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Keywords: dental caries, non cavitated lesions, remineralization, wattage, time.

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Caries Inhibition of Pit and Fissure Non Cavitated Lesions in Children by Low-Level Lasers– A Clinical Study

Lt Col Sonali Sharma [α], Lt Gen SM Londhe SM, [σ], Dr. Mithra N Hegde [ρ] & Dr. Vandana Sadananda [ω]

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In vitro studies were conducted on enamel samples prior clinical study to ascertain the optimum wattage and time.

Results: Control group had 14.7 failure rate where as Test group had no failures. The optimal wattage and time of irradiation was 0.5 watts and 30 secs respectively.

Conclusions: LASER irradiation leads to caries inhibition as indicated by fall in LASER fluorescence values after irradiation.

Keywords: dental caries, non cavitated lesions, remineralization, wattage, time.

I. Introduction

Dental caries is a dynamic, progressive disease with varying phases of demineralization and increased remineralization.[1-3] To harness the remineralization potentiality of an incipient non-cavitated lesion, it becomes imperative that carious lesions are detected and diagnosed well in time and evaluated regularly for any change in the lesion activity. The carious process is a continuum disease process and to monitor it at a specific point will give an unfounded, inaccurate and unrealistic result.[1, 4]

Since over a century, fluoride has been the cornerstone caries preventive strategy. Topical fluoride application had varying success rate. Oral fluoride administration in the form of tablets though effective has documented instances of overdose leading to dental fluorosis, nausea, diarrhea, and abdominal cramps.[5, 6] Safe drinking water is a constitutionally guaranteed right in India and millions of rupees are invested in water sanitization, however, centralized systems of water supply and monitoringare ineffective. Inappropriate fluoride levels in drinking water have led to dental and skeletal abnormalities. [7,8]

Thus, an effective preventive regime would be one that brings about a change in enamel crystal and makes it more resistant to acidic challenges of the oral cavity. One such modality is the inclusion of LASERs in the prevention of caries. High powered LASERs are discontinued for caries inhibition owing to their high cost, bulky equipment and lack of any evidence-based therapeutic application.[9,10]Low powered LASER has many soft tissue oriented clinical applications. Today they are being explored as an alternative to high powered LASERs in hard tissue applications.[11]Hence this study has been designed to explore the possibility of using a low-level LASER as caries preventive and inhibitory tool in caries prone population. The findings of the analysis shall help in making standard treatment guidelines, suitable recommendations for caries prevention and inhibition, to benefit children and help in reducing the caries burden.

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II. Material & Method

1. LASER fluorescence device (DIAGNOdent pen 2190 KaVo, Biberach, Germany)
2. LASER - Aluminium Gallium Arsenide (Whitestar TM, Creation, Verona, Italy)
3. Casein Phosphopeptide- Amorphous Calcium Phosphate Fluoride (CPP-ACPF) paste (GC Tooth Mousse)
4. 37 % phosphoric acid gel (Total Etch™– Ivoclar Vivadent AG, Schaan /Liechtenstein)

III. Procedure for Clinical Study

Ethical clearance and informed consent were undertaken. Patients of either sex, between the age of 6 years to 18 years reporting to OPD having initial pit and fissure caries on first or second mandibular molar were included in the study, and its respective contralateral tooth served as control. The sample size was 102 in both groups. Oral prophylaxis was done. Suspected incipient site1 size 0 lesions, as per Mount and Hume classification, were shortlisted. Patients who scored V0, V1 or V2 in Ekstra and criteria for visual scoring for dental caries and 0, 1 or 2 in ICDAS index and R0 or R1 in the Ekstra and criteria for radiographic scoring were included in the study. But the diagnosis was more conclusive by a more sensitive method of caries diagnosis that is the LASER fluorescence method. The LASER fluorescence pen (DIAGNOdent pen 2190, KAVO, Birbech Germany) scanned the area of interest on the tooth surface. A diagnostic readout as per Lussi criteria served as a baseline reading. The LASER fluorescence method values were recorded for both test and control. The test group teeth - (Group B) were irradiated with 810 nm AlGaAsdiode LASER of 0.5 watts (WhitestarTM, Creation, Verona, Italy) for 30 seconds, followed by the application of remineralizing paste CPP-ACP F (GC –Tooth Mousse). The contralateral tooth type served as a control- (Group A), in which, after recording LASER fluorescence device values, only remineralization paste had been applied. (Fig 1 & 2) The values were then noted on the seventh day, the test tooth was irradiated again, and the remineralizing paste was applied. After that, the follow up at six-monthly intervals included serial scanning with LASER fluorescence pen and comparison with baseline reading. The readings determined if caries had been inhibited, arrested, or progressed. A radiograph was then taken to assess if caries has progressed at the end of the study. (Fig3) The results were then tabulated and computed. Statistical analysis was carried out. (Graph 1)

