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A micro- computed tomographic study of the anatomic danger zone in mesial roots of permanent mandibular first and second molars

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Abstract

Background To investigate the geometric characteristics of the danger zone in the mesial roots of mandibular molars using micro-computed tomography (micro-CT).

Methods A total of 75 extracted mandibular first (50 were 2-rooted [2RM1] and 25 were 3-rooted [3RM1]) and 35 2-rooted mandibular second molars (2RM2) were collected and evaluated using micro-CT. The morphological aspects of the mesial roots associated with the danger zone (the canal curvature, minimum mesial [MWT] and distal canal wall thickness [DWT], depth and level of root concavities) were evaluated. One-way analysis of variance (ANOVA) was used for multiple group comparisons. Student's *t*-test or paired *t*-test was used to test the means between two groups.

Results All mesial roots curved severely (81.8%, 90/110) or moderately (18.2%, 20/110) towards the furcation side, and the mean angle was 25.3 ± 7.2 degrees. The presence of a distolingual root only had limited influence on the geometricgeometry of the mesial root. In the majority of cases, the mean DWT was less than the MWT, and statistical significance (all $p < 0.05$) was detected at 0–3 mm (MB and ML of 3RM1), 0–4 mm (MB of 2RM1, and MB and ML of 2RM2), 0–5 mm (single mesial canals of mandibular first and second molars), and 0–6 mm (ML of 2RM1) below furcation. The mean depth of distal concavities is always greater (all $p < 0.05$) than the mesial ones at each root level. Generally, the mean depth of distal concavities increased apically in the cervical portion, reaching the maximum value at 2 mm below furcation, and then declined gradually in the apical portion.

Conclusions The mesial roots of mandibular first and second molars often exhibit severe distal curvature, with a mean Schneider's angle of 25.3 degrees, and the thinnest dentin wall is typically on the distal side. Distal root concavities are significantly deeper than mesial ones, with the maximum depth generally located 2 mm below the furcation. When identifying the danger zone, factors such as DWT should not be considered in isolation.

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Canal curvature, distal root concavities, and the type of instrument used are also critical in affecting the likelihood and location of strip perforation, though their precise roles warrant further investigations.

Keywords Mandibular molar, Micro-computed tomography, Mesial root, Danger zone, Root canal curvature, Root concavity

Background

The ideal preparation for a root canal involves creating a continuously tapering funnel from the orifice to the apex while preserving the original canal curvature. This approach facilitates effective irrigation and complete canal obturation [1]. However, during the instrumentation of curved canals, various procedural errors may arise, including ledging, zipping, canal transportation, and strip perforation, all of which can compromise the treatment outcomes [2]. Mandibular first and second molars are frequently affected by various types of lesions and are commonly treated with root canal therapy [3–5]. In addition to their complex root canal systems, the canal curvature in the mesial root presents another significant endodontic challenge. The mesial root is inherently curved towards the distal side [5–7], making it a common focus of studies on curved canal preparation [6, 8, 9]. Moreover, previous studies have shown that the mesial canals are not centrally located within the root but are positioned closer to the furcation side [10, 11]. Abou-Rass et al. [12] were the first to describe the thinner distal dentin wall as the "danger zone", which is particularly susceptible to over-instrumentation and strip perforation. They recommended using an anti-curvature filing technique to prepare these canals. Several other studies demonstrated that the danger zone was typically found 4–6 mm below the canal orifice [13], and the length of the teeth and the depth of root concavities may both affect the thickness of the danger zone [14]. Compared to shorter teeth, the danger zone was thinner, and the distal root concavity was also deeper in longer teeth [14]. Contrary to the general consensus, De-Deus et al. [15] demonstrated that the smallest dentine thickness could also be located on the mesial side of the roots in approximately 40% of the canals. Despite these insights, our understanding of the danger zone in mandibular molars remains limited and, at times, controversial. While it is recognized that multiple factors, including anatomical characteristics and parameters related to endodontic instruments employed, can affect the occurrence and location of strip perforations, few researchers have comprehensively examined these factors using reliable methods, such as micro-CT. Variations in methodologies and study designs contribute to the inconsistencies observed across different studies.

