

**LaserWorld Science: 31 new abstracts, July 2005.**

### **1. Laser acupuncture special**

Litscher G, Rachbauer D, Ropele S, Wang L et al. **Acupuncture using laser needles modulates brain function: first evidence from functional transcranial Doppler sonography and functional magnetic resonance imaging.** *Lasers Med Sci.* 2004; 19 (1): 6-11.

The paper by Litscher presents an experimental double-blind study in laser acupuncture research in healthy volunteers, using a new optical stimulation method. 18 healthy volunteers (mean age +/- SD: 25.4 +/- 4.3 years; range: 21-30 years; 11 female, 7 male) were included in a randomized controlled cross-over trial using functional multidirectional transcranial ultrasound Doppler sonography (fTCD; n = 17) and performed functional magnetic resonance imaging (fMRI) in one volunteer. Stimulation of vision-related acupoints resulted in an increase of mean blood flow velocity in the posterior cerebral artery measured by fTCD [before stimulation (mean +/- SE): 42.2 +/- 2.5; during stimulation: 44.2 +/- 2.6; after stimulation: 42.3 +/- 2.4 cm/s, n.s.]. Mean blood flow velocity in the middle cerebral artery decreased insignificantly. Significant changes ( $p < 0.05$ ) of brain activity were demonstrated in the occipital and frontal gyrus by fMRI. Optical stimulation using properly adjusted laser needles has the advantage that the stimulation cannot be felt by the patient (painless and no tactile stimulation) and the operator may also be unaware of whether the stimulation system is active. Therefore true double-blind studies in acupuncture research can be performed.

Litscher G, Nemetz W, Smolle J, Schwarz G et al. **[Histological investigation of the micromorphological effects of the application of a laser needle--results of an animal experiment].** *Biomed Tech (Berl).* 2004; 49(1-2): 2-5. (in German)

In an experimental animal study Litscher investigated the effects of the new technique of laser needle stimulation (wavelength: 685 nm; energy density: 4.6 kJ/cm<sup>2</sup> per point; application duration: 20 min). The results revealed changes in microcirculatory parameters of the skin resulting in an increase in blood flow. However, the quality and intensity of the laser light did not induce micromorphological alterations in the skin.

Zalewska-Kaszubska J, Obzejta D. **Use of low-energy laser as adjunct treatment of alcohol addiction.** *Lasers Med Sci.* 2004; 19 (2): 100-104.

Zalewska-Kaszubska reports that fifty-three alcoholics were treated with two types of laser stimulation in four sessions. Each session consisted of 20 consecutive daily helium-neon laser neck biostimulations and 10 auricular acupuncture treatments with argon laser (every 2nd day). The Beck Depression Inventory-Fast Screen (BDI-FS) was used to assess their frame of mind before the session and after 2 months of treatment. Moreover, beta-endorphin plasma concentration was estimated five times using the radioimmunoassay (RIA) method. Improvement in BDI-FS and increase in beta-endorphin level were observed. These results suggest that laser therapy may be useful as an adjunct treatment for alcoholism.

O'Reilly B A, Dwyer P L, Hawthorne G et al. **Transdermal posterior tibial nerve laser therapy is not effective in women with interstitial cystitis.** *J Urol.* 2004; 172 (5 Pt 1): 1880-1883.

O'Reilly studied the effect of at-home laser acupuncture therapy. Interstitial cystitis (IC) is a debilitating condition which causes irritative bladder symptoms, pain and a decrease in health status. The pathophysiology is poorly understood so therapeutic options are diverse. Women meeting the National Institutes of Health National Institute for Diabetes and Digestive and Kidney Diseases criteria for IC were prospectively recruited and randomized to treatment (29) or placebo (27) cohorts in a double-blind trial. At home the patient performed laser therapy daily for 30 seconds over the SP6 acupuncture point for 12 weeks. Measures at baseline and at 84-day follow-up included the 7-day voiding diary, the Interstitial Cystitis Problem Index, Interstitial Cystitis Symptom Index and RAND 36-Item Health Survey questionnaires. There were no significant differences between the treatment and control cohorts on any of the measures. However, there was a significant decrease between baseline and 12-week follow-up in the amount voided, symptom problems and severity, and on all 8 SF-36 scales. There was no significant effect of fluid intake. This study demonstrated no difference between the active and sham device. However, it is interesting that treatment and control cohorts experienced similar improvements, suggesting that the control cohort improvements may have been due to participants' belief that they were receiving active treatment from the stimulator.

Gruber W, Eber E, Malle-Scheid D, Pflieger A et al. **Laser acupuncture in children and adolescents with exercise induced asthma.** *Thorax.* 2002; 57 (3): 222-225.

