

## EFFECTS OF PULSED ELECTROMAGNETIC FIELD VERSUS LOW LEVEL LASER THERAPY ON COLONY COUNT ON VENOUS ULCERATION

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### ABSTRACT

The purpose of this study was to evaluate the efficacy of pulsed electromagnetic field (PEMF) versus low level laser therapy (LLLT) on venous ulcer by using colony count (CC). Thirty patients with venous ulcer were participated in the present study and randomly divided into two equal groups; group A, which received pulsed electromagnetic field with their traditional medical treatment and group B, which was treated with low level laser therapy with traditional medical treatment. CC had been measured before treatment and after 6 weeks from the beginning of treatment for both groups. Results showed that there were NO significant differences in CC in LLLT group versus PEMF group.

**KEY WORDS:** Venous Ulcer, Pulsed Electromagnetic Field, Low Level Laser Therapy, And Colony Count.

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### INTRODUCTION

Venous ulcers are coetaneous wounds that result from chronic venous insufficiency; they are responsible for approximately 80%–85% of all ulcers.

That occur on the lower limbs. Venous ulcers represent a serious public health problem throughout the world. These wounds are difficult to heal because of their recurrence, high risk of infection and the high cost of treatment. These ulcers cause significant morbidity, pain, work productivity loss and decrease the quality of life in affected patients [1].

Chronic venous ulceration is defined as a break in the skin below the knee that has not healed within two weeks [2].

Venous ulcers are a common health concern for many older adults. The estimates of the prevalence of venous ulcers vary; however, the

prevalence of venous ulcers in a hospitalized population was higher at above 6% [3]. Venous stasis commonly presents as a dull ache or pain in the lower extremities, swelling that subsides with elevation, eczematous changes of the surrounding skin, and varicose veins. Venous ulcers often occur over bony prominences, particularly the gaiter area (over the medial malleolus) [4].

The pathophysiology of venous ulceration is contentious, and detailed consideration of the microcirculatory changes leading to tissue necrosis [5].

Patients with venous leg ulcers are best managed in the community for two reasons:[6].

- Maintenance of independence and mobility is important in this elderly population.
- The number of patients would overwhelm hospital services.

Venous ulcer treatment has two objectives: to heal the lesion and to avoid a recurrence. Wound care is very important. Surgical removal of fibrin and devitalized tissue from the base and the borders of the ulcer is needed. This procedure reduces the risk of infection and favors healing. Afterwards the wound must be kept humid by using occlusive dressing, for instance hydrocolloid dressings, hydro gels and alginate, which favor autolytic debridement [7].

It is believed that pulsed electromagnetic fields and low level laser therapy play their roles in wound healing. Pulsed electromagnetic field therapy is a physical therapy modality that has been widely used for increasing permeability of the cell membrane and blood circulation, increasing oxygen supply, stimulating healing process and epithelialization of the injured tissues, accelerating bone healing, improving fibroblastic as well as osteoblastic activities, plus its anti-inflammatory and analgesic effects[8].

The use of low-frequency pulsed electromagnetic field (PEMF) for selective control of cellular function has given biology and medicine a new dimension, so it is called the electromagnetic medicine and there are successful results in its use in the treatment of un-united fractures[9].

Low level laser therapy (LLLT), also called soft laser, is known to supply direct bio stimulative light energy to body cells. The absorbed laser energy stimulates molecules and atoms of cells but does not cause rapid or significant increase in tissue temperature[10].

Low level laser therapy involves interaction of a low power monochromatic light with biological systems in order to initiate biomodulative effects. Photon induced biomodulation (i.e., photobiomodulation) was the subject of several studies over the past few years. LILT exposures have produced both stimulatory and inhibitory effects in-vitro and in-vivo, depending on the energy dose [11]. The Purposes of this study were to: evaluate the therapeutic efficacy of PEMF and LLLT, detect which one of them would be more effective and better than the other in accelerating healing of venous ulcer and share in designing the optimal and ideal protocol for the treatment.

