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A Scanning Electron Microscopic (SEM) study of the evaluation between the results by Gracey curette and Er,Cr:YSGG Laser on periodontally involved root surfaces

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Keywords: Er,Cr:YSGG Laser, periodontal therapy, root planing, scaling. Abstract: Er,Cr:YSGG lasers are considered one of the most successful non-surgical treatment modalities for periodontium that help attain better periodontal regeneration than conventional non-surgical treatment. This study aims to evaluated the effect of an Er,Cr:YSGG laser therapy in periodontally involved root surfaces and compared its efficiency of calculus removal with that of conventional hand instruments through scanning electron microscope (SEM) analysis on the instrumented root surfaces. This comparative study comprised a total of 40 specimens that were divided into Group A [20 specimens (extracted teeth)- scaling and root planing were done by Gracey curettes] and Group B [20 specimens (extracted teeth)- Scaling and root planning were performed with 1.5 Watt Er,Cr:YSGG lasers]. The intergroup comparison of the mean values of SLR between both groups statistically significant result (p<0.01) in the specimen of the Laser group (Group B). Chi-square test revealed that group B (with Laser) produced better results with less time taken for calculus removal and thus, more teeth specimens were cleaned compared to the Gracey curettes. The mean-time taken for Root Surface Instrumentation with Er,Cr:YSGG Laser was much lesser (146.50 \pm 42.53; p<0.01) than the hand scaler group. The removal of calculus index was seen to be much lower with the Er,Cr:YSGG Laser group as compared with the hand scaler group and there were small patches of calculus visible in almost all the teeth specimen of Group A (hand instrumentation). It can be concluded a clinician's expertise is to be relied upon for selecting the suitable instrumentation at a given specific condition. Since a single type of instrumentation is not efficient enough to produce exact results, hence it is advised to select a combination of procedures to yield better results.

Introduction

Laser (Light Amplification by Stimulated Emission of Radiation) is a photon beam emitted after an excited atom's stimulation that generates a coherent, monochromatic, collimated form of light. After the advent of lasers in dentistry by a dermatologist, Dr. Leon Goldman, in 1964, different variants like CO₂, Argon, Neodymium-Doped Yttrium Aluminium Garnet (Nd: YAG), Erbium-Doped Yttrium Aluminum Garnet (Er: YAG) and Erbium, Chromium doped Yttrium Scandium Gallium Garnet (Er,Cr:YSGG) lasers have been applied in treatment of both oral soft and mineralized tissue lesions. A Nd: YAG laser was first used for root surface debridement and pocket curettage in the early and mid-1990s. Several *in-vitro* and *in-vivo* studies demonstrated that the Nd:YAG laser was unable to achieve root surface debridement satisfactorily, due to carbonization or melting of hard tissue (Aoki et al., 2004), whereas Er: YAG has been demonstrated to be very useful for hard tissue as well as soft tissue applications (Kelbauskiene and Maciulskiene, 2007). Er,Cr:YSGG is the latest version of the Erbium family that was introduced in 1994

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with a wavelength of 2,780 nm and was fundamentally designed for use in various dental fields such as periodontics, endodontics and oral surgery. This is a promising tool for better periodontal regeneration than conventional non-surgical treatment (Kelbauskiene and Maciulskiene, 2007; Riaz et al., 2022). Several investigations showed that the effect of Er,Cr:YSGG laser on root surface might be comparable to those Er: YAG Laser (Dean et al., 2010; Eversole and Rizoiu, 1995; Tunar et al., 2021). Therefore, the present study evaluated the effect of an Er,Cr:YSGG laser therapy in periodontally involved root surfaces and compared its calculus removal efficiency with conventional hand instruments through scanning electron microscope (SEM) analysis on the instrumented root surfaces.

Materials & Methods

The study was carried out in the department of Periodontics and Conservative & Endodontics at Dr. R. Ahmed Dental College and Hospital, Kolkata. Scanning Electron Microscopic analysis was done at the SEM department of The Bose Institute, Kolkata. The institutional ethical committee approved the study. The collected data was analysed at The Indian Statistical Institute, Kolkata.

Study design

The present comparative study comprised a total of 40 specimens which were divided into two groups.

