Characterization of the Correct Mandibular Premolar Region for Delayed Dental Implantation in Beagle Dogs

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For delayed dental implantation into the mandible, the implant size should be chosen according to the characteristics of that bone. This study investigated anatomic features of the mandible in beagle dogs, to develop recommendations regarding the correct implantation region and available bone area for delayed dental implantation surgery. We used 20 healthy male beagle dogs to create delayed dental implantation models. The dogs' mandibles underwent cone beam CT (CBCT) imaging; the locations of the middle mental foramen and canine root apex were measured on CBCT images. The dogs then were euthanized and their mandibles measured by using a digital vernier caliper. In addition, the correct implantation region and available bone areas were evaluated. The data obtained by using the 2 measuring methods were compared statistically. The results showed that the positions of the middle mental foramen and canine root apex were relatively fixed, with little variation. The implantation and available bone regions showed little variation among dogs and did not differ significantly between the 2 measuring methods. In conclusion, the correct implantation region (mean ± 1 SD) in the beagle mandible for delayed dental implantation surgery was 17.53 \pm 0.46 mm in width. The recommended available bone areas (height \times width) were 7.22 \pm 0.68 mm $\times 5.32 \pm 0.49$ mm (P2), 8.21 ± 0.71 mm $\times 5.81 \pm 0.56$ mm (P3), and 9.17 ± 0.65 mm $\times 6.39 \pm 0.56$ mm (P4) in the premolar region.

Abbreviation: CBCT, cone-beam computed tomography

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Dental implantation is a favorable treatment option for oral rehabilitation of people who have lost teeth. To improve dental implantation treatment options, preclinical studies that evaluate all related factors are needed. Animal models have served as tools to study dental implantation, playing a key role in preclinical implantology studies in recent decades. Various species of animals, including mice, guinea pigs, dogs, sheep, goats, and NHP, have been used as models of dental implantation. However no single animal model is ideal for studying human bone development and repair.¹⁹ Among available models, pigs are most similar to humans in terms of tooth shape, bone density, anatomic structure, and rate of remodeling and healing.^{4,11} But pigs are not the first choice as animal models because of their high growth rates, body size, and excessive weight.¹⁵ The jaws of beagle dogs possess many similarities to human jaws in physiology and pathology and can often hold human-sized implants, which make them a better choice as models to study dental implantation.¹⁹ Increasing attention has been paid to the beagle model since the first attempt to put implants into beagles' mandibles in 1975.6

The mandibular premolar region of beagles is the most commonly used area to study dental implantation. In this region lie the mental foramen, canine teeth, and mandibular canal, which are remarkably important and should be avoided when performing implantation. However little dental implantation research that is performed in strict accordance with the anatomic characteristics of the beagle mandible, which are incompletely defined, and involving the mental foramen, canine teeth, or mandibular canal can lead to a poor implantation outcome. Therefore, defining the anatomy of the beagle mandibles is crucial. In dental implantology studies, implantation is often delayed,

in dental implantology studies, implantation is often delayed, given that early resorption of the alveolar margin crest can lead to unsatisfactory osseointegration after immediate implantation in beagles.^{7,17} However, before delayed implantation is performed, various anatomic marker features of beagle mandibles can disappear as the extraction socket heals. Specifically, the socket can be sealed by the alveolar margin of the cortical bone, with replacement of the woven bone by bone marrow.² Thus, without accurate anatomic markers, implants can't be precisely inserted into the correct location. Therefore, we sought to localize the mental foramen, canine root apex, and mandibular canal and determine the correct implantation and available bone areas in a delayed implantation model using beagles.

To study the anatomic features of the beagle mandible, we created delayed dental implantation models. Crucial landmarks, including the mental foramen, canine root apex, and mandibular canal, were measured on cone-beam CT (CBCT) images and anatomic specimens. We hope that the data derived from this study provides useful information regarding delayed dental implantation in beagles.