## SUBJECTS, MATERIALS AND METHODS

**Subjects:** Thirty patients (male and female) with venous ulcer recruited in this study. Their age will range from 25-40 years; they will be free from any other diseases that may affect or influence the results. They were selected from the outpatient clinic Kaser El Aini hospital .

Patients were divided randomly into 2 groups of equal numbers (A and B), 15 in each group .

**Group A:** It included 15 venous ulcer patients. They will receive the application of pulsed electromagnetic field in addition to traditional medical treatment +nursing care.

**Group B:** This group included 15 patients with venous ulcer and they will receive the application of low level laser therapy in addition to traditional medical treatment +nursing care.

The treatment was conducted 3 times / week for 6 weeks.

**Inclusive criteria:** Patients will of both sexes (males and females). Their ages will range from 25-40 years. Patients have venous ulcer (stagell). Patients entered the study after having their informed consent. All patients were approximately the same age. All patients received a routine of medical treatment drugs and nursing care. The treatment was conducted 3 times / week for 6 weeks.

**Exclusive Criteria:** Patients who have contraindications for pulsed electromagnetic field. Patients who have other skin diseases which may affect the treatment outcomes. Concomitant psychiatric disorders e.g. major depression, anxiety or personality disorders.

## Equipment

### Measuring equipment

**Colony count:** Is a unit used to estimate the number of viable bacteria. Viable is defined as the ability to multi play via binary fission under the controlled conditions [13].

### Procedures of study

#### Measurement Procedures:

**Colony count:** The planktonic cells were removed from each well of micro plates, vortexed, serially diluted (1:10) on TSB, plated on Triptych Soy Agar (TSA) (Oxoid, Milan, Italy) and incubated aerobically for 24 hours at 37°C;

the bio-filmss were rinsed with PBS; then, 100 µl of PBS was added to each well to collect cells using a cell scraper Each sample was then vortexed, serially diluted (1:10) on TSB, plated on TSA and incubated aerobically for 24 hours at 37æ% C. The concentration was calculated as mean value of five replicates for both mono and polymicrobial bioûlms and reported as CFU/ml [15].

**Therapeutic Procedures:** All patients in both group (A) and (B) were received the same traditional medical treatment.

Therapeutic Procedure for Group A (PEMF)

**Group A:** This group included 15 patients with venous ulcer and they will receive the application of pulsed electromagnetic field in addition to traditional medical treatment +nursing care. The treatment was conducted for 25 minute each patient 3 times /week for 6 weeks.

**Therapeutic Procedure for Group B(LLLT):** This group included 15 patients with venous ulcer and they will receive the application of low level laser therapy +traditional medical treatment +nursing care. The treatment was conducted for 18 minute each patient 3 times /week for 6 weeks.

**Statistical Analysis:** Statistical analysis was conducted using SPSS for windows, version 18 (SPSS, Inc., Chicago, IL). The current test involved two independent variables. The first one was the tested group that had two levels (group A receiving magnetic therapy and group B receiving laser therapy). The second one was the treatment periods, which had two levels (pre and post). The dependent variable (colony account). Normality test of data using Shapiro-Wilk test was used, that reflect the data was not normally distributed for colony account, so non parametric statistical tests in the form of (Wilcoxon Signed Rank tests) was used to compare between “pre” and “post” treatment for each group and “Mann-Whitney tests” was conducted to compare colony account between both groups in the “pre” and “post” treatment. The alpha level was set at 0.05.

## RESULTS

**Baseline and demographic data:** There were no statistically significant differences (P>0.05) between subjects in both groups concerning age

(Table 1). Also, Chi square revealed there was no significant differences between both groups in sex distribution (p>0.05) (Table 1).

