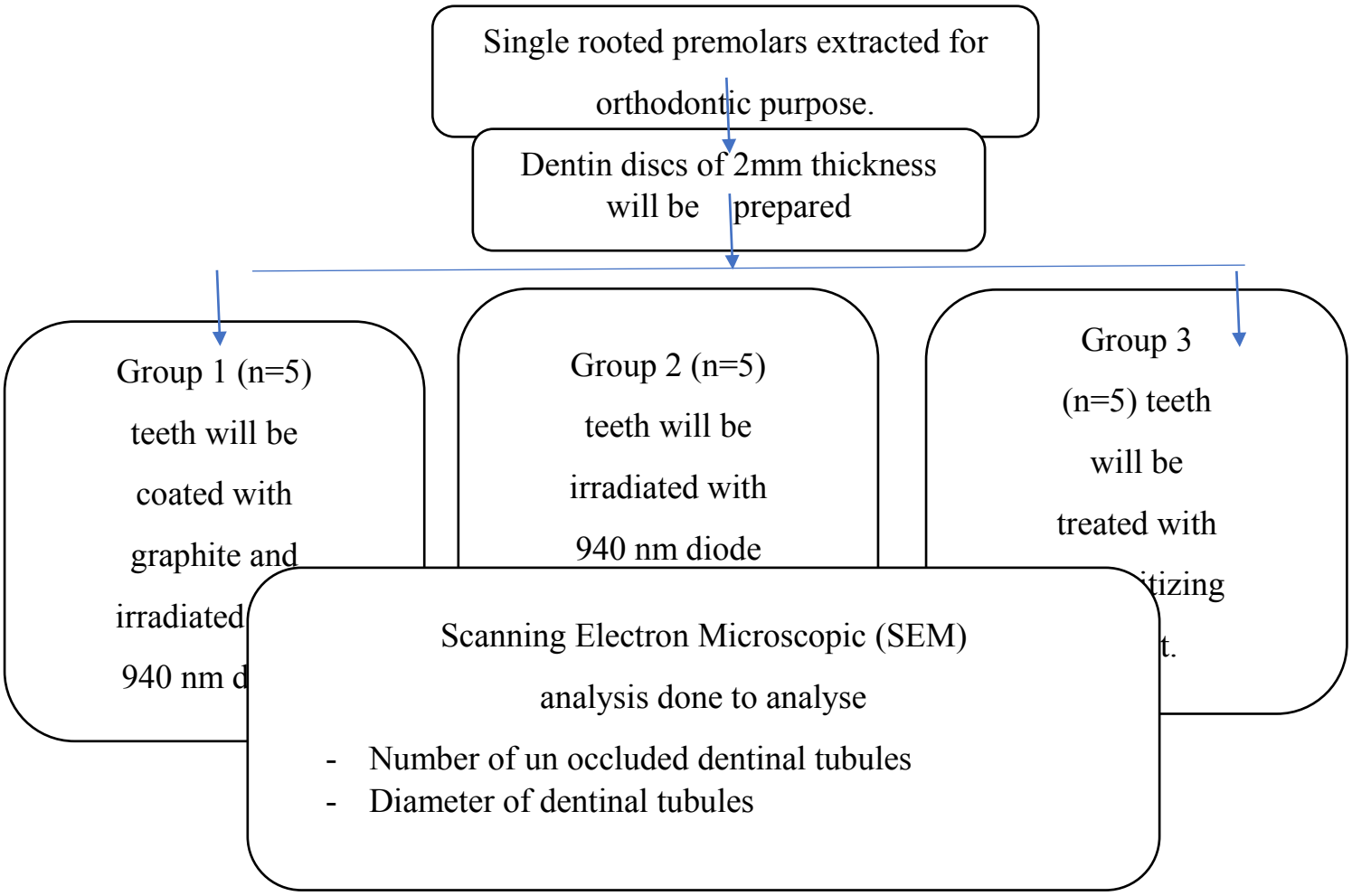

Figure 5 Study Design

SCANNING ELECTRON MICROSCOPY (SEM) ANALYSIS

The samples were attached to aluminium stubs and gold (25 nm thick), using a metallizer (model SCD 005, Bautec,Germany). They were then dehydrated at room temperature. The samples were examined with a 2500x magnification SEM for analysis.

