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Non-Thermal, Non-Ablative CO_2 Laser Therapy (NACLT): A New Approach to Relieve Pain in Some Painful Oral Diseases

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1. Introduction

CO_2 laser has been used as a very useful device in surgery for ablation, coagulation and cutting the tissues for the last four decades. It is interesting to know that this high power laser can also be used as a therapeutic laser for immediate pain reduction in some oral lesions without any visible side effects such as ulceration, erosion formation and even erythema.

Recently few case reports and clinical trials have been published about using CO_2 laser in non-ablative manner to reduce pain in oral lesions. In these studies, the oral painful lesions were irradiated through a layer of transparent, non-anesthetic gel with high water content to reduce the beam absorption by the soft tissue. The patients reported immediate and significant pain relief after laser irradiation. The procedure was painless and anesthesia was not required. This technique was called non-thermal, Non-Ablative CO_2 Laser Therapy (NACLT). The results of powermetry and thermometry demonstrated the low power nature of NACLT. However there are some differences between analgesic effects of NACLT and the other classical low power lasers which will be discussed in the next sections.

To provide a comprehensive understanding of NACLT, this chapter is organized in several sections. First, due to low level therapeutic nature of NACLT, conventional low power therapeutic lasers, their biological effects and their pain relieving properties are reviewed. Then, a discussion about the interesting analgesic effects of CO_2 lasers is presented. In the next section, NACLT as a new low level laser therapy procedure and its pain relieving applications in painful oral lesions is discussed. Finally, the presumed mechanisms of analgesic effects of NACLT are covered.

2. Low level laser therapy (laser phototherapy)

2.1 History

Low-level laser (or light) therapy (LLLT) has been investigated and used clinically for over 40 years. However, it is only in relatively recent times that LLLT has become scientifically and clinically accepted by even a fraction of the medical community (Hamblin 2010).

The history of the use of laser phototherapy in medicine goes back to the late 1960s, only eight years after the invention of the first laser (Ruby laser) by Theodore Maiman. In 1967, Endre Mester in Semmelweis University, Budapest, Hungary decided to test if laser radiation might cause cancer in mice. He shaved the dorsal hair of the mice, divided them into two groups and irradiated the shaved areas with a low powered ruby laser (694-nm) in one group. They did not get cancer and to his surprise the hair on the treated group grew back more quickly than the untreated group. This was the first demonstration of "laser biostimulation" (Hamblin, Waynant et al. 2006).

In early 1960's, the first low level laser, Helium-Neon was developed by Professor Ali Javan. It emits visible, red light with a wavelength of 632.8nm. This low power laser has been used extensively in experimental and therapeutic studies. Today, the semiconductor lasers, including InGaAlP lasers (633-700nm), GaAlAs lasers (780-890nm, invisible, near infrared area), GaAs laser (904nm, invisible, near infrared area) are widely used by researchers and clinicians.

LLLT originally thought to be a peculiar property of laser light (soft or cold lasers); the subject has now broadened, using non-coherent light (light-emitting diodes, LEDs). Today, medical treatment with coherent-light sources (lasers) or noncoherent light (LEDs) has passed through its childhood and adolescence (Hamblin, Waynant et al. 2006). Currently, low-level laser (or light) therapy (LLLT) is practiced as part of physical therapy in many parts of the world. Although LLLT was used mainly for wound healing and pain relief, the medical applications of LLLT have broadened to include diseases such as stroke, myocardial infarction, and degenerative or traumatic brain disorders (Hashmi, Huang et al. 2010).

Although many experimental and clinical studies have reported the positive effects of phototherapy to promote wound healing , pain relief and anti-inflammatory effects, some negative reports also have been published, further confounding the issue (Demidova-Rice, Salomatina et al. 2007), for instance regarding the application of laser phototherapy on wound healing (Posten, Wrone et al. 2005). This controversy seems to be due to two main reasons; first of all, the basic biochemical mechanisms underlying these biological effects are not completely understood. Secondly, the complexity of rationally choosing amongst a large number of laser irradiation parameters (such as wavelength, fluence, power density, pulse structure and treatment timing), inappropriate anatomical treatment location and concurrent patient medication (such as steroidal and non-steroidal anti-inflammatories which can inhibit healing) has led to conflicting results and publication of a number of unfavourable, as well as many favourable, studies. In particular a biphasic dose response has been frequently observed where low levels of light have a much better effect than higher levels (Gigo-Benato, Geuna et al. 2005; Aimbire, Albertini et al. 2006; Hamblin, Waynant et al. 2006; Goncalves, Souza et al. 2007; Huang, Chen et al. 2009; Hamblin 2010).

It should be noticed that LLLT has a diversified terminology. It is also called "cold laser", "soft laser", "biostimulation" , "photobiomodulation", "low intensity laser therapy" , "low energy laser therapy", "laser phototherapy (LPT)", "laser therapy", and "non-ablative irradiation". Some investigators state that using frequent terms, such as "low power laser therapy" is misleading, since high power lasers, too, can be used for laser phototherapy (Tuner and Hode 2010) (as we will discuss in the next sections, CO_2 laser is applied as a low

level (therapeutic laser) in NACLT, too). Some of the researchers favour the term "laser phototherapy (LPT)" which is an emerging terminology (Tuner and Hode 2010).

2.2 A brief review on biological effects of low level therapeutic lasers

Low level laser (or light) therapy (LLLT) is the application of light (usually a low power laser or LED in the range of 1mW – 500mW) to a pathology to promote wound healing and tissue repair, reduce inflammation and relieve pain. The light is typically of narrow spectral width in the red or near infrared spectrum (600nm – 1000nm); at power densities (between 1mw-5W/cm2) (Huang, Chen et al. 2009), not associated with macroscopic thermal effects, in contrast to thermally mediated surgical applications (Chow, David et al. 2007). In using high power surgical lasers, the collimation of laser light leads to the emission of a narrow, intense beam of light and is used for precise tissue destruction (photothermal effect). However, in LLLT, light radiation intensities are so low that the resulting biological effects are ascribable to physical or chemical changes associated with the interaction of cells and tissues with the laser radiation, and not simply to a result of heating (Snyder, Byrnes et al. 2002; Gigo-Benato, Geuna et al. 2005).

The main areas of medicine where laser phototherapy has a known and major role are as follows: promoting wound healing, tissue repair and prevention of tissue death, relief of inflammation in chronic diseases and injuries with its associated pain and edema, relief of neurogenic pain and some neurological problems (Hamblin, Waynant et al. 2006).

The first law of photobiology states that for low power visible light to have any effect on a living biological system, the photons must be absorbed by electronic absorption bands belonging to some molecular photoacceptors, or chromophores (Sutherland 2002; Huang, Chen et al. 2009). Red and near infrared light is absorbed by photoreceptors contained in the protein components of the respiratory chain located in mitochondria, in particular cytochrome c oxidase and flavoproteins like NADH-dehydrogenase. This can lead to a short time activation of respiratory chain and oxidation of NADH pool leading to changes in the redox state of both mitochondria and cytoplasm, leading to increased ATP production, and biological responses at the cellular level through cascades of biochemical reactions (Karu 1989; Karu, Pyatibrat et al. 2004; Karu and Kolyakov 2005). These effects in turn lead to increased cell proliferation and migration, modulation in levels of cytokines, growth factors, inflammatory mediators, and increased tissue oxygenation. The results of these biochemical and cellular changes in animals and patients lead to valuable biological effects such as promoting wound healing and tissue repair, relief of inflammation, pain reduction, and amelioration of damage after heart attacks, stroke, nerve injury and even retinal toxicity (Hamblin, Waynant et al. 2006).

2.3 Pain relieving effects of low level therapeutic lasers

Low-level laser therapy (LLLT) is increasingly recognized as an appropriate option for pain relief. In fact, it is for this indication that biostimulative lasers have been approved for marketing by the U.S. Food and Drug Administration through the premarket notification/510(k) (Gigo-Benato, Geuna et al. 2005). Many studies have demonstrated the efficacy of phototherapy in various pain syndromes (Tuner and Hode 2010). Responding to the increasing levels of evidence, the World Health Organization's Committee of the Decay

of the Bone and Joint has also recently incorporated LLLT into guidelines for treatment of neck pain (Haldeman, Carroll et al. 2008; Chow, Armati et al. 2011).

> *"Efficacy of LLLT in painful clinical conditions has been established by several recent systematic reviews and meta-analyses [level 1 evidence, according to the Australian Government, NHMRC (1999)] (Chow, David et al. 2007). This level of evidence relates to chronic neck pain(Chow and Barnsley 2005), tendinitis (Bjordal, Couppe et al. 2001), chronic joint disorders (Bjordal, Couppe et al. 2003), and chronic pain (Enwemeka, Parker et al. 2004) . Randomized controlled trials (RCTs) provide level II evidence for the efficacy of laser therapy in chronic low back pain (Umegaki, Tanaka et al. 1989; Soriano and Rios 1998; Basford, Sheffield et al. 1999) . In other reviews of laser therapy for painful conditions such as rheumatoid arthritis (Brosseau, Robinson et al. 2005) and musculoskeletal pain (Gam, Thorsen et al. 1993; De Bie, De. Vet et al. 1998), the evidence is equivocal. Such variability in outcomes may be due to the multiplicity of parameters used, including wavelengths, energy, and power densities, with differing frequencies of application(Chow and Barnsley 2005)."* ***(Chow, David et al. 2007)***

2.4 Mechanisms of analgesic effects of low level laser therapy

The basic biological mechanisms behind the analgesic effects of conventional LLLT are not completely understood. Some of the explanations for these pain relieving effects are as follows:

- Reversible blockage of action potential generation of nociceptive signals in primary afferent neurons and specific reversible inhibition and functional impairment of Aδ and C fibers, which transmit nociceptive stimuli (Wakabayashi, Hamba et al. 1993; Kasai, Kono et al. 1996; Orchardson, Peacock et al. 1997; Chow, Armati et al. 2011).
- Increase in β-endorphin synthesis and release (Labajos 1988; Montesinos 1988; Hagiwara, Iwasaka et al. 2007).
- Inhibiting cyclooxygenase, interrupting conversion of arachidonic acid in to prostaglandins, especially prostaglandin E2 (PGE2) (Shimizu, Yamaguchi et al. 1995; Mizutani, Musya et al. 2004).
- Suppression of Substance P, a neuropeptide associated with nociception (Ohno 1997).
- Suppression of bradykinin activity, a pro-inflammatory neuropeptide that irritates nociceptors and is a key element in clinical pain and the associated inflammation (Maeda 1989; Jimbo, Noda et al. 1998)
- Increased production of serotonin, affecting negatively neurotransmission (Tuner and Hode 2010)
- Increased synaptic activity of acetylcholine esterase (Simunovic 2000)
- Involvement of nitric oxide in analgesic effects of therapeutic lasers (Mrowiec 1997)
- Single oxygen production, which in small amounts, is very important in biochemical processes and may be important in biostimulation
- Decreased inflammation and subsequently decreased inflammatory sensitization of small-diameter afferent nerve endings
- Improvement of local microcirculation, increased tissue oxygenation, shift of metabolism from anaerobic to aerobic pathways, decreased production of acidic metabolites which stimulate the pain receptors
- Increased lymphatic flow and consequently reducing edema, which leads to decreased sensitization of pain receptors

- Systemic effect, some researchers propose that laser phototherapy has both a local effect in the area treated by laser light, and a systemic effect through the release of metabolites (Tuner and Hode 2010).

