J Oral Pathol Med (2005) 34: 157–63

www.blackwellmunksgaard.com/jopm

Histologic evaluation of temporomandibular arthritis induced by mild mechanical loading in rabbits

Kazuma Fujimura, Susumu Kobayashi, Toshikazu Suzuki, Natsuki Segami

Department of Oral and Maxillofacial Surgery, Kanazawa Medical University, Ishikawa, Japan

BACKGROUND: We still lack knowledge of causative factors in arthritis related to temporomandibular disorders (TMD). The goal of the present study was to investigate whether applying a mechanical loading on the glenoid fossa can induce arthritis.

METHODS: Coil springs were placed in 24 rabbits so as to exert a force of 100 *g* between the orbital edge and the antegonial notch. At 1, 2, 4 and 8 weeks after the surgery, six samples of the temporomandibular joint (TMJ) were removed for histologic examination.

RESULTS: The results showed that mild synovitis began 1–2 weeks after the start of loading, and the degree of synovitis was significant at 4 weeks, and that morphologic changes occurred in the articular eminence and condyle, while type II collagen in the cartilage of the articular eminence degraded prior to that in the condyle.

CONCLUSIONS: Our results revealed that mild, continuous mechanical loading to the glenoid fossa induces synovitis of the articular capsule, and induces organic changes of the articular cartilage without destroying these tissues.

J Oral Pathol Med (2005) **34**: 157–63

Keywords: arthritis; mechanical loading; temporomandibular joint disorders

Introduction

Recently, many clinicians have elucidated the pathophysiology of temporomandibular disorders (TMD), and inflammatory changes have been clarified in the synovium inside the articular capsule of the temporomandibular joint (TMJ) (1–5). In the patients with arthralgia, inflammation of unknown etiology develops in the synovial tissue and disk displacement and/or disk deformities were coincidentally noticed during the clinical examinations. Loading on the glenoid fossa has long been assumed to be one of the causes of TMD. Loading occurs mainly at night because of bruxism, clenching, etc., and is considered to induce TMD (6). Recently, two hypotheses have been proposed; one stating that this overloading induces hypertrophic response in the subchondral bone and the posterior band of the disk resulting in a decrease of the joint space, alteration in the form of the joint surface and inducing disk displacement before long (7, 8), and the other stating that the free radicals which are produced under overloading of the TMJ, degrade hyaluronic acid which inhibits the phospholipase A_2 activity. Then, lysis of the surface-active phospholipids in association with lubricin, which acts as an extremely efficient boundary lubricator, occurs. Finally, friction between the disk and the surface of glenoid fossa is generated. The surface of the articular eminence becomes exposed with subsequent disk displacement (9, 10). Although we still do not know distinct causes for TMD, loading is at least considered as one of the indispensable candidates. It has been reported that the oral splint therapy is effective in 70–90% of patients with TMD (11) that it mitigates the articular symptoms by reducing the load on the glenoid fossa (6) and inhibiting any abnormal muscle spasms, and that reducing the loading and increasing the joint space by intra-oral vertical ramus osteotomy result in improvement of the joint pain and joint dysfunctions (12, 13). These findings might support the aforementioned hypotheses. However, although many clinical studies have been conducted, there have been no insights into the pathophysiology of TMD. There are two reasons for this. The first reason is the versatility of the clinical manifestations presented by the patients with TMD because of the complex anatomical structure and specific articular movement. The intra-articular disk separates the structure of the TMJ into an upper joint compartment and a lower joint compartment (14), which is considerably different from the structure of the other joints, with the exception of the sternoclavicular joint. When there is a joint morbidity, the disk is displaced anteriorly or medially, and various signs and symptoms also present (15). The second reason is that performing biopsy of the pathologically changed tissue is useful to elucidate the etiology of TMD, but the

Correspondence: Kazuma Fujimura, Department of Oral and Maxillofacial Surgery, Kanazawa Medical University, Kahoku-gun, Ishikawa 920-0293, Japan. Tel.: +81 76 286 2211 (ext. 7019). Fax: +81 76 286 2010. E-mail: 1-kkk@kanazawa-med.ac.jp
Accepted for publication September 21, 2004

quantities of tissue that can be obtained, and the biopsy sites are limited. For example, the tissues that can be taken include an extremely small amount of synovial tissue, obtained during arthroscopy; also the saline-diluted synovial fluid with a low protein content, obtained during arthrocentesis; and the disk and synovial tissue, obtained during meniscectomy, which is a rarely performed procedure. These specimens are usually taken from patients with advanced clinical stages, and so it is also impossible to obtain normal articular tissue. Therefore, in order to assess the sequential pathophysiology of TMD and to develop new treatments, it is necessary to establish a reproducible model of TMD. For assessment of the etiology of synovitis or degeneration in TMJ, the arthritis model produced by extra-articular procedure is favorable. We established the following hypothesis: mild continuous mechanical loading via the condyle to the glenoid fossa induces mild non-bacterial inflammation, aggravates the intra-articular environment, which consists of proteins and glycoproteins of the synovial fluid, and increases the friction coefficient during the articular movement. Consequently, denaturation and defensive reactions in the articular cartilage and bone occur in real time. In our present study, we created minor disorders of occlusion and mastication by exclusively applying a mild, continuous mechanical load to the glenoid fossa, and observed the morbid changes in the intra-articular tissues.