- Group A consisted of 20 specimens (extracted teeth), in which Gracey curettes did scaling, and root planing and this group served as control.
- Group B consisted of 20 specimens (extracted teeth), in which scaling and root planing were done by Er,Cr:YSGG Laser at a power of 1.5 watts.
- Apart from the above two groups, 2 other teeth were used for study of the baseline SEM index: 1 otherwise healthy tooth (extracted due to orthodontic reason) without any calculus; and 1 periodontally involved extracted tooth, the surface of which was covered with calculus were collected and stored separately.

Inclusion criteria

Periodontally affected teeth of extremely poor prognosis (grade III mobility and dense calculus deposits) that were considered for extraction were procured from the department of Oral and Maxillofacial Surgery of Dr. R. Ahmed Dental College & Hospital.

Exclusion criteria

Abrasive lesions present on root surfaces of teeth with root surface caries and root restorations. A tooth with root fractures, or a cementum defect that is clinically evident within six months of extraction, should receive mechanical therapy within that time frame.

Procedure

After extraction, the soft tissue attached to the tooth surface was removed carefully without any damage to the root surface, cleaned, and stored in normal saline immediately._Proximal and labial surfaces of the teeth below the cemento-enamel junction were used in the present study. Cemento-enamel junctions were marked to limit the boundary of the_experimental portions.

> Mechanical debridement (Instrumentation)

The experimental portion of the root was scaled, and root planned with Gracey curette No. 1-2, 3-4, 5-6, 11-12, and 13-14 until an absolutely smooth surface was obtained. The time for thorough calculus removal was noticed by a stopwatch (Fig. 1). In this case, the strokes were directed from the junction of cement and enamel apico-coronally. The areas were frequently flushed with water to avoid dryness of the instrumented surface (Haghighati and Arefi., 2002).



Figure 1. Image shows scaling of the tooth specimen done using Gracey curette.

Laser treatment

The Er,Cr:YSGG Waterlase Laser (MBA 36507a16100138, made in USA) by Biolase Technology fitted with a fiber-tipped handpiece (fiber beam diameter 0.6mm), was used in this study. The Er,Cr:YSGG laser emitted light of 2780 nm wavelength, 300mJ, 20Hz in a 150µs pulsed mode. The power of the Laser which was used for this study was 1.5 Watts, at 15% water, 11% air spray and the distance from the tip of the beam to the surface of the specimen was 5 cms to perform a non-contact exposure (Fig. 2). The surfaces were exposed until all the calculus was removed.



Figure 2. Image shows scaling of the tooth specimen done using Er,Cr:YSGG Laser

Sectioning procedures

A longitudinal tooth sectioning was done and the two halves were separated. After the specimens had been collected, they were stored in sterile, clean, labelled, and distilled water bottles until further procedures could be performed.

> Preparation of teeth for SEM Analysis

After experimental procedures were completed, all groups A, B specimens and healthy and diseased teeth (for baseline SEM analysis) were fixed with 1% Glutaraldehyde in 0.2M phosphate buffer (pH 7.2) at room temperature for 24 hours. Following fixation, Postfixation was performed in phosphate-buffered 1% Osmium tetroxide for 1 hour after specimens were washed twice with the same buffer. Following this, the specimens were washed twice with phosphate buffer solution. The specimens were then dehydrated using ascending grades of aqueous Ethanol solution with the following concentrations 50%, 70%, 85%, 95% and 100% for 10 minutes each. Air-drying was then performed overnight on the specimens. All the specimens were dried before they were placed for SEM analysis. The SEM study used Quanta-200, Scanning Electron Microscope (FEI-Netherland). Scanning Electron Microscope consists of 4 systems (Illumination or imaging system, information system, display system, and vacuum system). A total of 4 specimens were examined and analyzed at a given time (Fig. 3 and 4). The teeth specimens were numbered and examined in a Qunta-200, FEI (made in Netherlands), with large field detection (LFD) scanning electron microscope at 0° tilt angle, operated at an emission current of 40 µA under low vacuum condition (80Pa). The characteristics of the surfaces to be examined were focused and adjusted under various magnifications ranging from 50x to 1000x. For statistical analysis, scores to estimate the remaining calculus, loss of tooth substance, roughness, and cleaning efficiency were calculated directly from the monitor of the SEM depending upon the healthy and diseased tooth (which were used for the baseline SEM index for comparison). All the scores were noted in the SEM analysis sheet and the average scores were finally calculated for each group.

