Decreased Temporomandibular Joint Range of Motion in a Model of Early Osteoarthritis in the Rabbit

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ABSTRACT

Purpose: Analysis of mandibular biomechanics may help with understanding the mechanisms of temporomandibular joint (TMJ) disorders (TMJDs), such as osteoarthritis (TMJ-OA), by investigating the effects of injury or disease on TMJ movement. The objective of this study was to determine the functional kinematic implications of mild TMJ-OA degeneration caused by altered occlusion from unilateral splints in the rabbit.

Methods: Altered occlusion of the TMJ was mechanically induced in rabbits via a unilateral molar dental splint (n=3). TMJ motion was assessed through three-dimensional skeletal kinematics twice, once before and once after 6 weeks of splint placement with the splints removed allowing three days of recovery. The relative motion of the condyle to the fossa was tracked, as well as the distance between the incisors.

Results: An overall decrease in the range of joint movement was observed at the incisors and in the joint space between the condyle and fossa. The incisor movement decreased from 7.0 ± 0.5 mm to 6.2 ± 0.5 mm right-left, from 5.5 ± 2.2 mm to 4.6 ± 0.8 mm anterior-posterior, and from 13.3 ± 1.8 mm to 11.6 ± 1.4 mm superior-inferior ($p < 0.05$). The total magnitude of the maximum distance between the points on the condyle and fossa decreased from 3.6 ± 0.8 mm to 3.1 ± 0.6 mm for the working condyle and from 2.8 ± 0.4 mm to 2.5 ± 0.4 mm for the balancing condyle ($p < 0.05$). The largest decreases were seen in the anterior-posterior direction for both condyles.

Conclusions: Determining the changes in condylar movement may lead to a better understanding of early predictors in the development of TMJ-OA, and determining when symptoms become a chronic, irreversible problem.

INTRODUCTION

For patients suffering from pain associated with temporomandibular joint (TMJ) disorders (TMJDs), normal life activities such as eating, talking, and even sleeping may be drastically impaired. TMJDs are multifactorial and complex [1]. Signs and symptoms of TMJDs include joint pain, limited mouth opening, deviation of the jaw, clicking, locking, dislocation, and pain in the masticatory muscles during jaw movement [2]. While a heterogeneous syndrome that may involve the joint as well as the muscles of mastication, osteoarthritis (OA) is present in up to 15% of those suffering from TMJD [3], and pain is the primary complaint associated with the loss of function [1, 4, 5]. TMJ-OA is the pathological process of joint degeneration including irreparable abrasion of articular cartilage and thickening and remodeling of underlying bone [6]. A number of potential causes of TMJ-OA have been implicated, including trauma and parafunctional habits such as teeth clenching and grinding.

We hypothesize that altered joint movement/travel path (kinematics), caused by trauma or parafunctional habits, are one of the underlying mechanisms of progressive TMJ-OA. Establishing the link between altered TMJ kinematics and joint degeneration may have an immediate clinical impact. Understanding this relationship may provide guidance to surgeons on the window in which interventions to normalize malocclusion associated with trauma may be most successful. This knowledge may support the use of TMJ kinematic analysis as a diagnostic tool with which to identify the basis for TMJ pain in at least a subpopulation of TMJD patients. They may also suggest that strategies to normalize altered TMJ kinematics may be effective for the treatment of TMJ pain in these patients.

Establishing a link between kinematics and TMJ-OA has been hindered by the dearth of data linking the changes observed in preclinical animal models to the development and

manifestation of the pathology of the human state. Of ongoing debate is the question of whether TMJ disc displacement precedes degeneration of the condylar cartilage. As a potential explanation for the relatively limited efficacy of interventions focused on the TMJ disc, results from our rabbit TMJ-OA model have shown that condylar cartilage remodeling (stiffening and loss of the subchondral layer) occurs with a disc in its proper anatomical position [7]. As the condylar cartilage resembles early stages of OA, it is not known whether TMJ kinematics were altered. If the joint kinematics do not return to normal, they can be a primary driver for further joint degeneration and end stage OA.

The objective of this study was to determine the functional kinematic implications of the TMJ with mild OA degeneration. In our model, OA is caused by altered occlusion from unilateral splints placed for six weeks. It was hypothesized that kinematic analysis would identify an overall decrease in joint motion after six weeks of splinting.