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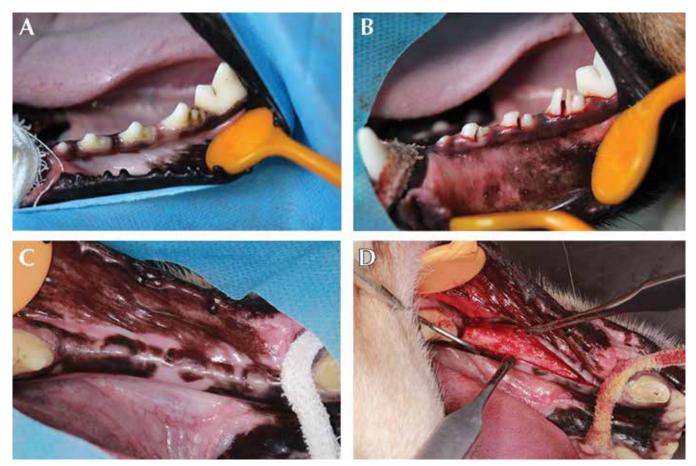


Figure 1. Mandibular premolars (P1, P2, P3, and P4) were extracted atraumatically by using an elevator.

Materials and Methods

Animals and surgical procedure. All research protocols were conducted in strict accordance with the principles of medical ethics and were approved by the IACUC of Chongqing Medical University, China. Purebred male beagle dogs (n = 20; age, 12 to 14 mo; weight, 10 to 12 kg) supplied by the Department of Laboratory Animals at Chongqing Medical University underwent maxillary sinus lift surgery. The dogs had normal dentition and had no periodontal disease. All dogs were fed a nutritious diet and maintained in separate cages (20 to 26 °C; 30% to 60% humidity).⁸

Before surgery, animals' oral cavity was cleaned using a toothbrush. All the dogs were anesthetized by intramuscular administration of 3% pentobarbital sodium (0.5 mL/kg; Merck, Darmstadt, Germany) combined with 4% xylazine hydrochloride (0.05 mL/kg; Huamu Animal Health Care, Jilin, China) according to previous reports.^{9,18} Articaine was used to provide local anesthesia in the surgical region. After induction of anesthesia, the dogs' vital signs, including heart rate, respiration, blood pressure, and body temperature, were monitored. An external heat source was used to keep the dogs warm. During surgery, to keep dogs' respiratory tracts unobstructed, their oral cavities were kept open, and excessive oral discharge was removed quickly. An endotracheal tube was placed for oxygen administration in the event of respiratory complications. Emergency protocols, including intravenous infusion, normal saline, nikethamide, and sodium hydrogen carbonate, were in place in case of anesthetic complications.

Bilateral mandibular premolars (P1, P2, P3, and P4) were extracted atraumatically by using an elevator after the double

roots were sectioned by using a diamond-coated bur (Figure 1).¹⁰ The sockets were sutured after irrigation with normal saline. To prevent infection after surgery, penicillin (Huabei Pharmacy, Shijiazhuang, China) and meloxicam (0.2 mL/ kg; Boehringer-Ingelheim, Ingelheim am Rhein, Germany) were administered for 3 d. The operative region was washed frequently with aqueous hydrogen peroxide solution to prevent infection.

Sample preparation. After 3 mo for healing, all dogs were euthanized by intravenous overdose of 3% pentobarbital sodium (1.5 mL/kg; Merck, Darmstadt, Germany) combined with xylazine hydrochloride (0.15 mL/kg; Huamu Animal Health Care). The healed mandibles were disarticulated from the temporomandibular joint bilaterally. All mandibles were fixed in 4% paraformaldehyde solution at 4 °C after sharp dissection of soft tissues.

Macroscopic observation. The positions and characteristics of the mental foramen, canine, premolar region of the mandible, alveolar crest, and mandibular canal were examined visually, recorded, and measured by using vernier calipers (Figure 2).

CBCT scanning and measurement. For CBCT scanning, the dogs were in horizontal position with the heads fixed by a head-positioning device. The bilateral mandibles were scanned by CBCT (QR-DVT 9000, NewTom, Verona, Italy) using a cesium iodide flat-panel detector, with the center rays of CBCT targeted to the center of the sample. The scanning parameters were 120 kVp; 10 mA; field of view, 16 cm (diameter) × 11 cm (height); voxel, 0.25 mm³; and scan time, 24 s. The locations of the mental foramen, canine teeth, and mandibular canal and the vertical height and horizontal width of the correct implantation region on the CBCT images were measured (Figures 3 and 4).