IV. Procedure to Ascertain Optimal Wattage

Thirty freshly extracted teeth, which were caries-free and without any structural defect, had been selected. They were then sectioned mesiodistally to obtain 60 samples, and the tooth section was coated with nail varnish to obtain windows of 3mm X 3mm on the facial surface, which aided in standardizing the sample's dimensions. The samples were divided into ten groups for each power setting which was being evaluated. Each group had six samples each. The power settings range from 0.1 to 1 watts was selected. For each sample, the preoperative value of the exposed tooth section was evaluated by LASER fluorescence method, and this served as control. The tooth section was surface treated with 37% acid etchant gel for 20 seconds. The LASER fluorescence device's values were then noted. Then they were irradiated with aluminium gallium arsenide LASER of 810nm for 30 sec, and each group were then individually evaluated for different power settings power setting from 0.1 watts to 1 watt. The power setting which were evaluated were 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1. After irradiation for 30 seconds, the LASER fluorescence method reading was again noted. The results were then tabulated and computed. Statistical analysis was carried out. (Graph 2)

V. Procedure to Ascertain the Time of Irradiation

Thirty-six samples surfaces were prepared as before and evaluated with a LASER fluorescence device to obtain the baseline values. The samples were then surface treated for twenty sec with 30 % phosphoric acid, to simulate surface demineralization. The LASER fluorescence device was then used to record the values of demineralized samples. The samples were then divided into six groups of six samples each. The sectioned surfaces of the samples were thereafter irradiated with aluminium gallium arsenide LASER of 0.5 watts for 5 secs, 10 secs, 15 secs, 30 secs, 45 secs, and 60 secs, respectively. The LASER fluorescence device recorded values post-irradiation. Statistical analysis was carried out. The exposure time in which the post-irradiation values came closest to preoperative values was then recoded as the optimal time of exposure. (Graph 3)

VI. Summary of Results

The results are being summarized in the graphs (1-3) and tables (1).In this clinical study, the caries preventive and caries inhibitory potentiality of low-level LASERs was evaluated and was compared with the conventional method, i.e, application of remineralization paste. LASER fluorescence was then used for monitoring the effect of the LASER, and remineralizingagent on incipient non-cavitated carious lesions. Based on the result it was finally concluded that the parameters of the low-level LASER of 810 nm, i.e wattage and time, which brings about the optimal results, were 0.5 watts and 30 secs, respectively. In the

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test group, there was a 100 percent success rate. The control group had 15 failures. The results were statistically significant.

VII. Discussion

Dental caries is the most prevalent oral pandemic disease of dental hard tissues. Acevedo et al. observed that 33 percent of children had non-cavitated lesions between age 11-15 years.[12] SK Jain et al. in an epidemiological study, found that currently, there is a change in pattern and trend of dental caries, and thus, there is an increase in the number of non-cavitated lesions as compared to the cavitated carious lesion, and it is more common at the age of 14 years.[13]The tooth which is most prone to dental caries is the first mandibular molar, especially young permanent molars. Hence in this study, the sample population was drawn from six years to eighteen years, and categorically, the first and second molars were included in the study. The sample population was equidistributed amongst both genders. The test and the control are in the same person, and test tooth is the contralateral tooth type of that of the control; thus, making challenges of the oral environment similar.

