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Effects of Maxillary Molar Intrusion on the Nasal Floor and Tooth Root Using the Skeletal Anchorage System in Dogs

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ABSTRACT

The skeletal anchorage system (SAS) was developed to provide intraoral absolute anchorage for the intrusion or distalization of molars. The purpose of this study was to verify the effects of remarkable molar intrusion on the tooth root and the maxillary sinus floor. Six adult female beagles with fully erupted dentition were used. Titanium miniplates were implanted bilaterally above the maxillary second premolar root apices using pentobarbital anesthesia. The second premolars were intruded for four or seven months after three months of healing after implantation. Standardized dental radiographs were taken periodically to evaluate the amount of tooth movement and root resorption. After the experimental animals were fixed by perfusion at the end of each experimental period, the second premolars were dissected along with the surrounding alveolar bone. Undecalcified (60 µm thick) and decalcified (five µm thick) sections were prepared. The average extent of intrusion was 1.8 mm after four months and 4.2 mm after seven months. The root apices of the intruded molars penetrated into the nasal cavity. Remodeled bone around the intruded molar roots was rich in woven bone on the buccal side, whereas that on the palatal side was rich in lamellar bone. Nasal floor membrane and a thin layer of newly formed bone, which lifted intranasally, covered the intruded molar root. Root resorption partly reached into the dentine without the formation of reparative cementum, and little or no serious pathological changes were seen in the pulp of the intruded molars. SAS effectively intruded maxillary molars, but some moderate root resorption was observed.

KEY WORDS: Skeletal anchorage system, Intrusion, Nasal floor, Alveolar bone remodeling, Root resorption.

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INTRODUCTION Return to TOC

Controlling the posterior dentoalveolar height predominantly contributes to the vertical correction of skeletal open-bite. Because traditional biomechanical techniques cannot effectively control the vertical height of the molars, skeletal open-bite has been an indication for orthognathic surgery, especially in adult patients. We have developed a skeletal anchorage system (SAS) to provide an intraoral absolute anchorage for orthodontic tooth movement¹⁻³ in the distalization and/or intrusion of molars. SAS uses transmucosal titanium anchor plates as a temporary absolute anchorage to intrude molars without serious clinical iatrogenic problems. Because the application of SAS for molar intrusion is a fairly new technique in orthodontic treatment, the effects of molar intrusion on root resorption and the maxillary sinus are unknown.

We previously demonstrated mandibular molar intrusion using SAS in beagle dogs and reported that (1) anchor plates can be maintained in the oral cavity without severe inflammation around the implantation site, (2) root resorption was minimal and was repaired by the formation of new cementum, and (3) the inferior alveolar neurovascular bundle was not damaged because it seemed to reposition with tooth movement. Thus, we concluded that the SAS could be a useful clinical tool for mandibular molar intrusion without creating serious iatrogenic problems.⁴ The stability and osseointegration of titanium implants were extensively studied in the field of dental implants and good results were obtained.⁵ Many clinical investigations and experimental studies have indicated that endosseous implants inserted into the alveolar bone or the anterior palate are resistant to orthodontic forces and can be used to provide stable anchorage.^{6.7} But there is very little information available regarding anchor plates that are temporarily implanted in the maxilla through the oral mucosa for orthodontic anchorage.

The purpose of this study were:

- to clarify the effect of maxillary premolar intrusion on the bony nasal floor, which is histologically similar to the human maxillary sinus;
- to study root resorption and iatrogenic pulpal reactions induced by intrusion.

We used an animal model system with dogs to histologically elucidate the effects of maxillary molar intrusion and hypothesized that maxillary molar intrusion induced the remodeling of basal cortical bone of the nasal floor.

MATERIALS AND METHODS Return to TOC

Experimental animals and titanium miniplates

Six adult female 12-month-old beagles weighing 9.5–10 kg were used in this investigation. In all the dogs, the permanent teeth were already fully erupted and general growth was complete. The titanium anchor plates used for orthodontic anchorage were specifically designed as described in our previous study (Figure 1a).⁴ The anchor plates used in this study were developed by Sankin Kogyo Ltd (Tokyo, Japan) and the titanium bone screws (two mm diameter, five or seven mm long) were from Leibinger Co (Freiburg, Germany).

