



# Osseous alterations in the condylar head after unilateral surgical directional change in rabbit mandibular condyles: Preliminary study



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## ABSTRACT

The purpose of this study was to investigate bony changes in the mandibular condyle when the surface not normally subjected to masticatory forces was subjected to functional loading using a unilateral surgical experiment. Fifteen male New Zealand white rabbits, divided into two groups, were used. Oblique vertical body osteotomies of the mandible and counterclockwise rotation (CCWR) of the proximal segment (PS) [six with 1 mm (group I), six with 3 mm (group II)] were performed on the right side. Osseous changes of condyles were analyzed using micro-computed tomography and histological evaluation four weeks postoperatively. The comparison was performed between condyles on the right and left sides. Since the left condyle (control) might be affected by the operation on the right side, the results were also compared with the healthy control (group III,  $n = 3$ , 6 condyles). CCWR of the PS led to osteoporotic changes of the condyle including significantly reduced bone volume and bone mineral density ( $p < 0.05$ ), thin and small number of trabeculae ( $p < 0.05$ ). In addition, thinning of condylar cartilage and reduced density of cartilaginous cells were observed. However, these changes were not affected by the amount of CCWR of the PS.

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## 1. Introduction

Degenerative joint disease, also known as osteoarthritis or osteoarthrosis, is the most common and severe form of temporomandibular disorder affecting the temporomandibular joint (TMJ). The disease, which is often brought on by mechanical overloading of the joint, is characterized by deterioration and abrasion of articular cartilage and disc surfaces and by a thickening and remodeling of the underlying bone (Tanaka et al., 2008; Machon et al., 2011; Lee et al., 2012).

Idiopathic condylar resorption (ICR), a kind of osteoarthritis, usually occurs in females with pre-existing TMJ dysfunction during the second or third decade of life (Hoppenreijns et al., 1999; Wolford, 2001). ICR results from dysfunctional articular remodeling, due to functional overloading and/or reduced adaptive capacity of the TMJ articular structure (Arnett et al., 1996). The disease has an obvious skeletal pattern characterized by decreased condylar head volume, shortened ramus height, and development or worsening of the class II occlusal relationship, especially in individuals with a high

mandibular plane angle (Arnett et al., 1996; Hoppenreijns et al., 1999; Wolford, 2001; Mercuri, 2008; Gunson et al., 2012). This tendency seems to be related to a decrease in the number and density of condylar trabeculae in patients with a skeletal class II open-bite (O’Ryan and Epker, 1984).

The trabecular patterns of a bone reflect its functional loading patterns. If there is a difference between trabecular and functional loading patterns, the trabecular pattern should change to adapt to the mechanical environment (O’Ryan and Epker, 1984). Among patients with skeletal class II, those whose condylar neck is posteriorly inclined are likely to have condylar resorption because the anatomically less dense, unloaded anterosuperior surface of the condyle is subjected to functional loading. During mandibular advancement with sagittal split ramus osteotomy, counterclockwise rotation (CCWR) of the proximal segment (PS) usually occurs, which is caused by cranial repositioning of the inferior border of the PS in order to avoid a postoperative antegonial notch (Hwang et al., 2000a). The amount of CCWR of the PS shows significant correlation with the amount of mandibular advancement and decrease of the mandibular plane angle (Hwang et al., 2000b). The possibility of postoperative condylar resorption increases if there is CCWR of the PS following mandibular advancement surgery. Loading of the normally unloaded surface and compressive force by tension of the

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suprahyoid muscle may have synergistic effects on resorptive changes of the condyle. This explanation is supported by previous studies showing that ICR also occurs after isolated Le Fort I osteotomy with autorotation of the mandible (O’Ryan and Epker, 1983; Arnett and Tamborello, 1990; Huang et al., 1997). Hwang et al. reported that patients with posterior inclination of the condylar neck and surgically induced CCWR of the PS show a greater tendency to develop postoperative condylar resorption after mandibular advancement surgery (Hwang et al., 2000a,b, 2004).

Numerous animal models have been developed to study degenerative joint diseases. Intra-articular injection of an antigen such as ovalbumin can cause osteoarthritis (Tominaga et al., 2001; Kristensen et al., 2011; Ohtani et al., 2012). Mechanical loading on the TMJ by traction of the mandible with wire or elastics can also induce osteoarthritis (Imai et al., 2001; Fujimura et al., 2005; Zhao et al., 2010). In addition, the removal of 4–5 mm of the antero-lateral meniscus of the knee joint (Colombo et al., 1983) or 2 × 2 mm defect on the cartilage on the surface of the mandibular condyle of a rabbit (Taylan Filinte et al., 2011) have been proposed as a method to induce osteoarthritis. However, these animal models do not mimic the characteristics of ICR which is affected by skeletal changes of the mandible and direction of the condylar trabeculae. In order to further clarify the pathophysiology of ICR, the development of animal models with similar mechanical patterns is required.