A double blind, placebo controlled, crossover study was performed by Gruber to investigate the possible protective effect of a single laser acupuncture treatment on cold dry air hyperventilation induced bronchoconstriction in 44 children and adolescents of mean age 11.9 years (range 7.5-16.7) with exercise induced asthma. Laser acupuncture was performed on real and placebo points in random order on two consecutive days. Lung function was measured before laser acupuncture, immediately after laser acupuncture (just before cold dry air challenge (CACH)), and 3 and 15 minutes after CACH. CACH consisted of a 4 minute isocapnic hyperventilation of -10 degrees C absolute dry air. Comparison of real acupuncture with placebo acupuncture showed no significant differences in the mean maximum CACH induced decrease in forced expiratory volume in 1 second (27.2 (18.2)% v 23.8 (16.2)%) and maximal expiratory flow at 25% remaining vital capacity (51.6 (20.8)% v 44.4 (22.3)%).

Wozniak P, Stachowiak G, Pieta-Dolinska A, Oszukowski P. **Laser acupuncture and low-calorie diet during visceral obesity therapy after menopause.** *Acta Obstet Gynecol Scand.* 2003; 82 (1): 69-73.

Wozniak reports that laser acupuncture can reduce postmenstrual obesity. The study population consisted of 74 postmenopausal females with visceral obesity that was divided into two groups according to an employed 6-month slimming procedure. In the first group (n = 36) a low-calorie diet was applied, while women in the second group (n = 38) were on the same kind of diet, having additionally one cycle of laser acupuncture procedure at the same time. At baseline and at the end of the study, body weight, body mass index and waist-to-hip ratio were determined in all women. After 6 trial months both groups exhibited a statistically significant drop in body weight, body mass index and waist-to-hip ratio. The mean reduction of body weight, body mass index and waist-to-hip ratio was significantly higher in the second group of women (laser acupuncture plus low-calorie diet).

Allais G, De Lorenzo C, Quirico P E, Lupi G et al. **Non-pharmacological approaches to chronic headaches: transcutaneous electrical nerve stimulation, laser therapy and acupuncture in transformed migraine treatment.** *Neurol Sci.* 2003; 24 Suppl 2: S138-142. In an open, randomized trial, Allais evaluated transcutaneous electrical nerve stimulation (TENS), infrared laser therapy and acupuncture in the treatment of transformed migraine, over a 4-month period free of prophylactic drugs. Sixty women suffering from transformed migraine were assigned, after a one month run-in period, to three different treatments: TENS (Group T; n=20), infrared laser therapy (Group L; n=20) or acupuncture (Group A; n=20). In each group the patients underwent ten sessions of treatment and monthly control visits. In Group T patients were treated for two weeks (5 days/week) simultaneously with three TENS units with different stimulation parameters (I: pulse rate = 80 Hz, pulse width = 120 micros; II: 120 Hz, 90 micros; III: 4 Hz, 200 micros). In Group L an infrared diode laser (27 mW, 904 nm) was applied every other day on tender scalp spots. In Group A acupuncture was carried out twice a week in the first two weeks and weekly in the next 6 weeks. A basic formula (LR3, SP6, LI4, GB20, GV20 and Ex-HN5) was always employed; additional points were selected according to each patient's symptomatology. The number of days with headache per month significantly decreased during treatment in all groups. The response in the groups differed over time, probably due to the different timing of applications of the three methods. TENS, laser therapy and acupuncture proved to be effective in reducing the frequency of headache attacks. Acupuncture showed the best effectiveness over time.

Ebneshahidi N S, Heshmatipour M, Moghaddami A, Eghtesadi A P. **The effects of laser acupuncture on chronic tension headache - a randomised controlled trial.** *Acupuncture in Medicine.* 2005; 23 (1): 13-18.

Fifty patients with chronic tension-type headache were randomly allocated to treatment or placebo groups. Patients in the treatment group received laser acupuncture to LU7, LI4, GB14, and GB20 bilaterally. Points were irradiated for 43 seconds, and the intensity was 1.3J (approximately 13 J/cm<sup>2</sup>). Ten sessions were given, three per week. The placebo group was treated in a similar way except that the output power of the equipment was set to zero. The outcome variables were headache intensity (VAS), duration of attacks, and number of days with a headache per month, by daily diary, assessed monthly to three months after treatment. There were significant differences between groups (P<0.001) in changes from baseline in months one, two and three, in median score for headache intensity (treatment group -5, -3 and -2, placebo group -1, 0 and 0), median duration of attacks (treatment group -6, -4 and -4, placebo group -1, 0 and 0 hours), and median number of days with headache per month (treatment group -15, -10 and -8, placebo group -2, 0 and 0).

