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# Evaluation the Effect of 805 nm Wavelength Diode Laser on Repair of Mandibular Bone Repair and Skin Incisions in Rabbits

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**Abstract**: The long healing time of bone after tooth extraction in order to construct artificial teeth is uncomfortable to the patient because of aesthetic or masticatory problems in addition to the daily visit to dental clinic. The objective of this study was to evaluate the effect of 805 nm diode laser with long time intervals on repair of bone and skin incisions in rabbits through biochemical, radiological and histological findings. Eighteen New-Zealand rabbits were undergone surgical operations to make a cavity in the bone of the lower jaw, the rabbits were divided into two groups:- Group A (control group) containing nine rabbits. Group B (lased group) containing nine rabbits in which two cavities were done, one on the right side and the other on the left side of the mandible. The cavities were subdivided into two groups according to the exposure time. Group $B_1$ (the right side) which was underwent treatment with 805 nm continuous diode laser with output power of 900mW and exposure time of 5 min, every 72 h for two weeks. Group $B_2$ (the left side) which was underwent treatment with 805 nm continuous diode laser with output power of 900mW and exposure time of 10 min, every 72 h for two weeks. The diode laser with a wavelength of 805 nm, power 900 mW and operating on continuous mode was applied directly over the site of the cavity according to the group. Radiological findings, histological and biochemical evaluations for both bone and skin were done for all groups after 7, 14 and 28 days of follow up. The histological results showed that there was a complete wound healing and bone repair at day 28 postoperatively in sub group $B_2$ which is represented by the group treated with 805 nm diode laser with exposure time of 10 min, every 72 hours. In conclusion, the 805 nm continuous wave diode laser with power density of 1.79 W/cm$^2$ and exposure time of 10 min, every 72 h for two weeks was beneficial to stimulate healing in bone and skin.

## Introduction

One of the oldest therapeutic methods is enhancing wound healing and bone remodeling was by light therapy, later as color light therapy and UV therapy further more the use of laser as light source was the last step in the development of light therapy (Tiina I and Karu 2003). The first publications about low-power lasers therapy (then called laser biostimulation) appeared before 1970s when Ender Mester tried to study the carcinogenic effect of laser on mice. This was the first demonstration of biostimulation and the first studies focused on visible light helium-neon (He-Ne), argonium, or kryptonium lasers, then semiconductors such as gallium-arsenide (Ga-As) and gallium-aluminum-arsenide (Ga-Al-As) diode lasers have been available and intensively used (Diego S. B., et al., (2008). After that, several in vitro and in vivo studies were done to evaluate the effect of laser on cells (Diego S. B., et al., (2008). Acceleration of bone healing provides patients with a shorter post operative period (2), and in dentistry it provides shorter time for denture or artificial teeth construction.

## Materials and Method

Eighteen New-Zealand rabbits, fourteen females and four males were used in this study, their mean weight was 1.5-2 kg and their mean age was 9-12 months, the females were separated from males. All animals were weighted to calculate the dose of anesthesia and antibiotic which would be given to them.

Rabbits were divided into two groups, one side of the lower jaw of the first group was prepared to be the control group (group A) while for the other group (B) both sides were prepared; the right side for receiving treatment with diode laser emitting at 805nm wavelength, 900mW power and exposure time 5 min. (group B1), while the left side was prepared for receiving treatment with the same laser but exposure time was 10 min. (group B2). The lower jaws were prepared by clipping, shaving and washing.

General anesthesia (Combelen 0.5 mg/kg) was injected intramuscularly. After 10 min. a mixture of Ketamin (50 mg/kg) and Xylazine (20 mg/kg) were injected intramuscularly. The prepared skin was cleaned with 10% povidon iodine, and then it was drapped with sterile towels which were properly fixed by towel clips.

The skin and the Masseter muscle were incised by the blade to expose the body of the mandible, and then the periostium was reflected. Drilling was done by prosthetic hand-piece with stainless steel round bur (2mm in diameter) and rotation speed about 5 RPM accompanied with irrigation by distilled water to clean the drilling site from bone debris and to prevent over heating of the surrounding tissue.

The cavity was 2 mm in diameter and a stopper was placed at the end of the ball of the bur to ensure that the depth of 2 mm of the cavity had been reached. The cavity was done about 1cm anterior to the projection in the lower border of the mandible to ensure the accurate site of laser radiation.

After cavity preparation, it was thoroughly washed using distilled water to remove bone debris. The Masseter muscle was sutured by 3/0 catgut suture with continuous matris suture, the skin was sutured by 3/0 black silk suture with vertical matris suture. Laser irradiation was done immediately after operation according to the group. After the first laser radiation, a post operative x-ray was taken.