Surgical procedure and application of intrusive force on teeth

The procedures used in the present study were basically the same as those described in our previous report.⁴ In brief, for the surgical application of anchor plates, experimental animals were anesthetized and a local anesthetic was applied at the implantation sites of the anchor plates. A two-cm mucosal incision was made over the buccal vestibule in the second premolar region, the mucoperiosteal flap was reflected, and the anchor plates were fixed on the buccal side at the lateral cortical bone of the maxilla with three titanium monocortical bone screws. An additional monocortical bone screw was fixed at the middle of the palatal bone of the second premolar region (Figure 1c **O**). Furthermore, two titanium bone markers were inserted into the buccal cortical bone parallel to the margin of the alveolar bone. The bone markers were used as reference points to measure tooth movement by superimposing the tracings of standardized radiographs. The mucoperiosteal flap was repositioned and the surgical wound was sutured, with the long arm of the anchor plates exposed intraorally. The plate on the left side was used as an anchor for tooth movement and the plate on the other side was a nonloaded control.

To apply intrusive force to the premolar, a cast metal crown was attached and a superelastic closing coil spring (TOMY International Co, Tokyo) was tied to both the head of the palatal bone screw and the hook on the buccal anchor plates (Figure 1b,c). After a three-month healing period, an intrusive force (80–100 g) was applied to each maxillary premolar for four or seven months, and the coil springs were adjusted or changed every three weeks. Five second premolars were intruded for four months, five second premolars for seven months, and two second premolars and three third premolars remained unintruded as controls (Table 1). The experimental periods refer to the duration of the actual clinical application of intrusive force in open-bite cases for 5–9 months¹ and to the bone remodeling period in dogs.^{8.9} During the experimental period, the tooth and transmucosal portions of the anchor plates were cleaned by brushing and irrigation with 0.2% aqueous chlorhexidine.¹⁰ The protocols described above were reviewed and approved by the Animal Ethics Committee, Tohoku University School of Dentistry, and animal care was in accordance with their guidelines.

Radiographic evaluation of tooth movement and root resorption

To evaluate the amount of molar intrusion, standardized lateral radiographs were taken using a holding box attached to a removable bite plate that was designed to fix the head of each experimental animal. During the radiographic procedures, the dogs were under general anesthesia. Standardized radiographs with a 240-mm focus-object distance and a 24-mm object-film distance were taken every four weeks. Exposures were made at 90 kV and 200 mA for 1.5 seconds. The radiographic films were magnified five times with a mounted slide projector. The titanium anchor plates, screws, teeth, maxillofacial bones, and bone markers were traced. The amount of tooth movement at the five second premolars, defined as the distance between the two lines connecting the mesial and distal root apices and two bone markers, was measured by superimposing those tracings on the titanium bone markers. The amount of root resorption was measured as

the distance between the top of the bifurcation of the root to the line connecting the distal and mesial root apices. Actual tooth movement and root resorption were calculated by dividing the measurements by 5.

Statistical analysis

A Student's *t*-test (unpaired) was used for the statistical analysis of tooth movement between the four-month group and seven-month group. Paired *t*-test was used to evaluate root shortening before and after tooth intrusion. Tooth movement and root resorption were considered significant at *P* values of .05.

Vital staining to analyze bone remodeling during molar intrusion

Two experimental animals underwent the vital staining procedure.^{11–14} These two dogs were treated with 5% oxytetracycline (Sigma, St Louis, Mo), 0.16% Alizarin red (WAKO Chemicals, Osaka), and 1% Calcein (Dojin Kagaku, Tokyo). Each vital staining reagent was diluted in saline, fructolact solution (Otsuka Seiyaku, Tokyo) and saline, and then each dog was subcutaneously injected a dose of 20, 75, and 8 mg/kg of body weight, respectively. The experimental animals in which premolars were intruded for seven months received oxytetracycline injections during the first seven weeks, Alizarin red injections for the next four weeks and, after a one-week interval, Calcein tetracycline injections during the first six weeks every week after a one-week interval from the beginning of tooth movement, and then Calcein injections every week for six weeks.

Tissue preparation and histological evaluation of tooth movement and remodeling of the nasal floor

The experimental animals were killed under general anesthesia and perfused with 4% paraformaldehyde in pH 7.4 phosphate-buffered saline (PBS) by intravenous injection into the bilateral jugular veins. Their skulls were cut coronally through the bilateral second and third premolars, and specimens were fixed in the same fixative for another two days at 4°C. The specimens were divided into two groups that were subjected to a decalcified (n = 4) or undecalcified histological evaluation by contact microradiography (n = 2).