Ilbuldu E, Cakmak A, Disci R, Aydin R. **Comparison of laser, dry needling, and placebo laser treatments in myofascial pain syndrome.** *Photomedicine and Laser Surgery.* 2004; 22 (4): 306-311.

Ilbuldu [1459] performed a placebo controlled, prospective, long-term follow up study with 60 patients who had trigger points in their upper trapezius muscles. The patients were divided into three groups randomly. Stretching exercises were taught to each group and they were asked to exercise at home. Treatment duration was 4 weeks. Placebo laser was applied to group 1, dry needling to group 2 and laser to group 3. HeNe laser was applied to three trigger points in the upper trapezius muscles on both sides. The patients were assessed before, post-treatment, and at 6 months after treatment for pain, cervical range of motion and functional status. The investigators observed a significant decrease in pain at rest, at activity, and

increase in pain threshold in the laser group compared to other groups. Improvement according to Nottingham Health Profile gave the superiority of the laser treatment. However, those differences among the groups were not observed at 6-month follow up.

## **2. Tissue repair**

Byrnes KR, Waynant RW, Ilev IK, Wu X, et al. **Light promotes regeneration and functional recovery and alters the immune response after spinal cord injury.** *Lasers Surg Med.* 2005; 36 (3): 171-185.

The effect of laser therapy on spinal cord injuries has been addressed by Byrnes. Adult rats underwent a T9 dorsal hemisection, followed by treatment with an 810 nm, 150 mW diode laser (dosage = 1,589 J/cm<sup>2</sup>). Axonal regeneration and functional recovery were assessed using single and double label tract tracing and various locomotor tasks. The immune response within the spinal cord was also assessed. Laser therapy with 6% power penetration to the spinal cord depth, significantly increased axonal number and distance of regrowth. It also returned aspects of function to baseline levels and significantly suppressed immune cell activation and cytokine/chemokine expression. These results demonstrate that light, delivered transcutaneously, improves recovery after injury and suggests that light will be a useful treatment for human spinal cord injury.

Ng G Y, Fung D T, Leung M C, Guo X. **Comparison of single and multiple applications of GaAlAs laser on rat medial collateral ligament repair.** *Lasers in Surgery and Medicine.* 2004; 34 (3): 285-289.

In the study by Ng sixteen rats were studied, with 12 receiving surgical transection to their right MCL and 4 receiving a sham injury. Group 1 (n = 4) received a single dose of GaAlAs laser therapy (wavelength 660 nm, average power 8.8 mW, pulse 10 kHz, dosage 31.6 J/cm<sup>2</sup>) directly to their MCL during surgery. Group 2 (n = 4) received 9 doses of GaAlAs laser therapy applied transcutaneously on alternate days (wavelength 660 nm, average power 8.8 mW, pulse 10 kHz, dosage 3.5 J/cm<sup>2</sup>). The controls (Group 3, n = 4) received one session of placebo laser at the time of surgery, with the laser equipment shut down, while the sham injured Group 4 (n = 4) received no treatment. Biomechanical tests for structural stiffness, ultimate tensile strength (UTS), and load-relaxation were done at 3 weeks after injury. The stiffness and UTS data were normalized by expressing as a percentage of the left side of each animal before statistical analysis. The load-relaxation data did not show any differences between the groups. The normalized stiffness levels of Groups 2 (81.08±11.28%) and 4 (92.66±13.19%) were significantly higher (P = 0.025) than that of the control Group 3 (58.99±15.91%). The normalized UTS of Groups 2 (81.38±5.68%) and 4 (90.18±8.82%) were also significantly higher (P = 0.012) than that of the control (64.49±9.26%). Although, Group 1 had higher mean stiffness and UTS values than the control, no statistically significant difference was found between these two groups. Multiple laser therapy improves the normalized strength and stiffness of repairing rat MCLs at 3 weeks after injury. The multiple treatments seem to be superior to a single treatment when the cumulative dosages are comparable between the two modes of application.

Lanzafame R J, Stadler I, Coleman J, Haerum B, Oskoui P, Whittaker M, Zhang R Y. **Temperature-controlled 830-nm low-level laser therapy of experimental pressure ulcers.** *Photomedicine and Laser Surgery.* 2004; 22 (6): 483-488.