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Figure 2. Profile of the beagle mandible. (A) Maxillofacial view. (B) Buccal view. (C) Lingual view. (D) Right lingual view. (E) Left lingual view.

To localize the canine teeth, the following parameters were used (Figure 5). The distance between the distal point of the alveolar crest (B) and the mesial point of the first molar alveolar crest (A) was defined as D1 (the straight line between A and B). The distance between the distal point of the alveolar crest (B) and projective point (C') of the apex (C) of the canine root on D1 was defined as D2 (the line between B and C'). D2/D1 was defined as P1, the relative position of the canine root apex. To locate the mental foramen, the distance between the distal point of the alveolar crest (B) and the distal point (D) of the middle mental foramen and its projective point (D') on D1 was defined as P3 (the line between B and D'). D3/D1 was defined as P2, the relative position of the mental foramen. The caudal mental foramen wasn't studied given its relative unimportance in dental implantation.

To measure the vertical height of the correct implantation region, the distance between A and D' (D4) was used. D4 was divided into 3 equal segments: S1, S2, and S3. At the center of each segment, the cross-sectional plane was selected as the region of interest to measure the vertical height. The distance between the most coronal point of the alveolar crest and the most coronal point of the mandibular canal was defined as the vertical height. The vertical heights obtained from S1, S2, and S3 were named as H1, H2, and H3, respectively.

To measure the horizontal width of the correct implantation region, cross-sectional planes (which were 2 mm below the crest of the alveolar margin) at the centers of S1, S2 and S3 were selected, and the marginal widths (W1, W2, and W3, respectively) were measured. The crest of the alveolar margin, which was about 2 mm in height, was excluded due to its irregular thin-blade shape. Therefore, the vertical height for

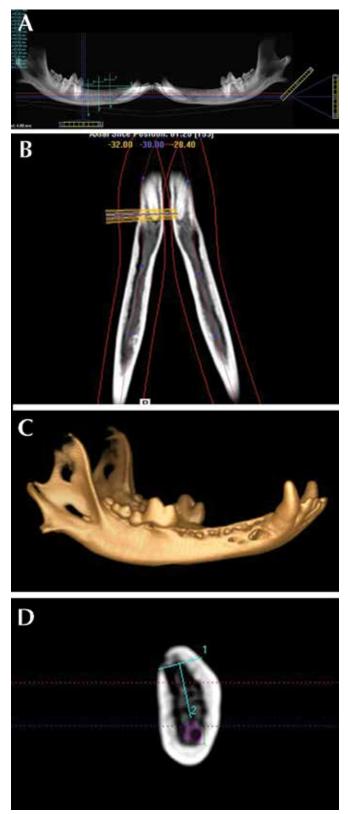


Figure 3. CBCT images of the beagle mandible. (A) Lingual view. (B) Maxillofacial view. (C) Buccal view. (D) Cross-sectional view.

implantation was the distance between the most coronal point of mandibular canal and the point 2 mm below the crest of the alveolar margin (Figure 4 B).

Anatomic measurement. The same parameters measured on CBCT images were also measured on the mandible specimens.

The cross-sections through the center of the alveolar margin crest of S1, S2, and S3 in the correct implantation region were obtained. The buccal bone plate of the canine was removed to expose the canine apex. All measurements were obtained by using a digital vernier caliper. All measurements were done 3 times by 2 operators independently. The average values were used for statistical analysis.

Statistical analysis. Statistical analysis was performed using SPSS 22.0 (SPSS, Chicago, IL). Data were normally distributed and are presented as mean ± 1 SD for continuous variables and as frequencies with percentages for categorical variables. Independent *t* tests were used to compare the measurements from the 2 methods. A *P* value less than 0.05 was considered to be statistically significant.

