

Bone Healing – Formation – Resorption

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[Evidences of physical agents action on bone metabolism and their potential clinical use]

[Article in Portuguese]

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The action of physical agents such as low level laser therapy, low-intensity pulsed ultrasound and electrical and electromagnetic fields on bone have been often studied, showing that they are able to promote osteogenesis, accelerate fracture consolidation and augment bone mass. The use of these therapeutic modalities was first based on the finding that bone is a piezoelectric material, that means it can generate polarization when deformed, transforming mechanical energy into electric energy, and this has widen therapeutic possibilities to bony tissue. The present work aims to present evidences of physiologic effects and mechanisms of action of these physical agents on bone metabolism, based on articles published in international scientific literature.

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Bone Stimulation by Low Level Laser - A Theoretical Model for the Effects

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Abstract

The anecdotal and researched evidence for the effects of Low Level Laser Therapy (LLLT) on the stimulation of bone have been reported for over 20 years. This has been in the form of local as well as systematic effects - including contra-lateral effects. Reports of stimulation of rabbit radii fractures and mice femurs were made as early as 1986 and 1987 with irradiated bones healing faster than controls and contra-lateral non-treated fractures similarly demonstrating faster healing times. Over the following decade and a half, further studies have also investigated and demonstrated that LLLT is effective for the stimulation of bone tissue.

The reasons for this have been attributed to the general effects of LLLT and its ability to increase the rates of healing through mitochondrial ATP production and alteration in the cellular lipid bi-layer. Additional hypothesis include the subsequent capacity of irradiated cells to alter their ion exchange rate and thus influence the catalytic effects of the specific enzymes and substrates. These in turn initiate and promote the healing process completing the cascading cycle of events.

In the area of bone specific research, Dr. Tony Pohl of the Royal Adelaide Hospital in South Australia, has provided a new theory that postulates that the majority of fluid transfer and exchange within living bone is predominantly influenced by the lymphatic circulation.

LLLT is well documented and known as having effects that influence the lymphatic circulation and wound healing process. A coupling of these two areas of theory can demonstrate a positive description and explanation of the predominant effects of LLLT in bone stimulation. In reality, LLLT's effects on bone may well be a further consequence of its actions on the lymphatic circulation.

Reports of stimulation of Rabbit radii fractures were made by Tang in 1986 and similar reports by Trelles in 1987 on mice femurs. In both situations the irradiated bones healed faster than the controls. In another study by Hernandez-Ros, in 1987, LLLT demonstrated stimulation of fresh fractures on Sprague-Dawley rats that were fractured bilaterally. The unexpected results of this study were that the contra-lateral fractured non-treated limb also healed faster than the control group. Over the following decade and a half further studies (Yamada 1991; Pyczek, Sopala et al. 1994; Ozawa 1995; Horowitz 1996; Yaakobi 1996; Saito and Shimizu 1997) have also investigated and demonstrated that LLLT is effective for promoting the stimulation of bone healing. Recently Nicolau and colleagues (2002) from Brazil demonstrated the positive effect of LLLT on the stimulation of bone in mice with latent promotion of bone remodulation at injury sites without changes in bone architecture, increased bone volume and increased osteoblast surface through increased resorption and formation of bone with higher apposition rates. A positive effect on bony implants has been demonstrated by Dörtbudak (2002) and Guzzardella (2003). The effect of laser irradiation on osteoblastic cells has been reported by Yamamoto (2001) and Guzzardella (2002).

The reasoning for this amelioration in all experimental circumstances, based on electron microscopy as well as macroscopic histological evidence, was concluded to be due to i.a. improved vascularisation as a consequence of blood vessel formation, absorption of the haematoma, macrophage action, fibroblast proliferation, chondrocyte activity, bone remodeling from increased osteoblastic activity and deposition of calcium salts.

These changes and evidence based studies attribute the macro- and microscopic effects to the known and accepted general actions of LLLT and its ability to increase rates of healing through stimulation of ATP production, (Karu 1989; Smith 1990) promoting repair and polarization of the cellular lipid bilayer (Fenyo 1990) as well as LLLT's

capacity to affect cells through alterations in their exchange rate of ions (Robinson and Walters 1991) and influences the catalytic effects of the specific enzymes and substrates (Pouyssegur 1985; Karu 1988) which in turn initiate and promote the healing process.

