Effects of Age on the Ability of the Rat Temporomandibular Joint to Respond to Changing Functional Demands
M. Bouvier

DOI: 10.1177/00220345880670091101

The online version of this article can be found at:
http://jdr.sagepub.com/content/67/9/1206

Published by:
SAGE
http://www.sagepublications.com
On behalf of:
International and American Associations for Dental Research

Additional services and information for Journal of Dental Research can be found at:

Email Alerts: http://jdr.sagepub.com/cgi/alerts
Subscriptions: http://jdr.sagepub.com/subscriptions
Reprints: http://www.sagepub.com/journalsReprints.nav
Permissions: http://www.sagepub.com/journalsPermissions.nav
Citations: http://jdr.sagepub.com/content/67/9/1206.refs.html

>> Version of Record - Sep 1, 1988

What is This?
Effects of Age on the Ability of the Rat Temporomandibular Joint to Respond to Changing Functional Demands

M. BOUVIER

Department of Anatomy, 1100 Florida Avenue, Louisiana State University, School of Dentistry, New Orleans, Louisiana 70119

This investigation examined the ability of the tissues of the temporomandibular joint (TMJ) to adapt to changing functional demands in young, growing rats compared with mature rats. Functional demands on the TMJ were varied by feeding diets with different physical consistencies. The first group was fed a soft diet for the experimental period. The second group was fed a hard diet, and the third group was initially fed the soft diet, then switched to the hard diet at the midpoint of the experimental period. Gross dimensions of the condyle, mandible, and maxilla were measured with calipers. Thickness of the articular, proliferative, transitional, and hypertrophic zones of the condylar cartilage, and the amount of bone in the subcondylar region and condylar neck were measured on histological sections. Gross dimensions of the condyle were significantly smaller in the soft-diet group compared with the hard- and soft/hard-diet groups in both growing and mature rats. The individual zones of the condylar cartilage were also significantly narrower in the soft-diet group in both growing and mature rats. However, the soft/hard-diet group of mature rats showed only a significant reduction in the thickness of the articular zone of the condylar cartilage compared with the hard-diet group. There were also narrower proliferative and transitional zones in the mature rats fed a soft/hard diet. In contrast, all of these zones showed full recovery in the young rats fed a soft/hard diet. The data presented here suggest that increasing age may diminish the capacity of the TMJ to adapt to altered function and consequently may play a significant role in the development of degenerative joint disease.


Introduction.

Experimental studies in mice, rats, rabbits, and non-human primates have shown that mechanical loads are vital for maintaining normal growth, morphology, and function of the secondary cartilage of the temporomandibular joint (TMJ). For example, lowered loads on the TMJ decrease condylar cartilage cell proliferation and matrix synthesis (Petrovic et al., 1975; Copray et al., 1985 a, b, c; Glineburg et al., 1982; Hinton and Carlson, 1986) and result in a small condyle with a thin cartilaginous articular covering (Watt and Williams, 1951; Simon, 1977; Beecher and Corrucini, 1981; Corrucini and Beecher, 1984; Bouvier and Hylander, 1982, 1984; Lydiatt and Davis, 1985; Hinton and Carlson, 1986; Bouvier and Zimny, 1987). Enzyme activity associated with bone mineralization within the hypertrophic layer of the condylar cartilage and also the amount of subcondylar trabecular bone drop when loads are lowered (Bouvier, 1987; Bouvier and Hylander, 1982, 1984; Hinton and Carlson, 1986). A return to normal loading results in recovery of cartilage thickness on the condylar articular surface as well as recovery of subcondylar trabecular bone volume, matrix synthesis, and enzymatic activity (Bouvier, 1987; Bouvier and Hylander, 1982, 1984; Glineburg et al., 1982). In vitro studies confirm that normal mechanical loading stimulates cell division, matrix synthesis, and enzyme activity in the tissues of the TMJ (Copray et al., 1985 a, b, c). In addition, epidemiological, anthropological, and autopsy studies suggest that the human TMJ also responds adaptively to normal mechanical loads, with hypertrophy and hyperplasia of the tissues of the condyle and articular eminence (Hinton, 1981; Zarb and Carlsson, 1979).

As the tissues of this joint age, however, they may lose some or all of their ability to adapt to changing functional loads, which then may play a role in the development of degenerative joint disease (Lubsen et al., 1987; Weiss et al., 1986; Richards et al., 1984; Zarb and Carlson, 1979). TMJ tissues in older individuals may be particularly susceptible to mechanical trauma because of this diminished capacity to respond to increases in mechanical loads. The purpose of this

Fig. 1—Gross mandibular and maxillary measurements. CL, condyle length; CW, condyle width; ML, mandible length; MAW, maxillary arch width.