3. Pain relieving effects of carbon dioxide lasers

Laser phototherapy is a relatively new application of carbon dioxide lasers, in spite of the fact that papers on the subject were published as early as the mid-eighties (Tuner and Hode 2010). CO_2 laser biostimulative and pain relieving effects can be assessed in the following two main groups:

- The lower post operative pain following CO_2 laser surgery compared to conventional surgery, which is attributed to the simultaneous low level laser therapy (photobiomodulation) effects of high power CO_2 laser irradiation (Tuner and Hode 2010).
- Application of CO_2 laser, as a phototherapeutic laser. "For using CO_2 laser as a low power therapeutic laser, one needs to make the beam wide enough not to burn. Another option is to scan rapidly over the lesion with a narrow beam. Therefore the power density or average power is kept low enough to avoid burning and their incident energy and power density are set within the low intensity biostimulative laser therapy range" (Tuner and Hode 2010). NACLT is a new technique, in which CO_2 laser is used in non-thermal, non-ablative manner as a biomodulative, low intensity laser for immediate and significant pain relief. (Zand, Ataie-Fashtami et al. 2009; Zand, Mansouri et al. 2009; Zand, Najafi et al. 2010).

This section is organized as follows. At first some clinical studies which demonstrate the pain relieving effects of high power surgical CO_2 lasers are reviewed, and then low power biostimulative CO_2 laser studies are briefly reviewed. In the next section, NACLT as a new low power, biomodulative laser therapy protocol for pain reduction is introduced.

3.1 Pain relieving effects of carbon dioxide laser as a high power surgical laser

Some investigators who used the classical thermal effects of CO_2 laser (vaporization, cutting and coagulation) reported less post-operative pain following CO_2 laser surgery, and a reduced requirement for post-operative analgesics (Duncavage and Ossoff 1986; Colvard and Kuo 1991; Demidov, Rykov et al. 1992; Chia, Darzi et al. 1995; Kaplan, Kott et al. 1996; Andre 2003).

Kaplan, one of the pioneers of CO_2 laser surgery, attributed the excellent healing and lower post operative pain experienced with CO_2 laser surgery compared to conventional surgery to the simultaneous laser therapy effects of CO_2 laser irradiation. Kaplan stated that laser surgery and laser therapy should be regarded *as two sides of the same coin* (Kaplan, Kott et al. 1996; Tuner and Hode 2010).

Duncavage reported that the advantages of the CO_2 laser surgery included homeostasis, precise visualization, and less edema and pain than the conventional techniques (Duncavage and Ossoff 1986).

Colvard and Kuo used high-power, ablative CO_2 laser at a power output of 4 W under local anesthesia for painful minor aphthous ulcers of 14 patients. In all, 88.8% of the patients in the study were completely pain free following anesthetic resolution, and none of the patients required post-operative medication for pain relief. (Colvard and Kuo 1991).

Demidov and his colleagues used high-energy CO_2 laser as a laser scalpel in 120 cases of breast surgery including 70 operations for cancer. They reported reduced pain sensitivity in the region of the wounds in the postoperative period (Demidov, Rykov et al. 1992).

In another study Chia reported that high-power CO_2 laser haemorrhoidectomy was associated with a reduced requirement for post-operative analgesia (Chia, Darzi et al. 1995).

Andre used classical high power CO_2 laser at 10–15 W in continuous mode under local anesthesia for treating ingrowing nails of 302 patients. He reported that ingrowing nails were easily operated with the CO_2 laser; bleeding was minimal, infection was rare, the wounds healed without exhudative drainage and cosmetic results were good. In addition the immediate post-operative pain was less severe than after classical nail surgery with scalpel (Andre 2003).

In another study, Tada et al. compared the clinical effects and postoperative course of the scanning CO_2 laser surgery and conventional surgical method to evaluate the clinical effectiveness of the former for the treatment of ingrowing nail deformities. They demonstrated that statistically, the operating time and the duration of postoperative pain were reduced significantly by the scanning CO_2 laser. Furthermore, patients treated with CO_2 laser were able to return to daily life significantly sooner (Tada, Hatoko et al. 2004).

Kaviani et al. investigated whether the CO_2 laser surgery was superior to conventional surgical techniques for minor breast surgery in a randomized clinical trial. They demonstrated that application of the CO_2 laser in breast mass biopsy had some advantages, including a lower requirement for local anesthetic and a lower rate of intraoperative bleeding; however it did not reduce the postoperative pain grade severity (Kaviani, Fateh et al. 2008).

Demetriades used ablative CO_2 laser in painful oral aphthous ulcer of a patient with Behçet's Syndrome. His experience showed transient pain relief following ablative CO_2 laser irradiation (Demetriades, Hansford et al. 2009).

3.2 Pain relieving effects of carbon dioxide laser as a low level (therapeutic) laser

It is interesting to know that in addition to classical low level therapeutic lasers, surgical lasers could also be used as therapeutic instruments, for example; defocused CO_2 laser 10,600 nm, defocused ruby laser 694 nm and defocused Nd:YAG laser 1064 nm can be used for photobiomodulation. "When high power laser are used for biomodulation, one only needs to make the beam wide enough not to burn. The patient will then feel only a mild heat. An alternative is to scan rapidly over the lesion with a narrow beam. Therefore the power density or average power is kept low enough to avoid burning and their incident energy and power density are set within the low intensity laser therapy range" (Tuner and Hode 2010).

The famous investigation of Mester with a low powered ruby laser (694-nm) on the shaved areas of the mice and quickly growing back of hairs can be an example of using a surgical laser as a therapeutic, biostimulative one (please see 2.1. History).

At Uppsala Academic Hospital, a CO_2-laser was tested successfully for biostimulative treatment of epicondylitis. This method was called EDL (Emitted Defocused Laser-light). It should be noted, however, that CO_2-lasers were not used as surgical lasers in this study;

their incident energy and power density were set within the laser therapy range by spreading out the beam over such a large surface that the laser did not cause burning (Tuner and Hode 2010).

Nicola used CO_2 low power laser treating chronic pharyngitis. 85 patients with non-specific chronic pharyngitis were elected to be treated: Group I, 40 patients with predominance of hyperaemic aspect; and group II, 45 patients, predominance of hypertrophied aspect. Both groups were treated for 8 to 10 sessions. They concluded that this method was very suitable for the treatment of chronic pharyngitis (Nicola and Nicola 1994).

In another study, 846 patients with different types of fibromyositic rheumatisms were submitted to defocalized laser therapy from 1980 to 1995. They employed Diodes and CO_2 lasers. Control groups were used to compare results with those of traditional methods. Results were evaluated on the basis of subjective (such as local pain) and objective criteria. On the whole, results were positive in comparison with other methods both as regards recovery time and persistence of results. Approximately 2/3 of the patients benefited from the treatment indicated that there were greater advantages in use of laser over other presently available methods. Longo and his collogues recommended that standardalization of treatment protocols deserves further studies (Longo, Simunovic et al. 1997).

The CO_2-laser can also be used as an acupuncture tool. Simulation of acupuncture points has been carried out both with biostimulating power densities (e.g.100mW/cm^2) and burning/coagulation/ evaporation power densities. Some clinics state that CO_2 lasers give better results on acupuncture points than HeNe lasers. "As the CO_2 laser's beam cannot penetrate more than around 0.5 mm into tissue, the effects must be due to the influence of the laser energy on the cells encountered, so that signal substances are released and then circulate in the organism. This indirectly confirms the hypothesis that conventional laser therapy has both a local effect in the area treated by laser light, and a systemic effect through the release of metabolites. It is well known that these kinds of secondary effect also occur at the traditional wavelengths of 633, 830, and 904 nm" (Tuner and Hode 2010).

4. NACLT (Non-Ablative CO_2 Laser Therapy)

Recently, there have been few reports about using CO_2 laser in non-ablative manner to reduce pain in painful mucosal lesions (Elad, Or et al. 2003; Sharon-Buller and Sela 2004; Zand, Ataie-Fashtami et al. 2009; Zand, Mansouri et al. 2009; Zand, Najafi et al. 2010).

In a report, Elad et al suggested that CO_2 laser treatment could be of benefit to control pain in severe oral chronic graft-versus-host-disease (Elad, Or et al. 2003). In this study, the oral lesions of four patients were irradiated by CO_2 laser during 17 sessions. The CO_2 laser was applied, over mucosal lesions, using 1W power for 2-3s/1mm^2. The treated mucosa was kept wet during the process. The treatment was pain free, and anesthesia was not required. The average VAS scores before, during, and immediately after CO_2 laser treatment were 8.09, 3.47, and 4.88 respectively. There was no visual damage to the oral mucosa and no aggravation of the oral lesions (Elad, Or et al. 2003).

In another case report, aphthous ulcers of two patients were irradiated with CO_2 laser at 1.0-1.5 W power, with a defocused hand piece for 5 seconds. Before laser irradiation, a thin film

of Elmex Gel (a transparent gel with high water content) was placed on the lesions to reduce the beam absorption by the soft tissue. Anesthesia was not required since the treatment was not painful. The patients reported immediate pain relief after laser irradiation (Sharon-Buller and Sela 2004).

