Histomorphometric Evaluation of Cortical Bone Thickness Surrounding Miniscrew for Orthodontic Anchorage

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ABSTRACT

Background: Recently, the use of miniscrews as an anchorage device has become a routine approach in the orthodontic field. However, there is no report that has analyzed the healing process of the miniscrew, such as the thickness of the cortical bone, in the past.

Purpose: In the present study, to histologically assess the healing process of the osseous tissue surrounding miniscrews used as an orthodontic anchorage, the change in the thickness of the cortical bone was analyzed after 3, 6, and 12 weeks after the placement. Furthermore, the change in the bone-implant contact in different regions of the miniscrew during the initial healing period was also investigated.

Materials and Methods: Ninety-six miniscrews were placed in eight beagle dogs. After 3, 6, and 12 weeks of healing, a force of 200–300 g was applied to the force-applied groups for 12 weeks. Non-forced groups remained in the jaw without force application.

Results: In the non-forced groups, a significant amount of cortical bone was formed at the head of the miniscrew at the initial stage of the healing process in the maxilla. However, less cortical bone formation was observed in the mandible. After the force application, increased bone formation was observed within 1 mm of the miniscrew compared to other regions in both jaws. In the mandible, significantly less cortical bone was observed 3 and 6 weeks after the force application. Bone-implant contact revealed that the osseous tissue surrounding the miniscrew matured from the apex toward the head of the miniscrew.

Conclusion: We suggest that this sufficient amount of cortical bone at the initial stage of healing enables the immediate loading in miniscrews to resist against orthodontic force. Furthermore, less amount of cortical bone formed at the head of the miniscrew may be one reason for the higher failure rate in the mandible.

KEY WORDS: cortical bone, dog, histomorphometric, miniscrew, orthodontics

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Reprint requests: Dr. Teruko Takano-Yamamoto, Division of Orthodontics and Dentofacial Orthopedics, Tohoku University Graduate In the case of dental implants, long-term maintenance is required, and immediate loading is not recommended. In order to obtain adequate strength to resist heavy occlusal force, "osseointegration" is required in conventional dental implants.^{1,2} The concept of "osseointegration" has been redefined by several authors since Brånemark used the term in 1977.³ The recent definition of osseointegration is described as "direct

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bone apposition onto the surface of an implant: no interposed fibrous connective tissue between the implant and supporting bone; no movement when used for orthodontic anchorage."⁴ From the light microscopic histological prospective, the characteristic of osseointegration is that bone opposes the implant surface without an intervening organized collagenous and fibroblastic matrix.^{2,5} One of the histomorphometric indices that indicate the degree of osseointegration is known to be the bone-implant contact. However, successful osseointegration occurs even with low bone-implant contact in dental implants.⁶

In recent years, the use of miniscrews for anchorage has dramatically increased in orthodontic treatment.^{7,8} In contrast with conventional dental implants, immediate loading is known to be possible when miniscrews are used for anchorage in orthodontic treatment.^{7,8} In the case of miniscrews, long-term maintenance is unnecessary, and the implant will not be subjected to heavy occlusal force during the initial healing phase. Thus, "osseointegration" may not be necessary in the initial phase of orthodontic treatment using miniscrews. Furthermore, miniscrews have successfully served as orthodontic anchorage with less than 10% of bone-implant contact.9 Therefore, we suggest that sufficient mechanical interdigitation between the miniscrew and the cortical bone is an important factor that affects the stability of the screw-type implant anchor and enables immediate loading. However, there has been no study that has histomorphometrically analyzed the change in thickness and the amount of cortical bone and degree of osseointegration during the healing process when miniscrews are used for orthodontic anchorage.

Clinically, miniscrews utilized for orthodontic anchorage tend to have a lower successful rate in the mandible than in the maxilla.⁸ Furthermore, our animal data also showed that all failed miniscrews occurred in the mandible.⁹ From an anatomical perspective, the mandibular alveolar bone is known to have more compact and dense bone compared to that of the maxilla.¹⁰ Thus, if the amount of osseous tissue is related to the implant success rate, mandibular implants should have a lower failure rate. However, the reason for the higher failure rate of miniscrews in the mandible compared to the maxilla has not been investigated.