The rabbits were given antibiotic which was a combination of streptomycin and penicillin intramuscular injection (0.25 ml/day) for 7 days. The rabbits were divided into two groups (A and B). In group B two cavities were done one on the right side and the other on the left side. The cavities were subdivided into two groups (B1 and B2).

The power density was calculated by calculating the area

area = $r^2 . \pi$

where r is the radius.

spot diameter was 8 mm → r = 4 mm

so the area is equal to 50.24 $mm^2$ = 0.5024 $cm^2$

power density = power / area = 1.79 W / $cm^2$

Laser irradiation was done by applying the laser probe with the beam perpendicular to the skin and with the long axis of the cavity prepared on the mandible with the first exposure immediately after operation. The irradiation was sited by putting laser probe about 1cm anterior to the projection on the lower border of the mandible. The projection was easily palpated by finger, in direct contact to the skin. This site is also with in an imaginary line passing across the anterior angle of the eye and perpendicular to the lower border of the mandible.

The animals were examined after 7, 14, 28 days post-operatively after sacrificed. A radiograph was taken to evaluate cavity prognosis, then a blood sample was taken from their hearts for alkaline phosphatase examination and finally the animal was sacrificed by over dose (0.5 ml) thiopental sodium.

## Results

The results of the two groups include the radiological, biochemical and histological findings for both skin and bone. Radiographical pictures at day 14 post-operatively in the control group and the laser treated group with exposure time 5 min. showed radiolucent spot at the drilling site while in lased group with exposure time 10 min. showed normal anatomy at the drilling site. At day 28 postoperatively the radiographical pictures of laser treated groups showed normal anatomy at drilling site in comparison with the presence of well-defined radiolucent spot at the drilling site in control group Figures 1-4.

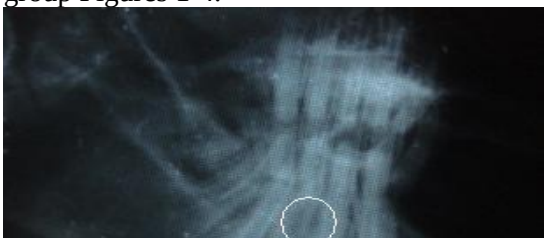

**Fig. (1):** Radiographical picture at day 14 for control group shows well-defined radiolucent spot at drilling site

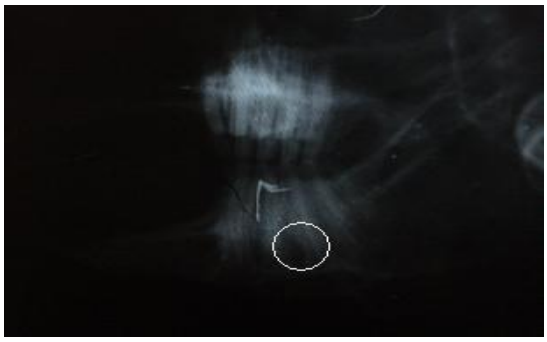

**Fig. (2):** Radiographical picture at day 14 for irradiated group with exposure time 10 min. shows normal anatomy at drilling site

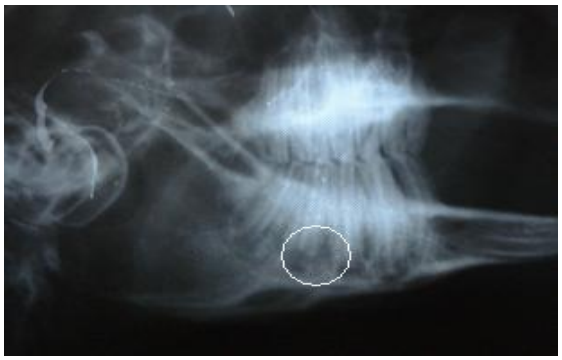

**Fig. (3):** Radiographical picture at day 28 for control group shows well defined radiolucent spot at drilling site

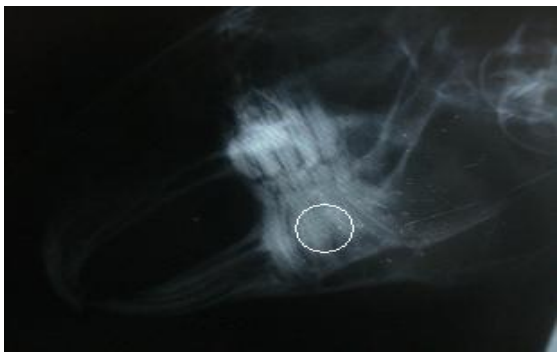

**Fig. (4):** Radiographical picture at day 28 for irradiated group with exposure time 10 min. shows normal anatomy at drilling site.