Materials and methods

This study was conducted in accordance with the guidelines for animal experiments in Kanazawa Medical University.

Preparation of loading-induced arthritis

Twenty-four Japanese male white rabbits, weighing 2.9–3.1 kg, were anesthetized by injecting 30 mg/kg of sodium pentobarbital into the marginal vein of the ear. Following infiltrating local anesthesia, and the inferior margin of the left mandibular ramus was exposed. After two holes were made in the antegonial notch, a stainless steel wire (0.5 mm in diameter) was introduced through the holes, which were reinforced with a stainless steel plate (0.1 mm in thickness). On the contrary, the orbital edge and the proximal area of zygomatic arch, about 7 mm far from the TMJ capsule, were exposed, two holes were made, and stainless steel wire (0.4 mm in diameter) was introduced. Then, the subcutaneous tissue and skin were sutured tightly, and a coil spring was placed between these wires with 100 ***g*** force. In these animals, loading was applied unilaterally to the TMJ using the coil spring, which was 'Helical Extension Spring' (SUS304WPB, JIS-G 4314, Accurate Sales Co. Ltd, Saitama, Japan). These rabbits were divided into four groups as follows: 1-week group, 2-week group, 4-week group, and 8-week group ($n = 6$ each). No antibiotic was administered to any animal. Six joints in another six rabbits that served as a control group were not subjected to any treatment.

Histologic examination

At 1, 2, 4 and 8 weeks after the surgery, the six animals in each group were killed by intravenous injection of excess pentobarbital. Their heads were fixed in 4% paraformaldehyde for 2 days. Samples of about 20 × 20 mm in size including the TMJ on the treated side were removed and fixed again in 4% paraformaldehyde for 1 day, then decalcified with 10% ethylenediaminetetraacetic acid (EDTA) solution for 4 weeks and embedded in paraffin. Then, serial sagittal sections (3–5 μm in thickness) were sliced and stained with hematoxylin and eosin (H & E) for microscopic examination. Staining for type II collagen that constituted the articular cartilage was done, too. The endogenous peroxidase was blocked with 1% hydrogen peroxide in methanol for 30 min at room temperature. The sections were treated with proteinase K (Dako, Carpinteria, CA, USA) for 6 min and treated with antihuman collagen II (Fuji Chemical Co. Ltd, Toyama, Japan; 1:50 diluted rate) for 1 h at room temperature. Color development was performed with Envision (Dako) for 30 min and DAB Kit (Dako). Counterstaining was performed using hematoxylin. Histologic photomicrographs were taken with a digital camera (Olympus BX41® and DP12®; Olympus Co. Ltd, Tokyo, Japan), and image processing was undergone with the software PHOTOSHOP (version 7.01; Adobe Systems, Inc., Park Avenue, CA, USA).

Histopathologic scores

Histopathologic grading of the synovial tissue was performed according to Gynther system (16). We previously evaluated the accuracy of this histologic grading system for assessing the degree of synovial inflammation in the TMJ in arthroscopically obtained synovial tissues, and we found it reasonable for evaluating synovitis in TMD (17). The intensity of inflammation in the synovium was quantified with following criteria; synovial lining cell layers: (i) normal, one to two cell layers (0 point), (ii) two to three cell layers (1 point), and (iii) three to five cell layers (2 points), five or more cell layers (3 points). Vascularity: (i) a limited number (< 5) of blood vessels/mm^2 (0 point), (ii) five to 10 small blood vessels/mm^2 (1 point), (iii) 11–15 small blood vessels/mm^2 (2 points), and (iv) more than 15 small blood vessels/mm^2 (3 points). Inflammatory cells: (i) zero to two inflammatory cells/mm^2 (0 point), (ii) three to 10 inflammatory cells/mm^2 (2 points), (iii) 11–50 inflammatory cells/mm^2 (5 points), and (iv) more than 50 inflammatory cells/mm^2 (10 points). Also quantitative grading of the cartilage of the condylar heads and articular eminences was performed according to a modified Mankin scoring system (18) and a modified Wilhelmi rating system (19), which was used for osteoarthrosis in the human hips or the knee joints. Structure: (i) normal (0 point), (ii) superficial irregularity and erosion (1 point), (iii) defect in the deep cartilage (2 points), and (iv) defect of the entire cartilage extending to the calcified tissue (3 points). Cartilage staining in the immunohistologic examination: (i) normal (0 point), (ii) slight or moderate reduction of the stained area (1 point), (iii) severe reduction (2 points), and (iv) no dye (3 points).

