EFFECTS OF LOW LEVEL LASER (DIODE- 830NM) THERAPY ON- HUMAN BONE REGENERATION:

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Abstract

Effects of low level laser(Diode-830 nm) therapy on human bone regeneration:

By- DR. MD NAZRUL ISLAM

Objective:

Laser (Semiconductor diode, Ga-Al-As, 830nm) is effective in human bone regeneration, i.e. it enhances bone fracture healing.

Background Data:

Tissue healing is a complex process that involves both local and systemic responses, and the healing process of bone is much slower than that of soft tissues which is a great challenge of medical science. The use of Laser Therapy (LLLT) for wound /bone healing has been shown to be effective in modulating both local and systemic response by enhancing- cellular & mitochondrial ion exchange, bone mineralization, nitric oxide formation, lymphatic circulation, osteoblast proliferation, effects on osteoblast gene expression, osteoclast inhibition (prevents bone mineral resorption) and by bone engraftment on synthetic materials.

Methods:

40 (Twenty in laser & Twenty in control group) otherwise healthy men and women with, closed appendicular bone fracture (Radius/ ulna, or Femur / Tibial shaft /Clavicle / Meta carpal /Meta-tarsal) was enrolled for fracture management by laser therapy adjunctive to regular management, and was assed by clinical and radiological findings (X-ray)/at 2nd , 3rd, 4th and 6th week post fracture: assessment included fracture line/margins, fracture gap, external callus appearance, callus-to-cortex ratio, bridging, and radiologic union as well as clinical assessment of the fracture- compliance of patient, and onwards follow-up of patients, in comparison to controlled group.

Results:

Early significant bone regeneration /callus formation achieved by early application of Low Level laser therapy (Ga-Al-As, 830 nm) on human fractured long (appendicular) bone.

Conclusions:

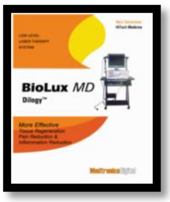
Treatment with 830 nm diode laser has substantially reduced the fracture healing time as well as improved the quality/quantity of callus formation of the patient, thus enhancing fracture healing. Laser biostimulative effects on bone could be a new dimension for bone regeneration which significantly reduce healing period, lessen cost of treatment, and enhance patient compliance in medical science.

▪ Materials & Methods

Duration of study: The duration of the study: 2years (from April- 2008 to March- 2010)
Type of study: Prospective randomized case control study.
Place of study: Shaheed Suhrawardy Medical College Hospital in the Department of Orthopedics and Traumatology , Dhaka-1207, Bangladesh.

Materials

Machine used in this work: The laser machine used on this research was BioLux MD (Ga-Al-As-830 nm) Diode Laser.



830 bone probe

Tip of 830 probe

BioLux MD Ga-Al-As Laser (830 nm) Machine with Probe.

Machine Technical Specification:

▪ Laser/Photon source	Semiconductor device (Ga-Al-As).
▪ Laser Type-	Class iii B
▪ Emission mode	CW/Pulsed
▪ Exposure control	Integrated digital
▪ Treatment Planning System	Software based
▪ Cooling System	Aerial
▪ AC source rating	220V & 50 Hz

Probe specification:

Part number	Integration	Probe Type	Wavelength	Emission	Application
1. LP 81200c	Standard	200mW Laser probe	810 nm	Coherent	Pain Care/Tissue Regeneration
2. LP 83500c	Optional	500mW Laser probe	830 nm	Coherent	Bone Regeneration

▪ Sample collection & distribution

The sample was collected randomly from admitted patients with bone fracture in superior and inferior extremity in Shaheed Suhrawardy Medical College Hospital in the Department of Orthopedics and Traumatology, Dhaka-1207, Bangladesh. A total of 40 patients randomly collected; among which 20 were in the Laser group (L1,L2) and 20 were in the control group (C1,C2). The patients were briefed about the study and written consent (Informed consent) was obtained from all patients/ medico legal guardian for other patients.

