A first experience of Low Level Laser Therapy as an adjunct to accelerate wound healing in chronic leg ulcers



Authors:
Harikrishna KR Nair, HarMohan Pall Singh Sing, Sylvia SY Chong

Objective: To demonstrate that photo-biomodulation accelerates wound healing through using Low Level Laser Therapy (LLLT) as an adjunct to conventional wound treatment in chronic non-healing leg ulcers. **Method:** Three patients with chronic wounds, i.e. venous ulcer, post-surgical non-healing ulcer and diabetic foot ulcer, were recruited for this evaluation. **Results:** The effectiveness and efficacy of LLLT was measured by comparing the pre- and post-treatment percentage of reduction in surface area of the ulcer by digital imaging. All three ulcers showed a significant reduction after 4 weeks of laser therapy. No adverse events were reported. **Conclusion:** The results of LLLT as an adjunct therapy for chronic wounds show that there was significant acceleration of wound healing as demonstrated by digital imaging. More clinical trials in the near future are needed to further investigate the mechanism of wound acceleration by photo-bioactivation.

***Harikrishna KR Nair** is Head of Wound Care Unit, Department of Internal Medicine, Hospital Kuala Lumpur; **Prof HarMohan Pall Singh HMP Sing** is Clinical Professor, Specialist in Laser Medicine, PU-RCSI, Perdana University, Malaysia; **Sylvia SY Chong** is Research Assistant, Wound Care Unit, Department of Internal Medicine, Hospital Kuala Lumpur*

Wound healing is a complex, highly regulated process that is critical in maintaining the barrier function of skin. With numerous disease processes, the cascade of events involved in wound healing can be affected, resulting in chronic, non-healing wounds that cause the patient significant discomfort and distress, while requiring a large amount of resources from the medical system. The healing of a superficial wound requires many factors to work in concert, and wound dressings and treatments have evolved considerably to address possible barriers to wound healing, ranging from infection to hypoxia. Even optimally, wound tissue never reaches its pre-injured strength and multiple aberrant healing states can result in chronic non-healing wounds (Han and Ceilley, 2017).

Chronic wounds are defined as wounds that have not proceeded through an orderly and timely reparation to produce anatomic and functional integrity after 3 months. All wound types have the potential to become chronic and, as such, chronic wounds are classified by cause, identification and treatment of which are essential (McInnes et al, 1998; Royal College of Nursing, 2006; 2008; Werdin et al, 2008). Venous or arterial insufficiency, diabetes, and local-pressure effects are the most common pathophysiological causes, whereas systemic factors, such as compromised nutritional status, infection, and altered immunological status further contribute to poor wound healing (Werdin et al, 2008).

Chronic wounds include, but are not limited, to diabetic foot ulcers, venous leg ulcers, and pressure ulcers. They are a challenge to wound care professionals and consume a great deal of healthcare resources around the globe (Frykberg and Banks, 2015). Chronic wounds represent a significant burden to patients, healthcare professionals, and the US healthcare system, affecting 5.7 million patients and costing an estimated $20 billion annually (Branski et al, 2008; Frykberg and Banks, 2015; de la Torre and Molnar, 2019).

It has been established that nearly 12–15% of patients with diabetes mellitus will develop a foot ulcer at some point in their life. In view of increasing morbidity, gangrene, amputation, increasing treatment cost and poor quality of life, it becomes imperative that proper wound care is given using different treatment options for complete healing (Gordois et al, 2003).

Use of adjunctive therapy, such as photo-biomodulation by LLLT, to conventional wound treatment has been shown to accelerate wound healing, thereby not only reducing treatment time and increasing patient compliance by reducing the number of visit and healing time, but also preventing infection, gangrene and amputation. This will also reduce the cost of treatment significantly.

Even though LLLT has been proven to be effective as adjunctive therapy in the treatment of diabetic foot ulcers, it has still not found its place as one of the standard treatments in ulcer management. The reason could be due to a lack of trials and studies to document its efficacy. Our initial case study is set out to address these issues by not only proving the beneficial effect of combining photo-biomodulation with conventional therapy, but also demonstrating accelerated wound healing by using laser therapy as an adjunctive treatment.