Fig. 2—Histological measurements on condylar cartilage. a, articular zone; d, disc; h, hypertrophic zone; p, proliferative zone; t, transitional zone. The arrowhead indicates the inferior joint space.

Received for publication November 13, 1987
Accepted for publication April 14, 1988
Materials and methods.

A total of 77 Sprague-Dawley rats was used in this experiment, including 20 young male and 26 young female rats and 31 adult male rats. The young rats were 21 days old at the beginning of the experiment. The adults weighed an average of 467 ± 48 g each and were approximately four months old at the start of the experiment. Upon arrival, the rats were weighed, housed in hanging cages, and divided into three groups.

The first group of rats was fed ad libitum on ground rat chow moistened with water for four weeks (16 young, growing rats) or 12 weeks (10 mature, adult rats). This group was designated the soft-diet group. Previous investigations of condylar adaptations to diet in young, growing rats indicated that the maximal levels are reached by 2–4 weeks, and the percentage of change in the dimensions is constant thereafter (Hinton and Carlson, 1986; Bouvier and Hylander, 1984; Bouvier, 1981). Comparable data are not available for older rats; therefore, a longer period was chosen to ensure that sufficient time had elapsed for detection of changes, particularly in the recovery (soft/hard-diet) group. The second group (16 and 12, respectively) was fed ad libitum on hard, pelleted rat chow, also for 4 or 12 weeks. This group was designated the hard-diet group. The third group was initially fed ad libitum on moist, ground chow for the first two weeks (14 growing rats) or six weeks (nine mature rats) of the experiment, and then for the remaining two weeks (growing rats) or six weeks (mature rats) of the experiment, on hard pelleted chow. This group was designated the soft/hard-diet group. The soft diet was presented in glass dishes which were placed inside the cages. These dishes were emptied, cleaned, and refilled every morning. Fresh water was always available for all rats.

The rats were weighed at the mid-point of the experiment (i.e., after two or six weeks), again at the end of the experiment, and then killed by ether overdose. Both dentaries along with the head were immediately removed. The head and right dentary were placed in 10% neutral buffered formalin. The left dentary was fixed in 2% glutaraldehyde and used for either scanning or transmission electron microscopy (Bouvier and Zimny, 1987). After 72 hr, the right dentary was removed from the fixative, washed thoroughly, measured, and decalcified in buffered formic acid for histological analysis. The head was stored in fresh 10% neutral buffered formalin for gross morphological analysis.

The following gross measurements were made on the right dentary before decalcification and on the head by use of sharp pointed calipers accurate to 0.01 mm (Fig. 1): (1) anteroposterior length from the most posterior to the most anterior point on the condylar articular surface; (2) maximum mediolateral width of the condylar articular surface; (3) mandibular length from the most posterior point on the condyle articular surface to the most anterior point on the incisor alveolus; and (4) maxillary arch width between the most lingual surfaces of the left and right upper second molars.

After several weeks to permit adequate decalcification, the posterior portion of the right dentary, including the condyle, was washed thoroughly, dehydrated in a graded series of alcohols and toluene, embedded in paraffin, and sectioned at 10 μm in the coronal direction. Slides were stained with hematoxylin and eosin.

The following measurements were made on every tenth section from the central portion of the condyle at the point of maximum cartilage thickness at 100 x by use of a calibrated eyepiece micrometer (Fig. 2): (1) thickness of the articular zone; (2) thickness of the proliferative zone; (3) thickness of the transitional zone; and (4) thickness of the hypertrophic zone. Total cartilage thickness was determined by summing the thickness of each individual zone, including the fibrous articular zone. A total of five sections spaced 100 μm apart was examined for each specimen. Measurements from the five sections were then averaged.

In addition, the following areas on the same five sections for each specimen were measured at 40 x and averaged by the Olympus Cuc-2 image analysis system (Fig. 3): (1) condylar cartilage area was measured by means of tracing over the entire condylar surface from medial to lateral poles and continuing along the junction between the hypertrophic zone and the mineralizing zone; (2) total subcondylar area was measured by means of tracing over the entire junction between the hypertrophic and mineralizing zones from medial to lateral poles and connecting these end-points with a straight line; and (3) total condylar neck area was measured by means of connecting the medial and lateral poles of the condyle with a straight line and then continuing to trace over the surface of the condylar neck present within the 40 x microscopic field.