The histological findings at day 14, skin development was more advanced in laser treated group than control group with normal skin tissue in subgroup B2 at day 28. The histological feature of skin in control group at day 28 was similar to that in subgroup B2 Figures 5-9.

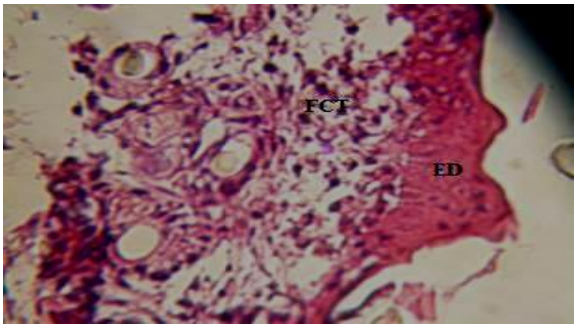


**Fig. (5):** Transmission light microscope for a histological section in skin at 14 days post-operatively control shows cellular fibrous connective tissue FCT infiltrated with mononuclear cells in incision line covered by irregular thickness of epidermal layer ED with out rete-ridges (H&E x40).

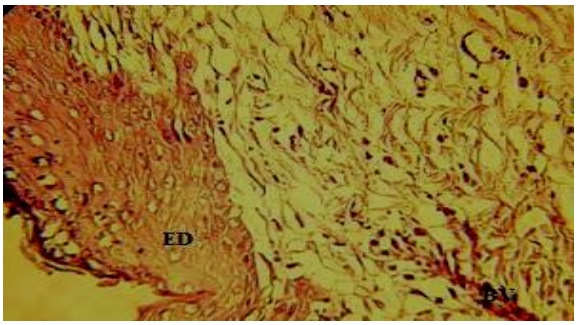


**Fig. (6):** Transmission light microscope for a histological section in skin at 14 days post-operatively treated with diode laser 5 min. reveals mononuclear cells infiltration mainly macrophages and lymphocytes in dermis particularly around blood vessels BV with thick layer of epidermis ED (H&E x40)

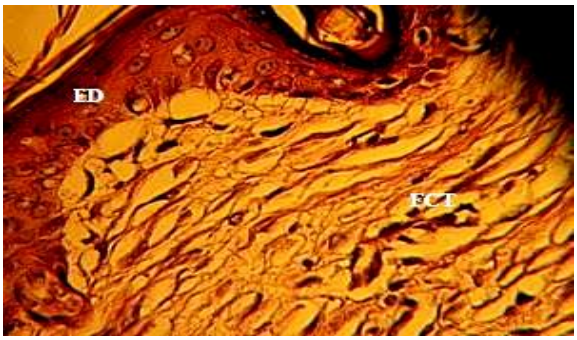


**Fig. (7):** Transmission light microscope for a histological section in skin at 14 days post-operatively treated with diode laser 10 min. reveal mature fibrous connective tissue FCT infiltrated with few mononuclear cells with moderate thickness of epidermal ED layer which shows rete-ridges (H&E x40)

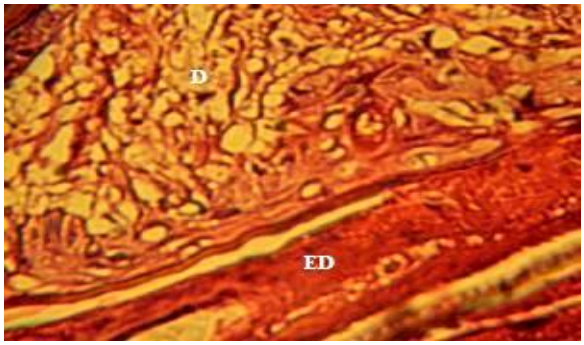


**Fig. (8):** Transmission light microscope for a histological section in skin at 28 days post-operatively control shows moderate mature fibrous connective tissue in the dermis D covered by thick epidermal layer ED under the cellular debris (H&E x40)