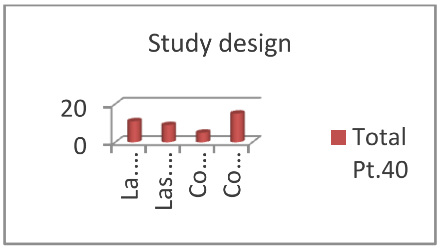
Inclusion Criteria

1. Patient suffering from recent (1 -7 days old) axial bone fracture of male and female patients, 2. Age between 15- 95 years, 3. Are not taking any pain medications, 4. Whether taking any bone supplement or not, 5. Aren't pregnant, haven't any previous fracture history, 6. Have not systemic or psychological disorder.

Exclusion Criteria

1. Bone fracture with open wound were excluded, 2. Bone fracture with any active infection, 3. Bone fracture with previous Surgery.

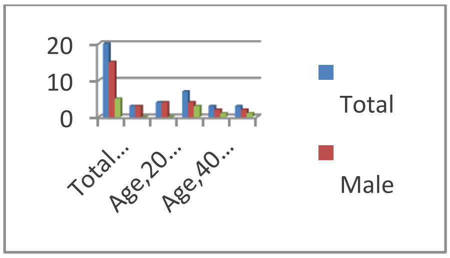
- **Patient distribution: General**



Patient distribution according to study design.

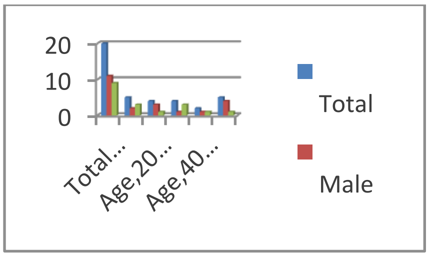
- **Patient distribution: By age-**

Laser Group (L1&L2)



Bar-graph showing-patient distribution according to age in study (laser) group.

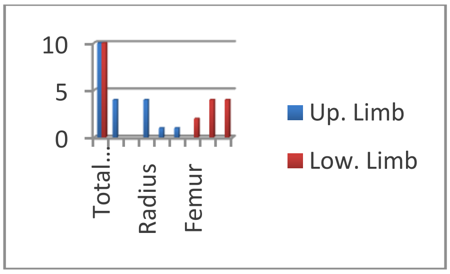
Control Group (C1&C2)



Bar-graph showing-patient distribution according to age in non-study (Control) group.

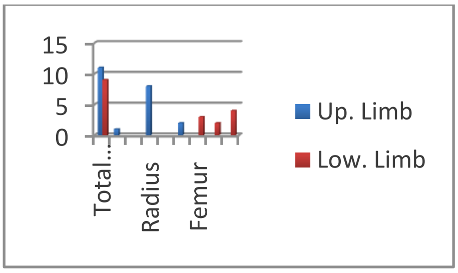
- **Patient distribution: By bone**

Laser Group (L1&L2)



Bar-graph showing patient distribution according to type of bone in study (laser) group.

Control Group(C1&C2)



Bar-graph showing patient distribution according to type of bone in control group.

▪ Treatment Protocols:

❖ Laser Treatment Protocols used in this work:

- 4- 8 joules /cm2
- 4 points/ session
- Power-500mw
- Point spacing is every 2-4 sq. cm.
- Treatment schedule: daily for the first week, followed by alternate day in the second week (9 days total).

❖ Treatment Chart maintained in this work:

Laser Therapy schedule-				
Group Adult	**Days**	**Dose Joule**	**Duration Minutes**	**Observation**
Ø Laser: L1,L2 (Adult)	1	32	5.33	
	2	32	5.33	
	3	32	5.33	
	4	32	5.33	
	5	32	5.33	
	6	32	5.33	
	7	24	4	
	8	24	4	
	9	24	4	

Laser Treatment Protocol in adult group (used in this work).

