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Ultrasound Techniques for Treatment of Bone Fractures: A Review of Mechanisms of Actions

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ABSTRACT

Ultrasound (US) waves have recently shown therapeutic effects for different fractures including fresh, non-union, and delayed fractures shown promising potentials as alternative or adjunctive treatment for bone fractures. However, the mechanisms of actions of this technique in bone fractures healing are not fully understood. This paper aimed to comprehensively review the biological interactions and mechanism of action of US waves in the treatment of bone fractures. The databases of PubMed (1990-2016), EMBASE (1990-2016), Web of Sciences (1990-2016), and Google Scholar (1980-2016) were searched using the set terms. The title and abstract of the collected results were reviewed by two authors and the relevant papers were selected for further evaluations. The available evidence showed the therapeutic efficiency of US waves in bone fractures. The mechanisms of action depend on the US physical parameters as well as exposure factors including duration. The main mechanisms of US waves for enhancing the healing process of bone fractures healing are increasing fibroblasts and decreasing osteoclasts, stimulating the collagen formation, enhancing extensibility of collagen, circulation, and pain threshold. Low US frequencies show more therapeutic efficiency in bone fractures healing compared with high frequencies.

Key words: Ultrasound Wave, Bone fractures, Treatment, Mechanisms of Action, Biological Interactions

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INTRODUCTION

Bone fracture is one of the most common diseases worldwide. The fracture healing process is a relatively long period mechanism. In addition, a relatively large portion of fractures become non-union or delay which increase the healing period or even impede the healing process completely [1]. Therefore, developing an intervention for speeding up the fracture healing process or preventing the formation of non-union or delay fracture has been always as one of the main research avenues in orthopedic and alternative medicine. Ultrasound (US) waves have shown promising therapeutic effects in shortening the fracture healing period as well as in treating non-union and delayed fracture.

These characteristics have motivated the researchers to develop and introduce new techniques as alternative or adjunctive treatments for reducing the healing process of bone fractures. The most important of them are US stimulation, direct current, capacitive coupling, inductive coupling (pulsed EMF), static and combined magnetic fields. However, the mechanisms of action of each technique are controversial and they are not yet completely understood. Nowadays ultrasound is widely used in medicine as an adjunctive treatment [2-16].

Low intensity pulsed ultrasound stimulation (LIPUS) is a biophysical intervention on a fracture healing process [17]. Most of the studies that investigated biological effect of LIPUS on the bone-healing process showed positive effects of ultrasound on fracture healing [18-20]. Several clinical and experimental studies have shown the potentials of LIPUS in enhancing the bone cells synthesis during fracture healing [18, 21-23]. Therapeutic exposures with ultrasound because of its ability to both image and deliver makes it a very practical and useful technique [24]. The present study comprehensively reviews the applications of US based techniques for enhancing bone fracture healing including fresh non-union and delayed fracture. In addition, recent advancements of these techniques as well as mechanisms of actions of these techniques in enhancing bone fracture healing process are discussed.

2. Method

The databases of PubMed (1990-2016), EMBASE (1990-2016), Web of Sciences (1990-2016), and Google Scholar (1990-2016) were searched using the set terms. The search terms included "ultrasound wave", "bone fractures", "mechanisms of action", and "biological interactions". The obtained records were reviewed for the title and abstract by two authors and they came to consensus whether the studies are related to the review. Animal and human studies in both in vivo and in vitro designs that evaluate the therapeutic effects and/or mechanisms of action of US waves on different bone fractures were included for further evaluations. Any studies that evaluate the effects of US waves on one of the physiological, metabolic, morphological, or physical characteristic of wounds were reviewed. Because of the immense body of literature and variance in the methodology, this study aimed to provide a comprehensive and descriptive overview of the recent advances in applications of US waves for treatment of bone fractures and their mechanisms of action and biological interactions with living tissues.

3. Results

3.1. Bone fracture healing process

Bone can repair itself with a self-regulating mechanism when is damaged. This Phenomenon is the resulting of both microscopically and macroscopically process and also is extremely complex. The process of bone healing is

influenced by many factors such as biological and mechanical factors. Biological factors related to tissues and mechanical factors associated with forces and motions at the fracture site.

3.2. The process of bone healing

Bone fracture healing process can be summarized in several main steps: hematoma formation, inflammation, cellular proliferation and differentiation, ossification and finally remodeling. Hematoma formation rich in platelets formed and released of cellular signaling molecules. In inflammation stage blood flew and vascular permeability is increased, chemotaxis and migration of inflammatory cells is occurred and lead to further cytokine release and MSC accumulate in fracture site. In next stage, stem cells proliferate and differentiate based upon mechanical and biological signals. Then vascular invasion and new vessel is formed. During the ossification stage collagen, fibrils are randomly laid down in random orientation. In this way, collagen fibrils ossification, callus bridges fracture, and union occur with woven bone. The final step is remodeling. Remodeling step is ongoing process in normal bone progresses to fracture site. In this stage, coupled action bone resorption and deposition based on mechanical stresses. Finally, woven bone replaces lamellar restoring micro-architecture [1, 25].