Results

Macroscopic observation. All beagle dog models were created successfully. Each side of the mandible had a middle mental foramen, which was bigger, and a caudal mental foramen, which was smaller. The lingual surface of premolar region of the mandible tended to be relatively flat in the inferior margin, tilting gradually toward the crest of the margin. The alveolar crest line protruded to the buccal side gradually from P1 to P4; M1 was located at the middle of the crest of the margin. The alveolar margin crest was buccolingually narrow. The canal occupied the majority of the lower part of the mandible, thus differing greatly from that in humans. The relatively large root of the canine tooth curved distally.

Location of the canine root apex and mental foramen according to mandible specimens and CBCT images. The mean distance between the canine teeth and the first molar, D1, was 41.55 ± 1.75 mm according to anatomic specimens and was 41.70 ± 1.61 mm according to CBCT (Table 1). Measurements for D2, D3, P1, and P2 were similar between the 2 measuring methods. None of the data differed significantly between measurement methods. The positions of the canine root apex and mental foramen were rather fixed, with little variation.

Vertical height and horizontal width of the available implantation bone region. To obtain the correct implantation region, the available vertical height and horizontal width were measured (Table 2). None of the measurements differed between the 2 methods.

Discussion

Dental implantation typically is done in beagle dogs, even though precise mandibular anatomic characteristics have not been available.¹⁹ The bone–implant contact value, which is derived from the lower segment of the overlong implant into the mandibular canal, is unreliable.³To our knowledge, the current study is the first research on anatomic structures in a beagle model of delayed dental implantation. In this study, we chose male beagles 12 to 14 mo old and weighing 12 to 15 kg because, at this age, beagles' teeth have erupted completely, and their mandibles are mature.¹⁴ In addition, younger dogs have greater healing ability.⁵

In this study, we measured various landmarks on both CBCT images and mandible specimens. The 2 sets of data obtained from the 2 measuring methods had no significant difference (P > 0.05). A previous study of beagles' anatomic and morphologic characteristics used X-rays, which are less accurate than CBCT and anatomic specimens.¹⁶ As previously reported, CBCT scans and anatomic specimens showed high reliability in an

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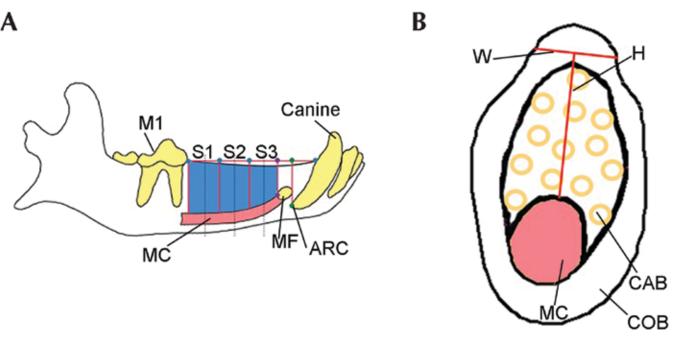


Figure 4. Diagram illustrating the measuring method. (A) Buccal view of the mandible. The blue area is the correct implantation area. ARC, apex of the root of canine; M1, first molar; MC, mandibular canal; MF, mental foramen. (B) Diagram showing the vertical height and horizontal width of the correct implantation region. The cross-sections passing the center of the alveolar margin crest of S1, S2 and S3 in the correct implantation region were used to measure the vertical height (H1, H2, and H3) and horizontal width (W1, W2, and W3). CAB, cancellous bone; COB, cortical bone; MC, mandibular canal.

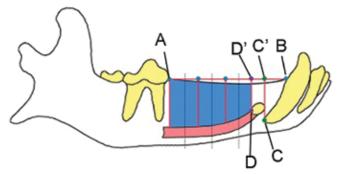


Figure 5. The points and lines used for localization of the canine root apex and mental foramen. A, Mesial point of the first molar alveolar crest; B, distal point of the canine alveolar crest; D1, the straight line between A and B; C, the apex of canine root; C', the projective point of the canine root apex on D1; D2, the line between B and C'; D, the distal point of the mental foramen; D', the projective point of the distal point of the line between A and D'; and D4, the line between A and D'.