Areas occupied...
by bone in the subcondylar and condylar neck regions were
determined by computer thresholding of stained areas (bone)
versus unstained areas (marrow spaces), by means of the
Olympus Cue-2 system. Total condylar head area was
determined by addition of condylar cartilage area to total subcon-
dylar area. Condylar cartilage area was then expressed as a
percentage of total condylar head area. Subcondylar bone area
was expressed as a percentage of total subcondylar area. Con-
dylar neck bone area was expressed as a percentage of total
condylar neck area.

Descriptive statistics were computed for each measurement
within the dietary, age, and sex groups. It was possible to
combine measurements from the growing male and female rats
from the same diet group for histological parameters, since it
was determined that they did not differ significantly (Student’s
t test, p > 0.05). These data along with data from the mature
rats were then subjected to one-way analyses of variance with
post hoc comparisons between means for significant differences
(p < 0.05). There were significant differences between
growing male and female rats for some gross measurements
(p < 0.05); therefore, these, along with data from the adults,
were analyzed separately by ranked one-way analyses of vari-
ance with post hoc comparisons of means for significant differ-
ces (p < 0.05).

Results.
No significant differences in body weight were found among
the different dietary groups of growing or mature rats (Table
1). The only significant differences in gross measurements were
for condylar dimensions. Condylar dimensions were greatest
in the hard-diet group, intermediate in the soft/hard-diet group,
and least in the soft-diet group (Figs. 4, 5). Differences be-
tween hard- and soft/hard-diet group means were not signifi-
cant except for condyle width, which showed a small but
significant difference.

Among the histological measurements, all were greatest in
the hard-diet groups and least in the soft-diet groups for both
weanlings and adults (Figs. 6, 7; Table 2, p < 0.05). In most
cases, histological measurements were intermediate in the soft/
hard-diet group. However, the differences between hard- and
soft/hard-diet group were not significant except for the thick-
ness of the articular zone in adult rats.

Table 3 shows percentages of cartilage and bone found in
the condylar head and neck. Mature rats showed a relative
decrease in the amount of cartilage and a concomitant increase
in bone compared with growing rats. Absolute values for these
parameters showed a pattern identical with that found for gross
condylar dimensions, with areas being greatest in the hard-diet
group, lowest in the soft-diet group, and intermediate in the
soft/hard-diet group. These findings recapitulate the existence
of gross-dimensional changes as a result of diet in both in-
mature and mature rats, and therefore, are not reported here.
Table 3 demonstrates that there was a relative decrease in the
percentage of bone found in the subcondylar region in the soft-
diet group in both growing and mature rats. In addition, young
soft/hard-diet rats had a significantly greater percentage of car-
tilage in the condylar head compared with soft-diet rats.

Discussion.
Results for growing and mature rats were generally similar.
Condyle measurements were the only gross dimensions to show
significant differences among the dietary groups in both grow-
ning and mature rats (Table 1). These findings indicate that there
is a local effect on the TMJ but not a general effect on growth

---

**TABLE 1**

MEANS AND STANDARD DEVIATIONS FOR LINEAR MEASUREMENTS (mm) AND BODY WEIGHT (g) FOR GROWING AND MATURE RATS ON DIFFERENT DIETS

