BIOSCIENCES BIOTECHNOLOGY RESEARCH ASIA, March 2015. Vol. **12**(Spl. Edn. 1), p. 185-195

Electromagnetic Fields and Ultrasound Waves in Wound Treatment: A Comparative Review of Therapeutic Outcomes

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DOI: http://dx.doi.org/10.13005/bbra/1622

(Received: 05 February 2015; accepted: 10 March 2015)

Different techniques have been proposed for wound treatment including laser, electric therapy, EMF therapy (EMFT), ultrasound (US), magnetic field, etc. Recently, EMFs and US have shown great potential for wound treatment. Despite of several evidences about the beneficial effects of EMF and US in wound healing, no definite dose-response existed on the clinical trials applications of these techniques. In addition, the therapeutic outcomes of these techniques can be influenced by different parameters such as intrinsic properties of living organs as well as physical parameters of stimulations. For further development of EMF and US based treatments for wound healing it is necessary to develop more quantitative assessments for wound healing. This paper aims to provide an comparatively overview of the EMFT and US based techniques for wound healing, and highlight their main procedures and underpinning physical principles exerting therapeutic outcomes background. In addition, it compares their efficacies on wound healing.

Key words: Electromagnetic fields, Ultrasound Waves, Wound treatment, Therapeutic outcomes.

Wounds are divided into two groups; acute and chronic. Acute wounds are usually treated by direct union whereas treating of chronic wounds remains for a long time. If treating procedure of a wound prolongs almost up to six weeks, it is considered a chronic wound[1].

Various techniques have been developed for healing of wounds including conventional medications, pressure relieving beds, cushions, medicinal plants. These techniques are commonly utilized for prevention and healing of pressure wounds. Due to different traditional methods for wound healing have some disadvantages such as high cost and elimination of reimbursement for various wounds like burns, venous leg ulcers, scientists pursued new ways of medical treatment of wounds. in last 10 years, various methods were employed for chronic and acute wounds treatment such as laser, electricity magnetic, ultrasound(US), light and electromagnetic field (EMF)[2-10].

Recently, EMF and electric field stimulations have gained much attention for treatment healing wounds which are non-healing. In this area, electrical stimulation was mainly applied for accelerate healing of decubitus ulcers and vein insufficiency[11].

The utilization of invasive electric field for treatment of bones and nerves has been developed for patients[12-16]. Recently, Noninvasive EMF therapy machines have appeared as alternatives to invasive electric field therapy. In this technique, a magnetic field is generated. Afterward this induces an electrical current in around of the desired area.

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Principle concept of the effect of electric field and EMF is that cells which are involving in wound healing are electrically charged, so applying endogenous electricity lead to facilitation cellular migration to the wound area and other wound healing procedures[17]. With respect to this theory, during the delaying of wound healing process, applying external electrical stimulation can produce bioelectric effects and lead to progression of wound healing procedure. Also, applying EMF stimulation may interact with the wound currents or with related signal transduction processes[18], therefore re-stimulating can retard healing of wound. For example, Carley *et al* (1985) showed that applying direct currents in the range of 200-800 ¼A caused to accelerate wound healing[19].

Addition to EMF, Ultrasound is one of other methods which acquired promising results for treatment wound. Techniques which are based on US have great advantages as compared with conventional and other alternative techniques. As compared with other models, US waves can penetrate into the beyond of the wound and reached to more deep tissues. In addition, US waves can be steered and focused significantly more than other methods.

The history of researches on the interactions between high frequency US waves and living organs and tissues draw back to 70 years ago[20]. As long as the realizing of the therapeutic potential of US energy, several US based techniques have been employed for treatment of different disorders such as skin wounds, malignant tumors[21, 22]. In this regard, many trials have demonstrated different physiological effects of US on living tissues and organs[23-26]. Similarly there are great evidence to express the therapeutic effects of these waves in the soft tissue disorders[27-29].

