EVALUATION AND COMPARISON OF THE MORPHOLOGICAL AND HISTOPATHOLOGICAL CHANGES INDUCED BY ER:YAG LASER AND BURS ON ENAMEL, DENTIN AND PULP TISSUE

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CERTIFICATE

This is to certify that this dissertation titled " **EVALUATION AND COMPARISON OF THE MORPHOLOGICAL AND HISTOPATHOLOGICAL CHANGES INDUCED BY ER:YAG LASER AND BURS ON ENAMEL, DENTIN AND PULP TISSUE**" is a bonafide dissertation performed by **SHAIK MOHAMED SHAMSUDEEN.S.S** under our guidance during the postgraduate period 2009-2012.

This dissertation is submitted to THE TAMILNADU DR. M.G.R MEDICAL UNIVERSITY, in partial fulfillment for the degree of **MASTER OF DENTAL SURGERY** in **ORAL PATHOLOGY AND MICROBIOLOGY, BRANCH VI**. It has not been submitted (partial or full) for the award of any other degree or diploma.

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CONTENTS

Light Amplification by Stimulated Emission of Radiation **(LASER)** is unidirectional, monochromatic and coherent in nature that covers electromagnetic spectrum between 0.1 to 10 µm. Laser was applied in dentistry by Leon Goldman in 1964[1]. Since then it has been modified and used for dental caries removal and prevention[2], etching of the tooth surface[3], preparation of teeth for restorative purpose[4], treatment of dental hypersensitivity[5] and soft tissue surgeries[6]. Laser has the advantage of being painless, time saving, precise and easy to manipulate. In soft tissue surgeries, it gives excellent haemostasis[7], improves visibility and causes minimal damage to adjacent tissues[8]. It reduces the post operative swelling, pain and infection[9] and also reduces scarring and wound contracture[10].

Dental lasers are classified as either soft tissue or hard tissue lasers based on the tissue applications. When laser light interacts with tissues, they get absorbed, reflected, scattered or transmitted[11]. The effect of laser irradiation depends on the energy absorbed by the tissue, the temperature between the energy deposited and conducted away as heat. The energy absorbed by tissues can have absorbed photochemical, photothermal, photomechanical, photoelectrical effect or a combination of these effects[12].

The soft tissues lasers are used in dentistry have photothermal and photochemical effects. The CO_2 and diode lasers are commonly

used for soft tissue procedures like excision. The semiconductor diode lasers used for soft tissue surgeries.

Conventional methods of cavity preparation with low and high speed hand piece and burs have the advantage of precision cutting for clearly identifiable margins and good tactile perception. However they are noisy and cause uncomfortable vibrations and stress for patients[13]. These disadvantages have led to search for new techniques for dental hard tissue removal.

At present, several laser types with wavelength in the middle infra red region of the electromagnetic spectrum are available for cavity preparation and caries removal. Examples are Er: YAG (Erbium: Yttrium Aluminium Garnet), Er: YSGG (Erbium: Yttrium Scandium Gallium Garnet) and Er Cr: YSGG (Erbium Chromium: Yttrium Scandium Gallium Garnet). The efficiency of ablation is greatest with the Er: YAG laser. When laser comes into contact with the hard tissue, they interact with water which rapidly absorbs the heat. Simultaneous intense absorption of energy within microspores of hard tissue also heats the water instantaneously, which changes its phase to vapour increasing the pressure inside the microspore. This effectively breaks the hydroxyapaptite structure leading to micro explosion that eject dental hard tissue away from the surface.[14].

Removal of tooth structure in cavity preparation by laser is simple and advantageous. The surface of the cavity at micro level

varies with the use of conventional air powered burs and lasers. The type of restoration may influence the method that is appropriate. Amalgam restorations requires clear identifiable margins with obtuse cavosurface, while resin based restoration requires absence of smear layer with increased surface roughness, making the bur appropriate for amalgam and lasers for resins.

This study was done to ascertain the surface changes and pulpal response to the cavity preparation using Er:YAG laser and diamond bur.

Aim:

The aim of the study was to evaluate and compare the morphological and histopathological changes in enamel, dentin and pulp tissue of the teeth treated with Er: YAG laser, conventional diamond burs and tungsten carbide burs.

Hypothesis:

Null hypothesis:

Enamel, dentin and pulp will not undergo morphological and histological alterations, when cavity preparation is done using Er: YAG laser and conventional bur techniques.

Alternate hypothesis:

Enamel, dentin and pulp will undergo morphological and histopathological alterations when cavity preparation is done with Er:YAG laser and conventional burs.

Objectives:

1) To study and evaluate the following morphological alterations in the cavity of teeth prepared using Er: YAG laser and conventional burs.
   (a) Alterations of the cut surface of the cavity walls
   (b) Surface microcracks
   (c) Dentinal tubule changes such as opening of tubules and presence of smear layer.

2) To study and evaluate the following histopathological changes in the pulp of the teeth prepared using Er: YAG laser and conventional burs.

   (a) Odontoblast cell changes such as degeneration and shrinkage of the odontoblastic layers

   (b) Inflammatory cell infiltrate

   (c) Vascularity of pulp tissue

ARMAMENTARIUM:

For cavity preparation:

Mouth mirrors- Size-5

Explorers

Tweezers

Hand piece –NSK No.G 7100145, Japan

Speed -300000 rotations per minute with water coolant.

Burs – Diamond Burs -No.SI 47

Tungsten carbide bur – No. FG 245

Suction tips.

Laser equipments:

Model Fidel Plus III- Er: YAG-Fotona, Germany

Scanning electron microscope:

EDS detector system Super Dry, Hitachi

Hard tissue microtome:

Leica SP 1600, Saw Microtome

Rotating speed: 600 RPM, Germany

Materials

10% Formalin LR

10% Formic acid LR

Distilled water LR

Haematoxylin and Eosin stain LR

METHODS:

Study subjects:

Inclusion criteria: Patients undergoing extraction of permanent teeth (premolars and canines) for orthodontic purpose.

Exclusion criteria: Decayed teeth, Restored teeth and malformed (any pathosed) teeth are excluded from the study.

Sample:

Sample size: 40 extracted teeth.

Procedure:

Ethical clearance was obtained from the Institutional Review Board. Informed consent was obtained from the patient after detail explanation of the study to the patient. Patients were randomly divided into three groups.

Group 1 – The cavity was prepared using Laser. (20 samples)

Group 2 –The cavity was prepared using diamond burs. (18 samples)

Group 3 – The cavity was prepared using tungsten carbide burs (2 samples).

The tungsten carbide bur was used in two samples which used as a comparison for the SEM study only.

Cavity preparation:

Conventional Class I cavity was prepared with 1.2mm in width which extended beyond the dentino enamel junction[15]. The

cavity was prepared on the occlusal surface of the teeth to be extracted. After cavity preparation the teeth were extracted under local anaesthesia.

Group 1- The cavity was prepared using Laser.

Laser specifications:

Type of laser: Er: YAG laser

Wavelength: 2970nm.

Laser power: 600 Watt (W)

Laser energy: 300 milli joules(mJ)

Water spray: 8

Air spray: 4

Hand piece: R02

The patient interaction protocols and safety precautions were followed including the use of eye goggles and sufficient water coolant.

Group 2- The cavity was prepared using conventional diamond burs No.SI 47 in 18 samples. The initial cavity was prepared using diamond burs by no. 12. Then the cavity was extended using straight fissure bur SI 47. The speed of the air turbine was 300,000 rotations per minute with the water coolant.

Group 3- The cavity was prepared using tungsten carbide bur. FG-245 in 2 samples with standard methods. [15]

Time required for cavity preparation:

The time taken for cavity preparation was determined to nearest minutes in all instances.

Morphological analysis of prepared cavity:

(a)Using Scanning Electron Microscope:

Samples – 12 teeth.

Group 1- 5 teeth

Group 2- 5 teeth

Group 3- 2 teeth

After extraction, the teeth were immediately stored in distilled water till processing. The teeth were dehydrated with grades of 50%, 70% and 90% alcohol and dried in vacuum chamber. The electron microscope utilized an EDS detector system. After drying tooth in the vacuum chamber the prepared tooth sample was coated with fine gold particles. The sample was loaded in the SEM and connected to the computer software in which the image was captured. Photomicrograph was obtained for morphological analysis. The cavity margins, the cavity surface was analysed between the groups as regular or irregular in nature. The presence of smear layer, surface roughness was graded as mild or severe, microcracks, glazing and craters was marked as present or absent.

(b)Using hard tissue microtome:

The hard tissue microtome used was Leica SP 1600 with speed of 600 rotations per minute. The section thickness can be adjusted from 100 microns to millimetre.

Samples – 10 teeth.

Group 1 -5 teeth.

Group 2- 5 teeth.

After extraction, the teeth were immediately stored in distilled water till processing. The teeth were mounted in acrylic base and sectioned in the hard tissue microtome with the section thickness of 100µm. The teeth were mounted in the slide using DPX and studied under microscope. The following parameters were observed like the structure of enamel, enamel lamellae running from the surface and tufts, a leaf like structure from dentino enamel junction, dentino enamel junction, the dentin structure and the presence or absence of dead tracts.

Histopathological analysis of pulp tissue:

Sample -18 teeth.

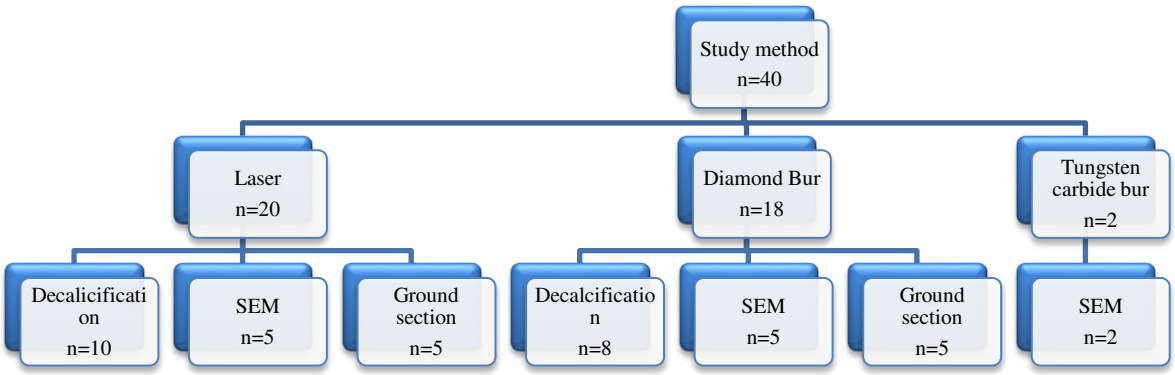
Group 1-10 teeth.

Group 2 –8 teeth.

The extracted teeth were kept in 10% formalin solution to fix the pulp. The teeth were sectioned with safe sided diamond disc at a distance of 2mm from the apex. The teeth were decalcified using

10% formic acid. After adequate decalcification and tissue processing, the teeth were embedded in paraffin wax, sectioned, stained with haematoxylin and eosin. Then the sections were mounted on the slide. They were observed under light microscope to observe the presence of inflammatory cells and odontoblastic degeneration. The arrangement of the fibres in the pulp tissues was observed. Vascularty of the pulp tissue was also observed.

Flow chart of the study sample:



Statistical Analysis:

Data entry and analysis was done using Statistic package for Social Service Package (Version 11.0). The statistical analysis for the gender distribution of the samples was done in percentage. The morphological and histopathological analysis was done in percentage. The descriptive analysis was used. The time difference between the groups was done by one way ANOVA. A $p \leq 0.05$ was taken as significant.

Development of Lasers in dentistry

The first laser used in medicine was a ruby laser by **Maiman in 1960**[16] (wavelength 694 nm). The ruby laser is a solid state laser with a ruby rod as the lasing medium. The histological effects of the ruby laser on the dental pulp were first reported by **Taylor, Shklar G, Rober F in 1965** [17]. The lab animals were exposed to ruby lasers in the energy range of 35-55J. They studied the dental pulp. It showed extensive haemorrhage, necrosis as well as disruption of odontoblastic layer. Damage to the adjacent teeth and surrounding structures was also reported as a result of scattering of the laser beam. They concluded that ruby laser can damage the pulp. This was later confirmed by **Adrian JC, Bernier JL, Sprague WG in 1971**[18], that application of ruby laser resulted in thermal damage to vital tissues.

The first gaseous laser used for surgery was the carbon dioxide (CO_2) laser. **Katola Laine E, Taina T in 1973**[19] studied the ability of CO_2 laser to induce resistance to acid penetration of enamel in attempts to use this laser for the sealing of pits and fissures. They were unsuccessful due to the excessive high temperature generated of the process. **Melcers J, Chaunette F, Melcer F et al in 1984**[20] worked on the clinical application of the CO_2 laser for the vaporization of caries. They further reported successful treatment of 1000 patients in clinical trials of caries removal. They further concluded that CO_2 laser could induce

secondary dentin formation, sterilization of dentin and exposed pulp. The CO_2 laser had many advantages in the soft tissue procedures like improved haemostasis.

The first report of dental application of the Nd:YAG laser to vital oral tissue was by **Yamamoto H, Ooyg K in 1974**[21].They observed that Nd:YAG laser was an effective tool for inhibiting the incipient caries both *in vitro* and *in vivo*. This laser proved useful for minor dental surgery, with a good coagulator effects. Application as a substitute for the "dental drill" attracted much public attention, but was not a great success. The drawbacks were due to the application of a dark dye to the tooth structure before drilling and because the process was very slow.

With the advent of the Er: YAG lasers in the late 1990s that application of lasers for removal of hard dental tissue became more widely adopted. The Er: YAG laser was tested for its ability to ablate dental hard tissues by **Gimbel in 2000**[22]. Enamel and dentine cavities were successfully prepared using the Er:YAG laser. Since then this laser has been used for caries removal and cavity preparation, soft tissue minor surgery and scaling. These versatile lasers can penetrate dental hard tissue at almost the same rate as a high-speed turbine drill. A major advantage is that little or no analgesia is necessary.

The most recent additions to the dental laser family are the diode lasers. These typically emit at wavelengths of 808, 940 or 980

nm, with outputs ranging from 3-7 watts. The light is transmitted through an optical fibre. They are commercialised for soft tissue Management[23] but are also used for endodontic decontamination[24]and sulcular debridement[25]. The diode lasers are much smaller than Nd: YAG and Er:YAG lasers and less expensive.

Effects of Er: YAG laser on Enamel and Dentin: SEM study

Hossain M, Yamada Y, NakamuraY et al in 2003[26] analysed the surface roughness and the surface morphology of the cavities prepared by Er: YAG laser *in vitro*. In each of the 30 human extracted teeth, two shallow cavities were prepared. One was prepared with Er: YAG laser on buccal surface, and other on the lingual surface with high speed bur. The surface roughness was analysed using colour laser three dimensional 3D microscope. It showed significant increase of roughness in the lased tooth. The SEM study showed that the lased cavities had irregular surface with absence of smear layer and opening of dentinal tubules. The cavities were also subjected to micro leakage test after regular composite restoration. It showed no significant difference between the lased and bur cavities. Crosscut sections of the cavities with no micro leakage showed no gap at the interface. They concluded that laser cavity was ideal for a composite restoration because of its good adaptation and increased surface roughness.

Jucaira in 2004[27]assessed the Er:YAG lased cavities prepared in the enamel of primary teeth *in vitro*. The rounding of the

cavosurface margin and the angle of the concavity at the base of the cuts were evaluated in 12 exfoliated teeth. Radiation was perpendicularly applied for 10s at a focal distance of 13mm with pulse repetition rate of 10 Hz and energy set at either 200mJ or 300mJ. Cross section of the cut surface was examined by SEM. It showed that there was a reduction in the concavity of the angles of the cavity. When the energy of the laser was increased, the cavosurface angle was altered and this leads to reduced microleakage of the restorative material.

Rubens CR, Patricia M, Otsuki M et al in 2005[28] studied the dentinal morphological changes irradiated with Er: YAG laser with different parameters such as energy output, contact or non contact mode and incidence angles. They used 45 dentin disks obtained from molars. They divided the dentin disks into 9 groups with different energy levels of 150mJ, 90mJ, 70mJ and contact vs. non contact mode, incidence angle 45^0 and 90^0. The SEM analysis revealed that different groups have different dentin morphology. The contact mode with higher energy density showed rough surface and opened dentinal tubules. The other groups showed fewer such changes. The inter tubular dentin showed more ablation than the peritubular dentin. An incidence angle of 90^0 used resulted in changes like greater dentin ablation forming micro holes than the 45^0 incidence angle with other parameters remaining the same. Hence it was concluded that operators should be aware of the best irradiation settings for specific clinical application.

Hussein A, Mclntyre J, Abbott J et al in 2006[29] studied the ultra structure of the dentin treated by Er: YAG laser in 21 extracted molar teeth. The coronal crown was sliced to expose the dentin. The dentin was irradiated with different energy output with maximum 600mJ and minimum of 40mJ. The highest frequency was 50Hz and minimum of 2Hz. These were used in very short pulse, short pulse, long pulse and very long pulse. The samples were prepared for SEM analysis. Within the Er:YAG laser parameters tested, several characteristic features were evident on the dentin surface. The features included were absence of smear layer, open dentinal tubules, micro roughness and crater like appearance. It was found that the inter tubular dentin is more prone to ablation than peritubular dentin. SEM analysis indicated that the optimal parameters which produced the cleanest and smoothest treated dentin surface were around 250mJ energy level with very short pulse mode and 10 Hz frequency.

Nishimoto Y, Otsuki M, Yamauti M et al in 2008[30] studied the dentin ablation using Er: YAG laser at various pulse durations on ten human dentin disks. The dentin disks were irradiated at 1pulse per second for 3 seconds at pulse duration of 100-150 milli sec (ms) with 150mJ. The depth and diameter of the ablated dentin were measured and the ablation volume was estimated. The depth of the removed dentin increased and the diameter of the spot decreased without a change in the estimated volume at increased pulse

durations. SEM observations showed that there were no morphological differences when the pulse duration changed. The cross sectional specimens showed dark layers under the irradiated surface. These showed the influence of pulse duration in caries removal and in the adhesion of lased dentin to adhesive restoration.

Effects of Er:YAG laser on Enamel and Dentin: TEM study

Ceballos L, Toledana M, Osorio R et al in 2002[31] compared Er: YAG laser irradiation and acid etching on dentin surface *in vitro*. Superficial or deep dentin from human molars was: (a)acid etched with 35%phosphoric acid (b) Irradiation was done with Er: YAG laser at 2Hz and 180mJ with water cooling and(c) combined laser and acid etch. Then the composite was bonded to prepared surface and studied in Transmission Electron Microscope (TEM) for bond strength. Acid etching alone yield shear bond strength values that were significantly higher than those achieved with laser ablation or in combination with acid etching. Hence they concluded that the laser bonded surface had modified layer which was not as good as acid etching for adhesion of materials.