The negligible effect of the presence of heat in laser therapy has been well demonstrated by Lanzafame. Pressure ulcers were created in mice by placing the dorsal skin between two round ceramic magnetic plates for three 12-h cycles. Animals were divided into three groups (n = 9) for daily light therapy, 830 nm, 5.0 J/cm<sup>2</sup> on days 3-13 post ulceration in both groups A and B. A special heat-exchange device was applied in Group B to maintain a constant temperature at the skin surface (30 degrees C). Group C served as controls, with irradiation at 5.0 J/cm<sup>2</sup> using an incandescent light source. Temperature of the skin surface, and temperature alterations during treatment were monitored. The wound area was measured and the rate and time to complete healing were noted. The maximum temperature change during therapy was 2.0 +/- 0.64 degrees C in Group A, 0.2 +/- 0.2 degrees C in Group B and 3.54 degrees C +/- 0.72 in Group C. Complete wound closure occurred at 18 +/- 4 days in Groups A and B and 25 +/- 6 days in Group C. The percentage of the wound closure at 14 days was 75.4 +/- 7.2% and 77.7 +/- 5.6% for Groups A and B, respectively (NS differences). However, animals in Group C demonstrated a wound closure of 36.3 +/- 4.8%. These results demonstrate that the salutary effects of laser therapy on wound healing are temperature independent in this model.

Stadler I, Lanzafame R J, Oskoui P, Zhang R Y, Coleman J, Whittaker M. **Alteration of skin temperature during low-level laser irradiation at 830 nm in a mouse model.** Photomedicine and Laser Surgery. 2004; 22 (3): 227-231.

The temperature increase in tissue of black as well as white mice was investigated by Stadler. Irradiation at 830 nm and 5.0 J/cm<sup>2</sup> fluence induced a small temperature increase at the surface and at 1 mm in depth. The smaller effects seen in white mice might be due in part to reflection. This suggests that the thermal effects of colour should be considered, particularly at higher fluences.

Ribeiro MS, Da Silva Dde F, De Araujo CE, De Oliveira SF, Pelegrini CM, Zorn TM, Zezell DM. **Effects of low-intensity polarized visible laser radiation on skin burns: a light microscopy study.**

This study was carried out to investigate the influence of low-intensity polarized visible laser radiation on the acceleration of skin wound healing. Background Data: Low-level laser therapy at adequate wavelength, intensity, and dose can accelerate tissue repair. However, there is still unclear information about light characteristics, such as coherence and polarization. Some studies indicate that linearly polarized light can survive through long propagation distance in biological tissue. Three burns about 6 mm in diameter were created on the back of rats with liquid N<sub>2</sub>. Lesion "L(//)" was irradiated by He-Ne laser (lambda = 632.8 nm), D= 1.0J/cm<sup>2</sup>, with linear polarization parallel to the spinal column of the rat. Lesion "L(inverted v)" was irradiated using the same laser and dose, but the light polarization was aligned perpendicularly to the relative orientation. Lesion "C" was not irradiated in order to be considered as control. The animals were sacrificed at day 3-17 after lesion creation. Samples were collected and prepared for histological analysis. Histological analysis showed that the healing of irradiated wounds was faster than that of non-irradiated wounds. Moreover, it was observed that skin wound repair is dependent on polarization orientation with respect to a referential axis as the animal's spinal column. Consequently, "L(//)" was completely healed after 17 days, whereas "L (perpendicular) " showed a moderate degree of healing after the same period. These results indicate that the relative direction of the laser polarization plays an important role in the wound healing process when highly coherent He-Ne laser is used.

Nussbaum E L, Lilge L, Mazzulli T. **Effects of low-level laser therapy (LLLT) of 810 nm upon in vitro growth of bacteria: relevance of irradiance and radiant exposure.** J Clin Laser Med Surg. 2003; 21 (5): 283-290.

In an in vitro study by Nussbaum Escherichia coli, Pseudomonas aeruginosa, and Staphylococcus aureus were irradiated using a wavelength of 810 nm at irradiances of 0.015 W/cm<sup>2</sup> (0-50 J/cm<sup>2</sup>) and 0.03 W/cm<sup>2</sup> (0-80 J/cm<sup>2</sup>). Bacteria were counted after 20 h of incubation. Laser effects varied significantly with species. P.aeruginosa growth decreased overall dependent on an interaction of irradiance and radiant exposure; greatest inhibition was produced using high irradiance delivering radiant exposures in the range of 1-20 J/cm<sup>2</sup> In contrast, E. coli growth increased overall regardless of irradiance; greatest effects were produced using low radiant exposures (1-20 J/cm<sup>2</sup>). There was a main effect for irradiance on S. aureus growth; however, growth was not different compared with controls. Additional analysis showed that there were differences in growth of .aeruginosa when comparing samples that were matched by exposure times (66, 329, 658, 1316, 1974, and 2632 sec) rather than radiant exposure; this suggests that irradiance rather than exposure time was the significant factor in P. aeruginosa inhibition.: These findings have immediate relevance in the use of laser for infected wounds. Exposure to 810 nm irradiation (0.03 W/cm<sup>2</sup>) could potentially benefit wounds infected with P. aeruginosa. However, increased E. coli growth could further delay recovery.