More recent work by Dr. Tony Pohl, an internationally renowned Orthopaedic Surgeon from the Royal Adelaide Hospital in South Australia and lecturer at the Adelaide and South Australian Universities, has given rise to a new theory on bone circulation through reconsideration of fluid and protein transfer within bone (Pohl 1999). This theory suggests that the general understanding of the circulatory action within bone has been incorrect. Pohl postulates that the majority of fluid transfer and exchange within the living bone is predominantly influenced by the lymphatic rather than the vascular circulation. This is justified through studies on bone fluid input and output levels that have demonstrated that the venous and arterial aspect of circulation alone cannot account for the demonstrated levels of output nor the presence of free radical molecules which exceed those of the vascular input. Furthermore, the diameter of large protein cells within the bone exceed the diameter of the vessels that form the terminal aspects of the circulatory system making it impossible for them to have been delivered via this system. Consequently, an additional circulatory system must be present that will account for both the increased output and the presence of the large diameter protein cells as well as the free radicals.

If LLLT is then considered within the context of this new theory on bone circulation and the contribution of the lymphatic circulation then a further logical reasoned deduction for the action of LLLT on bone stimulation can be made. LLLT has a well documented and known effect influencing the lymphatic circulation. This has been demonstrated from the early works of Lievens, (1985) that demonstrated the influence of "Laser Irradiation" on the motricity of the lymphatic system and on the wound healing process. This is supported by several wound studies that demonstrate that the levels of protein rich exudates in non-healing wounds increase markedly from exposure to LLLT. This demonstrated action is determined to be as a result of the increase in lymphatic circulation (Robinson and Walters 1991; Gabel 1995). More recent work at the Flinders Medical Center in Adelaide South Australia has been completed and presented at the World Association of Laser Therapy conference in Tokyo Japan (Anderson, Carati et al. 2002). This study has demonstrated the positive effects of LLLT on the lymphatic circulation and its consequential benefits to the post mastectomy patient.

An understanding of the existing knowledge of the effects of LLLT on the lymphatic system combined with the hypothesis of bone fluid transport provides a coupled theory that would demonstrate a positive description and explain of the predominant effects of LLLT in bone stimulation.

In the trauma situation of direct or indirect damage to the bone, including fractures and periosteal induced damage such as stress fractures, the tissue damage leads to compromises that include but are not limited to, physical blockage from the trauma and waste / debris, increased fluid and circulatory viscosity from added cellular content within the lymphatics, lower speed motility and energy deficit in the tissue and cells from

the loss of ATP production as a general effect from the trauma, cell changes and inability of mitochondria to function at the normal higher level to promote self repair and regeneration.

LLLT with its known general effects and specific direct effects on the lymphatic system would act to stimulate mitochondria ATP that increases cellular and circulatory motility as well as directly influencing lymphatic flow. LLLT also promotes increased permeability in interstitial tissue and facial layers (Gabel 1995) reducing stagnation and blockage. These actions would assist the increase in lymphatic flow and consequently the circulation within the affected bone. There is also a hypothetical potential that the presence of LLLT by increasing lymphatic circulation does so by virtue of an increase in the diameter of the lymphatic vessels, not just by increased flow rates within the vessel at an unchanged diameter. This diameter increase, if definitively present, would also explain the presence of large diameter protein cells within the normal bone circulation that cannot be attributed to the vascular circulation and would additionally explain a facilitated process for removal of debris and larger protein cells passing out of traumatized areas that is additionally stimulated by the use of LLLT.

Stimulation of bone healing by LLLT has till now has been generally classified as a consequence of the general healing effects of LLLT. In reality LLLT's effect on bone may well be a further consequence of its actions on the lymphatic circulation.

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J Clin Laser Med Surg. 2003 Dec;21(6):383-8.

Effect of 830-nm laser light on the repair of bone defects grafted with inorganic bovine bone and decalcified cortical osseous membrane.