Effects of Er: YAG laser on dental hard tissues:

Dastalova T, Jelinkova H, Krejsa O et al in 1998[32] compared the etching effects of the Er: YAG laser on the enamel with 37% phosphoric acid treatment in the extracted permanent molars. The surface roughness was evaluated in 41 extracted non carious permanent human molars. An oval shaped cavity was prepared on

the buccal surface of each tooth with the steel burs. The edges of the cavity were irradiated with Er: YAG laser with energy output of 105mJ in one group and with 208mJ in other group. The radiation was focussed by CaF2 lens and samples were placed in focus, in front of and behind the focal plane .10 samples were etched by phosphoric acid. The comparison of samples showed that the laser treatment also caused desired roughness of the enamel as compared to phosphoric acid.

Shigetani Y, Tate Y, Okamoto A et al in 2002[33] studied the marginal leakage of composite resin restoration using Er:YAG lased cavity in 60 extracted human premolars. The observation of dentin surface after the application of laser showed rough surface similar to fish scales and exposed dentinal tubules without striations. The smear layer was not observed in lased cavity in contrast to rotary cutting device. There is no leakage difference in the marginal seal for enamel and dentin between the laser and air turbine group.

Theodoro LH, Garcia VG, Hayped P et al **in 2002**[34] studied the effects of Er: YAG laser on the root surface of the teeth after scaling and root planning in 18 extracted teeth. They divided the teeth into 3 groups. Group 1-without treatment, Group 2-irraddiated with Er:YAG laser with 47mJ/10Hz in focussed mode with air /water spray for 15s and Group 3- irradiated with 83mJ/10Hz focussed mode with air/water for 15s. The scanning electron microscopy study showed absence of smear layer, with no fissures,

cracks or carbonized areas in group 3 as compared to other groups. In group 3, the surface was irregular. It was concluded that the most efficient removal of smear layer was seen in group 3.

Araki AT, Ibraki Y, Kawakami T et al in 2006[35] compared the effects of Nd: YAG and Ho: YAG on the enamel and dentin surfaces was with different energy levels and different spot sizes. The Ho: YAG laser with energy density of 4160 J/cm^2 with spot size 25µm produced cleaner puncture in dentin with less melting of the surrounding tissues.

Badry AS, Sadr A, Takahashi H et al in 2007[36] analysed the chemical characterises of dentin after irradiation with Er: YAG laser with various output energies with or without water irrigation using attenuated total reflectance Fourier transform infra red spectroscopy and X ray diffraction (XRD) were used for the same. The infra red intensities were compared. The energy output of 100mJ/pulse with water irrigation was not causing any detectable changes in dentin whereas higher energy output with absence of water, affected the organic portion of dentin. XRD results showed no difference between the control and irradiated group. They concluded that the irradiation of 100mJ laser with water irrigation can be used safely for dentin ablation.

Kameyama A, Junji K, Koya A et al in 2008[37] analysed the bond strength of the laser irradiated and non irradiated bovine enamel by three one step adhesive G bond and clearfil tri S and two step self

etch adhesive. In the study 80 extracted bovine enamel surfaces were prepared separately. One half was irradiated with laser. The surfaces were bonded to resin composite with different adhesives. After 24hrs, the bond strength was measured. The results showed no difference between the lasers irradiated group and four adhesives. Each adhesive showed no significant difference between laser irradiated and non irradiated surface. Hence it was concluded that Er: YAG laser irradiations of enamel did not affect the bond strength of one step and two step self adhesives.

The Holium: YAG and Er: YAG laser ablation was studied on alumina balls as hard tissue model by **Watanabe T, Iwai K, Katagiri et al in 2010** [38]**.** The two lasers had different ablation effects because of the difference in the absorption coefficient of water contained in the tissues. When combined irradiation was done, the ablation depended on the time delay between the pulses of the two lasers. The Er:YAG laser was irradiated at 200µm after the Holium :YAG laser. It had ablated surface which was 40% deeper than independent radiation. Heat generated by the Holium: YAG laser decreased the absorption coefficient of water at the Er:YAG laser. Then Er: YAG laser penetrated deeper and ablated the tissue deeper. The same effects were seen on the human dentin.

Effects of other lasers on Enamel and Dentin –SEM study

McCormack SM, Fried D, Featherstone JDB et al in 1995[39] analysed the effects of CO_2 laser on the dental enamel using

scanning electron microscope. 10 extracted bovine and human enamel were irradiated with pulsed CO_2 laser with the following wavelengths (9.3, 9.6, 10.3, 10.6µm) at absorbed fluencies of 2,5,10 and 20J/cm^2 and pulse width of 50, 100, 200and 500µs. Scanning electron microscope revealed evidence of melting, crystal fusion and exfoliation in a wavelength –dependent manner. Crystal fusion occurred at absorbed fluencies as low as 5J/ cm^2 per pulse at all wavelengths except in 10.6µm. The total number of laser pulses delivered to the tissue did not significantly affect surface changes as long as minimum of 5 to 10 pulses were used.

Lin C Lee BS, Lin F et al in 2001[40] conducted a study to determine the composition and morphological changes of the dentin with the irradiation of the Nd: YAG laser. The irradiation parameters used were 150mJ /pulse 10-4s to 150 mJ/ pulses 30 -4s. The x-ray diffractometer showed peak that corresponds to α tri calcium phosphate (tcp) and β tri calcium phosphate. The peaks increased gradually with the elevation of the irradiated energy. The energy which disperses x rays showed that there was increase of calcium phosphorus ratio in the irradiated areas. The energy density of 150mJ/pulse, 30 pulse per second (pps), and 4s and 150mJ/pulse 20 pps 4s resulted in the α tcp formation site and collagen break down, glass like melted surface without tri calcium phosphate formed with 150mj 10pulse pps-4s. SEM showed melting of dentin without cracking used at 150mj pulse 10 pps-4s. So it was suggested that the

irradiation energy of Nd:YAG could be used to fuse a low melting point bioactive glass to dentin with 150mJ/Pulse10pps-4second.

Al Shaher, Al Nazhan S in 2002[41] compared the effect of Nd:YAG laser and Endox –endodontic system(a new endodontic machine used to vaporise the inflamed pulp tissue) on the dentin using SEM. Twenty freshly extracted teeth were studied. The crown was cut at the cemento enamel junction. They were divided into 2 groups for each system. The Nd: YAG laser of wavelength 1.34µm with 269mJ, 10 Hz for 150µs was used in one system and Endox system was used in the other group. The teeth sectioned sagittally at the end of the treatment and root canals were examined by SEM. In group 1- Nd:YAG laser system, the specimen showed normal and regular appearance of dentinal tubules and dentin structures were covered by smear layer. In group 2- Endox system, the specimen showed total evaporation of the smear layer without causing damaging to the dentin structure. There was no smear layer in both systems. Hence both systems can be used in the dentin structure for effective results.

Kwon YH, Kwon O, Hyung-Kim et al in 2003[42] studied the acid resistance of the enamel surface after treatment by Nd: YAG laser. They analysed by the structure, mineral distribution and fluorescence radiance of the enamel surface. The formation of tri calcium phosphate was noticed in the laser ablated areas. The lased surfaces showed increase of calcium phosphorus ratio due to mineral redistribution and acid etching showed decreased ratio

because of mineral loss. The lased areas showed increase of fluorescence radiances and less clear laser con focal scanning microscope image. Hence the Nd: YAG laser irradiation enhances the acid resistance and retard the caries activity in the tooth.

Rode AV, Gsmaly EG, Davies Bl et al in 2003[43] studied the ultra short pulsed lasers for the precision laser ablation of 10 freshly extracted teeth. The laser beam used was 800nm, 1 KHz. The laser beam focused to a diameter of 50µm and was scanned over the tooth surface. The surface preparation of the ablated tooth showed no apparent cracking in optical and scanning electron microscope.

Magalhaes M, Matson E, Wagner R et al in 2004[44] analysed the efficiency of Nd: YAG laser in sealing dentinal tubules in 20 human extracted teeth. Cervical cement was removed with No.575 bur to expose the dentin. Buccal surface of the tooth was irradiated with laser and other surface was used as control. The irradiation parameters used were 30mJ, 0.3W (Group A) and 40mJ, 0.4 W (Group B). 10 samples underwent SEM study and remaining was studied for laser penetration depth by SEM. The irradiated dentin surface showed obliteration of dentin tubules and solidification of dentin surface in SEM. The depth of laser effect varied from 1 to 7µm depending on laser irradiation parameter. Hence they concluded that Nd:YAG laser was very effective measure for obliterating dentinal tubule openings

Bedini R, Manzon L, Fratto G et al in 2010[45] analysed the Nd: YAG laser irradiated enamel for hardness and morphological changes with different energy levels. The energy levels used were 0.6W, 1.2W, 2.4W. The micro hardness test showed no significant difference between treated and non treated samples. SEM study on the enamel samples showed vertical scratches and glass like areas in 0.6 W and crater and cracks in the other two groups. It showed that enamel should be lased at a low energy to preserve its integrity, while high energy creates retentive surface for sealant and composite resin fillings.

Effects of other lasers on dental hard tissues

Mckee in 1993[46] observed the effect of the CO_2 laser on the bone, enamel and dentin in the Wistar rats. The Wistar rats alveolar bone along with the inferior mandible was exposed to CO_2 laser in continuous mode and micro manipulator were used to penetrate the bone and lower incisors. The animals used were perfuse and fixed at either 10minutes or 10 days post operative. The ablated tissue was studied for light microscope and TEM. At ten minutes, the lesion showed organic debris in the ablated areas with voids. The surrounding tissue was slightly damaged but intact. At 10 days lesion in the bone, dentin and odontoblast layer showed morphological evidence of tissue repair presented by the presence of inflammatory cell infiltrate. The new bone formation and reparative dentin concludes the process of tissue repair. In lesions that were created during the secretory stage of amelogenesis that

moved into maturation stage, there was evidence of incomplete maturation related to enamel organ affected by laser. In the bone lesion at 10 days, new bone formation was observed. It was concluded that application of laser is an alternate method for exposing unerupted dental tissues and laser irradiation be useful for mineralized tissue repair.

Luciana, Greghi SL in 2004[47] studied the effectiveness of the Gallium Aluminium Arsenide Diode Laser in the treatment of hypersensitivity in 32 patients aged between 20-54years. They randomly distributed into two groups one treated with laser and the other with placebo using curing light. The treated group was exposed to 6 laser applications within a period of 48-72 hours. The result showed significant reduction of pain observed between the initiation phase and after the laser treatment methods, but such reduction was also seen in the placebo group. It was observed that the statistically significant difference was seen between the initiations of the treatment, but no significant difference was observed between the treated and the control groups.

Effects of lasers on pulp tissues:

Eversole LR, Riziou I, Kimmel AI et al in 1997[48] studied the pulpal response to cavity preparation using Er, Cr: YSGG laser power hydrokinetic system (HKS) by cutting dental hard tissue. The study involved the continuously erupting open apex incisors of albino rabbits and constricted apex of beagles. The effects of HKS

produced by lasers were compared with tapered fissure burs in dental pulp. The results showed that no pulpal inflammatory responses could be identified immediately and 30 days after the surgery, in which enamel and dentin was removed without exposure of pulp.

Glocknerk N in 1998[49] studied the effect on the temperature in the pulp on extracted human incisors and canines in class I cavities. The temperature during preparation with the Er:YAG was compared with diamond bur. The measuring probe was kept at constant temperature of 37^0 C. The results showed that there was a temperature drop from 37^0 C to 25^0 C then to 30^0 C because of cooling effect of water and air. Even with trepanation, there was increase in temperature in the pulp if the laser beam was directly exposed to pulp tissue. With conventional preparation in comparison, even before the trepanation there was a rise in temperature to more than 60^0 C. It was concluded that the reduction in pain with the clinical use of the Er:YAG laser for class V cavities showed lesser increase of intra pulpal temperature in comparison to the conventional bur.

Elliot et al in 1999[50] studied the response of the human primary pulp to CO_2 laser and formocresol for vital pulp therapy in 15 children with intact primary cuspids with at least $2/3^{rd}$ roots remaining who scheduled for orthodontic extraction. They were assigned for pulpotomy with CO_2 laser. The treated teeth were

clinically and radio graphically evaluated at 28 and 90 days post treatment prior to extraction. The extracted teeth were histological evaluated for pulp response. The formocresol treated pulp shows inflammatory cell infiltrate in the coronol pulp which is with moderate to severe in nature at 28 days. At 90 days the most severe change showed coronal pulp necrosis and heavy infiltrate of chronic and acute inflammatory cells. The laser showed zone of oedema, mild chronic inflammatory cell infiltrate at 28 days.

Jayawardena in 2001[51] observed the pulpal response to the Er:YAG laser and slow speed conventional bur after accidental exposure of the pulp in 76 maxillary first molars of male Wistar rats. Rats were killed immediately at 3 days, 1week and 2 weeks after the pulp exposure. Histopathological examination of the pulp showed no bleeding and no dentin chips at pulp exposed sites immediately after the pulp exposure. After 2 weeks of pulp exposure, the Er:YAG laser group formed more dentin bridges at the exposure site than the bur group. Good healing capacity with the formation of dentin bridges and reparative dentin was observed in the laser group.

Aldo BJ, Fatima Z, Marchesan M et al in 2002[52] studied the intra pulpal temperature changes in the teeth using Er: YAG laser in the preparation of Class V cavities *in vitro* in 10 incisors, 10 canines, 10 premolars and 10 molars. The parameters used were the short pulse 250ms/pulse and very short pulse 80-12-ms/pulse with 10 Hz, 500mJ for 6 seconds with 10mm distance and 25ml/min water flow

at 23^0C with humidity 65%. The greatest increase in temperature was found in incisors (2.8^0C). The results showed that the increase of temperature was less in very short pulse than short pulse.

Oeligieserd in 2003[53] observed the intra pulpal temperature changes during cavity preparation by Er: YAG laser in 175 fresh extracted caries free human teeth and in 42 caries teeth. Class I and Class V cavity were prepared in the samples with thermocouples inserted into the pulp chambers. Lasing was done with different energies and pulse frequencies. The temperature changes in the pulp chamber were measured. When the cavity preparation done using laser, it was noticed that the highest pulpal temperature increase was observed in the class 1 cavity preparation (3.31^0C), medium values in class V (2.10^0C) and lowest values were seen in the cemental preparation (1.21^0C).

Mollica FB, Camargo FP, Zamboni SC et al in 2008[54] compared the intra pulpal temperature produced between high speed hand pieces, Er: YAG laser and ultrasound tips during cavity preparation. They used 30 bovine mandibular incisors with enamel dentin thickness of 4mm at the buccal surface. Class V cavities were prepared to a depth of 3.5mm measured with periodontal probe. A thermocouple which was inserted in the pulp was used to measure the temperature changes. The mean temperature rise for high speed hand piece was 1.10^0C for Er: YAG laser at 0.84^0 C and for ultrasound tip at 3^0C. There was no statistical difference between the hand piece and Er: YAG laser. But ultra sound tips showed that

there was increase in temperature which is below the critical level of 5.5^0 C. The results showed that the use of Er:YAG laser and high-speed hand piece for cavity preparation showed similar temperature increase. Although ultrasound tips had higher intrapulpal temperature it remained below critical value and was considered safe for use.

Freitas P, Geraldo DS, Silva ACB et al in 2008[55] analysed the intra pulpal temperature variation during irradiation for caries prevention in third molar *in vitro*. They were grouped into 1) - Er, Cr: YSGG laser with 0.25W at 20Hz. 2) With 0.50W and 3) with 0.75W. The irradiation was done on the enamel with thermocouple attached to the inner surface. The intra pulpal temperature variation was observed as less than 0.1^o C for all irradiation. It showed that an acceptable temperature increase in pulpal chamber was observed when enamel was irradiated with Er, Cr: YSGG lasers for caries prevention methods.

Gallogos M, Arnabat-Dominguez et al in 2009[56]recorded the thermal increment recorded at the external root canal surface during application of Er, Cr: YSGG laser with endodontic tips. Twenty human canines and incisors were used in the study; irradiation was carried out using glass fibre endodontic tips. The teeth were irradiated for 1W and 2W for 30sec without water spraying. The result showed that at 1W power the mean temperature increment was 3.84^0 C versus 5.01^0C at 2W. The application of Er, Cr: YSGG

significantly increased the temperature at the external root surface; it would appear to damage the tissue in proximity to the treated tooth.

Krmek SJ, Miletic I, Simeon P et al in 2009[57] studied the temperature change in the pulp chamber during cavity preparation using Er: YAG laser by employing thermocouple. Nine groups of ten intact molars were used. The class V cavity were prepared to levels enamel and dentin. The enamel was lased in different energy levels in contact mode for 10 seconds. The dentin also irradiated with different modes for 7seconds. Cooling was done with water spray. The highest temperature rise in the pulp (1.99^0C) was seen in the enamel irradiation with 400mJ, 15Hz and lower (0.90^0C) was seen in 320mJ with 10Hz. In dentine the highest temperature increase in the pulp was achieved (1.37^0C) with 340mJ and the lowest (0.43^0C) was 200mJ with 5Hz. The enamel and the dentin showed that the influence of energy on temperature increase was stronger than frequency. In conclusion cavity preparation with Er:YAG laser with very short pulse mode did not cause significant increase in temperature in human molar

A study was conducted by **Tanabe K, Yoshiba K, Yoshiba N et al in 2002**[58] to determine the pulpal response of the teeth in which Er:YAG laser and conventional bur techniques were used. Cervical cavities were prepared in the upper first molars of rats, using either an Er:YAG laser or a conventional tungsten-carbide bur. At intervals of 5 min, 6 hours, 12 hours, 1 day, 3 days and 7

days after cavity preparation, the teeth were processed for immuno histochemical analyses of tissue non-specific alkaline phosphatase, OX6-positive major histocompatibility complex class II antigen-expressing cells and PGP 9.5-immunoreactive nerve fibers. DNA fragmentation was detected by the terminal deoxynucleotidyl transferase-mediated dUTP nick end labeling (TUNEL) method. Tissue non-specific alkaline phosphatase was observed mainly in the subodontoblastic layer under the cavity lesion, from 5 min, in both groups. The immunoreactivity was more pronounced in the laser group, but by 7 days no significant differences were recognizable. At 12 hours, TUNEL-positive cells were detected around the odontoblastic layer in both groups. From 3 days to 7 days, a limited number of positive cells were still visible in the group that underwent standard treatment. Clear similarities in the distribution patterns of OX6-immunopositive cells and PGP 9.5-immunoreactive nerve fibers were also noted. From 12 hours to 1 day, OX6-positive cells accumulated along the pulp-dentin border, extending their processes into the dentinal tubules. Numerous bead-like PGP 9.5-immunoreactive nerve fibers were observed under the odontoblastic layer at 7 days. These results demonstrated that there was no appreciable difference in the manner in which pulp tissue responded to treatment with either Er:YAG laser or a conventional drill. This would seem to indicate the usefulness of the Er:YAG laser system in the removal of caries and cavity preparation.