In these two reports, water (Elad, Or et al. 2003) and transparent gel with high water content (Sharon-Buller and Sela 2004) were used to reduce the beam absorption by the soft tissue.

These interesting results encouraged us to conduct a randomized controlled clinical trial to confirm the pain-relieving effect of CO_2 laser in minor aphthous ulcers as a prototype of painful oral lesions (Zand, Ataie-Fashtami et al. 2009). The results of this clinical trial demonstrated that a single session of low-intensity, non-thermal, CO_2 laser irradiation could reduce pain in minor aphthous ulcers immediately and significantly, with no visible side effects (Zand, Ataie-Fashtami et al. 2009), the technique was called NACLT (Non-Ablative CO_2 Laser Therapy) afterwards.

In order to use the CO_2 laser as a phototherapeutic laser for NACLT, the CO_2 laser beam is irradiated through a thick layer of transparent gel with high water content to reduce the beam absorption by the tissue. In addition, the CO_2 laser is operated with a de-focused hand piece 5–6 mm distant from the mucosal surface, scanning rapidly over the lesion with circular motion (Zand, Ataie-Fashtami et al. 2009; Zand, Mansouri et al. 2009; Zand, Najafi et al. 2010). The results of the powermetry have shown that the final laser power output, after passing through the gel, is reduced to 2-5 mW, which is in the range of low power lasers. Thermometry has also shown no significant temperature rise on the surface of the ulcers and under the gel layer during the laser irradiation, supporting the low power nature of the applied CO_2 laser in NACLT(Zand, Ataie-Fashtami et al. 2009). It appears that due to high water content of the gel, it absorbs CO_2 laser irradiation considerably, resulting in significant drops in the power output, by a factor of 200-500. In fact by irradiation of CO_2 laser through a transparent gel with high water content, CO_2 laser can be used as a non-destructive, non-thermal laser to reduce pain in some oral lesions immediately and significantly. This technique was called non-ablative CO_2 laser therapy (NACLT), in order to avoid any confusion with classical high power thermal CO_2 laser effects. This technique could also be called non-thermal CO_2 laser therapy (NTCLT) to avoid misinterpretation with fractional non-ablative lasers used for skin rejuvenation (Zand, Ataie-Fashtami et al. 2009).

NACLT is a pain free procedure and neither systemic nor topical anesthesia is required. The patients don't feel warmth in their lesions during the procedure, in contrast to conventional defocused CO_2 laser therapy in which the patients feel mild warmth. Up to now, in the series of studies about the analgesic effects of NACLT, we have observed no visual effects of thermal damage to the oral mucosa such as tissue ablation, ulceration, erythema or aggravation of the lesions following the careful application of the technique (Zand, Ataie-Fashtami et al. 2009; Zand, Mansouri et al. 2009; Zand, Najafi et al. 2010). So that the before-after NACLT pictures of the lesions cannot be differentiated from each other easily (Figure 1). Since there is no tissue ablation and plume formation during NACLT, in contrast to the classical ablative CO_2 laser surgery, it seems rational to conclude that this procedure has no potential for carrying viral particles to the surgeon and other operating room staff (Zand, Ataie-Fashtami et al. 2009).

There are some differences between analgesic effects of NACLT and the other classical low power lasers. The analgesic effect in LLLT is usually gradual, cumulative, and multi-session (Pinheiro, Cavalcanti et al. 1998; Gur, Karakoc et al. 2002; Gur, Sarac et al. 2004; Nes and Posso 2005; Chow, Heller et al. 2006; Djavid, Mehrdad et al. 2007; Bjordal, Bensadoun et al. 2011; Iwatsuki, Yoshimine et al. 2011; Ribeiro, de Aguiar et al. 2011). In contrast, the pain relieving effect of NACLT is immediate, dramatic and more sustained than conventional phototherapeutic lasers, so that immediately after NACLT, the patients of the studies have been able to eat and drink easily (Zand, Ataie-Fashtami et al. 2009; Zand, Mansouri et al. 2009; Zand, Najafi et al. 2010).

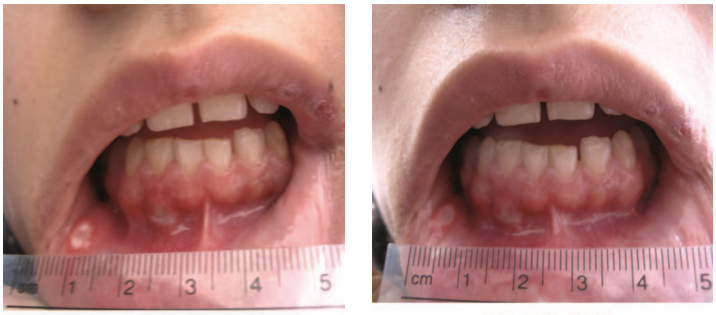


Fig. 1. Minor aphthous ulcers before and immediately after NACLT

4.1 NACLT protocol

At first, all standard precautions of laser utilization should be considered. Before laser irradiation, the patient and surgical staff should be given appropriate protective eye shields and eye glasses matched to the laser wavelength (10,600 nm) to protect inadvertent laser impact.[1] Before laser irradiation, a layer of a transparent, non-anesthetic gel with high water content is placed on the lesion. In our studies, we use a transparent gel (Abzar Darman Co., Iran) with 87.5% water content, with a thickness of 3–4 mm on the lesion, is used. The CO_2 laser is operated at 1W power, with a de-focused hand piece, 5–6 mm distant from the mucosal surface, in continuous mode, scanning rapidly over the lesion with a circular motion. The irradiation time depends on the size of the lesion. For example, in our studies the irradiation time for minor aphthous ulcers is about 5-10 seconds. When using NACLT for larger lesions, such as; pemphigus vulgaris, every one centimeter square of the lesion has been irradiated for 5 seconds in each pass, and repeated immediately if the contact pain of the lesion persists. Between the passes, the prior gel should be wiped gently and replaced by a new layer of gel, otherwise the water content of the gel will decrease which may lead to increasing the beam absorption by the lesion and subsequent tissue injury and even burning.

[1] In NACLT studies, we used eye glasses matched to the CO_2 laser wavelength (10,600 nm), but we presume that it might be safer to use eye glasses matched to both the 10,600 nm and the guiding beam to protect the eyes from the reflected beam from the surface of the gel, the presumption that should be evaluated in further studies.

4.2 NACLT applications in clinical studies

4.2.1 Recurrent oral aphthous stomatitis

4.2.1.1 Definition

Recurrent aphthous stomatitis (RAS) is a common oral disorder of uncertain etiopathogenesis (Scully, Gorsky et al. 2003), characterized by painful, round or ovoid ulcers with circumscribed margins, yellowish fibrinoid base, surrounded with erythematous haloes. The lesions typically first presenting in childhood or adolescence, recur at varying intervals throughout life (Jurge, Kuffer et al. 2006). The frequency and severity of the ulcerations usually decreases with age (Arikan, Birol et al. 2006). RAS occurs worldwide although it appears most common in the developed world (Jurge, Kuffer et al. 2006).

Recurrent aphthous stomatitis (RAS) is classified into three clinical forms, namely minor (miRAS), major (maRAS) and herpetiformis. Minor aphthous ulcers, which comprise over 80–90% of cases (Shashy and Ridley 2000), are less than 1 cm in diameter, last up to 7–14 days, and they heal without scar formation. Major aphthous ulcers are over 1 cm in diameter, and their healing may take 20 to 30 days at a time and often heal with scarring. Herpetiform ulcers (HUs) are multiple, clustered, 1–3 mm lesions that may coalesce into larger ulcers. They typically heal within 15 days (Prolo P 2006).

Although the exact underlying pathophysiology of RAS is not completely known, some evidences propose that aphthous ulcers are related to a focal immune dysfunction in which T lymphocytes have a significant role (Shashy and Ridley 2000; Jurge, Kuffer et al. 2006). Many etiologic, predisposing, and precipitating factors, such as genetic factors, immunologic problems, trauma, hypersensitivity to foods and drugs, hormonal changes, hematological deficiencies, cessation of smoking, and psychological stresses have been propsed (Shashy and Ridley 2000; Arikan, Birol et al. 2006).

Since there is no consensus regarding the cause of recurrent oral aphthous ulcers, it is difficult to have completely effective treatment or prevention (Shashy and Ridley 2000). There are currently few agents that have been found in randomized controlled clinical trials to cure aphthous ulcers (Jurge, Kuffer et al. 2006). As a result, the management of RAS is directed largely toward symptomatic relief. The main problem with aphthous ulcers is their pain which may be so severe. Many different therapeutic agents, including topical corticosteroids, mouth rinses, antibiotics, local anesthetic gels or pastilles, and treatment modalities, such as silver nitrate cautery and cryotherapy, have been tried for pain control in miRAS patients (Alidaee, Taheri et al. 2005; Arikan, Birol et al. 2006).

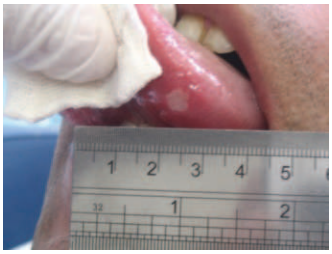


Fig. 2. Minor aphthous ulcer

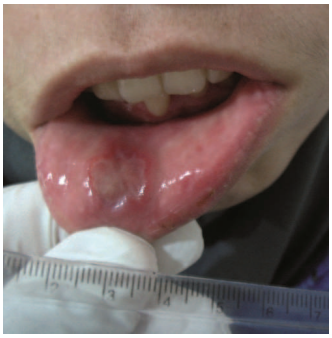


Fig. 3. Major aphthous ulcer

4.2.1.2 NACLT and minor oral aphthous stomatitis

A randomized controlled clinical trial was designed to evaluate the pain relieving effects of a single-session of NACLT in minor recurrent aphthous stomatitis as a prototype of painful oral ulcers. Fifteen patients, each with two discrete aphthous ulcers, were included. One of the ulcers was randomly allocated to be treated with NACLT and the other one served as a placebo. In each patient, the laser lesion was treated with NACLT, while the placebo lesion was irradiated with the same laser, but with an inactive probe. The patients scored and recorded the pain severity of their lesions on a 10-grade visual analogue scale (VAS) up to 4 days post operatively. In the laser group, the pain severity scores of the lesions were dramatically declined immediately after irradiation ($p<0.001$), whereas there were no changes in the mean scores in the placebo lesions at the same time. The reduction in pain scores was significantly greater in the laser group than in the placebo group in all of the follow up periods ($p<0.001$). The procedure itself was not painful, so anesthesia was not required. The patients reported no warmth in their lesions during laser treatment. There was no visual effect of thermal damage to the oral mucosa such as ablation, coagulation or erythema. The results showed that a single-session of NACLT reduced pain in minor aphthous ulcers immediately and significantly, without any visible side effects (Zand, Ataie-Fashtami et al. 2009).

