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Extracorporeal Shockwave Shows Regression of Osteoarthritis of the Knee in Rats

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Background. **This study investigated the effects of extracorporeal shockwave technology (ESWT) in osteoarthritis of the knee in rats.**

Materials and Methods. **Thirty-six Sprague-Dawley rats were randomly divided into three groups with 12 rats in each group. Group I was the control group and received neither anterior cruciate ligament transection (ACLT) nor ESWT. In groups II and III, ACLT was performed in left knee and osteoarthritis (OA) was verified at 12 wk. Group II received no ESWT, and group III received ESWT at 12 wk after ACLT. Radiographs and bone mineral density (BMD) were obtained at 0, 12, and 24 wk. The animals were sacrificed at 24 wk. One half of the animals were subjected to bone strength test, and the other half for histomorphologic examination and immunohistochemical analysis.**

Results. **Radiographs of the left knee showed progressive OA changes at 12 and 24 wk in group II, whereas, very subtle OA changes were noted in group I and group III. BMD and bone strength were significantly lower in group II compared with groups I and III, but no difference was noted between group I and group III. The cartilage degradations were significantly higher in group II compared with groups I and III, but no difference was noted between group I and group III. The subchondral bone remodeling was significantly less pronounced in group II compared with groups I and III, but no difference was noted between group I and group III.**

Conclusions. **Application of ESWT to the subchondral bone of the medial tibia condyle showed regression of osteoarthritis of the knees in rats.**

Key Words: **osteoarthritis; knee; subchondral bone; shockwave.**

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INTRODUCTION

Osteoarthritis (OA) of the knee has long been considered primarily a cartilage disease associated with cartilage loss and degradation. However, OA is usually accompanied by changes in the subchondral and periarticular bone such as sclerosis, bone cyst. and osteophyte formation [1, 2]. The relationship between the subchondral bone changes and the initiation and progression of OA is still debated [3–5]. It was suggested that increased subchondral bone stiffness reduces the ability of knee joint to dissipate the load and distribute the forces within the joint, and increases the force load on the overlying articular cartilage, which in turn accelerates the cartilage damage over time [6, 7]. Therefore, the functional integrity of the articular cartilage depends on the mechanical properties of the subchondral bone. Some authors proposed the potential role of subchondral bone in the initiation and progression of OA [8]. Emerging evidence indicates that bone turnover increases in patients with OA [6, 9–11]. Using microfocal computed tomography in a dog model, some reports showed subchondral bone loss in the early stage of OA and bone sclerosis in the late stage of OA [6]. As the OA progressed, increased bone resorption reduces the subchondral bone volume, and finally the process is followed by increased bone formation and

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increased subchondral bone volume (sclerosis) and formation of periarticular osteophytes [4, 5, 12].

The results of another study demonstrated that early application of extracorporeal shockwave technology (ESWT) to the subchondral bone of the medial tibia condyle showed chondroprotective effects in the initiation of OA changes of the knee in rats (unpublished data). We hypothesized that application of ESWT to the subchondral bone may ameliorate the progression of osteoarthritis of the knee. The purpose of this study was to evaluate the effects of ESWT in osteoarthritis of the knee in rats.

MATERIALS AND METHODS

Study Design

The Institutional Review Board on animal experiment approved this study. All studies were performed in accordance with the guidelines in the study and the care of animals in experiment.

Thirty-six male 8 wk-old Sprague-Dawley rats with body weight ranging from 250 to 275 mg were used in this study. The anterior cruciate ligament transacted (ACLT) osteoarthritic knee model in rat was used in this study [8, 10]. The rats were randomly divided into three groups with 12 rats in each group. The rats in group I received neither ACLT nor ESWT and served as the baseline control. The rats in group II underwent ACLT, but received no ESWT at 12 wk, and were observed for the progression of OA changes until 24 wk. The rats in group III underwent ACLT and received ESWT to the subchondral bone of the medial tibia condyle at 12 wk after ACLT when OA changes of the knee was verified radiographically.

Anterior Cruciate Ligament Transection (ACLT)

The rats in groups II and III underwent ACLT under anesthesia using intraperitoneal injections of phenobarbital (50 mg/Kg body weight). The left knee was prepared and draped in surgically sterile fashion. A straight anterior incision was made and the knee joint was opened through medial parapatellar arthrotomy. The anterior cruciate ligament was transacted with a scalpel. The knee was irrigated and closed in routine fashion. The animals were returned to the housing cages and were under the care of a veterinarian. The rats in group I did not receive surgery for ACLT.