Effects of laser on oral mucosa:

Ibarguren IC, Espana-Tost A, Dominguez JA et al in 2010[59] compared the thermal effect produced on the soft tissue by CO_2, Er, Cr: YSGG and diode laser. The porcine oral mucosa was irradiated with Er, Cr: YSGG lased at 1W with without water spray and diode laser at 2W, 5W and CO_2 laser with different energy levels. The thermal damage was evaluated by measuring the width of damaged tissue adjacent to the incision and stained with H&E, Masson tri chrome stains. The result showed lowest thermal effect was seen in Er, Cr: YSGG laser with water followed by CO_2 and diode laser. They concluded that the wavelength of each laser determines the absorption rate of every tissue and thermal effect.

Effects of burs on dental hard tissues:

The effects of steel and tungsten carbide burs have been studied under the scanning electron microscope on human enamel and dentin during preparation of mesio-occluso-distal cavities *in vitro* in 50 teeth with cutting speeds varying from 5 000 to 120 000 rotation per minute by **Comtle in 1983**[60]**.** All the cavities presented clean and regular walls. However, the final quality of the cavity borders, and in particular the cavo-surface angles of the box walls, was directly related to the speed and rotation direction of the bur. The cavo-surface angles at the gingival planes showed in all cases fractures of the superficial prisms. The cutting characters displayed in dentine surfaces were mainly dependent on the orientation of the tubules in relation to the surface of the cavity preparation and the degree of

sclerosed dentine. The cavities prepared in the absence of water and air showed the presence of tissue debris and cracks.

A study was conducted by **Sekimoto T, Derkson GD, Richardson AS in 1999** [61] to compare dentin permeability and the morphology of the dentin surfaces prepared with diamond and carbide steel burs after etching with 6% citric acid. Twenty-four freshly extracted human third molars were sectioned, mounted on plexiglass, and connected to the dentin-permeability measuring apparatus. The permeability of dentin was measured by fluid filtration and expressed as hydraulic conductance. There were two study groups of 12 teeth. Each tooth had one occlusal cavity preparation prepared but utilized three depths: the original was prepared just into the dentin, the second one 0.5 mm deeper than the first, and the third 0.5 mm deeper than the second. One group had the first cavity prepared with a diamond, the second deepened using a steel bur, and then the third depth was made by use of the diamond. The other group had the first cavity preparation prepared with a steel bur, deepened 0.5 mm again using a diamond and then deepened again using a steel bur. Dentin permeability was measured after cavity preparation, then after 2 minutes of acid etching. Analysis of variance and Duncan's multiple range tests were used to establish whether differences were significant at the 0.05 confidence level. Prepared and acid-etched surfaces were characterized using a scanning electron microscope to identify any differences between the two groups. After acid etching with 6%

citric acid, the permeability of dentin cavities prepared with diamond burs was significantly less than the permeability of cavities prepared with carbide steel burs. After etching, there were differences in the appearance of the dentin surfaces prepared with diamonds and steel burs. Dentin bonding agents may have their effectiveness reduced when placed following cavity preparation by use of a diamond burs.

Luciana ML, Cristiane M, Lourdes S et al in 2006[62] conducted a study to determine the cutting ability of chemical vapor deposition (CVD) diamond burs coupled to an ultrasonic dental unit hand piece for minimally invasive cavity preparation. One standard cavity was prepared on the mesial and distal surfaces of 40 extracted human third molars either with cylindrical or with spherical CVD burs. The cutting ability was compared regarding type of substrate (enamel and dentin) and direction of hand piece motion. The morphological characteristics, width and depth of the cavities were analyzed and measured using scanning electron micrographs. Statistical analysis using the Kruskal-Wallis test ($p < 0.05$) revealed that the width (1,199µm) and depth (181µm) of the cavities were significantly greater when they were prepared on dentin. Wider cavities were prepared when the cylindrical CVD bur was used, and deeper cavities resulted from preparation with the spherical CVD bur. The direction of handpiece motion did not influence the size of the cavities, and the CVD burs produced precise and conservative cutting.

Patient acceptance:

Keller in 1998[63] observed the patient's perception and response to cavity preparation between conventional mechanical preparation and Er: YAG laser preparation in dental hard tissue in 103 patients with 206 preparations. The laser treatment was found to be more comfortable than the mechanical preparation. It was found that 80% of the patients perceived conventional preparation as more uncomfortable than laser treatment and 82% of the patients indicated that they would prefer the Er: YAG laser preparation for further caries treatment. Hence the application of the Er: YAG laser system was perceived comfortable alternative or adjunct than conventional mechanical cavity preparation.

Evans DJP, Mathews S,Pitts NB et al in 2000[64] conducted a randomised controlled trial to determine the acceptability of cavity preparation in patients and dentists using Er: YAG laser with conventional burs. They randomly selected 15 dentists who treated 77 patients with age range from 3.5-68yrs who had two similar cavities. One cavity was prepared by conventional method and the other by laser. The dentist and patient preference was determined by a questionnaire. The dentist showed preference for conventional cavity preparation as compared to lasers which was significant ($p<0.001$). The difficulties encountered with the laser were access and slow speed of cutting. Patients aged more than 10years

expressed laser treatment as their choice and difference was significant ($p<0.001$). Patient aged less than 10years did not show any preference for either technique.

The cavity preparation by air turbine with burs and Low powered system, Erbium, Chromium: YSGG were compared by **Hadley in 2000**[65] using split mouth design. Class I, II and V cavities were prepared and restoration was done with resin materials. 75 teeth of 68 subjects were included in the study. The pulp vitality, the recurrent caries, pain , discomfort and restoration retention were analysed on the day of procedure, 30 days after and 6 months post operatively. The results showed that there was no statistically difference between the two groups in all parameters except in patient discomfort level. Patient felt better when laser was used in the cavity preparation.

Boj in 2005[66] assessed the psychology of the patient undergoing dental procedures like restorative and surgery were done under local anaesthesia with waterlase Er, Cr: YSGG. 49 patients were participated in this study and were divided into two different groups- restorative (33 patients) and oral surgery (16 patients). The degree of pain felt was assessed using Wong –Baker facial image scale. It showed six different faces numbered from 0-5. The patient had to choose one face only. Scores on the pain scale were low in cavity preparation. None of them needed local anaesthesia. Twelve patients underwent surgery without local anaesthesia and four

needed infiltration. These results showed that the pain perception was less in the primary teeth than permanent teeth.

Time duration:

Aoki A, Ishikawa I, Yamada T et al in 1998[67] studied the time difference *in vivo*, in removing caries between Er:YAG laser and conventional bur. They used 31 freshly extracted teeth ,in which half of the carious root were cleaned using bur while the rest was cleaned with Er:YAG laser. The bur group was powered by low speed micro motor. The time required to remove caries and histopathological observation of decalcified sections were compared. The result showed that the longer time is needed for Er: YAG laser compared to burs.

Shigetani Y, Okamoto A, Abubakr N et al in 2002[2] compared the time taken for the laser and bur to cut enamel and dentin in ten teeth for each group. The laser parameters used were 0.4mm, 200mJ/pulse and 100mJ/pulse. The burs used were round stainless steel of 0.44mm in diameter. The average time taken for the laser to cut the enamel was 33 seconds and the bur was 23.5 seconds. The cutting time of the laser in dentin took 35.3 seconds while the bur took 35seconds. There was no significant difference in the time taken between Er: YAG laser and bur in cutting dentin, but difference exists in removing the enamel.

Forty teeth from eighteen first visit dental patients formed the study group. Of the 18 patients, 16 were females and 2 were males. The age of the patients range from 13 years to maximum of 24 years with mean age of 16.8±2.3 years. Out of 40 samples 2 were upper canines. The 38 remaining samples were 1st premolars. Among them, 22 were upper premolars and 16 were lower premolars. Patients were randomly divided into three groups.

Group 1-The cavity preparation was done by Er:YAG laser using 300mJ at 20Hz in 20 samples.

Group 2- The cavity preparation was done by air turbine hand piece with diamond bur no.SI 47 in 18 samples.

Group 3- the cavity preparation was done with air turbine hand piece with tungsten carbide bur no.245 in 2 samples.

In all the groups, patients tolerated well to the procedures.

The gender distribution of the samples in group1 (n=20) is as 80% females and 20% males. In group 2 (n=18) showed 94.5% females and 5.5%males. In group 3 (n=2) all the samples were from females. The overall distribution (n=40) contains 87.5% females and 12.5% males. (Table 1 and Graph 1)

The time required to prepare the cavity between the groups were recorded. In group 1 (n=20) the time taken to prepare the cavity was 6.50±1.63 minutes (Table 2). The required time to prepare the cavity using diamond bur in group 2 (n=18) was

4.65±0.93 minutes (Table 2). The required time to prepare the cavity in group 3 (n=2) was 4.50±0.70 minutes (Table 2). The difference in time taken for the three groups were statistically significant (p =.001).The time difference between the group 1 and 2 was statistically significant.

The morphological changes between the groups were compared between Group 1 and 2 with 3 using SEM. It showed that all samples in the laser group (n=5) showed ill defined cavity margins with irregular surface. (Table 4) (Fig 4)The diamond bur group (n=5) showed well defined margins with regular surface (Fig 5) (100%). The third group tungsten carbide (n=2) also showed the same result (Fig 14) (100%). The smear layer was present in both the bur groups (100%) but was indistinct in laser group (Table 4). Cracking was seen in all the three groups. The surface roughness was mild in bur groups (Fig 6) but was severe in laser group (Fig 7). The dentinal tubules were seen in both the bur groups (100%). It was obliterated in the laser group. Microcracks were seen in all the groups. Glazing was seen in the laser group (60%) (Table 4) (Fig 13). The other two groups showed no glazing. Craters were present in laser group and diamond bur group equally (20%).

The histopathological examination was done on the decalcified section of the laser group (n=10) and diamond bur group (n=8).There were no histopathological difference between the laser group and the diamond bur group. The pulp stones was seen in the

laser group(Fig 30) (n=1) (20%) and in the diamond bur group (n=1) is 12.5% (table 5).

The morphological changes between the laser group (n=5) and diamond bur group (n=5) was studied using ground section of the teeth. They showed no difference between the two groups. The structure of the enamel and dentin were normal in appearance (Fig 38). The dentino enamel junction showed normal scalloped margin. The dentin showed dead tracts in both the groups (Table 6).

A. **Scanning electron microscopic study of cavity walls of the group 1.**

1) **Sample 1.** –The time required to prepare the cavity was 4 minutes. The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

   On SEM photomicrograph showed the occlusal cavosurface margins of the cavity were ill defined with irregular cavity surfaces(20X).The enamel cavity wall showed uniform scaly appearance with severe roughness(500X) The dentinal cavity wall showed indistinct smear layers [Fig-11]. The obliteration of dentinal tubules were seen (500X) [Fig-9]. The floor of the cavity showed microcracks with irregular craters (1500X).

2) **Sample 2**- The time required to prepare the cavity was 6 minutes. The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

On SEM photomicrograph showed the occlusal cavosurface margins of the cavity were ill defined with irregular cavity surfaces. (20X).The enamel cavity wall showed scaly appearance with severe roughness (500X) [Fig-7]. The cavity showed evidence of glazed areas in the base (200X) [Fig-13]. The dentin of the cavity showed indistinct smear layer with obliteration of dentinal tubules (1000X). The floor of the cavity showed irregular craters. (1500X).

3) **Sample 3**- The time required to prepare the cavity was 6 minutes. The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

   On SEM photomicrograph showed the occlusal cavosurface margins of the cavity were ill defined with irregular cavity surfaces (23X) [Fig-5]. The enamel wall showed uniform scaly appearance with severe roughness. The dentin of the cavity showed indistinct smear layer with obliteration of dentinal tubules (270X). The base of the cavity showed irregular surfaces (320X).

4) **Sample 4**- The time required to prepare the cavity was 4 minutes. The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

   On SEM photomicrograph showed the occlusal cavosurface margins of the cavity were ill defined with irregular cavity surfaces (35X). The enamel wall showed scaly appearance with severe roughness. The dentin of the cavity showed

indistinct smear layer with obliteration of dentinal tubules (1000X).

5) **Sample 5-** The time required to prepare the cavity was 5 minutes. The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

   On SEM photomicrograph showed the occlusal cavosurface margins of the cavity were ill defined with irregular cavity surfaces (35X).The enamel cavity wall showed scaly appearance with severe roughness. The dentin of the cavity showed indistinct smear layer with obliteration of dentinal tubules (1000X). The floor of the cavity showed uniform appearance of irregular surface (1500X).

B. **Group 1- Histopathological study**

6) **Sample -6**- The time required to prepare the cavity was 6 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

   On light microscopic examination under *4x* – showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber. One area showed artefactual separation of odontoblastic cells from dentin. The pulp tissue was seen in the pulp chamber and pulp canal.

   On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

7) **Sample -7**- The time required to prepare the cavity was 7 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

   On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber. One area showed artefactual separation of odontoblastic cells from dentin. The pulp tissue was seen in the pulp chamber and pulp canal.

   On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres.

   0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

8) **Sample -8-** The time required to prepare the cavity was 5 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

   On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining

the pulp chamber. The pulp tissue was seen in the pulp chamber and pulp canal.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cell.

9) **Sample 9**- The time required to prepare the cavity was 6 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

   On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber. One area showed artefactual separation of odontoblastic cells from dentin. The pulp tissue was seen in the pulp chamber and pulp canal.

   On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres.

   0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

10) **Sample 10**- The time required to prepare the cavity was 6 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light

microscope. On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin [Fig-27]. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber [Fig-33]. One area showed artefactual separation of odontoblastic cells from dentin [Fig-29]. The pulp tissue was seen in the pulp chamber [Fig-31].

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres [Fig-35]. The blood vessels were lined by endothelial cells.

11) **Sample 11**- The time required to prepare the cavity was 7 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber. The pulp tissue was seen in the pulp camber and pulp canal.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

12) **Sample -12**- The time required to prepare the cavity was 5 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber. The pulp tissue was seen in the pulp camber and pulp canal.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres. Homogenous haematoxyphilic calcified structure - pulp stones were seen in the pulp canal.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

13) **Sample -13**- The time required to prepare the cavity was 5 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin [Fig-19]. The dentin showed normal dentinal tubules [Fig-21]. The odontoblastic cells were lining the pulp chamber [Fig-23]. One area showed

artefactual separation of odontoblastic cells from dentin. The pulp tissue was seen in the pulp camber and pulp canal.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres [Fig-25]. The blood vessels were lined by endothelial cells. Homogenous haematoxyphilic calcified structure - pulp stones were seen in the pulp canal.

14) **Sample -14**- The time required to prepare the cavity was 5 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber. The pulp tissue was seen in the pulp chamber.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

15) **Sample 15**- The time required to prepare the cavity was 7 minutes. The tooth was extracted and decalcified using 10%

formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber. The pulp tissue was seen in the pulp chamber.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

C. **Group 1 – Ground section**

16) **Sample -16**- The ground section of the tooth showed well defined cavity margin [Fig-37]. The enamel appeared normal in structure [Fig-39]. The enamel lamellae and tufts were seen. The dentino enamel junction appeared normal with scalloped margins [Fig-41]. The dentin showed dead tracts.

17) **Sample -17**- The ground section showed well defined cavity margins. The enamel appeared normal in structure. The enamel lamellae and tufts were seen. The dentino enamel junction appeared normal with scalloped margins. The dentin showed dead tracts.

18) **Sample -18**- The ground section showed well defined cavity margins [Fig-43]. The enamel appeared normal in structure

[Fig-45]. The enamel lamellae and tufts were seen. The dentino enamel junction appeared normal with scalloped margins. The dentin showed dead tracts [Fig-47].

19) **Sample-19**- The ground section showed well defined cavity margins. The enamel appeared normal in structure. The enamel lamellae and tufts were seen. The dentino enamel junction appeared normal with scalloped margins. The dentin showed dead tracts.

20) **Sample -20**- The ground section showed well defined cavity margins. The enamel appeared normal in structure. The enamel lamellae and tufts were seen. The dentino enamel junction appeared normal with scalloped margins. The dentin showed dead tracts.

D. **Group -2 Scanning electron microscopic study**

21) **Sample 21**- The time required to prepare the cavity was 3 minutes. The cavity was prepared using diamond bur no. SI 47.The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

On SEM, the occlusal cavosurface margin showed well defined cavity margins with regular cavity surfaces (20X). The enamel wall showed scaly surface with mild roughness. The dentin of the cavity showed presence of smear layer with opening of dentinal tubules (500X). The base of the cavity showed flat base with smooth surface (300X) [Fig-12].

22) **Sample 22**- The time required to prepare the cavity was 5 minutes. The cavity was prepared using diamond bur no. SI 47.The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

On SEM, the occlusal cavosurface margin showed well defined cavity margins with regular cavity surfaces. (20X). the cavity enamel showed scaly surface with mild roughness. The dentin of the cavity showed presence of smear layer with opening of dentinal tubules (300X) [Fig-8]. The base of the cavity showed flat base with glazed areas (1500X).

23) **Sample 23**- The time required to prepare the cavity was 5 minutes. The cavity was prepared using diamond bur no. SI 47. The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

On SEM, the occlusal cavosurface margin showed well defined cavity margins with regular cavity surfaces. (20X) [Fig-4]. The cavity enamel showed indentation of the bur with mild roughness [Fig-6]. The dentin of the cavity showed presence of smear layer with opening of dentinal tubules (500X). The base of the cavity showed flat base with microcracks (300X).

24) **Sample 24**- The time required to prepare the cavity was 4 minutes. The cavity was prepared using diamond bur no. SI 47.The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

On SEM, the occlusal cavosurface margin showed well defined cavity margins with regular cavity surfaces. (20X). the cavity enamel showed scaly surface with mild roughness. The dentin of the cavity showed presence of smear layer with opening of dentinal tubules (500X). The base of the cavity showed flat base. (1000X).

25) **Sample 25**- The time required to prepare the cavity was 4 minutes. The cavity was prepared using diamond bur no. SI 47.The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

On SEM, the occlusal cavosurface margins showed well defined cavity margins with regular cavity surfaces with the indentation of bur on the surfaces (35X). The cavity enamel showed scaly smooth surface with mild roughness. The dentin of the cavity showed presence of smear layer with opening of dentinal tubules (1000X) [Fig-10]. The base of the cavity showed flat base with microcracks (200X).

E. **Group 2- Histopathological study**

26) **Sample 28**- The cavity preparation was done by air turbine hand piece using diamond bur no.SI 47. The time required to prepare the cavity was 4 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed

normal dentinal tubules. The odontoblastic cells were lining the pulp chamber.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres. The pulp chamber was scant in pulp tissue.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

27) **Sample 29**- The cavity preparation was done by air turbine hand piece using diamond bur no.SI 47. The time required to prepare the cavity was 6 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin [Fig-18]. The dentin showed normal dentinal tubules [Fig-20]. The odontoblastic cells were lining the pulp chamber [Fig-22].

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres. The pulp chamber was scant in pulp tissue.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres [Fig-24]. The blood vessels were lined by endothelial cells.