For decalcified samples, the specimens were thoroughly rinsed in PBS, and then decalcified in Plank and Rychlo solution or formalinformic acid solution for four weeks or four months, neutralized in 5% sodium sulfate, rinsed thoroughly in distilled water, dehydrated in a graded series of ethanol, and embedded in paraffin. Serial sections, six-µm thick, were cut longitudinally and stained with hematoxylin and eosin (HE) dye. On the other hand, undecalcified specimens were thoroughly dehydrated and embedded in Rigolac (WAKO Chemicals, Tokyo). Undecalcified sections were cut longitudinally (100 µm thick) using a saw microtome SP 1600 (Leica, Bensheim, Germany) and polished to a thickness of 60 µm, and observed under fluorescent microscopy.

Contact microradiographic analysis

Undecalcified sections were prepared as described above and subjected to contact microradiography to analyze bone formation around the intruded premolar roots and root resorption. The sections were microphotographed at 60 kVp and 25 mA for 10 minutes using a soft X-ray system (Softex, CMR type, Tokyo). Contact microradiograph films were developed in D19 (Eastman Kodak, Rochester, NY) for five minutes, allowed to dry for one minute, and then fixed in Fujifix (Fuji Film Co Ltd, Tokyo) for five minutes.

RESULTS <u>Return to TOC</u>

Tooth movement and alveolar remodeling

Maxillary premolars were intruded into the bony nasal floor in the seven-month experimental group (Figure 2 \bigcirc). During the first month of intrusive force (Figure 3 \bigcirc), tooth movement was minimal, and intrusion became apparent only in the second month. The average amount of intrusion increased linearly, and was 1.8 mm after four months and 4.2 mm after seven months. The maximum amount of intrusion was 4.3 mm and the minimum value was 3.8 mm after seven months of force application. There was no significant difference between the four-month and seven-month groups (P > .05).

Vital labeling samples and CMR (Figure 4 \bigcirc), histological analysis (Figure 5 \bigcirc) showed that the remodeling bone around the intruded second premolar roots might be rich in woven bone on the labial side, whereas bone deposition on the palatal side might be rich in lamellar bone. The vital staining dyes labeled the alveolar bone parallel to the surface of the tooth root and a new Haversian system formation was slightly observed on the palatal side as shown in the controls in Figure 4a \bigcirc . The experimental sample showed an atrophied alveolar bone and a new Haversian system formation on the palatal side of the tooth; a partial increase of osseous tissue might have been induced by bone remodeling on the buccal side (Figure 4b \bigcirc). Remodeled bone around the intruded premolar roots was rich in woven bone on the palatal side was rich in lamellar bone. The alveolar bone around the intruded premolar roots was rich in woven bone on the palatal side was rich in lamellar bone. The alveolar bone around the intruded premolar roots was rich in woven bone on the buccal side (Figure 4b \bigcirc). Remodeled bone around the intruded premolar roots was rich in woven bone on the buccal side, whereas bone deposition on the palatal side was rich in lamellar bone. The alveolar bone around the intruded premolar roots remodeled to encompass the root apices (Figures 4d and 5b,c \bigcirc).

Nasal floor remodeling

In the samples in which the premolars had intruded for four or seven months, the root apex of the second premolars penetrated the bony floor of the nasal cavity, as shown in Figure 5 •. Compared with the unintruded controls, the nasal floor membrane was lifted into the nasal cavity and covered the root apex of the intruding premolars in the four- and seven-month experimental groups. The normal nasal floor membrane consists of a pseudostratified ciliated epithelium with ciliated cells, goblet cells, basal cells, and some intermediate cells on a basal membrane. Below the basal membrane is the lamina propria, which contains serous glands and vessels. The thin bony wall was closely related to the apex of the maxillary premolar (Figure 5d •) in the controls. In the four- and seven-month groups, around the root apex of the intruding molars, the nasal floor membrane was lifted intranasally and the epithelium degenerated to a striated epithelium without serous gland cells (Figure 5e •).

Root resorption

Intruding premolars were resorbed at their apices; in some cases this extended into half of the dentin. The resorbed surfaces of the dentin or cementum were not repaired by new cementum in either of the groups (Figure 5b.c \bigcirc). There were no statistically significant differences in root length measured on radiographic films (Table 2 \bigcirc) between before and after intrusion for four or seven months. The amount of root resorption was 0.18 ± 0.18 mm (mean ± SD) in the seven-month group and was 0.1 ± 0.14 mm in the four-month group.

