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ORIGINAL RESEARCH

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Fractal analysis of mandibular bone in children with delayed formation or agenesis of the mandibular second premolar germ

Fatma Saraç[1,*], Sinem Öztürk[1], Fatmanur Erdem Mert[1], Seda Nisa Uçar[1], Sera Derelioğlu[1], Aybike Baş Öztürk[1]

[1]Department of Paediatric Dentistry, Faculty of Dentistry, Atatürk University, 25240 Erzurum, Türkiye

***Correspondence**
saracfatma@atauni.edu.tr
(Fatma Saraç)

Abstract

Background: The presence of dental developmental anomalies, such as hypodontia or delayed tooth germ formation, is a common etiological factor contributing to malocclusion. Understanding whether these anomalies affect mandibular trabecular bone structure is crucial for risk assessment and interceptive orthodontic planning. This study aimed to evaluate whether delayed formation or congenital absence of the mandibular second premolar germ alters the trabecular architecture of the mandible in children and adolescents. **Methods**: Two groups were studied (n = 48): delayed formation group (n = 24) and agenesis group (n = 24), age- and sex-matched at fractal dimension (FD) analysis. Standardized panoramic radiographs were processed using ImageJ software to calculate FD values for two regions of interest: the furcation area and the mandibular angulus. **Results**: There were no significant differences in age or gender between the groups at the time of FD analysis (mean age = 5.75 years in both groups). No significant differences in FD values were found between the two groups ($p > 0.05$). However, both groups exhibited significantly higher FD values in the mandibular angulus region compared with the furcation area ($p < 0.05$). **Conclusions**: These results suggest that the distinction between delayed formation and agenesis of the mandibular second premolar germ was not associated with major differences. Larger longitudinal studies are needed to further explore the relationship between trabecular architecture and dental developmental anomalies.

Keywords

Hypodontia; Malocclusion; Tooth agenesis; Fractal dimension; Pediatric dentistry; Mandibular trabecular bone

1. Introduction

The term hypodontia refers to the developmental absence of one or more teeth in either the primary or permanent dentition, and is one of the most common dental developmental anomalies [1]. The reported prevalence of hypodontia in the permanent dentition (excluding third molars) is approximately 6.4% (95% Confidence Interval (CI): 5.7–7.2%) according to a recent systematic review [2]. The most frequently missing permanent teeth, in descending order, are the third molars, second premolars, and maxillary lateral incisors; whereas agenesis of the maxillary central incisors, canines, and first molars is very rare [3, 4]. Hypodontia can lead to malpositioned teeth, malocclusion, impaired chewing function, periodontal damage, insufficient development of maxillary and mandibular bone height, and negatively affect esthetics, phonation, and overall quality of life [5].

The development of second premolars shows greater individual variation compared with other permanent teeth. Nolla reported that the dental crypt of the mandibular second premolars becomes evident at around the age of 3 years [6]. Another longitudinal study indicated that the development of second premolars starts between the ages of 3 and 3.5 years in most individuals [7]. However, the literature also emphasizes that in some individuals, the development of these teeth may be delayed until the age of 9–10 years [8, 9]. This highlights the importance of carefully considering age when clinically diagnosing “absence of tooth germ”.

Congenital absence or delayed development of posterior teeth, especially the mandibular second premolar, is strongly associated with mesial drift of adjacent teeth, arch length discrepancies, and space loss, which can lead to complex malocclusion patterns and long-term occlusal instability [9–11]. Longitudinal studies have reported that early detection of hypodontia and timely decision-making regarding space maintenance or closure significantly affect treatment outcomes and long-term occlusal stability [8, 11].

Management of congenitally missing teeth generally in-

cludes either orthodontic space closure or prosthetic replacement (implants or fixed prostheses) [12]. Retention of primary teeth may be considered if root conditions are favorable, although root resorption can occur later. The choice of treatment is strongly influenced by the timing of diagnosis, as late germ formation may complicate treatment planning [13].