<table>
<thead>
<tr>
<th>Group</th>
<th>N</th>
<th>Condyle Length</th>
<th>Condyle Width</th>
<th>Mandible Length</th>
<th>Mandible Arch Width</th>
<th>Body Weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>Growing Rats-Females</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hard Diet</td>
<td>9</td>
<td>3.6 (0.1)</td>
<td>1.8 (0.1)</td>
<td>24.3 (0.7)</td>
<td>6.0 (0.2)</td>
<td>184.2 (11.0)</td>
</tr>
<tr>
<td>Soft Diet</td>
<td>9</td>
<td>3.2 (0.2)</td>
<td>1.5 (0.1)</td>
<td>23.9 (0.9)</td>
<td>6.0 (0.2)</td>
<td>184.2 (11.0)</td>
</tr>
<tr>
<td>Soft/Hard Diet</td>
<td>8</td>
<td>3.5 (0.2)</td>
<td>1.8 (0.1)</td>
<td>24.2 (1.1)</td>
<td>6.0 (0.1)</td>
<td>181.5 (15.4)</td>
</tr>
<tr>
<td>*</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td></td>
<td>7.57</td>
<td>36.86</td>
<td>0.43</td>
<td>0.09</td>
<td>0.13</td>
</tr>
<tr>
<td>F²</td>
<td></td>
<td>0.40</td>
<td>0.76</td>
<td>0.04</td>
<td>0.01</td>
<td>0.01</td>
</tr>
<tr>
<td>Growing Rats-Males</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hard Diet</td>
<td>7</td>
<td>4.2 (0.3)</td>
<td>2.1 (0.1)</td>
<td>25.5 (0.8)</td>
<td>6.3 (0.1)</td>
<td>315.0 (28.7)</td>
</tr>
<tr>
<td>Soft Diet</td>
<td>7</td>
<td>3.7 (0.2)</td>
<td>1.6 (0.1)</td>
<td>25.2 (0.3)</td>
<td>6.3 (0.1)</td>
<td>294.3 (12.9)</td>
</tr>
<tr>
<td>Soft/Hard Diet</td>
<td>6</td>
<td>3.9 (0.2)</td>
<td>1.9 (0.1)</td>
<td>25.3 (0.9)</td>
<td>6.3 (0.2)</td>
<td>293.3 (18.9)</td>
</tr>
<tr>
<td>*</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td></td>
<td>3.59</td>
<td>21.20</td>
<td>0.22</td>
<td>0.77</td>
<td>1.17</td>
</tr>
<tr>
<td>F²</td>
<td></td>
<td>0.44</td>
<td>0.82</td>
<td>0.05</td>
<td>0.08</td>
<td>0.21</td>
</tr>
<tr>
<td>Mature Rats-Males</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hard Diet</td>
<td>12</td>
<td>4.3 (0.3)</td>
<td>1.9 (0.1)**</td>
<td>32.3 (0.7)</td>
<td>7.4 (0.2)</td>
<td>642.9 (36.8)</td>
</tr>
<tr>
<td>Soft Diet</td>
<td>10</td>
<td>3.6 (0.4)</td>
<td>1.5 (0.1)</td>
<td>31.7 (0.5)</td>
<td>7.3 (0.2)</td>
<td>636.8 (44.6)</td>
</tr>
<tr>
<td>Soft/Hard Diet</td>
<td>9</td>
<td>4.2 (0.2)</td>
<td>1.8 (0.1)**</td>
<td>31.9 (0.5)</td>
<td>7.3 (0.4)</td>
<td>647.7 (40.2)</td>
</tr>
<tr>
<td>*</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td></td>
<td>16.29</td>
<td>45.29</td>
<td>3.10</td>
<td>0.30</td>
<td>0.17</td>
</tr>
<tr>
<td>F²</td>
<td></td>
<td>0.54</td>
<td>0.76</td>
<td>0.18</td>
<td>0.02</td>
<td>0.01</td>
</tr>
</tbody>
</table>

*Means separated by an asterisk are significantly different (p<0.05).
**Means are significantly different (p<0.05).
Fig. 4—Histology of the condyle in young, growing rats. A, hard diet; B, soft diet; C, soft/hard diet.

Fig. 5—Histology of the condyle in older, mature rats. A, hard diet; B, soft diet; C, soft/hard diet.
Fig. 6—Histology of the superior region of the condyle in young, growing rats. A, hard diet; B, soft diet; C, soft/hard diet.

Fig. 7—Histology of the superior region of the condyle in older, mature rats. A, hard diet; B, soft diet; C, soft/hard diet.
of the mandible and maxilla, which corroborates earlier findings for growing male Long Evans rats (Bouvier and Hylander, 1984; Bouvier, 1981). Among mature rats, there was a small but statistically significant deficit in condylar width in the soft/hard-diet group compared with the hard-diet group.

Histological data indicated that all zones of the condylar cartilage were affected by changes in dietary consistency in both growing and mature rats (Table 2). Among growing rats, all zones showed recovery to nearly normal thickness in the soft/hard-diet group. However, among mature rats, the articular zone thickness was not completely recovered in the soft/hard-diet group compared with the hard-diet group. In addition, means for both proliferative and transitional zone thickness in the soft/hard-diet group were not significantly different from those in either the hard- or soft-diet groups. This suggests that in the mature soft/hard-diet group, there may have been a slight net depression in proliferation and/or maturational activity as a result of the initial period on the soft diet which was not completely compensated for during the period on the hard diet.

Reduction in subcondylar trabecular bone area in the soft-diet group compared with both hard- and soft/hard-diet groups indicated that significant porosity developed as a result of reduced function associated with the soft diet. However, in both growing and mature rats, there was complete recovery of subcondylar trabecular bone area in the soft/hard-diet groups. Thus, trabecular bone formation was apparently not affected by age in this study.

