Biomechanical Effect of Nd.YAG Laser Ablation on Commercially Pure Titanium Dental Implant (In Vivo Study)

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Abstract: ***Background**: A lot of studies have been done on dental implant that aim to fasilitate bone formation and enhance osseointegration and hence reduce the unloading period after insertion of dental implant which may last for 3-6 months render the patient be disturb and uncomfortable. **Aims of the study**: This study evaluate the effect of modification of dental implant topography by laser ablation on the force needed to remove implant from bone and osseointegration. **Materials and methods**: At first pilot study was done in order to select the energy of Nd.YAG laser, according to roughness measurement and SEM feature of Ti surface Thirty six screw like implants were prepared from cpTi .Eighteen implants were ablated with Nd.YAG laser to get optimal uniform roughness ,and the other 18 implants are left machined .The implants were implant in rabbit tibia ,each rabbit tibia received 2 implants ;one ablated implant (as an experimental) and the other machined implant (as a control).After 2 and 4 weeks of healing ,the removal torque was measured to evaluate the bone strength between implant and bone for 8 implants and histological analysis is done for one implant for each interval period. **Results**: The results revealed that there was high significant difference between removal torque of the laser ablated implant and that of machined implant over the two period time. There was an increase in the bone strength (removal torque) of bone implant interface with time in the same group. **Conclusion**: Ablation of cpTi implants with Nd.YAG laser was efficient in enhancing removal torque value and hence enhancing osseointegration when compared with machined implants.*

Keywords: laser ablation, implant, Nd.YAG laser

1. Introduction

The surface modifications that can shorten the osseointegration of dental implants which is relatively long (3-6 months) would achieve a decreased healing time, a lower failure rate and minimal discomfort for the patients. Also improved or modified surfaces will open the probability of implantation for people in relatively poor health status. Implants made of Ti and titanium alloy need to be surface treated to become bio-active , and the oxide layer on the surface plays a key role in improveing the connectivity between the surface of a medical implant and the surrounding tissues (1) Overall published studies indicated that surface roughness plays an important role in providing effective surface for bone implant contact, cell proliferation, and removal torque, despite having Rapidly development in nanotechnology was increased in restoration dentistry by adding nanoparticles to an acrylic base to improve some physical and mechanical properties of PMMA[23], like add nanosilver particles to improve thermal conductivity and compressive strength of PMMA[24] and add 1wt% Tio_2 nanoparticle show increase tensile strength.[25] mechanical properties .Thus a combination of a good surface roughness and mechanical properties of titanium could lead to successful dental implants (2)Laser is used in a wide scope in medicine and dentistry dependent on its type. In implantology research of laser surface treatments on bio-medical implants in recent years has increased noticeably (1)Studies on the laser machining of dental implants revealed that an appropriate structure with minimum contamination could be achieved by means of laser treatment(4). Surface modification of Ti and Ti alloy using Nd:YAG laser in air may result in melting and rapid resolidification developing a new surface textures(5) The results showed that a laser process-ing can attain desirable phase formation and improved oxide layer formation in cpTi (6)The ablated implant surface control and organize osteoblast during healing process. Osteogenic cells migrate, attached and differentiate into circumferential to the ablated implant surface, osteoblasts produced bone microstructure with tubercular attachment parallel to microgross , these structure mineralized during healing process (7).The present study attempt to evaluate the effect of Nd YAG laser ablation of cp Ti screw on osseointegration when positioned in rabbit tibia; this evaluation was done through torque measurement as well as histological study.