In a previous study, the trabecular structure of the mandibular bone in individuals with and without tooth agenesis was evaluated using fractal analysis, and differences were identified [14]. In contrast, Créton *et al*. [15] reported no significant difference in the trabecular structure of the mandibular bone between individuals with hypodontia and healthy controls. Although several studies have evaluated mandibular trabecular bone in individuals with and without hypodontia, to the best of our knowledge, there are no studies focusing specifically on individuals exhibiting late tooth germ formation.

The morphological characteristics of bone tissue can be assessed using various methods, including invasive histomorphometry [16], panoramic indices [17] on conventional radiographs, advanced imaging such as micro-computed tomography (micro-CT) [18], and densitometric methods like dual-energy X-ray absorptiometry [19]. However, both micro-CT and dynamic histomorphometry require either invasive biopsy or expose the patient to high radiation, making them ethically unsuitable for pediatric research [16]. The fractal dimension (FD) observed in radiographic images has been reported to reflect changes in trabecular bone density and mineral loss [20–22]. As the structural complexity increases, so does the FD value. The box-counting method introduced by Rudolph *et al*. [23] is one of the most commonly used techniques in fractal dimension analysis for evaluating the morphology of trabecular bone and marrow spaces. In this method, a box form ranging from 2 to 64 pixels is generated based on the radiographic image, and the FD is calculated from the slope of the log-log plot of the number of boxes versus box size.

In this context, the aim of our study was to evaluate the trabecular structure of the mandibular bone in children who exhibited late tooth germ formation—a relatively unexplored area in the literature—using fractal analysis, and to compare them with age- and gender-matched individuals without tooth germ formation. The unique aspect of this study lies in performing the fractal analysis during the early developmental period, prior to the initiation of mandibular second premolar development, to compare the trabecular structure of the mandibular bone between children with delayed tooth germ formation and those with agenesis. The hypothesis of the study was that the trabecular structure of the mandibular bone differs between these two groups.

2. Materials and methods

This retrospective cross-sectional study was conducted with the approval of the Non-Interventional Ethics Committee of the Faculty of Dentistry, Atatürk University (15.04.2025/41). The data were obtained from patients who presented for follow-up and treatment at the Pediatric Dentistry Clinic of Atatürk University Faculty of Dentistry, who were either diagnosed with late formation of the mandibular second premolar germ or found to have no germ formation, and who had consecutive panoramic radiographs taken between June 2018 and April 2025.

To ensure standardization, only developmental anomalies of the mandibular second premolar were included in the study group. The classification of late tooth germ formation was based on the stages defined by Nolla [6] as well as the developmental stages of the other teeth in the same individual. Fractal analysis was performed during the stage when neither group exhibited any clinical or radiographic signs of tooth germ formation (Nolla Stage 0). This approach is allowed for enabling an early comparative evaluation of fractal dimension values between groups.

Group 1 consisted of 24 children and adolescents with delayed mandibular second premolar germ formation, while Group 2 included 24 age- and gender-matched children and adolescents with agenesis of the mandibular second premolar. Each individual in both groups was matched according to age and gender based on the date of the panoramic radiograph used for fractal analysis.

Radiographs with inadequate image quality, patients diagnosed with systemic conditions affecting bone metabolism or using related medications, those with syndromes or genetic disorders, and individuals with apical or periodontal pathology in the region of the missing tooth were excluded from the study.

2.1 Sample size calculation

Sample size was calculated in G*Power 3.1.9.3 (Heinrich Heine University Düsseldorf, NRW, Germany) for an independent-samples t test (two-tailed) with $\alpha = 0.05$ and power $(1 - \beta) = 0.80$. The anticipated effect size (Cohen's $d = 1.208$) was derived from the closest available study in the literature [24], using the reported means and standard deviations of FD between hypodontia and controls. The required total sample size was 32 participants (16 per group); to increase power, we enrolled 48 participants (24 per group).

2.2 Radiographic examination and fractal analysis

All digital panoramic radiographs were acquired using the same panoramic unit (ProMax®, Planmeca Oy, 00880 Helsinki, Finland). Exposure settings were standardized at approximately 65 kVp, 5 mA, and 16.2 seconds. Patients were positioned according to the manufacturer's instructions, with the Frankfort plane parallel to the floor and the sagittal plane aligned with the vertical red laser line of the orthopantomogram machine. All panoramic radiographs were saved in Digital Imaging and Communications in Medicine (DICOM) format. Each pixel corresponded to 28.4 μm in size.