In therapeutic applications of US, non-ionizing radiation is delivered to the desired tissues in the mechanical waves form. This delivering caused to creation heats the tissue. Two important factors can affect therapeutic efficacies of US including dose (W/cm^2 time) and dosage (frequency of application, series)[30]. Therapeutic US is commonly applied at two frequencies of 1.0 MHz and 3.0 MHz and it mostly produced heating modality which can reach depths of 5 cm and more below the surface of body. US waves can apply in the model of pulsed or continuous waves to produce thermal and non-thermal effects[30]. It is clear that selection of correct parameters of US techniques are highly depends on the required effects and location of the tissue.

Recent years, it was reported that high frequency US can be used for some treatment applications such as treatment of tendon injuries and relief of the short-term pain[31-33]. In addition, it was revealed that US can facilitate healing of some acute bone fractures, venous and surgical incisions and pressure ulcers[31, 34, 35]. In applying US, if inappropriate parameters are used, it may lead to burning or damaging the endothelial[36,37]. Utilization of high frequency US in medicine may have the risk of tissue heating. With the advancement of technology, mercantile modalities which are based on low frequency US were supplied consequently, scientists have performed great effort to use optimal US parameters.

This paper aims to provide an overview of the EMFT and US comprehensive account of methods of wound healing, and highlight their main procedures and their background. In addition, it compares their efficacies on wound healing.

Electromagnetic Field and Wound Healing

Cellular interactions of Electromagnetic field (EMF) have attracted the attention researchers in various fields such as embryology, molecular biology and, physiology. It was demonstrated that Pulsed electromagnetic fields (PEMFs) could be applied as an adjunctive or alternative treatment of both delayed union fractures and chronic wounds. Among different EMFs gradients, low frequency EMF has great potential for treatment of wide variety of diseases such as diseases[38], cancer treatment[39], wounds[40-42]. Many researches have been reported the effects of EMFs on organs, tissues, and cells[40, 43, 44]. EMF can affect several biological systems such as bone, skin and hematologic[45, 46]. In physical medicine, application of low frequency EMF has been used for healing of wounds and certain musculotendinous lesions. It is worth mentioning that the base of majority of these clinical studies is on experience rather than scientific evidence[47, 48].

The devices which are utilized for these applications applied an external, non-invasive PEMF to create short electrical current in wound or injured tissue without generating heat or reacting with muscle function or nerve. Increased perception of PEMF therapy has led to development of novel PEMF devices with greater efficiency. With the advancement of knowledge in EMF researchers started investigating therapeutic application of EMF for wound. For example, Foley-Nolan *et al* (1992) revealed that applying PEMF could have significant benefit for management of various soft tissue injuries particularly when treatment was conducted in the acute phase[49, 50] Junger *et al* (2008) evaluated the effect of low frequency pulsed current on healingin chronic venous ulcers. They divided the patients into 2 groups including placebo and treatment (treated with the low-frequency pulsed). Their results showed no significant differences between both group in reduction of ulcer area, but in treatment group, pain reduction was rather than placebo group[51].

Aziz *et al* (2010) investigated the effect of EMF on wound healing. Their results could not confirm significant effects of applying EMF on pressure ulcers healing[52]. In 2005, a Blue Cross Blue Shield Technology reported that there is not enough evidence to judge about the efficacy of EMF as adjunct treatments for wound healing[53].

These findings were not reliable and required verification through a controlled and large sample studies.

Strategies of EMFT for Wound Healing

Almost all of electromagnetic devices use time varying or pulsed signals, some of which modulate a carrier frequency (usually 27.12 MHz). A further distinction between heating effects of pulsed radio frequency is associated to the energy which they deliver to the tissue. In this regard, commercially EMF devices can evaluate device average or peak power but these cannot determine the field strength which is delivered to the target tissues. Majority of commercially devices generated same pulse width and shape[65-95] ¼ sec), with the power per pulse reined by varying pulse amplitude.

EMF devices that act in non-thermal and thermal modality can allow both variable pulse width and rates (Magnatherm, 700-7000 pps), while other devices cannot control these feature. It is thought that tissue thermal effects are minimized by using low duty cycles. It is assumed that high power single, short pulses lead to dissipation the heat during a much longer off-time between consecutive pulses.