Brosseau L, Wells G, Marchand S, Gaboury I et al. **Randomized controlled trial on low level laser therapy (LLLT) in the treatment of osteoarthritis (OA) of the hand.** Lasers Surg Med. 2005; 36 (3): 210-219.

Low level laser therapy (LLLT) offers promising symptomatic relief of osteoarthritic (OA) pain. We examined efficacy of active LLLT versus sham LLLT on finger joints and three superficial nerves. OA-patients randomly assigned, received three treatments per week for 6 weeks of LLLT (n = 42) or sham LLLT (n = 46). Pain relief, morning stiffness, and functional status did not significantly improve for LLLT versus placebo. No significant differences were found in finger range of motion, except carpometacarpal opposition (P = 0.011), grip strength, and patient global assessment which improved for active LLLT participants (P = 0.041). LLLT is no better than placebo at reducing pain, morning stiffness, or improving functional status for OA-hand patients.

Al-Watban F A, Zhang X Y. **The comparison of effects between pulsed and CW lasers on wound healing.** J Clin Laser Med Surg. 2004; 22 (1):15-18.

Al-Watban compared the wound healing effect of pulsed and continuous 635 nm laser. Continuous beam provide the best results, and the 100 Hz frequency the best among the different frequencies used. And this frequency happens to be the one closest to continuous among the frequencies used, which further underlines the superiority of the continuous beam for wound healing procedures.

Ueda Y, Shimizu N. **Pulse irradiation of low-power laser stimulates bone nodule formation.** Journal of Oral Science. 2001; 43 (1): 55-60.

The effect of pulsing is studied by Ueda. Osteoblastic cells isolated from fetal rat calvariae were irradiated once with GaAlAs laser (830 nm, 500 mW) in two different irradiation modes; continuous irradiation (CI), and 1 Hz pulsed irradiation (PI). The author then investigated the effects on cellular proliferation, bone nodule formation, alkaline phosphatase (ALP) activity,

and ALP gene expression. Laser irradiation in both groups significantly stimulated cellular proliferation, bone nodule formation, ALP activity, and ALP gene expression, as compared with the non-irradiation group. Notably, 1 Hz markedly stimulated these factors, when compared with the continuous group. In this study the lowest possible pulse mode was compared to continuous, and the possible effect of higher frequencies is not known.

Demir H, Yaray S, Kirnap M, Yaray K. **Comparison of the effects of laser and ultrasound treatments on experimental wound healing in rats.** *J Rehabil Res Dev.* 2004; 41 (5): 721-728.

Demir performed a randomised controlled study investigating the effects of ultrasound and laser treatments on wound healing in rats. The duration of the inflammatory phase decreased with both laser and ultrasound treatments; however, laser was more effective than ultrasound, with more significant results. The proliferation phase showed, for both treatments, an increase in the level of hydroxyproline and the number of fibroblasts, as well as stimulation of the collagen synthesis and the composition. Laser treatment was again more effective than ultrasound. The wound breaking strength was significantly higher with both treatments, and no statistically significant difference emerged between the laser and ultrasound groups, although laser treatment provided a much greater increase in the wound breaking strength than ultrasound. Both treatments have beneficial effects on the inflammatory, proliferation, and maturation phases of wound healing. Both can be used successfully for decubitus ulcers and chronic wounds, in conjunction with conventional therapies such as debridement and daily wound caring. However, laser treatment was more effective than ultrasound in the first two phases of wound healing.

Demir H, Menku P, Kirnap M, Calis M, Ikizceli I. **Comparison of the effects of laser, ultrasound, and combined laser + ultrasound treatments in experimental tendon healing.** *Lasers Surg Med.* 2004; 35 (1): 84-89.

In another study by Demir 84 healthy male rats were divided into three groups consisting of 28 rats, the left Achilles tendons were used as treatment and the right Achilles tendons as controls. The right and left Achilles tendons of rats were traumatised longitudinally. The treatment was started on post injury day one. The US treatment was applied with a power of 0.5 W/cm<sup>2</sup>, a frequency of 1 MHz, continuously, 5 minutes daily. A GaAs laser was applied with a 904 nm wavelength, 6 mW average power, 1 J/cm<sup>2</sup> dosage, 16 Hz frequency, for 1 minute duration. Although US, L, and combined US + L treatments increased tendon healing biochemically and biomechanically more than the control groups, no statistically significant difference was found between them. Also the researchers did not find significantly more cumulative positive effects of combined treatment.