28) **Sample 30**- The cavity preparation was done by air turbine hand piece using diamond bur no. SI 47. The time required to

prepare the cavity was 5 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin**.** The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber. The pulp canal showed pulp stones.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres. The pulp chamber was scant in pulp tissue.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

29) **Sample 31**- The cavity preparation was done by air turbine hand piece using diamond bur no. SI 47. The time required to prepare the cavity was 4 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope. On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin [Fig-26**]**. The dentin showed normal dentinal tubules. One area showed artifactual separation of odontoblastic layer [Fig-28**]**. The odontoblastic cells were lining the pulp chamber. The pulp canal showed pulp stones.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres. The pulp

chamber was scant in pulp tissue. In pulp canal the pulp stones were seen.

0n 40x – the odontoblastic cells with nucleus were seen [Fig-32]. The pulp tissue showed fibroblastic cells and fibres [Fig-34]. The blood vessels were lined by endothelial cells. Homogenous haemotoxyphilic calcified structure pulp stones were seen [Fig-30].

30) **Sample 32**- The cavity preparation was done by air turbine hand piece using diamond bur no. SI 47. The time required to prepare the cavity was 6 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres. The pulp chamber was scant in pulp tissue.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

31) **Sample 33**- The cavity preparation was done by air turbine hand piece using diamond bur no. SI 47. The time required to prepare the cavity was 4 minutes. The tooth was extracted and

decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres. The pulp chamber was scant in pulp tissue.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

32) **Sample 34**- The cavity preparation was done by air turbine hand piece using diamond bur no. SI 47. The time required to prepare the cavity was 4 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres. The pulp chamber was scant in pulp tissue.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells.

33) **Sample 35**- The cavity preparation was done by air turbine hand piece using diamond bur no SI 47. The time required to prepare the cavity was 5 minutes. The tooth was extracted and decalcified using 10% formic acid. The decalcified section was studied using light microscope.

On light microscopic examination under *4x* –it showed well defined cavity margins in the dentin. The dentin showed normal dentinal tubules. The odontoblastic cells were lining the pulp chamber.

On 10x- the normal dentinal tubules with pulp chamber showed loose connective tissue with collagen fibres. The pulp chamber was scant in pulp tissue.

0n 40x – the odontoblastic cells with nucleus were seen. The pulp tissue showed fibroblastic cells and fibres. The blood vessels were lined by endothelial cells

F. **Group 2- Ground section**

34) **Sample 36-** The ground section showed well defined cavity margins [Fig-42]. The enamel appeared normal in structure [Fig-44]. The enamel lamellae and tufts were seen. The dentino enamel junction appeared normal with scalloped margins. The dentin showed dead tracts [Fig-46].

35) **Sample 37-** The ground section showed well defined cavity margins. The enamel appeared normal in structure. The enamel lamellae and tufts were seen. The dentino enamel junction appeared normal with scalloped margins. The dentin showed dead tracts.

36) **Sample 38-** The ground section showed well defined cavity margins. The enamel appeared normal in structure. The enamel lamellae and tufts were seen. The dentino enamel junction appeared normal with scalloped margins. The dentin showed dead tracts.

37) **Sample 39-** The ground section showed well defined cavity margins. The enamel appeared normal in structure. The enamel lamellae and tufts were seen. The dentino enamel junction appeared normal with scalloped margins. The dentin showed dead tracts.

38) **Sample 40-** The ground section showed well defined cavity margins [Fig-36]. The enamel appeared normal in structure. [Fig-38]. The dentino enamel junction appeared normal with scalloped margins. The dentin showed dead tracts [Fig-40].

G. **Group 3 –SEM study**

39) **Sample 26**- The time required to prepare the cavity was 4 minutes. The cavity was prepared using tungsten carbide bur no.245.The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

On SEM, the occlusal cavosurface margin showed well defined cavity margins with regular cavity surfaces (27X) [Fig-14]. The cavity enamel showed scaly smooth surface with mild roughness. The dentin of the cavity showed presence of smear layer with opening of dentinal tubules (100X) [Fig-15].

40) **Sample 27**- The time required to prepare the cavity was 5 minutes. The cavity was prepared using tungsten carbide bur no.245.The tooth was extracted and the cavity preparation was studied using scanning electron microscope.

On SEM, the occlusal cavosurface margin showed well defined cavity margins with regular cavity surfaces. (21X). the cavity enamel showed regular smooth surface with mild roughness (850X) [Fig-17]. The dentin of the cavity showed presence of smear layer with opening of dentinal tubules (500X) [Fig-16].

Lasers have revolutionized the world in all the fields during the 21st century. The acceptance of lasers in medicine and dental fields was one of the events that created new vision in scientific techniques and procedures in the research based studies. Lasers have become important tool for diagnosis and treatment of diseases. Dental lasers are being using in various dental specialities for both hard tissue and soft tissue procedures.

Graham JM in 2009 [68] used laser for preservation of the tooth structure in minimal interventional operative dentistry. Dental caries is one of the common diseases that irreversibly affect the hard tissues of the tooth. Various methods were introduced in removal of the diseased tooth structure and restoration of the lost structure. The techniques for the preparation of cavities have been modified in recent years with various techniques including rotary instruments and lasers.

The conventional cavities were prepared using diamond abrasives and tungsten carbide burs with the high speed rotary devices. Margolis F in 2009[69] stated that dentists preferred use of this procedure because of its précised cutting, ease of control good tactile perception with good visibility and cost. The rotary cutting instruments have the disadvantage of causing pain, noise and over cutting the tooth structure. Dull and worn out burs produce heat and could potentially damage the pulp tissue [69]. Lasers were introduced to overcome these drawbacks.

Er: YAG laser:

The wavelength of laser determines its properties and penetration in different tissues by its absorption in the tissues. According to Castellan et al in 2007 [70], the thermal and the mechanical effects of the laser tissue interaction were based on the tissue target and the laser pulse characteristics. Lasers are available from wavelength of 488nm to of 10,600nm. The current choice is Er: YAG laser which had a wavelength of 2970nm. Walsh LJ in 2003 [71] reported that Er:YAG laser is highly absorbed by water and hydroxyapaptite, which are present in the dental hard tissue. The intense absorption within microspores heats the water instantaneously change its phase to vapour increasing the pressure inside the microspore. This effect breaks the hydroxyapaptite structure leading to micro explosion that eject dental hard tissue away from the laser interacting volume. Hence it is useful for the removal of caries with less damage to the surrounding tissues.

Rubens et al in 2005 [28] considered the optimal temperature measurements of the laser parameters. He considered 300mJ energy output causes effective removal of dental tissue with minimal damage to surrounding tissues. The laser parameters used in our study were based on temperature measurements from study of Ruben et al.

Patient acceptance:

Studies by Evans DJP et al in 2000[64],Keller U in 1998 [63] and Harashima et al [72] have shown that the patients are comfortable and have less pain with laser than bur . The psychological assessment of the patient study conducted by Boj in 2005[66] confirmed these findings. The requisition of local anaesthesia is reduced in the laser cavity preparation when compared to bur as observed by Glocknerk N in 1999 [49]. The fear of anxiety to needles can then be avoided in laser preparation. One of the parameters that cause pain perception to the patient was the vibration speed which is minimised with laser. Jakamoosik et al in 2003[73] reported the mean vibration speed that occurs in laser is 166±38 µm/second and the high speed dental drill induces a 100 times higher vibration. Though the present study did not assess the patient psychology between the study groups, the overall acceptance was similar in all the groups. The patients cooperated well to the above procedures. For small cavities the advantage of laser is similar to rotary drills.

Time duration:

In our study the required mean time to prepare the cavity using diamond bur was 4.65±0.93 minutes. The required mean time to prepare the cavity in tungsten carbide bur was 4.50±0.70 minutes. The mean time needed to prepare the cavity with laser was 6.5±1.63 minutes. The time taken to prepare the cavity in lasers were slight longer than the burs which was statistically significant ($p<0.000$). This observation is similar to that report of Shigetani Y et al in

2002 [2] that the time required to prepare the cavity with laser was longer than the burs. Between the groups 2 and 3 no difference in time was observed when similar procedures done. The time with lasers can be reduced by increasing the energy densities, but the risk of pulpal damage may be increased. The time required to prepare the cavity depends on several factors such as patient co-operation, visibility of the patient fields, accessible areas and the operator's skill and type of cavity.

SEM analysis: Morphological changes

In our study the morphological alterations of the cavity walls differ between the three groups. On the SEM examination of the cavity preparation, the laser group showed irregular occlusal cavosurface margins. The pattern was similar in all the samples. The surface of the enamel cavity wall was irregular as described by Hossain M in 2003[26] in the literature. Hibst et al [74] showed that the lased cavity surface produces rough appearance of the enamel. In the dentin surface the dentinal tubules were exposed and these surfaces appears as scaly or flaky these irregular surface occurs because of the micro explosion of the hard tissue by Er:YAG laser.

The conventional bur preparation in our study showed regular well defined occulso cavo surface margins. The indentation of the abrasive particles was seen on the tooth structure. The cavity was uniform in shape because of good tactile sensation. The cavity

enamel wall showed regular surface with smooth uniform running of the diamond abrasives.

In our study, the surface roughness was severe in the laser group compared to the bur group. The enamel wall appears as scaly as described in the study by Hibst et al [74]. Aoki A et al in 1998[67] showed craters with white chalky appearance in the cavity walls of the enamel. Surface roughness was significantly increased in the study conducted by Hossain M in 2003[26]. The surface roughness increased because of the interaction of the laser beam with hard tissue result s in irregular surface. The enamel roughness cannot be assessed quantitatively using SEM analysis. The surface roughness in the enamel helps to increase the adhesion of the restorative material to the cavity walls, and may be beneficial with composite restorations.

The enamel surface showed microcracks and craters in cavity prepared by lasers. The craters and microcracks increase, when the energy output of the laser beam increases. This is due to the process of hard tissue melting and the following solidification with generation of voids, craters and cracks, the surface roughness and glazed areas. The craters and cracks occur as result of increase of temperature up to the sublimation point of the material and subsequent by during solidification it change its structure. In our study as we used optimum energy output, at which the microcracks are the least. The glazing is seen at the of the lower energy output.

In the bur preparation, the microcracks were present, but cracking and craters was absent.

In the bur group, the dentinal wall of the cavity showed sharp cavity margins with opened dentinal tubules with smear layer. On the contrary, laser group exhibited ill defined cavity margin the relatively less smear layers. In the study using Er:YAG laser by Hossain N et al in 1999 [75] they that the prepared cavities showed an irregular surface with the absence of smear layer. Enamel prism and dentinal tubules were seen. The dentinal surface of the lased cavity showed irregular surface with the obliteration of the dentinal tubules. In our study we were not able to appreciate the dentinal tubules opening in the laser group, but observed the reduction of smear layer. The floor of the cavity showed irregular surface.

Isabel et al in 2009[76] suggested that the obliteration of the dentinal tubules would reduce dentinal hypersensitivity. This can reduce the passage of fluid across the dentinal tubules. The conventional cavity showed the open dentinal tubules which leads to pulpal damage and failure of the restoration. The dentinal surface showed absence of smear layer in the lased cavity. The smear layer which forms on the dentin surface of the cavity interferes with the bonding ability of the restoration. So in the lased cavity the bonding of the restoration is good with better adaptation to the restoration particularly composite resins. In the conventional cavity preparation the dentinal surface has smear layer with the opening of dentinal

tubules which interferes with adaption of the material to the cavity. So removal of the smear layer is mandatory for better bonding strength for the restoration.

SEM analysis –tungsten carbide bur:

The tungsten carbide bur is used with the high speed dental drill with more than 300,000 rotations per minute and produces a clean cut. It is very strong in its action to cut the metal and remove the older restorations. It is not commonly used in the minimal intervention cavity preparation as reported by Graham JM in 2009[63]. They often produce micro cracks in the enamel (Welchm AJ in 1989)[12]. In our study we used 2 samples with the tungsten carbide burs. The SEM analysis showed well defined cavity margins and microcracks in the enamel. The dentin layer showed debris and smear layer. The floor of the cavity was flat. There is not much difference between the diamond bur and the tungsten carbide bur preparation.

Ground section:

In the ground section of the cavity prepared teeth showed no difference between the laser group and the bur group. The enamel appears normal on both the groups. The enamel lamellae and tufts were similar in all groups. The dentino enamel junction appeared scalloped as in normal tooth structure. In the dentin the dead tracts were seen below the cavities in all the groups. The ground section of the tooth did not show any zones which can be seen in the dental

caries because of alteration in the organic and inorganic materials. There was no difference observed in the ground section of both the groups.

Histopathological analysis:

Overheating of the teeth can cause pulp damage and inflammatory response of the pulp tissue. The previous studies by Glocknerk N in 1999[49] in determining the pulpal response showed increase in temperature was recorded with laser. The pulpal tissue directly exposed to laser treatment shows no bleeding. After direct exposure with the 34mJ/pulse no inflammation or resorption was found by Kimura et al in 2003[77] .A good healing capacity of the laser exposed pulp tissue was observed with subsequent formation of dentin bridges and reparative dentin by Jayawardena in 2009[51.] *In vivo* observation by Nair e al in 2003[69] showed direct dentin apposition adjacent to the laser irradiated sites. In our study the histopathological examination of the decalcified section shows that the margins of the cavity appeared regular and normal in both the groups. The structure of the dentinal tubules appeared normal without any changes like vacuolization.

The odontoblastic layers in both the groups were normal in appearance. There were no signs of odontoblastic degeneration and shrinkage. In laser group 3 samples showed arte factual separation of odontoblastic layer in one area. The cells of the odontoblast were normal as observed in the above studies. There were no

vascular changes noted in the pulp tissue. There was no hyperaemia of pulp. The blood vessels in the pulp tissue were normal in appearance in all groups. The pulp stone in the laser group (20%) and bur group (12.5%) was an incidental finding.

The connective tissue showed normal collagen fibres arranged in the loose extra cellular matrix. There were no inflammatory cells observed in the pulp chamber. In some areas inflammatory cells were seen apically. There was no difference observed regarding the pulpal response of the teeth between the groups. Hence with the ideal laser parameters there was no pulpal damage of the teeth was seen in the conventional cavity preparation between laser groups and bur groups.

In our study, there were no morphological alterations of the enamel and dentin between laser and bur groups. The histopathological results showed that the laser is effective in the removal of dental hard tissue with minimal damage to the pulp tissues.

- The cavity preparation between the conventional methods and lasers were compared in 18 patients with 40 samples.
- The patient acceptance was good with the laser procedures like that of the conventional methods.
- The time required to prepare the cavity with laser was (6.50±1.63 minutes) slight longer than the conventional methods (4.65±0.93minutes).
- On the Scanning Electron Microscope analysis the surface of lased cavity was irregular in appearance with absence of smear layer.
- The ground sections of the teeth in both methods showed no differences.
- The histopathological study of the decalcified sections showed that there was no demonstrable pulpal hyperaemia and inflammation in both the methods.

In conclusion, there were no significant differences in the ground section and decalcified section with respect to laser and bur. However with Scanning Electron Microscope there were significant surface changes between the groups with the laser showing a surface that is beneficial for adhesive restoration and features which suggest that they could reduce the post operative hypersensitivity.

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ANNEXURE -1

Time schedule

S. No.	Ref .no	Sample no.	Name	Age/sex	Tooth	Technique	Date	Starting time	Finishing time	Duration
1	4278	Sample 28	Maria	24/f	23	Bur	10-5-11	19.10	19.14	04mts
2	4279	Sample 35	Parveen	19/f	14	Bur	10-5-11	19.30	19.35	05mts
3	4280	Sample 29	Maria	24/f	13	Bur	16-5-11	19.00	19.06	06mts
4	4298	Sample 6	Zareena	17/f	34	Laser	11-6-11	11.45	11.51	06mts
5	4299	Sample 7	Zareena	17/f	24	Laser	11-6-11	11.53	12.00	07mts
6	4300	Sample 8	Zareena	17/f	14	Laser	11-6-11	12.02	12.07	05mts
7	4301	Sample 9	Zareena	17/f	44	Laser	11-6-11	12.09	12.15	06mts
8	4302	Sample 10	Jesanth	13/m	34	Laser	11-6-11	11.10	11.16	06mts
9	4303	Sample 11	Jesanth	13/m	24	Laser	11-6-11	11.17	11.24	07mts
10	4304	Sample 12	Jesanth	13/m	14	Laser	11-6-11	11.25	11.30	05mts
11	4305	Sample 13	Jesanth	13/m	44	Laser	11-6-11	11.31	11.36	05mts
12	4311	Sample 30	Krithika	19/f	34	Bur	15-6-11	18.30	18.34	04mts
13	4312	Sample 31	Yuvashri	15/f	34	Bur	15-6-11	19.00	19.05	05mts
14	4313	Sample 32	Pavithra	15/f	14	Bur	16-6-11	19.00	19.06	06mts
15	4314	Sample 33	Pavithra	15/f	24	Bur	16-6-11	19.08	19.12	04mts
16	4319	Sample 34	Jayanthi	20/f	14	Bur	22-6-11	19.10	19.04	04mts
17	4334	Sample 14	Pavitra S	15/f	24	Laser	3-7-11	10.35	10.41	06mts
18	4335	Sample 15	Pavitra S	15/f	14	Laser	3-7-11	10.43	10.50	07mts
19	4336	Sample 5	Mubeena	15/f	14	Laser	5-7-11	11.00	11.05	05mts
20	4337	Sample 4	Mubeena	15/f	24	Laser	5-7-11	11.06	11.11	05mts
21	4338	Sample 1	Mubeena	15/f	44	Laser	5-7-11	11.13	11.17	04mts
22	4339	Sample 2	Mubeena	15/f	34	Laser	5-7-11	11.18	11.26	06mts
23	4340	Sample 25	Jayanthi	20/f	34	Bur	5-7-11	18.30	18.34	04mts
24	4385	Sample 21	Shanthi	17/f	44	Bur	8-8-11	19.00	19.03	03mts
25	4386	Sample 24	Shanthi	17/f	34	Bur	8-8-11	19.04	19.08	04mts
26	4387	Sample 23	Shanthi	17/f	24	Bur	8-8-11	19.09	19.14	05mts
27	4388	Sample 22	Shanthi	17/f	14	Bur	8-8-11	19.15	19.20	05mts

S No.	Ref .no	Sample no.	Name	Age/sex	Tooth	Technique	Date	Starting time	Finishing time	Duration
28	4401	Sample 26	Fathima	18/f	44	Tc bur	24-8-11	18.00	18.04	04mts
29	4402	Sample 27	Fathima	18/f	14	Tc bur	24-8-11	18.06	18.11	05mts
30	4404	Sample 40	Monica	17/f	34	Bur	24-8-11	18.45	18.50	05mts
31	4405	Sample 26	Monica	17/f	24	Bur	24-8-11	18.51	18.56	05mts
32	4406	Sample 37	Monica	17/f	14	Bur	24-8-11	18.58	19.02	04mts
33	4407	Sample 38	Manoj	20/m	14	Bur	24-8-11	19.30	19.37	07mts
34	4408	Sample 39	Lakshmi	18/f	14	Bur	24-8-11	20.00	20.04	04mts
35	4418	Sample 3	Sultana	16/f	14	Laser	07-9-11	10.30	10.36	06mts
36	4419	Sample 16	Sultana	16/f	44	Laser	07-9-11	10.37	10.45	08mts
37	4420	Sample 17	Sultana	16/f	24	Laser	07-9-11	10.47	10.57	10mts
38	4421	Sample 18	sudha	18/f	44	Laser	07-9-11	19.00	19.06	06mts
39	4422	Sample 19	Raji	20/f	24	Laser	07-9-11	11.30	11.40	10mts
40	4423	Sample 20	Raji	20/f	44	Laser	07-9-11	11.42	11.50	08mts

ANNEXURE -2
SEM table

S. No	Reference no.	Sample no.	technique	Cavity margins	Cavity surfaces	Smear layer	Cracking	Surface roughness	Cavity base	Dentinal openings	Micro cracks	others
1	4336	Sample 5	Laser	ID	IR	A	P	S	IR	OB	P	Glazing
2	4337	Sample 4	Laser	ID	IR	A	P	S	IR	OB	P	Glazing
3	4338	Sample 1	Laser	ID	IR	A	P	S	IR	OB	P	Craters
4	4339	Sample 2	Laser	ID	IR	A	P	S	IR	OB	P	Glazing
5	4340	Sample 25	Bur	WD	R	P	P	M	SM	P	P	
6	4385	Sample 21	Bur	WD	R	P	A	M	SM	P	P	Craters
7	4386	Sample 24	Bur	WD	R	P	P	M	SM	P	P	
8	4387	Sample 23	Bur	WD	R	P	P	M	SM	P	P	
9	4388	Sample 22	Bur	WD	R	P	A	M	SM	P	P	
10	4401	Sample 26	TC Bur	WD	R	P	A	M	SM	P	P	
11	4402	Sample 27	TC Bur	WD	R	P	A	M	SM	P	P	
12	4418	Sample 3	Laser	ID	IR	A	P	S	IR	OB	P	

Cavity margins - **D**-well defined, **ID**- ill defined.
Cavity surface - **R**-regular .**IR**-irregular
Cavity base - **SM**-smooth, **IR**- irregular.
Surface roughness - **M**- mild, **S**-severe
Smear layer - **P**-present, **A**- indistinct
Cracking - **P**-present, **A**- absent.
Dentinal openings - **P**-present, **OB**-obliterated.