In recent decades, micro-computed tomography (micro-CT) has become a widely utilized tool in ex vivo studies of human teeth, owing to its high resolution and non-destructive nature [7, 16, 17].Unlike traditional methods such as sectioning, clearing, or radiography, micro-CT captures intricate three-dimensional (3D) internal structures and variations within roots while preserving sample integrity. This approach facilitates comprehensive quantitative and qualitative analysis without altering the specimen. Consequently, micro-CT offers significant advantages in examining the danger zones of tooth roots by providing sub-millimeter detail and enabling robust 3D reconstruction and geometric analysis.

This study aims to provide detailed geometric information about the danger zone in mandibular molars using micro-CT. To address the ongoing academic debate on danger zones, we focus on the curvature of the mesial root canals and their relationship with the thinner dentin walls adjacent to distal root concavities. Additionally, we conduct detailed disto-mesial comparisons concerning the canal wall thickness, as well as the depth and occurrence of root concavities.

Materials and methods

Collection of sample teeth

Approval for use of extracted teeth was obtained from the institutional Ethics Committee for Human Research (Issuing Number: KY2022-089–01). All subjects were native Chinese, and the teeth were extracted due to periodontal disease, non-restorable caries, trauma, or prosthodontic reasons. The tooth type (permanent mandibular first and second molar) was accurately identified and recorded immediately after tooth extraction. The exclusion criteria were as follows: (a) teeth with open root apices, (b) teeth with root canal calcification, (c) teeth that had been endodontically treated previously, (d) teeth with root carious, fractures, internal or external resorption, and (e) teeth with a C-shaped or fused root. The sample size was determined using G*Power software (ver. 3.1.9.7; Heinrich-Heine-Universität Düsseldorf, Düsseldorf, Germany). The analysis indicated that at least 27 samples per tooth group were required to achieve an effect size of 0.50 with 80% power and a 95% confidence interval. Due to the susceptibility of the distolingual (DL) root to fracture during extraction, obtaining a large, defect-free sample of extracted three-rooted mandibular first molars (3RM1) is particularly challenging. Consequently, this study included a total of 75 mandibular first molars, comprising 50 with two roots (2RM1) and 25 with three roots (3RM1), along with 35 two-rooted mandibular second molars (2RM2).

Micro-CT scanning of extracted teeth

Each tooth was scanned along its axis in a micro-CT device (SkyScan1174; Bruker-microCT, Kontich, Belgium) with a voxel size of 32 µm, set at 800 mA, 50 kVp, 180˚ of rotation with steps of 0.7˚. The frame average was set to 1, and a 1-mm thick aluminum filter was used. The micro-CT data files were then transferred to Mimics 21.0 (Materialise, Leuven, Belgium) software to perform 3D reconstruction of the teeth and root canal systems. The root canal systems in the mesial roots were classified according to Vertucci's methods [18].

Odontometric analysis

The volume of interest was selected from the pulp floor level to the anatomical apex of the mesial roots. The length of root trunk was assessed as the vertical distance from the orifice to the furcation level. To define the long axis of the mesial root cone, a spline curve was drawn by fitting a series of (8–10) geometric centroids of root sections from the furcation to the apex. The entire length of the spline was considered the length of the mesial root (cone). The mesial root was then resliced along the spline line at 1-mm intervals (Fig. 1). For each slice, the following measurements were taken (Fig. 2): (a) The measurements of mesial (MWT) and distal canal wall thickness (DWT) for the MB, ML, and single mesial canals were performed as described by Lim et al. [19], representing the minimum dentin thickness from the inner border of the canals to the external root surface on the mesial and distal side. (b) The depth of mesial/distal root concavity was measured as the distance between a line linking two points on the convex root surface and the deepest point of the root concavity [14]. It is essential to establish a threshold between a slight depression and a typical root concavity. We arbitrarily defined a root concavity as having a depth of at least ≥ 0.1 mm.

