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Electromagnetic Fields in the Treatment of Wound: A Review of Current Techniques and Future Perspective

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Electromagnetic fields (EMFs) have shown a promising potential for treatment of different wounds. Different techniques have been proposed for wound treatment including electric current therapy (ET), EMF therapy (EMFT), static magnetic field, and combined magnetic field. The present study reviews the most current EMF based methods for wound treatments and compare their efficiency for each wound. In addition the proposed mechanisms of action of these techniques were reviewed. Among different techniques, ET shows more promising effects on wounds. Furthermore, different parameters influence the therapeutic performance of ET and EMFT including electrical intrinsic properties of living organs as well as physical parameters of stimulations. For further development of EMF based treatments for wound it is necessary to develop more quantitative assessments for wound healing.

Key words: Electric current therapy, Wound treatment, Electromagnetic field, Mechanism of action.

Wound healing, a complex tissue repairing process, is a continuing challenge in rehabilitation medicine. Although several recent advances in comprehending its basic principles have been made, problems in wound healing have remained unchanged with significant morbidity and mortality[1]. The usage of electric current and electromagnetic field (EMF) stimulation to enhance wound healing is not new. The pioneer clinical studies originated in late 1960s [2]. A large number of studies have been performed on the wound healing speed, attainment of normal breaking power, scar formation and hypertrophic scar and keloid formation. The understanding of the biological and pathologic events in wound healing has led to three areas of treatment that are currently indicated for the treatment of chronic wounds in the clinic practice[3]: grow factors, tissue engineered skin, and physical devices. Despite the wide interest in growth factors, cytokine biology and their potential in terms of wound healing, clinical trials aiming to accelerate chronic wound healing in most cases have been disappointing[4, 5]. However, several studies have demonstrated that the application of growth factors may lead to the increased speed of cutaneous wound healing in animal models[6]. Not only does tissue engineered skin offer the possibility of creating physiologically compatible human skin, but it also is successfully used on burn wounds to prevent bacterial infection and gives wounds the chance to be healed by normal reparative processes. Unlike in patients with burned wounds, the condition in patients with chronic wounds results from underlying diseases; therefore, closing wound with skin substitutes would not be sufficient to initiate the wound healing[7].

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The endogenous electric fields (EFs) effect on wound healing has also been proposed as a main indication directing the cells to migrate in wound healing process[8-11]. Studies in the last decade have been shown that there is a role for EFs in wound healing[12-18].

Many aspects of the role of EFs in wound healing have been expressed in some recent reviews[12, 18-20]

For soft tissue wound healing, the most common application is for patients with chronic wounds, non- or slow-healing, or otherwise intractable wound to standard treatments.

Interactions of electrical fields are regulator of many basic physiological processes ranging from conformation of molecules within a cell membrane bilayer to the macroscopic mechanical properties of the tissues. Anyway, there is no well-established mechanism that can describe how electric field and electromagnetic field (EMF) applications influence the action of living cells and tissues.

In recent years, electric current and EMF stimulations have become increasingly popular treatment modalities of non-healing wounds. Electrical stimulation was primarily used to accelerate wound healing of decubitus ulcers and vein insufficiency. Studies showed that pressure ulcers react better on electrical stimulation than other types of wounds[21].

At the macroscopic level, naturally applying current loops of about 10 µA have been measured in the human legs[22], and at the microscopic level, membrane function is largely determined by intrinsic electrical processes. Because skinny wounds interrupt normal transepithelial potential within injury sites, developing electric field and injury current is postulated to play an important role in the healing process[23, 24].

The pivotal concept of this theory is that cells involving in wound healing are electrically charged, so that endogenous bioelectricity can facilitate cellular migration to the wound area and might be involved with angiogenesis[25] and other wound healing processes. According to this theory, upon the delaying of wound healing, external electrical stimulation may mimic one or more of the bioelectric effects and help to trigger a renewed healing progression. Furthermore, the externally applied EMF may interact directly with the wound currents or with related signal transduction processes[26], thereby re-stimulating retarded or arrested wound healing. Wound healing acceleration induced by direct currents in the range of 200-800 µA, applied by a portable unit, is an example of such a process[27].

Exogenous extremely low frequency electric fields applied in fixed juxtaposition to their target tissues (animals and cells of eukaryote and prokaryote origins) have been found to initiate mitogenesis-related signals[28].