Background

Photo-biomodulation therapy is defined as a form of light therapy that utilizes non-ionizing light sources, including lasers, light emitting diodes, and/or broadband light, in the visible (400–700 nm) and near-infrared (700–1,100 nm) electromagnetic spectrum. It is a nonthermal process which involves endogenous chromophores eliciting photophysical (i.e. linear and nonlinear) and photochemical events at various biological scales. This process results in beneficial therapeutic outcomes, including, but not limited, to the alleviation of pain or inflammation, immunomodulation, and promotion of wound healing and tissue regeneration (Anders et al, 2015). Phototherapy, also known as photo-biomodulation by LLLT, involves the application of light (often laser light of a specific wavelength or a light emitting diode, LED) to stimulate cellular processes (Nemeth, 1993). The exact mechanisms of action that follow laser irradiation are not well understood, and a number of theories exist; the most studied and best understood being that of cytochrome-c oxidase (cyt a/a 3), the terminal enzyme in the eukaryotic mitochondrial respiratory chain (complex IV). Cytochrome c oxidase facilitates the transfer of electrons to molecular oxygen. The end product of this complex is the production of adenosine triphosphate (ATP) (Karu, 1999; Karu et al, 2008; 2010). Laser is absorbed by mitochondria leading to increase in ATP, reactive oxygen species, nitric oxide and intracellular calcium. These changes lead to increased cell survival and accelerate wound healing (Tuner and Hode, 2002; Pinheiro et al, 2002).

Different laser wavelengths have different depths of penetration into human tissue (Ebrahimi et al, 2012). Red laser has a deeper penetration depth than violet, blue, green, or yellow. Infrared and near infrared light are not visible, but it has been shown to penetrate human tissue deeper than visible red light (Litscher et al, 2004). Bichromatic laser needles combine wavelengths, for example, of red (685 nm) and infrared (785 nm) light. Their penetration depth in human skin is 2–3 cm. Patients usually do not feel the beginning of the treatment, but 5–10 minutes later many patients report a pleasant warm and sometimes vibrating feeling in some treated areas. Blue light (405 nm) is supposed to have a bactericidal effect on the tissue surface. Cell studies with cultured human keratinocytes, endothelial cells, and fibroblasts indicated potential effects of near-infrared light in the treatment of chronic skin ulcers. After irradiation of the cells, the production of transforming growth factor (TGF)-1 and matrix metalloproteinase (MMP)-2 was examined by enzyme immunoassay, zymography, and reverse transcription polymerase chain reaction (PCR). A biostimulatory effect of near-infrared irradiation was shown by a significant elevation of TGF-1 and MMP-2 content in the medium of cultured cells. Irradiated fibroblasts also showed an upregulated amount of MMP-2 mRNA. These results suggest that near- infrared irradiation may accelerate wound closure (Houreld and Abrahamse, 2010; Danno et al, 2001).

Houreld and Abrahamse (2010) tested the positive effect of low-intensity laser irradiation (LILI) of different wavelengths on cellular migration, viability, and proliferation in diabetic wounded and unwounded human skin fibroblast cells. They compared cellular morphology and migration (determined microscopically), cellular viability (determined by ATP luminescence), and proliferation (determined by basic fibroblast growth factor expression and alkaline phosphatase activity). While diabetic wounded cells irradiated at 1,064 nm showed a lesser degree of migration, viability, and proliferation, cells irradiated at 632.8 nm showed a higher degree of haptotaxis and migration, as well as ATP luminescence, compared to cells irradiated at 830 nm (Houreld and Abrahamse, 2010). In conclusion, diabetic wounded cells have more benefit in wound healing from irradiation in the visible range

(632.82 nm) than from the infrared range (Mester et al, 2009).

A study by Hopkins et al (2004) showed that wound size significantly reduced after application of low-level laser on follow up at days 6, 8 and 10 days. Gupta et al (1998) reported that the ulcer area reduced significantly (p=0.0002) in the study group compared with the control group. In a double-blind randomized placebo-controlled study, the healing effect of combined 660 and 890 nm LED laser treatment on 23 diabetic leg ulcers was tested by Minatel et al (2009). Mean ulcer granulation and healing rates were significantly higher in the treatment group than in the placebo group at each of 15, 30, 45, 60, 75 and 90 days of treatment. Placebo-treated ulcers were cleaned, dressed with 1% silver sulfadiazine cream, and treated with placebo laser radiation <1.0 j/cm^2. During the initial 30 days, they even worsened. The ulcers in the treatment group got the same treatment, but a 3 j/cm^2 dose. At day 30, the ulcers in the treatment group had achieved 56% more granulation and 79.2% faster healing than the placebo group, and similarly higher rates of granulation and healing were maintained throughout. In the treatment group, 58,3% of ulcers had healed fully and 75% had achieved 90–100% healing by day 90. In contrast, in the placebo group, only one ulcer healed fully and no ulcer attained more than 90% healing (Minatel et al, 2009).