The performance of caries detection methods will be assessed by considering two fundamental parameters: reproducibility and validity. The visual and tactile technique depends on the clinical acuity of the clinician, and it is not reproducible even for the same clinician leave alone for other observers. 30 to 40% percent of mineral loss must take place before any carious lesion can be detected on radiograph.[14]Hence a sensitive diagnostic adjunct is required. Current diagnostic aids use the alteration in LASER fluorescence, reflectance, electrical conductance or impedance, and ultrasound transmittal properties of enamel concerning demineralization and remineralization during the continuum of a carious lesion over a lengthy period.[14]

Studies by Lussi et al., Novaes TF, Atrill, Fung L, Burin et al., have all inferred that LASER fluorescence methodis a predictive diagnostic tool and is more sensitive than the traditional method of visual and radiographic examination and. It not only detects early and hidden caries but also aids in longitudinal assessment and monitoring of caries activity.[15-19]Hence in this study LASER fluorescence was selected as an assessment tool to study the effect of low-level LASER on the enamel surface.

At the ultrastructural level, the enamel is formed of closely apposition edhydroxylapatite crystals. Outer 50-100 microns of enamel layer are heavily impregnated with fluoride. Below this zone, there is more concentration of carbonate ions and less of fluoride ion concentration. The super saturation at the surface layer chokes out fluoride from permeating into subsurface layers. The incipient carious lesion and non-cavitated lesions are thus characterized by intact surface layer due to more fluoride in the 50-100 microns of the tooth's outer surface and subsurface dissolution due to decreased fluoride ion concentration.[1, 5-8,20]

The advancement of caries may be impeded or foreshortened using risk modifiers. Fluoride, an efficacious therapeutic risk modifier, present in saliva and biofilm does not affect the biofilm formation and sugar metabolism but reduces demineralization by concurrent precipitation of fluoro-hydroxyapatite, a phase more stable than hydroxyapatite at any given pH. Thus, remineralization is a consequence of fluoride application, not an effect of it.[1,5-8,20] Ten cate et al. have inferred that to remineralize subsurface lesions 150microns deep 5000ppm of fluoride is required, which is not practical and excessive doses can lead to toxic side effects and even be fatal.[20]

Hence an alternative remineralizing paste that could substantiate the effect of fluoride is sought. Casein phosphopeptide amorphous calcium phosphate plus fluoride (CPP-ACP F), has been shown to increase fluoride's uptake into plaque and subsurface enamel by providing bio-available calcium ions, phosphate ions and fluoride ions in the correct molar ratio to form fluorapatite.[21] Hence in this study, we have used CPP ACP F as remineralizing paste both in test and control group.

The limitations of remineralizing studies are that most of the evidence-based studies are invitro and is very different when translated to the complex dynamic oral milieu. The other deterrents for remineralization to occur within the body of a subsurface lesion are calcium and phosphate ions must penetrate the surface layer of the enamel. The highly mineralized and charged nature of the surface layer poses a challenge for ion penetration. To infiltrate to subsurface carious layer a very high molar concentration, of approximately 5000 ppm of fluoride ions, are needed. [1, 20]CPP ACP products if, ingested in significant quantities, will cause side effects. The potential risk increases with patients who have allergic diathesis. The effectiveness of CPP ACP in the remineralizing subsurface lesion isquestionable.[5, 7-8, 20-23] The other drawback of incorporating a paste based remineralization strategy is that a regular and repetitive replenishment of paste to overcome the loss of calcium and phosphate ion. Thus, an effective remineralizing protocol would be one that brings about a change in enamel crystal and makes it more resistant to acidic challenges of the oral cavity.

LASERs, since its introduction in the 1960s, have been studied and piloted to bring about caries prevention on inhibition by increasing acid resistance.[9-11]Ana et al deduced that under specific conditions LASER irradiation can change crystallographic properties of apatite crystals and increasing the acid resistance of lased tooth as well as increased fluoride

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uptake.[24] The LASERs which have been investigated in the past for caries inhibition are CO2, Nd:YAG, Er:YAG, ErCr:YSGG. These LASERs had innumerable encumbrances like that of exorbitant price tag, unwieldy, heavy cumbersome and besides, the result obtained was also highly debatable. Additionally, the majority of the experiments were lab-based and further the diode LASER had never been evaluated as a caries inhibitory technique. Clinical studies at best have been pilot studies or short term. [9-11, 24-26]