Alvarez *et al* (1983) was one of the first groups to demonstrate that direct electric currents promote epidermal resurfacing and dermal collagen biosynthetic capacity in partial- thickness wounds in a pig model[29]. The usage of invasive electric field therapy for bones and nerves has been widely embraced as useful for patients[30-34]. Noninvasive EMF therapy machines have appeared recently as alternatives to invasive electric field therapy. However, research behind these products has consisted mostly of anecdotal clinical reports with very few well-controlled laboratory mechanistic studies.

To date, wound studies have typically been performed on skin ulcers induced by arterial or venous dysfunction, diabetes related ulcers, pressure ulcers and surgical and burn wounds.

Human testing and clinical trials have demonstrated the therapeutic properties of some of non-drug methods in triggering the healing process of “stalled” soft tissue ulcers or wounds. Human studies showing a positive privilege of EMFT range from those on a single subject with multiple experimental wounds[35] through (a limited number of) randomized controlled trials.

Animal studies have also indicated therapeutic potentials of ET and EMFT in wound healing[20, 36-38]. However, most of these studies were based on wound models that diverge in one or more important aspects from human “chronic” wounds, those in which repair is stalled and difficult to manage. Yet these are precisely the types of wounds that would most likely benefit from such adjunctive treatments. However, many EMFT-related effects on cells, tissues, and tissue repairing processes have been convincingly shown to occur. The scientific basis for detecting the possible correlation between EMFT and soft tissue wound repair processes is neither complete nor fully accredited. But, based on many specific clinical trials, experiments and cellular observations, a clear linkage between ECT/EMFT and wound healing is strongly suggested.

This paper aims to provide comprehensive review of the ECT and EMFT exhaustive account of methods of wound healing, to sketch their major procedures and their background. Moreover, it aims to compare their effect on wound healing.

Electromagnetic Field and Wound Healing

Electromagnetic field (EMF) gradients have been linked with cellular reactions in a variety of fields including embryology, molecular biology and, physiology. Pulsed electromagnetic fields (PEMFs) have shown characteristics as an adjunctive or alternative therapy for both delayed-union fractures and chronic wounds treatment[39]. Low frequency EMFT has been successfully used for a wide variety of diseases including musculoskeletal diseases[40], cancer treatment[41], neurological disorders[41, 42], wounds[37, 43, 44]. There is an increased public awareness about the potential health hazards of exposure to electromagnetic fields (EMF)[36,38,45]. The effects of magnetic and electric fields on organs, tissues, and cells have been explored during the past 30 years[43, 46, 47]. EMF is reported to influence a wide variety of biological systems such as bone, skin and hematologic. For example, some in vitro studies have showed that EMF affects the cellular physiology of many types of human cells[48, 49]. In physical medicine, EMF of low frequency has clinically been applied for early healing of wounds and certain musculotendinous lesions. However, majority of these clinical studies are based on experience rather than scientific evidence.

The epidermis energy associated with radiation can directly affect the outermost layer of the skin[50, 51]. In this regard, several studies have demonstrated that EMF can modify cell morphology of normal keratinocytes and interfere with their differentiation and cellular adhesion[50, 51,52-54]. These relatively simple devices use an external, non-invasive PEMF to generate short bursts of electrical current in injured tissue without producing heat or interfering with nerve or muscle function. Recently, increased comprehension of the mechanism of action of PEMF therapy has permitted technological advances yielding economical and disposable PEMF devices. Using these devices, PEMF therapy has been broadened to embody the treatment of postoperative pain and edema in both outpatient and home settings offering the physician a more versatile tool for patient management[55]. In noninvasive EMF therapy, a controlled magnetic field is generated which, in turn, induces an electrical current in the surroundings of the target.

Electromagnetic Therapy

In a randomized, double-blinded study, Czyz *et al* (2012) investigated the advantages of electromagnetic energy in eyelid wound healing in 57 patients who underwent upper blepharoplasty. There was no difference in patient pain rating when comparing placebo with the electromagnetic energy patch. Patients reported 6% less edema and 10% less ecchymosis with the active patch eye than in control eye[56]. The authors demonstrated that the use of pulsed electromagnetic energy did not have any significant effect on postoperative pain, edema, or ecchymosis as rated by patients and physicians. They noted a statistically significant reduction in physician-graded erythema for active patch eyes versus placebo[56]. The significance of these conclusions is restricted by an extremely small sample size. These findings suggested that treatment with pulsed electromagnetic field may improve healing rates in venous or pressure ulcers and in the donor site following skin linkage, compared with standard wound care[57, 58].