Table 1. The locations of the canine teeth and mental foramen obtained from the 2 measuring methods

	CBCT images	Anatomic specimens	Р
D1 (mm)	41.70 ± 1.61	41.55 ± 1.75	0.269
D2 (mm)	11.79 ± 1.04	11.63 ± 1.09	0.102
D3 (mm)	14.44 ± 0.95	14.38 ± 1.0	0.546
P1 ^a	$28.4\% \pm 2.5\%$	$28.0\% \pm 2.8\%$	0.144
P2 ^b	$34.8\% \pm 2.5\%$	$34.7\% \pm 2.7\%$	0.877

^aP1 was D2/D1, which represented the relative position of the canine root apex.

^bP2 was D3/D1, which represented the relative position of the mental foramen.

Table 2. The vertical height (H) and horizontal width (W) of the correct implantation region obtained by using the 2 methods

Parameters	CBCT images	Anatomic specimens	Р
H1 (mm)	7.25 ± 0.78	7.22 ± 0.68	0.775
H2 (mm)	8.22 ± 0.52	8.21 ± 0.71	0.956
H3 (mm)	9.26 ± 0.79	9.17 ± 0.65	0.423
W1 (mm)	5.22 ± 0.59	5.32 ± 0.49	0.104
W2 (mm)	5.73 ± 0.59	5.81 ± 056	0.266
W3 (mm)	6.28 ± 0.41	6.39 ± 0.56	0.126

investigation of the correct interradicular zones for miniscrew implantation in beagles.²⁰

We determined the anatomic positions of mental foramen, canine root apex, and mandibular canal to evaluate the correct implantation area. The root apex of the canine teeth is in front of the middle mental foramen. The bulky curved roots of the canine teeth renders the P1 site unsuitable for holding a human-sized implant. In a CT study of brachycephalic dogs, the height of the mandibular canal decreased slightly from the mental foramen to the molar region.^{12,13} We inferred from the edentulous mandibles specimens that the variation tendency of the mandibular canal in beagles was similar to brachycephalic dogs: the course between alveolar margin crest and the mandibular canal increases gradually from premolar region to molar region. Unlike that in humans, the mandibular canal of beagles is quite large, with thin buccal and lingual bone walls, and the canal occupies nearly all of the lower part of mandible horizontally. Avoiding the mandibular canal when performing dental implantation is crucial. We do not recommend using the molar region as an insertion site in beagles because it is difficult to reach, even with the help of a mouth gag. In addition, we thought prudent to retain the molars because they are very important for maintaining normal occlusion.

In regard to the vertical aspect, we considered the available bone height (the distance between the alveolar margin crest and upper wall of the mandibular canal) as the optimal vertical area. The vertical area for implantation increased from the P2 to P4 site (Table 2). According to the proposed scheme for classifying human dental implants on the basis of length, we consider that 'short' (length, 6 to 10 mm) and 'very short' (shorter than 6mm) implants are relatively appropriate for implanting into beagles, but we do not recommend using 'long' (that is, 10 mm or longer) implants.¹ The implant's size should be chosen depending on the insertion site. Therefore, our study likely provides guidance regarding choosing an appropriately sized dental implant and maximizing the available jaw height without damaging the mandibular canal.

The present study showed that the width of alveolar margin crest in beagle mandibles (Table 2) was narrower than in humans. According to the proposed scheme for classifying dental implants on the basis of diameter, we consider that 'narrow' (diameter, 3.0 to 3.75 mm) and 'very narrow' (less than 3.0 mm) to be appropriate for implanting into beagles, but we do not recommend using 'standard' or 'wide' (diameter, 3.75 mm and larger).¹ In addition, the choice of dental implant for use in beagles might be limited by what is commercially available for humans. Considering the greater thickness of the cortical bone in beagle dogs than that in humans, narrow implants (with smaller diameters) are preferable.

This study had several limitations. Further study is needed to confirm the efficacy of the suggested implantation region. Given the data derived from this study, we are conducting delayed implantation experiments using custom-designed implants. In conclusion, the correct implantation region in the beagle mandible for delayed dental implant surgery was 17.53 \pm 0.46 mm in width. The recommended available bone areas were 7.22 \pm 0.68 mm \times 5.32 \pm 0.49 mm (P2), 8.21 \pm 0.71 mm \times 5.81 \pm 0.56 mm (P3), and 9.17 \pm 0.65 mm \times 6.39 \pm 0.56 mm (P4) in the premolar region.

