

Influence of malocclusion on the development of masticatory function and mandibular growth

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ABSTRACT

Objective: To verify the hypothesis that appropriate acquisition of masticatory function and normal growth of the mandible are modified by malocclusion.

Materials and Methods: Eighteen Jcl:ICR mice were divided into two groups. In one group we shifted the mandible laterally using an occlusal guidance appliance, creating a posterior crossbite at 5 weeks of age. The other group served as control. After 10 weeks, three-dimensional jaw movements and muscle activities were recorded simultaneously during mastication. Microcomputed tomography scans were obtained in vivo to evaluate morphometric changes in the mandible.

Results: (1) The jaw movement pattern in the sagittal plane showed significantly less anteroposterior excursion in the malocclusion group during the late-closing phase (power phase). (2) Electromyography showed significantly less masseter activity in the malocclusion group. (3) The condylar width and mandibular bone mineral density (BMD) were significantly reduced in the malocclusion mice compared to the normal mice.

Conclusions: These findings suggest that optimization of the chewing pattern and acquisition of appropriate masticatory function is impeded by malocclusion. Altered mechanical loading to the mandible may cause significant reduction of condylar width and mandibular BMD. (*Angle Orthod.* 2013;83:749–757.)

KEY WORDS: Mastication; Jaw movement; EMG; Masseter; Micro-CT; BMD

INTRODUCTION

It has long been believed that malocclusion may affect not only morphology but also masticatory

function. Several studies have speculated that malocclusion during early childhood could greatly affect the subsequent growth and development of the jaws and face not only morphologically^{1,2} but also functionally.^{3,4} Basic masticatory patterns begin to form soon after weaning and are established during early childhood.^{5,6} Malocclusion during this period may have a significant impact on development of normal masticatory function, resulting in an abnormal masticatory pattern.

The basic rhythm of masticatory movement and rhythmic muscle activities synchronized with jaw movements is thought to be directed by the masticatory central pattern generator (mCPG) located in the brainstem.⁷ In addition to this basic movement and rhythm pattern, jaw movements during mastication are controlled by sensory input from teeth, temporomandibular joints (TMJ), and masticatory muscles. Once an abnormal masticatory pattern is established, it generally remains and tends to affect jaw growth.^{3,4}

Several studies have suggested an association of malocclusion with morphology^{1,2} or function^{3,4} of the jaws and face, but only a few have attempted to study these aspects in combination because of the difficulty of investigating growth and development longitudinally.

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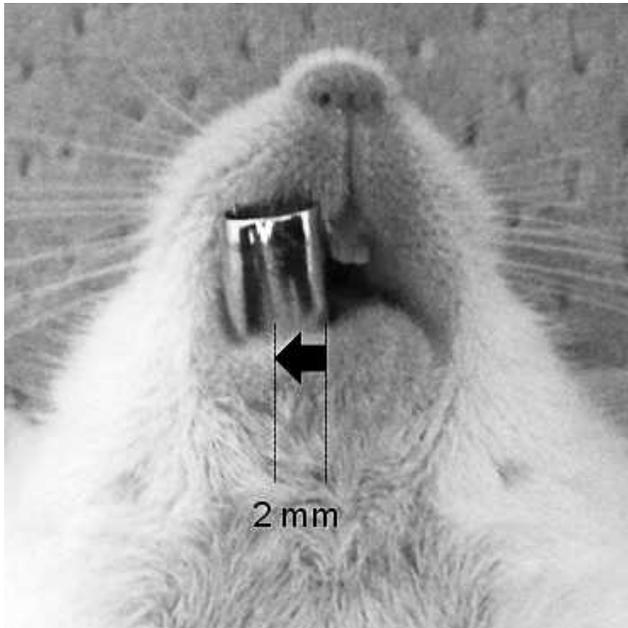


Figure 1. Mouse model for malocclusion. Note the excursion of the lower incisors 2 mm to the right.

In this study, we aimed to clarify the influence of malocclusion on mandibular growth and development of stomatognathic function. For this purpose, we took in vivo microcomputed tomography (micro-CT) scans continuously to observe the growth and development in mice in which malocclusion was induced in early childhood.

MATERIALS AND METHODS

Experimental Animals and Feeding

The Animal Welfare Committee of Nagasaki University approved this study based on the Animal Care Standards of this institution. Eighteen male Jcl:ICR mice (Clea, Tokyo, Japan) were randomly divided into normal ($n = 9$) and malocclusion ($n = 9$) groups. At

5 weeks of age, an occlusal guidance appliance was placed in the malocclusion group mice. The appliance was modified from those used in previous studies^{8,9} to induce a 2-mm lateral functional shift of the mandible to the right side (Figure 1). The appliances, constructed from band material (Rocky Mountain Morita, Tokyo, Japan), were attached to the lower incisor with composite resin (Clearfil Liner Bond II; Kuraray, Okayama, Japan). The same appliance was used continuously from 5 until 15 weeks of age. Then, micro-CT (R-mCT; Rigaku, Tokyo, Japan) scans were obtained every 2 weeks with the animals under anesthesia. All of the mice were fed a powdered diet of CE-2 (Clea) until age 15 weeks because the malocclusion group animals could not grind the CE-2 pellets.