Sant'anna has conducted an FT Raman spectroscopy study to evaluate low-level diodes as a caries inhibitory tool.[27] Thus, in this clinical study, we have evaluated low-level LASER therapy as a caries inhibitory tool. Comparing test and control it was inferred, to begin with, that the preoperative LASER fluorescence values for all were in the same range. LASER fluorescence values over time have increased in control group thereby indicating demineralization but decreased in test group thereby indicating remineralization, and thus it can be deduced that LASER irradiation brings about changes in the crystallographic properties of enamel apatite and increases the uptake of CPP ACP F paste much more than when the paste is used alone as in control. Thus, our study is confirmation with other reviewed in vitro studies and short term pilot studies. [24-27,29] (Graph 1)

Today, aluminium gallium arsenide LASER is currently being investigated as an alternative to high powered LASERs. The hypothesized mechanism of action is that these wavelengths selectively target and remove carbonate ions from hydroxyapatite crystals which results in increased acid resistance of enamel. Additionally, the altered mineral has greater uptake of topically applied fluoride and leads to remineralization of non-cavitated lesions.[11]Thus, in this study, we have selected low-level LASER to increase the acid resistance and the remineralizing potentiality of incipient non-cavitated lesions.

For any clinical procedure using a LASER, its optical interaction with enamel and dentin must be thoroughly understood. The LASER interaction with dental hard tissues depends upon irradiation parameters such as wavelength, pulsed or continuous emission, pulse duration, repetition rate, beam spot size, and delivery method. All reviewed studies agreed upon the caries inhibitory role of LASERs, but none ascertained the parameters to bring about the desired result. [26-30] Thus, in vitro studies were first conducted to determine an optimum wattage and exposure time. The least amount of deviation there is from control i.e. the closest the LASER fluorescence value to that of the control, the more successful is the treatment. Thus 0.5 watts and 30 secs is the most optimal value as it has shown LASER fluorescence values closest to the corresponding control. The results are statistically significant. (Graph 2& 3)

Another challenge in this study was to determine the sequence of the protocol of LASER and remineralizing paste application. Carounanidy has opined that the LASER activated fluoride uptake method can enhance the remineralization potential of an acid challenged tooth.[3] Further, the scientific world is divided on whether fluoride application should precede or follow LASER irradiation.[28-29] In the present study, on the test group, the fluoride application succeeds the LASER treatment. There has been a fall in the LASER fluorescence values in the test group (Group A)which was irradiated by LASER followed by fluoride application. This decrease in LASER fluorescence numerics was seen throughout the follow-up period. However, in the control group (Group B) in which only CPP ACP F was applied, the digital numerics initially fell and then again increased. (Graph1) Thereby, indicating that LASER irradiation followed by remineralizing paste is more effective than remineralizing paste used alone.

Each study has its limitations, and no research is flawless, nor should it be expected, as is envisaged in this study. The most reliable way to assess the clinical impact of a novel technique is through its endpoint, such as reversal of carious lesion or survival of the tooth after LASER caries inhibition. However, this standard may be impractical for the evaluation of new therapies, because long periods with large sample size are required for these clinical endpoints to be objectively achieved. A surrogate endpoint can expedite the trial process. A surrogate endpoint is identified as a biological marker that is designed to proxy for a clinical termination. Clinical endpoints, on the other hand, are distinct, direct measurements that reflect how a patient responds to treatment. Clinical endpoints are the most credible characteristics used in the assessment of the benefits and risks of a therapeutic intervention. In this study, we have used a clinical endpoint over a long follow up of eighteen months.[30] Since dental caries is a dynamic disease, a surrogate endpoint should be determined to evaluate the validity of this novel technique.

Another limitation is that enamel chemistry studies should also be included to support promising results. The synergism of the low-level LASER and remineralizing paste should be studied at a molecular level.

Thus, this novel concept of combining low-level LASERs with remineralizing paste seems like a promising preventive and inhibitory modality for non-cavitated initial caries provided that they are diagnosed early. For early detection, a LASER fluorescence device is found to be a reliable and accurate tool.