Laser Therapy schedule-				
Group Child	**Da ys**	**Dose Joule**	**Duration Minutes**	**Observation**
Ø Laser: L1,L2 (Child)	1	16	2.66	
	2	16	2.66	
	3	16	2.66	
	4	16	2.66	
	5	16	2.66	
	6	16	2.66	
	7	12	2	
	8	12	2	
	9	12	2	

Laser Treatment Protocol in child group (used in this work.)

Methods:

This randomized clinical trial (RCT) has been done at Shaheed Suhrawardy Medical College Hospital, Sher-E-Bangla Nagor, Dhaka-1207, Bangladesh. Fourty (40) patients with appendicular bone fracture in the range of 15- 95 years of age were randomly divided to the laser treatment group (Group L1 & L2) and non-laser/control group (Group C1 & C2). The laser group went under treatment for 6 times in a week for the first week and three times/ week (alternate day) for the next week of total 9 sessions

A patient on LLLT (Diode-830nm) therapy.

Applied laser in laser treatment group was continuous infrared laser with BioLux MD with 830 nm wavelength and 8 J/cm2 dose (energy) of total dose 8*4*9 J/cm2 for adult & 4 J/cm2 dose-(energy) of total dose 4*4*9 J/cm2 for child, and was irradiated on the fracture- side in pointing-method, in 4 anatomical locations at 500 mW; 0.5 centimeter away from radiological fracture line, two points in each site of line/ day. The irradiation was performed transcutaneously and the first session was performed on the 5th day after surgery/ incidence; based on previous research work which proved that laser works best on the proliferative stage of tissue healing.

At first, demographic data such as age and sex and subsequently pain and functional specifications were assessed and documented. Pain functional assessments were based on- Visual- Analogue Scale, and bone union was assessed on clinical point of view and by radiological assessment of callus formation.

The data's were routinely processed, by measuring the callus/ new bone formation. The best sets of weekly x-ray images of each patient from each group were selected for this analysis, and data's are also shown in datasheets and bar graphs. Efficacies of treatment were evaluated with pain questionnaire, clinical assessment and serial weekly radiograph starting from 1st up to 4th week and on the 6th week.

The patients were analyzed by-

- **Clinical assessment**

  Assessments of Clinical parameters were:

  a. Pain & inflammation level.
  b. Stability of fracture side.
  c. Movement of fracture side.
  d. Immobilization duration.
  e. Patient compliance.

- **Radiological assessment**

  a. Radiographic Scoring System (by Lane and Sandhu) of fracture site, done weekly.
  b. Densitometer assessment of Callus in the radiograph of fracture site, taken weekly.

- Radiographic Scoring System (by Lane and Sandhu) of fracture site:

Bone formation (Periosreal)	Score
No evidence of bone formation	0
Bone formation occupying 25% of defect	1
Bone formation occupying 50% of defect	2
Bone formation occupying 75% of defect	3
Full gap bone formation	4
Bone Union	
Full fracture line	0
Partial fracture line	2
Absent fracture line	4
Bone Remodeling	
No evidence of remodeling	0
Remodeling of intramedullary canal	2
Remodeling of cortex	4

Radiographic Scoring System by Lane and Sandhu: Based o-n
- Bone formation
- Bone Union and
- Bone Remodeling.

Table: 5.2.10-Radiographic Scoring System (by Lane and Sandhu) of fracture site, taken weekly.

- **Densitometer assessment of Callus in the radiograph of fracture site, taken weekly**.

Figure: 5.2.11-Optical densitometer used in this work.