All panoramic radiographic measurements were performed by a single trained observer (FS) using ImageJ 1.3 software (National Institutes of Health, Bethesda, MD, USA). Images were upscaled four times in both x and y directions using bilinear interpolation. For each patient, a region of interest (ROI) measuring 25 $\times$ 75 pixels was standardized. Two different ROIs were selected on each panoramic film:

ROI 1: The furcation area of the second primary molar located in the region of the missing or undeveloped mandibular

second premolar.

ROI 2: Below the mandibular canal in the mandibular angle region.

Anatomical structures, such as the mandibular canal, cortical bone, lamina dura, and periodontal space, were excluded from the ROI areas (Fig. 1).

A modified version of the method originally described by White and Rudolph [23] for analog panoramic radiographs was adapted for use with digital panoramic radiographs. The ROIs in each image were selected and duplicated twice. A Gaussian filter (sigma = 2) was applied to the second duplicate to eliminate brightness fluctuations associated with the superimposition of soft and various bone tissues. The blurred image was then subtracted from the first duplicate that retained the original image. The image, originally in 16-bit lossless DICOM format, was converted to 8-bit grayscale and saved in Tagged Image File Format (TIFF) format. To isolate the bone marrow spaces and trabeculae, a gray Red Green Blue (RGB) value of 128 was added, and the image was binarized using a threshold value of 128, enabling easier identification of trabecular structures and marrow spaces. Subsequently, the image was inverted and skeletonized. The resulting skeletonized structure was overlaid onto the original image to visually confirm the alignment of the skeleton with the trabecular structure. The FD of the skeletonized image was then calculated using the box-counting method (Figs. 2,3).

To assess intra-observer reliability, 24 radiographs were reanalyzed by the same examiner after a two-week interval. The reliability of repeated measurements was evaluated using the Intraclass Correlation Coefficient (ICC) with a two-way mixed effects model and absolute agreement definition. In addition, Cronbach's Alpha was calculated to test internal consistency.

2.3 Statistical analysis

Data analysis was performed using IBM SPSS Statistics 29.0.1.0 (IBM Corp., Armonk, NY, USA). The normality of data distribution was evaluated using the Shapiro-Wilk test. For comparisons between independent groups, either the Independent Samples *t*-test or the Mann-Whitney U test was used. For comparisons between dependent groups, the Wilcoxon Signed-Rank Test was applied. The distribution of gender between the groups was analyzed using the Chi-Square test with Continuity Correction (Yates' correction). A *p*-value of less than 0.05 was considered statistically significant.

3. Results

The study included a total of 48 participants, with 24 individuals in the delayed tooth germ formation group (Group 1) and 24 individuals in the tooth agenesis group (Group 2). Since Group 1 and Group 2 were matched by age and gender at the time of the radiographs used for fractal analysis, no statistically significant differences were found between the groups in terms of age and gender (p age = 0.473, p gender = 1.000). The distribution of mean ages at the time of the radiographs used for fractal analysis is presented in Table 1. The mean age at the time of radiographic diagnosis of hypodontia was 10.6 ± 1.7 years.

It was observed that the fractal values measured from the furcation (ROI 1) and angulus (ROI 2) regions were similar between the groups ($p > 0.05$, Table 2). No significant differences in fractal measurements were observed between the groups based on gender ($p > 0.05$).

The mean ROI 2 values measured in Group 1 were found to be higher than the mean ROI 1 values ($p = 0.003$, Fig. 4). Similarly, the mean ROI 2 values measured in Group 2 were higher than the mean ROI 1 values ($p < 0.001$, Fig. 4).