The effective parameters for EMF include generated power, stimulation frequency, pulse width, duplication rate and duty cycle, carrier frequency, prevalent magnitude, and intensity of magnetic field. In addition, some differences occurred due to specific features of the excitation patterns, for example whether stimulation is sinusoidal or not, continuous or pulsed, biphasic or monophasic, symmetrical or asymmetrical, and high voltage or low voltage[54,55]. Due to this wide variety of excitation parameters, the relationship between optimal physical parameters and wound healing is hardly achieved. However, using pulse radio frequency EMF (PREMF), with its inductive coupling to tissue caused to creation a more uniform and predictable electromagnetic field signal in the target tissue as compared with the signal which acquired with surface contact electrodes[56]. Also, it was demonstrated that the broad spectrum of PREMF provide more possibilities for coupling of the field in order to produce effects in a wider range; however exact biological processes are not determined.

Effect of EMF on cell proliferation and cell cycle

Cell Proliferation

The exact mechanism of EMF stimulation of cell proliferation and wound healing has not been determined. Therefore, to determine the effect of EMF on cell proliferation, some scientists reported that some EMF characteristics signals lead to increase of cell proliferation of keratinocytes [47, 48, 57-60]. In this regard, Manni *et al* (2002) found that applying EMF at 50 Hz could increase the human keratinocyte cell growth[47]. As well as some other research demonstrated that applying low frequency EMF could enhance the proliferation of human keratinocyte[58, 60]. Also it was reported that the keratinocytes proliferation from cutaneous stem cells are necessary for wound healing[61, 62]. Epidermal stem cells can heal damaged tissues by stimulating mobilizing signals[63]. For instance, human epidermal stem cell proliferation can be enhanced by expression of epidermal growth factor receptor (EGFR)[63]. As well as it was demonstrated that a magnetic field at Hz produces EGFR clustering[64]. Zhang *et al (2013*) surveyed the effect of low frequency EMF on proliferation of human epidermal stem cell (hESC). They demonstrated that low frequency EMF modulated the hESC proliferation[63]. Similarly, some other studies reported that applying EMF could increase the keratinocyte growth[47, 57]. Huo *et al* (2010) demonstrated that noninvasive EMFs affect the cell proliferation seem keratinocyte-specific without any effect on dermal fibroblasts[60].

Cell Cycle

The cell cycle has three main phases including G0/G1 phase, S phase, and G2/M phase. Due to the low frequency EMF at low frequency can augment cell proliferation; a lot of researches were performed to find the effect of EMF on the cell cycle, despite some of these results were controversial[65-67]. These results are not enough for clarify the relationship between cell proliferation which occurred by applying EMF and distribution of cell cycle[68]. Commonly, in order to determine the potential of cell proliferation, the proportion of S-phase cells is considered. It was reported that applying low frequency EMF caused to significantly decrease of the percentage of cells which are in the G1 phase while it can increase the number of those in the S phase, affect the cell cycle, enhance cell growth and increase the proportion of cells[63]. It was reported that low frequency EMF did not promote apoptosis[63]. Due to absence of high quality trials, EMF therapy remained investigational for wounds healing[53].

Characteristics of Therapeutic US

US waves are generated by piezoelectric effect. Indeed, in transducer of US the electrical energy is converted to acoustic energy. These waves are transformed in tissue by diffusion and vibration of molecules. The US energy is lost due to attenuation during passage through the tissue. There are several characteristics such as frequency, intensity, amplitude, focus, and beam uniformity for US waves. This features determine the amount of energy attained by a particular site[69].

The therapeutic US waves have the frequency in the range of between 0.75 and 3MHz. More penetration depth and less focused are two important features of low frequency US waves. Therefore the low frequency waves are suitable for deeper injuries and fatty patients[70, 71]. In vitro and in vivo studies have demonstrated that the therapeutic effects of low frequency US are depend on exposure levels. For example low levels are useful whereas the cell death can be induced by High intensities. Also high intensity can induce delay healing[72, 73] and low level can accelerate the rate of repair and reduce the time of healing[74, 75].

The main feature of tissue that can influence on interaction of US with matter is acoustic impedance. The less portion of the US wave will transmit through the interface between two tissues with large acoustic impedance difference.