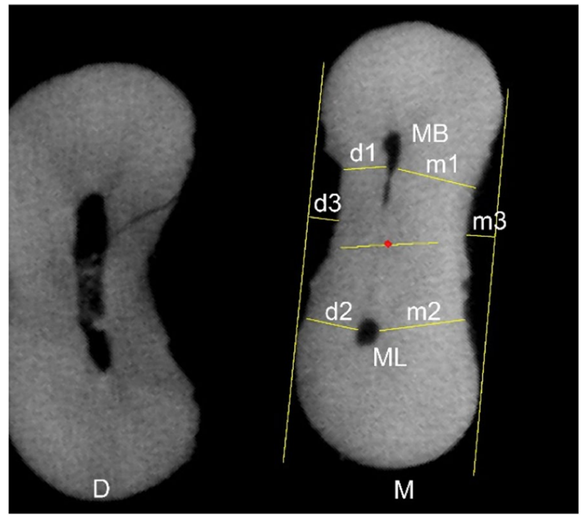

Fig. 2 Measurement of the minimum distal and mesial canal wall thickness of the MB (d1, m1) and ML canals (d2, m2), and the depth of distal and mesial root concavities (d3, m3) in a micro-CT cross-section

In the Mimics interface, the 3D tooth models were displayed in parallel projection mode [7]. The position of

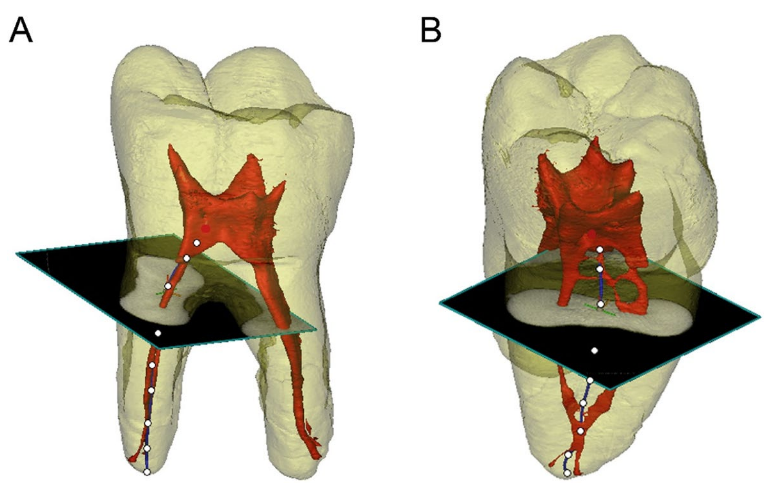

Fig. 1 Representative micro-CT images of a two-rooted mandibular first molar (the mesial root is resliced along the long axis of the root at 1-mm intervals). **A** A buccal view of the mesial root, the cross-section is perpendicular to the root axis, and the mesial canals are closer to furcation side at the danger zone; **B** A mesial view

the tooth model was further adjusted (by rotation) until the images of the MB and ML canals were completely overlapped (exactly in the buccal view of the mesial root) (Fig. 1A). Screenshots were saved in TIFF format with a resolution of 1280 × 770 pixels and then analyzed using Image-Pro Plus 6.0 (Media Cybernetics, Silver Spring, MD, USA). After calibration, the degrees of canal curvature in the mesial root were measured using Schneider's method [20]. In this method, a line is drawn parallel to the longitudinal axis of the coronal part of the canal, and a second line is drawn from the starting point of the canal curvature to the apical foreman. The acute angle formed by the intersection of these two lines is measured to evaluate canal curvature. Curvatures are categorized into three groups: straight (≤ 10 degrees), moderate (10–20 degrees), and severe (≥ 20 degrees). The canal length between the orifice to the furcation level was also measured (canal length in the root trunk).

Statistical analysis

All data analyses were performed using SPSS 21.0 (SPSS Inc., Chicago, IL, USA). One-way analysis of variance (ANOVA) followed by the post hoc Fisher's least significant difference test (LSD) test was used for multiple group comparisons. Student's *t*-test or paired *t*-test was applied to compare the means between two groups. The Chi-square test was used to compare frequencies. The significance level of 5% was set for all tests.