Statistical analysis
The data of the synovitis grading values or the quantitative grading values were analyzed using Whelch's *t*-test, with $P < 0.05$ being accepted as significant.

Results

None of the rabbits treated died during the study period. Postoperative infection occurred in two rabbits at the metal plates in the antegonial notch and in one rabbit at the proximal area of the zygomatic arch. However, the joints of these rabbits were intact because of the distance between the TMJ and the operated area.

Histologic findings
Typical histologic findings in each group are shown in Figs 1–5, although the microscopic findings varied among the individual animals. Type II collagen that composed the articular cartilage was detected with the antihuman type II collagen antibody. In the control group, the articular disk that intervenes between the articular eminence and condyle had thick posterior band and anterior band separated by a thinner intermediate zone (Fig. 1a) (20, 21). The synovial membrane in the upper joint compartment has generally one or two cell layers. No inflammatory cells under the synovial lining were observed (Fig. 1b). The surfaces of the articular eminence and condyle were covered with thin fibrous tissue and were smooth with no clefts. The cartilage layers of the condyle and articular eminence had a constant thickness, and the following three stratums of cartilage were seen: articular surface zone, proliferative zone, and hypertrophic zone. However, these stratums were observed more distinctly in the condyle than in the articular eminence (Fig. 1a). Type II collagen was stained prominently in the surface of the articular eminence and condyle, and most of the deep area was not stained (Fig. 1c). In the 1-week group, the synovial

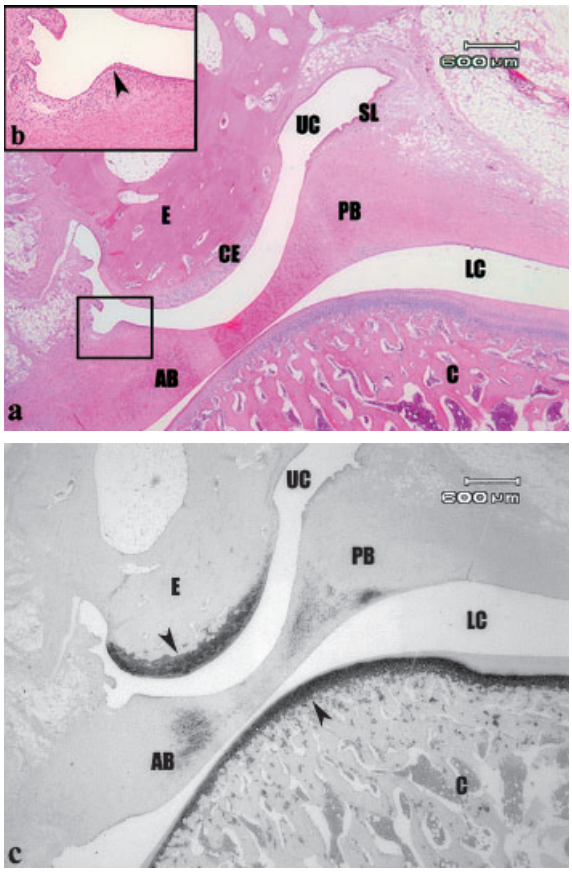

Figure 1 (a) Low-power microphotograph of a mid-sagittal joint view in the control group [hematoxylin and eosin (H & E) stain, original magnification ×12.5] and (b) high-power microphotograph of boxed area in (a) (×100). The surfaces of the articular eminence and condyle that covered with an inner fibrous tissue and were smooth (a). One or two cell layers and no inflammatory cells under the synovial lining were observed (arrowhead) (b). The surfaces of the articular eminence and condylar head were stained prominently (arrowheads), and most of the deep area in the condyle and eminence were not stained (c; immunologic staining for type II collagen, original magnification, ×12.5). AB, anterior band; C, condylar head; CC, condylar cartilage; CE, cartilage of eminence; DS, dilated synovium; E, articular eminence; LC, lower compartment; PB, posterior band; SL, synovial lining cells; SS, synovial stroma; UC, upper compartment.