Dental pulp reaction

The pulp tissue in the control teeth showed a normal histological appearance (Figure 6a •). As shown in Figure 6c •, odontoblasts showed a characteristic columnar shape overlaid on the predentine layer. Although the pulp of intruding teeth showed no remarkable changes (Figure 6b,d •), the vacuolization of odontoblasts, mild disruption of odontoblastic layers, and a decrease in the thickness of the predentine layer were observed in the root pulp.

DISCUSSION Return to TOC

Experimental procedures

SAS has been used previously in clinical orthodontic treatment.^{1.2} In the present investigation, miniplates with three monocortical bone screws fixed in the maxilla provided absolute anchorage under orthodontic force loading. In reports concerning the maintenance of undisturbed osseointegration of dental implants, only slight inflammatory changes were reported in the peri-implant soft tissue when oral hygiene was good.^{15,16} In the present experiment because we maintained good oral hygiene by irrigating with 0.2% aqueous chlorhexidine and brushing around the transmucosal area of the anchor plates, although slight inflammatory changes were observed, there was no bone resorption that decreased the stability of the implants. Meticulous oral hygiene is important for maintaining favorable soft tissue conditions around the anchor plates for clinical orthodontic treatment with SAS.³

This experiment was carried out to show the effect of premolar intrusion on the maxillary sinus region. The maxillary sinus floor and nasal floor in humans are covered by a single layer of ciliated epithelium above the cortical bone of the maxilla. Similar to the human maxillary sinus floor, the nasal floor in dogs is covered by a single layer of ciliated epithelium on the cortical bone. Therefore, the nasal floor of the dog can be considered the equivalent of the human maxillary sinus, and our results may provide insight into the relevant processes in humans.

Molar intrusion and its influence on the nasal cavity

We believe this to be the first report in animals where maxillary premolars were intruded an average of 3.4 mm. Although the method of measuring tooth movement does not take into consideration possible angular changes in the tooth with intrusion, we were able to effectively intrude maxillary premolars. In both the four- and seven-month groups, modeled bone around the intruded molar roots was predominately in woven bone on the buccal side, whereas that on the palatal side was predominately in lamellar bone when compared with the other side in control samples. As Gorski (1998) described,¹⁷ woven bone can be remodeled more rapidly than lamellar bone, and, therefore, the alveolar bone on the buccal side could be remodeled more rapidly than that on the palatal side according to the intrusion of maxillary molars. Woven bone implies that bone formation is occurring in the area. The predominance of woven bone on the buccal side is most likely because of some osteogenic stimulus that may be a lingual root tip of these teeth during intrusion.

In samples in which the premolars had intruded, the root apex of the second premolars penetrated the bony floor of the nasal cavity. As the premolars intruded, the nasal mucosa was lifted and a thin epithelial layer covered the root apex. In the seven-month groups, the bone formation occurred abundantly around the premolar root in the direction at the root apices. A thin bony layer covered the root apex of intruding molars. In previous studies, investigators attempted to insert implants into the maxillary alveolar bone in dogs until the nasal floor was penetrated. Their results demonstrated that the hard and soft tissues around the penetrating implants were covered with connective tissue and coated with respiratory mucosa,¹⁸ and did not show any signs of adverse tissue reaction at the resolution level of the radiograph.¹⁹

After surgical removal of the sinus mucosa, the denuded sinus lining was re-epithelized by a flattened ciliated epithelium on lamina

propria displaying fibrosis and lacking serous glands.²⁰ The number of vessels in the regenerated mucosa significantly increased and there was no difference in blood flow between the operated cavities and their control sides.²¹

These results suggest that even after the intruding molars penetrated the maxillary sinus floor, tissue regeneration, including alveolar bone, could occur to reestablish normal periodontal conditions.

Root resorption and pulpal reaction

Orthodontic tooth movement including excessive intrusion of the anterior teeth produces different types of root resorption.^{22–24} Root blunting is a common type of root resorption and is usually corrected by the formation of cementum,^{25–31} except when the roots are extensively resorbed. Additionally, there is no loss of tooth stability or mobility from this type of root blunting.³² In our present investigation, the intruded premolars showed small-to-moderate amounts of apical root resorption and showed periodontal attachment. There was no stability or mobility from root blunting.

We previously reported⁴ that the resorptive surface of intruding mandibular molars was repaired by the formation of new cementum, even after excessive intrusion. But in the present study, reparative cementum in the resorption cavities was not observed in any of the intruding maxillary molars. These results suggest that the formation of reparative cementum in the resorption cavities could be inhibited under compressive force generated by the nasal membrane under orthodontic force loading. Because our study does not take into consideration the changes that would have occurred in root healing if the forces were stopped and provided for healing, further investigations are needed to clarify these points.