VIII. Conclusion

1. The optimal wattage and the optimal time of irradiation, which could bring about the caries

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inhibitory potentiality of low-level LASERs clinically were 0.5 watts and 30 seconds respectively, in the age group 6 years to 18 years.

2. The test group, in which the teeth had been irradiated with low-level LASERs followed by application of remineralizing paste i.e. CPP ACP F, had a 100 % success rate. The control group, in which only remineralizing gel had been applied, had a failure rate of 14.7 %, which necessitated operative intervention.
3. The low-level LASERs increased the uptake of remineralizing paste in irradiated teeth. Thus, LASERs followed by remineralizing paste seems to be the appropriate sequence to be followed.
4. Remineralizing paste alone did not have sufficient long term caries inhibitory potentiality.
5. The low-level LASERs can be used as a viable option to the conventional method, as an accepted modality in preventive and caries inhibitory regimen of dental caries in routine dental practice.
6. For subsurface incipient carious lesions, the LASER fluorescence device is found to be a reliable and accurate diagnostic tool.

- Conflict of interest statement: The authors declare there has been no conflict of interest.
- Ethical Approval: Institutional Ethical clearance has been obtained.
- Informed Consent: Informed consent was obtained from all individuals participating in the study.

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Table 1

Succes & Failure in Control

	Frequency	Percent
Failure	15	14.7
Success	87	85.3
Total	102	100

Sex	Frequency	Percent
Male	51	50
Female	51	50
Total	102	100

		GENDER		Total	Pearson Chi-Square	p-value
		Male	Female			
Failure	Failure	1	14	15	13.21	<0.001
	Success	50	37	87		
Total		51	51	102		

Failure	N	Mean	Std. Deviation	t-value	p-value
Failure	15	14.4	3.869	1.68	0.096
Success	87	12.7471	3.458		

The percentage of success in control is 85.3 percent and failure is 14.7 percent. The sample population is equally represented by both the genders. Correlation of gender and failure shows that 14 failures are seen in females and 1 failure in males. This inference is statistically significant. Age has no bearing on the outcome. The comparison for test group is not computed as there are no failures in test group.

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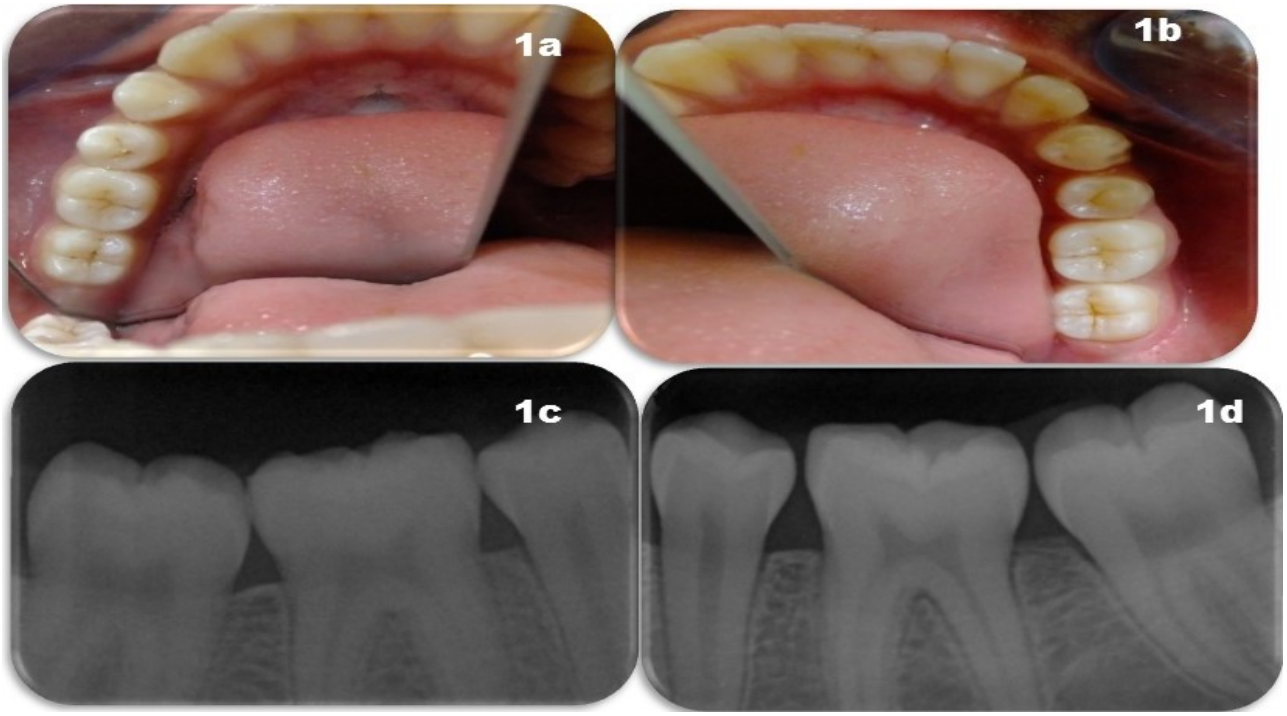