Another earlier randomized controlled trial failed to find a significant treatment effect of electromagnetic therapy for patients with chronic venous ulcers, although there was a trend toward improved healing in the intervention group[59]. Some scientists such as Foley *et al* (1992) and Pennington *et al* (1993) who controlled earlier randomization examined pulsed electromagnetic energy therapy for management of different types of soft tissue injuries, including whiplash, ankle sprains, and hand/finger lacerations[60, 61]. In these studies, tendencies were found toward significant benefit to the intervention groups with respect to swelling, pain, and mobility, particularly when treatment was applied in the acute phase (within 3 to 4 days of injury). However, aside from examining the same technology, the trials differed so much in the type of injury and treatment protocol that no overall conclusions regarding the efficacy of pulsed electromagnetic energy therapy can be made.

Gupta *et al* (2009) evaluated the effectiveness of pulsed electromagnetic field therapy (PEMF) in the healing of pressure ulcers in patients with neurological disorders in a randomized double blind control trial[62]. Their study included 12 patients (M: F, 9:3) with pressure ulcers who were 12-50 years of age. Six patients with 13 ulcers received PEMF therapy and the remaining 6 patients with 11 ulcers received placebo treatment, for 30 sessions (45 minutes each) using the equipment 'Pulsatron'. The frequency of PEMF was set at 1 Hz with sinusoidal waves and current intensity of 30 mA. Whole body exposure was given to both groups. Bates-Jensen wound assessment tool (BJWAT) score was exploited as the main outcome measure and scores at the end of session were compared with initial scores. Similarly, National Pressure Ulcer Advisory Panel (NPUAP) scores were compared and analyzed as secondary outcome measure. Thirteen ulcers were in stage IV and 11 were in stage III at the start of the study. Significant healing of ulcers was noted. However, when comparing the groups, healing was not significant. A similar trend was noted with NPUAP scores with no significant difference between the treatment and placebo groups at the completion of study. The investigators concluded that no significant difference in pressure ulcer healing was observed between PEMF treatment and placebo group in this study[62].

Junger *et al* (2008) assessed 39 patients in a placebo-controlled, double blind study on the effect of low-frequency pulsed current on healing in chronic venous ulcers. The patients were treated with the low-frequency pulsed or a placebo for 4 months. Ulcer area was reduced in both groups, but pain reduction was better in the treatment group. These findings were not reliable and required verification in a larger study[63].

In another review, Aziz *et al* (2011) assessed the effects of electromagnetic therapy (EMT) on the healing of venous leg ulcers[64]. Three randomized controlled trials (RCTs) of variable quality involving 94 people were included in the review. All the trials compared the use of EMT with sham-EMT. In the two trials that reported healing rates; one small trial (n=44) reported that significantly more ulcers healed in the EMT group than the sham-EMT group; however, this result was not robust to different assumptions about the outcomes of participants who lost to follow up. The second trial that reported numbers of ulcers healed found no significant difference in healing. The third trial was also small (n=31) and reported significantly greater reduction in ulcer size in the EMT group; however, this result may have been influenced by differences in the prognostic profiles of the treatment groups. The authors concluded no high quality evidence that electromagnetic therapy increases the rate of healing of venous leg ulcers, and further research is warranted[64].

In another review, Aziz *et al* (2010) evaluated the effects of EMT on the pressure ulcers healing[65]. Two randomized controlled trials (RCTs), involving 60 contributors, at unclear risk of bias were included in the review. Both trials compared the use of EMT with sham EMT, although one of the trials included a third arm in which only standard therapy was applied. Neither study found a statistically significant difference in complete healing in people treated with EMT compared with those in the control group. The results provided no strong evidence of benefit in using EMT to treat pressure ulcers[65]. However, the possibility of a beneficial or harmful effect cannot be ruled out because there were only two trials, both with methodological limitations and small numbers of participants, and so the authors recommended further research.

In 2005, a Blue Cross Blue Shield Technology Evaluation Center[66] reported that the evidence is not sufficient to allow conclusions on the efficacy of electromagnetic therapy as adjunct treatments for wound healing[67]. Well-designed and well conducted sham placebo-controlled randomized controlled studies are needed that consistently show better results for active treatment over placebo. ET and EMFT for chronic wounds did not meet the TEC criteria (Blue Cross Blue Shield Association, 2005)[67].