In this study, we histologically analyzed the changes in the thickness of cortical bone and bone-implant contact in three different locations of the miniscrews during the initial healing stage, and compared the maxilla and mandible in dog alveolar bone.

MATERIALS AND METHODS

A total of eight dogs (8-month-old males) were used in this study. In each maxilla and mandible, three out of six miniscrews (Titanium screw, Stryker® Leibinger, Kalamazoo, MI, USA, 5.0 × Ø1.0 mm) were subjected for experimental tooth movement, while the other three miniscrews served as control (total of 12 miniscrews for each dog). In the maxilla, miniscrews were placed between the first and second premolar, second and third premolar, or between the roots of the fourth premolar. In the mandible, miniscrews were placed between the first, second, third, and fourth, or between the roots of the fifth premolar. All of the miniscrew was placed at the attached gingiva. A small pilot hole with a round bar followed by a drill with a diameter of 1.0 mm was drilled into the cortical bone while using external irrigation. After the placement of the miniscrew, orthodontic force (200-300 g) by elastomeric chain was applied for 12 weeks after three different healing durations (3, 6, 12 weeks). Thus, only force-applied implants remained in the jaw for an additional 12 weeks of force application. Because the miniscrew was placed perpendicular to the teeth, we presume that the force was applied in the lateral direction.

The study protocol was reviewed and approved by the Indiana University Review Committee for animal care and use.

Maxillary and mandibular block specimens were harvested and dehydrated in an ascending series of ethyl alcohols, and cleared in xylene. The specimens were infiltrated with methylmethacrylate, containing 3% dibutyl phthalate, in a Shandon Hypercenter XP[™] automatic tissue processor (Shandon; Pittsburgh, PA, USA). The embedded specimens were serially sectioned at 110 to 120 µm in the sagittal plane with a Leica 1600 Saw Microtome[™] (Deerfield, MA, USA) or an EXAKT[™] cutting/grinding system (EXAKT Medical Instruments, Oklahoma City, OK, USA) and polished to approximately 100 µm for bright field microscopic examination.

Histomorphometric Analysis

Cortical Bone Thickness. The thickness of cortical bone was measured in three (1 mm within implant, 1–2 mm away from the implant, 3–4 mm away from the implant)



Figure 1 Schema of where the cortical bone thickness (A) and bone-implant contact (B) was analyzed. Cortical bone thickness was measured within 1 mm, 1–2 mm, and 3–4 mm from the miniscrew (A). Bone-implant contact was measured in three different locations (U, upper one-third; M, middle one-third; L, lower one-third).

locations of the alveolar bone surrounding the miniscrew (Figure 1A). Average thickness of the right and left side of the miniscrews was identified as the cortical bone thickness. In addition, the control data were obtained from similar locations in both maxilla and mandible without miniscrews. All measurements were performed using an Olympus BX60 microscope combined with an Olympus DP70 Digital Camera (Tokyo, Japan) and WinROOF image processing software (Mitani Corp., Tokyo, Japan).

Bone-Implant Contact in Different Locations. Histomorphometric analysis was performed using a Nikon FXA equifluorescent microscope (Nikon Inc., Melville, NY, USA) using stereological point-hit and linear intercept methods at magnifications of ×100 with a 10×10 point ocular square grid.¹¹ The measurements and calculations followed standard nomenclature and formulas (bone-implant intercept/total intercept %).¹² Measurements were performed in three different locations (upper one-third, middle one-third, lower one-third) in the miniscrew (see Figure 1B). Microradiographic images were produced using a FaxitronTM (Hewlett-Packard, Beaverton, OR, USA).

Statistical Methods

One-way analysis of variance (ANOVA) models and Fisher's protected least significant difference for *post hoc*

comparison were used at the level of p < .05 to compare the following parameters: difference of cortical bone thickness between different healing duration and locations, and bone-implant contact between different locations, healing stage, and between jaws.