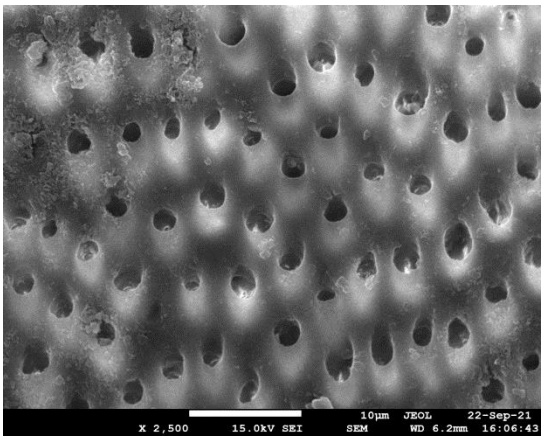

Figure 6 (View of unlased dentin under scanning electron microscopy (SEM) that has only received tetracycline as treatment. The dentin is not covered by the smear layer. Open tubules can be seen. 2500x magnification. The tubules are open. Magnification: 2500×.)

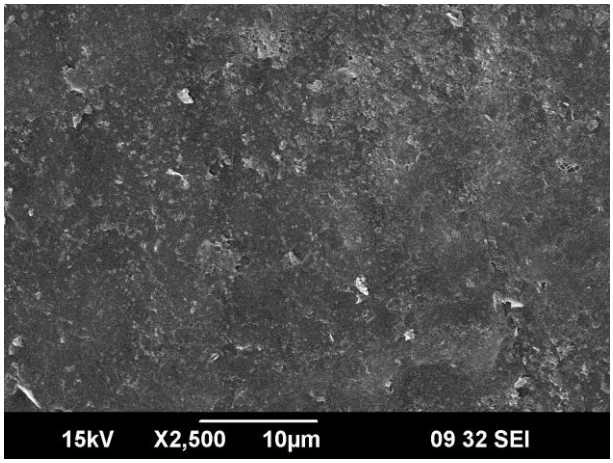

Figure 7 Group 1 (View of treated dentin by diode laser (940 nm) at 1 W with graphite under scanning electron microscopic (SEM). A narrowing of dentinal tubules can be noted. Only a few tubules are completely obliterated. Magnification: 2500×.)

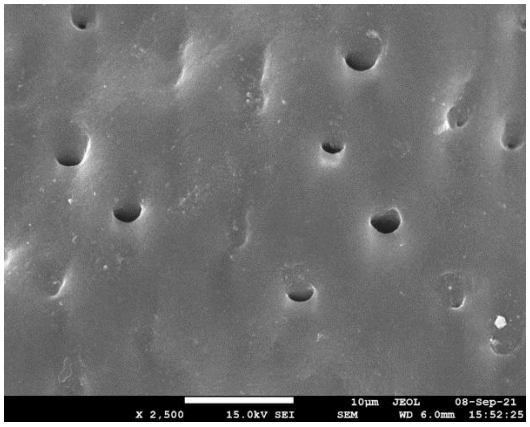

Figure 8 Group 2 (View of treated dentin by diode laser (940 nm) at 1 W under Scanning electron microscopic (SEM). A narrowing of dentinal tubules can be noted. Only a few tubules are completely obliterated. Magnification: 2500×.)

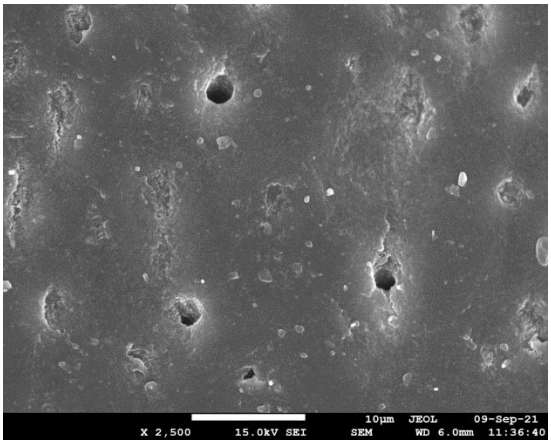

Figure 9 Group 3 (View of treated dentin by desensitizing past under Scanning electron microscopic (SEM). A narrowing of dentinal tubules can be noted. Magnification: 2500×.)