Landau et al (2011) investigated the effect of broadband visible light (400–800 nm) in a double-blind, placebo-controlled, randomized study on 16 patients suffering from diabetic or venous foot ulcers. The treatment group received wound illumination twice daily with 43.2 j/cm^2, while the placebo group received wound illumination in the same device with only 2.4 j/cm^2, which was declared as nontherapeutic. All patients received conventional wound care. At the end of the follow up, all of the wounds in nine of the treatment-group patients were closed (90%), whereas in the placebo group, only two of six patients (33%) had closed wounds, judged by Wagner's classification for foot ulcer and wide/length measurement. There were no adverse therapy effects (Landau et al, 2011).

The study done by Kajagar et al (2012) compared diabetic ulcer healing dynamics in 68 patients. They were randomized into a LLLT plus conventional care group which was compared with conventional care alone. Depending on the ulcer size, the duration of exposure was calculated to deliver 2–4 j/cm^2 at 60 mW, 5 kHz, daily for 15 days. The ulcer floor and edge were irradiated. There was a significant percentage of ulcer reduction in the LLLT group compared with conventional care alone (Kajagar et al, 2012). A double blinded, randomized controlled clinical trial of 23 patients with a diabetic foot ulcer by Kaviani et al (2011) showed that four weeks after laser therapy, the size of the ulcers significantly decreased and by 20 weeks a greater number of patients treated with laser showed complete healing than the control group with lower mean healing time (Kaviani et al, 2011).

A number of recent studies have demonstrated the beneficial effect of laser therapy on diabetic foot ulcers, and the underlying mechanisms in *in-vitro* as well as *in-vivo* trials. Ramos et al (2018) demonstrated that laser treatment helps in decreasing the wound area and increasing the collagen fiber. Another study by Bagheri et al (2018) also showed similar results, along with an increase in tensiometric properties in the healing wound. Whereas in a study conducted by El-Makakey et al (2017), patients who had undergone appendectomy, demonstrated an increase in the activation of extracellular signal-regulated kinase and a decrease in pain sensation.

Method

Chronic wounds exhibit slow healing and increased morbidity due to hypoxia, neuropathy and pre-gangrenous state and extracellular matrix imbalance. The risk of developing foot ulcers is increased up to seven-fold in diabetic patients with neuropathy, as compared to non-neuropathic diabetic patients (Young et al, 1994). Despite progress in conventional wound healing many patients have to undergo amputations. In these case studies, the authors are demonstrating acceleration of wound healing by LLLT to prevent amputation in non-infective, non-ischemic ulcers. Three patients for this initial trial were included in the study. The wound assessment was done under the following parameters at weekly intervals:

- Clinical assessment
- Digital imaging.

No controls were used in this case series because it was an observational trial, which involved a particular intervention on patients with chronic wounds who received standard of care. This study conformed with the guidelines set out in the Declaration of Helsinki for Ethical Principles for Medical Research involving Human Subjects. The study was approved by the Kuala Lumpur Hospital Review Board (local institution board). The study's objectives and potential risks were explained to the patient in detail. Informed consent and permission to use wound photographs and case details for publication/research purposes were obtained, and the data was stored safely in the Kuala Lumpur Hospital.

Device used in the study

The laser used for this case study was Theralase® LLLT (Toronto, Canada), which had FDA approval and was provided by the Quick Stop Solutions (QSS) company (Kuala Lumpur, Malaysia).

Recruitment of patients

Study participants who were either attending for their routine treatment visits or referred for treatment in Wound Care Unit in Hospital Kuala Lumpur were recruited by simple randomization. Three patients with chronic wounds, comprising of two males and one female, were selected based on inclusion criteria.