Functional Recordings

At 15 weeks of age, recordings of jaw movements and masticatory muscle activities were obtained. The methodology for recording jaw movements and masticatory muscle activities from freely moving animals has been published previously.¹⁰⁻¹² For electromyography (EMG) recordings, bipolar electrodes consisting of Teflon-coated stainless steel wires with 2-mm exposed tips and 1-mm inter-polar distance were implanted in bilateral masseter muscles and unilateral digastric muscle. EMG signals were amplified with AC amplifiers and stored in a computer memory through a 12-bit A/D converter. Sampling rate of recordings was fixed at 2000 Hz.

Data Analysis

The masticatory sequence comprised food intake and chewing periods. Further analyses addressed the chewing period of rhythmical cycles. The parameters were calculated from 10 chewing cycles for each animal. Previously, we found that jaw closing from the occlusal view could be divided into two phases: early

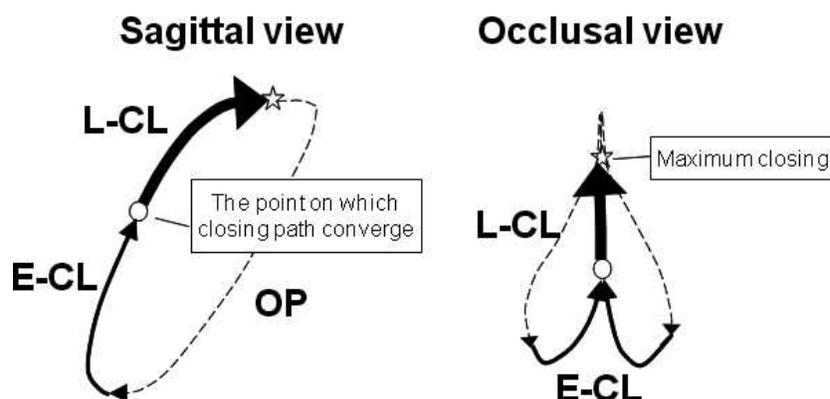


Figure 2. Classification of three-dimensional jaw movement. OP indicates opening phase; E-CL, early-closing phase; L-CL, late-closing phase.

Table 1. Definitions of Parameters for Analyzing Jaw Movements

Parameters	Definition
(1) Total cycle duration (TCL)	Time between one point of maximum jaw-opening and the next
(2) Opening phase duration (OP)	Time between maximum closing and the next maximum opening
(3) Closing phase duration (CL)	Time between maximum opening and the next maximum closing
(4) Early-closing phase duration (E-CL)	Time between the maximum opening and the point at which closing paths converge
(5) Late-closing phase duration (L-CL)	Time between the point at which closing paths converge and maximum closing
(6) Gape size (GAPE)	Opening/closing excursion between maximum opening and maximum closing
(7) Early-closing excursion (E-CL excursion)	Distance along the curvature corresponding to the early-closing phase
(8) Late-closing excursion (L-CL excursion)	Distance along the curvature corresponding to the late-closing phase
(9) Number of chewing cycles	Need to swallow standardized portions

closing and late closing (Figure 2). The early-closing (first) phase is composed of anterior and medial movements. It is defined as the section between the maximum opening and the point at which the closing paths converge. The late-closing phase is the section between the point at which the closing paths converge

and maximum closing. Parameters for analyzing jaw movements are shown in Table 1.

Jaw muscle activity was analyzed in terms of (1) duration and (2) average amplitude. The duration of EMG activity was the time in seconds for which the EMG amplitude was greater than two standard devia-