4.2.1.3 NACLT and major oral aphthous stomatitis

A pilot randomized controlled clinical trial was designed to evaluate the analgesic effects of a single-session of NACLT in major recurrent aphthous ulcers. Five patients, each with two discrete major aphthous ulcers were included. One of the ulcers was randomly allocated to be treated with NACLT and the other one served as a placebo. The lesions in laser group were irradiated with CO_2 laser (λ = 10,600 nm; Lancet-2, Russia) through a thick layer of transparent, non-anesthetic gel (Abzar Darman Co., Iran) with 87.5% water content, with a thickness of 3–4 mm. The CO_2 laser was operated at 1W power, with a de-focused hand piece, 5–6 mm distant from the mucosal surface, in continuous mode, scanning rapidly over the lesion with circular motion. The patients' idiopathic (non-contact) and contact pain severity scores were recorded before and immediately after NACLT. These scores were also recorded up to 4 days post- operatively. The results of the study demonstrated that in the laser group, both the non-contact and contact pain severity scores of the lesions were dramatically declined immediately after irradiation ($p<0.001$), whereas there were no

changes in the mean scores in the placebo lesions at the same time. The reduction in pain scores was significantly greater in the laser group than in the placebo group in all of the follow up periods ($p<0.001$). There were not any visible side effects following NACLT. None of the patients reported warmth feeling in their lesions during laser treatment. The results of the study suggested that a single-session of NACLT could reduce pain in major aphthous ulcers immediately and significantly, without any visible side effects (Zand, Ataie-Fashtami et al. 2009). This study is in progress.

4.2.1.4 Literature review

Colvard and Kuo evaluated the potential efficacy of the high-power, surgical CO_2 laser for pain relief in 28 painful minor aphthous ulcers of 14 patients. Their anesthetic protocol included pre-operative pain medication (oral administration of ketoprofen) and local anesthesia by infiltration of 1:200,000 2% isocaine with 1:200000 neocobefrin to overcome the painful nature of the procedure. During the procedure, CO_2 laser was used as a classical, ablative manner with power output 4 W and as much necrotic tissue as possible was removed. Over all 88.8% of the patients were completely pain free following anesthetic resolution, and none of the patients required post-operative medication for pain relief. The authors concluded that CO_2 laser should be included as an alternative modality for the treatment of miRAS, due to its ability to reduce or eliminate pain (Colvard and Kuo 1991). In this study, CO_2 laser was used in classical, high power ablative manner. However the post operative analgesic effects of the procedure demonstrated the simultaneous biomodulative effects of CO_2 laser irradiation. The same concept Kaplan stated that laser surgery and low level laser therapy should be regarded as two sides of the same coin.

Fekrazad et al. evaluated the effects of Nd: YAG laser (power: 3 W, energy: 100 mj, pulse repetition rate: 30Hz, irradiation time: 60 sec) in 138 patients with aphtous ulcers. The patients were randomly assigned into three groups, as follows: (1) treatment with a focalized beam; (2) treatment with a non-focalised beam and (3) placebo treatment. In group (1) the laser beam was administered from a distance of 6 mm from the centre of the ulcer without using a clear and defined point of irradiation. In group (2) a well defined point beam of the laser was irradiated from a distance of 2 mm from the center of the ulcer, in a helical fashion. In group (3) the HeNe Laser was used as placebo with inactive probe. In group (1) and (2) a significant reduction of pain was observed compared to group (3). The duration of pain and the duration of recovery period were shortest in group (2) (Fekrazad, Jafari et al. 2006).

De Souza TO et al. assessed the effect of low-level laser therapy on pain control and the repair of recurrent aphthous stomatitis. Twenty patients with recurrent aphthous ulcers were divided into two groups. The patients in Group I (n = 5) treated with topical triamcinolone acetonide and the patients in Group II (n = 15) received laser treatment with an InGaA1P diode laser (670 nm, 50 mW, 3 J/cm^2 per point) in daily sessions on consecutive days. All patients were assessed daily, and the following clinical parameters were determined during each session: pain intensity before and after treatment and clinical measurement of lesion size. The results showed that 75% of the patients reported a reduction in pain in the same session after laser treatment, and total regression of the lesion occurred after 4 days. Total regression in the corticosteroid group was from 5 to 7 days. They concluded that LLLT with these laser parameters demonstrated analgesic and healing effects with regard to recurrent aphthous stomatitis (De Souza, Martins et al. 2010).

Title	Author	Study	Type of irradiation	Need to Anesthesia	Number of patients
Managing aphthous ulcers: Laser Treatment Applied	Colvard M, Kuo P	Before-after clinical trial 1991	Ablative CO_2 laser surgery	+	18
CO_2-laser treatment of ulcerative lesions	Sharon-Buller A, et al.	Case report 2004	CO_2 laser irradiation of the lesions through a thin film of transparent gel with high water content	–	2
Relieving pain in minor aphthous stomatitis by a single session of non-thermal carbon dioxide laser irradiation	Zand N, Ataie-fashtami L. et al.	Randomized controlled clinical trial (RCT) 2008	CO_2 laser Irradiation of the lesions through a thick (3-4mm) layer of transparent gel with high water content; (NACLT)	_	15
Analgesic effects of single session of Non-Ablative CO_2 Laser Therapy (NACLT) in major aphthous ulcers: (a preliminary study)	Zand N, Ataie-fashtami L. et al.	Randomized controlled clinical trial (RCT) 2009	CO_2 laser Irradiation of the lesions through a thick (3-4mm) layer of transparent gel with high water content; (NACLT)	-	5

Table 1. Irradiation of aphthous ulcers with CO_2 laser

4.2.2 Behçet's disease

4.2.2.1 Definition

Behçet's disease (BD) which is classified among vasculitides is a complex, multisystem inflammatory disease characterized by oral and genital aphthae, cutaneous lesions, arthritis, ocular, gastrointestinal, and neurologic manifestations (Meador, Ehrlich et al. 2002; Suzuki Kurokawa and Suzuki 2004; Lin and Liang 2006).

The most common clinical feature is the presence of recurrent and usually painful mucocutaneous ulcers (Lin and Liang 2006). Oral aphthosis is the most frequent and

constant manifestation of Behçet's disease (Davatchi, Shahram et al. 2005) and usually the initial presenting symptom in most, if not all, patients (Lin and Liang 2006) . The distinct difference between the clinical features of aphthous ulcers of RAS and Behçet's Syndrome remains unclear. The aphthous ulcers of Behçet's disease are typically painful punched-out ulcers with a white yellowish fibrinoid base, surrounded with erythematous halo. They range in size from a few millimeters to 2 cm. These ulcers typically heal spontaneously within 1 to 3 weeks,usually without scarring (Ghate and Jorizzo 1999; Lin and Liang 2006).

Genital ulceration occurs in approximately 75% of the patients with Behçet's disease (Lin and Liang 2006). The genital aphthous ulcers are morphologically similar to the oral ulcers, except that lesions are usually larger, more painful, heal more slowly, recur less frequently and can have scarring tendency (Davatchi, Shahram et al. 2005; Lin and Liang 2006). In females they are often larger than 10 mm, and deeper than oral lesions. They are localized on the vulva, vagina, and rarely cervix. The giant aphthous lesion of the vulva is frequent, causing dysfunction and leaving sometimes indelible cicatrix. In males, genital aphthosis is often seen on the scrotum, but may be seen also on the shaft of penis or on the meatus. Sometimes they become giant lesions (Davatchi, Shahram et al. 2005) . Genital ulcerations of Behçet's disease may be very painful, exert a negative impact on the patient's quality of life, and these lesions are often refractory to multiple treatments(Kasugai, Watanabe et al. 2010).

Treatment of Behçet's disease is based on the clinical symptoms and severity of systemic involvement, including topical therapies as well as colchicine, dapsone, thalidomide, and immunosuppressants, interferon-alpha/beta, anti-tumor necrosis factor antibody, the latter specially in treatment for the cases with poor prognosis including eye, intestine, vessel and central nervous system involvement (Suzuki Kurokawa and Suzuki 2004).

The mucosal lesions, especially genital lesions can often become refractory to multiple treatments and present challenges to physicians. Topical or intralesional corticosteroids, oral pentoxifylline, sucralfate, dapsone, colchicine, and systemic low-dose corticosteroids, used either alone or in combination, are safe and having varying evidence for effect in mild to moderate mucocutaneous disease. Azathioprine or methotrexate can be used if the lesions are refractory to the previously mentioned therapies. Tumor necrosis factor (TNF) inhibitors such as infliximab or etanercept should be considered as the next step treatments . Tacrolimus, cyclosporine, and interferon-alpha-2a should be used generally only if TNF inhibitors have failed as a result of their toxicities (Lin and Liang 2006).

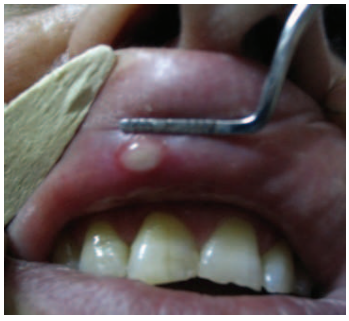


Fig. 4. Behçet's Disease

4.2.2.2 NACLT and oral aphthous ulcers of Behçet's disease

A pilot before-after clinical trial was designed to evaluate the analgesic effects of a single-session of NACLT in painful aphthous ulcers of Behçet's disease. Up until the time of this publication,, three patients with known Behcet's disease have been eligible and consented to participate in the study according the inclusion/exclusion criteria.

Four painful oral aphthous ulcers of the three patients were treated by NACLT. The pain severity of the lesions were dramatically declined immediately after irradiation ($p<0.001$). This analgesic effect was consistently sustained during the five days follow-up periods. Up until the time of this publication, the results of this pilot study suggest that a single session of NACLT could relieve pain in oral aphthous ulcers of Behçet's disease immediately and significantly without visible side effects of thermal damage or aggravation of the lesions. Similar to the other NACLT studies, the procedure itself was pain free and no anesthesia was required. The study is in progress.