### **3. Dental**

Marsilio AL, Rodrigues JR, Borges AB. **Effect of the clinical application of the GaAlAs laser in the treatment of dentine hypersensitivity.** *J Clin Laser Med Surg.* 2003; 21(5): 291-296.

In the study by Marsilio 25 patients, with a total number of 106 cases of dentinal hypersensitivity (DH), were treated with GaAlAs laser therapy. 65% of the teeth were premolars; 14% were incisors and molars; 6.6% were canines. The teeth were irradiated with 3 and 5 J/cm<sup>2</sup> for up to six sessions, with an interval of 72 h between each application, and they were evaluated initially, after each application, and at 15 and 60 days follow-up post-

treatment. The treatment was effective in 86.53% and 88.88% of the irradiated teeth, respectively, with the minimum and maximum energy recommended by the manufacturer. There was a statistically significant difference between DH and after a follow-up of 60 days for both groups. The difference among the energy maximum and minimum was not significant.

Almeida Lopes L, Lopes A. **Using Laser Therapy on the lymphatic drainage technique.** Photomedicine and Laser Surgery. 2005; 23 (1). Abstracts from the 5th Congress of the World Association for Laser Therapy, São Paulo, Brazil, November 2004. Abstract no 042, p.100.

Almeida Lopes points out that laser irradiation might enhance not only tissue but also microorganisms in infected areas. In these cases, and in particular for immune depressive patients, a lymph drainage technique can be used. With knowledge of the lymphatic anatomy the lymph nodes involved in the inflammatory process can be irradiated in cases such as periocoronitis, herpes simplex, endodontic abscesses and avleolitis. The author suggests a dose around 70 J/cm<sup>2</sup> with a two-day interval.

#### **4. LED therapy special**

Schubert V. **EFFECTS OF PHOTOTHERAPY (LLLT) ON PRESSURE ULCER HEALING IN ELDERLY PATIENTS AFTER A FALLING TRAUMA. A PROSPECTIVE, RANDOMIZED, CONTROLLED STUDY.**

The effects of infrared and red pulsed monochromatic light, with varied pulsations and wavelengths, on the healing of pressure ulcers were evaluated in this prospective, randomized, controlled study. Elderly patients (> or =65 years) with Stage 2 or 3 skin ulcers were enrolled and assigned to one of two groups. Both groups were given the same standard ulcer therapy. One group was also given phototherapy with pulsed monochromatic infrared (956 nm) and red (637 nm) light. Treatments lasted 9 min each time using a regimen with pulse repetition frequency varied between 15.6 Hz and 8.58 kHz. Patients were followed for 10 weeks or until the ulcer was healed, whichever occurred first. The ulcer surface area was traced weekly. Patients treated with pulsed monochromatic light had a 49% higher ulcer healing rate, and a shorter time to 50% and to 90% ulcer closure compared with controls. The results are encouraging as pulsed monochromatic light increased healing rate and shortened healing time. This will positively affect the quality of life in elderly patients with pressure ulcer.

Eells J T, Henry M M, Summerfelt P, Wong-Riley M T, Buchmann E V et al. **Therapeutic photobiomodulation for methanol-induced retinal toxicity.** Proc Natl Acad Sci U S A. 2003; 18; 100 (6): 3439-3444.

The study by Eells was undertaken to test the hypothesis that exposure to monochromatic red radiation from light-emitting diode (LED) arrays would protect the retina against the toxic actions of methanol-derived formic acid in a rodent model of methanol toxicity. Using the electroretinogram as a sensitive indicator of retinal function, it was demonstrated that three brief (2 min, 24 s) 670 nm LED treatments (4 J/cm<sup>2</sup>), delivered at 5, 25, and 50 h of methanol intoxication, attenuated the retinotoxic effects of methanol-derived formate. This study documents a significant recovery of rod- and cone-mediated function in LED-treated, methanol-intoxicated rats. It is further shown that LED treatment protected the retina from the histopathologic changes induced by methanol-derived formate. These findings provide a link between the actions of monochromatic red to near-IR light on mitochondrial oxidative

metabolism in vitro and retinoprotection in vivo. They also suggest that photobiomodulation may enhance recovery from retinal injury and other ocular diseases in which mitochondrial dysfunction is postulated to play a role.