METHODS

Animal Model

Skeletally mature, female, New Zealand White rabbits approximately 1 year in age weighing between 5-7 kg were purchased from Charles River Laboratories International, Inc. (Wilmington, MA). All rabbits were examined by a veterinarian prior to use in the study and were found to be in good health. All animal procedures were approved by the Institutional Animal Care and Use Committee at the University of Pittsburgh and in accordance with the National Institutes of Health guidelines for the use of laboratory animals.

Unilateral dental splints were chosen as the method for altering the occlusion because the splints could be removed at a later point in time and the joint space was not penetrated [8-10]. For three separate procedures, impressions, splint placement, and splint removal, all rabbits were sedated with ketamine and xylazine, and a surgical plane of anesthesia was maintained with isoflurane. Impressions were taken of the upper and lower right molars of the rabbits to create a unique mold for casting the splints as crowns made from non-precious metals (Figure 1). The thickness of each casted splint was approximately 1 mm. During a second procedure, the right maxillary and mandibular molar arches were cleaned and primed, and then the splints were attached to the respective molars with dental cement. Splint placement was verified after 1 week. Splints were removed at 6 weeks, three days prior to the final kinematic data collection. Kinematic data was collected twice, once before impressions and splint placement, and once 6 weeks after splint placement, with the splints removed for three days on three rabbits (n=3). After the second kinematic data collection, the rabbits were euthanized and the whole head was frozen and saved for micro-computed tomography (CT).

Kinematics

While biaxial x-ray systems have been used to assess the impact of injury on the structure-function relationship of knees and shoulders in humans [11-13], it was necessary to determine whether this system could be used for this purpose in the rabbit. Using high resolution micro-CT, we were able to demonstrate the feasibility of this system for the analysis of rabbit TMJ movements during chewing [14]. We determined that rabbit TMJ kinematic were measureable and repeatable, including maximal distances and movement paths for both the incisors and the condyle-fossa relationship.

As per these published methods [14], TMJ kinematics were assessed using a unique high-speed stereo-radiographic system consisting of two sets of 110 kW pulsed X-ray generators (CPX 3100CV; EMD Technologies, Quebec, Canada), 40 cm image intensifiers (Thales, Neuilly-sur-Seine, France) using a 20 cm field of view, and high-speed 4 Megapixel digital video cameras (Phantom v10, Vision Research, Wayne, New Jersey, USA). The rabbits were placed in a radiolucent ventilated cage (Figure 2A) and fed small pieces of dried fruit. Radiographic images were collected at 170 frames/s for 1 s with 1 ms exposures at 4 Megapixel image resolution (Figure 2B). At least three data acquisitions per rabbit were collected. Three rabbits were tested before splint placement and after 6 weeks of splint placement with the splints removed three days prior to the second data collection.

The markerless, model-based technique used for determining 3D kinematics has been extensively described, validated, and tested [11, 13-16], including testing of the rabbit TMJ [14]. Briefly, a 3D model of each bone to be tracked was derived from a CT scan (as described below) (Figure 2C). A virtual model of the biplane x-ray system was created for generating a pair of digitally reconstructed radiographs (DRR's) via ray-traced projection through the CT bone model. The bone model was automatically repositioned within the virtual model until a best match was achieved between the simulated DRR's and the actual radiographic image pair (Figure 2D). This technique determined the 3D positions and orientations of the mandible and the skull for each motion frame. After euthanasia, the entire head, including the mandible and skull, was scanned at high resolution (105 μm isotropic voxels) using a micro-CT system (Inveon micro-CT system by Siemens located at the Rangos Research Center Animal Imaging Core, Children's Hospital of Pittsburgh of UPMC). The mandible and skull bones (with teeth)

were segmented from each other and from soft tissue and reconstructed into both surface and volumetric models using Mimics software (Materialise, Inc. Leuven, Belgium) (Figure 2C).

Anatomical coordinate systems were set up similar to the systems used by Brainerd et al. [17] and in our previous study [14] to determine the joint translations and rotations. The transverse (x) axis was aligned through the center of the condyles, the longitudinal (y) axis was parallel to a plane through the occlusal plane, and the vertical (z) axis was the cross product of x and y (Figure 3A).