STATISTICAL ANALYSIS

SPSS Version 22 was used for statistical analysis of data. Mann- Whitney test was to evaluate the statistical significant difference in the mean number of remaining un occluded dentinal tubules and Kruskal-Wallis test was used to evaluate the statistical significant difference in the mean number diameter of remaining dentinal tubules in the three different groups (Diode plus graphite, diode laser and desensitizing paste). P value of <0.05 was considered as statistically significant.

RESULTS

The mean number of remaining un occluded dentinal tubules after the usage of Diode laser with graphite was found to be 1.40±1.949.

The remaining un occluded dentinal tubules after the usage of Diode laser and Desensitizing agent was found to be 6.00±3.317 and 4.00±2.73 respectively.

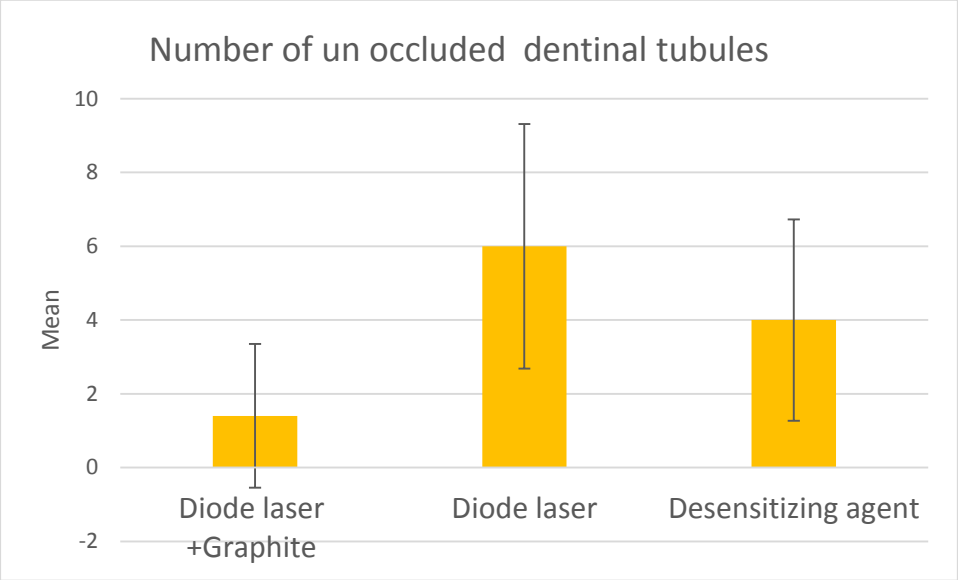

Figure – 10 Bar graph of mean and standard deviation of number of un occluded dentinal tubules of all the three groups.

Groups	n	Mean ±SD	Median	P value
1.Diode laser	5	6.00±3.317	5.00	P=0.45
2.Desensitizing agent	5	4.00±2.73	4.00	

Table 1 – Mean and standard deviation of un occluded dentinal tubules of diode laser group and desensitizing agent.

Groups	n	Mean ±SD	Median	P value
1.Diode laser +Graphite	5	1.40±1.949	0.00	P=0.12
3.Desensitizing agent	5	4.00±2.73	4.00	

Table 2 - Mean and standard deviation of un occluded dentinal tubules of diode laser group with graphite and desensitizing agent.

Desensitizing agent group (4.00±2.73) showed lesser number of un occluded dentinal tubules than diode laser group (6.00±3.317) but no statistical significance was found between the two groups (p = 0.45)

Diode laser with Graphite group (1.40±1.949) showed less number of un occluded dentinal tubules than desensitizing agent (4.00±2.73). There was no statistical significance between two groups (p value = 0.12)

Independent – samples median test showed that, there was no statistical difference between 3 groups diode laser with graphite group, diode laser and desensitizing agent.