RESULTS

Cortical Bone Thickness

Non-Forced Group. After 3 weeks of healing, significant amount of woven bone formation was observed at the alveolar bone surrounding the miniscrew. In the maxilla, compared to the control, significantly thinner cortical bone was observed in 1–2 mm and 3–4 mm away from the implant in the 3-week group (Table 1) (Figure 2, A and B).

In the mandible, significantly thinner cortical bone was observed in all of the measurement in 3-week groups compared to the control (see Table 1). In some sections, there was almost no bone formed at the upper region of the miniscrew in the mandible. Thus, significantly less cortical bone was observed within 1 mm (see Figure 2, C and D) compared to that in 1–2 and 3–4 mm away.

Force-Applied Group. In the maxilla, thicker cortical bone was noticed within 1 mm from the implant at 12 weeks compared to 3–4 mm away (Table 2) (see

TABLE 1 C	TABLE 1 Cortical Bone Thickness in Non-Forced Groups																			
	3-week								6-we	12-week										
	Control		ontrol <1 mm		1–2 mm		3–4 mm		<1 mm		1–2 mm		3–4 mm		<1 mm		1–2 mm		3–4 mm	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Maxilla																				
Cortical bone thickness	832.4	161.0	728.5	226.4	566.3*	189.1	525.5*	136.7	766.4	409.9	725.9	220.2	782.6	225.2	850.9	142.1	871.4	88.2	853.5	101.2
Mandible																				
Cortical bone thickness	1,357.1	360.2	471.2 ^{*†}	124.0	857.4*	226.7	944.7*	227.1	1,123.6	131.4	1,161.2	131.4	1,158.0	257.7	1,441.7	423.4	1,535.6	230.8	1,409.0	271.3
unckness																				

*Significant difference compared to the control (p < .05). †Significant difference compared to 1–2 and 3–4 mm away (p < .05).

Figure 2, E and F). However, the thickness gradually decreased to the control level as further away from the implant.

In all locations of 3- and 6-week mandible, thinner cortical bone was noticed when compared with the control group (see Table 2) (see Figure 2, G and H).

Bone-Implant Contact

Non-Forced Group. In the 3-week middle and lower regions of the maxilla (Figure 3A), significantly higher bone-implant contact was observed compared to that in the 6- and 12-week groups (Table 3). In the upper region of 6- and 12-week (see Figure 3B) groups, significantly higher bone-implant contact was observed compared to that in the middle and lower regions. Furthermore, significantly higher bone-implant contact was observed in the maxillary upper region of the 3-week group compared to the mandible.

In the mandible 3-week group (see Figure 3C), significantly higher bone-implant contact was observed in the middle and lower regions compared to the upper region (see Table 3). In the upper region, 6- and 12-week (see Figure 3D) groups were significantly higher than the 3-week group. In the lower region, the 3-week group was significantly higher than the 6- and 12-week groups, and the 6-week group was significantly higher than the 12-week group. In the 12-week group, the upper region was significantly higher than that of middle and lower regions.

Force-Applied Group. In the maxilla, significantly more bone-implant contact was observed in the upper region compared to the middle and lower regions in all maxillary groups (Table 4) (see Figure 3E). Significantly more bone-implant contact was observed in the middle region compared to the lower region in all maxillary groups.

TABLE 2 C	TABLE 2 Cortical Bone Thickness in Force-Applied Groups																			
	3-week								6-w	eek			12-week							
	Control		<1 mm		1–2 mm		3–4 mm		<1 mm		1–2 mm		3–4 mm		<1 mm		1–2 mm		3–4 r	nm
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Maxilla																				
Cortical bone	832.4	161.0	847.9	302.8	694.3	230.1	714.7	123.3	887.0	152.3	765.8	153.2	761.6	63.7	1,117.0*	258.6	947.4	309.2	921.1	101.2
thickness																				
Mandible																				
Cortical bone	1,357.1	360.2	951.9*	174.8	752.1*	104.5	775.6*	127.6	950.6*	332.8	949.3*	279.7	948.8*	239.5	1,401.1	193.5	1,141.0	276.8	1,095	212.8
thickness																				

*Significant difference compared to the control (p < .05).