4.2.2.3 NACLT and genital aphthous ulcers of Behçet's disease

In another case report that is being published, the extremely painful genital aphthous ulcers of a 23-year-old female with Behcet's disease were irradiated by NACLT. Before laser irradiation the pain of the lesions was so severe which impeded daily functions, such as sitting, walking, and even sleeping and did not respond to conventional analgesics. The non-contact and contact visual analogue scale (VAS) pain scores of the left genital ulcer were 8 and 10 and the scores of the right sided ulcer were 6 and 10 respectively. Immediately after NACLT of the ulcers and its surrounding erythematous rim, the contact pain of the lesions relieved completely (so that she could even walk downstairs without difficulties). Similar to the other prior NACLT investigations, there were no visual side effects of thermal damage to the lesions, such as tissue ablation or aggravation of the lesions following NACLT. The procedure was painless and neither systemic nor local anesthesia was required. This analgesic effect of NACLT was sustained during the healing period and she experienced no problem in daily functions and did not require topical or systemic analgesics. She just had mild burning sensation during urination for the first three days which relieved after healing of the lesions. It should be noted that concomitantly, treatment with prednisolone 30mg/day and colchicine 2mg/ day was initiated in the hospital. Additionally, the depth of the genital ulcers decreased remarkably two days after NACLT. The ulcers healed completely within 11 days which seemed to be much shorter than what was expected. Interestingly in spite of the large size of the ulcers, they left a very small (6mm) scar. The results of this case report suggest that NACLT could be potentially considered as an alternative method for pain relief in painful genital aphthous ulcers of Behçet's disease without any complications.

It should be noticed that Behçet's syndrome is a serious multisystem disease, which in some cases it may lead to systemic complications such as; severe ocular problems (even blindness), intestinal, central nervous system,… involvement. Therefore the patients must be warned that NACLT should not substitute the systemic therapy of the disease in spite of its significant pain relieving effect.

Certainly, controlled clinical trials with larger sample sizes are necessary to further evaluate the efficacy and safety of NACLT in reducing the pain of oral and genital aphthous ulcers of

Behcet's disease. In addition such studies can demonstrate whether NACLT could accelerate wound healing in these lesions and specially prevent scar formation in genital aphthous ulcers of Behcet's disease or not.

4.2.2.4 Literature review

Demetriades used ablative CO_2 laser in four painful oral aphthous ulcers of a patient with Behçet's Syndrome. Before laser irradiation, the lesions were infiltrated with a minimal amount of lidocaine 2% with 1:100,000 epinephrine. A CO_2-laser set at 2W superpulse mode with a 0.4 mm ceramic tip was used, in a defocused way to lightly char the surface of the ulcers. The patient tolerated the procedure well. On subsequent follow-up, one week after the procedure, the patient reported considerable relief of symptoms on most of the treated ulcers. The oropharyngeal ulcer displayed only moderate response, but the patient reported an overall improvement of his quality of life (Demetriades, Hansford et al. 2009).

Title	Author	Study	Type of irradiation	Need to Anesthesia
General manifestations of Behçet's syndrome and the success of CO_2-laser as treatment for oral lesions: A review of the literature and case presentation	Demetriades M et al	Case report 2009	Ablative defocused CO_2-laser irradiation	+
Relieving pain in painful genital ulcers of Behcet's disease by a single session of non thermal, Non-Ablative CO_2 Laser Therapy(NACLT): A Case Report	Zand N, Fateh M. Et al.	Case report, under publish	NACLT	–
Immediate pain relief of oral aphthous ulcers of Behcet's disease by non-thermal, Non-Ablative CO_2 Laser Therapy (NACLT)	Zand N, Fateh M. Et al.	Pilot before-after clinical trial/ under publish	NACLT	-

Table 2. Irradiation of aphthous ulcers of Behcet's disease with CO_2 laser

4.2.3 Pemphigus vulgaris

4.2.3.1 Definition

Pemphigus Vulgaris (PV) is a rare, potentially life-threatening, autoimmune blistering disease of the skin and mucous membranes. Although the disease can affect anyone, it is most prevalent in people of Mediterranean or Jewish ancestry(Bystryn and Rudolph 2005). The prevalence of the disease is 30/100,000 and annual incidence has been reported between 1 and 5 in 100,000 according to different studies in Iran (Chams-Davatchi, Valikhani et al. 2005; Asilian, Yoosefi et al. 2006). The lesions are characterized by intra-epidermal vesicles with acantholysis and an intact basal layer. In the majority of patients, painful mucous membrane erosions are the presenting sign of pemphigus vulgaris and may be the only sign for weeks to months before any bullous skin lesion develops. The mucous membranes most often affected are those of the oral cavity, in which intact blisters are rare, probably because they are fragile and break easily, leaving scattered and often extensive erosions. The lesions are usually multiple, superficial, and irregular in shape, and arise from mucosa of healthy appearance. Although any surface can be involved, the most common sites are the buccal and labial mucosa, the palate, and the tongue (Bystryn and Rudolph 2005).

Oral lesions in pemphigus vulgaris may be so painful during the active period of the disease that may interfere with their eating, drinking and even speaking (Black, Mignogna et al. 2005; Bystryn and Rudolph 2005).

High doses of systemic corticosteroids plus immunosuppressive agents have dramatically declined the mortality rate of the disease. Understandably, owing to the life threatening nature of PV, the main focus of the peer reviewed literature has been on suppression and remission of PV (Rashid and Candido 2008). However, remission is not instantaneous and takes time to achieve. This delay in remission allows ample opportunity for complications to develop, secondary to the pain associated with PV. This can be highlighted by cases of repeated dehydration and malnutrition seen in PV patients (Rashid and Candido 2008). Therefore it seems necessary to obtain new modalities for pain control of these oral lesions during conventional systemic therapy.

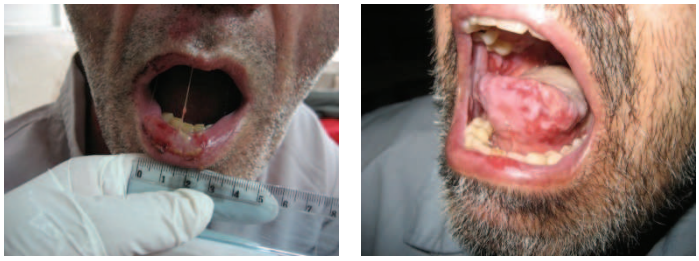


Fig. 5. Pemphigus vulgaris

4.2.3.2 NACLT and oral lesions of pemphigus vulgaris

A pilot before-after clinical trial was designed to evaluate the analgesic effects of application of a single session of NACLT in oral lesions of PV. Thirty eight painful oral lesions of ten patients with PV were irradiated with CO_2 laser by NACLT protocol. The patients scored and recorded the pain severity of their lesions on a visual analogue scale (VAS) up to 7 days

post operatively. Immediately after NACLT, the severity of idiopathic (non-contact) and contact pain were dramatically declined ($p<0.001$), so that the patients could eat and drink without any difficulties. This analgesic effect was sustained during follow-up periods. There was no visual effect of thermal damage to the oral mucosa or aggravation of the lesions. The results of this pilot study suggested that a single session of NACLT could reduce pain in oral lesions of pemphigus vulgaris immediately and significantly, without visible side effects (Zand, Mansouri et al. 2009). We recommend that in further studies, the pain severity of the lesions should be followed up for longer periods of time.

It should be noted that due to the life threatening nature of PV without appropriate systemic treatment, the patients must be warned that NACLT should not alter their conventional treatment at all, in spite of its significant analgesic effect, as we instructed our patients to comply with their prescribed medical regimen.

4.2.3.3 Literature review

In a case report, the oral lesions of two patients with recalcitrant oral pemphigus vulgaris (who were under systemic treatment) were irradiated with CO_2 laser at 1-1.5 W in a defocused mode for 5-10 seconds. The patients reported no pain after treatment. Recall examination after 1 month, 3 months and 5 month revealed complete healing of the lesions with no recurrence (Bhardwaj, Joshi et al. 2010).

The pictures of the paper demonstrate the themal, ablative nature within the procedure. Its pain relieving effects can be explained by its simultaneous biomodulative effects of CO_2 laser irradiation.

Title	Author	Study	Type of irradiation	Need to Anesthesia	Number of patients
Relieving pain in painful oral lesions of pemphigus vulgaris by a single session, Non-ablative 10600 nm CO_2 Laser irradiation (pilot study)	Zand, N., Mansouri, P. et al.	Before-after clinical trial 2009	NACLT	_	Ten patients/38 lesions
Management of recalcitrant oral pemphigus vulgaris with CO_2 laser-Report of two cases	Bhardwaj, A. et al.	Case report 2010	CO_2 laser Irradiation of the lesions in a defocused mode (thermal)	?	Two patients/? lesions

Table 3. Irradiation of oral lesions of pemphigus vulgaris with CO_2 laser

4.2.4 Post chemotherapy oral mucositis

4.2.4.1 Definition

Oral mucositis is a common, debilitating, and potentially serious complication of chemoradiotherapy. Studies have shown that mucositis will develop in about 40% of chemotherapy patients, 80% of bone marrow transplant patients, and 100% of patients treated with radiotherapy to the head and neck (Berger & Kilroy 1997; Sonis et al. 1999).

It presents as erythema, edema, ulceration, bleeding along with pain. The pain of the lesions is aggravated by the patient's swallowing and normal oral functioning. Consequently, oral intake difficulties lead to loss of weight. The progression of oral lesions and their impact on the patient's general condition may require nasogastric tube feeding or temporary discontinuation of the treatment or modification of the therapeutic plan (Arora, Pai et al. 2008).

Pathologic evaluation of mucositis reveals mucosal thinning leading to a shallow ulcer thought to be caused by inflammation and depletion of the epithelial basal layer with subsequent denudation and bacterial infection. The wound healing response to this injury is characterized by inflammatory cell infiltration, interstitial exudate, fibrin and cell debris producing a "pseudomembrane" (Sonis 2004).