Dosage of US can also be altered by changing of wave amplitude and intensity. Moreover, therapeutic US can be generated in pulse or continuous form. The thermal effects of continuous US are higher than pulse form. The both form induce non-thermal effect at low intensity[76].

Biological effects of US

In classifying US, high power, high frequency US is considers as US at 0.5-10 MHz and with intensity up to 1500W/cm^2 while low power, low frequency US is described as an US of 20-120 kHz and 0.05-1.0 W/cm^2. When low frequency, low intensity US is applied, it chiefly reflected in the wound surface or skin and only little fractions of the US energy are delivered to the profound tissue layers. The main effect in this condition is mechanical effect. In contrast, the main effect in applying high frequency US is combination of mechanical and thermal effects.

Different researches indicated beneficial effects of low frequency US on wound healing which are dependent on the exposure levels. In this frequency, high intensities lead to cell death, whereas at low intensities useful effects are emerged.

Therapeutic effects of US through the range of intensity are achieved by both thermal and non-thermal interaction mechanisms. At low intensities, acoustic flows are highlighted; while at considerable effects of US at higher levels are thermal and acoustic cavitations. Despite of the beneficial therapeutic effects of US are clinically examined, the exact mechanism of US are not thoroughly understood.

US techniques could apply as superseded diathermy therapy in competition with radiofrequency and microwave techniques to create middle heating. With improving the understanding of the therapeutic mechanisms of US, therapeutic models are trying to utilize any beneficial non-thermal mechanisms which may be existed[77].

In order to optimize the therapeutic efficiency of US for wound, understanding the exact mechanism of US waves on desired tissue is required. Some studies which performed on the therapeutic US have demonstrated no a dose-response relationship[78, 79]. Nevertheless, Laakso *et al* (2002) revealed that the spatial average temporal average dosage with the range of 0.5 W/ cm^2 to 3 W/cm^2 caused to minimize side effects[79]. Similarly, applying US with the dosage of 1 W/ cm^2 to 1.5 W/cm^2 had considerable therapeutic effects[80-82].

Strong evidences confirmed that US with high intensity can hurt bone or delay healing[72, 73] and low intensities facilitated repair and reduced the treatment period[74, 75]. Also pulsed US with low intensity revealed great potential to heal some disorders such as bone fracture, pain relieving and osteoporosis[83, 84].

As mentioned above, there are various evidences to confirm therapeutic effect of very low intensity US on bone and soft tissue treating. At low intensities, thermal effects are not responsible action mechanism. Applying US causes to increase the penetration of pharmacologically-active drugs through the skin which is called sonophoresis or phonophoresis[85, 86]. Comparing with high frequency, US at low frequencies have more therapeutic potential for wound healing.

It is important to note that acoustic streaming and cavitation are preferred in aqueous in vitro environment which is different with US exposure. This means that the action mechanisms of acoustic streaming and cavitation are different into two mediums.

It was reported that US at low frequency can increase penetration into the skull. US with high intensity enhance the platelet and fibrin deposition. Nilsson *et al* (1995) revealed that US at 0.5–1 W/cm2 created clot lysis and US at 4 W/ cm^2 created fewer clot lysis as compared with the attendance of fibrinolytic factors[87].

Effect of US during different stages of wound healing

Applying US in first phase of healing: Although US causes no a direct anti-inflammatory effect, it seems that applying US during the primary 'inflammatory' phase of repairing tissue increase the speed of this phase.

'Proliferative' stage is the second healing phase. During this step, this stage includes migration the cells to the injury area, forming the granulation tissue and also beginning to divide. As well as collagen is created by fibroblasts. In this regard, US can repair fibroblasts and epithelium. This repairing increases collagen synthesis[88-90].

'Remodeling' is the last phase of tissue repairing. It was explained that applying US have beneficial potential to wounds healing.

Recent findings of clinical experiments as well as in vivo and in vitro studies have reported that US treatment can accelerates various wounds healing[91-93] .In addition, US at low frequency has been applied for treat burn wounds[94, 95]. In addition to these effects, since US can generate and diffuse nitric oxide, it helps to palliate the pain during the process of wound healing[96, 97].