Fig. 1: Preoperative

Fig.1a: Preoperative Clinical Picture of Control 47; Fig.1b: Test 37 Fig.1c: Radiograph Control 47; Fig.1d: Test 37



Fig. 2: Clinical Procedure

Fig 2a: Laser Fluorescence Preoperative Value of 21 in Control 47; Fig 2b: Laser Fluorescence Preoperative Value 26 in Test 37, Fig 2c: Laser Irradiation, Fig 2d: Laser fluorescence value 25 in Test 37, Fig 2e: Remineralizing paste applied

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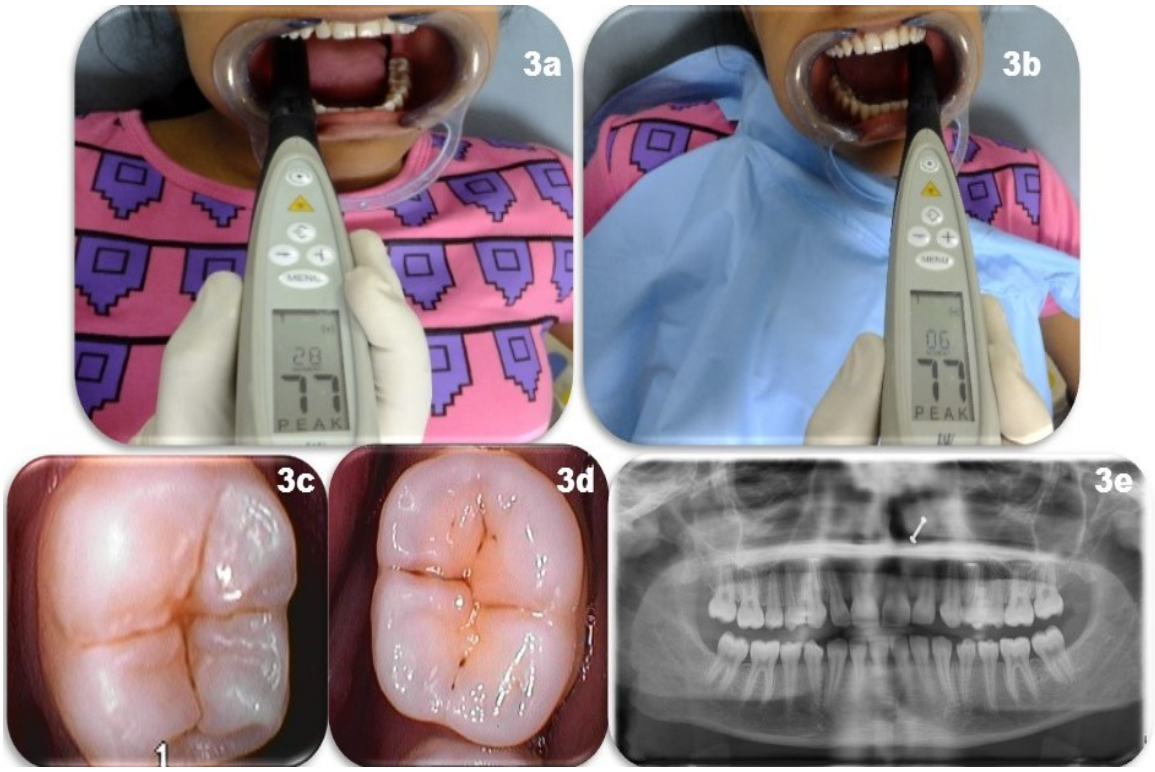