Various preventive measures causing alteration of mucosa (cryotherapy, allopurinol, pilocarpine, leucovorin), modification of mucosal proliferation (beta-carotene, glutamine, cytokines), and antimicrobial or anti-inflammatory action (chlorhexidine, corticosteroids) have been tried. General oral care, diet, topical mucosal coating agents (sucralfate, magnesium hydroxide), topical anesthetics, and systemic analgesics (opiods and non opioids) (Arora, Pai et al. 2008), recombinant human keratinocyte growth factor (palifermin) and Amifostine have also been suggested (Kuhn, Porto et al. 2009). However, currently no definitive preventive or therapeutic intervention exists that is completely successful at preventing oral mucositis and treatment for this complication has thus been symptomatic (Arora, Pai et al. 2008).

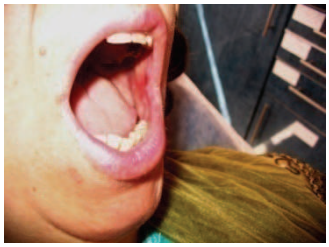


Fig. 6. Post Docetaxole chemotherapy oral mucositis

4.2.4.2 NACLT and mild to moderate post chemotherapy oral mucositis

A pilot before-after clinical trial was designed to evaluate the effects of single session of non-thermal, Non-ablative CO_2 Laser therapy (NACLT) to reduce pain in mild to moderate oral mucositis following breast cancer chemotherapy with Docetaxole. Six patients were included and their oral lesions were irradiated by NACLT. The patients reported the idiopathic (non-contact) and contact pain of their lesions on VAS (visual analogue scale) before and immediately after laser and up to 7 days post operatively. The results of the

study showed that dramatically after NACLT, the severity of pain declined immediately and it was sustained during follow-up periods (P<0.001). Similar to the other NACLT clinical trials, the procedure itself was painless and anesthesia was not required. There was no visible side effect such as ulceration, erosion and even erythema following NACLT. The results of the study suggested that single session of NACLT could reduce pain in lesions of mild/moderate post Docetaxole oral mucositis immediately and dramatically without visible side effects (Zand, Najafi et al. 2010).

4.2.4.3 A brief literature review

Some studies have shown that laser phototherapy (LPT) can be useful in prevention or treatment of oral mucositis. The principle behind using laser phototherapy (LPT) is that it accelerates wound healing and has anti-inflammatory effects (Arora, Pai et al. 2008). In addition, pain relieving properties of low power lasers seem to be of value in management of painful lesions of oral mucositis (Arora, Pai et al. 2008).

Some researchers state that prophylactic laser application seems more successful than curative laser application, although the reason is not entirely clear (Arora, Pai et al. 2008). Most studies of LLLT in cancer patients have focused on oral mucositis prevention (Bensadoun, Franquin et al. 1994; Bensadoun, Franquin et al. 1999; Rubenstein, Peterson et al. 2004; Bensadoun, Le Page et al. 2006; Kuhn, Porto et al. 2009; Clarkson, Worthington et al. 2010; Bjordal, Bensadoun et al. 2011) . Since the subjects of this section refers to analgesic effects of low power lasers in established chemotherapy-induced oral mucositis (COM), we briefly review some articles in which LLLT has been used for relieving pain in patients with chemotherapy-induced oral mucositis and not the prophylactic laser protocols.

Cauwels and Martens evaluated the capacity of analgesic effect and wound healing of low level laser therapy in 16 children suffering from chemotherapy-induced oral mucositis. All children were treated using a GaAlAs diode laser (wavelength: 830 nm, potency: 150 mW). The energy released was adapted according to the severity of the oral lesions. The same protocol was repeated every 48 hours until healing of each lesion occurred. The results of the study demonstrated that immediately after irradiation of the oral mucositis, pain relief was noticed. Depending on the severity of oral mucositis, on average, 2.5 treatments per lesion in a period of 1 week were sufficient to heal a mucositis lesion. They concluded that LLLT could reduce the severity and duration of mucositis and to relieve pain significantly (Cauwels and Martens 2011).

Nes and Posso investigated the pain relieving effect of LLLT among 13 patients who have developed moderate chemotherapy-induced oral mucositis. The laser used was GaAlAs (830 nm, power: 250 mW, energy density: 35 J cm^{-2}). The patients were treated during a 5-day period, and the pain was measured before and after each laser application. There was a significant (P= 0.007) 67% decrease in the daily average experience of pain felt before and after each treatment, confirming that LLLT can relieve pain among patients who have developed chemotherapy-induced oral mucositis (Nes and Posso 2005).

4.2.5 Other NACLT studies

We have evaluated the pain relieving effects of NACLT in some other painful mucosal lesions such as painful oral lesions of Stevens Johnson Syndrome, etc. as case reports, the

results of which are being published. In addition we have used NACLT in few ulcerated and non-ulcerated skin diseases with variable degrees of success. In some lesions, such as post herpetic neuralgia, NACLT acts like other conventional therapeutic lasers, in which the pain relieving effect is completely short standing and needs the several sessions of NACLT to be performed frequently.

In addition, we have evaluated the effects of NACLT in promoting wound healing in few studies with variable degrees of success. Although in few studies, NACLT has shown some valuable results in this field we are not yet ready to express our view in this regard. Certainly, controlled clinical trials with larger sample sizes are necessary to further evaluate the promoting wound healing effects of NACLT.

It should be mentioned that since the biological effects of NACLT and their mechanisms are not fully known, it seems ethically questionable to use NACLT in diseases with malignant potential, such as oral lichen planus, consequently we have not assessed the effects of NACLT in such illnesses.

4.3 Presumed mechanisms of pain relieving effects of NACLT

In order to develop an understanding of the mechanisms of analgesic effect of NACLT, powermetry and thermometry were performed in prior studies, the results of which demonstrated the low power nature of the applied CO_2 laser (Zand, Ataie-Fashtami et al. 2009). (Please see also 4. NACLT)

Since the analgesic effect of NACLT is immediate, we assume that at least in part, physiological neural changes such as blockage of action potential generation and conduction of nociceptive signals in primary afferent neurons might take part in this analgesic effect (Zand, Ataie-Fashtami et al. 2009). Destruction of nerve endings is less probable to be induced by NACLT, because, even in the studies in which CO_2 laser has been used as a surgical scalpel, there have been no statistically significant differences in the number of intact peripheral nerve structures in laser-treated sites in comparison with sites treated with electrocautery and scalpel (Rocha, Pinheiro et al. 2001). It is not known, whether the other mechanisms such as increase in β-endorphin synthesis and release, changes in bradykinin, prostaglandins, substance P, serotonin, acetylcholine, nitric oxide, singlet oxygen production, and the other biochemical events- which have been proposed to play a part in pain relieving effect of conventional low power lasers - are responsible for analgesic effect of NACLT or not. (Please see also 2.4.Mechanisms of analgesic effects of low power laser therapy) Further basic studies are necessary to elucidate the mechanisms of this analgesic effect.

On the other hand, there are some differences between analgesic effects of NACLT and the other classical low power lasers. The analgesic effect in LLLT is usually gradual, cumulative, and multi-session (Pinheiro, Cavalcanti et al. 1998; Gur, Karakoc et al. 2002; Gur, Sarac et al. 2004; Nes and Posso 2005; Chow, Heller et al. 2006; Djavid, Mehrdad et al. 2007; Bjordal, Bensadoun et al. 2011; Iwatsuki, Yoshimine et al. 2011; Ribeiro, de Aguiar et al. 2011). In contrast, the pain relieving effect of NACLT is immediate, dramatic and more sustained than conventional low level therapeutic lasers (Zand, Ataie-Fashtami et al. 2009; Zand, Mansouri et al. 2009; Zand, Najafi et al. 2010). Therefore one could presume that the mechanisms of analgesic effect of NACLT might have some differences from that for conventional low power lasers, an assumption that should be assessed in further basic studies.

In some ulcerated oral lesions such as aphthous ulcers, the pain of the lesions derives from inflammatory sensitization of small-diameter afferent nerve endings that form a plexus at the junction of the epithelial and subepithelial layers. Branches of this plexus extend upward, into the epithelial layer; producing a superficial, focal, inflammatory lesion that is directly associated with exposed sensory nerve endings. Therefore, in such ulcers, CO_2 laser irradiation can reach the exposed nerve endings easily and as we assume, for example, block the action potential generation and conduction of nociceptive signals in primary afferent neurons. On the other hand, in other under-publish studies, we have used NACLT for reducing pain in few non-ulcerated lesions, such as pre-aphthous lesions with moderate-good results. As the CO_2 laser's beam has a very limited depth of penetration in to the tissue, explaining this analgesic effect of NACLT seems more complex.

Tuner and Hode state that the therapeutic effects of defocused CO_2 laser must be due to the influence of the laser energy on the cells encountered, so that signal substances are released and then circulate in the organism. This indirectly confirms the hypothesis that conventional laser therapy has both a local effect in the area treated by laser light, and a systemic effect through the release of metabolites (Tuner and Hode 2010). We don't know whether such mechanisms may at least in part, take part in the analgesic effect of NACLT or not.

Further fundamental studies are necessary to elucidate the mechanisms of analgesic effect of NACLT.

5. Conclusion

CO_2 laser has been used as a very useful high power, thermal laser in surgery for cutting, ablation and coagulation of the tissues for many years. In contrast, in non- thermal, Non-Ablative CO_2 Laser Therapy (NACLT), the CO_2 laser is used as a low level (phototherapeutic) laser to reduce pain in some oral mucosal lesions without any visual effects of thermal damage to the oral mucosa such as ablation, ulceration or aggravation of the lesions. The results of powermetry and thermometry have demonstrated the low power nature of the applied CO_2 laser in NACLT.

As discussed above, in order to use the CO_2 laser as a phototherapeutic laser for NACLT, the CO_2 laser beam is irradiated through a thick layer of transparent, non-anesthetic gel with high water content. In addition, the CO_2 laser is operated with a de-focused hand piece, scanning rapidly over the lesion with circular motion. With these considerations, CO_2 laser can be used as a non-destructive, non-thermal, phototherapeutic laser (NACLT) to reduce pain in some oral mucosal lesions immediately and dramatically, so that after NACLT, the patients of the studies have been able to eat and drink easily at once. So far, in the series of NACLT studies, we have not observed any visible side effects following careful performance of the technique.

Certainly, controlled clinical trials with larger sample sizes will be able to prove the analgesic effects of NACLT more definitely. We recommend that in further studies, the pain severity of the lesions would be followed up for longer periods of time.