ANNEXURE -3

Decalcification table

Sample no.	Slide no.	Structure of dentinal tubules			Changes in odontoblast layer			Changes in vascular tissue	Changes in connective tissues			Others
		Margin	Staining of dentinal tubules	Vacuolization in dentinal tubules	Degeneration of odontoblastic layer	Shrinkage of odontoblastic layer	Odontoblastic cell changes	Pulp hyperaemia	Inflammatory cells	matrix		
									Present /absent	Fibres	arrangement	
28	4278	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
35	4279	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
29	4280	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
6	4298	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
7	4299	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
8	4300	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
9	4301	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
10	4302	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
11	4303	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
12	4304	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	Pulp stone
13	4305	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	Pulp stone
30	4311	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
31	4312	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	Pulp stone
32	4313	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
33	4314	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
34	4319	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
14	4334	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	
15	4335	Regular	Normal	Absent	Absent	Absent	Absent	Absent	Absent	collagen	Loose	

Margins – Regular, Irregular
Staining- Normal, Abnormal
Vacuolization- Present, Absent.

Degeneration of odontoblast- Present, Absent.
Shrinkage of odontoblast- Present, Absent
Pulpal hyperaemia- Present, Absent

Inflammatory cells-Present, Absent.
Matrix –fibrous- Collagen, Others.
Arrangement – Loose, Dense.

ANNEXURE -4

Ground section

S. No	Sample No	Ref No	Technique	Enamel			DEJ	Dentin	
				Structure	Tufts	Lamellae		Structure	Dead tracts
1	16	4419	Laser	Normal	Present	Present	Normal	Normal	Present
2	17	4420	Laser	Normal	Present	Present	Normal	Normal	Present
3	18	4421	Laser	Normal	Present	Present	Normal	Normal	Present
4	19	4422	Laser	Normal	Present	Present	Normal	Normal	Present
5	20	4423	Laser	Normal	Present	Present	Normal	Normal	Present
6	36	4405	Diamond Bur	Normal	Present	Present	Normal	Normal	Present
7	37	4406	Diamond Bur	Normal	Present	Present	Normal	Normal	Present
8	38	4407	Diamond Bur	Normal	Present	Present	Normal	Normal	Present
9	39	4408	Diamond Bur	Normal	Present	Present	Normal	Normal	Present
10	40	4404	Diamond Bur	Normal	Present	Present	Normal	Normal	Present

CERTIFICATE

This is to certify that this dissertation titled " **EVALUATION AND COMPARISON OF THE MORPHOLOGICAL AND HISTOPATHOLOGICAL CHANGES INDUCED BY ER:YAG LASER AND BURS ON ENAMEL, DENTIN AND PULP TISSUE**" is a bonafide dissertation performed by **SHAIK MOHAMED SHAMSUDEEN.S.S** under our guidance during the postgraduate period 2009-2012.

This dissertation is submitted to THE TAMILNADU DR. M.G.R MEDICAL UNIVERSITY, in partial fulfillment for the degree of **MASTER OF DENTAL SURGERY** in **ORAL PATHOLOGY AND MICROBIOLOGY, BRANCH VI**. It has not been submitted (partial or full) for the award of any other degree or diploma.

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The dissertation topic titled 'Evaluation and comparison of morphological and histopathological changes induced by ER:YAG laser and burs on enamel dentin and pulp tissue' submitted by Dr.Shaik Mohamed Shamsudeen .S. S has been approved by the Institutional Review Board of Ragas Dental College and Hospital on 14th March 2011.

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Secretary,

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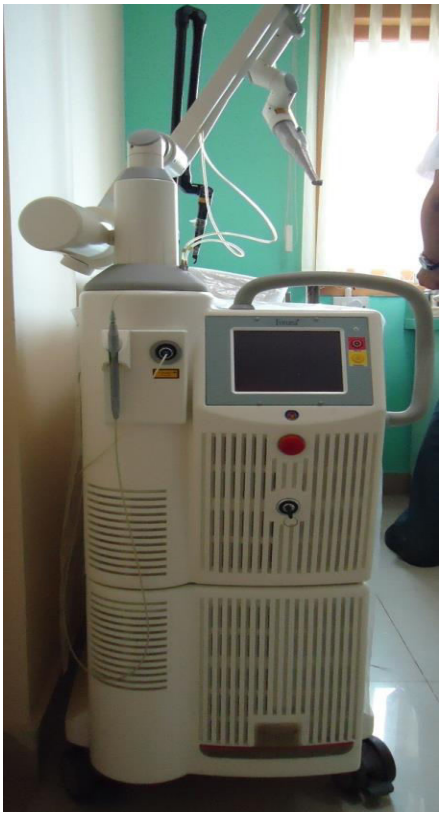
Dr.S.Ramachandran

Chairman,

Ragas , IRB

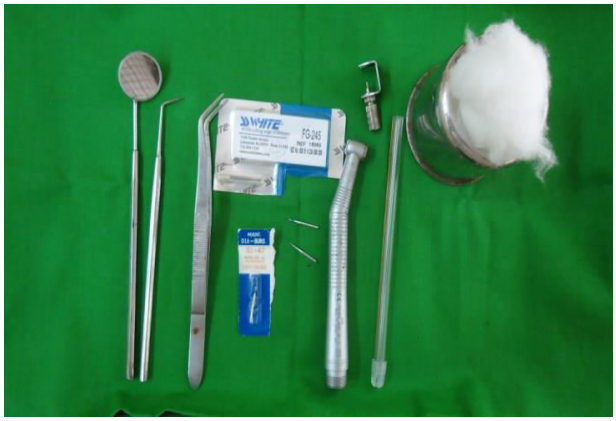
ARMAMENTARIUM USED IN CAVITY PREPARATION

Fig 1-a



Er:YAG laser

Fig 1-b



Conventional methods using burs

Fig 2

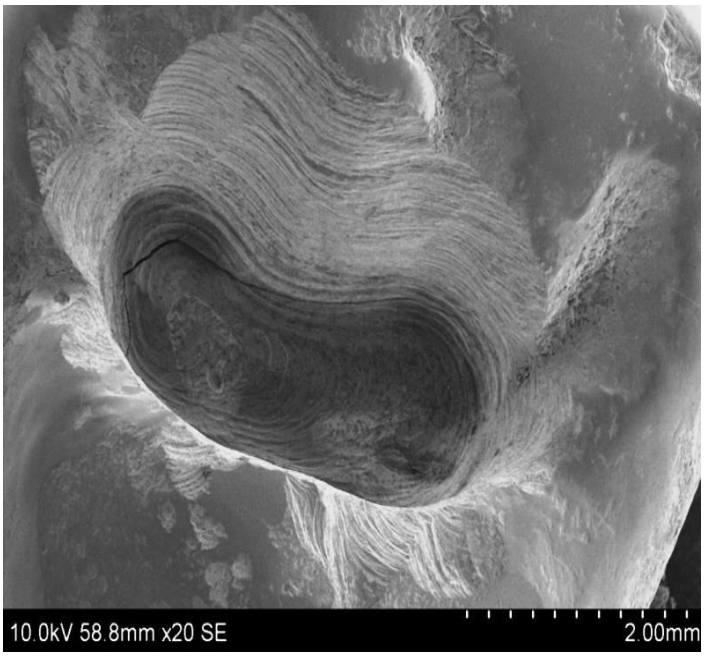
Scanning Electron Microscope

Fig 3

Hard tissue microtome

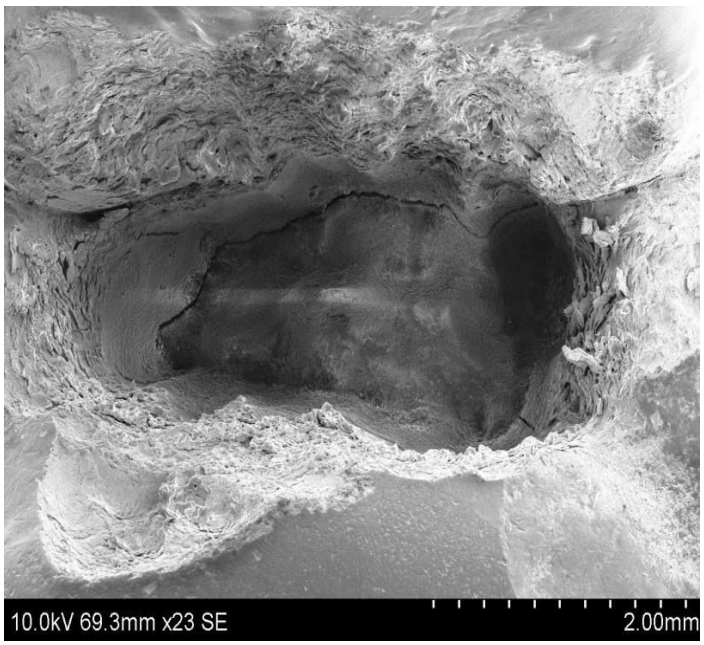
SCANNING ELECTRON PHOTOMICROGRAPH

Fig -4 (sample 23): Cavity prepared by Bur



The occluso cavosurface margin of the cavity was well defined with regular surface

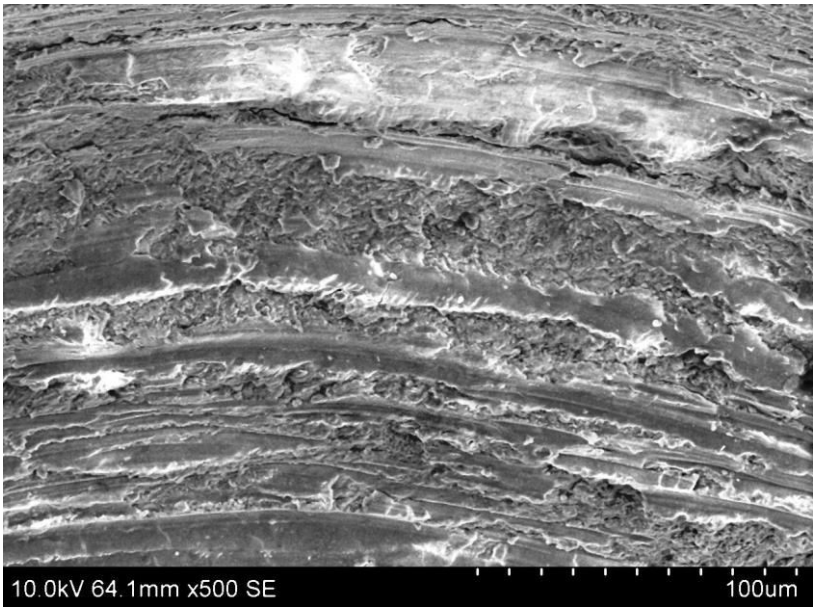
Fig -5 (sample 3) :Cavity prepared by Laser



The occluso cavosurface margins of the cavity was ill defined with irregular surface

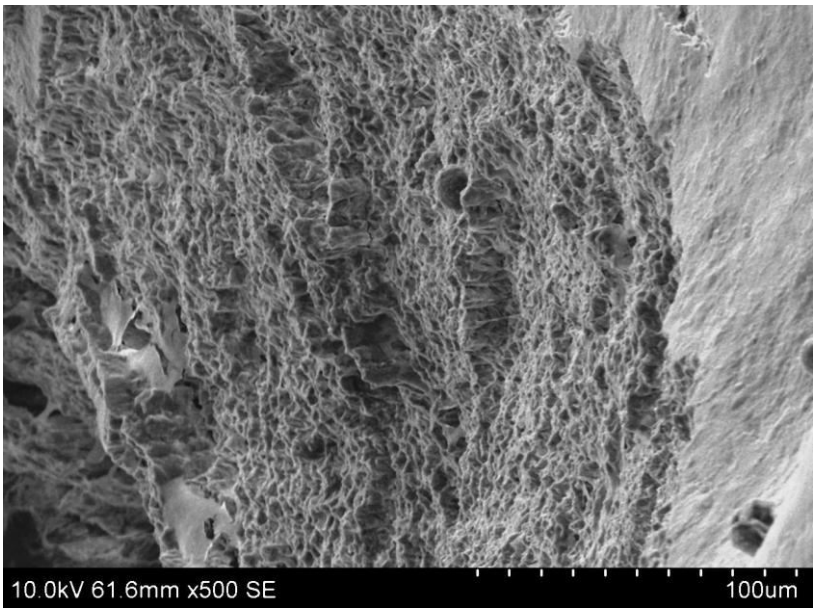
SCANNING ELECTRON PHOTOMICROGRAPH

Fig -6 (sample 23): Cavity prepared by Bur



The enamel wall of the cavity showed indentation of the bur with mild roughness

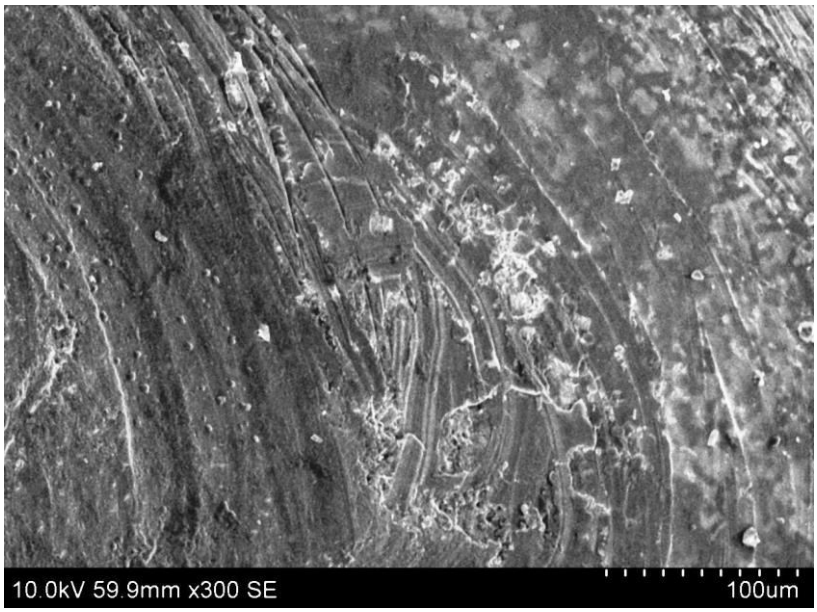
Fig -7(sample 2): Cavity prepared by Laser



The enamel wall of the cavity showed scaly appearance with sever roughness

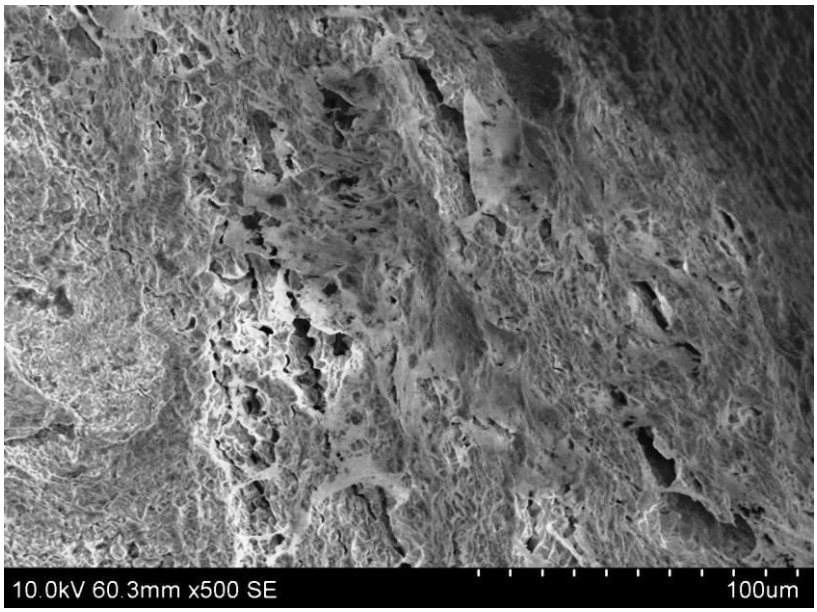
SCANNING ELECTRON PHOTOMICROGRAPH

Fig- 8 (sample 22): Cavity prepared by Bur



The dentinal wall of the cavity showed opening of dentinal tubules and smear layer

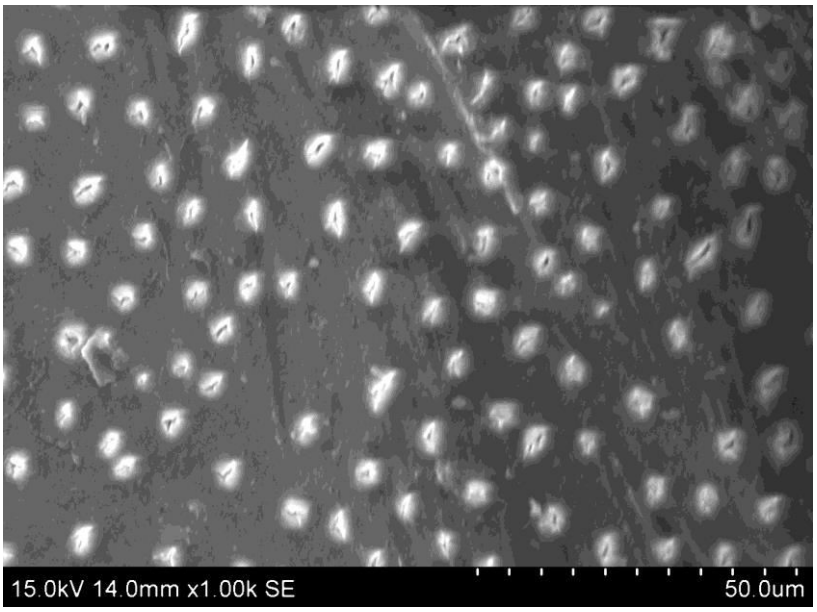
Fig- 9 (sample 1) :Cavity prepared by Laser



The dentinal wall showed indistinct smear layer and dentinal tubules.