Shockwave Application

A pilot study was first performed to determine the optimal dosage of ESWT in osteoarthritis of the knee in rats. Using the lowest 14 kV of the shockwave device, the results showed that 800 impulses at 14 kV produced the better effects than 200 and 400 impulses, respectively. Such protocol was subsequently utilized in the current study.

ESWT was administered to the rats in group III at 12 wk after ACLT. The animals in group III were anesthetized with intraperitoneal injection of phenobarbital (50 mg/Kg body weight) for receiving ESWT, whereas the rats in groups I and II received neither anesthesia nor ESWT. The source of shockwave was from an OssaTron orthotriptor (Sanuwave, Alpharetta, GA), a high-energy electrohydraulic focused shockwave device with a conical shockwave tube of 13 cm in diameter at the base, and 6.5 cm in vertical length. The focus of shockwave treatment was 0.5 cm below the medial tibia plateau in anteroposterior view and 0.5 cm from the skin edge in lateral view (Fig. 1). Ultrasound lubricate gel was applied to the skin in contact with the shockwave tube. Application of 800 impulses of shockwave at 14 kV (equivalent to 0.18 mJ/mm^2 energy flux density) was delivered to medial tibia condyle in a single session. The rats were observed in the housing cage. The gait patterns of the rats were not affected following ESWT.

Radiographs and Bone Mineral Density (BMD)

Serial radiographs of the left knee in anteroposterior and lateral views were obtained at 0, 12, and 24 wk. Bone mineral density (BMD) of the knee with region of interest (ROI) focused on the medial proximal tibia was performed at 0, 12, and 24 wk using DEXA (dual energy X-ray absorptiometry) device.

Blood and Urine Samples

Two milli-litters of blood were obtained *via* cardiac puncture at 24 wk before animals were sacrificed. Serum levels of cartilage oligometric protein (COMP), alkaline phosphatase, and osteocalcin were measured from the blood samples. Urine samples were also collected and urinary concentrations of C-telopeptide of type II collagen (CTX-II) were assessed.

Bone Strength Test

The animals were sacrificed at 24 wk. Six rats from each group were subjected to bone strength test, and the other six for histomorphologic examination and immunohistochemical analysis. A 1.0 cm × 1.0 cm bone block was harvested from the medial half of the proximal tibia. The bone strength test was performed on material testing system) (MTS) machine using a slow load compression technique. The compression strength of the proximal tibia bone was measured accordingly. The torsional strength of the bone was tested with torque load to determine the maximal bone strength before fracture occurs.

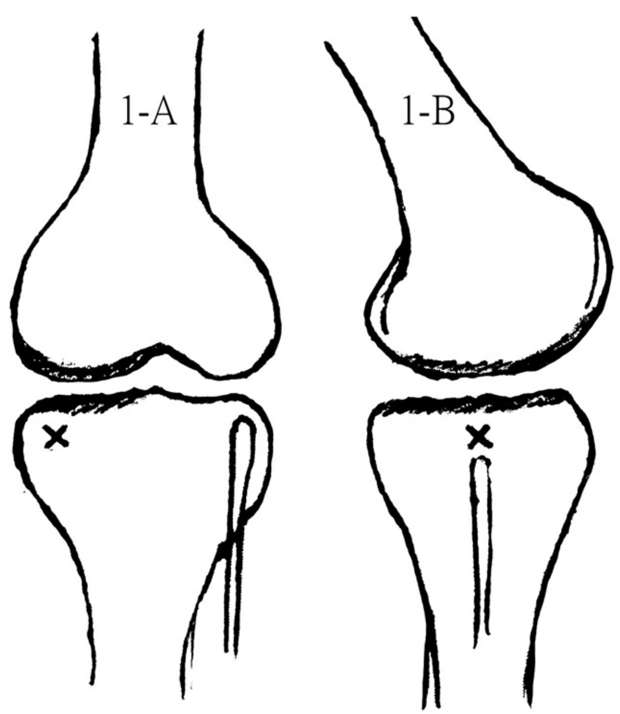

FIG. 1. A sketch of the left knee showed the location of shockwave application in the medial tibia condyle in anteroposterior view (A) and lateral view (B).