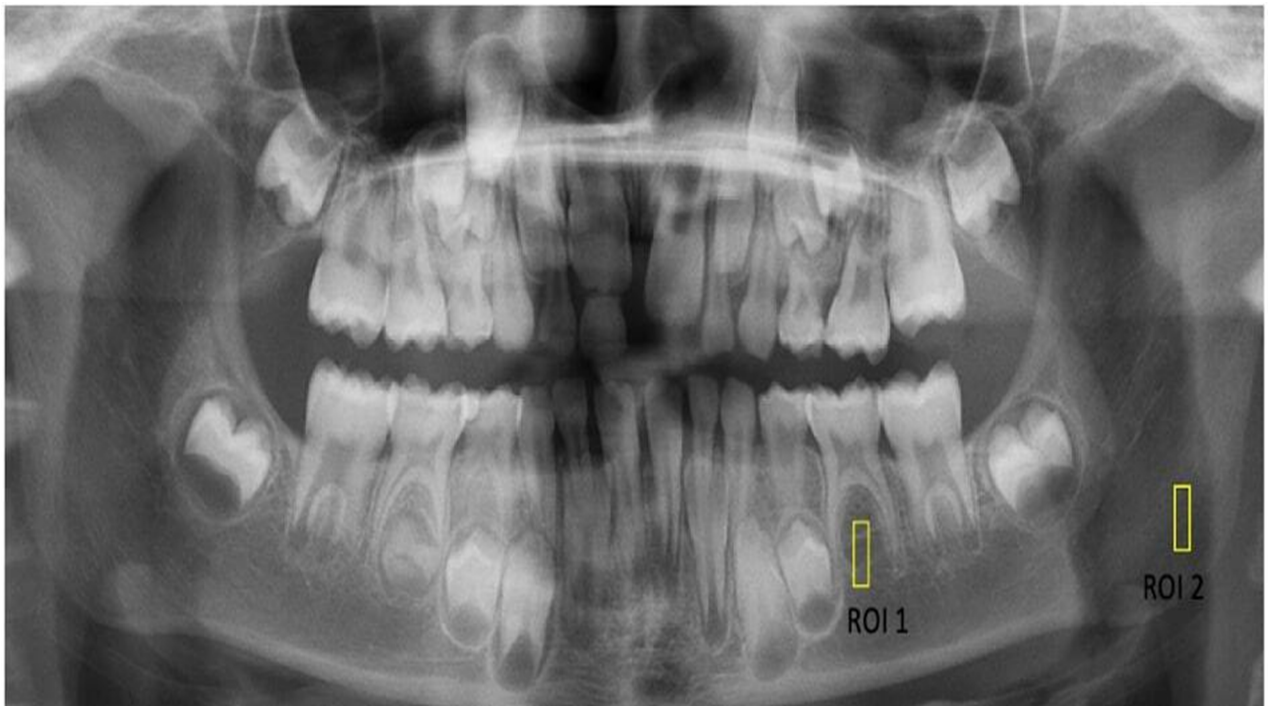

FIGURE 1. Selection of ROI 1 and ROI 2 on a panoramic radiograph. ROI: region of interest.

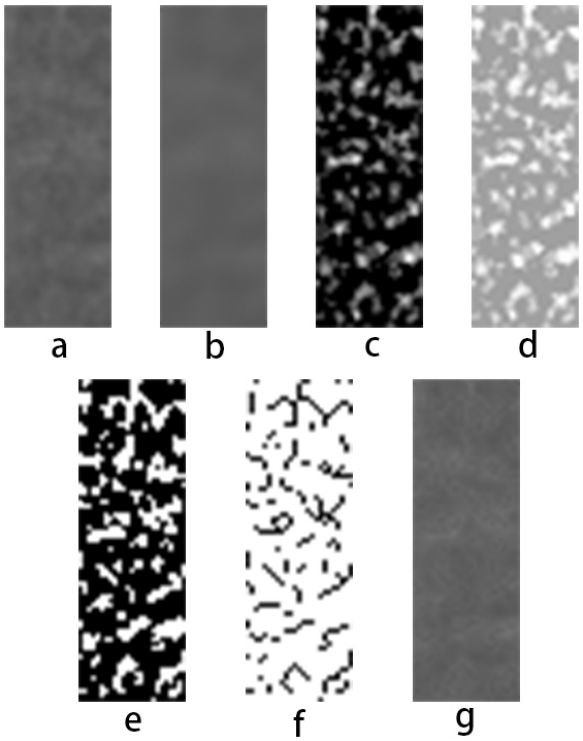

FIGURE 2. Steps of fractal analysis on panoramic radiographs. (a) Raw image before processing. (b) Gaussian blurred image. (c) Result of subtracting blurred image from raw image. (d) Grayscale offset. (e) Binarized. (f) Invert, skeletonize. (g) overlay.