DIAMETER OF THE DENTINAL TUBULES

Groups	n	Mean ±SD	Median	P value
1.Diode laser +Graphite	5	0.2760±0.39068	0.000	P = 0.006
2.Diode laser	5	1.53±0.20579	1.530	
3.Desensitizing agent	5	1.9220±0.17441	2.000	

Table 3

Sample 1-Sample 2	Test statistic	Significance	Adj Sig
Diode laser +graphite – Diode laser	10.000	0.002	0.005
Diode laser + graphite – Desensitizing agent	10.000	0.002	0.005
Diode laser - Desensitizing agent	3.600	0.058	0.173

Table 4

Table 3 ,4 Mean and standard deviation of diameter of remaining dentinal tubules of all the three groups.

Between three intervention groups there was statistically difference was observed for the remaining diameter of dentinal tubules as p value is 0.006.

Post hoc revealed there is statistical difference between Diode laser with graphite 0.2760±0.39068 and Diode laser 1.53±0.20579 group. Diode laser with graphite 0.2760±0.39068 and Desensitizing agent 1.9220±0.17441.

There is no statistical difference between diode laser 1.53±0.20579 and desensitizing agent1.9220±0.17441.

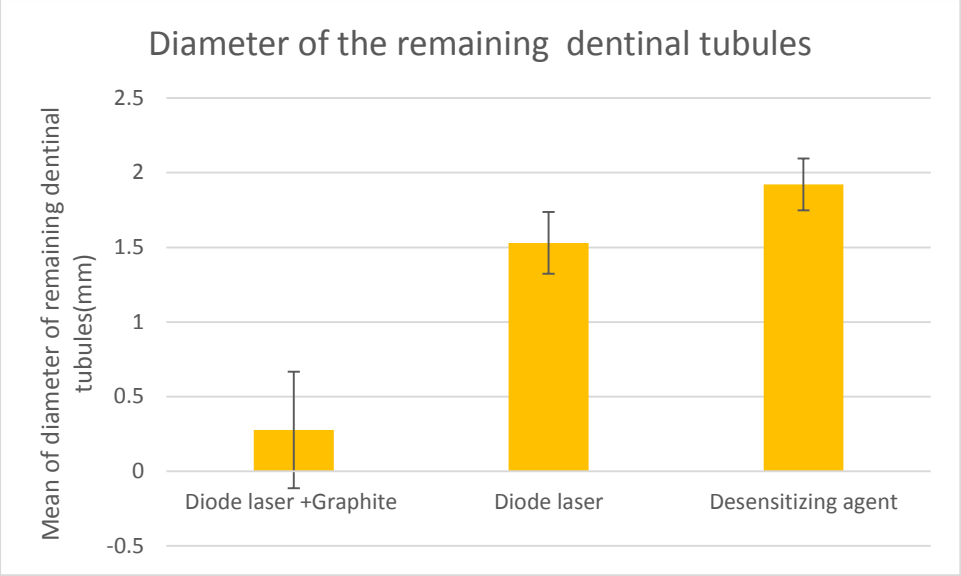

Figure – 11 Bar graph of mean and standard deviation of diameter of remaining dentinal tubules of all the three groups.

The mean diameter of remaining open dentinal tubules after the usage of Diode laser + graphite was measured to be 0.2760±0.39068.

The mean diameter of remaining open dentinal tubules after the usage of Diode laser and Desensitizing agent was measured to be 1.53±0.20579 and 1.9220±0.17441 respectively.

DISCUSSION

There are many methods which have been tried for the treatment of dentinal hypersensitivity including chemical agents like fluoride, potassium nitrate, strontium acetate , calcium sodium phosphosilicate etc which are often incorporated in some toothpastes, mouthwashes, and dentine adhesive sealers.(9)

The use of lasers in the treatment of dentinal hypersensitivity is relatively a recent concept which has shown promising results. Hard tissue lasers like CO_2, Er: YAG has been used in treatment of dentinal hypersensitivity .The dentinal surface melts and disarranges the crystalline arrangement as a result of thermochemical ablation, which is the fundamental working principle of a laser.(10) Hard tissue lasers are not commonly used because of high equipment cost.