Indices used in the present study

- Time for calculus removal (TCR)
- Remaining calculus index (RCI)
- SEM Roughness index
- Loss of tooth substances (LTS)
- Cleaning efficiency
- Presence or absence of smear layer Chi-Square Test and Independent Sample 't'-Test were





Figure 3. Image depicts the SEM photograph of the presence of a smear layer and calculus on the tooth treated by Gracey curette (500x)



Figure 4. Image depicts the SEM photograph of the absence of smear layer and calculus on the tooth treated by Er,Cr:YSGG Laser (500x)

Results

The intergroup comparison of the mean values of SLR between both the groups showed a significant result (p<0.01) in the specimen of the Laser group (Group B), as is shown in Table 1. In Table 2, the Chi-square test revealed that group B (with Laser) produced better results with less time for calculus removal; thus, more teeth specimens were cleaned than the Gracey curettes. Table 3 depicts that the mean time taken for Root Surface Instrumentation with Er.Cr:YSGG Laser was much lesser $[146.50 \pm 42.53; p < 0.01]$ than the hand scaler group. The removal of calculus index was seen to be much lower with the Er,Cr:YSGG Laser group as compared with the hand scaler group and there were small patches of calculus visible in almost all the teeth specimen of Group A (hand instrumentation) (Table 4). Table 5 and 6 show that loss of tooth structure and surface roughness were observed more in Group B (with laser) and the mean values of these parameters were 1.35 ± 0.49 and $1.45\pm$ 0.51, respectively (Table 8 & 9). The mean score of the RCI index was seen to be lower in Group B (0.35 ± 0.49) as compared to that of Group A (1.00±0.00), with 't' value of 5.94 (significant), as is shown in Table 7.

Table 1. Intergroup comparison of Smear Layer Removal (SLR) means values with Hand Scaler and Er,Cr:YSGG Laser by Chi-square test

		Total					
Instrument (groups)	Absent		P	Present		0/	
	n	%	n	%	11	/0	
Hand Scaler (Group A)	5	25.00	15	75.00	20	100	
Laser (Group B)	17	85.00	3	15.00	20	100	
Total	22	55.00	18	45.00	40	100	
Chi-square value	14.54**						
	n= number ; **p<0.01						

Table 2. Intergroup comparison of the Time for calculus removal (Chi-square test)

	Time for calculus removal(sec)								
Instrument	Score 1 (60-140)		Score 2 (141-225)		Score 3 (226-310)		Total		
	n	%	n	%	n	%	n	%	
Hand Scaler	2	10.0	8	40.0	10	50.0	20	100.0	
Laser	12	60.0	6	30.0	2	10.0	20	100.0	
Total	14	14 35.0 14 35.0 12 30.0 40 100.0							
Chi-square value	12.762**								
				n= num	ber ; **	p<0.01			

Table 3. Intergroup comparison of mean time (in seconds) taken for complete calculus removal from root surface between Hand Scaler and Er,Cr:YSGG laser [Independent Samples 't'-Test]

Root SurfaceInstrumentationVariable(Control, Gr1)		Root S Instrumen Er,Cr:YS (Study Gre	Inter- group Comparison (Independent Samples 't'-Test)					
	No of Teeth	Mean±SD	No of Teeth	Mean±SD	t-value			
Time (Second/s)	20	$220.15{\pm}50.47$	20	146.50± 42.53	4.99**			
**p<0.01								

Table 4. Intergroup comparison of variation for Removal of Calculus Index (RCI) by Chi-square test

		R	Total				
Instrument	No calculus (0)		Small pa	tches (1)	n	0/	
	n	%	n	%	11	/0	
Hand Scaler	0	0.00	20	100.0	20	100.0	
Laser	13	65.30	7	35.0	20	100.0	
Total	13	32.5	27	67.5	40	100.0	
Chi-square value	19.259**; RCI= Removal of Calculus Index ; n= Number						

Instrument	No loss of tooth substance (0)		LTS Slightly loss of tooth substance (1)		Definite areas of loss of tooth substance (2)		Total	
	n	%	n	%	n	%	n	%
Hand Scaler	9	45.0%	1	55.0%	0	0.0%	20	100.0%
Laser	0	0.0%	13	65.0%	7	35.0%	20	100.0%
Total	9	22.5%	24	60.0%	7	17.5%	40	100.0%
Chi-square value	16.167**							
				n= number:	; **p<0.01	-		

Table 5. Intergroup comparison of Loss of tooth substance (LTS) by Chi-square test.

Table 6. Intergroup comparison of Roughness by Chi-square test.