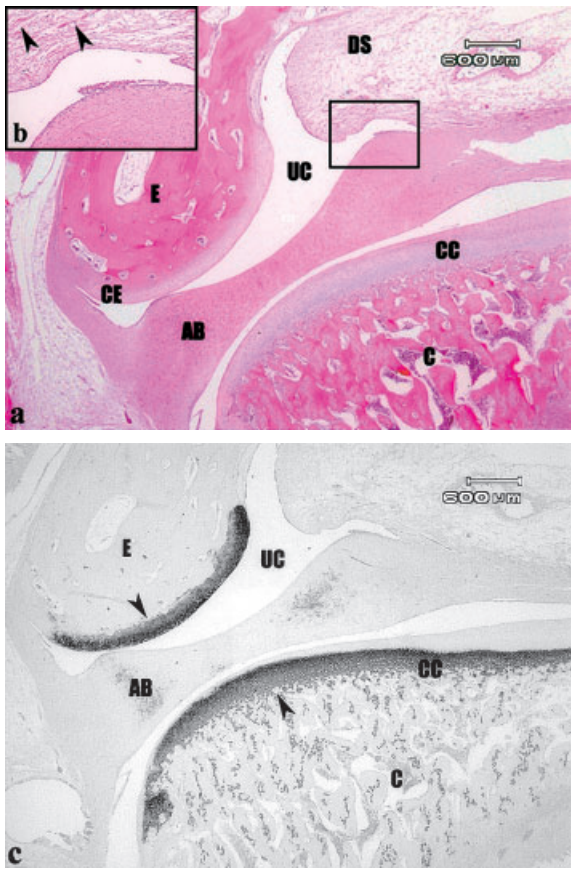

Figure 2 (a) Low-power microphotograph of a mid-sagittal joint view in the 1-week group [hematoxylin and eosin (H & E) stain, original magnification ×12.5] and (b) high-power microphotograph of the boxed area in (a) (×100). In general, the synovial lining cell layers slightly increased in parts of the anterior or posterior synovial portion of the upper joint cavity (a), and usually, one to four synovial cell layers were observed. The synovial stroma with a few capillary vessels was dilated (arrowheads) (b). There were no changes in the cartilage of articular eminence and condylar head (arrowheads) (c; immunologic staining for type II collagen, original magnification, ×12.5).

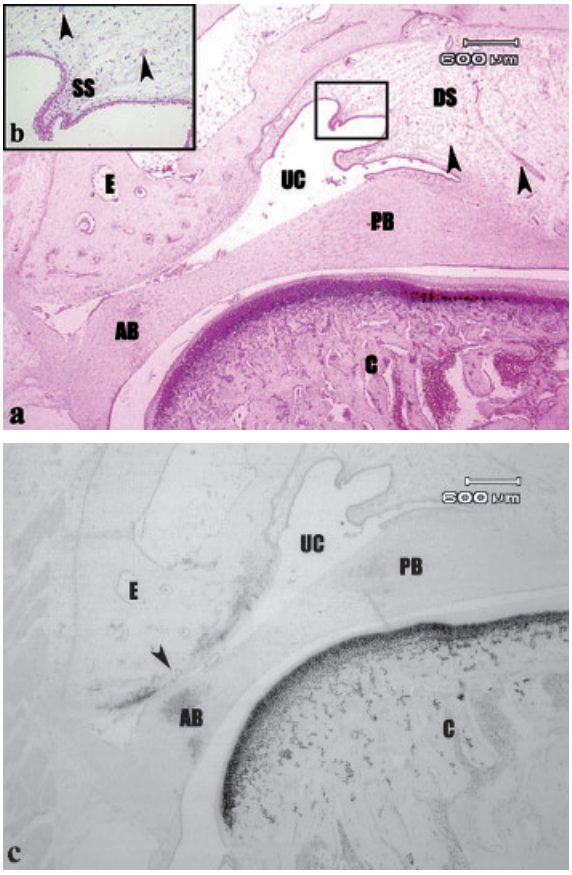

Figure 3 (a) Low power microphotograph of a mid-sagittal joint view in the 2-week group [hematoxylin and eosin (H & E) stain, original magnification, ×12.5]. (b) High-power microphotograph of the boxed area in (a) (×100). The synovial stroma with a vascular dilatation (arrowheads) increased in the upper cavity (a). Two to five of synovial lining cells were observed and capillary vessels (arrowheads) were also observed under the synovial lining (b). Thin cartilage layers and some defects of cartilage in the eminence were observed. The type II collagen in the vicinity of the surface of the articular eminence (arrowhead) was faintly stained in spite of no apparent configurative alternation as shown (c; immunologic staining for type II collagen, original magnification, ×12.5).