Strategies of EMFT for Wound Healing

In electromagnetic instruments, electrode and target tissues need not be exposed to electric and magnetic fields and their associated induced currents. Among electromagnetic devices, all use time varying or pulsed excitation, some of which modulate a carrier frequency, commonly 27.12 MHz. A further distinction among pulsed radio frequency devices is made with respect to their potential tissue heating effects which are related to the energy they deliver to the tissue. Commercially existing EMF devices usually determine device average or peak power but these do not specify the energy or strengths of field delivered to target tissues. Pulse breadth and shape generated by most commercial devices is fixed (65-95 µsec), with the power per pulse generally reined by varying pulse amplitude. Devices that function in non-thermal and thermal ranges may allow both variable pulse width and rates (Magnatherm, 700-7000 pps), whereas other devices do not provide control feature. Tissue thermal effects are thought to be minimized by use of low duty cycles, on the assumption that heating due to high power single, short pulses will be dissipated during a much longer off-time between successive pulses. In general, for EF or EMF, the parameter variants conclude generated power, stimulation frequency, width of pulse, duplication rate and duty cycle, carrier frequency, prevalent magnitude, and intensity of magnetic field. Furthermore, there are variants with respect to specific specifications of the excitation patterns, i.e., whether stimulation is continuous or pulsed, biphasic or monophasic, galvanic or frequency modulated, symmetrical or asymmetrical, sinusoidal or not, and high voltage or low voltage stimulation is used[68, 69]. Because of this wide range of physical excitation parameters, it has been impossible to correlate specific features with wound healing efficacy. However, the use of pulse radio frequency EMF (PREMF), with its inductive coupling to tissue, is said to prepare for a more uniform and predictable electromagnetic field signal in the target tissue than is currently achieved with surface contact electrodes[70]. Thus, the dose of tissue is more reliable. It has also been found that, due to the large spectral span of PREMF, there are more possibilities for coupling of the field to produce effects in a wider range of possible, but as yet unspecified, biological processes. More detailed technical descriptions may be found in several sources[68-71].

Cell Proliferation

The mechanisms of EMF-induced stimulation of cell proliferation and wound healing remain unclear. Some studies reported that different characteristics of EMF signals can persuade cell differentiation or enhance proliferation of cell of keratinocytes[50-52]. A few studies analyzed the efficacies of low frequency EMF stimulation on keratinocytes in vitro[53, 54, 72]. Manni *et al* (2002) found that EMF at 50 Hz increased human keratinocyte cell growth[50]. Other research also demonstrated that low frequency EMF significantly increased human keratinocyte proliferation[53, 54]. The human epidermis is assumed to be the large supply of stem cells[73, 74]. Motlik *et al* (2007) and Larouche *et al* (2010) found that healing a wound requires the proliferation and differentiation of new keratinocytes from cutaneous stem cells[75, 76]. In particular, epidermal stem cells can fix various damaged tissues by stimulating mobilizing signals like endogenous and exogenous factors. For example, over expression of epidermal growth factor receptor (EGFR) can increase human epidermal stem cell proliferation[77]. Ke *et al* (2008) also reported that a 50 Hz magnetic field induces EGFR clustering[78]. In this regard, Zhang *et al* investigated the effect of low frequency EMF on expanding hESC in vitro. Their results showed that low frequency EMF modulated the proliferation of hESC[77]. These results are in agreement with other reported studies showing an enhancement of keratinocyte growth by EMF applied at different frequencies[50, 52].

Huo *et al* (2010) demonstrated that the effects of noninvasive EMFs on cell migration and proliferation seem keratinocyte-specific with no such effects on dermal fibroblasts[54].

Cell Cycle Analysis

The cell cycle consists of three main phases including G0/G1 phase, S phase, and G2/M phase. Since low frequency EMF augment proliferation of cells, further experiments were carried out to define the effect of EMF on the cell cycle. Some groups have tested the effects of EMF on the cell cycle in different lines of cell, although some controversial outcomes on cell proliferation have been reported[79-81]. These results indicate a lack of knowledge on the clear relationship between cell proliferation which induced by EMF and distribution of cell cycle[82]. Commonly, the proportion of S-phase cells is considered to demonstrate the proliferative potential of a cell population. Low frequency EMF application significantly decreased the percentage of cells in the G1 phase whereas it increased the percentage of those in the S phase and EMF significantly affected the cell cycle, increasing the proportion of cells, synthesizing DNA, and increasing cell growth[77]. Zhang *et al* found that low frequency EMF did not promote apoptosis usually stem cells apportion infrequently because the duration of their cell cycle is long or some cells are arrested in the G0 phase[77]. For treatment with electromagnetic field, there is a lack of high-quality randomized reined trials. Therefore, this treatment is considered investigational for the treatment of wounds[67].