Instrument	Smooth and even root surface (0)		Slightly roughened or corrugated (1)		Definitely corrugated areas (2)		Total	
	n	%	n	%	n	%	n	%
Hand Scaler	7	35.0%	12	60.0%	1	5.0%	20	100.0%
Laser	0	0.0%	11	55.0%	9	45.0%	20	100.0%
Total	7	17.5%	23	57%	10	25.0%	40	100.0%
Chi- square value	13.443**							
				n= number	; ** <u>p</u> <0.0)1		

Table 7. Intergroup comparison of Remaining Calculus Index (RCI) mean values with Hand Scaler and Er,Cr:YSGG laser by Independent Samples 't'-Test

Variable	Root S Instrum with Hai (Gro	Surface centation nd Scaler up A)	Root Su Instrumenta Er,Cr:YSG (Grouj	Intergroup Comparison (Independent Samples 't'- Test)				
	Tooth No.	Mean±SD	Tooth No.	Mean±SD	't'-value Significance level			
Remaining Calculus Index (RCI)	20	1.00 ± 0.00	20	0.35 ± 0.49	5.94**			
**p<0.01								

 Table 8. Intergroup comparison of mean values of Loss of Tooth Substances (LTS) with Hand Scaler and Er,Cr:YSGG laser by Independent Samples t-Test

Variable	Root S Instrum with Har (Grou	furface entation nd Scaler up A)	Root S Instrumen Er,Cr:YS (Grou	Inter-group Comparison (Independent Samples t- Test)				
	Tooth No.	Mean±SD	Tooth No.	Mean±SD	't'-value Significance level			
Loss of Tooth Substances (LTS)	20	0.55 ± 0.51	20	1.35 ± 0.49	5.06**			
**p<0.01								

 Table 9. Intergroup comparison of mean values of Root Surface Roughness (R) with Hand Scaler and

 Er,Cr:YSGG laser by Independent Samples 't'-Test

Root SurfaceInstrumentationwith Hand ScalerVariable(Group A)		Root S Instrumen Er,Cr:YS (Grou	Inter- group Comparison (Independent Samples t- Test)						
	Tooth No.	Mean±SD	Tooth No.	Mean±SD	't'-value Significance level				
Root Surface Roughness (R)	20	0.70 ± 0.57	20	1.45 ± 0.51	4.378**				
**p<0.01									

Discussion

The etiotropic phase or phase I of periodontal therapy plays a significant role in its success, wherein instrumentation of the root surface is of prime importance. Root surface instrumentation comprises subgingival scaling and root planing. The root planing procedure must be systematic and thorough for the complete removal of all irritants, however, the clinician should not be overzealous to invite iatrogenic root sensitivity owing to the excessive removal of tooth substance. Hence, various instruments are available and used for root planing, yet none is considered the ideal one (Lavespere et al., 1996).

Manual instrumentation is useful during root planing but uncomfortable. The pain of local anaesthesia and bleeding during the procedure with curettes make the patient anxious. Modern surgical techniques, including dental surgery, utilize minimally invasive concepts. Several procedures are available today for carrying out minimally invasive techniques. Of them, lasers are the recent version and Er,Cr:YSGG laser is the latest type.

Several studies have reported the effectiveness of Laser alone or Laser adjunct to hand instruments on root surface instrumentation in periodontal therapy. But there are limited studies on the efficacy of Er,Cr:YSGG Laser alone for root surface instrumentation. So, the present study was undertaken to see the efficacy of Er,Cr:YSGG Laser as a root planing instrumentation and to compare it with conventional hand instruments. In this study it was decided to use an *in-vitro* test model so that direct access could be obtained and root treatment could be performed by Gracey curettes and Er, Cr: YSGG laser under identical conditions. Both the methods of instrumentations were compared on the basis of time and cleaning efficiency by clinically and remaining calculus index, loss of tooth substance, presence or absence of smear layer, and roughness index by SEM study.

The present in-vitro study found that Er,Cr:YSGG Laser required the shortest time to remove subgingival calculus when compared to hand instruments. The independent sample 't'- test was used to see the difference where 't'-value was 4.99, which was statistically significant at (p<0.01) (Table 3).