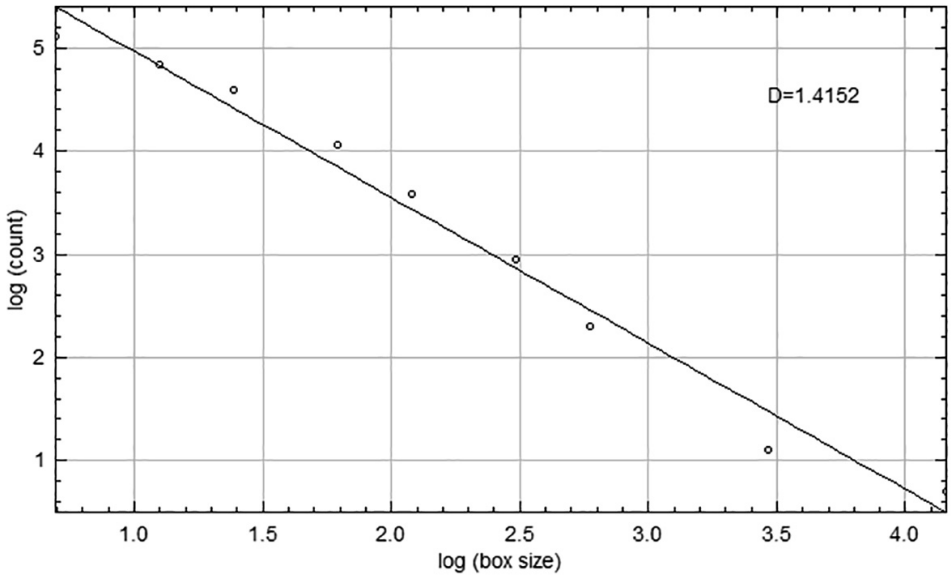

FIGURE 3. Fractal dimension calculation of the images with box counting method.

TABLE 1. Distribution of mean ages and gender at the time of radiographs used for fractal analysis.

Variable	Delayed premolar germ group (n = 24)	Premolar agenesis group (n = 24)	*p*-value
Age (yr) (mean ± SD)	5.75 ± 1.1	5.75 ± 0.9	0.473
Gender (F/M)	13/11	13/11	1.000

Independent Samples T-Test; F: Female; M: Male; SD: standard deviation.

TABLE 2. Mean fractal measurements by groups, mean ± SD.

ROI	Delayed premolar germ formation group (n = 24)	Premolar agenesis group (n = 24)	*p*-value (between groups)
ROI 1 (Furcation)	1.42 ± 0.03	1.41 ± 0.03	0.743
ROI 2 (Angulus)	1.45 ± 0.02	1.46 ± 0.02	0.516

Mann-Whitney U, Independent Samples T-Test. SD: standard deviation; ROI: region of interest.