3.3. Types of bone fractures

If the damage to the bone is severe enough bone fracture occurs. At first its motion should minimized by internal or external fixation. In Such conditions provided primary fracture healing can occur. Fractures are divided into several categories based on the recovery time. The bone fracture types include fresh fracture, closed or open fracture, union, non-unions and delayed fracture. Kind of a bone fracture is important factor for determining the treatment protocol [2].

Delayed union is occurred when the fracture site failure to heal between 3 and 9 months after fracture. In other words, bone fracture is called delayed fracture when the rate of bone healing has decelerating process. The U.S. Food and Drug Administration (FDA) labeling defined nonunion as follows: "A nonunion is considered to be established when a minimum of 9 months has elapsed since injury and the fracture site shows no visibly progressive signs of healing for minimum of 3 months." According to another definition of the FDA “a nonunion is considered to be established when the fracture site shows no visibly progressive signs of healing.” This type of fracture creates many problems for both patients and the orthopedic trauma surgeons. The rates of non-union fractures between 5% and 10% have been reported. Recently, the surgeons looking for the understanding of fracture healing to management of fractures and reduce the risk of non-union [1, 26].

3.4. Bone fracture treatment

Generally, the aim of fracture treatment should be at first relieving the pain and then restoring function to the body part [27]. The first step in the traditional treatment of bone fractures is local treatment. This method can restore alignment, obtain appropriate stability, enhance fracture site biology and eradicate infection [1, 28]. Furthermore local treatment, some of instructions to patients that may be beneficial in bone fracture treatments are improving nutritional status, cessation of smoking and avoidance of non-steroidal anti-inflammatory by patients [29]. Several factors that may affect the healing process are Infection, Smoking, Certain medications, Advanced age, Systemic medical conditions, Poor functional level, Venous stasis, Obesity, Alcohol abuse, Metabolic bone disease, Malnutrition, Vitamin deficiencies [1, 30, 31]. Sometimes the fracture healing process does not pass normally. The

recovery process may be take up to several months that in this case delayed union arises. If the delay in healing continues more than nine months is thus termed non-unions. To resolve this problem for improving fracture union rates and shortening the treatment period several new methods such as external stimulants, including electromagnetic fields, high-frequency low-magnitude mechanical stimuli and low intensity pulsed US have been introduced [32-34]. Among the advantages to using of this methods are relatively inexpensive, easy to use and carry and very low risk of complications [35]. LIPUS is a one of the promising techniques for the treatment of fracture that has been shown to improve fracture healing [19, 34, 36-38]. Both in vivo and in vitro studies have confirmed the positive stimulatory effect of LIPUS on biologic activities in fracture healing and improve healing process [15, 39, 40]. They have shown that it can affect cellular differentiation and functional activation of bone formation [41-43]. This effect is critical to begin a sequence of biologic activities to synthesize of new bone cells.

3.5. US and bone fracture treatments

Ultrasound waves are currently used in different fields of medicine such as diagnostic, operative and therapeutic settings [35]. Low-intensity US is an intervention method that has biophysical effects in mechanisms of fracture healing process. It can accelerate process of healing, improve callus formation and return of bone strength in all kind of fractures [44-47]. The energy used by LIPUS treatment is extremely low but the results have shown significant beneficial effects. Several histological studies have shown that LIPUS influences several cell types including osteoblasts, osteoclasts, chondrocytes and mesenchymal stem cells. LIPUS treatment can effect on cell membrane permeability and increase cellular activity [39, 44, 48, 49].

The effects and adsorption of US waves on tissues are proportional to the density of the tissues. With regard to this subject its effect on the fracture gap and bone healing is more than soft tissues. When the US waves pass through tissues, due to reflect radiation energy on desired location like the bone callus or bone muscle interfaces pressure variations create throughout the tissues. These effects can make changes in the cellular and molecular levels. The response of this process may be modulated cell function [35]. Studies considered the mechanism of action of US in bone healing in two parts: non-thermal mechanisms that influence on cellular activity and the biophysical mechanisms that have enhancing effect on bone regeneration [25]. Several potential mechanisms of LIPUS that effect on bone fracture healing are Mechanical signal transduction and induction of gene expression, activation of enzymes in response to heat energy, increased vascularity at the fracture site, modulation of intracellular calcium signaling, enhanced cartilage calcification and maturation [35]. Clinical and experimental studies investigated the therapeutic applications of LIPUS into ‘‘low power’’ includes physiotherapy, fracture repair, sonophoresis, sonoporation and gene therapy and ‘‘high power’’ involves high intensity focused US. Each of the different level of intensity has thermal and non-thermal interaction mechanisms such that at higher levels, heating and acoustic cavitation will predominate [24].