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OBJECTIVE: The aim of this study was to assess histologically the effect of LLLT (lambda830 nm) on the repair of standardized bone defects on the femur of Wistar albinus rats grafted with inorganic bovine bone and associated or not to decalcified bovine cortical bone membrane. **BACKGROUND DATA:** Bone loss may be a result of several pathologies, trauma or a consequence of surgical procedures. This led to extensive studies on the process of bone repair and development of techniques for the correction of bone defects, including the use of several types of grafts, membranes and the association of both techniques. There is evidence in the literature of the positive effect of LLLT on the healing of soft tissue wounds. However, its effect on bone is not completely understood. **MATERIALS AND METHODS:** Five randomized groups were studied: Group I (Control); Group IIA (Gen-ox); Group IIB (Gen-ox + LLLT); Group IIIA (Gen-ox + Gen-derm) and Group IIIB (Gen-ox + Gen-derm + LLLT). Bone defects were created at the femur of the animals and were treated according to the group. The animals of the irradiated groups were irradiated every 48 h during 15 days; the first irradiation was performed immediately after the surgical procedure. The animals were irradiated transcutaneously in four points around the defect. At each point a dose of 4 J/cm² was given (phi approximately 0.6 mm, 40 mW) and the total dose per session was 16 J/cm². The animals were humanely killed 15, 21, and 30 days after surgery. The specimens were routinely processed to wax, serially cut, and stained with H&E and Picosirius stains and analyzed under light microscopy. **RESULTS:** The results showed evidence of a more advanced repair on the irradiated groups when compared to non-irradiated ones. The repair of irradiated groups was characterized by both increased bone formation and amount of collagen fibers around the graft within the cavity since the 15th day after surgery, through analysis of the osteoconductive capacity of the Gen-ox and the increment of the cortical repair in specimens with Gen-derm membrane. **CONCLUSION:** It is concluded that LLLT had a positive effect on the repair of bone defect submitted the implantation of graft.

Lasers Med Sci. 2003;18(2):89-94.

Effect of low-power GaAlAs laser (660 nm) on bone structure and cell activity: an experimental animal study.

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Low-level laser therapy (LLL) is increasingly being used in the regeneration of soft tissue. In the regeneration of hard tissue, it has already been shown that the biomodulation effect of lasers repairs bones more quickly. We studied the activity in bone cells after LLLT close to the site of the bone injury. The femurs of 48 rats were perforated (24 in the irradiated group and 24 in the control group) and the irradiated group was treated with a GaAlAs laser of 660 nm, 10 J/cm² of radiant exposure on the 2nd, 4th, 6th and 8th days after surgery (DAS). We carried out histomorphometry analysis of the bone. We found that activity was higher in the irradiated group than in the control group: (a) bone volume at 5 DAS (p=0.035); (b) osteoblast surface at 15 DAS (p=0.0002); (c) mineral apposition rate at 15 and 25 DAS (p=0.0008 and 0.006); (d) osteoclast surface at 5 DAS and 25 DAS (p=0.049 and p=0.0028); and (e) eroded surface (

p=0.0032). We concluded that LLLT increases the activity in bone cells (resorption and formation) around the site of the repair without changing the bone structure.

Clin Oral Implants Res. 2003 Apr;14(2):226-32.

Osseointegration of endosseous ceramic implants after postoperative low-power laser stimulation: an in vivo comparative study.

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Stimulation with low-power laser (LPL) can enhance bone repair as reported in experimental studies on bone defects and fracture healing. Little data exist concerning the use of LPL postoperative stimulation to improve osseointegration of endosseous implants in orthopaedic and dental surgery. An in vivo model was used for the present study to evaluate whether Ga-Al-As (780 nm) LPL stimulation can improve biomaterial osseointegration. After drilling holes, cylindrical implants of hydroxyapatite (HA) were placed into both distal femurs of 12 rabbits. From postoperative day 1 and for 5 consecutive days, the left femurs of all rabbits were submitted to LPL treatment (LPL group) with the following parameters: 300 J/cm², 1 W, 300 Hz, pulsating emission, 10 min. The right femurs were sham-treated (control group). Three and 6 weeks after implantation, histomorphometric and microhardness measurements were taken. A higher affinity index was observed at the HA-bone interface in the LPL group at 3 (P<0.0005) and 6 weeks (P<0.001); a significant difference in bone microhardness was seen in the LPL group vs. the control group (P<0.01). These results suggest that LPL postoperative treatment enhances the bone-implant interface.