The relative motion of each condyle with respect to the fossa was tracked (Figure 3B and 3C). The distance between a fixed point on the condyle and a fixed point on the fossa was measured throughout the chewing cycles (Figure 2Ei). Points were placed at the superior most point of the condyle and in the convex region of the fossa. The relative motions of the centers of the two points were determined in all 3 anatomic planes. The measurement points were placed in the same location on the condyle and fossa for pre and post splint data collection for each rabbit. The pre and post splint motions were analyzed in each direction, as well as in the total magnitude of the vector (Figure 2Eii). The movement of the mandible was somewhat elliptical in the coronal plane and always started to the same side during an acquisition. As established in the literature, the side in which the mandible first moves is the working side, while the other is called the balancing side [18]. The condyles were analyzed by working and balancing condyles as previously established in the literature [14, 18]. Of the three rabbits, one rabbit consistently had the left condyle as the working condyle, one rabbit had the right condyle consistently as the working condyle, and the third rabbit the working condyle switched from the right condyle pre splint to left condyle post splint. Again, the splint was placed on the right molars.

The relative motion near the incisors was measured as well. Since, the incisor teeth were observed to grow and change shape from the treatment they were not directly included in the analysis. To be consistent with the changing incisors from pre and post splint, the incisor markers were instead placed on the skull and mandible at the center of the incisors at the interface where the teeth met the bone on the posterior side (Figure 3D).

Based on our previous study [14], some of the rotation-translation relationships were shown to be linear and repeatable, so the slopes of these relationships were also compared. The slope of the movement of the condyles in the superior-inferior direction with respect to the transverse rotation during the first 50% of closing was determined for both the working and balancing condyles. The slopes of the paths from the incisal point in the superior-inferior direction with respect to the transverse rotation and the incisal point in the anterior-posterior direction with respect to the vertical rotation were also determined. Only one acquisition was analyzed for each rabbit pre and post splint, if the acquisition had at least three complete chewing cycles. All kinematic data was reported as an average $\pm$ standard deviation. Data was tested for normality with an Anderson Darling Test for Normality and found to be normal. A repeated measures ANOVA with Tukey's post-hoc test ($p < 0.05$) was used to assess the differences for all kinematics values before and after splinting.

RESULTS

Kinematics

Overall, the altered occlusion from the splint caused the range of motion of the condyles and the teeth to decrease. The distances the condyles traveled on both the working and balancing sides significantly decreased after splinting (Table 1). The total magnitude of the maximum distance between the points on the condyle and fossa decreased by 15.0% for the working condyle and by 8.9% for the balancing condyle ($p < 0.05$). Figure 4A and B shows the total magnitude of the condyle-fossa distance over time and the decrease in distance traveled from the splint. Breaking the movement into the vector components, both the anterior-posterior displacement and the superior-inferior displacement significantly decreased ($p < 0.05$). The anterior-posterior displacement decreased by 20.5% and the superior-inferior by 8.9% for the working condyle and for the balancing condyle the displacements decreased by 14.9 and 13.6%, respectively (Table 1). Figure 4C and D show the superior-inferior motion with respect to the anterior-posterior motion, for the working and balancing condyles, pre and post splint. The right-left movement of the condyles was minimal and thus, not reported.

The paths of the condyles were analyzed by determining slope of the superior-inferior distance with respect to the transverse rotation during the first 50% of the closing cycle. The slopes for the working condyle significantly increased by 13.7% after splinting ($p < 0.05$) (Table 1). The shapes of the curves are shown in Figure 4E and F for the working and balancing condyles, respectively.

The distances the incisors traveled also significantly decreased after splinting ($p < 0.05$) (Table 2). The right-left movement decreased by 11.2%. The anterior-posterior movement decreased by 16.1%. The superior-inferior movement decreased by 13.0%. The shapes of the

curves are shown in Figure 5A and B. Figure 5C shows the change in movement from pre to post-splint in the coronal plane.