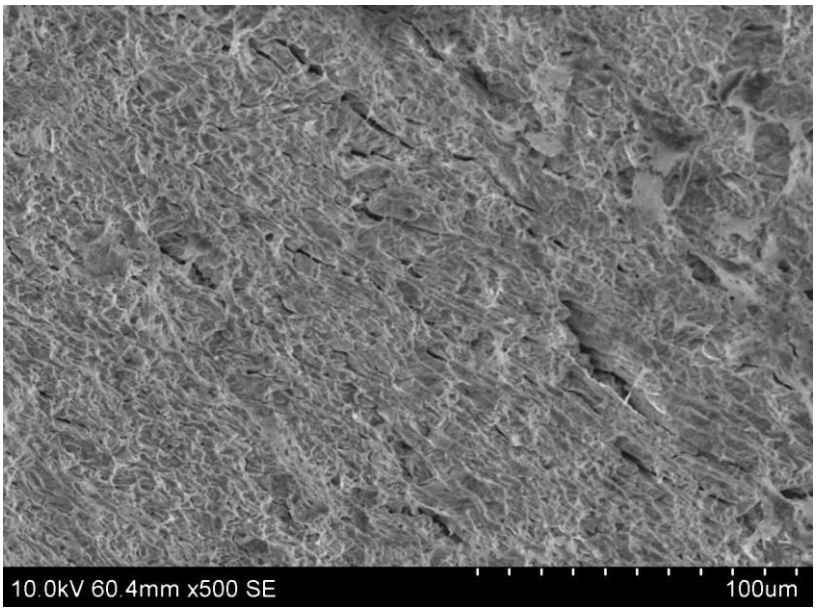
SCANNING ELECTRON PHOTOMICROGRAPH

Fig -10 (sample 25) Cavity prepared by Bur



The dentin showed smear layer with opening of the tubules

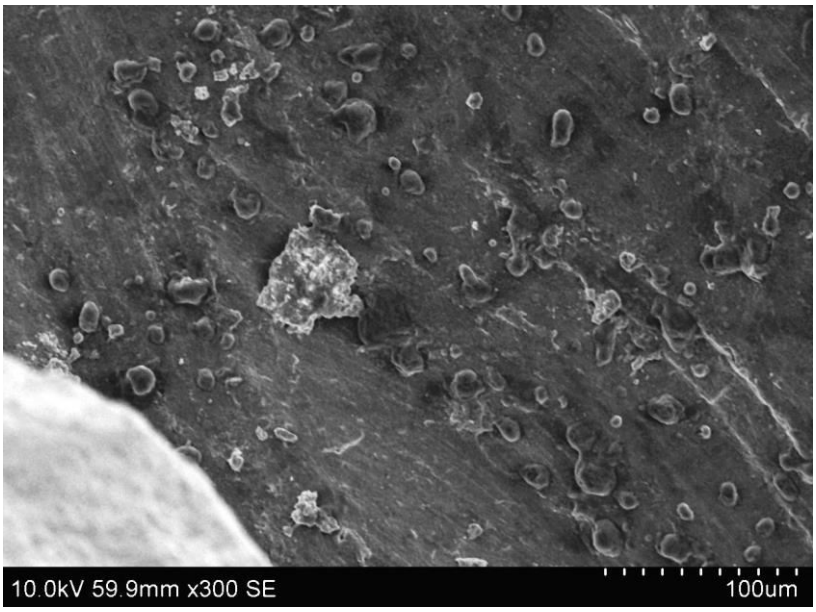
Fig -11 (sample 1): Cavity prepared by Laser



The dentin showed obliteration of dentinal tubules with indistinct of smear layer

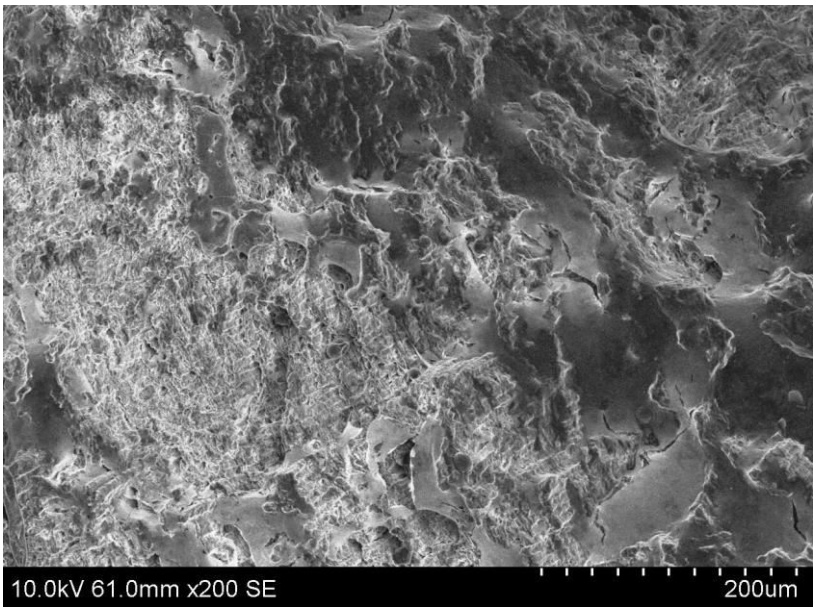
SCANNING ELECTRON PHOTOMICROGRAPH

Fig -12 (sample 21) Cavity prepared by Bur



The base of the cavity showed flat appearance with smooth surface

Fig 13(sample 2): Cavity prepared by Laser

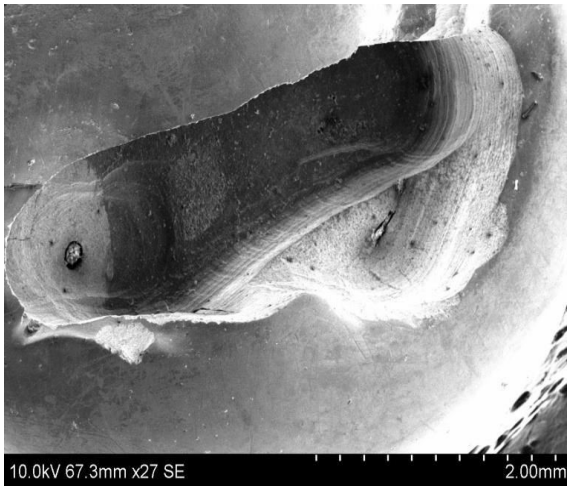


The cavity showed glazed areas in the base

SCANNING ELECTRON PHOTOMICROGRAPH

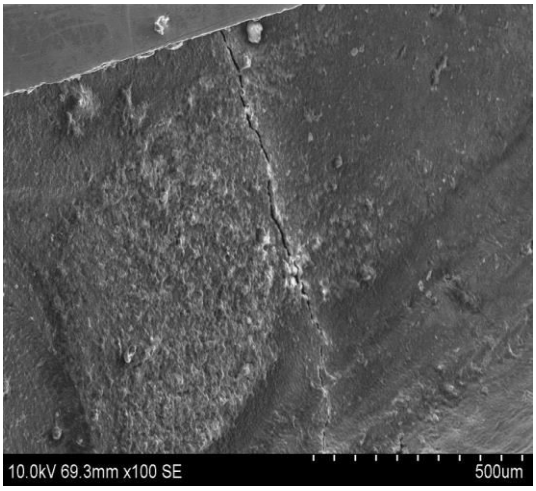
Cavity prepared by Tungsten carbide bur

Fig-14 (sample 26)



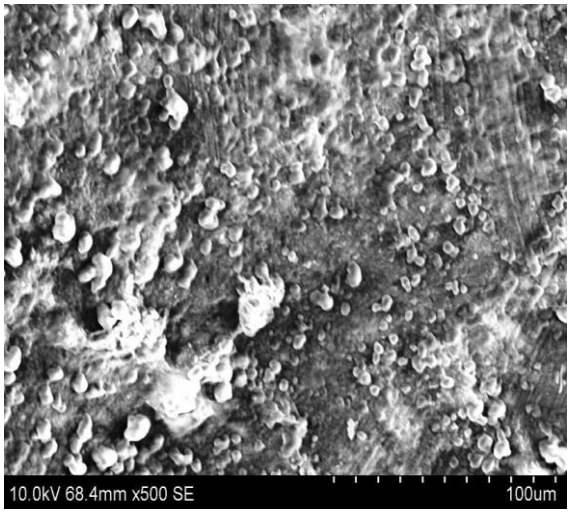
The cavosurface margin showed well defined with regular cavity surface

Fig-15 (sample 26)



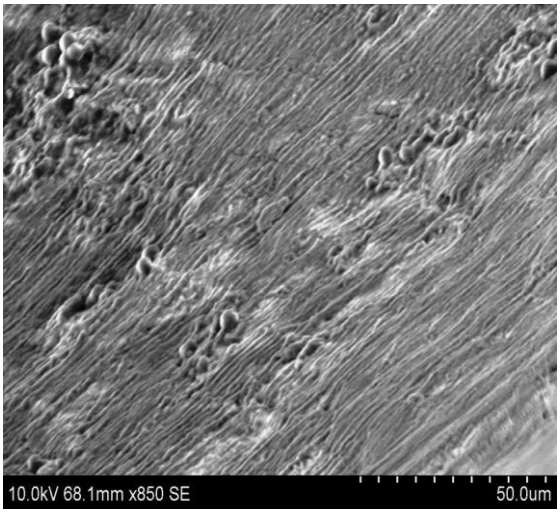
The dentin of the cavity showed smear layer with microcracks

Fig-16 (sample 27)



The base showed dentinal tubules with smear layer

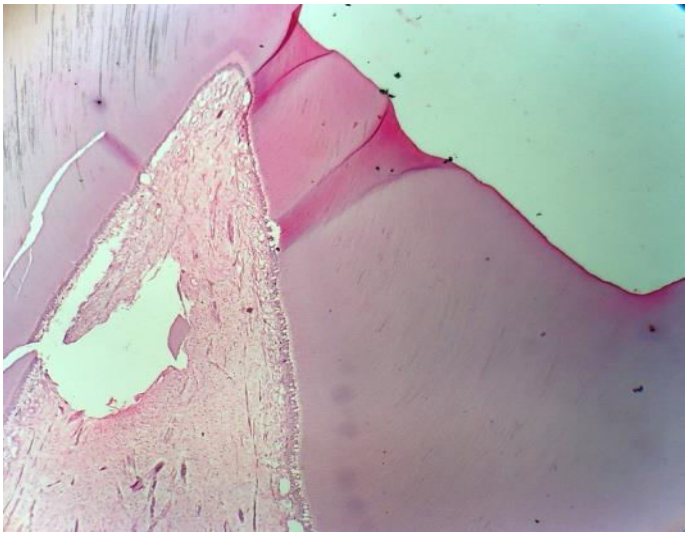
Fig-17 (sample 27)



The enamel wall showed regular cavity surface

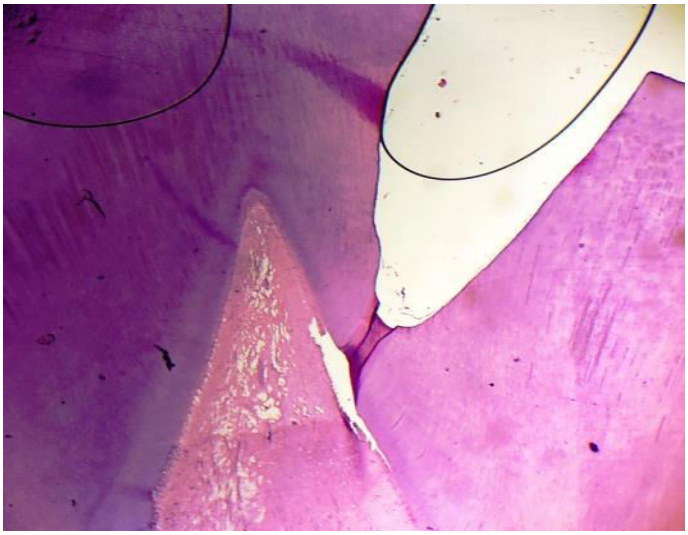
PHOTOMICROGRAPH OF HISTOPATHOLOGICAL FEATURES

Fig -18 (sample 29):Cavity prepared by bur



A well defined cavity margin with pulp tissue was seen

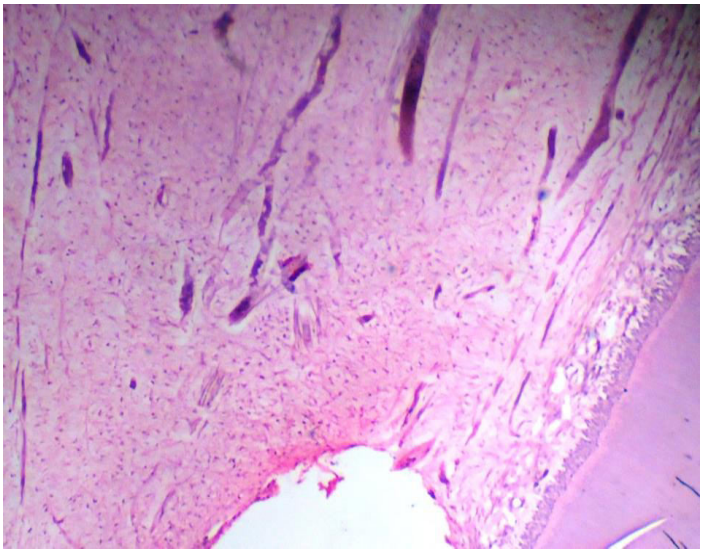
Fig -19 (sample 13):Cavity prepared by Laser



A well defined cavity margin with pulp tissue was seen.

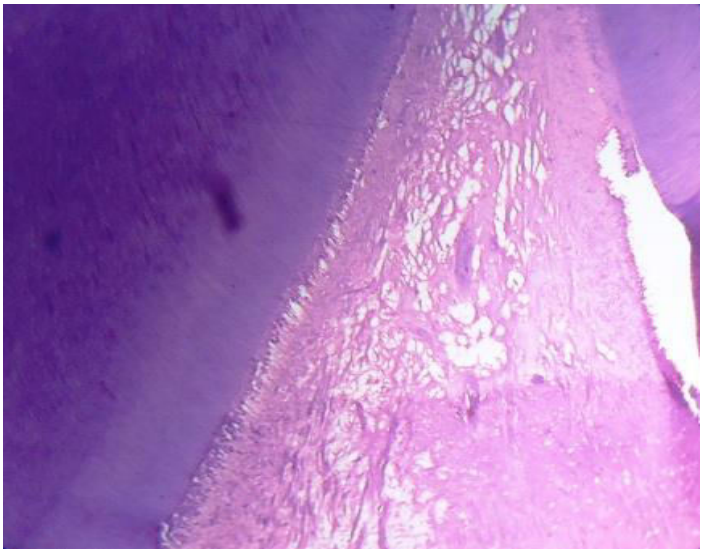
PHOTOMICROGRAPH OF HISTOPATHOLOGICAL FEATURES

Fig -20 (sample 29): cavity prepared by Bur



The dentinal tubules are lined by odontoblastic cells

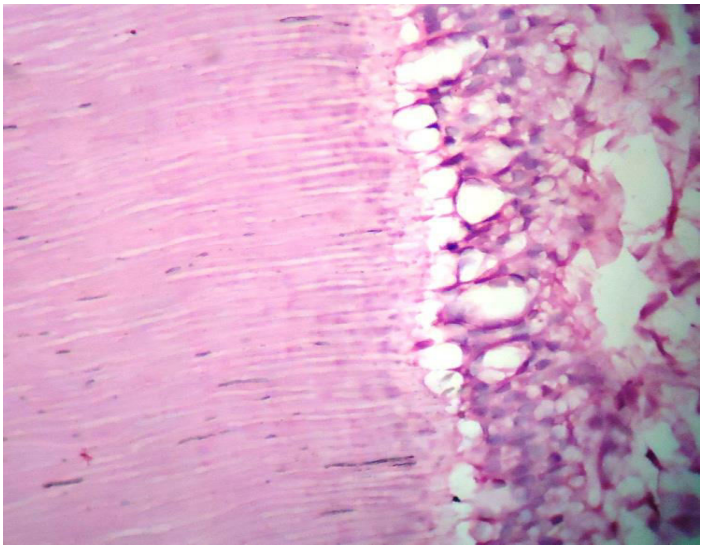
Fig -21 (sample 13):Cavity prepared by Laser