2. Materials and Methods

Commercially pure titanium (grade II) is used as sub-strate of this study. Titanium was cut into small circ-ular disc (6mm in diameter and 2 mm in thickness) , polished by using (Grinder_polisher; metkon, forcipo IIv, Turkey) to have mirror like surface , This sample was used to detect if Nd.YAG laser with its own para-meter has an ablation effect on Ti. The distance between the Nd.YAG laser source and sample was fixed at 10cm which is the focal length of the built- in lense Unislide was used to move the sample which placed on it, after each laser pulse the specimen would move 0.5 mm to get a uniform standard overlapping of laser pulses effect along the specimen The ablated line was gotten after laser ablation. Different voltage and accordingly different laser energy was used to induce different range of surface roughness of specimens. to gain the appropriate implant surface roughness of (1.38 ± 0.23 μm) according to Faeda(8)

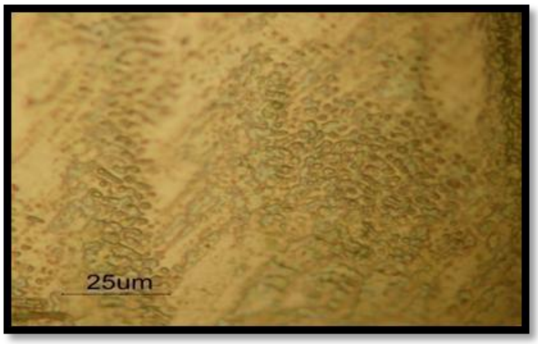

Figure 1: Ti ablated with 400 mJ Nd.YAG laser.under LM (1000x)

Results of pilot study indicated that the best energy that gave the most suitable roughness (1.38 µm) according to[8] is 400mJ. Also figure. (1) shows a uniform roughness surface in comparison with other, therefore it was decided to test the effect of laser ablation at 400mJ energy on the osseointegration of screw design cp ti implant fixture .Then machined Ti surface and that has been ablated with 400mJ(which give us the preferable roughness) and 580 mJ had been investigated by scanning electron microscope (SEM Tescan vegaIII,Czech),as shown in figure 2,3.

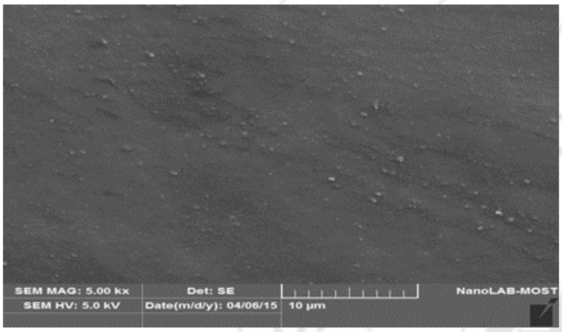

Figure 2: Machined Ti surface under SEM(5000x)

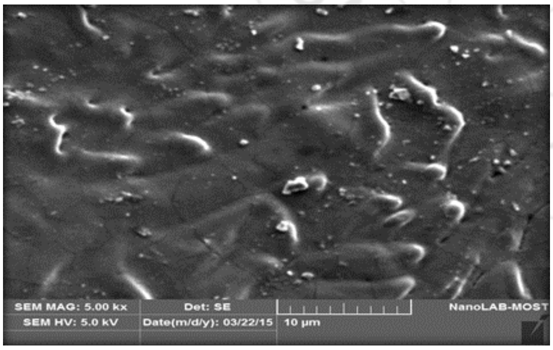

Figure 3: Ti ablated with 400 mJ Nd.YAG laser under SEM (5000x)

Implant Preparation: Implant machining: Thirty six screw shaped implants 3 mm in diameter and 8 mm in length(threaded part is 5 mm and smooth part is 3 mm), were machined from cpTi rods using Lathe machine. The head of the implant had a slit of 1mm depth to fit the screwdriver during insertion and removal by torque meter during mechanical testing. The screws were washed in ethanol in an ultrasonic cleaner for 15 minutes and dried at 100ºC for 15 minutes. Eighteen implants were left machined and served as controls The remaining 18 implants had their surface modified by a laser ablation process.

Laser Ablation A special tool was prpepared to standarized laser ablation procedure through out the samples area and all the samples used in the experimental group of the study. The tool allowed to rotate the fixture in circle around its long axis the base rotated manually by a nobe which move the implant in 22.5º . Finally the implant circumference was exposed to 16 laser pulse. The unislide stage was used to move the implant longtudinally every 0.5 mm .In this way all the implant surface (except smooth surface) was ablated .After laser ablation the experimental screws appeared brighter when compared with the control specimen Then every 2 implants were packed separately (one implant from the control group and the other from the experimental group) in airtight plastic pack.