Figure 2 Light microscopic photographs (A, C, E, G) and microradiograph (B, D, F, H) at the same location. After 3 weeks of healing in the maxilla (A, B), intense new woven bone (W) formation is observed under the original lamellar (L) cortical bone. After 3 weeks of healing in the mandible (C, D), significantly less bone was observed at the head of the miniscrew (arrow heads). After 12 weeks of force application in maxillary 12-week healing group (E, F), thicker cortical bone was formed at the head of the miniscrew compared to other areas. On the other hand, thinner cortical bone was observed at 6-week mandible force-applied group (G, H). Arrows indicate where the cortical bone thickness was measured.

In the mandible, significantly more bone-implant contact was observed in the upper region compared to the lower region in all groups (see Table 4) (see Figure 3F).

DISCUSSION

In this study, cortical bone thickness in dog maxilla was approximately 800 μ m, and 1,200 μ m in the mandible. The dog cortical bone thickness was about half compared that in humans measured by CT.¹³ In the maxilla in the early phase (after 3 weeks) of healing process, there was no change in the cortical bone thickness within 1 mm of the miniscrew. Histological findings indicated the formation of new woven bone tissue within 1 mm around the miniscrew after 3 weeks in the maxilla. We suggest that this woven bone tissue was formed in the early stage of the healing process, which maintained the thickness of the cortical bone. After 3 weeks, approximately 30% of bone-implant contact was observed in all regions (upper, middle, and lower) around the miniscrew. After 6 weeks, increased bone-implant contact was only observed at the upper, whereas it decreased in the middle and lower regions of the miniscrew. This indicates that as the surrounding alveolar bone heal, osseous tissue decrease in the lower region and increase in the upper region of the miniscrew, and returns to the original form of the cortical bone. Thus, a significant amount of bone maintained cortical bone thickness at the head of the implant even in the early stage of the healing process, enabling immediate loading in the maxilla.



Figure 3 Photographs of low magnification of non-forced (A–D) and force-applied (E, F) maxilla (A, B, E) and mandible (C, D, F) miniscrew. A significant amount of bone formation is observed after 3 weeks of healing (A), whereas after 12 weeks (B), bone formation is only observed at the upper region of the miniscrew. Notice less bone formation at the upper region of the miniscrew after 3 weeks of healing (D) in the mandible, a significant amount of bone was formed at the upper region as well as in the maxilla. After the force application, the amount of bone tended to increase at all regions in both the maxilla (E) and mandible (F).

Interestingly, miniscrews are known to have higher failure rate in the mandible compared to the maxilla.⁸ From the past reports, surgical technique (such as root proximity of the placed miniscrew)¹⁴ and the placement methods (self-drilling vs. self-tapping)¹⁵ are known to be one of the reasons for miniscrew failure. However, previous studies have not focused on the histological feature during the healing process between the two jaws, and have not histomorphometrically analyzed that might have a great impact on the stability of the miniscrew. In this study, 3 weeks later in the mandible, less cortical bone thickness was observed in all areas compared with the control. In contrast to the maxilla, bone-implant contact showed less bone at the upper level compared to middle and lower regions. This indicates that in the mandible, the recovery of cortical bone thickness has not been achieved after 3 weeks of healing. During the recovery period, the same tendency was observed in the mandible as in the maxilla, and finally achieved a normal cortical bone structure after 12 weeks. However, less bone formed around the head of the miniscrew in the early stage of implant healing in the mandible, and also after the force application, may be one reason for the higher failure rate of the

TABLE 3 Bone-Implant Contact in Non-Forced Groups																		
		3-w			6-we	12-week												
	Upper		Middle		Lower		Upper		Middle		Lower		Upper		Middle		Low	er
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Maxilla																		
Bone/implant	32.5*	4.1	30.5^{\dagger}	13.2	32.1^{\dagger}	12.3	39.9 [‡]	10.1	14.6*	8.7	11.4	5.3	40.1^{\ddagger}	8.2	8.5*	3.2	5.3	4.7
contact (%)																		
Mandible																		
Bone/implant	15.6†‡	4.3	31.6	7.4	41.6^{\dagger}	12.2	39.0 [§]	17.1	29.4	12.1	19.4 [¶]	5.5	43.8 [‡]	16.3	32.1	10.5	7.4	7.6
contact (%)																		

*Significant difference compared to the mandible (p < .05).