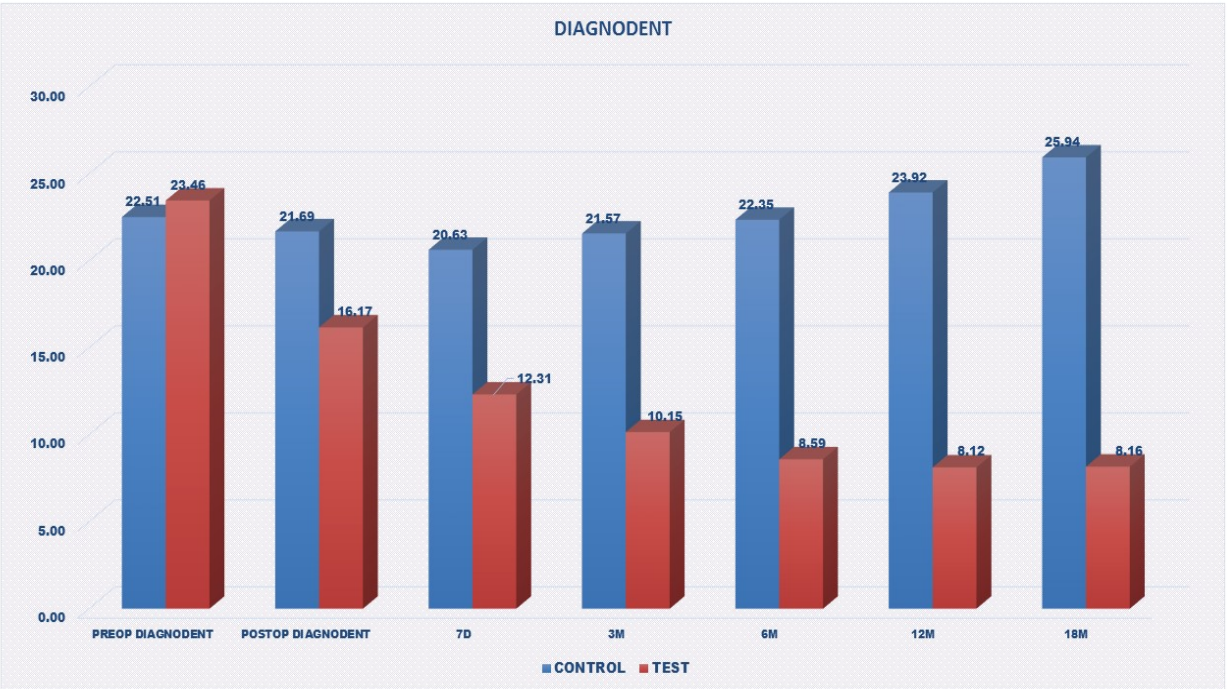


Fig. 3: 18 Month Follow Up
Fig 3a: 18 month follow up Laser Fluorescence Values of Control -28;
Fig 3b: Test- 06; Fig 3c: Tooth no 47; Fig 3d: tooth no 37; Fig 3e: OPG

GRAPH 1



GRAPH 1: Comparison of Laser Fluorescence Values

Laser fluorescence values over time have increased in control group thereby indicating demineralization but decreased in test group thereby indicating remineralization.

