



Sex dependent mechanical properties of the human mandibular condyle



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ABSTRACT

The mandibular condyle consists of articular cartilage and subchondral bone that play an important role in bearing loads at the temporomandibular joint (TMJ) during static occlusion and dynamic mastication. The objective of the current study was to examine effects of sex and cartilage on 1) static and dynamic mechanical analysis (DMA) based dynamic energy storage and dissipation for the cartilage-subchondral bone construct of the human mandibular condyle, and 2) their correlations with the tissue mineral density and trabecular morphological parameters of subchondral bone. Cartilage-subchondral bone constructs were obtained from 16 individual human cadavers (9 males, 7 females, 79.00 ± 13.10 years). After scanning with micro-computed tomography, the specimens were subjected to a non-destructive compressive static loading up to 7N and DMA using a cyclic loading profile (-5 ± 2 N at 2 Hz). After removing the cartilage from the same specimen, the series of loading experiments were repeated. Static stiffness (K) and energy dissipation (W), and dynamic storage (K'), loss (K'') stiffness, and energy dissipation ($\tan \delta$) were assessed. Gray values, which are proportional to degree of bone mineralization, and trabecular morphological parameters of the subchondral bone were also measured. After removal of the cartilage, static energy dissipation significantly decreased ($p < 0.009$) but dynamic energy dissipation was not influenced ($p > 0.064$). Many subchondral bone properties were significantly correlated with the overall mechanical behavior of the cartilage-subchondral bone constructs for males ($p < 0.047$) but not females ($p > 0.054$). However, after removal of cartilage from the constructs, all of the significant correlations were no longer found ($p > 0.057$). The current findings indicate that the subchondral bone is responsible for bearing static and dynamic loading in males but not in females. This result indicates that the female condyle may have a mechanically disadvantageous TMJ loading environment.

1. Introduction

Temporomandibular joint (TMJ) disorder (TMD) is the second most common musculoskeletal symptom, affecting up to 25% of Americans (Detamore et al., 2007). The incidence of TMD is higher in women than men (Halpern et al., 2007). About 10% to 15% of TMD patients have osteoarthritis, characterized by a degenerative joint that results from erosion of articular cartilage and degeneration of subchondral cortical and trabecular bone (Ingawale and Goswami, 2009; Tanaka et al., 2008a). The TMJ is a synovial joint that has a fibrocartilaginous articular disc located between the articular cartilage of the articular eminence of the temporal bone and the mandibular condyle (Ingawale and Goswami, 2009; Tanaka et al., 2008b). During static occlusion and dynamic mastication, muscle contractions associated with TMJ movement can provide combinations of loading in

various directions on the articular surface of the joint (Herring et al., 2002; Liu and Herring, 2000; Tanaka et al., 2008b). It was observed that the peak loading direction on the mandibular condyle bone is in compression during occlusion and masticatory cycles (Herring et al., 2002; Liu and Herring, 2000). Degeneration of the TMJ can develop when these applied loads surpass the adaptive capacity of the mandibular condyle (Chen et al., 2009; Milam, 2005; Zarb and Carlsson, 1999). As such, it has been indicated that the mechanical properties of the mandibular condyle construct are the most important factors in determining risk and progression of TMD (Ingawale and Goswami, 2009; Tanaka et al., 2008b).

The mandibular condylar cartilage plays an important role in controlling mechanical load transmission to the underlying subchondral bone (Singh and Detamore, 2009). Effective dissipation of the high static and dynamic loading energy during occlusion and mastication is

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important in protecting against the development of damage at the mandibular condyle and maintaining stability of the TMJ. Both cartilage and bone have viscoelastic properties that are responsible for time-dependent energy dissipation (Dong et al., 2004; Kim et al., 2010; Lamela et al., 2013; Tanaka et al., 2006; Yeni et al., 2006). Dynamic mechanical analysis (DMA) is a well-established method to assess viscoelastic properties of polymers (Menard, 1999; Stroede et al., 2012). DMA has successfully measured viscoelastic parameters including dynamic stiffness and energy dissipation capacity ($\tan \delta$) for cartilage using cyclic indentation loading (Lamela et al., 2013; Tanaka et al., 2006) and those for bone using non-destructive cyclic loading with different frequencies (Dong et al., 2004; Yeni et al., 2006). However, a lack of knowledge exists about the mechanical characteristics of the cartilage-subchondral bone construct of the human mandibular condyle of the TMJ.

It has been observed that tissue mineral density and trabecular morphological parameters of bone can explain its elastic, viscoelastic, and fracture behavior because these parameters are significantly correlated with mechanical properties of bone (Keaveny et al., 2001; Kim et al., 2012). We hypothesized that the mechanical behavior of the cartilage-subchondral bone construct may be sex-dependent and related to the subchondral bone properties including tissue mineral density and trabecular morphological parameters. Thus, the objective of the current study was to examine effects of sex and subchondral bone properties on 1) static and DMA based dynamic energy storage and dissipation for the cartilage-subchondral bone construct of the human mandibular condyle, and 2) their correlations with the tissue mineral density and trabecular morphological parameters of subchondral bone.

2. Materials and methods

Fresh human mandibles were obtained from 16 individual cadavers (9 males, 7 females, 79.00 ± 13.10 years) that were provided by the Body Donation Program at The Ohio State University (Fig. 1a). No records of temporomandibular joint disorder (TMD) or gross evidence of it were observed on the specimens. No fixation was performed on any mandibles. The mandibles were stored at -21°C until utilization. After thawing at room temperature, all soft tissues were removed from the mandibular bone surface. Then, mandibular condyles were dissected at the point of sigmoid notch concavity parallel to the occlusal plane in the transverse direction using a low speed saw under water irrigation. A total of 11 right and 5 left side condyles were randomly chosen for the current study.