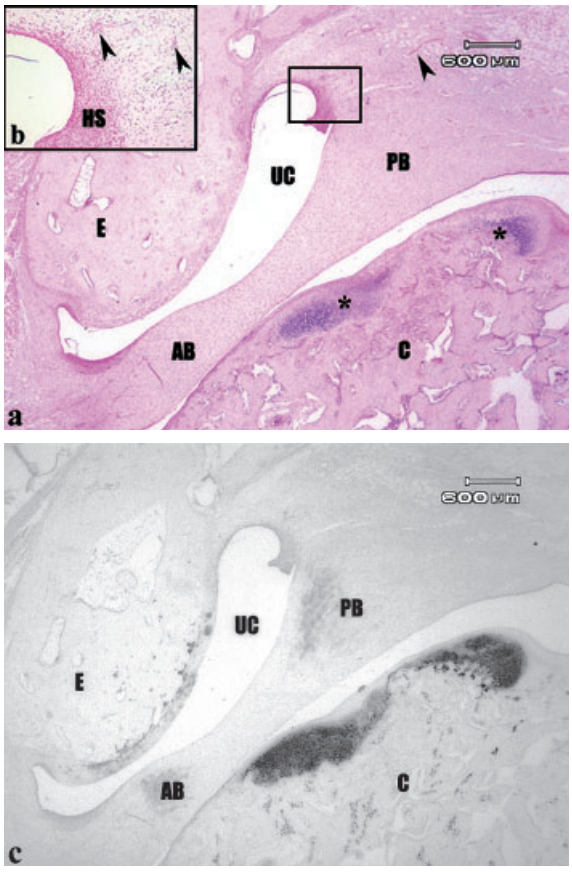

Figure 4 (a) Low power microphotograph of a mid-sagittal joint view in the 4-week group [hematoxylin and eosin (H & E) stain, original magnification, ×12.5] and (b) high-power microphotograph of the boxed area in (a) (×100). Hyperplasia of the synovium in the upper joint cavity and increased vascular dilatations (arrowheads) under the synovial lining cells also were observed (b). Thin cartilage layers in the crest of the eminence and disappeared cartilage in the condylar surface are observed (a). Asterisk indicates a cartilage of condylar head. Mottled staining of type II collagen is observed in the surface of the eminence and deformed condylar head (c; immunologic staining for type II collagen, original magnification, ×12.5).

lining cell layers slightly increased in parts of the anterior or posterior synovial portion of the upper joint cavity, but in general, one to four synovial cell layers were observed. The synovial stroma with a few capillary vessels was dilated (Fig. 2a, b). In general, there were no changes in the surface of the articular eminence and condylar head (Fig. 2c). In the 2-week group, the synovial stroma with a vascular dilatation projected into the mid-upper joint cavity and two to five synovial cell layers were observed (Fig. 3a, b). Up to 2 weeks, in general, slight irregularity of the surface of the fibrous layer in the condylar head and articular eminence was observed, but type II collagen was reduced in the articular eminence (Fig. 3c). In the 4-week group, hyperplasia of the synovium in the upper joint cavity and increased vascular dilatations under the synovial lining cells were observed (Fig. 4a, b). Hemocytes were frequently observed in the upper and lower joint cavities. Thin cartilage layers in the surface of the articular eminence and disappeared cartilage in the condylar surface were observed. The three stratums of cartilage were not observed in both the articular eminence and condyle (Fig. 4a). The type II collagen was faintly stained in the surface of the articular eminence. Mottled staining was observed in the surface of the condylar head (Fig. 4c). In the 8-week group, the synovial lining cells slightly increased in parts of the anterior or posterior synovial portion of the upper joint cavity, but usually two to four synovial cell layers were observed (Fig. 5a, b). Fibroblast hyperplasia with a lot of inflammatory cells, which seemed-like villous, was observed (Fig. 5b). In the articular eminence, type II collagen disappeared and the underlying trabeculae were exposed. Deformed condylar head and osteophyte-like projection with thick cartilage were observed (Fig. 5c). Condylar deformities including osteophyte-like projection were observed in three of six joints and predominantly occurred in the anterior region of the condylar head. In general, at 2–4 weeks, degeneration of the