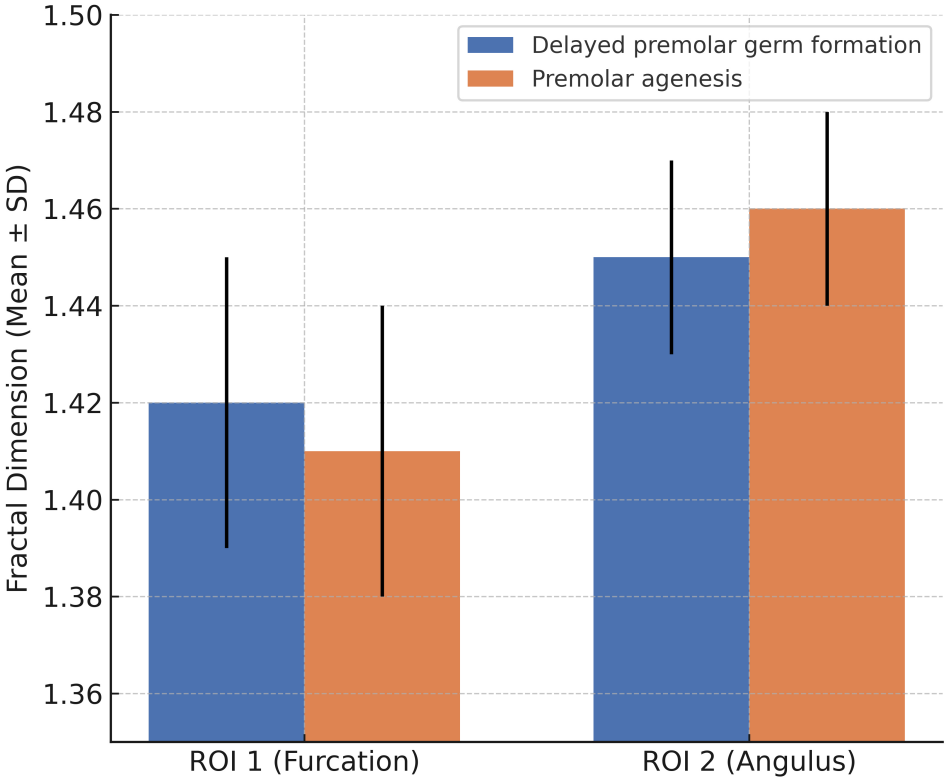

FIGURE 4. Graph of the mean fractal dimension measurements for ROI 1 and ROI 2 in each group. SD: standard deviation; ROI: region of interest.

The intra-observer reliability was found to be high. The ICC was 0.740 for single measures and 0.851 for average measures (95% CI: 0.424–0.963, $p = 0.005$), indicating good to excellent agreement. Cronbach's Alpha was 0.847, further confirming internal consistency.

4. Discussion

Hypodontia is a frequently encountered condition in dental practice, and its treatment protocol is complex. The management of hypodontia requires a multidisciplinary approach involving a combination of orthodontics, fixed/removable prosthetics, and implant surgery. Treatment strategies vary depending on the child's age and stage of dental development [8, 25]. In orthodontic treatment, a reduction in local alveolar bone density—resulting from enhanced remodeling in accelerated orthodontic protocols—has been associated with faster tooth movement and, thus, reinforces the need for increased anchorage in regions of low bone density [26]. Therefore, evaluating the bone condition in patients with hypodontia may provide valuable information for clinicians. Although radiographic evaluation is considered the standard for the definitive diagnosis of hypodontia, imaging should not be performed prematurely, as the mineralization of permanent mandibular second premolars may occur as late as 9–10 years of age [8, 9]. The distinction between late germ formation and complete agenesis is crucial for treatment planning. To the best of our knowledge, no previous study has evaluated the trabecular structure of the mandibular bone in patients with late-forming tooth germs compared with those with agenesis. According to our results, no statistically significant differences were found between the groups in the fractal analysis of orthopantomographs matched by age and gender. These findings suggest that late germ formation does not cause a measurable change in the trabecular structure of the mandibular bone. Therefore, our study hypothesis was rejected. However, the results still provide clinically relevant insights for future research.

Our study revealed no significant differences in the mandibular trabecular bone structure between individuals with late-forming dental germs and those without germ

formation when evaluated using fractal analysis. In the pediatric population, the mandibular bone differs significantly from that of adults due to ongoing growth and remodeling processes. In pediatric patients, the trabecular bone undergoes dynamic changes in both mineral density and trabecular organization [27]. The high bone turnover rate and incomplete skeletal maturation in children may allow the bone to adapt more rapidly to local dental stimuli. As a result, the effects of late germ formation on the trabecular structure may be masked by these adaptive responses. Future studies should consider conducting subgroup analyses based on bone developmental stages to improve interpretability. Although fractal analysis provides insights into bone structural complexity, it may be insufficient in detecting subtle and localized changes associated with late tooth germ formation. Similarly, Créton *et al*. [15] reported no significant differences in mandibular trabecular structure between individuals with and without hypodontia. Our results differ from Temur *et al*. [14]. Notably, their fractal analysis sampled three sites—(i) the center of the ramus above the mandibular foramen, (ii) a corpus region between the apical level of the mandibular molars and the superior border of the mandibular canal, and (iii) the missing-tooth region located in the apical third on the mesial side of the erupting or fully erupted first permanent molar. None of these sites corresponds to the interradicular furcation of the second primary molar, which was our primary, localized ROI. Because FD is sensitive to loading environment and trabecular orientation, differences in ROI location and scale can materially affect FD estimates. This methodological divergence likely contributes to the discrepant findings, and underscores the need for standardized, anomaly-adjacent ROIs and harmonized preprocessing protocols in future studies. From a clinical perspective, hypodontia and delayed tooth germ formation contribute to arch length discrepancies and space loss, which may complicate malocclusion patterns and treatment decisions. Accurate differentiation is therefore essential for orthodontic planning [5]. These results suggest that changes in trabecular architecture accompanying hypodontia may not be radiographically evident, and that functional and biological compensatory adaptations may occur in the bone tissue of these individuals.