Electric Fields and Wound Healing

Since three decades ago, it has been known that very small exogenously applied electrical currents created beneficial effects on wound healing process[34]. It was first showed by Bassett *et al* that inserting an electrode on each side of a bone nonunion and passing a weak electrical current through it would assist in the fusing of the fracture[83]. Naturally occurring electric currents at human skin wounds were measured over 150 years ago. Bois-Reymond (1818-1896), the founder of modern electrophysiology, wrote in details the electric activities associated with nerve excitation, muscle contraction and wounds[84]. Most recently developed micro needle arrays are able to measure the trans-dermal skin potentials at multiple sites simultaneously[85, 86]. The ET has shown the higher therapeutic effects than the other modalities. Various studies have shown that the electrical properties of wound and intrinsic electrical properties of other involved organs are the most effective factors on the wound treatment responses to electric and EMF stimulations. In addition to these intrinsic electrical properties, physical parameters of electric stimulation are important factors determining the wound treatment outcomes. The common modalities of electric and electromagnetic field stimulations include: direct current, low-frequency pulsed currents, monophasic high-voltage pulses, and pulsed electromagnetic fields, static magnetic field and combined magnetic field. The first technique is direct skin contact method that uses electrode, while the other methods are non-contact. The latter group is also called EMFT. In the following, the most common methods of ET and EMFT are discussed.

Direct Current Electrical Stimulation

Normally, human skin which was uninjured, a difference in ionic concentrations is actively maintained between the upper and lower epidermal layer, which can be measured as a difference of electrical potentials, ranging between 10 and 60 mV on different locations on the body surface. The positive terminal of this so-called epidermal battery is situated on the inside surface of the living layer of the epidermis[24]. After wounding, when the skin layers are interrupted, the epidermal battery at the wound site is short-circuited, creating a conducting pathway that permits ionic current to move through the sub-epidermal region out of the wound and return to the battery by flowing through the region between the dermis and the living layer. The injury current can only flow, as long as wound surface is moist. The active role of endogenous electrical phenomena in wound healing is indirectly confirmed by the fact that the healing of wounds, the surface of which remains wet, is more successful than in wounds that are left to dry out. Modeling of wound edge has shown relatively fast lateral voltage gradient across the edge, which implies that the cells on the wound edge are placed in an electric field[87]. Electric fields on order 100–200 mV/ mm have been measured lateral to wounds in mammalian epidermis. Endogenous wound-induced electric fields present in the cornea plays role in the healing process by helping guide the cellular movements that close wounds.

It has been shown that externally applied electrical fields of such ‘‘physiological’’ intensities can affect orientation, migration, and proliferation of cells[88] which are of key importance for healing, such as fibroblasts and keratinocytes[17, 89-91]. Several studies have confirmed that externally induced electrical fields with endogenous electrical conditions, positive electrode on the surface of wound, and negative on the healthy skin around the wound, speed up wound healing process. Electrical currents ranged from 0.2 mA to 1 mA. The negative electrode on wound surface has reportedly antimicrobial effect that can be advantageous in initial stage of wound development[92, 93].

Low Frequency Pulsed Electric Currents

Low-frequency pulsed electric current applications are entire popular in physical medicine. They are most commonly applied for functional electrical stimulation to arouse involuntary muscle contraction for intensification muscles atrophied by disuse and for eliciting functional movements in patients with motor dysfunction[94,95]. Such electric current pulses are also recognized as totalizing currents. Low-frequency pulsed electric currents were applied locally to the wound as well as to areas quite distant to the wound. The two main distant locations were the spinal cord and acupuncture points[96]. When low-frequency pulsed electric currents are applied locally both electrodes are located on the healthy skin surrounding the wound, the amplitude of pulses is set to value just below visible titanic contraction of surrounding muscles. This modality of treatment is noninvasive and simple to use. The formation of chronic wounds is principally the result of an insufficient supply of oxygen and nutrients to the tissues for poor blood flow. Every-day use of low-frequency pulsed electric current stimulation was found to significantly augment partial oxygen tension (pO2) around the chronic wound while when direct current electrical stimulation was used no significant changes of pO2 were located[97]. Increasing the pO2 during low-frequency pulsed electric current stimulation in patients with ischemic ulcers improved their microcirculation. It is presumed that hypoxia or releasing the metabolites during electrical stimulation is because of insufficient blood flow and increasing capillary growth.