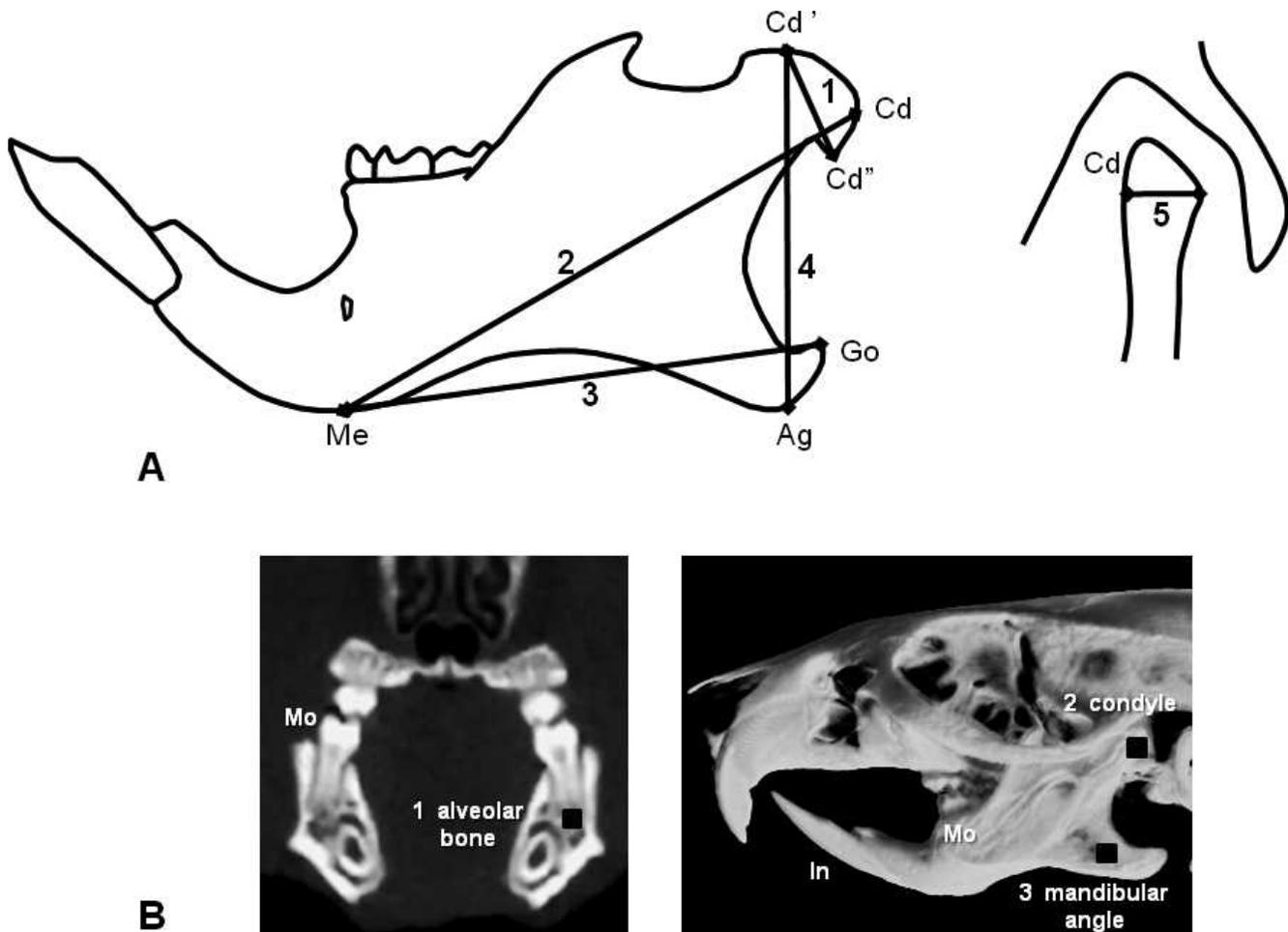


Figure 3. (A) The following five mandibular distances were measured. 1: condylar length, Cd'-Cd''; 2: mandibular body length, Cd-Me; 3: mandibular body length, Go-Me; 4: ramus height, Cd'-Ag; 5: condylar width, left-to-right thickness of the condyle. Cd indicates most posterior point of the condyle; Cd', highest point of the condyle; Cd'', lowest point of the condyle; Me, menton; Go, gonion; Ag, antegonion; In, incisor; Mo, molar. Right, sagittal view; left, frontal view. (B) BMDs were measured on the following three regions. 1: alveolar bone; 2: condyle; 3: mandibular angle. In indicates incisor; Mo, molar. Right, frontal view; left, sagittal view.

