Effects of low-intensity pulsed ultrasound on autogenous bone graft healing

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Objective. Pulsed ultrasonic waves have been shown to accelerate bone healing. The aim of this study was to evaluate the effects of low-intensity pulsed ultrasound on bone healing.
Study Design. Thirty-two femurs from 16 skeletally mature male Wistar albino rats were used. Two defects were produced in each femur with a 3-mm-diameter trephine bur and then autogenous cortical grafts were placed in 1 defect. The animals in the experiment group received a daily 20-minute ultrasound treatment for 15 days. All animals were killed on the 18th postoperative day.
Results. Dual-energy x-ray absorptiometry values and biochemical markers in the experimental side were significantly higher than those in the control side at the early period of bone healing.
Conclusions. Low-intensity pulsed ultrasound treatment accelerates bone healing both in bone defects and in bone defects filled with cortical autogenous block graft. (Oral Surg Oral Med Oral Pathol Oral Radiol 2014;117:e255-e260)

The restoration of bone defects in the maxillofacial region remains a challenge because of the body's limitations in healing bone defects on its own.[1] Such defects may be congenital, traumatic, periodontal, or iatrogenic, caused by bone resections such as oncological procedures. Research in the field of maxilla and mandible bone regeneration has yielded several ways of dealing with bone defects, such as the use of bone grafts.[2] Graft materials such as tricalcium phosphate, hydroxyapatite, or calcium sulfate were used in routine maxillofacial surgery operations. In reconstructive oral and maxillofacial surgery, autogenous bone grafts are one of the most favored grafts because of their osteogenic properties.[2,3]

Multiple methods for accelerating bone healing have been proposed, some of which are approved for use in the clinical setting.[4] Growth factors implanted at the bone defect site have been shown to increase rates of bone healing and decrease rates of infection.[5] Electrical stimulation, either by implanted direct current stimulators or by externally applied capacitative or inductive couplers, has been variably shown to be marginally effective.[6,7] Recently, approaches have focused on enhancing bone healing using different forms of biophysical stimulation such as ultrasound.[8]

Ultrasound is acoustic radiation at frequencies above the limit of human hearing. Ultrasound is used in medicine for diagnostic and therapeutic purposes.[8] Low-intensity pulsed ultrasound (LIPU) began to be widely used clinically to treat bone defects in the early 1990s. In the literature, LIPU generally entails a 20-minute treatment per day of 1-MHz waves repeating at 1 kHz, an average intensity of 30 mW/cm^2, and a pulse width of 200 ms[9] LIPU is a form of mechanical energy that is transmitted through and into living tissue as acoustic pressure waves. The micromechanical strains produced by these pressure waves in body tissue can result in biochemical events at the cellular level and may promote bone formation in a manner comparable with the bone responses to mechanical stress postulated by Wolff's law.[10]

The purpose of this study was to evaluate the effects of LIPU on cortical autogenous block bone grafts and defect healing in a rat model using dual-energy x-ray absorptiometry (DEXA) and biochemical analyses.

Statement of Clinical Relevance

With the use of low-intensity pulsed ultrasound (LIPU) applications in clinical cases, surgeons could accelerate maxillofacial bone healing in dental implant surgery and bone defect repair and increase the amount of alveolar bone in routine maxillofacial surgery practice to shorten the bone healing time. In addition, specific LIPU devices may be designed for home use in routine maxillofacial surgery practice.

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Fig. 1. Bone defects were created nearly 2.5 to 3 mm from each defect and prepared in the same diameter using a trephine bur.

Fig. 2. The ultrasound transducer was only attached to the lateral surface of the right femurs of the group A animals.

MATERIAL AND METHODS

The animal study protocol was approved by the Animal Studies Review Committee and was in accordance with institutional guidelines (University of Süleyman Demirel, Isparta, Turkey). Twenty-four adult male Wistar albino rats weighting 345 to 360 g at acquisition were used. All animals were older than 8 months of age. Specific attention was paid to selecting animals of uniform size to limit variability in DEXA scanning. These rats were randomly divided into 2 groups (group A, treated with ultrasound, and group B, a control group without ultrasound application). In group A, ultrasound treatment was applied only to the right femurs; the left femurs were untreated. To compare both femurs under the standard conditions, the same operations were performed on both femurs in group B without ultrasound application. The animals were housed in an experimental animal room (22°C, 55% humidity, and a 12-h light/dark cycle) and fed a standard laboratory diet and water.

Surgical procedure

After general anesthesia was delivered with 90 mg/kg xylazine HCl (Ketasol; Richter Pharma AG, Weis, Austria), the surgical area was shaved, and the skin was washed with 10% povidone iodine solution (Poviiodeks, Kimpa, Turkey).

After a skin incision in the femur area, the subcutaneous tissues were dissected down to the periosteum, and a periosteal incision was made on the femur. With a standard trephine bur under irrigation with saline, 2 pieces of cortical block bone grafts 3 mm in diameter were osteotomized from the femur. After the grafts were obtained, proximal bone defects were filled with these bone grafts in 1 piece, whereas the other piece was left empty. These bone defects were nearly 2.5 to 3 mm from each other (Figure 1). The grafts were placed in the proximal bone defects and stabilized with absorbable suture materials. The periosteum, muscle fascia, and skin were then sutured in separate layers. These procedures were performed on both the left and the right femurs of all rats. An analgesic (1 mg/kg tramadol, Contramal; Abdiibrahim, Istanbul, Turkey) and an antibiotic (25 mg/kg cefazolin, Cefamezine; Eczacıbas, Istanbul, Turkey) were administered intramuscularly preoperatively and twice per day for 3 postoperative days.

Ultrasound application

On the second postoperative day, LIPU application was started for the animals in group A. Ultrasound energy was provided by an ultrasound signal comprising a burst width of 200 μs containing 1.5-MHz sine waves, with a repetition rate of 1 kHz and a spatial average–temporal average intensity of 30 mW/cm^2.[8] Ultrasound applications were performed in a standard manner by the same device (Exogen 2000; Smith and Nephew Inc., Memphis, TN) after the animal was placed in a box to restrict excessive movement. The ultrasound transducer was attached only to the lateral surface of the right femurs of the group A animals (Figure 2). No sedative agent was given during the ultrasound treatments to avoid the negative effects of sedative agents. An investigator observed the whole ultrasound treatment session to ensure the continuity of the application. The 20-minute sessions were repeated on a daily basis for 15 days. The surgical procedure was applied to both group A and group B, but ultrasound treatment was applied only to the right femurs of group A. On the 18th postoperative day, all animals were sacrificed with an intravenous injection of 100 mg/kg sodium pentobarbitone (Pental; IE Ulagay, Istanbul, Turkey). Before the rats were sacrificed, 10-mL blood samples were collected for assays of biochemical markers. The right and