Monophasic High-Voltage Pulses

Muscle is contracted at application of low pulse amplitudes and longer durations as well as at large amplitudes and shorter pulse durations. The use of short high-voltage pulses could not be explained physiologically. The positive electrode is placed over the wound and voltage is set just below that being able to produce visible muscle tetanic contraction. If treatment reached the healing plateau, the polarity of electrode over the wound was reversed. Reversing electrode polarity was successful when wounds were infected. Negative electrode located on the wound has disinfection effect. Studies showed that high-voltage stimulation improves blood flow and therefore facilitate wound healing[44,98]. It is presumed that high-voltage pulses stimulation restores sympathetic tone and vascular resistance below the level of the spinal cord lesion, thereby increasing gradient of the perfusion pressure in the capillary beds. Therefore, high-voltage pulses stimulation could be used for preventing pressure ulcers[99].

Influencing Parameters in Electrical Wound Healing

Endogenous electric fields in the range of 40–200 mV/mm are naturally present near wounds and that skin cells respond to fields of this magnitude with directed motility. It is likely that the electric field may have a role to play in the stimulation of wound healing. Robinson *et al* conducted the first controlled experiment designed to investigate the role of the electric field in wound healing, utilizing a very simple system, the neurula-stage frog embryo[100-102]. Transected frog embryos healed completely within 7 h in a pond water medium that began with a rapid purse string-like contraction requiring microfilaments but not Naþ. This was followed by a slower phase that was blocked by either Naþ-free medium, or the addition of amiloride, benzamil, or ouabain, drugs that inhibit Naþ flux through the epithelium as showed by a rapid reduction in the transepithelial potential. This indicated that the slow phase of wound healing requires the endogenous Naþ-carried electric current and certainly supports a role for the electric field in wound healing.

The second well-controlled experiment was conducted by to Iglesia *et al* test the role of electric fields in wound healing in the newt[103]. They made a small skin wound in one hind-limb digit on both the right and left foot of notophthalamus viridescens and monitored the healing rate while changing the lateral electric field near the wounds by passing current through one digit, across the body and out the contralateral digit. The amount of current passed was adjusted so that the lateral field of one wound was zero while the contralateral wound had an enhanced field[103]. They observed that the wounds with the enhanced field healed more rapidly than the wounds with the zero fields. When digits on one side were treated with 30 M benzamil in the artificial pond water so that their wound fields were reduced to approximately zero, and the contralateral wounds were kept in artificial pond water without benzamil so that they had normal wound fields, there was significantly less epithelization of the benzamil-treated wounds than of the control wounds. This effect on wound healing was reversed by adding currents that restored the normal wound fields, but not by adding currents that reversed the wound fields to the opposite polarity. When currents were added to reverse the wound fields on one side of the animal, leaving the contralateral wounds free of added currents, the contralateral wounds with the normal fields healed much better than the wounds with reversed fields. These results are consistent with the hypothesis that the intrinsic lateral electric fields in the vicinity of wounds promote epithelization of these wounds. These experiments are the most elegant ones to date on this question and the overall conclusion is that in the absence of a lateral electric field, the rate of wound healing is reduced by about 25%[104, 105].

Iglesia *et al* assessed the increasing electric field on the wound healing rat and found that increasing the field from 40 to 80 and 100 mV/mm reduced the rate. They concluded that the newt epithelialization rate was nearly maximal at the normal field strength[103].