L2-2. Patient Name: Enamul Haque Shah,Age: 55 Years.Sex: Male,Diagnosis: Fracture Rt. Colles. Study group: L2

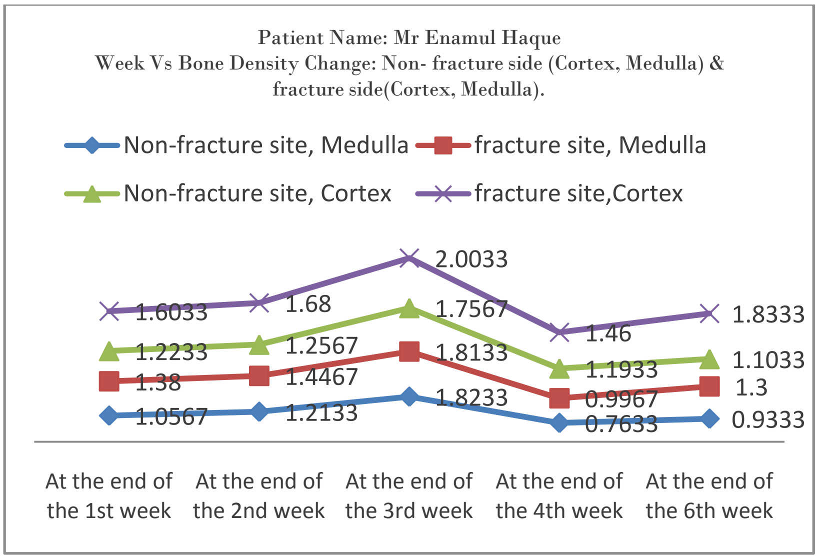
Week.	Non-Fracture Side, Cortex		Fracture Side, Cortex		Non-Fracture Side, Medulla		Fracture Side, Medulla	
	Reading	Mean	Reading	Mean	Reading	Mean	Reading	Mean
At the end of 1st week.	1.21		1.96		1.09		1.34	
	1.20	1.2233	1.90	1.6033	1.02	1.0567	1.26	1.3800
	1.26		0.95		1.06		1.54	
At the end of 2nd week.	1.28		1.88		1.22		1.37	
	1.22	1.2567	1.74	1.6800	1.25	1.2133	1.52	1.4467
	1.27		1.42		1.17		1.45	
At the end of 3rd week.	1.56		2.07		1.98	1.8233	1.80	
	1.80	1.7567	2.01	2.0033	1.83		1.83	1.8133
	1.91		1.93		1.66		1.81	
At the end of 4th week.	1.22		1.49		0.74		0.93	
	0.98	1.1933	1.55	1.46	0.78	0.7633	0.94	0.9967
	1.38		1.34		.77		1.12	
At the end of 6th week.	1.04		1.75		0.95		1.12	
	1.13	1.1033	1.83	1.8333	0.87	0.9333	1.31	1.30
	1.14		1.92		0.98		1.30	

▪ Observation & Result

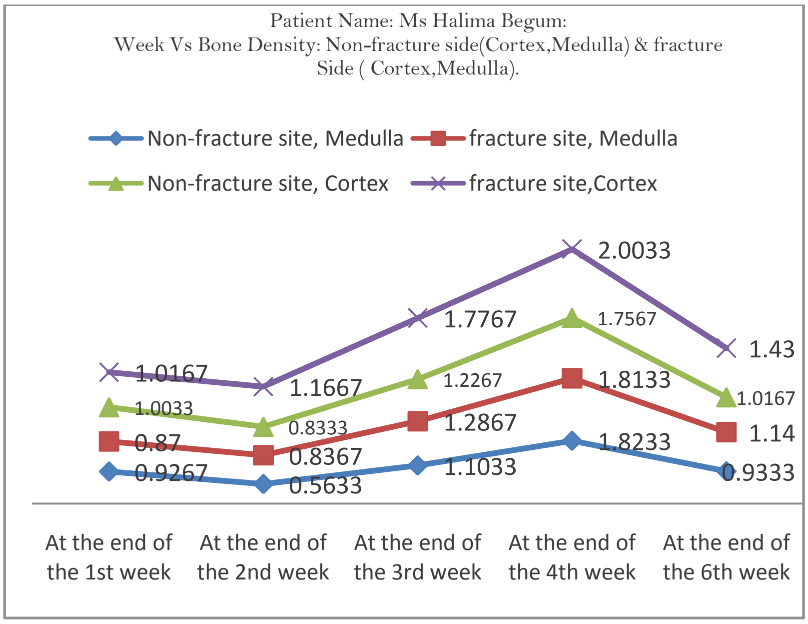
The aim of this work was to asses comprehensive (Both subjective & Objective) study/ evaluation of LLLT effect (Ga-Al-As 830nm@ Pw 500) on human bone.