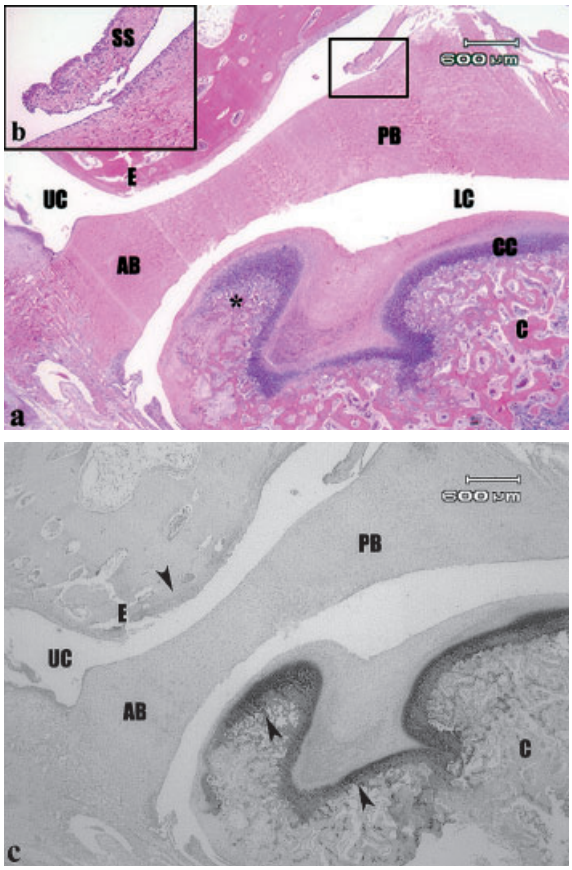

Figure 5 Low-power microphotograph of a mid-sagittal joint view in the 8-week group [hematoxylin and eosin (H & E) stain, original magnification, ×12.5] and (b) high-power microphotograph of the boxed area in (a) (×100). The synovial lining cells slightly increased in parts of the anterior or posterior synovial portion of the upper joint cavity (a), but usually two to four synovial cell layers were observed. Fibroblast hyperplasia and a lot of inflammatory cells in the synovial tissue that seemed like a villous were observed (b). The cartilage layer was thin or disappeared in the articular eminence. Condylar deforming and osteophyte-like tissue (asterisk) were observed (a). In the articular eminence, type II collagen disappeared in the surface of the eminence and the underlying bone was exposed (arrowhead). In the osteophyte-like projection in the condylar head, three stratums of cartilage were observed (arrowheads) (c; immunologic staining for type II collagen, original magnification, ×12.5).

cartilage in the articular eminence preceded that in the condyle.

Histologic scoring

The severity of synovitis in the H & E-stained section was evaluated on the basis of Gynther system. Most scores of the synovial lining cell layers, vascularity and presence of inflammatory cells in each group increased gradually with time, but were slightly reduced at 8 weeks (Table 1). There were significant differences between the control group and 4-week group. Most scores of the quantitative changes in the articular cartilages and the reduction of type II collagen also increased with time in both groups (Table 2). Type II collagen in the articular eminence cartilage was reduced earlier, and the reduction was severer than in the condylar head, whereas the collagen

Table 1 Histologic grading with Gynther system in the arthritis induced by mechanical loading (mean ± SD)

	Control	*1 week*	*2 weeks*	*4 weeks*	*8 weeks*
Synovial lining cell layers	0.4 ± 0.5	0.7 ± 0.6	0.9 ± 0.4	1.4 ± 0.5*	1.0 ± 0.7
Vascularity	0.2 ± 0.5	0.7 ± 0.6	1.3 ± 0.8	1.7 ± 0.8*	1.3 ± 0.6
Inflammatory cells	0.4 ± 0.9	1.3 ± 1.2	1.0 ± 1.1	2.1 ± 1.5*	1.0 ± 0.9

*The mean data of the synovitis grading values between control and 1, 2, 4 and 8 weeks were analyzed using Whelch's *t*-test (significant difference, $P < 0.05$).

Table 2 Histologic grading of cartilage degradation in the arthritis induced by mechanical loading (mean ± SD)

	Control	*1 week*	*2 weeks*	*4 weeks*	*8 weeks*
Articular cartilage grading					
Condyle	0	0.3 ± 0.6	1.3 ± 1.1*	1.8 ± 0.8*	1.6 ± 1.4
Eminence	0	0.7 ± 0.6	1.1 ± 1.1*	1.9 ± 0.7*	2.3 ± 0.6*
Reducing of type II collagen					
Condyle	0	0	0.9 ± 0.7*	1.2 ± 1.0	1.0 ± 0.6
Eminence	0	1.0 ± 0.0	1.1 ± 0.7*	1.7 ± 0.8*	1.7 ± 0.6*

*The mean data of the quantitative grading values between control group and 1, 2, 4 and 8 weeks groups were analyzed using Whelch's *t*-test (significant difference, $P < 0.05$).

in the condyle surface was reduced with time but slightly recovered in the 8-week group.

Discussion

In the present study, we created minor disorders of occlusion and mastication by exclusively applying a mild, continuous mechanical load (100 ***g***) to the glenoid fossa, and observed the morbid changes in the TMJ in rabbits. As a result, mild inflammation was observed from 1 week after the start of loading and peaked in the 4-week group. The Gynther system was used to assess the degree of synovitis, and most of the grading points in the 4-week group and the 8-week group were similar to those of specimens obtained arthroscopically from the patients with TMD (4, 17). In the 1-week group, chondral degeneration occurred in the articular eminence, and type II collagen was reduced before that in the condyle. In the 4-week group, there was little type II collagen, and pannus formation and condylar deformity were observed in some joints. In the 8-week group, many joints showed no cartilage in the articular eminence, and half of the joints showed thin condylar cartilage and severe condylar deformities. Synovitis reduced in comparison with the 4-week group, but chondral degeneration and deformity were markedly observed in the articular eminence and condyle. The chondral degeneration and destruction – as in osteoarthritis – were assumingly induced by continuous or intermittent mechanical loading. With overloading, the pathogenic mechanism is that type II collagen, which are considered to be among the main constituents of the cartilage, degenerate on the surface of the articular cartilage (22). Matrix metalloproteinases (MMPs) cleave

this type II collagen molecule at one of four and three of four of its initial length. Large amounts of MMPs are expressed in the osteoarthritic cartilage (23), and in particular, MMP-13 plays a key role in destroying the cartilage matrix (24). Interleukin (IL)-1 and tumor necrosis factor (TNF)-α are also involved in the development of osteoarthritis (25), and TNF-α receptors are expressed in sites where the cartilage has been destroyed (26). IL-1 and TNF-α might play a major role in the production of MMPs. IL-1, IL-6, IL-8, TNF-α (27, 28), MMP-1, and MMP-13 are expressed in the synovial fluid of patients with TMD (29), and are considered to be involved in the degeneration and destruction of the cartilage.