Laser therapy plays a role in bone healing Lasers Surg Med. 1998; 22: 97-102.

Luger et al. studied the effect of HeNe laser on the healing of tibial bone fractures in rats. 63 J (35 mW) was given transcutaneously daily over the fracture area. After 4 weeks the tibia was removed and tested at tension up to failure. The maximal load at failure and the structural stiffness of the tibia were found to be elevated significantly in the irradiated group, whereas the extension maximal load was reduced. In addition, gross non-union was found in four fractures in the control group, compared to none in the irradiated group.

Computerized morphometric assessment of the effect of low-level laser therapy on bone repair: an experimental animal study.

Silva Júnior AN, Pinheiro AL, Oliveira MG, Weismann R, Ramalho LM, Nicolau RA. J Clin Laser Med Surg. 2002; 20: 83-87

The aim of this study was to evaluate morphometrically the amount of newly formed bone after GaAlAs laser irradiation of surgical wounds created in the femur of rats. Low-level laser therapy (LLL) has been used in several medical specialties because of its biomodulatory effects on different biological tissues. However, LLL is still controversial because of contradictory reports. This is a direct result of the different methodologies used in these works. In this study, 40 Wistar rats were divided into four groups of 10 animals each: group A (12 sessions, 4.8 J/cm² per session, observation time of 28 days); group C (three sessions, 4.8 J/cm² per session, observation time of 7 days). Groups B and D acted as nonirradiated controls. The specimens were routinely processed to wax and cut at 6-microm thickness and stained with H&E. For computerized morphometry, Imagelab software was used. RESULTS: Computerized morphometry showed a significant difference between the areas of mineralized bone in groups C and D (p = 0.017). There was no difference between groups A and B (28 days; p = 0.383).

Effects of visible NIR low intensity laser on implant osseointegration in vivo.

Laser Med Surg Abstract issue, 2002: 11.
Blay A, Blay C C, Groth E B et al.

The effects of 680 and 830 nm lasers on osseointegration was studied by Blay. 30 adult rats were divided into three groups; two laser groups and one control. The rats in the two laser groups had pure titanium Frialit-2 implants implanted into each proximal metaphysis of their respective tibias, inserted with a 40 Ncm torque. The initial stability was monitored by means of a resonance frequency analyser. Ten irradiations were performed, 48 hours apart, 4 J/cm² on two points, starting immediately after surgery. Resonance frequency analysis indicated a significant difference between frequency values at 3 and 6 weeks, as compared to control. At 6 weeks the removal torque in the laser groups was much higher than in the control group.

Bone repair of the periapical lesions treated or not with low intensity laser (wavelength=904 nm).

Laser Surg Med. Abstract Issue 2002. abstract 303.
Sousa G R, Ribeiro M S, Groth E B.

The effect of bone repair in periapical lesions has been studied by Sousa []. 15 patients with a total of 18 periapical lesions were divided into two groups. One group received endodontic treatment and/or periapical surgery. The patients in the other group were submitted to the same procedure and in addition the lesions were irradiated by GaAs laser, 11 mW, 9 J/cm². This therapy was performed during 10 sessions with an interval of 72 hours. Bone regeneration was evaluated through X-ray examination. The results showed a significant difference between the laser and the control group in favour of the laser group.

J Photochem Photobiol B. 2003 May-Jun;70(2):81-9.

Low-power laser irradiation improves histomorphometrical parameters and bone matrix organization during tibia wound healing in rats.