Objective parameters were: Clinical (Qualitative) assessment included fracture line/margins, fracture gap, external callus appearance, callus-to-cortex ratio, bridging, and radiologic union/nonunion. Stability/ movement of the fracture site, functional ability, Pain scale, subsiding of inflammation, compliance of patient and onwards follow-up of patient, in comparison to controlled group. And Subjective parameters were: Quantitative assessment- done by weekly serial radiograph, and analyzed by Radiographic Scoring System by Lane and Sandhu, callus density measurement by optical densitometer, taken in-between treatment period & continued up to 6th week.

Clinically the laser group showed better stable fracture site, earlier movement of limb and removal of cast/plaster was performed. Pain & inflammation also subsided much earlier in the laser group (L1 &L2) than the control group (C1&C2).Radio-logically, this study compared degree of callus formation, callus density changes by weeks, and assessment of union, pain & inflammation parameters changes, with and without laser radiation (LLLT) in the post laser therapy period, starting from 2nd week up to 6th.



L 2-1. Patient Name: Mr Enamul Haque Shah ;Age: 55 Years.Sex: Male,Diagnosis: Fracture Rt. Colles.(Study group: L2) Dose Vs density bar-graph showing change of bone density in the fracture & non-fracture (two centimeter away from fracture line) cortex & medulla weekly in group L2 (Study group, operated).



C1-1.Patient Name: Halima Begum, Age: 55Years, Sex: Female,Diagnosis: Fracture Rt. Shaft of Femur(Study group:C1). Dose Vs density bar-graph showing change of bone density in the fracture & non-fracture (two centimeter away from fracture line) cortex & medulla weekly in group C1(control group, operated).

L1-2. Patient Name: Md Dalil Uddin,Age: 95 Years,Sex: Male, Diagnosis: Fracture Lt. Humerus (Shaft)-(Study group:L1)


Figure: 6.5 a-Before & starting of treatment.

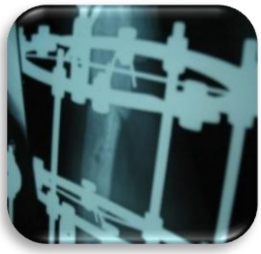
__Serial weekly x-rays of post-laser therapy of a L1 group patient –__

At 1st week

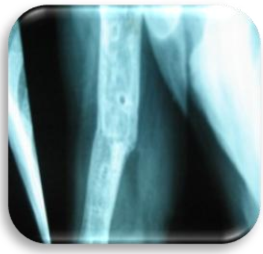
At 2nd week

At 3rd week

At 4th week



At 6th week



At 12th week

Stages of union/ callus formation by week (from 2nd to 12th week) of a study group (L1) patient.

In this study, at the end of 3rd week following laser treatment, the presence of callus/ new bone formation in fracture defects was more advanced in laser group (L-2-1) than in the control group (C1 & C2). In the control group (C1 & C2) similar callus/new bone formation was observed at the end of 4th week (C-1-1). There was a significant difference in the degree of callus formation and in the degree of bone union between study groups to which values were assigned to the different degrees of new bone formation. So, it can be inferred from this study that LLLT enhances bone regeneration/ callus formation in the early stages (Proliferative and reparative stages) of bone union which is compatible to earlier studies. Again, L2 group (Laser non-operated) showed earlier clinical & radiological union/ callus formation than L1 (Laser operated), which also infers that bone regeneration is faster in non-operated patient by LLLT. Within our study period, after 4th week, from 5th up-to 6th week in both study group (Laser & control) showed almost same degree of clinical & radiological bone healing parameters.