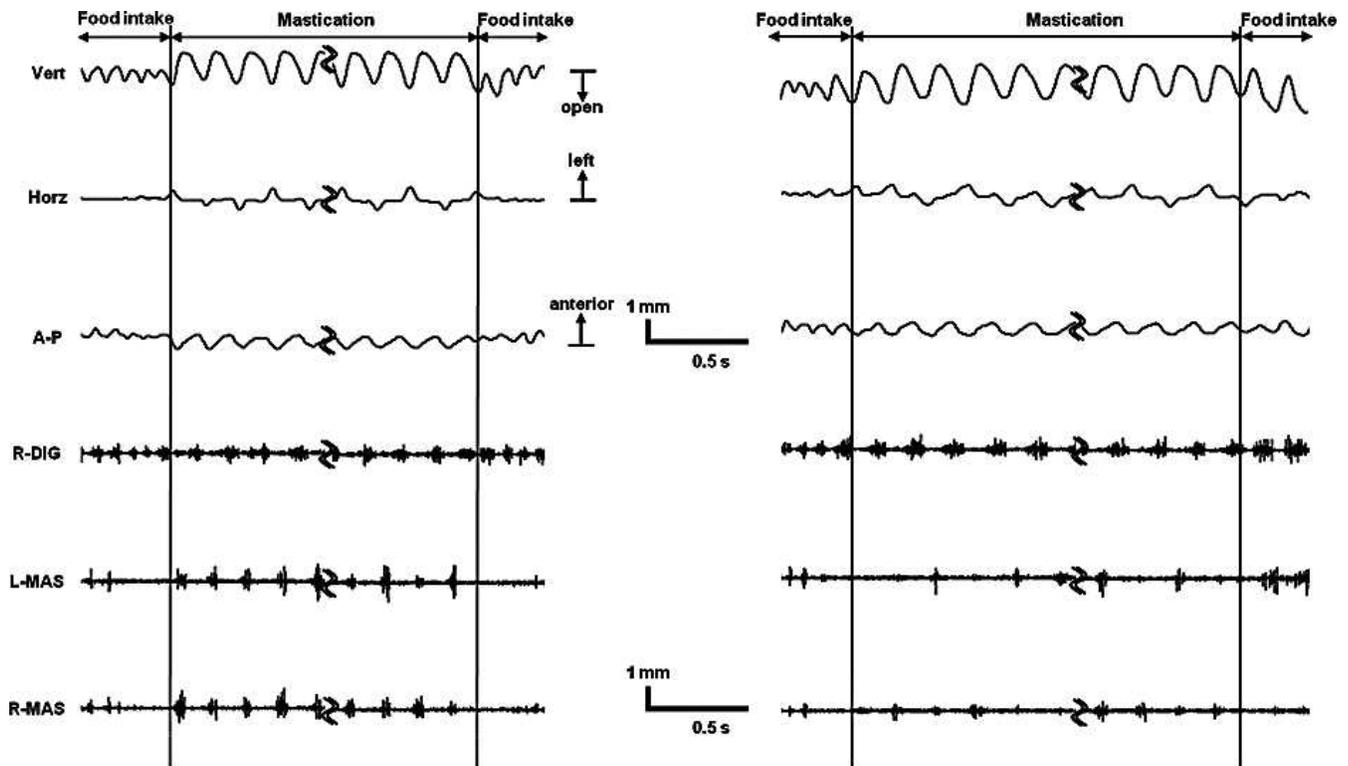


Figure 4. A typical masticatory sequence obtained from normal mice (right) and malocclusion mice (left) when chewing paste food. Upper three traces illustrate jaw movements in the vertical (Vert), horizontal (Horz), and antero-posterior (A-P) direction. Lower three traces show EMGs of the right digastric (R-DIG), left masseter (L-MAS), and right masseter (R-MAS) muscles. The sequence was divided into food intake and mastication periods. Horizontal and vertical calibrations of jaw movement are common for all tracings as with EMGs.

tions of the baseline. The average amplitude was calculated as the average of all electrical activity for the duration of the EMG burst. The Spike 2 software (Cambridge Electronic Design, Cambridge, UK) aided in the data analysis.

Morphometric Measurement

Under anesthesia, multiple scans of the mandible were obtained (90 kV, 100 mA, slice thickness 60 μ m) using micro-CT. Distances between morphometric landmarks were measured with image analysis software (i-view; J. Morita MFG, Kyoto, Japan). Five mandibular distances were measured (Figure 3). The mandible was reconstructed using TRI/3D-BON software (Ratoc, Tokyo, Japan) to evaluate the development of the mandibular area over the 15-week period. The same software was used to analyze the parameters of bone mineral density (BMD; Figure 3). The same investigator performed all measurements, and every measurement was repeated three times. The mean value was used as the final measurement.

Statistical Analysis

The Mann-Whitney *U*-test determined the significant difference between the normal and malocclusion

groups. $P < .05$ was considered significant. All values are displayed as means \pm standard error of the mean.

RESULTS

Jaw Movements

Movement amplitude. Jaw movement paths and jaw muscle activity during mastication were determined (Figure 4). The sequence was divided into food intake and chewing periods depending on the rhythm of the mandibular movement and the pattern of muscle activity.

The paths of three-dimensional jaw movement were reconstructed in two dimensions by projecting on the sagittal, frontal, and occlusal planes, respectively (Figure 5). In both groups, the closing movement consisted of arched, forward trajectory in the sagittal plane and medial movement of the jaw in the frontal and occlusal planes. The opening movements consisted of a nearly horizontal forward movement followed by backward depression in the sagittal plane and simultaneous lateral movement of the jaw in the frontal and occlusal planes. In the malocclusion group, the distance of the jaw movement was longer during the early-closing phase and shorter in the late-closing phase than those in the normal group (Table 2).