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The influence of daily energy doses of 0.03, 0.3 and 0.9 J of He-Ne laser irradiation on the repair of surgically produced tibia damage was investigated in Wistar rats. Laser treatment was initiated 24 h after the trauma and continued daily for 7 or 14 days in two groups of nine rats (n=3 per laser dose and period). Two control groups (n=9 each) with injured tibiae were used. The course of healing was monitored using morphometrical analysis of the trabecular area. The organization of collagen fibers in the bone matrix and the histology of the tissue were evaluated using Picosirius-polarization method and Masson's trichrome. After 7 days, there was a significant increase in the area of neoformed trabeculae in tibiae irradiated with 0.3 and 0.9 J compared to the controls. At a daily dose of 0.9 J (15 min of irradiation per day) the 7-day group showed a significant increase in trabecular bone growth compared to the 14-day group. However, the laser irradiation at the daily dose of 0.3 J produced no significant decrease in the trabecular area of the 14-day group compared to the 7-day group, but there was significant increase in the trabecular area of the 15-day controls compared to the 8-day controls. Irradiation increased the number of hypertrophic osteoclasts compared to non-irradiated injured tibiae (controls) on days 8 and 15. The Picosirius-polarization method

revealed bands of parallel collagen fibers (parallel-fibered bone) at the repair site of 14-day-irradiated tibiae, regardless of the dose. This organization improved when compared to 7-day-irradiated tibiae and control tibiae. These results show that low-level laser therapy stimulated the growth of the trabecular area and the concomitant invasion of osteoclasts during the first week, and hastened the organization of matrix collagen (parallel alignment of the fibers) in a second phase not seen in control, non-irradiated tibiae at the same period. The active osteoclasts that invaded the regenerating site were probably responsible for the decrease in trabecular area by the fourteenth day of irradiation.

Clin Orthod Res. 2001 Feb;4(1):3-14.

The effects of low level laser irradiation on osteoblastic cells.

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Low level laser therapy has been used in treating many conditions with reports of multiple clinical effects including promotion of healing of both hard and soft tissue lesions. Low level laser therapy as a treatment modality remains controversial, however. The effects of wavelength, beam type, energy output, energy level, energy intensity, and exposure regime of low level laser therapy remain unexplained. Moreover, no specific therapeutic window for dosimetry and mechanism of action has been determined at the level of individual cell types. The aim of this study was to investigate the effects of low level laser irradiation on the human osteosarcoma cell line, SAOS-2. The cells were irradiated as a single or daily dose for up to 10 days with a GaAlAs continuous wave diode laser (830 nm, net output of 90 mW, energy levels of 0.3, 0.5, 1, 2, and 4 Joules). Cell viability was not affected by laser irradiation, with the viability being greater than 90% for all experimental groups. Cellular proliferation or activation was not found to be significantly affected by any of the energy levels and varying exposure regimes investigated. Low level laser irradiation did result in a heat shock response at an energy level of 2 J. No significant early or late effects of laser irradiation on protein expression and alkaline phosphatase activity were found. Investigation of intracellular calcium concentration revealed a tendency of a transient positive change after irradiation. Low level laser irradiation was unable to stimulate the osteosarcoma cells utilised for this research at a gross cell population level. The heat shock response and increased intracellular calcium indicate that the cells do respond to low level laser irradiation. Further research is required, utilising different cell and animal models, to more specifically determine the effects of low level laser irradiation at a cellular level. These effects should be more thoroughly investigated before low level laser therapy can be considered as a potential accelerator stimulus for orthodontic tooth movement.

Stomatologiia (Mosk). 2001;80(2):33-5.

[Prevention of inflammatory complications after mandibular osteosynthesis by a combination of low-frequency ultrasound and laser exposure]

[Article in Russian]

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Clinical and laboratory study of the efficiency of separate and combined use of low-frequency ultrasound and laser exposure of the operative wound for prevention of pyoinflammatory complications during mandibular osteosynthesis was carried out. Clinical parameters of wound reparation in the course of healing and microbiological and cytological findings in various methods of treatment are presented. The results evidence a high efficiency of these physical methods, particularly of their combination.

Bull Exp Biol Med. 2001 Apr;131(4):399-402.

Healing of bone fractures of rat shin and some immunological indices during magnetic laser therapy and osteosynthesis by the ilizarov method.