More-over, there was no side effect/ negative effect of laser on the targeted bone site or on surrounding soft tissue. The Mann whitny and Kruskal-Wallis test were used for statistical analysis. In this analysis the control group had lower values of callus density / new bone formation in 2nd-3rd week, than the infrared laser groups ($p \leq 0.01$).

▪ Discussion

The result of this study reveals a better bone healing after irradiation with 830nm diode laser (Ga-Al-As).This study result also concludes that better bone healing after irradiation with Ga-Al-As, 830nm diode laser in human model as an adjunctive to regular fracture management that accelerates bone union significantly and enhances patient compliances. The results observed are similar to previous reports, which demonstrated increased vascularization activates cytokines, growth factors, necessary hormonal activities for tissue healing enhancement in the proliferative stage, thereby reduction of pain & inflammation, and increased fibroblast, chondrocyte and osteoblast proliferation that activities bone regeneration

Some previous reports do recognize that LLLT has positive effects on bone [1, 2]. These studies reflect the idea that non-differentiated mesenchymal cells could be biomodulated positively to osteoblasts that would more rapidly change to osteocytes [1]. This aspect may be possibly corroborated by several previous studies in which LLLT was used in fractures [3], bone defects [4], tooth extraction [5, 6, 7] and after the placement of dental implants [8]. On the other hand, LLLT seems ineffective when used on normal tissues [9]. In order to observe the biomodulating effects of LLLT, some level of tissue deficiency seems necessary 10].

It is known that the osteogenic potential of mesenchymal cells depends on several genetic factors and also on systemic and local inducer factors [11]. LLLT may act as an inducer factor. However, some reports [12], suggested that LLLT would improve bone matrix production due to improved vascularization, anti-inflammatory effect and enhanced Collagen synthesis.

These aspects would increase both the release of mediators and micro-vascularization, which in turn would accelerate bone healing. It was suggested that PGE2 activates wound healing [13], and increased level of PGE2 was observed by Messer et al. [14]. There is evidence that PGE2 is also produced by osteoblast and that its effects may be therapeutic or adverse [15].

Collagen is an important component of the extracellular matrix of bone. It is significantly increases by LLLT.The mechanism by which LLLT interferes in collagen synthesis is not fully understood; however, it may be because of alterations in the genetic regulation or in the modulation of enzymatic activity involved in the metabolism of the collagen as suggested previously (16).

Studies of bone healing response to infrared light show acceleration of osteoblast formation as well as calcium salt deposition under the influence of infrared light. (17, 18) Studies have- demonstrated that bone growth factors are stimulated by IR light. Osteoglycin is a small leucine rich proteoglycan (SLRP) of the extracellular matrix which was previously called the osteoinductive factor. SLRP are abundantly contained in the bone matrix, cartilage cells and connective tissues, and are thought to regulate cell proliferation, differentiation and adhesion in close association with collagen and many other growth factors.

In addition to the above mentioned references and application, Bone regeneration/ healing by Laser therapy has been proved in In-Vitro/ vivo models in hundreds of researches / applications over the past few decades both in animal & human model. Bone healing and bone engraftment by Infrared Low Level-Laser treatment is thought to be the cumulative coordinated effects of some specific physiological changes (in locally & systematically).They are -Ion Exchange and Bone Mineralization (19, 20), Nitric Oxide in Bone Formation (21, 22), Lymphatic Circulation (23, 24, and 25) and Bone Osteoblast Proliferation Increases Bone Formation (26, 27, 28, and 29), Osteoblast Gene Expression (30), and Osteoclast Inhibition Prevents Bone Mineral Resorption (31).