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References

- Al-Johany SS, Al Amri MD, Alsaeed S, Alalola B. 2016. Dental implant length and diameter: a proposed classification scheme. J Prosthodont 26:252–260.
- Araújo MG, Lindhe J. 2005. Dimensional ridge alterations following tooth extraction. An experimental study in the dog. J Clin Periodontol 32:212–218.

- Baoji B, Xie Z, Zhang Y, Lu C, Huang D. 2012. [Establishment of beagle dog model for the study of the dental implant.] J Chinese Oral Implantol 17: 58–61. [Article in Chinese].
- 4. **Boutrand J-P, editor.** 2012. Biocompatibility and performance of medical devices. Philadelphia (PA): Woodhead Publishing
- Cardoso HFV. 2008. Epiphyseal union at the innominate and lower limb in a modern Portuguese skeletal sample, and age estimation in adolescent and young adult male and female skeletons. Am J Phys Anthropol 135:161–170.
- Cranin AN, Schnitman PA, Rabkin SM, Onesto EJ. 1975. Alumina and zirconia coated vitallium oral endosteal implants in beagles. J Biomed Mater Res A 9:257–262.
- Discepoli N, Vignoletti F, Laino L, de Sanctis M, Muñoz F, Sanz M. 2014. Fresh extraction socket: spontaneous healing vs immediate implant placement. Clin Oral Implants Res 26:1250–1255.
- 8. Holmstrom SE, Bellows J, Juriga S, Knutson K, Niemiec BA, Perrone J; American Veterinary Dental College. 2013. 2013 AAHA dental care guidelines for dogs and cats. J Am Anim Hosp Assoc 49:75–82.
- Koshman YE, Herzberg BR, Seifert TR, Polakowski JS, Mittelstadt SW. 2018. The evaluation of drug-induced changes in left ventricular function in pentobarbital-anesthetized dogs. J Pharmacol Toxicol Methods 91:27–35.
- Lommer MJ. 2012. Complications of extractions, p 153–159. Chapter 16. In: Verstraete FJM, Lommer MJ, editors. Oral and maxillofacial surgery in dogs and cats. Philadelphia (PA): Saunders.
- Mardas N, Dereka X, Donos N, Dard M. 2013. Experimental model for bone regeneration in oral and craniomaxillofacial surgery. J Invest Surg 27:32–49.
- 12. Martinez LA, Gioso MA, Lobos CM, Pinto AC. 2009. Localization of the mandibular canal in brachycephalic dogs using computed tomography. J Vet Dent 26:156–163.
- Martinez LAV. 2010. Determination of the mandibular canal course by means of computerized tomography in mandibles of brachycephalic and mesaticephalic *Canis familiaris*. Vet Radiol Ultrasound 51:199–199.
- 14. **Pasupuleti MK, Molahally SS, Salwaji S.** 2016. Ethical guidelines, animal profile, and various animal models used in periodontal research with alternatives and future perspectives. J Indian Soc Periodontol **20:**360–368.
- Reichert JC, Saifzadeh S, Wullschleger ME, Epari DR, Schütz MA, Duda GN, Schell H, van Griensven M, Redl H, Hutmacher DW. 2009. The challenge of establishing preclinical models for segmental bone defect research. Biomaterials 30:2149–2163.
- Santos M, Carreira LM. 2016. Insight of dogs' inner mandible anatomy using mathematical models. Anat Histol Embryol 45:479–484.
- Schultes G, Gaggl A. 2001. Histologic evaluation of immediate versus delayed placement of implants after tooth extraction. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 92:17–22.
- Tang Z, Jiang LP, Wu JY. 2015. Effect of maxillary expansion on orthodontics. Asian Pac J Trop Med 8:944–951.
- Wancket LM. 2015. Animal models for evaluation of bone implants and devices: comparative bone structure and common model uses. Vet Pathol 52:842–850.
- Wang Z, Li Y, Deng F, Song J, Zhao Z. 2008. A quantitative anatomical study on posterior mandibular interradicular safe zones for miniscrew implantation in the beagle. Ann Anat 190:252–257.