Whelan H T, Buchmann E V, Dhokalia A, Kane M P et al. **Effect of NASA light-emitting diode irradiation on molecular changes for wound healing in diabetic mice.** J Clin Laser Med Surg. 2003; 21(2): 67-74.

The purpose of the study by Whelan was to assess the changes in gene expression of near-infrared light therapy in a model of impaired wound healing. Polyvinyl acetal (PVA) sponges were subcutaneously implanted in the dorsum of BKS.Cg-m <sup>+/+</sup> Lepr(db) mice. LED treatments were given once daily, and at the sacrifice day, the sponges, incision line and skin over the sponges were harvested and used for RNA extraction. The RNA was subsequently analyzed by cDNA array. The study revealed certain tissue regenerating genes that were significantly upregulated upon LED treatment when compared to the untreated sample. Integrins, laminin, gap junction proteins, and kinesin superfamily motor proteins are some of the genes involved during regeneration process. These are some of the genes that were identified upon gene array experiments with RNA isolated from sponges from the wound site in mouse with LED treatment.

Wong-Riley M T, Liang H L, Eells JT, Chance B et al. **Photobiomodulation directly benefits primary neurons functionally inactivated by toxins: role of cytochrome c oxidase.** J Biol Chem. 2005; 11; 280 (6): 4761-4771.

Previous studies using 670 nm light-emitting diode (LED) arrays suggest that cytochrome c oxidase, a photoacceptor in the NIR range, plays an important role in therapeutic photobiomodulation. If this is true, then an irreversible inhibitor of cytochrome c oxidase, potassium cyanide (KCN), should compete with LED and reduce its beneficial effects. This hypothesis was tested on primary cultured neurons in a study by Wong-Riley [1499]. LED treatment partially restored enzyme activity blocked by 10-100 micromol KCN. It significantly reduced neuronal cell death induced by 300 micromol KCN from 83.6 to 43.5%. However, at 1-100 mM KCN, the protective effects of LED decreased, and neuronal deaths increased. LED significantly restored neuronal ATP content only at 10 micromol KCN but not at higher concentrations of KCN tested. Pretreatment with LED enhanced efficacy of LED during exposure to 10 or 100 micromol KCN but did not restore enzyme activity to control levels. In contrast, LED was able to completely reverse the detrimental effect of tetrodotoxin, which only indirectly down-regulated enzyme levels. Among the wavelengths tested (670, 728, 770, 830, and 880 nm), the most effective ones (830 nm, 670 nm) paralleled the NIR absorption spectrum of oxidized cytochrome c oxidase, whereas the least effective wavelength, 728 nm, did not. The results are consistent the hypothesis that the mechanism of photobiomodulation involves the up-regulation of cytochrome c oxidase, leading to increased energy metabolism in neurons functionally inactivated by toxins.

## **5. Mechanisms**

Lubart R, Eichler M, Lavi R, Friedman H, Shainberg A. **Low-energy laser irradiation promotes cellular redox activity.** Photomedicine and Laser Surgery. 2005; 23 (1): 3-9.

In a search for chromophores responsible for photobiostimulation, endogenous porphyrins, mitochondrial and membranal cytochromes, and flavoproteins were found to be suitable

candidates This is described in the review article by Lubart. The above-mentioned chromophores are photosensitizers that generate reactive oxygen species (ROS) following irradiation. As the cellular redox state has a key role in maintaining the viability of the cell, changes in ROS may play a significant role in cell activation. In the present review, we summarize evidence demonstrating that various ROS and antioxidants are produced following laser illumination. It was found that very little evidence for NO formation in illuminated non-vascular smooth muscle cells exists in the literature. The author suggests that the change in the cellular redox state, which plays a pivotal role in maintaining cellular activities, leads to photobiostimulative processes.

Kujawa J, Zavodnik I B, Lapshina A, Labieniec M, Bryszewska M. **Cell survival, DNA, and protein damage in B14 cells under low-intensity near-infrared (810 nm) laser irradiation.** *Photomedicine and Laser Surgery.* 2004; 22 (6): 504-508.

Kujawa studied the cytotoxic effects of laser therapy. B14 cells and a suspension of human erythrocyte membranes were irradiated with near-infrared (810 nm) therapy laser at different radiant exposures (3.75-15.0 J/cm<sup>2</sup>) and light power (fluency rate) 200 mW at 22 degrees C. Laser induced cellular oxidative damage was measured in terms of cell survival, DNA damage, measured using the method of single cell gel electrophoresis, protein damage measured as protein carbonyls formation. No substantial changes of cell survival under B14 cells irradiation at radiant exposures 3.75-11.25 J/cm<sup>2</sup> were observed. Similarly, neither considerable light-induced DNA damage or protein carbonyls accumulation was revealed. On the contrary, laser irradiation has led to decrease of cell protein carbonyl groups level in a dose-dependent manner. Additionally, using human red blood cell membranes as model membranes and biological oxidant HOCl we observed that laser irradiation resulted in a decrease of the level of membrane protein carbonyl groups accumulated under oxidative HOCl treatment.