DISCUSSION

An overall decrease in the range of joint motion at both the condyles and the incisors was observed after placement of dental splints for six weeks. The persistent change in kinematics is likely due to the abnormal occlusion and the degeneration of the condylar fibrocartilage observed in our previous studies [7, 19]. The kinematics of the rabbit TMJ at the condyles and the incisors were easily measured with our non-invasive system before and after splint placement to determine changes in the joint movement. The decreased range in incisal movement post splint was comparable to a previous TMJ disc injury study [20]; however, the specific values could not be compared due to different measurement techniques. The pre splint control incisal data was also consistent with our previous study and with other normal rabbit kinematic studies [14, 18, 20, 21].

Determining the changes in condylar movement may lead to a better understanding of early movement predictors in the development of TMJDs. This knowledge of dynamic TMJ function through 3D skeletal kinematics is essential for investigating the effects of injury or disease in the TMJ. The altered occlusion model used in this study has been shown to produce condylar fibrocartilage degeneration that would likely not be detected by conventional MRI [7, 19]. Understanding the changes in joint movement that occur from various types of TMJDs could allow for earlier diagnosis and treatment, for example, if a certain signature of changes in movement are observed earlier before onset of severe joint degeneration. As such, our study

suggests that decreases in 20% anterior-posterior displacement of the working condyle might be indicative of condylar degeneration and early stage TMJ-OA.

A limitation of this study was that only one microCT scan could be obtained per rabbit head after the final time point. Ideally a microCT scan would be collected for each time point that kinematic data was collected, this would also allow for analysis of the effect of the splints on the bony structure. However, due to the size limitations of the scanner, the heads could only be scanned after euthanasia and decapitation. With only one scan, it was assumed that the bony structures of the joints were not affected by the treatment. Another assumption of this study was that the bony structure of the skull and mandible were rigid. However, it has been shown that the mandible can deform during joint motion [22, 23], which led to some minor errors in the DDR matching process. There were a few frames in each chew cycle where the mandible and fossa were barely touching along the edges of both of the condyles. Rabbits' teeth are also constantly erupting, so the teeth were ignored throughout the analysis. In future studies with longer time points the teeth may need to be filed down to their original height. The possible changes in muscle performance were also not considered in this study. Even with these limitations, this study provided an important initial understanding of what can change in the TMJ motion from inducing TMJ-OA with altered occlusion.

The splint model used in this study was advantageous because it allowed for removal of the splints, and the joint space was not penetrated. Future work will need to look at various lengths of time between splint placement and removal and allow time for recovery to understand if and when the change in joint movement has the potential to correct itself and the joint to heal or when the TMJ-OA becomes chronic and permanent. The potential exists with this type of TMJD animal model to combine the kinematic analysis with other assessments such as pain

measurements and mechanical assessments of the TMJ tissues, which will allow for multidisciplinary studies of various types of TMJDs. The combination of assessment methods will allow for a fuller understanding of the TMJ disease progression in this and other animal models.

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Conflict of Interest Statement

The authors have no conflicts of interest to disclose.

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Table 1.

Measurement	Pre Splint	Post Splint
<i>Working Condyle AP (mm)</i>	4.9 ± 0.8	3.9 ± 0.4*
<i>Working Condyle SI (mm)</i>	2.2 ± 0.5	2.0 ± 0.2*
<i>Working Condyle Magnitude (mm)</i>	3.6 ± 0.8	3.1 ± 0.6*
<i>Working Condyle SI vs Transverse Rotation (mm/°)</i>	0.21 ± 0.02	0.24 ± 0.04*
<i>Balancing Condyle AP (mm)</i>	4.3 ± 0.8	3.6 ± 0.2*
<i>Balancing Condyle SI (mm)</i>	1.8 ± 0.3	1.6 ± 0.14*
<i>Balancing Condyle Magnitude (mm)</i>	2.8 ± 0.4	2.5 ± 0.4*
<i>Balancing Condyle SI vs Transverse Rotation (mm/°)</i>	0.07 ± 0.02	0.08 ± 0.04
<i>Incisors AP (mm)</i>	5.5 ± 2.2	4.6 ± 0.8*
<i>Incisors SI (mm)</i>	13.3 ± 1.8	11.6 ± 1.4*
<i>Incisors RL (mm)</i>	7.0 ± 0.5	6.2 ± 0.5*
<i>Incisors RL vs Vertical Rotation (mm/°)</i>	-1.38 ± 0.06	-1.36 ± 0.09
<i>Incisors SI vs Transverse Rotation (mm/°)</i>	1.12 ± 0.08	1.14 ± 0.08*

Figure and Table Captions

Figure 1. Unilateral molar bite raising splints. A) The splints were made by first taking an impression of the molars. B) A plaster mold was made from the impressions. C) The metal splints were cast as crowns on the molds, and the superior and inferior views are shown.