Sterilization The implants were sterilized according to (AECL,1984) with gamma irradiation at 2.5-3 mega rad using gamma cell 220 with a CO60 source . 1.25 MeV was the energy of the used radiation with a dose rate of 90.4 rad/min and 80cm distance between source of radiation and dental implants.

Sample Grouping **:** The implants were categorized according to the test performed into two main categories:

Mechanical (torque measurements):(32 implants)The implants were subclassified into :

a) Control Group (16 implants) This group includes 8 implants for each healing interval (2 and 4 weeks).
b) Experimental Group (16 implants): This group includes 8 implants for each healing interval (2 and 4 weeks) (ablated with laser)

After incision and reflection of fascia and muscles to expose implants that has been fixed in rabbit's tibia, tibia was supported firmly to prevent any movement and the head of digital torque-meter (TQ8800) was engaged into slit of implant head to determine the peak torque to loosen implant from boney

Histological Test: (4 implants) in this test the implants were subclassified into:A-Control Group (2 implants): one implant for each healing interval (2 and 4 weeks) B-Experimental Group (2 implants):one implant for each healing interval (2 and 4 weeks

3. Results / Discussion

The removal torque values of implants after 2 and 4 weeks of implantation expressed in figure (4). The removal torque value needed to remove the experimental group more than that needed for removal of control group for the two interval ,and the torque for each group increase with time of healing period.

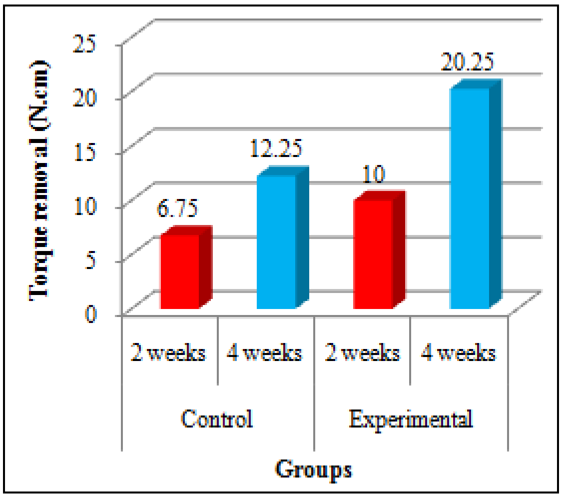

Figure 4: Bar chart shows mean of removal torque value in groups and duration

In table (1) the values of t-test showed that there is highly significant difference between the experimental and control group , and there is highly significant difference between the two intervals (2 weeks) and (4 weeks))

Table 1: Descriptive statistics of torque removal (N.cm) of dental implants in different groups and duration

Group		N	Mean	SD	SE
2 weeks	Control	8,00	6,75	1,66	0,58
	Study	8,00	10,00	2,08	0,74
4 weeks	Control	8,00	12,25	1,69	0,59
	Study	8,00	20,25	1,71	0,60

Analysis of the result using ANOVA test showed high significant (table-2) further comparism between every 2 group regarding time or test.

Table 2: Effect of experiment on the torque removal (N.cm) of dental implants in different groups

Group	*N*	*Mean*	*±SD*	*Independent T test*	*Df*	*Sig.*
Control	8,00	6,75	1,66	3.455	14	0.004
Study	8,00	10,00	2,08			
Control	8,00	12,25	1,69	9.402	14	0.000
Study	8,00	20,25	1,71			

The histological view of the rabbit's tibia in the microphotograph view at 2 weeks duration shows thread of implant impression within bone marrow tissue show osteoid tissue at the apex of the thread.