In this study, differences were observed between the FD values of different ROIs on the same side of individuals. In both groups, FD values obtained from ROI 2 (mandibular angle) tended to be higher than those from ROI 1. However, this difference is more likely to reflect normal anatomical variation rather than condition-specific changes. Therefore, the clinical relevance of ROI 2 findings is limited, and they should be interpreted as secondary observations [28, 29]. Based on this information, ROI 2 (mandibular angle) tended to show higher FD values compared to the other regions analyzed. This difference is more likely related to normal anatomical variation and differences in functional loading rather than condition-specific changes. Çitir *et al*. [30] likewise found higher FD values in the angulus region than in the corpus area close to the teeth. In this context, the findings of our study appear to be consistent with the literature, although the clinical relevance of ROI 2 remains limited.

In our study, the mean age at the time of radiographs diagnosing hypodontia was 10.6 ± 1.7 years. According to Nolla's stages, the development of mandibular second premolars typically begins around the age of 3–4 years [6]. However, delayed mineralization of the second premolars may lead to false-positive diagnoses of agenesis on radiographs [3, 31], and the establishment of a definitive diagnosis of premolar agenesis is not recommended before the age of 7 years [7]. For this reason, the mean age of 10.6 ± 1.7 years in our group with no germ development was considered clinically reliable to distinguish between delayed formation and agenesis.

This study has several limitations. First, its retrospective cross-sectional design prevents longitudinal assessment of trabecular changes. Second, a healthy control group with normal premolar development was not included in this study. In healthy children, the mandibular second premolar germ typically appears around 4–5 years of age, at which point the furcation area is already occupied by developing teeth, making fractal analysis technically unfeasible. Moreover, fractal analysis has primarily been applied to edentulous areas, where trabecular patterns can be more reliably assessed. For these reasons, the present study was limited to comparative analysis between delayed germ formation and agenesis groups. Lastly, despite a priori power analysis, the relatively small sample size (n = 24 per group) may have limited the ability to detect subtle differences. Future prospective studies with larger samples are warranted.

5. Conclusions

In this study, fractal analysis was conducted during the early developmental stage, prior to the formation of mandibular second premolars. Thus, the study aimed to compare trabecular bone characteristics between children with delayed germ formation and those with agenesis. The findings suggest that fractal parameters did not differ significantly between the two groups. Consequently, our results show that fractal analysis of mandibular trabecular bone at this early developmental stage is not a practical therapeutic technique for distinguishing between agenesis and delayed germ formation.

AVAILABILITY OF DATA AND MATERIALS

The datasets generated and/or analysed during the current study are not publicly available due to lack of parental consent for data sharing, but are available from the corresponding author on reasonable request.

AUTHOR CONTRIBUTIONS

FS, SÖ—Conceptualization. FS, SD, SNU, ABÖ—Methodology. FS, FEM, SNU—Data Collection. FEM, FS, SNU—Data Analysis. FS—Writing-Original Draft. FS, SD, SÖ, ABÖ—Writing-Review & Editing. FEM, SD, FS—Supervision.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

This study was conducted in accordance with the Ataturk University Faculty of Medicine Ethics Committee approval (Approval date: 15 April 2025, Session no: 04/2025, Decision no: 41) and guidelines of the Declaration of Helsinki. Prior to the study, informed consents in writing were obtained from all parents.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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