Figure 2. Overview of methods for kinematic data collection. A) High-speed biplane radiography was used for image collection. Rabbits were placed in a radiolucent box between two sets of x-ray generators and image intensifiers with high speed cameras. B) Sample radiographic image from the high resolution cameras at one frame in time. C) 3D bone model

created from a microCT scan of the rabbit head. D) For 2D to 3D registration, the collected radiographic images (pink) are matched frame by frame with the virtual radiographic images (green) from the CT scan. One of the two combined images is shown for each the mandible (i) and the skull (ii). E) 3D distances can be measured. (i) Vector indicating the distance between the condylar point and the fossa point (the condyle is labeled with C and the fossa with F). (ii) Sample displacement curve. The picture in E is labeled by directions: A-anterior, P-posterior, M-medial, L-lateral, S-superior, I-inferior.

Figure 3. Kinematic analysis tools were used to set up the measurements. A) The anatomical coordinate system for the mandible was set up with the transverse axis (x-red) aligned through the center of the condyles, the longitudinal axis (y-green) parallel to a plane through the occlusal plane (where the teeth meet), and the vertical axis (z-blue) was the cross product of the transverse and longitudinal axes. The skull coordinate system was set up in a similar manner (not shown). B) Condylar measurement point, anterior view of left condyle. C) Fossa measurement point, inferior view of left fossa. D) Mandible incisor measurement point, superior view. Pictures are labeled by directions: A-anterior, P-posterior, M-medial, L-lateral, Le-left, R-right, S-superior, I-inferior.

Figure 4. Condyle-fossa relationships pre and post splint. A) Working and B) Balancing side condylar kinematics showing the total condyle-fossa distance with respect to time, pre and post splinting. C) Working and D) Balancing side condylar kinematics showing the superior-inferior condyle-fossa distance with respect to the anterior-posterior condyle-fossa distance, pre and post splinting. E) Working and F) Balancing side condylar kinematics showing the superior-inferior

condyle-fossa distance with respect to the transverse rotation, pre and post splinting.

Representative curves from one rabbit shown.

Figure 5. Patterns of incisor movement pre and post splint. A) The change in distance of the mandible incisors from the skull incisors with relation to time pre splint and B) post splint. C) Relationship of the mandible incisors to the skull incisors in the coronal plane pre and post splint. Three chewing cycles are shown in each graph. Representative curves from one rabbit shown.

Table 1. Kinematics results for rabbits before and after splinting for 6 weeks, with the splint removed three days prior to testing. The data was separated into the working and balancing condyle and the incisors. AP: anterior-posterior movement. SI: superior-inferior movement. RL: right-left movement. The average and standard deviation values are presented.

*Differences were significant when $p < 0.05$.

Supplemental Video Information

Videos illustrating the chewing pattern of a rabbit pre and post splint have been included as supplementary online information.

Video Captions

Video 1: Illustrates the chewing pattern of one rabbit before splinting, from an anterior view. It should be noted that the CT scans are from post splinting.

Video 2: Illustrates the chewing pattern of the same rabbit in video 1 post splinting, with the splint removed for 3 days prior to data collection, from an anterior view.

Video 3: Illustrates the movement of the working condyle of the same rabbit pre splint, from a posterior and slightly superior view.

Video 4: Illustrates the movement of the working condyle of the same rabbit post splint, from a posterior and slightly superior view.

Figure1A

Figure1B

Figure1C

Figure2A

Figure2B

Figure2C

Figure2Di

Figure2Dii

Figure2Ei

Figure2Eii

Figure3A

Figure3B

Figure3C

Figure3D

Figure4A

Figure4B

Figure4C

Figure4D

Figure4E

Figure4F

Figure5A

Figure5B

Figure5C