Iglesia *et al* conducted a controlled experiment to determine the involvement of electric fields in wound healing[106]. They used bovine corneal lesions with a 1.5 mm circular wound. A decrease in the field strength by submersion of the lesions or by treating the lesions with the Naþ-channel blocker, benzamil, significantly hindered the healing. Increasing the field strength through increasing direct current increased the epithelialization. Epithelialization rate increased twofold when the field strength doubled (80 mV/mm). However, further increase of the field strength to 120 mV/mm reduced the epithelization rate. A similar pattern was also observed upon the reversal of field's polarity[106]. Iglesia *et al* showed for the first time that increasing the field strength as well as reversing its polarity at the wound site enhances corneal wound epithelialization. The final well-controlled study was also conducted on a cornea preparation in situ. McCaig *et al* used the rat cornea to study wound healing in response to a similar circular wound. They manipulated the endogenous lateral electric field near the wound by using drugs with differing actions. Furthermore, they found that the rate of wound closure was highly sensitive to the field strength. In addition to influencing the rate of wound closure, the wound-induced field influenced the orientation of cell division. Most epithelial cells divided with a cleavage plane parallel to the wound edge and perpendicular to the field vector. Increasing or decreasing the field pharmacologically, respectively increased or decreased the extent of oriented cell division. In addition, cells closest to the wound edge, where the field was highest, were oriented most strongly by the field. The frequency of cell division was also enhanced by the endogenous electric field. Because the endogenous field also influenced the wound-healing rate, it appears to be one force that can stimulate both cell migration and cell division during healing.

A further important observation made by McCaig *et al* was the effect of these endogenous fields on nearby nerve growth[13,14,107].The endogenous electric field near the wound shows a very strong orienting effect on the direction of sensory nerve sprouting and growth. Between 16 and 20 h after wounding a large number of nerve sprouts project directly towards the cut wound edge in a whole-mount rat cornea. Reducing the wound field with ouabain randomizes nerve fiber orientation, suggesting that the electric field is the main orienting influence for these nerve sprouts. It has been known for decades that nerve growth can be oriented by imposed electric fields[9-11, 108-111]. However, this is one of only a few well documented examples in which a naturally occurring electric field has been found to exert a strong influence over neuronal growth. In addition to nerve growth, the axis of cell division was also strongly influenced by this endogenous electric field. Treatments that enhanced the field resulted in a greater degree of mitotic spindle alignment perpendicular to the field lines.

Clinical Trials of Electrical Wound Treatment

While these well-controlled experiments are fairly recent, there are a large number of earlier clinical trials to improve wound healing with electrical stimulation. There exists an intense desire to find improved methods for wound management and in the last years alone, at least seven reviews have been made of the clinical trials using electrical stimulation[3, 112-117]. All of these reviews conclude that electro-therapy usually improves the rate of wound healing by 13–50% and sometimes precipitates the healing of chronic wounds for which other conventional therapies have not been successful. Vanable (1989) conducted the most extensive review on the clinical trials[25]. In this book, he describes the three negative reports in the literature[66, 118, 119] followed by 11 positive reports grouped by methodology. Some investigators placed the cathode in the wound during the experiment[66, 120-122]; some used the anode at the wound throughout[27] and others alternate polarity with the cathode in the wound first followed by the anode[29, 92, 93, 122-124]. The most popular approach for stimulating wound healing in humans uses both polarities of imposed fields sequentially, with the cathode in the wound first, followed by the anode. The rationale for using the cathode first is that this renders the wound free from infection[125,126]. There are reports of significant stimulation of healing in over 300 ulcers with this approach, but the control group was always a small fraction of the size of the experimental group. However, these studies demonstrate that electric fields promote the healing of chronic wounds. It is also clear that much work is needed to determine the optimal protocol for electric field application since so many different treatments have been used by these investigators. This will require further studies on model systems such as the mammalian cornea and skin as well as much more work applying fields to human skin wounds.

Evidence from small randomized controlled experiments on electro stimulation indicates improvements on some intermediate outcomes, such as decrease in wound size and/or the velocity of wound healing. However, these studies have not demonstrated Evidence from small randomized controlled trials on electro stimulation reports improvements on some intermediate outcomes, such as decrease in wound size and/or the velocity of wound healing. However, these studies have not demonstrated[67].

Wound-Healing Quantification Assessments

Although different research groups have demonstrated that electrical stimulation can accelerate wound healing, it is still not widely used. Universal effects of electrical stimulation, variety of small studies, unsuitable wound healing quantification methods, and not well established mechanisms that can explain how electrical stimulation affect the behavior of living cells and tissues are the main barriers for optimization of electrical stimulation in wound treatment. Different quantification methods make it impossible to make a quantitative analysis of the comparative advantages and disadvantages of different treatment modalities. To enable quantification and comparison of treatment efficacy, same measure of wound healing requires to be generally accepted, which ideally would fulfill the following criteria:
Simple calculation, suitable for statistical handling, transparency—evident physiological meaning, employability for different wound types, sizes, shapes, and healing and/or non-healing courses. Service providers are enabled to assess, improve, and individualize the treatment applied to each wound patient by quantitative measurement of wound healing. In order to correctly quantify wound healing wound has to be periodically assessed and wound healing process dynamics has to be taken into consideration[18, 85, 90].