In the recent past, diode laser has been tried for treating dentinal hypersensitivity. The sodium pump mechanism is affected, cell membrane permeability is altered, and the sensory axon ends are momentarily changed by diode laser. It has a melting effect which closes the dentinal tubules, thereby reducing dentinal hypersensitivity.(11)

The drawback of using diode LASER alone is that , the coefficient of absorption of diode lasers are low in dentin, hence it results in the propagation of the laser beam into the pulpal tissue .This will result in generation of heat due to high absorption by chromophores – melanin and haemoglobin causing undesirable side effects such as hyperaemia or irreversible pulpitis.(12)

In this study, the test group was irradiated with 940 nm diode laser 1 W, continuous mode,

irradiation speed: 1 mm/sec for 10 sec, tangentially mode (45º angle), laser fibre tip diameter of 200 μm with non-contact mode was used.

The 940 nm diode laser is absorbed by water at a higher rate, and the 810-nm wavelength is absorbed by melanin at a higher rate. The tooth pulp is less thermally affected by the 980-nm wavelength than by the 810-nm wavelength.(5)

1 W power was used, which is considered safe for the pulp and seals the dentinal tubules. Energy densities above 1.6 W results in destruction of dentin surface and eventually damage the dental pulp. Since the operator can more easily scan the entire dentin surface using continuous wave mode, it was chosen. Noncontact mode was used to safeguard the optical fibre from the graphite paste. To prevent the portion of the beam not absorbed by dentin from directly exposing the pulp, the tangentially mode (45º angle) was preferred.(5)

The application of graphite paste enhances the absorption of the laser beam superficially on to the dentinal surface which will prevent the propagation of light to the pulp and also leads to better dentinal tubules occlusion. It is important to note that the graphite paste will only evaporate rather than entering the dentinal tubules. In essence, the dentinal tubules' typical diameter is larger than the graphite paste's 5–25 micron particle size.(13)

The most common treatments for dentine hypersensitivity are desensitising toothpastes, as these toothpastes results in dentine tubule occlusion.(14) Potassium nitrate containing tooth paste is effective in occluding dentinal tubules. So, in this study thermoseal RA was used as control group. As its major ingredient is Potassium Nitrate 5%.

When compared to desensitising toothpaste, the use of a 980 nm diode laser by itself has an additional benefit for treating dentinal tubule occlusion.(15) But this was not in accordance with the present study as using 940 nm diode laser alone showed reduced dentinal tubular occlusion when compared to desensitizing agent but it was not statistically significant.

Graphite with 940 nm diode laser irradiation at 1 W, continuous mode, speed: 1 mm/second non-contact mode can lead to melting effect on dentin and narrowing of dentinal tubules. The results of Umana et al (5) seem to be supported by the current study's observation that group 1 diode laser with graphite had significantly less un occluded dentinal tubules and tubular diameters.

The SEM gives high-resolution magnified images of the surface topography and images can be statistically analysed .(16) In this study SEM analysis was done to evaluate the dentinal tubules occlusion and measure the diameter of dentinal tubules with image J software analysis with 2500x magnification.

According to the literature, there is a direct relationship between open dentin tubules, permeability and dentin hypersensitivity (Pinto et al., 2010; Sauro et al., 2010).

Parameters analysed were the un occluded dentinal tubules and also the diameter of the remaining dentinal tubules.

Results concluded that there was no statistical difference between the three groups - diode laser with graphite group, diode laser and desensitizing agent in the remaining number of un occluded dentinal tubule occlusion. Diode laser with Graphite group (1.40±1.949) showed reduced number of un occluded dentinal tubules. Statistically it was not significant it may be due to the small number of sample size.

There was statistical difference between three groups in diameter of remaining open dentinal tubules after the usage of Diode laser with graphite (0.2760±0.39068) compared to diode laser group and desensitizing agent group. This might be as a result of the use of graphite paste, which improves the laser beam's surface absorption and, by enhancing the superficial melting effect.

Reduced dentinal diameter of the remaining dentinal tubules were observed in group 1 due to irradiation of the diode laser on dentinal surface decrease tubule diameter markedly and thus reduce tubular fluid movement, an established mechanism to relieve dentin hypersensitivity.