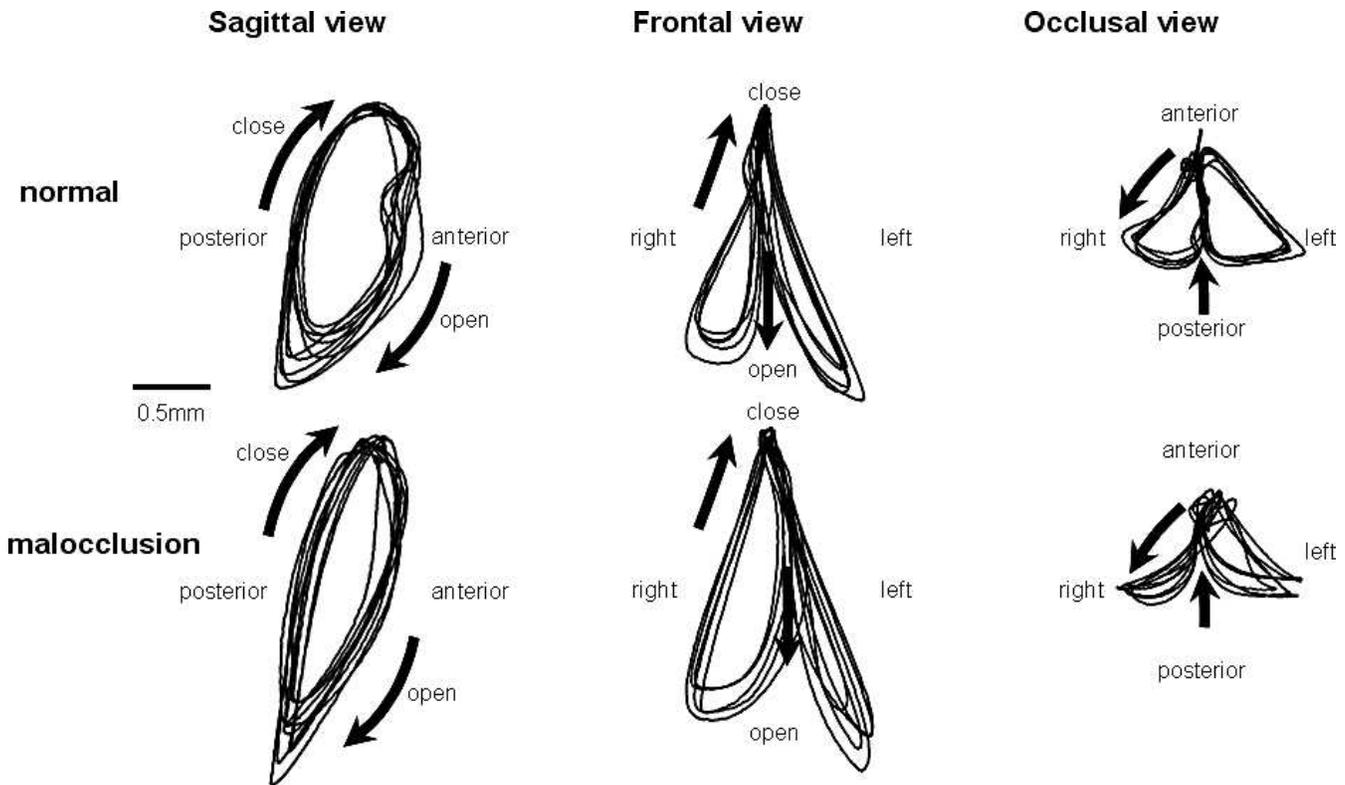


Figure 5. Jaw movement orbits in normal (upper) and malocclusion (lower) mice when chewing food. Tracings show 10 consecutive cycles superimposed. Left tracings: sagittal plane; center tracings: frontal plane; right tracings: occlusal plane. Horizontal and vertical calibrations are common for all orbits.

Cycle duration. The chewing cycle durations are shown in Table 3. The total cycle, opening phase, and early-closing phase were extended in the malocclusion group. When the phase durations were standardized as a percentage of the total cycle duration, the late-closing phase was shorter and the opening phase longer in the malocclusion group (Figure 6). The late-closing phase duration in relation to total cycle duration was $13.74\% \pm 3.03\%$ for the malocclusion group and $19.31\% \pm 3.58\%$ for the normal group ($P < .01$, Mann-Whitney *U*-test). There was no significant difference in the early-closing phase duration in relation to total cycle duration between the normal mice ($21.26\% \pm 2.45\%$) and malocclusion mice ($19.82\% \pm 3.89\%$). The number of chewing cycles was significantly increased in the malocclusion group (Table 2).

Muscle Activity

In a comparison of masseter and digastric muscle activity during mastication, the duration of masseter muscle bursts was 24.33% longer and the average amplitude was 26.68% lower in the malocclusion group (Table 4). There were no significant differences in jaw muscle activity between right and left masseter muscles in either group.

Morphology

The reconstructed three-dimensional micro-CT images are shown in Figure 7. In the ventral view, the mandible was shifted and rotated to the right, and the frontal view indicated that the molar relation was changed in the malocclusion group.

Table 2. Comparison of Jaw Movements Between Normal and Malocclusion Mice^a

	GAPE, μm	E-CL Excursion, μm	L-CL Excursion, μm	Number of Chewing Cycles
Normal (n = 9)	1705.27 \pm 189.09	1187.16 \pm 237.77	519.11 \pm 78.33	74.8 \pm 11.25
Malocclusion (n = 9)	1911.12 \pm 341.35	1511.47 \pm 303.33	393.72 \pm 68.92	123.45 \pm 26.23
Significance	NS	*	*	*

^a Values are presented as the standard error of the mean of each group. GAPE indicates gape size; E-CL excursion, early-closing excursion; L-CL excursion, late-closing excursion. Individual differences were tested using Mann-Whitney *U*-test.

* $P < .05$; NS indicates not significant.