Physiological Effects of US

The energy of US waves generates a mechanical pressure wave in soft tissue. Two main processes are initiated by pressure wave: in the firs process, microscopic bubbles are produced in living tissues. This process is associated with changes in cell membrane and intracellular activity. Acoustic streaming, bubble formation and microstreaming are three examples of cell membrane distortion mechanisms. The second process is related to thermal and non-thermal physical effects in tissues. Non-thermal effects can be happened with or without thermal effects. Thermal effects can be achieved when the tissue temperature increases to 40–45 °C for at least 5 min. Thermal effects of US can increase the blood flow, extensibility of collagen and pro inflammatory response in tissue[98-99]. This phenomenon can be divided in two categories: stable and unstable. Stable cavitation that need short pulses(At least, 1000 cycles at 1 MHz), is occurred when bubbles expand and contract, without growing to critical size but unstable cavitation does not form in therapeutic range of US intensity in healthy tissues except in lungs and intestines[100].

The changing of mechanical pressure within the US field also can induce Acoustic microstreaming. The function and permeability of cell membrane may alter by Microstreaming[101]. This can be used for enhancing tissue repair[102].

Some in vitro studies have reported the

role of cavitation and microstreaming as stimulates of fibroblast repair and collagen synthesis[23-25, 103], regeneration of tissue[24] and bone healing[75].

The recovery rate of wound tissue can accelerate by US. This ability is related to several mechanisms of action of US, including enhancing the fibrinolysis rate[26, 104], stimulating macrophage derived fibroblast mitogenic factors[105], escalating fibroblast recruitment[21], increasing angiogenesis[106], accelerating matrix synthesis[103], formation more dense collagen fibrils[107] and improving tissue tensile strength. These interactions can modulate inflamed tissues[25, 108, 109].

Moreover, the thermal effect of US can be employed for palliation of pain and muscle spasm. This ability in combination with stretching practice can provide the optimal tissue length[110]. Elis and coworkers have applied the thermal doses of US for Lengthening the ligament of normal knees[111].

US also can enhance vasodilatation, stimulates of angiogenesis, increase release of growth factors and greater amounts of high-quality collagen. The healing is accelerated by these cellular effects[112, 113]

Ultrasound and Wound Healing

According to in vitro and in vivo studies, ultrasound in two separate phases will lead to wound healing: Inflammatory Phase and Proliferative Phase

Inflammatory Phase

The non-thermal effects of US induce mast cells degranulation. Histamine as a chemical mediator is released from mast cells. Therefore the absorption of neutrophils and monocytes are increased in the injured site. The rate of acute inflammatory phase is accelerated. Because of this, the wound healing is promoted[21, 105, 106].

Proliferative phase

In this phase, after interaction of continuous US with tissue, fibroblasts release the collagen. Indeed, this event is due to thermal effect of US. Moreover, the extensibility of collagen, circulation, pain threshold, enzymatic activity, permeability of cell membrane and acceleration of nerve conduction are enhanced by US[114]. As a result, the healing of wound is due to improving circulation[115].

CONCLUSION

The present study has overviewed the most current techniques based on EMFs and US for wound healing and compared their efficacy. Furthermore, the proposed mechanisms of actions for these methods have been discussed.

There are different parameters which affect the therapeutic performance EMFT such as electrical intrinsic properties of living organs as well as physical parameters of stimulations.

Applying US in appropriate parameters can be beneficial for some wounds as alternative and adjunctive treatment options. The mechanical forces induced by US energy at molecular and cellular level show significant beneficial efficacies on the wound healing procedure such as decreasing bacterial activities of pathogens present in the wound.

Addition to accelerating the wound healing rate of wounds, low frequency US has great potential for early treatment for injuries which are suspected deep-tissue. Many researches have shown therapeutic effects of US in several wounds. Up to now the exact dose-response for therapeutic applications of US in different wounds is not determined. With respect to promising results of US in healing of different wounds, it is expected that US will be a novel standard method for wound healing.

For development of EMF and US techniques for wound healing it is necessary to examine more quantitative evaluations for wound healing and also performing more studies on the efficacies of US and EMF techniques on wound healing process.

ACKNOWLEDGEMENT

The present study was financially supported by Ahvaz Jundishapur University of Medical Sciences (Grant No.: U-93190).

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