In some joints with condylar deformities, osteophyte-like projections were observed in the 8-week group. Although the mechanism of osteophyte development has not yet been demonstrated, it is considered to be a physiologic reaction for dispersing and adapting to the mechanical load in the TMJ. When this stress dispersion mechanism breaks down, the articular cartilage degenerates and loses its viscoelasticity, and the mechanical loading is then transmitted almost directly to the subchondral bone, and subsequently may induce reactive ossification, or the chondrocytes regenerate after the osteochondral tissues have been absorbed, and osteophytes then form through the process of enchondral ossification. Interestingly, fibrous adhesions of the disk and articular eminence, accompanied by slight inflammation, were exclusively observed in 17% of the joints contralateral to the mechanically loaded joints (one joint in the 4-week group and one joint in the 8-week group) (findings are not shown), but were not observed in the control group. This fibrous adhesion has also been observed in other models that employed elastic force (30, 31). These results may suggest that mechanical loading is one of the key inducers of TMD.

In conclusion, we applied a mild mechanical load to the TMJ of rabbits for 8 weeks and periodically observed the morbid changes in the articular tissues. This observation showed that mild synovial inflammation began within 1–2 weeks, and became significant at 4 weeks after the start of loading. Organic changes in the articular cartilage of the articular eminence occurred prior to that in the condyle. We could not conclude that these changes reflect the morbid changes in the synovial tissues of the patients with TMD, but our study revealed that mild, continuous mechanical loading causes synovitis of the TMJ, and induces organic changes without destroying condylar and articular eminence. The observation period was 2 months, but a longer observation period and periodic biochemical analysis of the synovial fluid are required in the future.

References

1. Murakami K, Segami N, Fujimura K, Iizuka T. Correlation between pain and synovitis in patients with internal derangement of the temporomandibular joint. *J Oral Maxillofac Surg* 1991; **49**: 1159–61.
2. Holmlund AB, Gynther GW, Reinholt FP. Disk derangement and inflammatory changes in the posterior disk attachment of the temporomandibular joint. A histologic study. *Oral Surg Oral Med Oral Pathol* 1992; **73**: 9–12.
3. Gynther GW, Holmlund AB, Reinholt FP. Synovitis in internal derangement of the temporomandibular joint: correlation between arthroscopic and histologic findings. *J Oral Maxillofac Surg* 1994; **52**: 913–7.
4. Segami N, Suzuki T, Sato J, Miyamaru M, Nishimura M, Yoshimura H. Does joint effusion on T2 magnetic resonance images reflect synovitis? Part 3. Comparison of histologic findings of arthroscopically obtained synovium in internal derangements of the temporomandibular joint. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2003; **95**: 761–6.
5. Sato J, Segami N, Nishimura M, et al. Correlation between the arthroscopic diagnosis of synovitis and microvessel density in synovial tissues in patients with internal derangement of the temporomandibular joint. *J Craniomaxillofac Surg* 2003; **31**: 101–6.
6. Nitzan DW. Intra-articular pressure in the functioning human temporomandibular joint and its alteration by uniform elevation of the occlusal plane. *J Oral Maxillofac Surg* 1994; **52**: 671–9.
7. Werther JR, Hall HD, Gibbs SJ. Disk position before and after modified condylotomy in 80 symptomatic temporomandibular joints. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 1995; **79**: 668–79.
8. Hall HD, Navarro EZ, Gibbs SJ. One- and three-year prospective outcome study of modified condylotomy for treatment of reducing disc displacement. *J Oral Maxillofac Surg* 2000; **58**: 7–17.
9. Nitzan DW, Nitzan U, Dan P, Yedgar S. The role of hyaluronic acid in protecting surface-active phospholipids from lysis by exogenous phospholipase A(2). *Rheumatology (Oxford)* 2001; **40**: 336–40.
10. Nitzan DW. The process of lubrication impairment and its involvement in temporomandibular joint disc displacement: a theoretical concept. *J Oral Maxillofac Surg* 2001; **59**: 36–45.
11. Clark GT. A critical evaluation of orthopedic interocclusal appliance therapy: design, theory, and overall effectiveness. *J Am Dent Assoc* 1984; **108**: 359–64.
12. Bell WH, Yamaguchi Y, Poor MR. Treatment of temporomandibular joint dysfunction by intraoral vertical ramus osteotomy. *Int J Adult Orthodon Orthognath Surg* 1990; **5**: 9–27.
13. Hall HD, Navarro EZ, Gibbs SJ. Prospective study of modified condylotomy for treatment of nonreducing disk displacement. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2000; **89**: 147–58.
14. Dolwick MK, Sanders B. TMJ internal derangement and arthrosis, surgical atlas. In: Dolwick MK, Sanders B, eds. *Anatomy*. St Louis, Princeton, Toront, USA: The C. V. Mosby Company, 1985; 1–26.
15. Burakoff RP, Kaplan AS. Temporomandibular disorders: current concepts of epidemiology, classification, and treatment. *J Pain Symptom Manage* 1993; **8**: 165–72.
16. Gynther GW, Dijkgraaf LC, Reinholt FP, Holmlund AB, Liem RS, de Bont LG. Synovial inflammation in arthroscopically obtained biopsy specimens from the temporomandibular joint: a review of the literature and a proposed histologic grading system. *J Oral Maxillofac Surg* 1998; **56**: 1281–86.
17. Suzuki T, Segami N, Sato J, Nojima T. Accuracy of histologic grading of synovial inflammation in temporo-