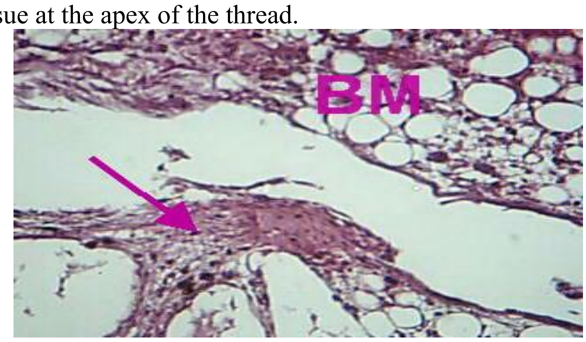

Figure 5: Microphotograph view for control group after 2 weeks duration shows thread (arrow) of implant impression within bone marrow tissue (BM).H&E×20

Microphotograph view for experimental group at 2 weeks duration shows thread of implant impression. Magnifying view for this impression shows bone trabeculae occupies apex of the thread that is surrounded by many active osteoblast .Bone trabeculae occupies base of implant bed close to basal bone,osteoblasts rimming bone and osteocytes can be detected.

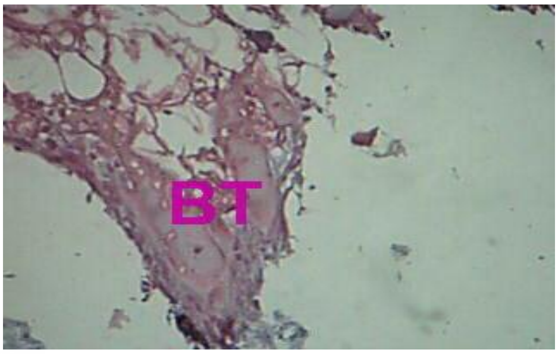

Figure 6: Microphotograph view forexperimental group after 2 weeks durationshows thread of implant impression bone trabeculae (BT) occupies apex of the thread. H&E×10

The histological view for control group at 4 weeks duration illustrate primitive bone formation , Microphotograph view shows thread with bone trabeculae and woven bone. Microphotograph view for experimental group at 4 weeks duration shows well developed boney thread, magnifying view for thread shows development of haversian canals with osteocytes, bone trabeculae coalesced with basal bone, osteoblasts rimming bone and osteocytes within bone trabeculae.

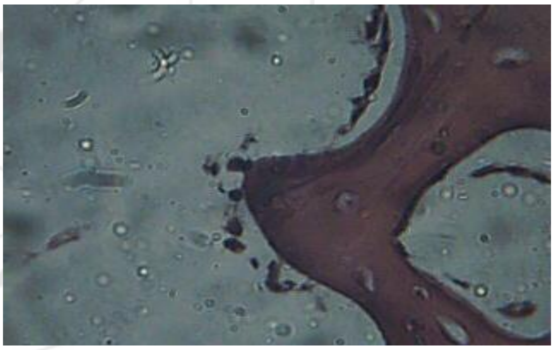

Figure 7: Microphotograph view for control group after 4 weeks duration shows thread with bone osteoblast rimming bone trabeculae .H&E×40

Figure 8: Microphotograph view for experimental group after 4 weeks duration shows well developed boney thread (arrow).H&E×10