[†]Significant difference compared to 6 and 12 weeks (p < .05).

^{*}Significant difference compared to middle and lower (p < .05).

[§]Significant difference compared to lower (p < .05).

[§]Significant difference compared to 12 weeks (p < .05).

mandible than the maxilla. Therefore, clinically, we may have to avoid immediate heavy loading in the mandibular miniscrews, and wait approximately 6 weeks of healing to obtain sufficient cortical bone for immediate loading.

In the maxillary force-applied group, significantly less cortical bone thickness in the area away from the miniscrew (1–2 and 3–4 mm) was observed compared to the control at 3 weeks. Thus, by loading in the early stage (3 weeks), less bone may form in the area away from the miniscrew in the maxilla. At 12 weeks after, significantly thicker cortical bone thickness was observed within <1 mm compared to both the control and 3–4 mm from the miniscrew in the maxilla. From the results of bone-implant contact in the maxilla, increased osseous tissue was observed only at the upper region. This result is consistent with the previous studies that indicated an increase in the amount of bone in immediate-loaded dental implants.^{16,17} Some studies have shown that orthodontic force applied to the miniscrew^{9,18} and dental implant¹⁹ increased the bone turnover rate, which resulted in bone apposition. We suggest that force does not have a negative impact on the healing process at the miniscrew but may increase the amount of osseous tissue surrounding the miniscrew in the maxilla.9 Furthermore, microdamage is known to occur by force application to the dental implant, resulting in increased bone formation rate.²⁰ Orthodontic force may cause microdamage, resulting in increased bone remodeling that may lead to an increased amount of osseous tissue surrounding the miniscrew. Therefore, clinically, immediate loading may be preferable in the maxilla,

TABLE 4 Bone-Implant Contact in Force-Applied Groups																		
		3-we		6-we		12-week												
	Upper		Middle		Lower		Upper		Middle		Lower		Upper		Middle		Low	/er
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Maxilla																		
Bone/implant	49.0*	7.6	26.4^{\dagger}	9.0	15.9	10.8	51.8*	12.8	26.5^{\dagger}	8.9	14.6	8.3	58.8*	9.5	29.0^{\dagger}	13.5	17.0	11.4
contact (%)																		
Mandible																		
Bone/implant	42.4^{\dagger}	11.6	38.6	11.7	30.8	10.2	47.3^{\dagger}	11.0	37.9	12.8	25.3	9.8	52.8†	13.3	39.6	12.3	29.3	9.4
contact (%)																		

*Significant difference compared to middle and the lower (p < .05). †Significant difference compared to lower (p < .05). because continuous orthodontic force might increase the bone turnover and as a result, enhance the stability of the miniscrew.

From this study, there were significant differences in the healing process of the alveolar bone surrounding the miniscrew between the maxilla and mandible. The bone-implant contact data in this study, in the mandible, significantly less bone-implant contact was observed at the head of the implant (upper one-third) compared to other areas. However, in the maxilla, there was no significant difference between the areas, and significantly more bone-implant contact compared to the same location (upper one-third) in the mandible. In addition, significantly higher bone turnover rate was observed in the maxilla compared to the mandible.²¹ Clinical data have indicated a lower successful rate in the mandible than in the maxilla with the use of miniscrews for orthodontic anchorage.^{7,8} Taken together, less boneimplant contact at the head of the implant and lower bone turnover rate in the mandible may be one reason for the lower stability of miniscrews during the initial phase of implant healing. Therefore, longer healing duration might be required in miniscrews that was placed in the mandible from clinical point of view.

In conclusion, a significant amount of cortical bone at the head of the miniscrew in the initial stage of the healing process may be a key factor for immediate loading as an anchorage in orthodontic loading. As healing progresses, cortical bone matures mainly at the upper region of the miniscrew, and returns to original shape of cortical bone. Furthermore, less cortical bone formed at the head of the miniscrew in the initial stage of healing may be one reason for the higher failure rate in the mandible.

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