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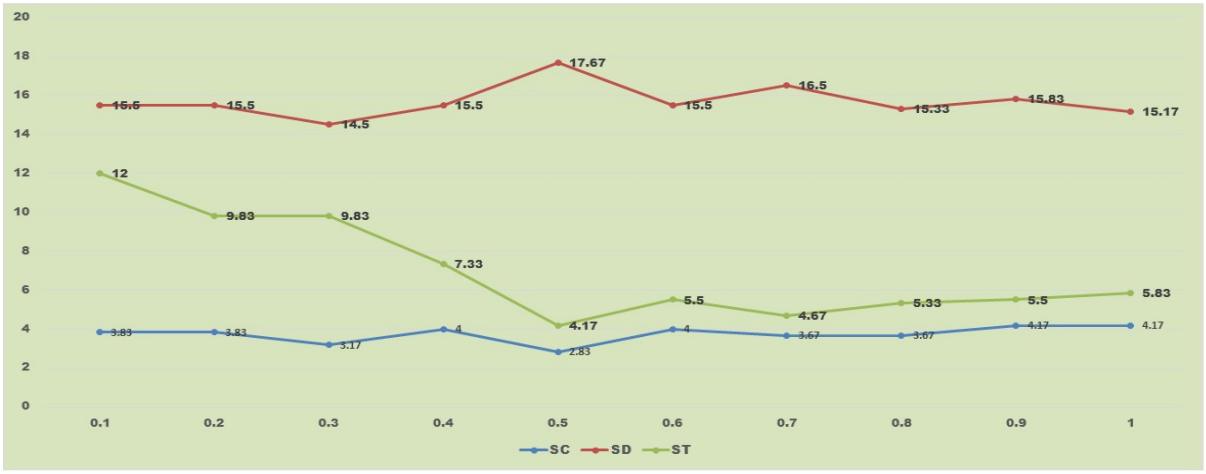
GRAPH 2

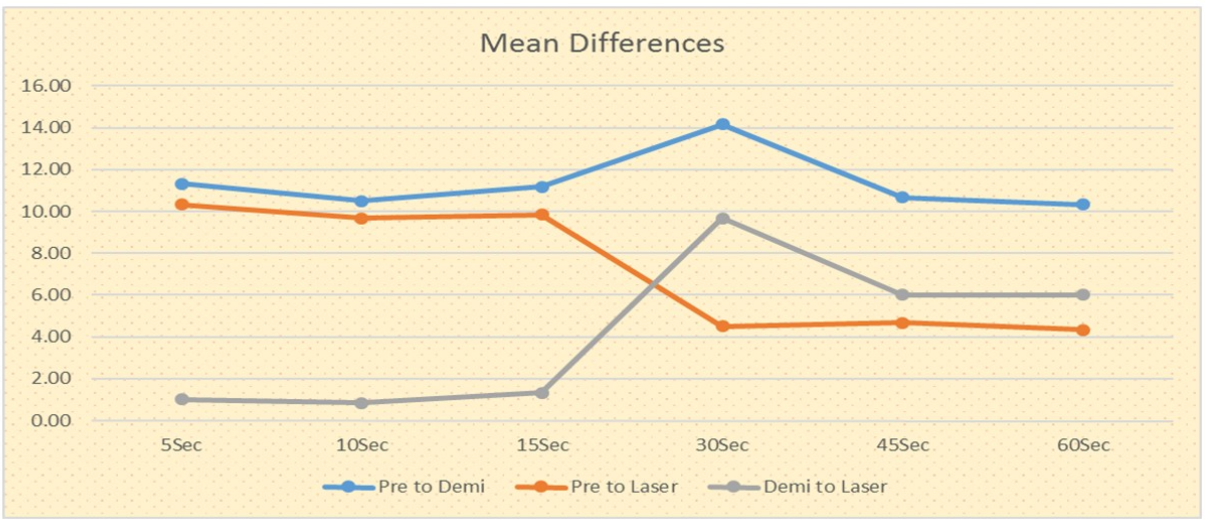


GRAPH 2: Optimum Wattage

There is an inverse correlation between increase in power and laser fluorescence i.e Laser fluorescence method values of irradiated surface. Indicating that the increase in wattage alters the tooth and makes it closer in its configuration in tooth structure to that of control, thus the values of control and treated group i.e laser irradiated by laser. Beyond 0.5 watts there is not much difference between control and laser irradiated values by laser fluorescence

SC Control; SD – Demineralized; ST - Treated

GRAPH 3



GRAPH 3: Optimal Time of Irridiation

There is significant difference between control and demineralized samples and treated and demineralized samples. The treated samples have laser fluorescence values closer to that of control. The time duration of laser irradiation which brings about optimal changes is 30 sec.

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