3.6. A Historical review of US and bone fractures

Several studies have investigated the out effects of US on bone healing in various qualifications and protocols [50-56]. Some of them survey and next compared two or more different positions and protocols with each other [34, 51, 57]. The positive feedback of experiments and their results led to extension use of LIPUS clinically and attention to its therapeutic applications. In this section some of them are reviewed. Buchtala (1950) investigated US therapy and its effect. He suggested that US might effect on bone healing process through stimulate osteogenesis [50]. Similarly,

Maintz in 1950 in a first examine studied effects of high intensity US on rabbit radial fractures. He survived effects of 4 high intensities of US on callus formation and bone healing. He investigated the results with histological and radiographic analysis. He reported that US stimulation in 500 mW/cm^2 intensity had minimal changes on fracture healing. He also survived other high intensities such as 1000, 1500, 2500 mW/cm^2 that the three protocols had negative effects and cause reduce callus formation [51]. Several studies used very high US intensities (between 2500 and 5000 mW/cm^2) for fracture treatment in dog femora. They observed that very high US intensities cause delayed bone healing, necrosis, and dense fibrous tissue formation [52-54]. In another studies that had investigated High intensity US between 200 and 3000 mW/cm^2 on fracture healing, they showed increase callus formation and accelerate bone healing [55, 56, 58, 59]. In 2002, change et al. used high intensity US with 500 mW/cm^2 intensity and observed significant increase in torsional stiffness of limbs stimulated (about 80%) and increase in new bone formation (about 30%) [60].

3.7. Biophysics of US induced bone osteogenesis

3.7.1. Thermal effects of US

LIPUS stimulation through transfer of energy can increase the temperature of tissues and it may be cause changes in tissues and cells [60-62]. LIPUS treatment with low level of energy leads to extremely low pressure waves [20, 63-65]. The observe of thermal effects of LIPUS may be expected more in the high intensity (higher than 1000 mW/cm^2), but studies reported little temperature variations of LIPUS on 20-50 mW/cm^2 intensities [34, 60]. Nevertheless, studies have shown even a small increase in temperature may affect some enzymes such as matrix metalloproteinase 1 that also known as interstitial collagenase or collagenase 1 [66, 67].

3.7.2. Non-thermal effects of US

The changes caused by US in addition to thermal effects may also be caused by non-thermal effects. Including non-thermal processes of LIPUS treatment in tissues and cells are acoustic streaming and cavitation [68, 69]. Several studies showed that LIPUS stimulation has biochemical events at the cellular level and it can increase the activity level of the cell [70-72]. The results of several studies have showed that LIPUS treatment can increase in protein synthesis and also have a direct effect on cell membrane permeability [48, 70, 73-75]. The effects of LIPUS on cell membrane permeability due to increase micromechanical blood pressure and this process may lead to accelerated fracture healing [75]. Webster et al. (1980) reported collagen synthesis observed in human fibroblasts that stimulated with US in 500 mW/cm^2 intensity may involve cavitation mechanisms [76]. Cavitation ‘‘involves the pulsation of gas or vapour-filled voids in a sound field’’ that are included stable and Unstable cavitation [71, 77]. The cellular reactions could associated with the effect of cavitation in the case of very low energy [25]. The cavities could be the concentration of acoustic energy and they can be obtain in shearing and microstreaming fields [69]. Acoustic streaming is ‘‘a small scale eddying of fluids near a vibrating structure such as cell membranes and the surface of stable cavitation gas bubble’’ [78]. Stable cavitation and acoustic streaming are cases affect diffusion rates and membrane permeability [25].

According to Wolff’s Law LIPUS can be considered as noninvasive force stimulating bone healing processes through producing mechanical stimulation followed by induce micro-motion [25, 79].

Influence and penetration of LIPUS on the tissues is related to the density of tissues and it is mostly reflected at tissue boundaries of soft and hard tissue such as connective tissue and cortical bone. Therefore, LIPUS stimulation does not have the ability to stimulate osteogenesis in intact bone [20, 63-65] or callus in the remodeling phase [15, 80, 81]. The stimulation by mechanical loading because of the differential absorption of it by tissues may establish a gradient of mechanical strain in the inflammatory and soft callus formation phases of fracture healing and also can predict specific locations of new bone formation [23, 82].

Several studies have suggested that US may influence localization and quantification of proliferating cells such as fibroblasts and osteoblasts [83-85]. Nevertheless, many in-vivo and in-vitro studies and their positive results about beneficial applications of LIPUS biophysical mechanisms and complex process of bone healing is still unknown and require further more research.

4. Conclusion

The available evidence showed the therapeutic efficiency of US waves in bone fractures. The mechanisms of action depend on the US physical parameters as well as exposure factors including duration. The main mechanisms of US waves for enhancing the healing process of bone are increasing fibroblasts and decreasing osteoclasts, stimulating the collagen formation, enhancing extensibility of collagen, circulation, and pain threshold.

Low US frequencies show more therapeutic efficiency in bone fractures healing compared with high frequencies fractures healing.

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