Differences in the responses of the cells of the condylar cartilage to changes in dietary consistency in the mature rats compared with the growing rats may have important consequences in TMJ adaptation to the functional environment throughout adult life. The articular zone of the condylar cartilage in mature animals demonstrated the least ability to re-

### Table 2

<table>
<thead>
<tr>
<th>Group</th>
<th>N</th>
<th>Total Cartilage Thickness</th>
<th>Articular Zone</th>
<th>Proliferative Zone</th>
<th>Transitional Zone</th>
<th>Hypertrophic Zone</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Growing Rats</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hard Diet</td>
<td>16</td>
<td>378 (60)</td>
<td>56 (11)</td>
<td>76 (19)</td>
<td>80 (24)</td>
<td>167 (28)</td>
</tr>
<tr>
<td>Soft Diet</td>
<td>16</td>
<td>251 (50)</td>
<td>39 (7)</td>
<td>53 (9)</td>
<td>54 (16)</td>
<td>114 (29)</td>
</tr>
<tr>
<td>Soft/Hard Diet</td>
<td>14</td>
<td>377 (40)</td>
<td>55 (7)</td>
<td>72 (10)</td>
<td>83 (13)</td>
<td>167 (29)</td>
</tr>
<tr>
<td><strong>F</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.59</td>
<td>0.48</td>
<td>0.38</td>
<td>0.35</td>
<td>0.45</td>
</tr>
<tr>
<td><strong>Mature Rats</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hard Diet</td>
<td>12</td>
<td>317 (55)</td>
<td>65 (14)**</td>
<td>70 (10)</td>
<td>49 (11)</td>
<td>133 (25)</td>
</tr>
<tr>
<td>Soft Diet</td>
<td>10</td>
<td>203 (16)</td>
<td>37 (11)</td>
<td>50 (9)</td>
<td>32 (9)</td>
<td>85 (16)</td>
</tr>
<tr>
<td>Soft/Hard Diet</td>
<td>9</td>
<td>282 (28)</td>
<td>49 (12)**</td>
<td>60 (13)</td>
<td>40 (14)</td>
<td>134 (18)</td>
</tr>
<tr>
<td><strong>F</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.64</td>
<td>0.50</td>
<td>0.28</td>
<td>0.30</td>
<td>0.58</td>
</tr>
</tbody>
</table>

*Means separated by an asterisk are significantly different (p<0.05).

**Means are significantly different (p<0.05).**

### Table 3

<table>
<thead>
<tr>
<th>Group</th>
<th>N</th>
<th>% Cartilage in Total Condyle Area</th>
<th>% Bone in Subcondyle Area</th>
<th>% Bone in Total Condyle Neck Area</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Growing Rats</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hard Diet</td>
<td>16</td>
<td>50.1 (6.3)</td>
<td>59.4 (4.4)</td>
<td>66.7 (6.6)</td>
</tr>
<tr>
<td>Soft Diet</td>
<td>16</td>
<td>45.5 (8.1)</td>
<td>53.8 (4.5)</td>
<td>65.7 (6.5)</td>
</tr>
<tr>
<td>Soft/Hard</td>
<td>14</td>
<td>53.6 (5.1)</td>
<td>57.8 (4.3)</td>
<td>64.5 (4.2)</td>
</tr>
<tr>
<td><strong>F</strong></td>
<td></td>
<td>0.20</td>
<td>0.21</td>
<td>0.03</td>
</tr>
<tr>
<td><strong>Mature Rats</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hard Diet</td>
<td>12</td>
<td>36.4 (7.5)</td>
<td>78.3 (4.6)</td>
<td>87.5 (3.2)</td>
</tr>
<tr>
<td>Soft Diet</td>
<td>10</td>
<td>36.5 (5.0)</td>
<td>69.9 (7.6)</td>
<td>89.2 (3.5)</td>
</tr>
<tr>
<td>Soft/Hard</td>
<td>9</td>
<td>36.3 (4.7)</td>
<td>77.9 (6.1)</td>
<td>87.8 (3.9)</td>
</tr>
<tr>
<td><strong>F</strong></td>
<td></td>
<td>0.00</td>
<td>0.62</td>
<td>0.70</td>
</tr>
<tr>
<td><strong>r²</strong></td>
<td></td>
<td>0.00</td>
<td>0.31</td>
<td>0.15</td>
</tr>
</tbody>
</table>

*Means separated by an asterisk are significantly different (p<0.05).
cover from functional alterations in joint loading and therefore may be the most vulnerable to early degenerative changes. In addition, the data presented suggest that condylar cartilage cells may have slightly diminished capacity for proliferation and maturation in adults in response to an altered functional environment. In contrast, recovery of trabecular bone density was not affected by age in this study.

REFERENCES