Shalabi et al concluded that micro-texture of the implant surface influenced attachment and growth of cells originating from human mandibular bone in vitro. The increase in roughness increase directly the surface area of implant ,improve cell migration and attachment to implant and enhance osseointegration process. From 1953-2003 almost all papers revealed an enhanced bone –to-implant contact with increasing surface roughness.[(17)(18)] Cell attachment and proliferation were surface roughness sensitive and increased as the roughness of Ti and Ti alloy increaseed [(19)(20)(21)(22)].That explain the increase in torque removal and quality and quantity of bone formation in experimental than control.Zinger et al found that the cells responded to nano-scale roughness by a higher cell thickness and a delayed apparition of the focal contacts ,so Zinger support this study by depend-ing on micrometer roughness and exclude work on nanometer rough-ness[(23)].The optimal roughness (S_a of about 1.5 μm) of such surfaces that can elicit the maximum bone response has been investigated [(24)(25).]Yeo who support the results in this study since the surface roughness level obtained by laser was within the above recommended value[(26)] .A great number *of the* experimental investigations have demonstrated that the bone response was influenced by the implant surface topography; smooth (S(a)<0.5 microm) and minimally rough (S(a) 0.5-1 mum) surfaces showed less strong bone responses than rougher surfaces. Moderately rough (S(a) 1-2 microm) surfaces showed stronger bone responses than rough (S(a)>2 microm) in some studies[(27).]Faeda et al showed a significant statistical difference (p<0.01)between the roughness of laser modified implants of (1.38±0.23μm)and machined implants ,that agree with this study which also showed highly significant difference between laser treated implants of roughness average of 1.36μm and machined implants[(8).] This study does not agree with Aparicio et al who proved that roughness value of 4.5 micrometer is favoured for osseointegration of dental implants at short and mid-term healing periods ,it may due to the different way of surface modification which was grit blasted[(28).] Thisstudy proved that modification of titanium dental implant surface morphology could enhance tissue response and decrease the waiting time for loading of implant , and this agree with[(2)] This is agree with[(7)] who proved increasing of attachment and differentiation of osteogenic cells to laser ablated implant surface. [(19)] agree with this study in proving that proliferation of osteoblast increase by increasing surface roughness. The removal torque of the Laser treated titanium implant upon rough implant surface induced by anodizing surface embedded in rabbit femoral metaphysis for 8 weeks was stronger than that of the anodized titanium implant *.[29]* The Alkaline phosphotase activity per cell number was improved by about four times in the laser-treated surface compared with that in polished surface . On the laser-treatedcommercially pure titanium(cp Ti) surface, itwas considered that the bone formation activity of osteoblasts was promoted without inhibiting cellproliferation . From the results of this study, it is possible to conclude that by treating cp Ti surfaces with a laser, a surface with good cytocompatibility can be created[30]. Joob-Fancsalyet al agree with this study concluded that the high energy laser treated implants needed higher removal torque and in some cases an osteogenic activity was observed around them in the medullary space as well[(31)].It was thought the advantage of laser surface treatment lies in special micromorphology and the increased cleanliness of the surface. Nd:YAG micro-texturing produce micro-size hole with reduced chemical modifications, increased the osteoblast attachment on the substrate by 64% compared to the untreated surface[(32)]This explain enhancement in osseointegration and increase torque removal in experimental group upon control .This study is agree with Peto et al who proved that laser treated implants have removal torque 20% more compared to the machined and blasted implants[(7)] . But at this study improvement was about 47% more in laser treated implants compared to machined. Faeda et al measured torque removal of laser modified implants and machined implants as control after 4 weeks and it was 23.28 N cm for machined implants and 33.0 Ncm for laser modified implants ;that agree with this study in increasing removal torque of implants of the laser modified implants upon machined implants[(8)] . Microroughened implant surfaces showed, qualitatively, more platelets than machined surfaces .Park &Davis believed that these early blood cell/implant interactions may play a key role in the osteoconduction stage of peri-implant bone healing response to microroughened implants[(35).] And that may interpretate the improvement of quality and quan-tity of osseointegration in experimental group after 2 and 4 weeks.This could be explained by the fact that laser-engineered porous titanium surface seems to promote , *in vitro*, the adsorption of albumin and fibronectin more than sandblasted or machined implants[(36)] The improved osseointegration in laser treated implants resulted in this study could be due to rougher and more opened titanium oxides enriched surfaces ,the laser treated samples were found to have surface more favourable Ca and P inducibility to form bone like apatite structures in the m-SBF(modified artificial simulated body fluid) solution according to Hong et al [(37)]

4. Conclusion

1-Laser ablation can be used to produce uniform repeatable and clean surface ablation of cp Ti material that provides suitable roughness range. 2-The ablated implant screw showed higher torque measurement after 2 and 4 weeks of implantation when compared with non-ablated one.3-Histological evaluation of implanted ablated screw in bone indicated that laser treatment provided favorable surface that enhance and promote osseointegration better than non-treated one

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