We designed an animal experiment with rabbits that mimics CCWR of the PS, which commonly occurs during mandibular advancement surgery in skeletal class II cases. CCWR can induce a difference between the trabecular pattern and functional loading vector. This occurs because the load-resistant anterosuperior surface is located in a more superior position, and the condylar anterior surface which is normally less loaded or unloaded is subjected to anterosuperiorly directed functional loading after CCWR while the normal trabecular pattern of the rabbit’s condyle is in the anterosuperior direction. The purpose of this study was to investigate bony changes of the mandibular condyle when the surface normally not subjected to significant loading was subjected to functional loading using a unilateral surgical experiment. In addition, we evaluated whether this experiment was suitable as an animal model of condylar resorption due to mechanical loading.

## 2. Material and methods

Fifteen adult male New Zealand white rabbits (weighing 3.0–3.5 kg) were used in this study. All experimental procedures were undertaken in compliance with the guidelines for the care and use of animals described in the guidelines of the local ethics committee of Seoul National University. The animals were kept at constant temperature and humidity during the experimental period. The 12 animals were allocated into two groups of six animals each: 1 mm (group I) and 3 mm (group II) rotation. For groups I and II, operations were only performed on the right side. Therefore, experimental and control condyles were on the right and left sides, respectively. Since the left condyle (control) might be affected by the operation on the right side, six condyles of three rabbits were used as healthy controls without operation (group III).

### 2.1. Surgical procedure for mandibular unilateral body osteotomy in rabbits

Animals in groups I and II underwent operation under general anesthesia using an intramuscular and intravenous injection of Zoletil (0.4 ml/kg; Virbac Laboratories, Carros, France) mixed with Rompun (10 mg/kg; Bayer HealthCare AG, Leverkusen, Germany). After disinfection of the right mandible with 10% Povidone Iodine topical solution (Gumi, Gimpo, Korea) and subcutaneous injection

of 2% lidocaine containing 1:100,000 epinephrine (Huons, Seongnam, Korea), an incision of about 4 cm in length was made along the mandibular inferior border from the back of the mouth on the right side. Subperiosteal dissection was done to expose the mandibular body from the mental foramen to anterior margin of the masseteric fossa. For unilateral oblique vertical body osteotomy (UOVBO) of the mandible, a buccal cortical osteotomy was performed vertically between the premolar and the molar with a fissure bur under copious irrigation with normal saline. This site on the osteotomy line can make the firm fixation on the mandibular body with adequate thickness, because the masseteric fossa is too thin to fix a miniplate. To obtain maximum bone contact during fixation, an oblique vertical osteotomy was performed from the buccal vertical osteotomy line toward the distolingual cortical bone for as far as possible. UOVBO was performed completely using a chisel and mallet. The PS was rotated counterclockwise. The bony interference between the proximal and distal segments was ground with a vulcanite bur. For group I, the proximal segments were fixed with the inferior border of the PS being 1 mm higher than that of the distal segment (Fig. 1A). For group II segments, the difference between the inferior borders was 3 mm (Fig. 1B). The amount of rotation was measured using a normal surgical ruler with 0.5 mm graduations during the surgery. Fixation was performed with a 4-hole titanium miniplate (Jeil Medical Corporation, Seoul, Korea). Sutures were made using a 4-0 absorbable suture (Ailee Co. Ltd., Busan, Korea). Postoperatively, all rabbits received antibiotics with an intramuscular injection of Cefazolin (55 mg/kg; Chongkundang, Seoul, Korea).

### 2.2. Micro-computed tomography analysis

Animals were sacrificed four weeks after surgery with an overdose of KCl (Jeil Pharmaceutical Co. Ltd., Daegu, Korea), and their mandibles were removed and fixed in 10% formalin for one week. Micro-computed tomography (micro-CT) scans were taken for quantitative evaluation of bony changes of the rabbit condyles using the SkyScan 1172VR microfocus X-ray system (SkyScanVR, Kontich, Belgium). The SkyScan 1172VR microfocus X-ray system is equipped with a microfocus X-ray tube with a focal spot of 2 μm, producing a cone beam that is detected by a 12-bit cooled X-ray camera CCD fiber-optically coupled to a 0.5 mm scintillator. The resulting images were 1000 × 1000-pixel square images with an aluminum filter used to produce optimized images. Reconstruction and analyses were performed using NRecon reconstruction and CTAn 1.8 software, respectively. To measure the bony structure of the mandibular condyle, a rectangular area was selected as the region of interest (ROI) in a two-dimensional image of the midsagittal plane of the mandibular condyle. The inferior border of the ROI was 6 mm inferior to the most superior point of the condyle. The anterior one third of the condyle was evaluated because this area was most affected by loading after rotational change of the PS. Only anterosuperior bony changes of the mandibular condyle were evaluated in the defined ROI. The pixel zone representing ossification in the defined ROI was then reconstructed three-dimensionally by creating a volume of interest in the lower and upper ranges of the threshold using grayscale units. Then, microarchitecture parameters, including bone volume (BV), bone volume/tissue volume (BV/TV), trabecular thickness (Tb.Th), trabecular number (Tb.N), and trabecular separation (Tb.Sp) were obtained using CTAn 1.8 software according to the manufacturer’s instructions. To measure bone mineral density (BMD), attenuation data for ROIs were converted into Hounsfield units and expressed as a value of BMD using phantom scans. BMD values were expressed in terms of grams per cubic centimeter of calcium hydroxyapatite in distilled water. A zero value for BMD corresponded

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