Table 3. Comparison of Cycle Duration of Jaw Movements Between Normal and Malocclusion Mice^a

	TCL, ms	OP, ms	CL, ms	E-CL, ms	L-CL, ms
Normal (n = 9)	184.77 ± 23.03	109.56 ± 15.35	74.34 ± 9.34	38.32 ± 4.53	36.03 ± 9.89
Malocclusion (n = 9)	234.31 ± 83.14	152.46 ± 62.53	81.74 ± 23.25	47.97 ± 7.24	33.75 ± 16.08
Significance	*	**	NS	*	NS

^a Values are presented as the standard error of the mean of each group. TCL indicates total cycle duration; OP, opening phase duration; CL, closing phase duration; E-CL, early-closing phase duration; L-CL, late-closing phase duration. Individual differences were tested using Mann-Whitney *U*-test.

* $P < .05$; ** $P < .01$; NS indicates not significant.

Mandibular Distances

The mandibular distances in the two groups are shown in Table 5. There were no significant differences between the two groups regarding condylar length, mandibular length, mandibular body length, or ramus height. On the other hand, condylar width was 13.61% smaller in the malocclusion group. No significant differences were found between the right and left sides in either group.

BMD

BMDs of the alveolar bone, condyle, and mandibular angle for the two groups are shown in Table 6. Alveolar bone was 35.73%, condyle was 18.1%, and mandibular angle was 22.74% less on both sides in the malocclusion group.

DISCUSSION

Jaw Movements

Movement pattern. There were significant differences in the paths of jaw movement between normal and malocclusion mice in the sagittal plane but not the frontal plane. Characteristics of jaw movement trajectory are more marked in the sagittal plane, so changes related to malocclusion or food textures are more readily observed there. A study of posterior crossbite in humans reported a higher frequency of abnormal chewing patterns in the frontal plane, including reverse, cross, and chopping.³ However, unlike what is observed in humans and according to the results

obtained here, there are no characteristic chewing patterns in the frontal plane in mice with malocclusion. These differences between humans and mice may be due to the different morphology of the TMJ and the cusps of molars.¹³

We previously found an interesting feature of the chewing pattern in mice.¹⁰ Because the jaw-opening and jaw-closing paths diverge widely in the sagittal plane in mice, masticatory performance can be increased by extending the grinding path of the late-closing phase (Figure 2). This phase started earlier during hard-food chewing than during soft-food chewing. In the present study, the distance of the late-closing phase was smaller and the early-closing phase was larger in the malocclusion group. A previous study indicated that the late-closing phase coincided with the “power phase” of the chewing cycle, where most of the food grinding and possibly some tooth contact take place.¹⁰ The reduced late-closing phase indicated that the occlusal phase was shortened. Occlusal interference and the reduced occlusal contact area caused by malocclusion may make it difficult to grind and crush food efficiently. These results indicate that malocclusion during the growing period hinders development of a normal chewing pattern.

Cycle Duration

The total cycle duration, opening phase, and early-closing phase were prolonged in the malocclusion group.

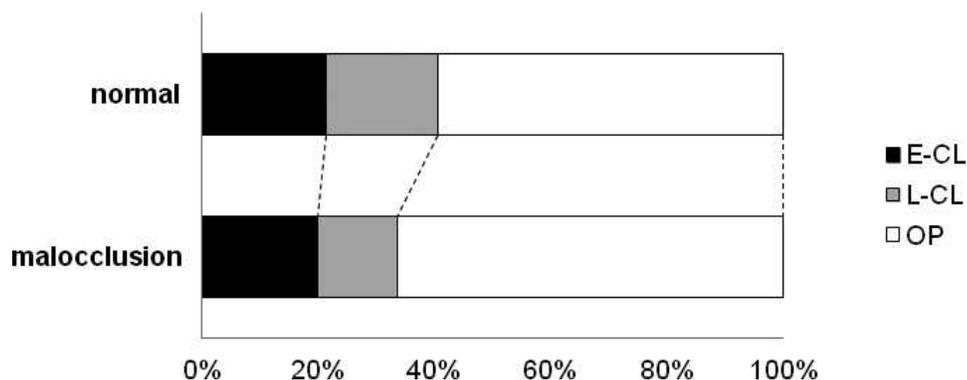


Figure 6. Percentage of the early-closing phase or late-closing phase in relation to total cycle duration in normal and malocclusion mice. E-CL indicates early-closing phase; L-CL, late-closing phase; OP, opening phase.

Table 4. Comparison of Muscle Activity Between Normal and Malocclusion Mice^a

	EMG Duration, ms			Average Amplitude, mV		
	R-MAS	L-MAS	R-DIG	R-MAS	L-MAS	R-DIG
Normal (n = 9)	60.82 ± 12.31	61.53 ± 9.49	121.67 ± 17.56	0.085 ± 0.019	0.079 ± 0.014	0.058 ± 0.028
Malocclusion (n = 9)	78.22 ± 18.95	73.90 ± 11.91	107.19 ± 21.33	0.067 ± 0.035	0.053 ± 0.036	0.078 ± 0.036
Significance	*	*	NS	*	*	NS

^a Values are presented as the standard error of the mean of each group. R-MAS indicates right masseter muscle; L-MAS, left masseter muscle; R-DIG, right digastrics muscle. Individual differences were tested using Mann-Whitney *U*-test.