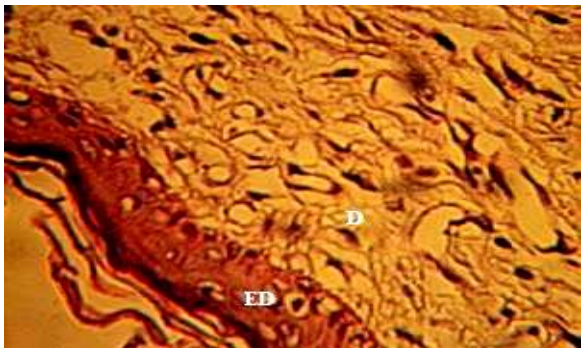


**Fig. (9):** Transmission light microscope for a histological section in skin at 28 days post-operatively treated with diode laser 10 min. shows mature loose connective tissue in dermis layer D with few mononuclear cells infiltration and thick epidermal layer ED (H&E x40)

For bone histological results also confirm that the laser in groups ($B_1$&$B_2$) accelerate bone repair. Histological findings represented by the presence of bone trabiculae in group $B_1$ (treated by 5 min. diode laser) at day seven which are thicker in group $B_2$ (treated by 10 min. diode laser) and thinner in group A (control group) Figures 10-13.

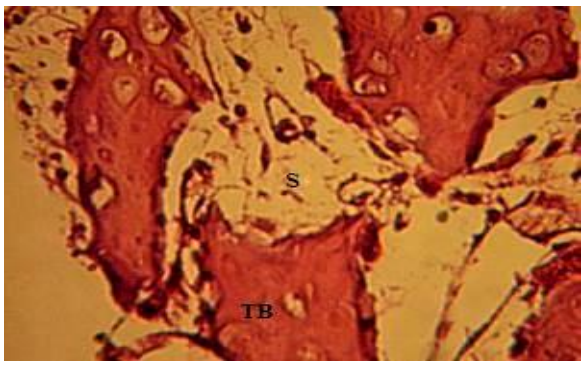


**Fig. (10):** Transmission light microscope for a histological section in bone at 14 days post-operatively control revealed thick trabicular bone TB lined with osteoblast cells surround large space S (H&E x40)

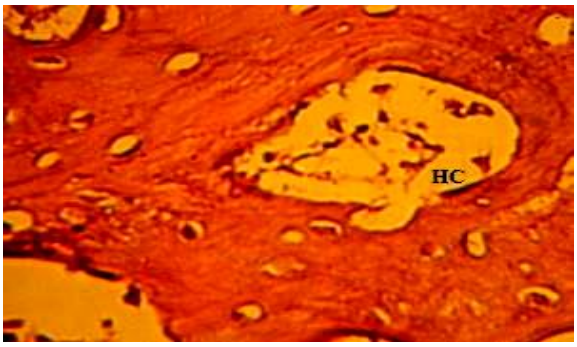


**Fig. (11):** Transmission light microscope for a histological section in bone at 14 days post-operatively treated with diode laser 10 min. shows compact bone with Haversian canal HC (H&E x40)

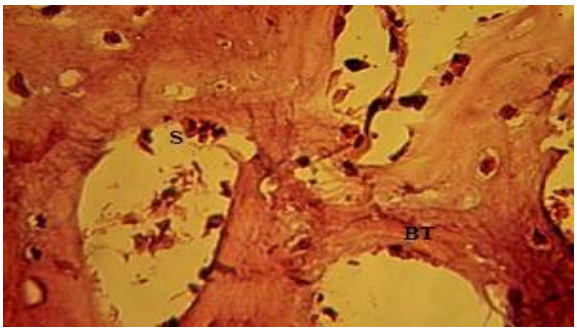


**Fig. (12):** Transmission light microscope for a histological section in bone at 28 days post-operatively control shows more thick bone trabiculae BT surrounds large space S lined by osteoblast cells (H&E x40)

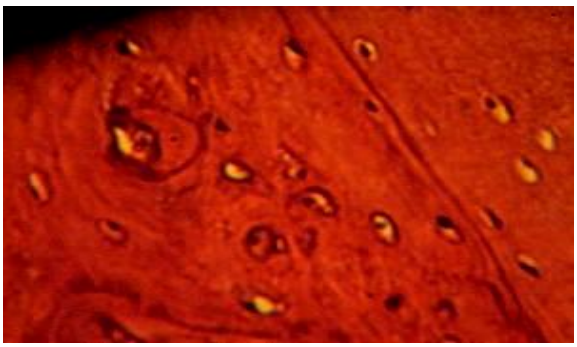

**Fig. (13):** Transmission light microscope for a histological section in bone at 28 days post-operatively treated with diode laser 10 min. shows compact bone with Haversian canal (H&E x40)

The laboratory values of alkaline phosphatase are illustrated in Table 1.