The dentinal tubules are lined by odontoblastic cells

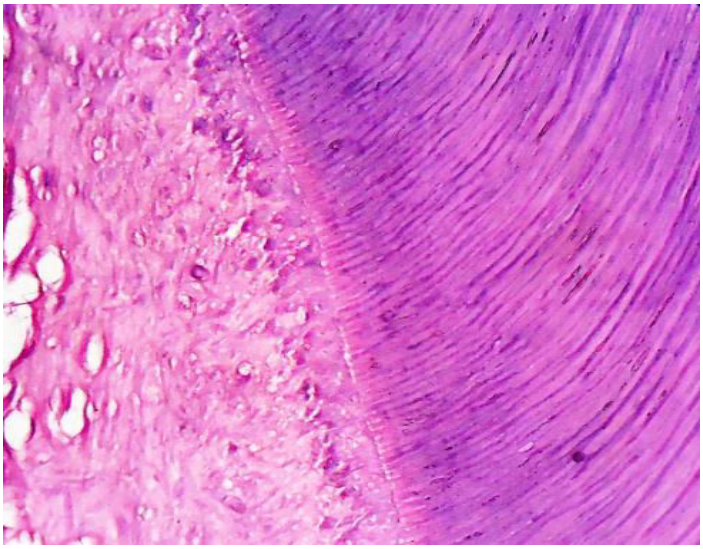
PHOTOMICROGRAPH OF HISTOPATHOLOGICAL FEATURES

Fig -22 (sample 29): Cavity prepared by Bur



The normal odontoblastic cells lining the pulp chambers were seen

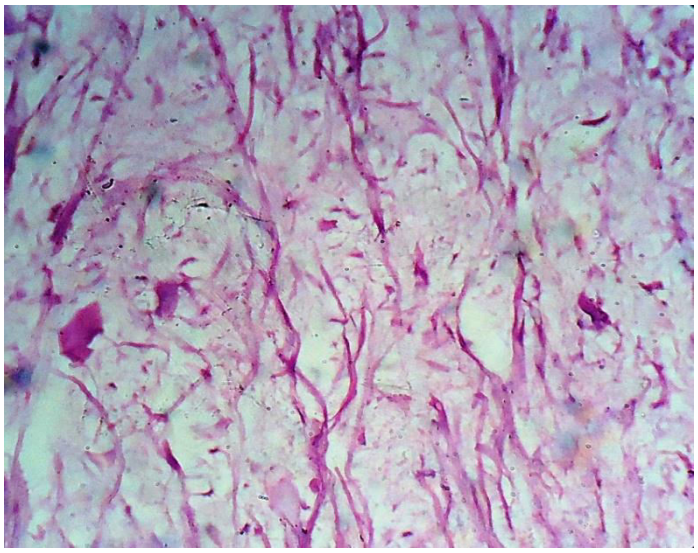
Fig -23 (sample 13): Cavity prepared by Laser



The normal odontoblastic cells lining the pulp chambers were seen

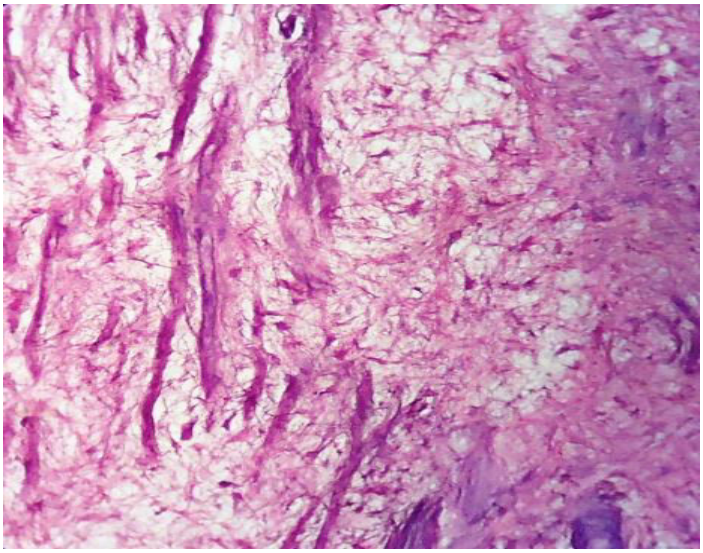
PHOTOMICROGRAPH OF HISTOPATHOLOGICAL FEATURES

Fig -24 (sample 29):Cavity prepared byBur



The pulp tissue showed normal collagen fibres with fibroblast cells

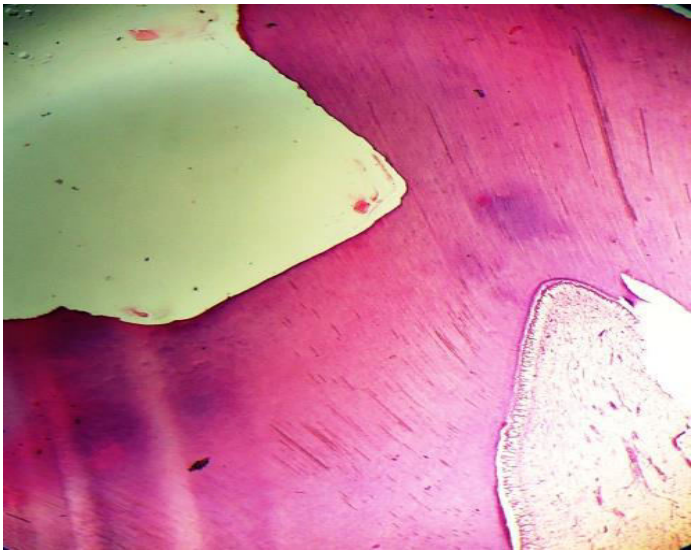
Fig -25 (sample 13): Cavity prepared by Laser



The pulp tissue showed normal collagen fibres with fibroblast cells

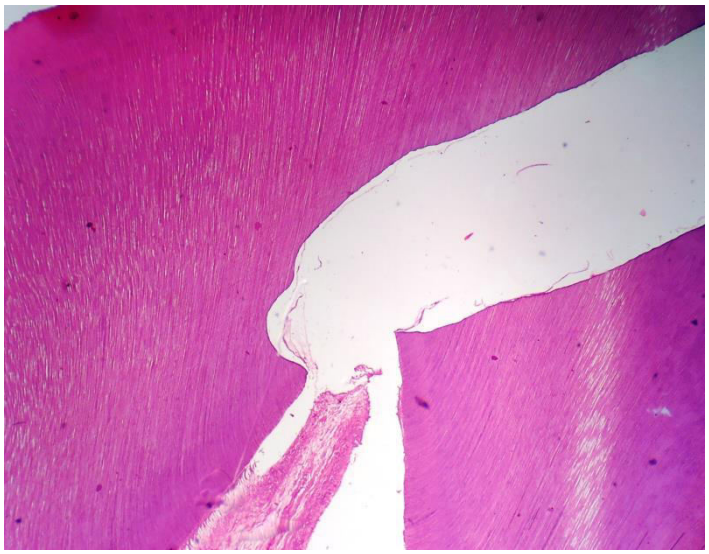
PHOTOMICROGRAPH OF HISTOPATHOLOGICAL FEATURES

Fig -26 (sample 31): Cavity prepared by Bur



A well defined cavity margin were seen in the dentin

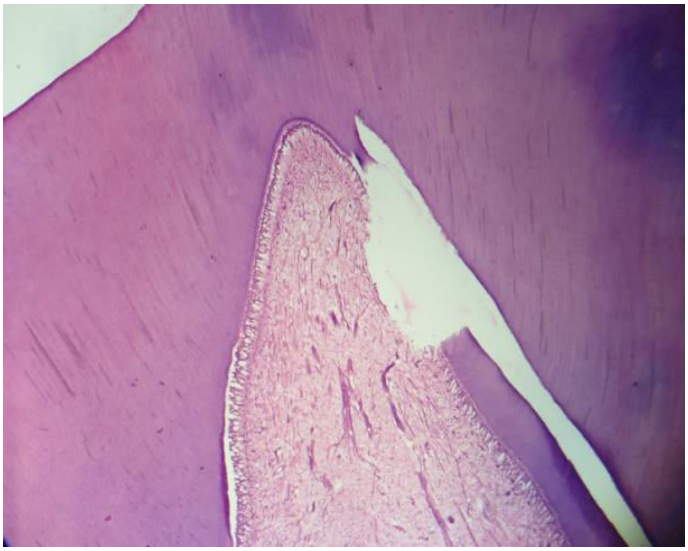
Fig -27 (sample 10): Cavity prepared by Laser



A well defined cavity margin were seen in the dentin.

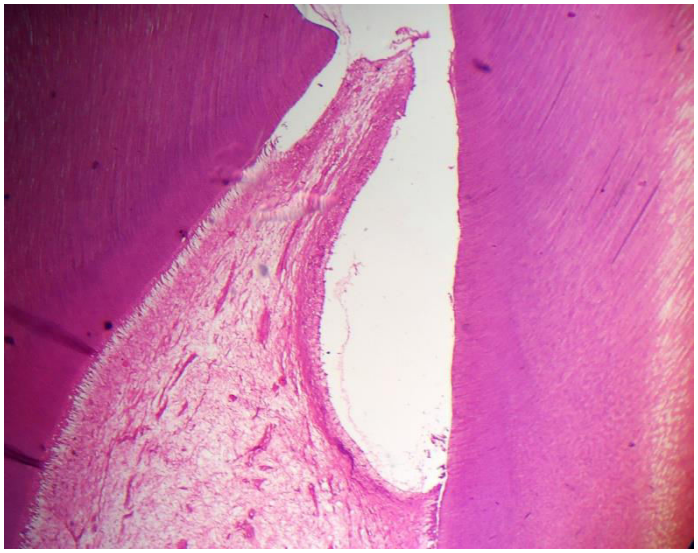
PHOTOMICROGRAPH OF HISTOPATHOLOGICAL FEATURES

Fig -28 (sample 31):Cavity prepared by Bur



One area shows artefactual separation of odontoblastic layer

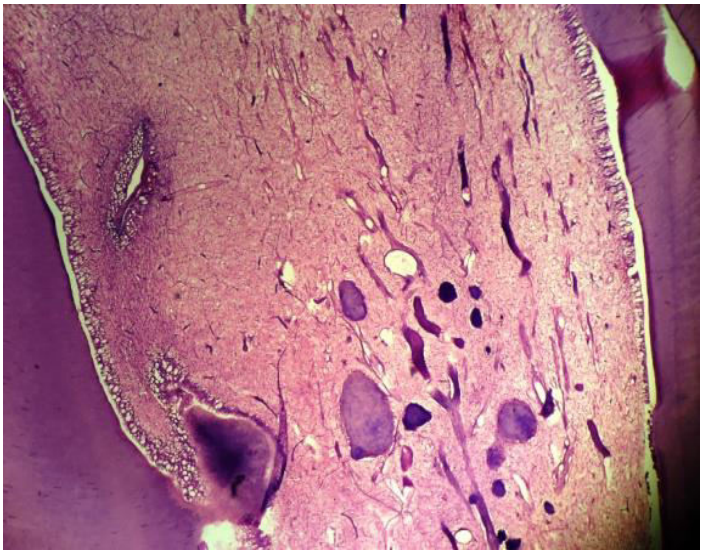
Fig -29 (sample 10):cavity prepared Laser



Artefactual separation of pulp tissue from dentin were seen

PHOTOMICROGRAPH OF HISTOPATHOLOGICAL FEATURES

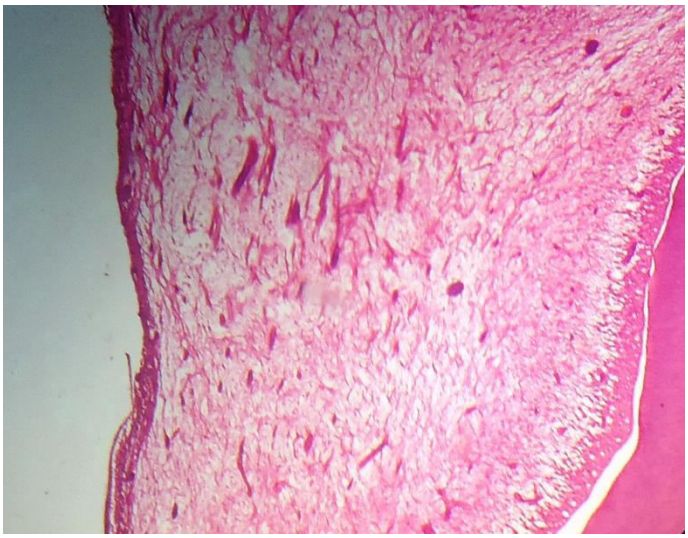
Fig- -30 (sample 31): Cavity prepared by Bur



The pulp chamber showed pulp tissues with normal collagen fibres.

Pulp stones were seen

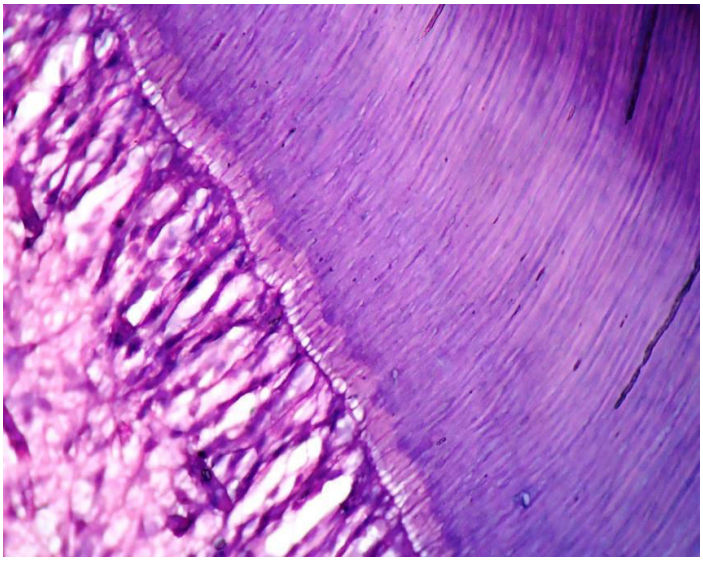
Fig -31 (sample 10): Cavity prepared by Laser



The pulp chamber showed pulp tissue with normal collagen fibres

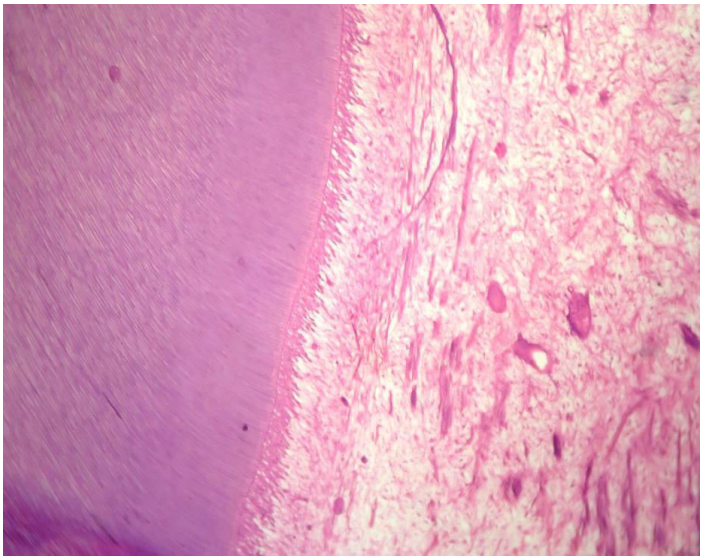
PHOTOMICROGRAPH OF HISTOPATHOLOGICAL FEATURES

Fig -32 (sample 31): Cavity prepared by Bur



The odontoblastic cells with the dentinal tubules were seen

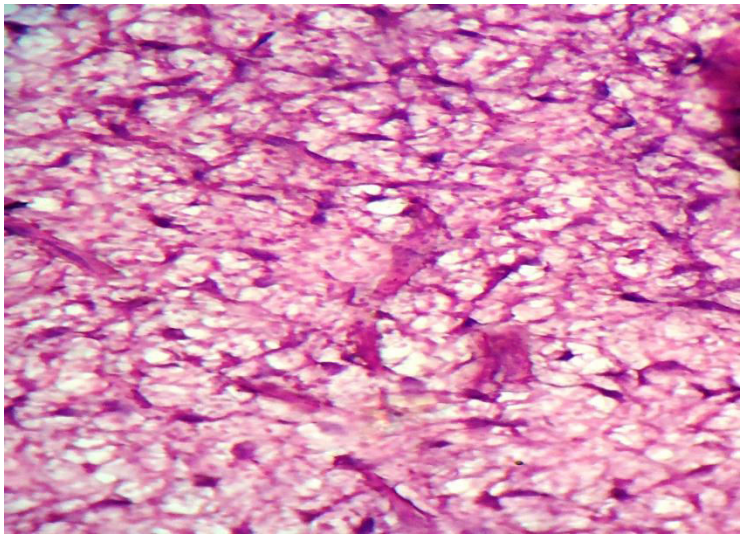
Fig 33 (sample 10): Cavity prepared by Laser



The odontoblastic cells with the dentinal tubules were seen

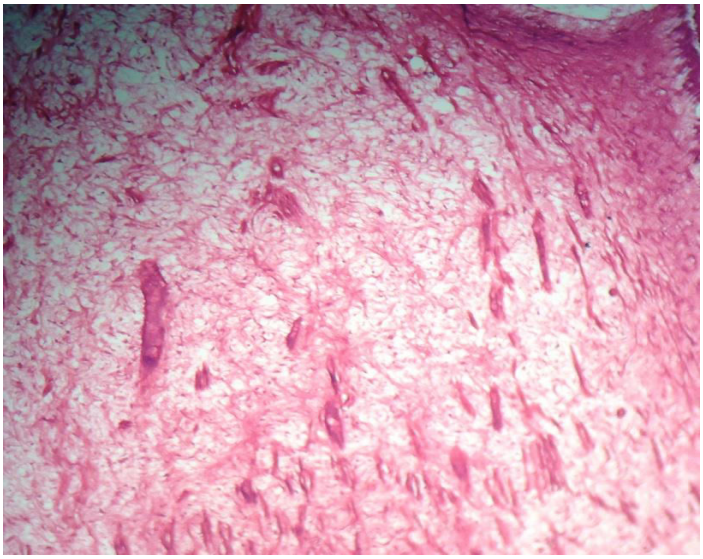
PHOTOMICROGRAPH OF HISTOPATHOLOGICAL FEATURES

Fig- 34 (sample 31): Cavity prepared by Bur



The pulp tissue showed fibroblastic cells with normal collagen fibres

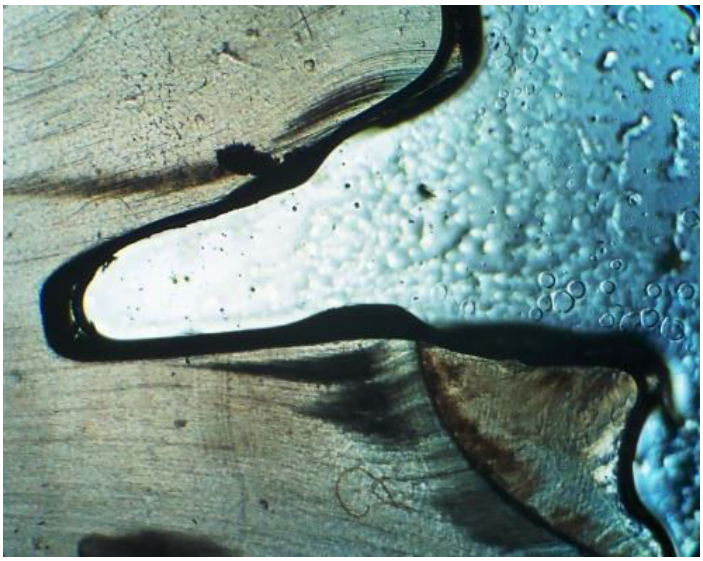
Fig -35 (sample 10): Cavity prepared by Laser