In addition, it should be emphasized that in serious diseases such as pemphigus vulgaris, Behcet's disease, etc., the patients must be warned that NACLT should not alter their conventional systemic treatment in spite of its significant analgesic effect.

6. References

Aimbire, F., R. Albertini, et al. (2006). "Low-level laser therapy induces dose-dependent reduction of TNFalpha levels in acute inflammation." *Photomed Laser Surg* 24(1): 33-37.

Alidaee, M. R., A. Taheri, et al. (2005). "Silver nitrate cautery in aphthous stomatitis: a randomized controlled trial." *Br J Dermatol* 153(3): 521-525.

Andre, P. (2003). "Ingrowing nails and carbon dioxide laser surgery." *J Eur Acad Dermatol Venereol* 17(3): 288-290.

Arikan, O. K., A. Birol, et al. (2006). "A prospective randomized controlled trial to determine if cryotherapy can reduce the pain of patients with minor form of recurrent aphthous stomatitis." *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 101(1): e1-5.

Arora, H., K. M. Pai, et al. (2008). "Efficacy of He-Ne Laser in the prevention and treatment of radiotherapy-induced oral mucositis in oral cancer patients." Oral Surg Oral Med *Oral Pathol Oral Radiol Endod* 105(2): 180-186, 186 e181.

Asilian, A., A. Yoosefi, et al. (2006). "Pemphigus vulgaris in Iran: epidemiology and clinical profile." *Skinmed* 5(2): 69-71.

Basford, J. R., C. G. Sheffield, et al. (1999). "Laser therapy: a randomized, controlled trial of the effects of low-intensity Nd:YAG laser irradiation on musculoskeletal back pain." *Arch Phys Med Rehabil* 80(6): 647-652.

Bensadoun, R., J. Franquin, et al. (1999). "Low-energy He-Ne laser in the prevention of radiation-induced mucositis." *Support Care Cancer* 7: 244–252.

Bensadoun, R., J. Franquin, et al. (1994). "Low-energy He/Ne laser in the prevention of radiation-induced mucositis. A multicenter phase III randomized study in patients with head and neck cancer." *Support Care Cancer* 7: 244-252.

Bensadoun, R., F. Le Page, et al. (2006). "Radiation-induced mucositis of the aerodigestive tract: prevention and treatment. MASCC/ISOO mucositis group's recommendations." *Bull Cancer* 93: 201-211.

Bhardwaj, M., M. Joshi, et al. (2010). "Management of recalcitrant oral pemphigus vulgaris with CO2 laser-Report of two cases." *Journal of Indian Society of Periodontology* 14(2): 132-135.

Bjordal, J., C. Couppe, et al. (2001). "Low-level laser therapy for tendinopathy: evidence of a dose-response pattern." *Phys Ther Rev* 6: 91-99.

Bjordal, J. M., R. J. Bensadoun, et al. (2011). "A systematic review with meta-analysis of the effect of low-level laser therapy (LLLT) in cancer therapy-induced oral mucositis." *Support Care Cancer* 19(8): 1069-1077.

Bjordal, J. M., C. Couppe, et al. (2003). "A systematic review of low level laser therapy with location-specific doses for pain from chronic joint disorders." Aust J Physiother 49(2): 107-116.

Black, M., M. D. Mignogna, et al. (2005). "Number II. Pemphigus vulgaris." *Oral Dis* 11(3): 119-130.

Brosseau, L., V. Robinson, et al. (2005). "Low level laser therapy (Classes I, II and III) for treating rheumatoid arthritis." *Cochrane Database Syst Rev*(4): CD002049.

Bystryn, J. C. and J. L. Rudolph (2005). "Pemphigus." *Lancet* 366(9479): 61-73.

Cauwels, R. G. and L. C. Martens (2011). "Low level laser therapy in oral mucositis: a pilot study." *Eur Arch Paediatr Dent* 12(2): 118-123.

Chams-Davatchi, C., M. Valikhani, et al. (2005). "Pemphigus: analysis of 1209 cases." *Int J Dermatol* 44(6): 470-476.

Chia, Y. W., A. Darzi, et al. (1995). "CO2 laser haemorrhoidectomy--does it alter anorectal function or decrease pain compared to conventional haemorrhoidectomy?" *Int J Colorectal Dis* 10(1): 22-24.

Chow, R., P. Armati, et al. (2011). "Inhibitory effects of laser irradiation on peripheral mammalian nerves and relevance to analgesic effects: a systematic review." *Photomed Laser Surg* 29(6): 365-381.

Chow, R. T. and L. Barnsley (2005). "Systematic review of the literature of low-level laser therapy (LLLT) in the management of neck pain." *Lasers Surg Med* 37(1): 46-52.

Chow, R. T., M. A. David, et al. (2007). "830 nm laser irradiation induces varicosity formation, reduces mitochondrial membrane potential and blocks fast axonal flow in small and medium diameter rat dorsal root ganglion neurons: implications for the analgesic effects of 830 nm laser." *J Peripher Nerv Syst* 12(1): 28-39.

Chow, R. T., G. Z. Heller, et al. (2006). "The effect of 300 mW, 830 nm laser on chronic neck pain: a double-blind, randomized, placebo-controlled study." *Pain* 124(1-2): 201-210.

Clarkson, J. E., H. V. Worthington, et al. (2010). "Interventions for treating oral mucositis for patients with cancer receiving treatment." *Cochrane Database Syst Rev*(8): CD001973.

Colvard, M. and P. Kuo (1991). "Managing aphthous ulcers: laser treatment applied." *J Am Dent Assoc* 122(6): 51-53.

Davatchi, F., C. Shahram, et al. (2005). "Behcet disease." *Acta Medica Iranica* 43(4): 233-242.

De Bie, R., H. De. Vet, et al. (1998). "Efficacy of 904 nm laser therapy in the management of musculoskeletal disorders: a systematic review." *Phys Ther Rev* 3: 59-72.

De Souza, T., M. Martins, et al. (2010). "Clinical evaluation of low-level laser treatment for recurring aphthous stomatitis." *Photomed Laser Surg* 28 Suppl(2): 85-88.

Demetriades, M., Hansford, et al. (2009). "General Manifestations of Behçet's Syndrome and the Success of CO2-Laser as Treatment for Oral Lesions: A Review of the Literature and Case Presentation." *Journal of the Massachusetts Dental Society* 58 (3): 24-27.

Demidov, V. P., V. I. Rykov, et al. (1992). "[The use of the carbon dioxide laser in the surgical treatment of breast cancer]." *Vopr Onkol* 38(1): 42-50.

Demidova-Rice, T. N., E. V. Salomatina, et al. (2007). "Low-level light stimulates excisional wound healing in mice." *Lasers Surg Med* 39: 706–715.

Djavid, G. E., R. Mehrdad, et al. (2007). "In chronic low back pain, low level laser therapy combined with exercise is more beneficial than exercise alone in the long term: a randomised trial." *Aust J Physiother* 53(3): 155-160.

Duncavage, J. A. and R. H. Ossoff (1986). "Use of the CO2 laser for malignant disease of the oral cavity." *Lasers Surg Med* 6(5): 442-444.

Elad, S., R. Or, et al. (2003). "CO2 laser in oral graft-versus-host disease: a pilot study." *Bone Marrow Transplant* 32(10): 1031-1034.

Enwemeka, C. S., J. C. Parker, et al. (2004). "The efficacy of low-power lasers in tissue repair and pain control: a meta-analysis study." *Photomed Laser Surg* 22(4): 323-329.

Fekrazad, R., S. Jafari, et al. (2006). *Evaluation of the effects of pulsed Nd:YAG laser on RAU*. The 6th international congress of the World Association of Laser Therapy, Lissamol, Cyprus.

Gam, A. N., H. Thorsen, et al. (1993). "The effect of low-level laser therapy on musculoskeletal pain: a meta-analysis." *Pain* 52(1): 63-66.

Ghate, J. V. and J. L. Jorizzo (1999). "Behcet's disease and complex aphthosis." *J Am Acad Dermatol* 40(1): 1-18; quiz 19-20.

Gigo-Benato, D., S. Geuna, et al. (2005). "Phototherapy for enhancing peripheral nerve repair: a review of the literature." *Muscle Nerve* 31(6): 694-701.

Goncalves, W. L., F. M. Souza, et al. (2007). "Influence of He-Ne laser therapy on the dynamics of wound healing in mice treated with anti-inflammatory drugs." *Braz J Med Biol Res* 40(6): 877-884.

Gur, A., M. Karakoc, et al. (2002). "Efficacy of low power laser therapy in fibromyalgia: a single-blind, placebo-controlled trial." *Lasers Med Sci* 17(1): 57-61.

Gur, A., A. J. Sarac, et al. (2004). "Efficacy of 904 nm gallium arsenide low level laser therapy in the management of chronic myofascial pain in the neck: a double-blind and randomize-controlled trial." *Lasers Surg Med* 35(3): 229-235.

Hagiwara, S., H. Iwasaka, et al. (2007). "GaAlAs (830 nm) low-level laser enhances peripheral endogenous opioid analgesia in rats." *Lasers Surg Med* 39(10): 797-802.

Haldeman, S., L. Carroll, et al. (2008). "The Bone and Joint Decade 2000-2010 Task Force on Neck Pain and Its Associated Disorders: executive summary." *Spine (Phila Pa 1976)* 33(4 Suppl): S5-7.

Hamblin, M. R. (2010). "Introduction to experimental and clinical studies using low-level laser (light) therapy (LLLT)." *Lasers Surg Med* 42(6): 447-449.

Hamblin, M. R., R. W. Waynant, et al. (2006). "Mechanisms for Low-Light Therapy." *Proc. of SPIE* 6140 614001-614010.

Hashmi, J. T., Y. Y. Huang, et al. (2010). "Role of low-level laser therapy in neurorehabilitation." *PM R* 2(12 Suppl 2): S292-305.

Huang, Y. Y., A. C. Chen, et al. (2009). "Biphasic dose response in low level light therapy." *Dose Response* 7(4): 358-383.

Iwatsuki, K., T. Yoshimine, et al. (2011). "Percutaneous diode laser irradiation for lumbar discogenic pain: a clinical study." *Photomed Laser Surg* 29(7): 459-463.