The mandibular condyle specimens were scanned by a three-dimensional (3D) micro-computerized tomography (micro-CT) scanner (SkyScan 1172-D, Kontich, Belgium) with the scanning and reconstruction voxel sizes set at $27 \times 27 \times 27 \mu\text{m}^3$. The same scanning conditions (70 kV, 141 μA , 0.4° rotation per projection, 8 frames averaged per projection and 120 ms exposure time) were used for all specimens. The subchondral cortical bone (CB) region and trabecular bone (TB) region of the mandibular condyle were digitally isolated using the 3D micro-CT image following the separation procedure illustrated in a previous study (Fig. 1b) (Kim et al., 2012). A heuristic algorithm introduced in previous studies (Kim et al., 2004; Kim et al., 2012) was used to remove non-bone voxels outside the CB maintaining all voxels in the CB and TB regions. Then, the entire TB region was masked using a compartmentalizing method (Buie et al., 2007; Kim et al., 2012). Bone voxels inside the whole mandibular condyle were segmented from non-bone voxels using the same heuristic algorithm. The masked TB region was subtracted from the segmented whole condyle image leaving the CB of the mandibular condyle. Finally, the TB was obtained by subtracting the CB from the segmented whole condyle. All of these steps were performed using open codes in Image J software (NIH).

The gray value of each bone voxel, which is proportional to the degree of bone mineralization (DBM), was identified in the 3D micro-

CT images during the segmentation process. Histograms of gray values were obtained for the whole mandibular condyle (Total), CB, and TB. The average gray values were computed by dividing the sum of gray values by the total number of bone voxels in each region using the gray value histograms of Total, CB, and TB (Fig. 1c, d). The standard deviation (SD) of the gray value histogram was also computed. Low and high gray values (Low_5 and High_5) were determined as the lower and upper 5th percentile values, respectively. In addition, TB morphological parameters were measured using the largest possible rectangular region of interest (ROI) in the TB region. The ROI volume was the same for all specimens. Trabecular bone volume fraction (BV/TV), specific bone surface (BS/BV), thickness (Tb.Th), number (Tb.N), separation (Tb.Sp), and connectivity density (Conn.D), and degree of anisotropy (DA) (Bouxsein et al., 2010; Doube et al., 2010; Kim et al., 2004). An open source code (BoneJ function of ImageJ) was used for bone image processing.

Following the non-destructive micro-CT scanning, static compressive tests and dynamic mechanical analyses (DMA) were performed for all of the mandibular condyles using a commercial loading device (ELF 3230, Bose, MN). The condyles were soaked in normal saline solution for 2 h before testing. Each condyle specimen was glued in the superior-inferior direction on the loading jig (Fig. 2a). All load and displacement data were obtained using a 450 N load cell and a high-resolution (15 nm) displacement transducer, which were integrated with the loading machine. A pre-load of 1 N was applied to secure the contact between specimen and loading jig. Static properties of each condyle specimen were characterized by non-destructive compressive static loading to 7 N and unloading to 0 N under force control with a loading rate of 2 N/second (Fig. 2b). Static compressive stiffness (K) was calculated as the slope of load-displacement during the loading period, and the amount of static energy dissipation (W) was calculated by subtracting the area under the unloading-displacement curve from that under the loading-displacement curve. DMA was consecutively performed following the static testing for the same condyle specimen. A non-destructive compressive cyclic loading (-5 ± 2 N) with scanning frequencies between 0.5 and 3 Hz, and a frequency of 2 Hz was chosen to be utilized regarding human chewing ranges from 0.94 to 2.17 Hz (Po et al., 2011). The phase shift angle (δ) was measured between the dynamic force (F^*) and displacement (d^*) curve during the cyclic loading (Fig. 2c). Dynamic complex stiffness (K^*) is composed of two parameters: the elastic (storage) stiffness (K') and the viscous (loss) stiffness (K''), as defined by $K^* = K' + iK''$, where $K' = K^* \cos(\delta)$ and $K'' = K^* \sin(\delta)$. The phase angle (δ) based tangent δ ($\tan \delta$) was also computed as K''/K' . The K' and K'' of bone represent its abilities to store and dissipate the dynamic loading energy, respectively. As such, the $\tan \delta$ accounts for the relative capacity of dynamic energy dissipation. After DMA, the cartilage of each mandibular condyle specimen was removed using a surgical scalpel. Caution was paid to avoid scratching the condylar surface. The specimens were kept hydrated for the whole process. After cartilage removal, the static loading and DMA tests were repeated.

A finite element model was constructed of one mandibular condyle specimen using the micro-CT image of a 63-year-old male. From the isosurface, a tetrahedral mesh was created in the open-source program PreView (Maas et al., 2012). The model had ~ 1.3 million elements and $\sim 400,000$ nodes. An additional model was created to simulate a cartilage layer on top of the condylar bone surface. In LS-PrePost (LSTC, Livermore, CA), the condylar bone surface was projected outwards to create a cartilage layer 0.22 mm in thickness (Stratmann et al., 1996). The bone and cartilage were modeled using elastic material properties. The Young's moduli of the bone and cartilage were prescribed as 2.29 GPa and 7.8 MPa, respectively (Kim et al., 2015; Singh and Detamore, 2009). The static compressive tests were used as the basis of the boundary conditions for the model. A rigid plate was placed in contact with the bone or cartilage and loaded with a prescribed displacement based on Fig. 2b. Displacement control was

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