The pulp tissue showed fibroblastic cells with normal collagen fibres

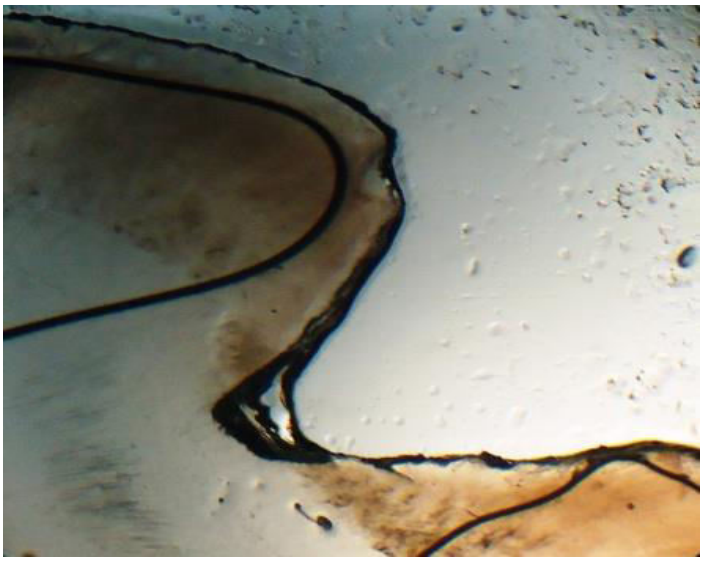
PHOTOMICROGRAPH OF GROUND SECTION OF TEETH

Fig -36 (sample 40): Cavities prepared by Bur



The ground section showed defined cavity margins

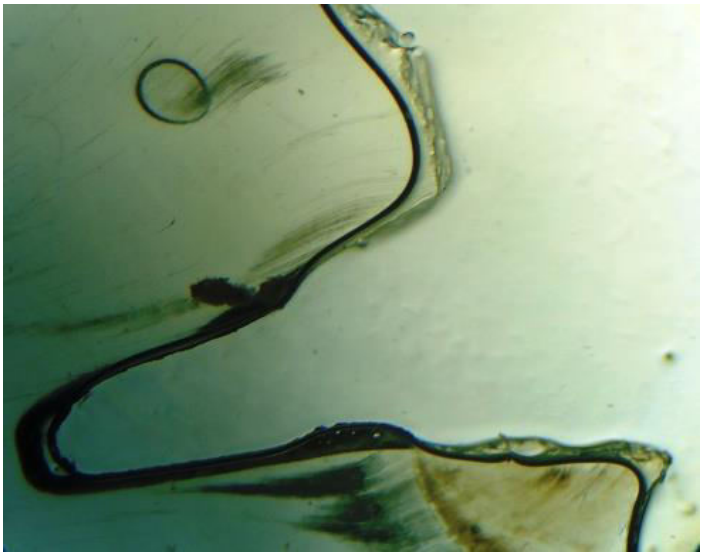
Fig -37 (sample 16): Cavity prepared by Laser



The ground section showed defined cavity margins

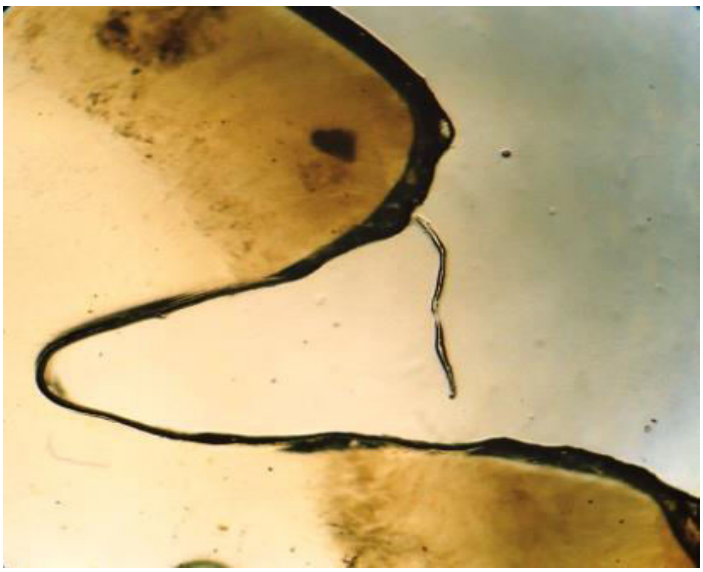
PHOTOMICROGRAPH OF GROUND SECTION OF TEETH

Fig -38 (sample 40): Cavity prepared by Bur



The enamel appears normal

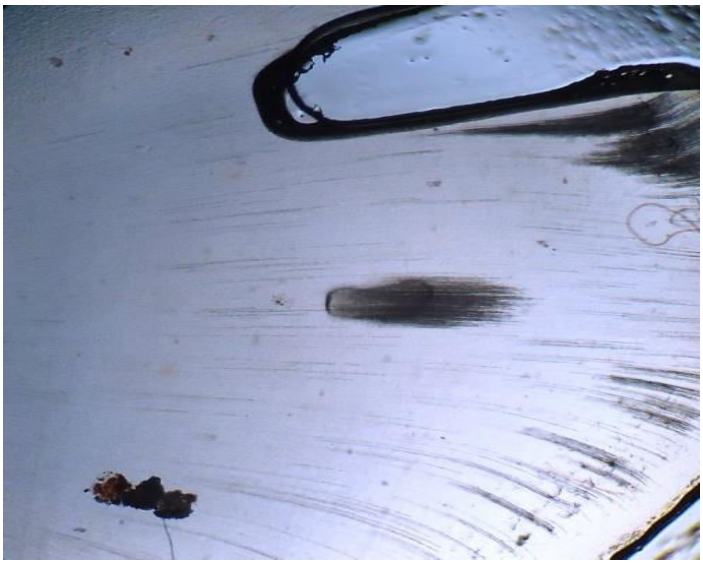
Fig -39 (sample 16): Cavity prepared by Laser



The enamel appears normal

PHOTOMICROGRAPH OF GROUND SECTION OF TEETH

Fig 40(sample 40): Cavity prepared by Bur



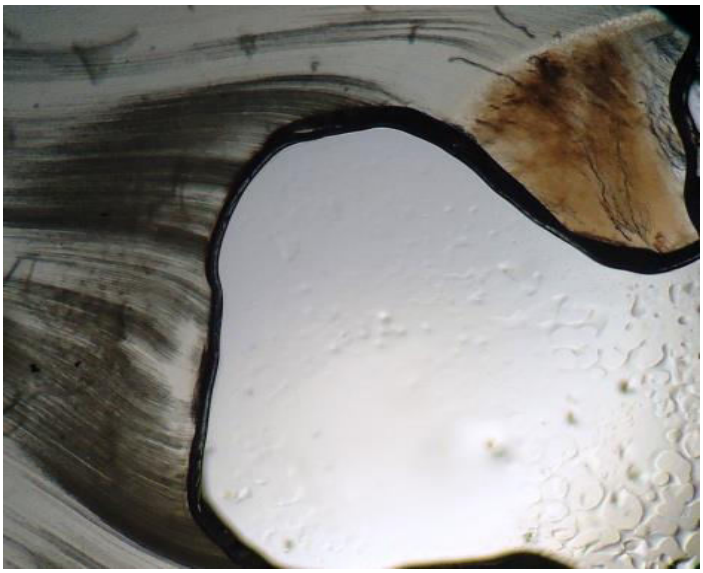
The dentin showed dead tracts

Fig -41 (sample 16): Cavity prepared by Laser

The dentinoenamel junction was scalloped in nature.

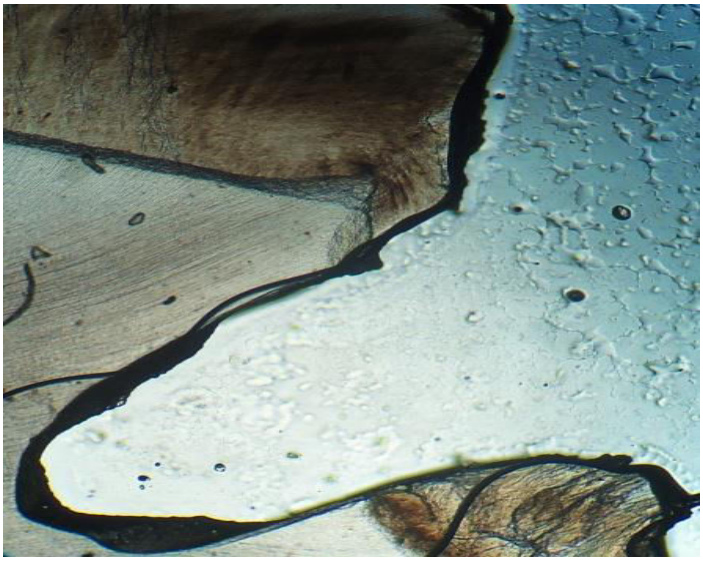
PHOTOMICROGRAPH OF GROUND SECTION OF TEETH

Fig -42 (sample 36): Cavity prepared by Bur



The section showed well defined cavity margin

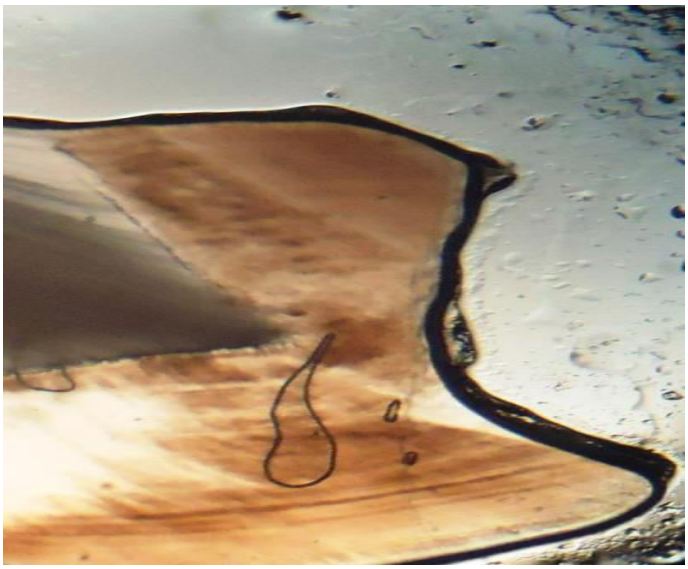
Fig 43(sample 18): Cavity prepared by Laser



The section showed well defined margin

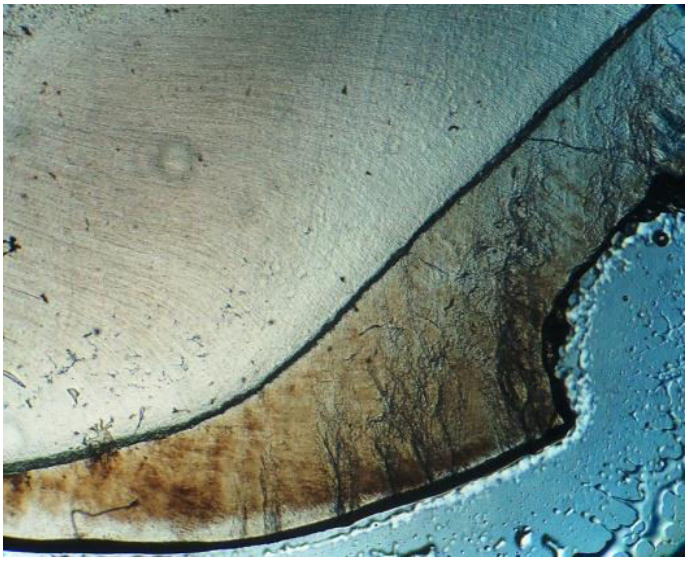
PHOTOMICROGRAPH OF GROUND SECTION OF TEETH

Fig -44 (sample 36): Cavity prepare by bur



The enamel appears normal in structure

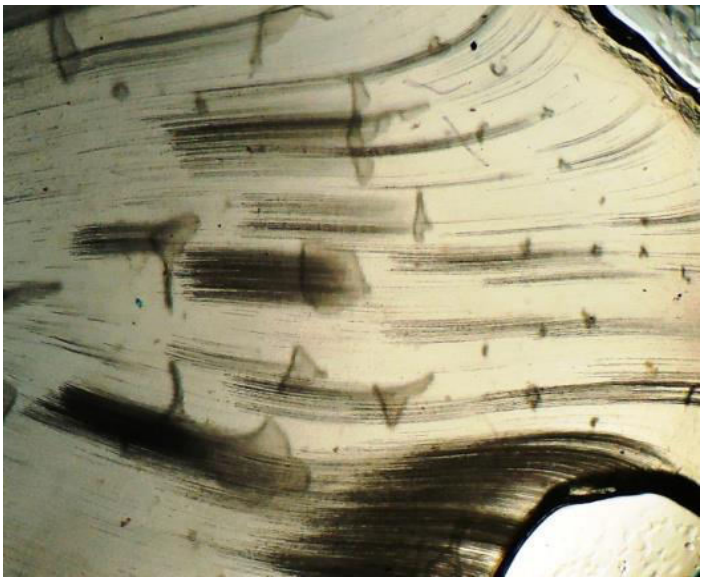
Fig -45 (sample 18): Cavity prepared by Laser



The enamel appears normal in structure

PHOTOMICROGRAPH OF GROUND SECTION OF TEETH

Fig- 46 (sample 36): Cavity prepared by Bur



The dentin showed dead tracts

Fig -47 (sample 18): Cavity prepared by Laser

The dentin structures are normal in appearance

Table 1: Gender distribution of samples in the study groups

Groups	Teeth no.	Male		Female	
Overall n=40	Upper teeth n= 24 Lower teeth n= 16	5	12.5%	35	87.5%
Group 1-laser n=20	Upper teeth n=10 Lower teeth n= 10	4	20%	16	80%
Group 2-bur n=18	Upper teeth n= 13 Lower teeth n=5	1	5.5%	17	94.5%
Group 3-TC bur n=2	Upper teeth n= 1 Lower teeth n= 1	0	0	2	100%

Graph 1: Gender distribution of samples in % in the study groups

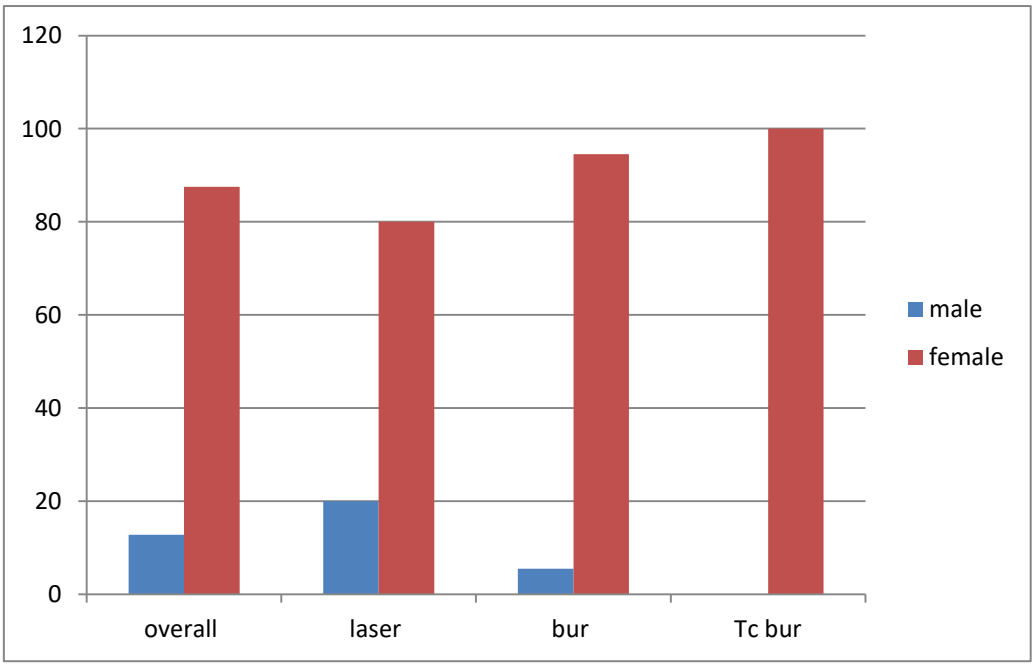


Table 2: Time taken for cavity preparation in the study groups

	N	Mean	Std.Deviation	95% Confidence interval for Mean		p value
				Lower bound	Upper bound	
Group 1	20	6.5000	1.6384	5.7332	7.2668	.001
Group 2	18	4.6667	.9701	4.1842	5.1491	
Group 3	2	4.5000	.7071	-1.8531	10.8531	
Total	40	5.5750	1.6154	5.0584	6.0916	

The time duration is statistically significant (p=0.001)

Table 3: Comparison of time difference between the three groups

Group	Mean difference	95% confidence interval		p value
		Lower bound	Upper bound	
Laser & bur	1.8333*	.7619	2.9047	.000
Laser & TC bur	2.000	-.4456	4.4456	.127
Bur & TCbur	.1667	-2.2913	2.6246	.985

*The mean difference is significant at the .05 level

Table 4: SEM analysis between groups

Morphology	Findings	Group 1-Laser (n=5)	Group 2-Bur (n=5)	Group 3-Tungsten carbide bur (n=2)
Cavity margins	Well defined	-	5(100%)	2(100%)
	Ill defined	5 (100%)	-	
Cavity surface	Regular	-	5(100%)	2(100%)
	Irregular	5(100%)	-	-
Smear layer	Present	-	5(100%)	2(100%)
cracking	Present	5(100%)	5 (100%)	2(100%)
Surface roughness	Mild	-	5(100%)	2(100%)
	Severe	5(100%)	-	-
Cavity base	Smooth	-	5(100%)	2(100%)
	Irregular	5(100%)	-	-
Present of dentinal tubules	Present	-	5(100%)	2(100%)
Microcracks	Present	5(100%)	5(100%)	2(100%)
Glazing	Present	3(60%)	-	-
Craters	Present	1(20%)	1(20%)	-

Table 5: Histopathological analysis between groups

Histopathology	**Findings**	**Laser group (n=10)**	**Diamond Bur (n=8)**
Margin	Regular	10 (100%)	8(100%)
Staining of dentinal tubules	Normal	10 (100%)	8(100%)
Vacuolization in dentinal tubules	Absent	10 (100%)	8(100%)
Degeneration of odontoblastic layer	Absent	10 (100%)	8(100%)
Shrinkage of odontoblastic layer	Absent	10 (100%)	8(100%)
Odontoblastic cell changes	Absent	10 (100%)	8(100%)
Pulp hyperaemia	Absent	10 (100%)	8(100%)
Inflammatory cells	Absent	10 (100%)	8(100%)
Fibres	Collagen	10 (100%)	8(100%)
Arrangement	Loose	10 (100%)	8(100%)
Pulp stones	Present	2 (20%)	1(12.5%)

Table -6: Analysis of the morphology by ground section between groups

Morphology	Findings	Laser group (n=5)	Diamond Bur (n=5)
Enamel structure	Normal	5 (100%)	5 (100%)
Tufts	Normal	5 (100%)	5 (100%)
Lamellae	Normal	5 (100%)	5 (100%)
Dentinoenamel junction	Normal	5 (100%)	5 (100%)
Dentin structure	Normal	5 (100%)	5 (100%)
Dead tracts	Present	5 (100%)	5 (100%)