Inclusion criteria

- Patients between 18 and 80 years old
- Ulcers persisted for more than 3 months
- Depth of ulcer was less than or equal to 1.5 cm (due to penetration depth of LLLT)
- No evidence of overt infection or ischemia.

Exclusion criteria

- Children and young adults under 18 years
- Infected foot ulcers
- Presence of osteomyelitis
- All ulcers less than 3 months duration
- Patients with comorbidities, such as end-stage renal disease, malignancy or tuberculosis.

Initial visit

The protocol described below was applied to all three patients during their first visit:

- Full medical history: clinical examination and detailed assessment of patient was done in respect of duration, medications, presence of other complications, ambulatory status and history of previous ulceration or amputation
- Assessment included: baseline measurement of greatest length and width to ascertain surface area using a grid ruler; measurement of depth of ulcer using sterile probe and digital radiography; state of granulation tissue, inspecting edge and margin of the ulcer; checking surrounding skin.

Treatment

Laser therapy was given in a special room in the wound care department provided for this purpose, all the safety protocols were strictly followed pertaining to safety goggles etc. The therapy was instituted under the guidance of trained laser technicians. The following protocols for laser therapy were followed:

- All the patients were subjected to laser therapy 3 times per week (Monday, Wednesday, Friday)
- The laser therapy was given by non-contact probe from a distance of 1 cm from the wound
- The exposure time of the laser depended on the wound size, with the aim to deliver an average energy of 4–5 j/cm^2
- The laser therapy was stopped if the primary endpoint of healing was achieved. This trial reflects the results of the 4 weeks only and is still ongoing.

Follow-up visits

Follow-up visits were scheduled three times a week, on Monday, Wednesday and Friday. Follow-up was for 4 weeks or stopped earlier if the primary end point had been achieved:

- Dressing was changed on each follow up visit
- The wound was assessed once a week on Wednesday in detail, e.g. to condition of dressing, surface area of the ulcer, state of granulation tissue, depth of ulcer
- A digital image of the ulcer was taken at initial visit, at 2nd week and at 4th weeks (or before then if primary end point achieved)
- The patient was assessed in respect of development of any new complaints, number of dressing changes and compliance to medicines
- Patient were informed about the importance of offloading to prevent mechanical stress to the ulcer during each visit for the diabetic ulcer and compression was used for venous ulcer, as per standard of care.

Ulcer dressing protocol

- The wound was cleaned and irrigated with distilled water during each visit
- Debridement, if required, was done with sharp instruments
- Dressings were changed at each follow-up visit
- Dressing changes were done under strict aseptic conditions, using collagen sheets as per standard of care by trained nurses in the wound care department of Hospital Kuala Lumpur
- A dressing card was given to the patient detailing the frequency and date of dressings.

Final evaluation

A final evaluation took place at 4th week or earlier if the wound had healed completely:

- The ulcer was assessed with respect to complete healing, which means no active discharge and complete epithelization
- Digital imaging was done at final visit.

Primary endpoint

The primary end point was complete healing of foot ulcer within specified time period of 4 weeks or earlier.

Secondary endpoints

Time to complete healing:

- The wound closure rate was assessed by comparing the wound size at 4 weekly intervals.
- Digital imaging was undertaken at the initial visit, 2nd week and 4th week.

Case study 1

A 74-year-old Indian gentleman, with a right chronic venous ulcer for 1 year, with underlying type 2 diabetes mellitus, hypertension and dyslipidaemia. Same dressing regimen (collagen sheet) were used throughout the study period as per standard of care.

The area of ulcer for pre-treatment is 17.2 cm^2 and area of ulcer post-treatment is 13.6 cm^2. The results showed that there was a significant percentage of ulcer reduction of 21.2%.

1st visit

5th visit

8th visit

12th visit

Case study 2

A 56-year-old Indian lady, with a post-surgical non-healing wound for 8 months, on her right medial malleolus with no known medical illness. Same dressing regimen (collagen sheet) were used throughout the study period as per standard of care.

The area of ulcer for pre-treatment is 1 cm^2 and area of ulcer post-treatment is 0 cm^2. The results showed that there was a significant percentage of ulcer reduction of 100%.