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The effect of magnetic and laser therapy on healing of bone fractures and blood levels of T and B lymphocytes was studied in rats during osteosynthesis by the Ilizarov method. Laser therapy induced changes in cells attesting to stimulation of reparative processes and normalization of immunological parameters.

J Clin Laser Med Surg. 2003 Dec;21(6):383-8.

Effect of 830-nm laser light on the repair of bone defects grafted with inorganic bovine bone and decalcified cortical osseous membrane.

Barbos Pinheiro AL, Limeira Junior Fde A, Marquez Gerbi ME, Pedreira Ramalho LM, Marzola C, Carneiro Ponzi EA, Oliveira Soares A, Bandeira De Carvalho LC, Vieira Lima HC, Oliveira Goncalves T. Laser Center, School of Dentistry, Federal University of Bahia, Salvador, Brazil. albp@ufba.br

OBJECTIVE: The aim of this study was to assess histologically the effect of LLLT (λ 830 nm) on the repair of standardized bone defects on the femur of Wistar albinus rats grafted with inorganic bovine bone and associated or not to decalcified bovine cortical bone membrane. **BACKGROUND DATA:** Bone loss may be a result of several pathologies, trauma or a consequence of surgical procedures. This led to extensive studies on the process of bone repair and development of techniques for the correction of bone defects, including the use of several types of grafts, membranes and the association of both techniques. There is evidence in the literature of the positive effect of LLLT on the healing of soft tissue wounds. However, its effect on bone is not completely understood. **MATERIALS AND METHODS:** Five randomized groups were studied: Group I (Control); Group IIA (Gen-ox); Group IIB (Gen-ox + LLLT); Group IIIA (Gen-ox + Gen-derm) and Group IIIB (Gen-ox + Gen-derm + LLLT). Bone defects were created at the femur of the animals and were treated according to the group. The animals of the irradiated groups were irradiated every 48 h during 15 days; the first irradiation was performed immediately after the surgical procedure. The animals were irradiated transcutaneously in four points around the defect. At each point a dose of 4 J/cm² was given (ϕ approximately 0.6 mm, 40 mW) and the total dose per session was 16 J/cm². The animals were humanely killed 15, 21, and 30 days after surgery. The specimens were routinely processed to wax, serially cut, and stained with H&E and Picrosirius stains and analyzed under light microscopy. **RESULTS:** The results showed evidence of a more advanced repair on the irradiated groups when compared to non-irradiated ones. The repair of irradiated groups was characterized by both increased bone formation and amount of collagen fibers around the graft within the cavity since the 15th day after surgery, through analysis of the osteoconductive capacity of the Gen-ox and the increment of the cortical repair in specimens with Gen-derm membrane. **CONCLUSION:** It is concluded that LLLT had a positive effect on the repair of bone defect submitted the implantation of graft.

J Clin Laser Med Surg. 2003 Oct;21(5):271-7.

Effects of pulse frequency of low-level laser therapy (LLLT) on bone nodule formation in rat calvarial cells.

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OBJECTIVE: The purpose of this study was to determine the effect of pulse frequencies of low-level laser therapy (LLLT) on bone nodule formation in rat calvarial cells in vitro. **BACKGROUND DATA:** Various photo-biostimulatory effects of LLLT, including bone formation, were affected by some irradiation factors such as total energy dose, irradiation phase, laser spectrum, and power density. However, the effects of pulse frequencies used during laser irradiation on bone formation have not been elucidated. **MATERIALS AND METHODS:** Osteoblast-like cells isolated from fetal rat calvariae were irradiated once with a low-energy Ga-Al-As laser (830 nm, 500 mW, 0.48-3.84 J/cm²) in four different irradiation modes: continuous irradiation (CI), and 1-, 2-, and 8-Hz pulsed irradiation (PI-1, PI-2, PI-8). We then investigated the effects on cellular proliferation, bone nodule formation, alkaline phosphatase (ALP) activity, and ALP gene expression. **RESULTS:** Laser irradiation in all four groups significantly stimulated cellular proliferation, bone nodule formation, ALP activity, and ALP gene expression, as compared with the non-irradiation group. Notably, PI-1 and -2 irradiation markedly stimulated these factors, when compared with the CI and PI-8 groups, and PI-2 irradiation was the best approach for bone nodule formation in the present experimental conditions. **CONCLUSION:** Since low-frequency pulsed laser irradiation significantly stimulates bone formation in vitro, it is most likely that the pulse frequency of LLLT an important factor affecting biological responses in bone formation.