* *P* < .05; NS indicates not significant.

Studies in humans reported that closing-phase duration was longer in the presence of crossbite than with normal occlusion.^{4,14} Our study indicates that the movement cycle was prolonged because malocclusion caused occlusal interference on the molars, which prevents smooth, effective jaw movements during mastication.

In contrast, a percentage of the duration of the late-closing phase in relation to the total cycle duration, corresponding to the occlusal phase, was reduced in

the malocclusion group. This finding was in agreement with a previous study, which suggested that occlusal interference reduces the duration of the occlusal phase in humans.¹⁵ It has been suggested that occlusal interference inhibits continuation of the isometric contraction during the occlusal phase via sensory input from the periodontal ligaments.

From the above, we concluded that the malocclusion group had difficulty establishing a smooth chewing

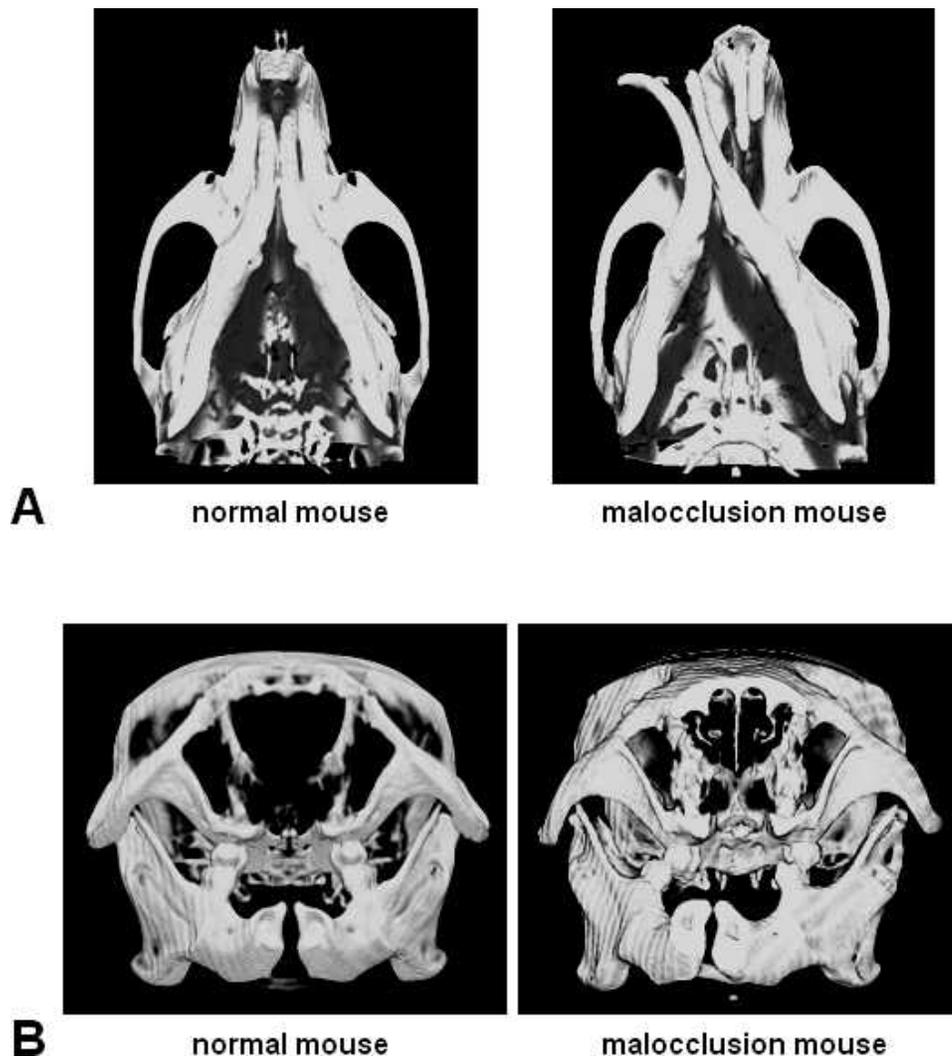


Figure 7. Micro-CT three-dimensional reconstruction image. (A) Ventral view. (B) Frontal view.

Table 5. Comparison of Mandibular Length Between Normal and Malocclusion Mice^a

	Normal Right Side (n = 9)	Malocclusion Right Side (n = 9)	Significance	Normal Left Side (n = 9)	Malocclusion Left Side (n = 9)	Significance
Condylar length, mm	1.78 ± 0.12	1.81 ± 0.26	NS	1.76 ± 0.16	1.75 ± 0.22	NS
Mandibular length, mm	10.54 ± 0.51	10.23 ± 0.35	NS	10.63 ± 0.39	10.23 ± 0.35	NS
Mandibular body length, mm	9.71 ± 0.38	8.96 ± 0.69	NS	9.82 ± 0.41	9.09 ± 0.68	NS
Ramus height, mm	5.97 ± 0.09	5.71 ± 0.39	NS	6.01 ± 0.31	5.68 ± 0.18	NS
Condylar width, mm	0.98 ± 0.12	0.83 ± 0.07	*	0.93 ± 0.07	0.82 ± 0.03	*

^a Values are presented as the standard error of the mean of each group. Individual differences were tested using Mann-Whitney *U*-test.