1st visit

5th visit

8th visit

12th visit

Case study 3

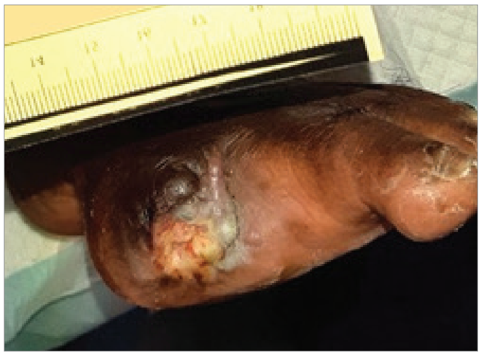
A 64-year-old Indian gentleman, with post-extended Ray's amputation with right 1st metatarsal hallux ulcer for 6 months with underlying type 2 diabetic mellitus and hypertension. Same dressing regimen (collagen sheet) were used throughout the study period as per standard of care.

The area of ulcer for pre-treatment is 3 cm^2 and area of ulcer post-treatment is 0.2 cm^2. The results showed that there was a significant percentage of ulcer reduction of 91.7%.

1st visit

5th visit

8th visit



10th visit

Discussion

Even though LLLT has been proven to be effective as adjunctive therapy in the treatment of diabetic foot ulcer, it has still not found its place as one of the standard treatments in ulcer management. The reason could be due to lack of trials and studies to document its efficacy. Our study aims to address these issues by not only proving the beneficial effect of combining LLLT with conventional therapy, but also demonstrating pathological changes suggestive of accelerated wound healing by using LLLT as adjunctive treatment.

Prevalence of chronic wounds are increasing in Malaysia. There is a need for a better controlled, well designed study to be carried out on the viability of low-level laser as a complimentary therapy to conventional wound treatment, so it can be accepted as a standard treatment for chronic wounds and benefit a large number of patients. Chronic wounds, with its associated complications in Malaysia, not only increases the healthcare cost of the hospital, but also places an economic burden on the patient, due to prolonged treatment. A well-designed study proving the efficacy of LLLT in conjunction with conventional wound treatment will lead to faster healing time, thereby reducing the treatment cost significantly.

Conclusion

Our initial results of LLLT as an adjunct therapy for chronic wounds showed that there was significant ulcer reduction and accelerated wound healing. In this case series, three patients with different wound aetiologies had LLLT applied to their wound as as an adjunct therapy to their standard care. The healing rates were quite impressive. A limitation of this study is the small sample size. A larger study needs to be conducted to show consistently good results. The LLLT is another modality which can be utilized as an adjunctive therapy in managing chronic complex wounds.

References

Anders JJ, Lanzafame RJ, Arany PR (2015) Low-level light/laser therapy versus photobiomodulation therapy. *Photomed Laser Surg* 33(4): 183–4

Bagheri M, Amini A, Abdollahifar MA et al (2018) Effects of photobiomodulation on degranulation and number of mast cells and wound strength in skin wound healing of streptozotocin-induced diabetic rats. *Photomed Laser Surg*. 36(8): 415–23

Branski LK, Gauglitz GG, Herndon DN, Jenschke MG (2009) A review of gene and stem cell therapy in cutaneous wound healing. *Burns* 35(2): 171–80

Danno K, Mori N, Toda K-I, Kobayashi T, Utani A (2001) Near-infrared irradiation stimulates cutaneous wound repair: laboratory experiments on possible mechanisms. *Photodermatol Photoimmunol Photomed* 17(6): 261–5

Ebrahimi T, Moslemi N, Rokn AR et al (2012) The influence of low-intensity laser therapy on bone healing. *J Dent (Tehran)* 9(4): 238–48

El-Makakey AM, El-Sharaby RM, Hassan MH, Balbaa A (2017) Comparative study of the efficacy of pulsed electromagnetic field and low-level laser therapy on mitogen-activated protein kinases. *Biochem Biophys Rep* 9: 316–321.