CONCLUSION

The results of this invitro SEM study showed test group (graphite with diode laser), when

compared to control group (desensitizing paste), is more effective in the occlusion of dentinal tubules thereby reducing the dentinal hypersensitivity. Hence it can be used as a therapeutic strategy for treating dentinal hypersensitivity. Further studies have to be done with increase in sample size, comparing different diode laser wavelength, power settings and timings.

REFERENCE

1. Marsilio AL, Rodrigues JR, Borges AB. Effect of the clinical application of the GaAlAs laser in the treatment of dentine hypersensitivity. J Clin Laser Med Surg. 2003 Oct;21(5):291–6.
2. Trushkowsky RD, Oquendo A. Treatment of Dentin Hypersensitivity. Dent Clin North Am. 2011 Jul;55(3):599–608.
3. Miglani S, Aggarwal V, Ahuja B. Dentin hypersensitivity: Recent trends in management. J Conserv Dent JCD. 2010;13(4):218–24.
4. Brännström M, Åström A. A Study on the Mechanism of Pain Elicited from the Dentin. J Dent Res. 1964 Jul;43(4):619–25.
5. Umana M, Heysselaer D, Tielemans M, Compere P, Zeinoun T, Nammour S. Dentinal tubules sealing by means of diode lasers (810 and 980 nm): a preliminary in vitro study. Photomed Laser Surg. 2013 Jul;31(7):307–14.
6. Aranha ACC, Eduardo C de P. Effects of Er:YAG and Er,Cr:YSGG lasers on dentine hypersensitivity. Short-term clinical evaluation. Lasers Med Sci. 2012 Jul;27(4):813–8.
7. Parker S. Verifiable CPD paper: laser-tissue interaction. Br Dent J. 2007 Jan 27;202(2):73–81.
8. Maleki-Pour MR, Birang R, Khoshayand M, Naghsh N. Effect of Nd:YAG Laser Irradiation on the Number of Open Dentinal Tubules and Their Diameter with and without Smear of Graphite: An in Vitro Study. J Lasers Med Sci. 2015;6(1):32–9.
9. García-Delaney C, Abad-Sánchez D, Arnabat-Domínguez J, Valmaseda-Castellón E, Gay-Escoda C. Evaluation of the effectiveness of the photobiomodulation in the treatment of dentin hypersensitivity after basic therapy. A randomized clinical trial. J Clin Exp Dent. 2017 May;9(5):e694.
10. Reddy GV, Akula S, Malgikar S, Babu PR, Reddy GJ, Josephin JJ. Comparative scanning electron microscope analysis of diode laser and desensitizing toothpastes for evaluation of efficacy of dentinal tubular occlusion. J Indian Soc Periodontol. 2017;21(2):102–6.
11. Kimura Y, Wilder-Smith P, Yonaga K, Matsumoto K. Treatment of dentine hypersensitivity by lasers: a review. J Clin Periodontol. 2000 Oct;27(10):715–21.
12. Zach L, Cohen G. PULP RESPONSE TO EXTERNALLY APPLIED HEAT. Oral Surg Oral Med Oral Pathol. 1965 Apr;19:515–30.
13. Namour A, Nammour S, Peremans A, Heysselaer D, De Moor RJG. Treatment of dentinal hypersensitivity by means of Nd:YAP Laser: a preliminary in vitro study. ScientificWorldJournal. 2014;2014:323604.
14. Arnold WH, Prange M, Naumova EA. Effectiveness of various toothpastes on dentine tubule occlusion. J Dent. 2015 Apr 1;43(4):440–9.
15. Doppalapudi H, Kancharla AK, Nandigam AR, Sheema Tasneem M, Gummaluri SS, Dey S. Comparative evaluation of diode laser alone and in combination with desensitizing toothpaste in occlusion of dentinal tubules - A SEM study. J Oral Biol Craniofacial Res. 2023;13(2):224–9.
16. West NX, Macdonald EL, Jones SB, Claydon NCA, Hughes N, Jeffery P. Randomized in situ clinical study comparing the ability of two new desensitizing toothpaste technologies to occlude patent dentin tubules. J Clin Dent. 2011;22(3):82–9.