* *P* < .05; NS indicates not significant.

cycle because of occlusal interference with jaw movement. Consequently, the appropriate chewing pattern was not established, and masticatory efficiency was reduced. The increased number of chewing cycles required for grinding a certain amount of food may also indicate reduced masticatory efficiency.

Muscle Activity

In the present study, the duration of masseter muscle activity was increased in the malocclusion group. It is suggested that unstable occlusion extends the duration of EMG activity. Also, patients with malocclusion have less area of occlusal contact in the intercuspal position, so masticatory performance is poor.¹⁶ It is possible that our malocclusion mice compensated for their reduced masticatory efficiency with a longer period of muscle activity.

The present study also showed that the average amplitude of the masseter muscle activity was decreased in the malocclusion group. It has been reported that the average amplitude of the masseter muscle activity in crossbite patients is significantly smaller on the crossbite side than in normal subjects.¹⁷ The presence of tooth contact in an unstable position can cause discomfort or pain. Therefore, an inhibitory-protective reflex might activate to avoid injury of the stomatognathic system. According to a previous study,¹⁸ there was a positive correlation between occlusal stability in the intercuspal position and masseter muscle activity during mastication. The correlation between occlusal stability and elevator muscle function is probably based on feedback from periodontal receptors. It is believed that (1) various

masticatory muscles produce rhythmic activity that is regulated by the mCPG, and (2) sensory feedback from mechanoreceptors in the maxillofacial area modifies the basic chewing pattern and coordinates movements of the tongue, lips, and jaw. Based on these findings, it was concluded that periodontal receptors are primarily responsible for facilitating jaw-closing muscle activity.^{19,20} When sensory input from periodontal ligaments is blocked, the occlusal force is significantly reduced, and masticatory efficiency is decreased. The loss of masticatory efficiency may be compensated for by increasing the number of chewing cycles in a masticatory sequence.²⁰ In rats, occlusal hypofunction induced by bite raising causes decreased alveolar bone volume and density.²¹ This indicates that decreased mechanical stimulation of the periodontal ligament and surrounded alveolar bone may cause reduction of BMD of the jaw.

It is, therefore, suggested that masseter muscle activity decreases following a reduction in masticatory stimulation of the periodontal ligament. Persistence of this condition might inhibit the growth and development of masticatory muscles and their function.

Morphology

Condylar width and mandibular BMD were significantly less in the malocclusion group. It has been suggested that feeding soft food may cause a decrease in condylar width in growing rats.²² In our experiment, malocclusion is considered to cause reduced masticatory demand on teeth and, thereby, decreased masticatory muscle activity, which in turn may have affected bone mass and bone quality of the

Table 6. Comparison of BMDs Between Normal and Malocclusion Mice^a

	Normal (n = 9)	Malocclusion (n = 9)	Significance
Right alveolar bone, mg/cm ³	633.6 ± 93.4	396.8 ± 129.7	**
Left alveolar bone, mg/cm ³	630.8 ± 65.3	415.8 ± 134.8	**
Right condyle, mg/cm ³	1077.5 ± 69.4	888.5 ± 104.1	*
Left condyle, mg/cm ³	1095.3 ± 77.7	891 ± 103.2	*
Right mandibular angle, mg/cm ³	1005.6 ± 78.7	793.3 ± 184.3	*
Left mandibular angle, mg/cm ³	1018.1 ± 118.1	769.7 ± 98.3	**

^a Values are presented as the standard error of the mean of each group. BMD indicates bone mineral density. Individual differences were tested using Mann-Whitney *U*-test.

* *P* < .05; ** *P* < .01.

condyle. Likewise, a reduced late-closing phase (power phase of the chewing cycle) indicates decreased masticatory muscle activity, which could inhibit condylar growth and decrease BMD.

We conclude that malocclusion affected both mandibular growth and stomatognathic functional development in growing mice. We suggested that malocclusion may promote the further abnormal growth of the maxillofacial bone and inhibit the acquisition of normal jaw function in growing mice. Although results from animal studies cannot be transferred immediately into clinical practice, the results of this study point in the direction that malocclusion in humans should be corrected as early as possible to promote normal morphological growth and functional development.

CONCLUSIONS

- Compared to normal mice, growing mice subjected to a malocclusion model displayed parameters of morphology and function indicative of decreased masticatory efficiency.
- The late-closing phase (“power phase”) had a shorter duration in mice with malocclusion than in normal mice.
- The masseter muscle’s activity level was significantly lower in the malocclusion group than in normal mice. It is, therefore, suggested that malocclusion interferes with optimizing the chewing pattern and establishing appropriate masticatory function.
- Also, altered mechanical loading of the mandible may significantly impede normal growth of the mandible.

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