It is acknowledged that the controversy observed in the literature are due to different protocols used in which different wavelengths, association of wavelengths, different modes of emission and several doses were utilized in different animal or cell models. It is recognized that each method has its advantage and disadvantage.

The finding of this study is not consistent with some other research groups, which did not show positive effects of LLLT on healing bone [31, 32, 33, 34, 35 and37.], they did not consider the systemic effect of LLLT [23, 26]. They used the contra-lateral side of the same subject as controls. On the other hand, the findings of this investigation is very close to a study which found intense activity and high numbers of osteoblast 5-6 days after the procedure was performed on bone defects in a similar model.
Previous work using 790nm laser at a similar dose used in the present investigation, demonstrated a 10% increase on the amount of mineralized bone at seven days following irradiation. Another study examined bone consolidation, increased formation of trabecular bone, and the number of osteoblast after the use of He-Ne laser (633nm, 1mW, f ~1.1mm). The experimental period was seven days and doses per treatment were 3.15, 31.5 and 94.7J/cm2. Positive responses were found at 31.5 and 94.7J/cm2 but not at lower doses. These values were higher than that used in this work. This may indicate a more effective effect of 830nm laser light in comparison to lasers emitting 632.8 or 790nm, since 830nm penetrates to a deeper level.
The doses used in this study are in agreement with several previous reports that suggested that 8-10 J/cm2 induces positive effects on both bone and soft tissues (38, 39, and 40). A total dose (adult) of 32 J/ session is in accordance with the clinical parameters recommended- by Pinheiro et al. (41).
The literature shows that biomodulatory effects are dose dependent (42). It is also recognized that other factors such as the phase of cell growth (43) and the frequency and number of sessions (44) also influence the final result of the use of LLLT.
It is concluded that the use of LLLT at 830nm significantly improves bone healing during the early stages. Further studies are needed on the effects LLLT on growth factors, BMPs, prostaglandin and bone forming genes.

▪ Conclusion

Laser therapy is a standard therapeutic procedure, with unambiguous indications and contraindications. Among the reasons for this are: positive Clinical experiences, scientifically verified objective changes in tissue equilibrium caused by laser, and above all, better understanding of the mechanisms of laser effects. Clinical and experimental experience shows that laser therapy has its greatest effects on cells/tissue/organs® affected by a generally deteriorated condition with the ph. value lower than normal.
During the last decade, it was discovered that low-power laser irradiation has stimulatory effects on bone cell proliferation and gene expression.
The purposes of this review are to analyze the effects of low- power laser irradiation on bone cells and bone fracture repair, to examine what has been done so far, and to explore the additional works needed in this area. The studies reviewed show how laser therapy can be used to enhance bone repair at cell and tissue levels. As noted by researchers, laser properties, the combinations of wavelength and energy dose need to be carefully chosen so as to yield bone stimulation. With better study designs, the results will be more credible, allowing for greater recognition of advances in bone repair using laser therapy.
Many studies on the effects of laser therapy on bone healing and fracture repair have used biochemical and histological methods, but those subjective assessments are not enough for practical use rather objective assessment (clinical) should be preferred. However, in final sentence, in order to establish the effects of laser treatment on bone healing/ regeneration, additional studies need to be performed using clinical, radiological, biomechanical point of view, the ultimate evidences of bone repair.
Recommendations for Future Studies
This study has demonstrated the potential of low level laser therapy in treatment of enhancement of human bone fracture union. A large multicentric study pointing important -subjective (i.e. mechanical, biochemical and histological) as well as objective (clinical) parameters in addition to- laser protocol (dose, duration, type of laser & mode of operation),patient selection criteria and procedure of therapy is highly desirable to make this non-invasive method of bone stimulation applicable in medical science.

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