Histological studies^{33–36} revealed that the application of continuous intrusive force caused changes in pulp tissue, such as disruption of the odontoblastic layer, vacuolization of odontoblasts, and circulatory disturbances. Stenvik and Mjor³⁷ indicated in their in vivo studies that vacuolization of the odontoblast cell layer could result in the retardation or inhibition of predentine formation. In previous studies, ^{38–40} investigators attempted to evaluate the pulpal status of teeth after posterior maxillary or mandibular segmental osteotomy. They concluded that the ischemia caused by segmental osteotomy resulted in pulpal degeneration. In our present investigation, excessive maxillary molar intrusion may have produced some pulpal ischemia, which could cause the disruption and vacuolization of odontoblasts, whereas the pulp tissue generally seemed to be healthy. Because these degenerative changes were limited to the root pulp, they could be reactive responses to the root resorption that accompanies molar intrusion.

CONCLUSION Return to TOC

In our present and previous studies, we have demonstrated that the SAS can provide absolute anchorage for maxillary molar intrusion, whereas the periodontal and peri-implant tissues mostly remained healthy. The resorbed root apices that penetrated to the nasal floor were not repaired and the alveolar bone around the root apices was remodeled. Further study will be needed to clarify these points.

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TABLE 1. The Number of Experimental and Control Molars

	Second	Third	
	Premolar	Premolar	
Control group	2	3	
Four month group	5	0	
Seven month group	5	0	
Total number of teeth	12	3	

TABLE 2. Amount of Root Resorption after Molar Intrusion (mm)

	Four months (n = 5)		Seven months (n = 5)	
	Mean	SD*	Mean	SD
Root length				
Before intrusion	6.85	0.21	6.95	0.23
After intrusion	6.75	0.2	6.77	0.19
Amount of root resorption	0.1	0.14	0.18	0.18

* SD indicates standard deviation; n, number of specimens per group.

FIGURES Return to TOC



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FIGURE 1. SAS components and application of intrusive force to teeth. Orthodontic titanium miniplates and bone screws are shown in (a). To apply intrusive force to the molar, a cast metal crown was attached to the molar crown and a superelastic closing coil spring was tied to both the head of the palatal bone screw and the hook on the buccal miniplates (b, c)



Click on thumbnail for full-sized image.

FIGURE 2. Intrusion of the second upper premolar in the seven-month experimental group as shown in oral photographs (a) before and (b) after intrusion



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FIGURE 3. Tooth movement averaged 4.2 mm in the seven-month group. The solid line indicates the time-course changes in tooth movement in the seven-month group, whereas the broken line indicates those in the four-month group with standard deviation bars. There was no statistically significant difference between the four- and seven-month groups



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FIGURE 4. Vital labeling (original magnification 50x) and CMR images. Newly formed Haversian systems (arrowheads) and osseous tissue (arrow) were labeled with Calcein and tetracycline in (b). CMR images of an unintruded second premolar (c) and that of the fourmonth group (d) show that the alveolar bone around the intruding molar roots (arrowheads) was remodeled to encompass the root apices, and a thin bony layer (arrows) covered the root apex of the intruding molars. Bs, buccal side and Ps, palatal side



Click on thumbnail for full-sized image.

FIGURE 5. The second premolar intruded into the bony nasal floor. Photomicrographs of HE-stained sections from the (a) control, (b) four-month and (c) seven-month groups are indicated (original magnification 50×). The nasal floor membrane and a thin layer of newly formed bone covered the intruding molar root. Root resorption (arrowheads) partially reached into the dentine without the formation of reparative cementum. The nasal floor membranes are indicated in the (d) control, (e) four-month, and (f) seven-month groups. The nasal floor membranes were lifted intranasally and the epithelium had degenerated into striated epithelium (arrows) without serous gland cells (original magnification 200×). Bs, buccal side and Ps, palatal side



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FIGURE 6. Photo micrographic images of dental pulp tissues of the second upper premolars in (a) the control teeth and (b) the sevenmonth group. The pulp tissues of intruding teeth showed no remarkable changes (HE, original magnification 200×). The odontoblastpredentine region of a control tooth (c) and that of the seven-month group (d). The vacuolization of odontoblasts (arrows) and a decrease in the thickness of the predentine layer (arrowheads) were observed in the root pulp (HE, original magnification 400×)

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