mandibular joints with internal derangement using Gynther's system. *J Oral Maxillofac Surg* 2001; **59**: 498–501.
18. Mankin HJ, Dorfman H, Lippiello L, Zarins A. Biochemical and metabolic abnormalities in articular cartilage from osteo-arthritic human hips: II. Correlation of morphology with biochemical and metabolic data. *J Bone Joint Surg Am* 1971; **53**: 523–37.
19. Wilhelmi G. Effect of C 21 524-Su (pirprofen) on spontaneous osteoarthrosis in the mouse. *Pharmacology* 1978; **16**: 268–72.
20. Tallents RH, Macher DJ, Rivoli P, Puzas JE, Scapino RP, Katzberg RW. Animal model for disk displacement. *J Craniomandib Disord* 1990; **4**: 233–40.
21. Mills DK, Daniel JC, Herzog S, Scapino RP. An animal model for studying mechanisms in human temporomandibular joint disc derangement. *J Oral Maxillofac Surg* 1994; **52**: 1279–92.
22. Hollander AP, Pidoux I, Reiner A, Rorabeck C, Bourne R, Poole AR. Damage to type II collagen in aging and osteoarthritis starts at the articular surface, originates around chondrocytes, and extends into the cartilage with progressive degeneration. *J Clin Invest* 1995; **96**: 2859–69.
23. Mitchell PG, Magna HA, Reeves LM, et al. Cloning, expression, and type II collagenolytic activity of matrix metalloproteinase-13 from human osteoarthritic cartilage. *J Clin Invest* 1996; **97**: 761–68.
24. Billinghurst RC, Dahlberg L, Ionescu M, et al. Enhanced cleavage of type II collagen by collagenases in osteoarthritic articular cartilage. *J Clin Invest* 1997; **99**: 1534–45.
25. Goldring MB. Osteoarthritis and cartilage: the role of cytokines. *Curr Rheumatol Rep* 2000; **2**: 459–65.
26. Webb GR, Westacott CI, Elson CJ. Chondrocyte tumor necrosis factor receptors and focal loss of cartilage in osteoarthritis. *Osteoarthritis Cartilage* 1997; **5**: 427–37.
27. Takahashi T, Kondoh T, Fukuda M, Yamazaki Y, Toyosaki T, Suzuki R. Proinflammatory cytokines detectable in synovial fluids from patients with temporomandibular disorders. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 1998; **85**: 135–41.
28. Segami N, Miyamaru M, Nishimura M, Suzuki T, Kaneyama K, Murakami K. Does joint effusion on T2 magnetic resonance images reflect synovitis? Part 2. Comparison of concentration levels of proinflammatory cytokines and total protein in synovial fluid of the temporomandibular joint with internal derangements and osteoarthrosis. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2002; **94**: 515–21.
29. Srinivas R, Sorsa T, Tjaderhane L, et al. Matrix metalloproteinases in mild and severe temporomandibular joint internal derangement synovial fluid. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2001; **91**: 517–25.
30. Imai H, Sakamoto I, Yoda T, Yamashita Y. A model for internal derangement and osteoarthritis of the temporomandibular joint with experimental traction of the mandibular ramus in rabbit. *Oral Dis* 2001; **7**: 185–91.
31. Yoda T, Sakamoto I, Imai H, Yamashita Y, Enomoto S. Fibrous adhesions in the joint compartment of the temporomandibular joint associated with experimental opposite drawing mandibular ramus in rabbit. *Cranio* 2001; **19**: 169–73.