Quantitative and Qualitative Measures for Wound Healing Assessments

The proof weather a method of treating wounds is successful is a matter of histological analyses of the affected soft tissue before and after treatment[127, 128]. Reports of histological analyses of electrical wound healing are rare especially in clinical trials. At the Institute of Rehabilitation a histological study of electrical wound healing was done recently[129]. The study enrolled 50 patients with spinal cord injury, suffering from decubital ulcers of III degree according to the Shape scale in the sacral area[129]. A half of wounds were treated according to described biphasic electrical stimulation treatment and another half received only conservative treatment. In five patients from each group a qualitative and a quantitative histological analyses of the tissue samples (about 4 mm) taken from the wound, on the line between the wound edge and freshly formed scar, were performed before the beginning of treatment and after around 2 months, when the formed scar formed during the electrical stimulation was of considerable size. Wound healing was followed as described in above clinical study section. Significantly faster healing of wounds in electrically stimulated group was observed. The histological preparations were analyzed by a quantitative stereological method. Content of surface collagen in the preparations stained according to Masson and the surface density of blood vessels was determined in the immune histochemically stained preparations. The surface percentage of collagen was determined by using test system M-42 and the number of blood vessels per surface unit by a semiautomatic IBAS 1000 image processing and analysis system. Wounds treated by electrical stimulation had lower inflammatory response, higher collagen density as well as more intense process of angiogenesis. In electrically healed group collagen density increased in average 23%, while in the control group decreased by2%of the initial surface in two months time period. The area density of blood vessels was higher in electrically stimulated wounds, and in post stimulation period the blood vessels were found to be reaching essentially higher towards the wound surface than in the non stimulated wounds, in fact, almost as far as the crust. In stimulated wounds endothelial cells were flat, the blood vessels lumina broad with erythrocytes clearly visible within them. In the control group, endothelial cells were thickened, cubically shaped, with round nuclei, and no erythrocytes were visible within blood vessels lumina. In addition, earlier in vitro studies reported flatter endothelial cells exposed to electromagnetic or electric field, which are cubic when not exposed to the field[91, 130]. Cukjati and Sávrin indicated that the intercellular substance is controlled by fibrin whereas more collagen was found in the sample preparations of electrically healed wounds[131]. The conclusion is that electrical stimulation may exert the release of mediators responsible for the increase in collagen synthesis in fibroblasts or the shrinking of myofibroblasts. Furthermore the study showed that electrical healing has a favorable effect on blood circulation in the wound, improves blood circulation in the tissue surrounding the wound and improves the quality of post-treatment scar[131].

Biological Effect of Electric Fields

There are many theories regarding the mechanisms of electrical field influencing the cells. One theory explains that the lateral electrophoresis and local clustering of membrane ion channels such as calcium channels increase local fluxes of ions, possibly inducing the cell to form local lamellipodia. Changes in actin stress fibers and microtubules can also affect cell shape and orientation[132, 133]. In many cells, DC electric field induced migration depends on the variations of intracellular $Ca^{2þ+}$ concentration and induction of alterations of membrane potential[134, 135]. Various biological interactions of electric fields with tissues have been proposed. However, most of these studies were performed with DC electric field devices on tumor cells. A recent study by Zhao highlighted the significance of physiological electric fields in wound healing, arguing that they act as important directional cues that can override other migration factors, such as chemotaxis and contact inhibition release, both in monolayer wound healing models and in stratified epithelia[107, 136].

CONCLUSION

The present study has reviewed the most current electric and electromagnetic field therapies for wound treatments and compared their efficiency for each wound. In addition the proposed mechanisms of action of these techniques were reviewed. Electric current and electric field stimulations have shown the more promising effects on wounds. Induction of lateral electrophoresis and local clustering of membrane ion channels such as calcium channels are among the main biological interactions of EMF and cells. Furthermore, different parameters influence the therapeutic performance of ET and EMFT including electrical intrinsic properties of living organs as well as physical parameters of stimulations. For further development of EMF based treatments for wound it is necessary to develop more quantitative assessments for wound healing as well as conducting controlled studies on the effects of different EMF modalities on wound healing process.

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