Jimbo, K., K. Noda, et al. (1998). "Suppressive effects of low-power laser irradiation on bradykinin evoked action potentials in cultured murine dorsal root ganglion cells." *Neurosci Lett* 240(2): 93-96.

Jurge, S., R. Kuffer, et al. (2006). "Number VI - Recurrent aphthous stomatitis." *Oral Diseases* 12(1): 1-21.

Kaplan, I., I. Kott, et al. (1996). "The CO2 laser in the treatment of lesions of the eyelids and periorbital region." *J Clin Laser Med Surg* 14(4): 185-187.

Karu, T. (1989). "Photobiology of low-power laser effects." *Health Phys* 56(5): 691-704.

Karu, T. I. and S. F. Kolyakov (2005). "Exact action spectra for cellular responses relevant to phototherapy" *Photomed Laser Surg* 23(4): 355-361.

Karu, T. I., L. V. Pyatibrat, et al. (2004). "Photobiological modulation of cell attachment via cytochrome c oxidase." *Photochem Photobiol Sci* 2004;3(2):211–216. 3(2): 211-216.

Kasai, S., T. Kono, et al. (1996). "Effect of low-power laser irradiation on impulse conduction in anesthetized rabbits." *J Clin Laser Med Surg* 14(3): 107-109.

Kasugai, C., D. Watanabe, et al. (2010). "Infliximab treatment of severe genital ulcers associated with Behcet disease." *J Am Acad Dermatol* 62(1): 162-164.

Kaviani, A., M. Fateh, et al. (2008). "Comparison of carbon dioxide laser and scalpel for breast lumpectomy: a randomized controlled trial." *Photomed Laser Surg* 26(3): 257-262.

Kuhn, A., F. A. Porto, et al. (2009). "Low-level infrared laser therapy in chemotherapy-induced oral mucositis: a randomized placebo-controlled trial in children." *J Pediatr Hematol Oncol* 31(1): 33-37.

Labajos, M. (1988). "β-endorphin levels modification after GaAs and HeNe laser irradiation on the rabbit. Comparative study " *Investigacion clinica laser* 1-2(6-8).

Lin, P. and G. Liang (2006). "Behcet disease: recommendation for clinical management of mucocutaneous lesions." *J Clin Rheumatol* 12(6): 282-286.

Longo, L., Z. Simunovic, et al. (1997). "Laser therapy for fibromyositic rheumatisms." *J Clin Laser Med Surg* 15(5): 217-220.

Maeda, T. (1989). "Morphological demonstration of low reactive laser therapeutic pain attenuation effect of the gallium aluminium arsenide diode laser" *Laser Ther* 1: 23-30.

Meador, R., G. Ehrlich, et al. (2002). "Behcet's disease: immunopathologic and therapeutic aspects." *Curr Rheumatol Rep* 4(1): 47-54.

Mizutani, K., Y. Musya, et al. (2004). "A clinical study on serum prostaglandin E2 with low-level laser therapy." *Photomed Laser Surg* 22(6): 537-539.

Montesinos, A. (1988). "Experimental effects of low power laser in encephalin and endorphin synthesis." *Journ Eur Med Laser Ass* 1(3): 2-7.

Mrowiec, J. (1997). *Analgesic effect of low-power infrared laser irradiation in rats*. SPIE.

Nes, A. G. and M. B. Posso (2005). "Patients with moderate chemotherapy-induced mucositis: pain therapy using low intensity lasers." *Int Nurs Rev* 52(1): 68-72.

Nicola, E. M. and H. Nicola (1994). *Low power CO2 laser in the treatment of chronic pharyngitis: a five- year experience* Proc. SPIE.

Ohno, T. (1997). "[Pain suppressive effect of low power laser irradiation. A quantitative analysis of substance P in the rat spinal dorsal root ganglion]." *Nippon Ika Daigaku Zasshi* 64(5): 395-400.

Orchardson, R., J. M. Peacock, et al. (1997). "Effect of pulsed Nd:YAG laser radiation on action potential conduction in isolated mammalian spinal nerves." *Lasers Surg Med* 21(2): 142-148.

Pinheiro, A. L., E. T. Cavalcanti, et al. (1998). "Low-level laser therapy is an important tool to treat disorders of the maxillofacial region." *J Clin Laser Med Surg* 16(4): 223-226.

Posten, W., D. A. Wrone, et al. (2005). "Low-level laser therapy for wound healing: mechanism and efficacy." *Dermatol Surg* 31(3): 334-340.

Prolo P, F. Z., Domingo D, Outhouse T, Thornhill M (2006). "Interventions for recurrent aphthous stomatitis (mouth ulcers)." *The Cochrane liberary* 3(10).

Rashid, R. M. and K. D. Candido (2008). "Pemphigus pain: a review on management." *Clin J Pain* 24(8): 734-735.

Ribeiro, A. S., M. C. de Aguiar, et al. (2011). "660 AsGaAl laser to alleviate pain caused by cryosurgical treatment of oral leukoplakia: a preliminary study." *Photomed Laser Surg* 29(5): 345-350.

Rocha, E. A., A. L. Pinheiro, et al. (2001). "Quantitative evaluation of intact peripheral nerve structures after utilization of CO2 laser, electrocautery, and scalpel." *J Clin Laser Med Surg* 19(3): 121-126.

Rubenstein, E. B., D. E. Peterson, et al. (2004). "Clinical practice guidelines for the prevention and treatment of cancer therapy-induced oral and gastrointestinal mucositis." *Cancer* 100(9 Suppl): 2026-2046.

Scully, C., M. Gorsky, et al. (2003). "The diagnosis and management of recurrent aphthous stomatitis: a consensus approach." *J Am Dent Assoc* 134(2): 200-207.

Sharon-Buller, A. and M. Sela (2004). "CO2-laser treatment of ulcerative lesions." *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 97(3): 332-334.

Shashy, R. G. and M. B. Ridley (2000). "Aphthous ulcers: a difficult clinical entity." *Am J Otolaryngol* 21(6): 389-393.

Shimizu, N., M. Yamaguchi, et al. (1995). "Inhibition of prostaglandin E2 and interleukin 1-beta production by low-power laser irradiation in stretched human periodontal ligament cells." *J Dent Res* 74(7): 1382-1388.

Simunovic, Z. (2000). pain and practical aspects of its management. *Lasers in medicine and dentistry*. Z. Simunovic. zagreb, AKD: 269-301.

Snyder, S., K. Byrnes, et al. (2002). "Quantification of calcitonin gene-related peptide mRNA and neuronal cell death in facial motor nuclei following axotomy and 633 nm low power laser treatment.." *Lasers Surg Med* 31: 216 –222.

Sonis, S. T. (2004). "The pathobiology of mucositis." *Nat Rev Cancer* 4(4): 277-284.

Soriano, F. and R. Rios (1998). "Gallium arsenide laser treatment of chronic low back pain: a prospective, randomized and double blind study." *Laser Ther* 10: 175-180.

Sutherland, J. C. (2002). "Biological effects of polychromatic light." *Photochem Photobiol* 76(2): 164-170.

Suzuki Kurokawa, M. and N. Suzuki (2004). "Behcet's disease." *Clin Exp Med* 4(1): 10-20.

Tada, H., M. Hatoko, et al. (2004). "Clinical comparison of the scanning CO2 laser and conventional surgery in the treatment of ingrown nail deformities." *J Dermatolog Treat* 15(6): 387-390.

Tuner, J. and L. Hode (2010). Biostimulation, Laser therapy with high output lasers. *The new laser therapy handbook* Prima books AB: 67-147.

Tuner, J. and L. Hode (2010). The mechanisms, effects on pain. *The new laser therapy handbook*, Prima books AB: 557-559.

Tuner, J. and L. Hode (2010). Medical indications. *The new laser therapy handbook*, Prima Books: 149-372.

Tuner, J. and L. Hode (2010). Some basic laser physics,Therapeutic lasers. *The new laser therapy handbook*, Prima books AB: 1-47.

Umegaki, S., Y. Tanaka, et al. (1989). "Effectiveness of low-power laser therapy on low-back pain – double-blind comparative study to evaluate the analgesic effect of low power laser therapy on low back pain." *Clin Rep* 23: 2838–2846.

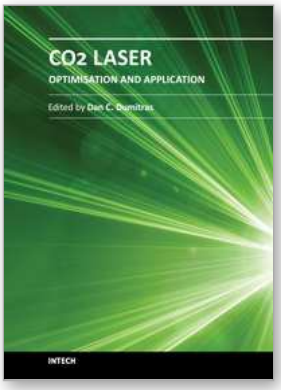
Wakabayashi, H., M. Hamba, et al. (1993). "Effect of irradiation by semiconductor laser on responses evoked in trigeminal caudal neurons by tooth pulp stimulation." *Lasers Surg Med* 13(6): 605-610.

Zand, N., L. Ataie-Fashtami, et al. (2009). "Relieving pain in minor aphthous stomatitis by a single session of non-thermal carbon dioxide laser irradiation." *Lasers Med Sci* 24(4): 515-520.

Zand, N., L. Ataie-Fashtami, et al. (2009). "Analgesic effects of single session of Non-Ablative CO2 Laser Therapy (NACLT) in major aphthous ulcers: (a preliminary study) " *Lasers in medicine* 6(4): 6-12.

Zand, N., P. Mansouri, et al. (2009). Relieving pain in painful oral lesions of pemphigus vulgaris by a single session, Non-ablative 10600 nm CO_2 Laser irradiation (pilot study). *The 29th Annual conference of the American Society for Lasers in surgery and medicine*. Harbor. 41: 67-68.

Zand, N., S. Najafi, et al. (2010). "NACLT (Non-ablative CO2 laser therapy): a new approach to relieve pain in mild to moderate oral mucositis following breast cancer chemotherapy (a pilot study) " *EJC supplements* 8(3): 166.



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The present book includes several contributions aiming a deeper understanding of the basic processes in the operation of CO2 lasers (lasing on non-traditional bands, frequency stabilization, photoacoustic spectroscopy) and achievement of new systems (CO2 lasers generating ultrashort pulses or high average power, lasers based on diffusion cooled V-fold geometry, transmission of IR radiation through hollow core microstructured fibers). The second part of the book is dedicated to applications in material processing (heat treatment, welding, synthesis of new materials, micro fluidics) and in medicine (clinical applications, dentistry, non-ablative therapy, acceleration of protons for cancer treatment).

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