**Table 1:** Descriptive statistics and unpaired t-tests for the mean age of the patients with for both groups.

Items	Group A	Group B	Comparison		Level of significant
	Mean ± SD	Mean ± SD	t-value	P-value	
Age (yrs)	55.13±2.64	54.66±3.59	0.405	0.689	NS

\*SD: standard deviation, P: probability, S: significance, NS: non-significant.

**Table 2:** Distribution of sex in both groups.

	Group A		Group B		Chi-Square	
	Females	Males	Females	Males	χ <sup>2</sup>	P-value
NO.%	11\ (73.3%)	4\ (26.7%)	6\ (40%)	9\ (60%)	3.394	0.065
Total.	15 (100%)		15 (100%)			

\*Significant at the alpha level (p < 0.05).

## Colony account

**Within groups:** The median score of colony account in the “pre “ and “post” treatment were 100000 and 1000 respectively in the group A. “ statistical analysis using the non parametric Wilcoxon Signed Rank tests” revealed that there was a significant decrease in the colony account in the “post” test in the group A (p < 0.05). Meanwhile, the median score of colony account in the “pre” and “post” tests were 100000 and 1000 respectively in the group B. statistical analysis using the non parametric “Wilcoxon Signed Rank tests” revealed that there was a significant decrease in the colony account in the “post” test in the group B (p < 0.05).

**Between groups:** Considering the effect of the tested group (first independent variable) on colony account, “ Mann-Whitney tests” revealed that the median score of the “pre” test between both groups revealed that there was no significant difference between the both groups ( p>0.05). as well as, the median score of the “post” test between both groups showed there was no significant difference between the both groups ( p>0.05) (Table(3)).

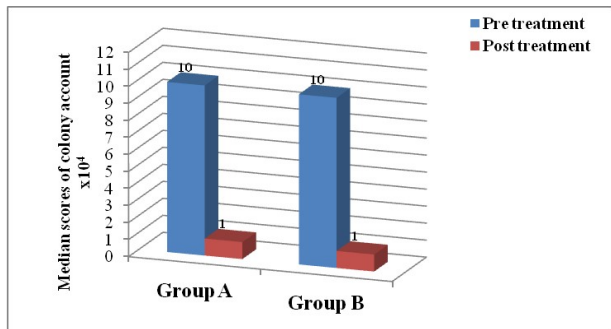
**Table 3:** Median score, U, Z, and P values of the colony account pre and post treatment in both group.

Colony account	Median score		Z-value	P- value
	pre	post		
Group A	100000	1000	-3.305	*0.001
Group B	100000	1000	-3.415	*0.001
U-value	96	80		
Z-value	-0.721	-1.39		
P- value	0.512	0.187		

\*Significant level is set at alpha level <0.05



**Fig. 1:** Median scores of colony account pre and post treatment between both groups.



## DISCUSSION

Pulsed electromagnetic field (PEMF) is a very effective biophysical modality used in physical therapy and utilized for acceleration therapeutic purposes as well as in the area of diagnoses. Published reports indicated that the PEMF stimulators may promote wound healing, but this depends on parameters such as frequency, intensity, exposure time and orientation of PEMF. Data from several small, randomized controlled trials suggest that pulsed high-frequency electromagnetic energy may promote healing of chronic ulcers and soft tissue injuries [16].

The results of this study concerning the effect of PEMF for promoting healing of dermal burn in humans confirm the observations of the following authors; Callaghan, studying wound healing under the influence of PEMF and he stated that PEMF accelerates wound closure and increases endothelial cell proliferation. Strauch et al., examined the effect of pulsing electromagnetic fields on the biomechanical strength of rat Achilles' tendons at 3 weeks after transection and repair. He showed that electromagnetic field enhanced Ca (2+) binding in the growth factor cascades involved in tissue healing, and achieved a marked increase of tensile strength at the repair site. They investigated the effects of extremely-low-frequency PEMFs on the synthesis of epidermal collagen. It was concluded that at 25 Hz under a field setting of 2 mt for the duration of 8 days, stimulation of skin at 2.5 h/day would cause increase in collagen synthesis in rat skin. In addition to the previous Athanasiou<sup>25</sup> investigated the effects of short duration PEMF on secondary healing of full thickness skin wounds in a rat model. According to their findings with the planimetry, there was a statistically significant acceleration of the heal-

ing rate for the first 9 days in the experimental group, whereas a qualitative improvement of healing progress was identified by histological examination at all-time points, compared to the control group.