**Table (1):** illustrates the laboratory values of alkaline phosphatase measured in KA units/100 ml in control group and lased group

| Days | Control group | Lased group |
|---|---|---|
| 0 | 45 | 45 |
| 7 | 63.9 | 85.2 |
| 14 | 86.5 | 120.7 |
| 28 | 261.8 | 199.6 |

## Discussion

The main objective of this study was to evaluate the effect of low level laser irradiation on the healing process of the surgical incision of skin and bone repair for long time intervals. Skin clinical and histological results confirm that the laser in groups (B1&B2) accelerate wound healing. The epithelial cells regeneration of epidermal layer is faster in lased group B2 than in lased group B1 which is faster in comparison with control group A. this result is in agreement with (Hawkins and Abraham 2007), the reason can be attributed to that laser stimulates cell proliferation by increasing oxygen uptake by the cell and ATP synthesis in mitochondria (Hermes P., et al., 2007, Rocha J. A. et al., 2006, Pinheiro A. L. B., 2003, Richard M., 2003, Grace W., 2004, and Denise H. and Heidi A., 2007). Bone radiological and histological results confirm that laser in subgroup B1 and B2 accelerate bone repair, this agrees with Hermes P., et al., (2007). The reason can be attributed to that different lasers have effects on ATP synthesis in bone cells (Hermes P., et al., 2007, Pinheiro A. L. B., 2003 and Mohammed A. A., 2004). The value of alkaline phosphatase increases with increasing time of exposure, which may indicate that increasing exposure time increases the activity of osteoblast (Paskalev M. et al., 2005 and Nazaroglou, et al., 2009). Reduction of alkaline phosphatase level at day 28 in lased group may be due to the formation of bone reaching the final stage; this result was confirmed by the histological findings which revealed the formation of compact bone so there is regression in cellular activity.

## Conclusion

From this research work it was concluded that diode laser at wavelength of 805nm and power density of (1.79 $W/cm^2$) can produce good healing in both bone and skin of rabbits. Half the healing time can be obtained with exposure time equal to 10min. every 72h.

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# تقييم تأثير ليزر الدايود 805 نانومتر على إصلاح عظام الفك السفلي والشقوق الجلدية في الأرانب

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**الخلاصة**

الوقت الطويل من أنتظار العظم للشفاء بعد قلع السن لغرض تعريضه بأسنان أصطناعية تسبب أزعاجا" الى المريض بسبب المشاكل الجمالية أو المضغية بالأضافة الى الزيارة اليومية الى عيادة الأسنان, لذا هدف هذه الدراسة كانت أن تقيم تأثير ليزر الصمام الثنائي 805 نانومتر بفترات طويلة من الزمن على ألتئام العظم والجروح الجلدية في الأرانب من خلال فحوصات سريرية وكيمياء حيوية ونسيجية. ثمانية عشر أرنب نيوزلندي خضعت لعمليات جراحية تحت المخدر العام لعمل تجويف في عظم الفك الأسفل, ألأرانب قسمت الى مجموعتين: مجموعة أ (مجموعة قياسية) تسعة أرانب. مجموعة ب (مجموعة الليزر) تسعة أرانب, في هذه المجموعة تم عمل تجويف واحد في الجهة اليمنى والاخر في الجهة اليسرى, قسمت الحفر الى مجموعتين ثانوية حسب وقت التعرض, مجموعة ب1 (الجانب الأيمن) مرت بالمعالجة مع ليزر الصمام الثنائي 805 نانومتر بوقت تعرض 5 دقائق, مجموعة ب2 (الجانب الأيسر) مرت بالمعالجة مع ليزر الصمام الثنائي 805 نانومتر بوقت تعرض 10 دقائق. بعد أغلاق الجرح تم تسليط ليزر الصمام الثنائي بطول موجة 805 نانومتر وقدرة 900 ملي واط وشعاع مستمر على موقع التجويف حسب المجموعة كل 72 ساعة ولمدة أسبوعين. ملاحظات سريرية ونسيجية توبعت للجلد بالأضافة الى فحوصات أشعاعية ونسيجية وتحليلات كيمياوية عضوية للعظم لكل المجاميع بعد 7, 14, 28 يوم من المتابعة. كشفت النتائج بأن ليزر الصمام الثنائي 805 نانومتر ذو الشعاع المستمر بالكثافة 1,7 واط / سم2 ووقت تعريض 10 دقائق كل 72 ساعة ولمدة أسبوعين (المجموعةب2) كان مفيدا لتحفيز ألتام الجلد والعظم.