Frykberg RG, Banks J (2015) Challenges in the Treatment of Chronic Wounds. *Adv Wound Care (New Rochelle)* 4(9): 560–82

Gordois A, Scuffham P, Shearer A, Oglesby A, Tobian JA (2003) The health care costs of Diabetic peripheral neuropathy in the US. *Diabetes Care* 26(6): 1790–95

Gupta A K, Filonenko , Salansky N, Sauder D N (1998) The use of low energy photon therapy (LEPT) in venous leg ulcers: a double-blind, placebo-controlled study. *Dermatol Surg* 24(12): 1383–6

Han G, Ceilley R (2017) Chronic Wound Healing: A Review of Current Management and Treatments. *Adv Ther* 34(3): 599–610

Hopkins JT, McLoda TA, Seegmiller JG, Baxter GD (2004) Low-level laser therapy facilitates superficial wound healing in humans: a triple-blind, sham-controlled study. *J Athl Train* 39(3): 223–9

Houreld N, Abrahamse H (2010) Low-intensity laser irradiation stimulates wound healing in diabetic wounded fibroblast cells (WS1) *Diabetes Technol Ther* 12(12): 971–8

de la Torre J I, Joseph A Molnar (2019) *Chronic Wounds*. Available at: https://emedicine.medscape.com/article/1298452-overview#a1 (accessed 16.01.2020)

Kajagar BM, Godhi AS, Pandit A, Khatri S (2012) Efficacy of low-level laser therapy on wound healing in patients with chronic diabetic foot ulcers—a randomised control trial. *Indian J Surg* 74(5): 359–63

Karu T (1999) Primary and secondary mechanisms of action of visible to near-IR radiation on cells. *J Photochem Photobiol B* 49(1): 1–17

Karu TI, Pyatibrat LV, Kolyakov SF, Afanasyeva NI (2008) Absorption measurements of cell monolayers relevant to mechanisms of laser phototherapy: reduction or oxidation of cytochrome c oxidase under laser radiation at 632.8nm. *Photomed Laser Surg* 26(6): 593–99

Karu TI (2010) Multiple roles of cytochrome c oxidase in mammalian cells under action of red and IR-A radiation. *IUBMB Life* 62(8): 607–10

Kaviani A, Djavid GE, Ataie-Fashtami L et al (2011) A randomized clinical trial on the effect of low-level laser therapy on chronic diabetic foot wound healing: a preliminary report. Photomed Laser Surg29(2): 109–14

Landau Z, Migdal M, Lipovsky A, Lubart R (2011) Visible light-induced healing of diabetic or venous foot ulcers: a placebo-controlled double-blind study. *Photomed Laser Surg* 29(6): 399–404

Litscher G, Wang L, Schikora D et al (2004) Biological effects of painless laser needle acupuncture. *Medical Acupuncture* 16(1):24–9

McInnes E, Cullum N, Nelson A, Duff L (1998) RCN guideline on the management of leg ulcers. *Nurs Stand* 13: 61-63

Mester E, Mester AF, Mester A (1985) The biomedical effects of laser application. *Lasers Surg Med* 5(1): 31–9

Minatel DG, Frade MAC, França SC, Enwemeka CS (2009) Phototherapy promotes healing of chronic diabetic leg ulcers that failed to respond to other therapies. *Lasers Surg Me* 41(6): 433–41

Nemeth AJ (1993) Lasers and wound healing. *Dermatologic Clinics* 11(4): 783–89

Pinheiro AL, Carneiro Nascimento S, De Barros Vieira AL et al (2002) Effects of low-level laser therapy on malignant cells: in vitro study. *Med Surg* 20(1): 23–6

Ramos FS, Maifrino LBM, Alves S et al (2018) The effects of transcutaneous low-level laser therapy on the skin healing process: an experimental model. *Laser Med Sci* 33(5): 967–76

Royal College of Nursing (2006) *Clinical Practice Guidelines: the Nursing Management of Patients with Venous Leg Ulcers*. Available at: https://gneaupp.info/wp-content/uploads/2014/12/The-nursing-management-of-patients-with-venous-leg-ulcers.pdf (accessed 16.01.2020)

Tuner J, Hode L (2002) *Laser Therapy. Clinical Practice and Scientific Background.* Prima Books, Grängesberg, Sweden:

Werdin F, Mayer Tenenhaus, Hans-Oliver Rennekampff (2008) Chronic wound care. *Lancet* 72(9653): 1860–2

Young MJ, Breddy JL, Veves A, et al (1994) The prediction of diabetic neuropathic foot ulceration using vibration perception thresholds: a prospective study. *Diabetes Care* 17(6): 557–60