Regarding the remaining calculus index, the present evaluation showed that in Group A samples (Gracey curettes), more remaining calculus was present under scanning electron microscope in comparison to Group B (Lasers) sample and which is statistically significant at p<0.01 (Table 7). Krause et al., 2007, also found a similar result. In an in- vitro study, Aoki et al., 1994, showed the similar efficiency of calculus removal by Er, YAG Laser. In another study, using histological and SEM analysis, laser scaling was found to remove calculus at a level similar to that which can be obtained using ultrasonic scaling (Aoki et al., 2000; Bertacci et al., 2021). An important finding in this study was removing the smear layer during the root planing procedure. 25% of Group A (Conventional hand scaling) samples showed the absence of the smear layer, whereas 85% of group B (Laser group) showed the absence of the smear layer. Under scanning electron microscope, open dentinal tubules indicate complete removal of the smear layer and diseased cementum. A study by Folwaczny et al., 2000 showed that laser-treated specimens showed an almost smooth surface without residual deposits or evidence of smear layers.

In contrast, root surfaces treated with manual scaling typically showed a superficial smear layer composed of necrotic cells, organic matrix and dentin particles. According to Frank et al., 1983 & Ulian et al., 2021, superficial demineralization is an important step in the natural healing process. In observation on Loss of tooth substance in the present study greater loss of tooth substance was found with Laser scaling and root planing in comparison with conventional hand instruments. By independent sample 't'-test, the t-value obtained was 5.06, which was statistically significant at p<0.01 (Table-8). Schwarz et al., 2003a & 2003b, stated that some amount of healthy cementum is inadvertently removed during the procedure of calculus and contaminated cementum removal using Laser, which may be clinically acceptable. Regarding roughness, in the present study, Er, Cr: YSGG Laser group showed a greater roughness value than Hand Instruments. In Hand scaling, most of the specimens under a scanning electron microscope showed slightly roughened areas with no obvious instrumentation marks. One of the specimens revealed corrugated areas with some instrumentation marks. whereas many specimens of Er, Cr: YSGG laser showed a corrugated appearance with instrumentation marks. In

Independent sample 't'-test compared to two groups, the 't' value was 4.378, which was statistically significant at p<0.01 (Table 9).

According to a study done by Arora et al., 2016, it was concluded that Er,Cr:YSGG laser produced severe changes in the microstructure of the tooth, especially on the root surface that affected the soft tissue attachments of the periodontium and also caused remarkable alterations in the plaque and calculus deposition. Dereci et al., 2016 demonstrated that Er,Cr:YSGG laser facilitated eradicating malodor from the oral cavity and thus improved the healing property of the periodontium compared to the conventional hand instruments.

Another similar study by Poormoradi et al., 2018 showed that the root conditioning effect by Er,Cr:YSGG laser enhanced Er,Cr:YSGG. The mean root coverage and the percentage of complete root coverage.

In this study regarding cleaning efficiency, the two groups showed similar results and there was no statistical difference between the two groups. Previous literature has documented that the detoxification and antimicrobial features of laser prove to be highly beneficial. Surface roughness of the root can be advantageous or detrimental for the periodontium, contingent on the area of the root surface that is instrumented (Aoki et al., 2004). From the present study, it was seen that each instrument had got its own merit over other. Hand instrument (Gracey curettes) gave smoother surfaces and less loss of tooth substances in almost all the cases, but the clinical time required to reach such smoothness was higher than Er, Cr:YSGG Laser. On the other hand Er, Cr: YSGG Laser showed more amount of tooth substance loss with rougher surfaces. However, it satisfactorily removed the calculus and smear layer and opened the dentinal tubules in less time than conventional hand instruments.

Conclusion

There is a great difference in the outcome of in vitro and in vivo studies. A direct vision, easy access and better control over the site of instrumentation can be achieved in case of in vitro study in comparison with in vivo study. Considering all the parameters used in this study and results obtained, it can be concluded that it is up to the clinician's judgment and expertise to select an appropriate type of instrument in a specific situation. As one type of instrument is insufficient to give accurate results, it is rational to prefer the combination of the instrument when required for a given situation rather than strictly biased toward a particular type of instrument. There remains incomplete and inadequate scientific data regarding the excellent clinical efficacy of Er,Cr:YSGG

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laser on the root surface when compared to other orthodox modes of scaling and root planing. Despite several fruitful experiments, there are also questions concerning the conclusions that influence the bacterial and cellular adhesion and their effect on clinical outcomes. Hence, further *in-vitro* and *in-vivo* investigations are suggested and required with a larger sample size to assess this aspect of the present study.

Conflict of interest

Nil

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