It is believed that electromagnetic fields play its role in healing by guiding cellular movements that close wounds. It has been shown that fields can affect orientation, migration and proliferation of cells such as fibroblasts, myofibroblasts and keratinocytes, which are of key importance in healing [17].

Magnetic therapy can help the body ward off such microbial invaders as viruses, bacteria, and fungi. It achieves this, in part, by increasing immune function through the oxygenation of white corpuscles, an important part of the immune system's arsenal. A magnetic field can also function like an antibiotic by lowering acidity, with the result that microorganisms have a more difficult time surviving. In addition, hormonal production is regulated, altering enzymatic activity and biochemical messengers of the immune system. For example, the pineal gland is one large electromagnetic entity. The net effect is to augment the body's natural ability to resist a variety of germs [18].

Low level laser therapy (LLLT), as a noninvasive, pain-free method with minor side effects, has been considered as a possible treatment option for venous ulcer.

Biometrical and histological analysis indicated "faster lesion contraction showing quicker re-epithelization and reformed connective tissue with more organized collagen fibers" in irradiated wounds. Laser therapy reduces the inflammatory reaction and provokes a greater proliferation of myofibroblasts in experimental cutaneous wounds [19].

Stimulation of cell division and cell growth of fibroblasts plays an important role in wound healing. Different wavelengths of low-intensity laser irradiation (LILI) have been tested on cellular migration, viability, and proliferation in diabetic wounded and unwounded human skin fibroblast cells. Cells irradiated at 632,8 nm showed a higher degree of haptotaxis and migration as well as ATP luminescence as compared to cells irradiated at 830 nm. These

results may lead to the conclusion that diabetic wounded cells have more benefit in wound healing from irradiation in the visible range than in the infrared range. However, since near-infrared light has a deeper penetration rate than visible red light, deeper ulcers in vivo might require the use of near infrared laser therapy.

Red light laser (630 nm) as well as blue light Laser (470 nm) can improve perfusion by release of nitric oxide from nitrosyl complexes with hemoglobin, enhanced epithelialization, and elevated keratin-10 mRNA level. Blue light also facilitated the recovery of mitochondria inhibited by NO gas by release of NO from mitochondrial complexes, so an improved wound healing via the NO pathway induces endothelial cell migration by activating growth factors, resulting in an increase keratin expression.

Anti-inflammatory effects of laser therapy can be explained by inhibition of prostaglandin, interleukin, and cytokine in cell and animal models [19].

In vitro experiments with a low-power laser (415 nm) showed a direct antibacterial effect on *S. aureus* and *E. coli* by induction of ROS. LLLT also can increase the diameter and blood flow velocity of the peripheral arterioles and can enhance the microcirculation.

On evaluating the effect of Helium-Neon (He-Ne) laser irradiation on the viability, adherence, phagocytic activity of the polymorphonuclears and monocytes. Also the level of metabolic processes in phagocytosing blood cells, monocytes and polymorphonuclears was estimated phagocytic activity was examined by classical method using microspherical hydrophilic particles. Small dosage of the laser irradiation increases the phagocytic activity, and after higher laser irradiation phagocytic activity is decreased. Also the changes of the level of metabolic processes in the phagocytizing cells are very similar with the changes of phagocytic activity. The viability of the blood cells examined after higher laser irradiation is gradually decreased [19].

**Conflicts of interest: None**

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