Ciprian Antipa, Mihail-Lucian Pascu, Viorica Stanciulescu, Mihaela Vlaiculescu, Elena Ionescu, Daniel Bordea. **Coherent and noncoherent low-power diodes in clinical practice.** *Proc. SPIE Vol. 2981*, p. 236-241, Coherence Domain Optical Methods in Biomedical Science and Clinical Applications; Valery V. Tuchin, Halina Podbielska M.D., Ben Ovrin; Eds. Clinical efficacy of the low power laser (LPL) in medical treatments is still not well established. In a double blind, placebo controlled study, we tried to find out first which type of LPL is more efficient, and second if coherence is an important character for clinical efficacy. We treated 1228 patients having different rheumatic diseases, with low power diode, used as follows: A group: IR coherent diode, continuous emission, 3 mW power; B group: IR coherent diode, pulsed emission, output power about 3 mW; C group: IR noncoherent diode continuous emission 9 mW power; D group: both IR diode lasers (continuous or pulsed) and HeNe laser, continuous emission, 2 mW power; E group: placebo laser as control group. The energy dose used for every group was the same, as well as the clinical protocols. The positive results were: 66.16% for A group; 64.06% for B group; 48.87% for C group; 76.66% for D group, and 39.07% for E group. Finally, we showed that LPL is really efficient in the treatment of some rheumatic diseases, especially when red and IR diode laser were used in combination. The type of emission (continuous or pulsed) is not important, but coherence is obviously necessary for clinical efficacy.

Silva JC, Lacava ZG, Kuckelhaus S, Silva LP et al. **Evaluation of the use of low level laser and photosensitizer drugs in healing.** *Lasers Surg Med.* 2004; 34 (5): 451-457.

The present study analyses the effect of low level laser InGaAlP (685 nm), radiation, either alone or combined with a phthalocyanine-derived photosensitizer (PS) in a gel base delivery (GB) system, on the healing process of cutaneous wounds in rats. The rats were divided into six groups: control (untreated) (CG), gel base (GB), photosensitizer (PS), laser (LG), laser+photosensitizer (LPS), and laser+photosensitizer in a GB (LPSG). Standardized circular wounds were made on the dorsum of each rat with a skin punch biopsy instrument. After wounding, treatment was performed once daily and the animals were killed at day 8. Tissue specimens containing the whole wound area were removed and processed for histological analysis using conventional techniques. Serial cross-sections were analyzed to evaluate the organization of the dermis and epidermis as well as collagen deposition. The animals of groups LG, PS, LPS, and LPSG presented higher collagen content and enhanced re-epithelialization as compared to CG (control) and GB rats. Connective tissue remodelling was more evident in groups LPS and LPSG. The results clearly indicated a synergetic effect of light + photosensitizer + delivery drug on tissue healing. PDT did not cause any healing inhibition or tissue damage during the healing process.

Karu TI, Pyatibrat LV, Kalendo GS. **Donors of NO and pulsed radiation at lambda = 820 nm exert effects on cell attachment to extracellular matrices.** *Toxicol Lett.* 2001; 8;121 (1): 57-61.

The adhesion of HeLa cells to a glass matrix was evaluated after the irradiation of the cell suspension with a pulsed near-infrared light-emitting diode ( $\lambda = 820$  nm, frequency 10 Hz, dose 8-120 J/m<sup>2</sup>) and treatment with two donors of nitric oxide, sodium nitroprusside (SNP,  $5 \times 10^{-4}$  M) and NaNO<sub>2</sub> ( $4 \times 10^{-4}$  M). It was found that in an irradiated cell suspension, the cell-glass adhesion increases in a dose-dependent manner (a bell-shaped curve with a maximum at 60 J/m<sup>2</sup>). The treatment of cells with SNP or NaNO<sub>2</sub> before the irradiation eliminates the radiation-induced attachment stimulation. Pretreatment of cells with SNP not only eliminates the radiation-induced attachment stimulation but inhibits the attachment of irradiated (but not non-irradiated) cells. It is suggested that a modulation of the activity of respiratory chain (probably the alteration of the activity of cytochrome c oxidase) is involved in radiation-induced increase of cell attachment