Lasers Med Sci. 2003;18(2):78-82.

Effect of low-level laser irradiation on osteoglycin gene expression in osteoblasts.

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Many studies have attempted to elucidate the mechanism of the biostimulatory effects of low-level laser irradiation (LLLI), but the molecular basis of these effects remains obscure. We investigated the stimulatory effect of LLLI on bone formation during the early proliferation stage of cultured osteoblastic cells. A mouse calvaria-derived osteoblastic cell line, MC3T3-E1, was utilised to perform a cDNA microarray hybridisation to identify genes that induced expression by LLLI at the early stage. Among those genes that showed at least a twofold increased expression, the osteoglycin/mimecan gene was upregulated 2.3-fold at 2 h after LLLI. 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. We investigated the time-related expression of this gene by LLLI using a reverse transcription polymerase chain reaction (RT-PCR) method, and more precisely with a real-time PCR method, and found increases of 1.5-2-fold at 2-4 h after LLLI compared with the non-irradiated controls. These results suggest that the increased expression of the osteoglycin gene by LLLI in the early proliferation stage of cultured osteoblastic cells may play an important role in the stimulation of bone formation in concert with matrix proteins and growth factors.

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Effect of low-power laser irradiation on bony implant sites.

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This study was designed to examine the effects of low-energy laser irradiation on osteocytes and bone resorption at bony implant sites. Five male baboons with a mean age of 6.5 years were used in the study. Four holes for accommodating implants were drilled in each iliac crest. Sites on the left side were irradiated with a 100 mW low-energy laser (690 nm) for 1 min (6 Joule) immediately after drilling and insertion of four sandblasted and etched (Frialit-2 Synchro) implants. Five days later, the bone was removed en bloc and was evaluated histomorphometrically. The mean osteocyte count per unit area was 109.8 cells in the irradiated group vs. 94.8 cells in the control group. As intra-individual cell counts varied substantially, osteocyte viability was used for evaluation. In the irradiated group, viable osteocytes were found in 41.7% of the lacuna vs. 34.4% in the non-irradiated group. This difference was statistically significant at $P < 0.027$. The total resorption area, eroded surface, was found to be 24.9% in the control group vs. 24.6% in the irradiated group. This difference was not statistically significant. This study showed that osteocyte viability was significantly higher in the samples that were subjected to laser irradiation immediately after implant site drilling and implant insertion, in comparison to control sites. This may have positive effects on the integration of implants. The bone resorption rate, in contrast, was not affected by laser irradiation.

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Laser technology in orthopedics: preliminary study on low power laser therapy to improve the bone-biomaterial interface.

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Low Power Laser (LPL) seems to enhance the healing of bone defects and fractures. The effect of LPL in other orthopedic areas such as osteointegration of implanted prosthetic bone devices is still unclear. In the present study, 12 rabbits were used to evaluate whether Ga-Al-As (780 nm) LPL stimulation has positive effects on osteointegration. Hydroxyapatite (HA) cylindrical nails were drilled into both distal femurs of rabbits. From postoperative day 1 and for 5 consecutive days, the left femura of all rabbits were given LPL treatment (Laser Group-LG) with the following parameters: 300 Joule/cm², 1 Watt, 300 Hertz, pulsating emission, 10 minutes. The right femura were sham-treated (Control Group-CG). At 4 and 8 weeks after implantation, histologic and histomorphometric investigations evaluated bone-biomaterial-contact. Histomorphometry showed a higher degree of osteointegration at the HA-bone interface in the LG Group at 4 ($p < 0.0005$) and 8 weeks ($p < 0.001$). These preliminary positive results seem to support the hypothesis that LPL treatment can be considered a good tool to enhance the bone-implant interface in orthopedic surgery.