Results

Root canal configurations of the mesial roots of mandibular molars

The root canal configurations of the mesial roots were summarized in Table 1. Type 1–1 canal configurations occurred in only 8% of the first molars, while the percentage increased to 31.4% in the second molars. For multi-canal configurations, both MB and ML canals were identified and measurable at different root levels.

Table 1 Root canal configuration of mesial roots (Vertucci's classification) *n* (%)

Type of root canal configuration	3RM1	2RM1	2RM2
Type I (1–1)	2 (8.0%)	4 (8.0%)	11 (31.4%)
Type II (2–1)	2 (8.0%)	9 (18.0%)	10 (28.6%)
Type IV (2–2)	18 (72.0%)	32 (64.0%)	9 (25.7%)
Type V (1–2)	2 (8.0%)	3 (6.0%)	1 (2.9%)
Others	1[a] (4.0%)	2[b] (4.0%)	4[c] (11.4%)
Total	25 (100.0%)	50 (100.0%)	35 (100.0%)

3RM1 is 3-rooted mandibular first molar, 2RM1 and 2RM2 are 2-rooted mandibular first and second molar, respectively.

[a] one case of type 3–3 canal

[b] one case of type 2–1–2 and one case of type 1–2–1 canal

[c] 4 cases of type 1–2–1 canals

Root length and canal curvature of the mesial roots

The measurements of the root (cone) length and canal curvature are presented in Table 2 and Fig. 3. The mean mesial root length of the 2RM1s was significantly longer than that of the 2RM2s (9.7 mm vs. 8.9 mm, $p < 0.05$). However, no statistical differences were observed in the canal length at the root trunk or in the degrees of canal curvatures among the three tooth groups (all $p > 0.05$). The means for canal length and curvature were 2.5 ± 0.5 mm and 25.1 ± 6.8 degrees, respectively ($n = 110$). All mesial canals curved either severely (81.8%, 90/110) or moderately (18.2%, 20/110) towards the furcation side, with no instances of straight canals observed.

Minimum mesial and distal canal wall thickness

In this study, the canal wall thickness was statistically analyzed irrespective of Vertucci's classification. All root slices were aggregated and reclassified into non-single- and single-canal forms at each root level. The measurements of MWT and DWT for the MB, ML and single mesial canals at each level are summarized in Tables 3 and 4, and Fig. 4. In the majority of cases, DWT was found to be smaller than MWT, with statistically significant differences (all $p < 0.05$) observed at the following levels: 0–3 mm (MB and ML of 3RM1), 0–4 mm (MB of 2RM1, and MB and ML of 2RM2), 0–5 mm (single mesial canals of mandibular first and second molars), and 0–6 mm (ML of 2RM1) below the furcation.

The depth of mesial and distal root concavities

All mesial roots of mandibular molars, except one, exhibited distal concavities. In contrast, mesial concavities were observed in only 76.0% (57/75) of the first molars and 60.0% (21/35) of the second molars, with a statically significant difference between the sides (both $p < 0.01$). Figure 5 presents the mean depth of root concavities

Table 2 The measurement result of the root (cone) length and canal curvatures of the mesial roots of mandibular molars

Tooth group	*n*	Root length (mm)		Curvature (degrees)	
		Mean ± SD	Range	Mean ± SD	Range
3RM1	25	9.17 ± 1.40^{ab}	7.01 ~ 11.94	23.67 ± 5.37^{A}	11.36 ~ 37.04
2RM1	50	9.68 ± 1.31^{a}	7.05 ~ 12.42	25.17 ± 5.96^{A}	11.00 ~ 40.11
2RM2	35	8.85 ± 1.37^{b}	6.10 ~ 11.67	26.05 ± 8.65^{A}	12.20 ~ 49.4

3RM1 is 3-rooted mandibular first molar, 2RM1 and 2RM2 are 2-rooted mandibular first and second molar, respectively. The values with the different superscript lowercase (root length) or uppercase (curvature) letters along the same column are significantly different ($p < 0.05$). The curvatures were measured by Schneider method

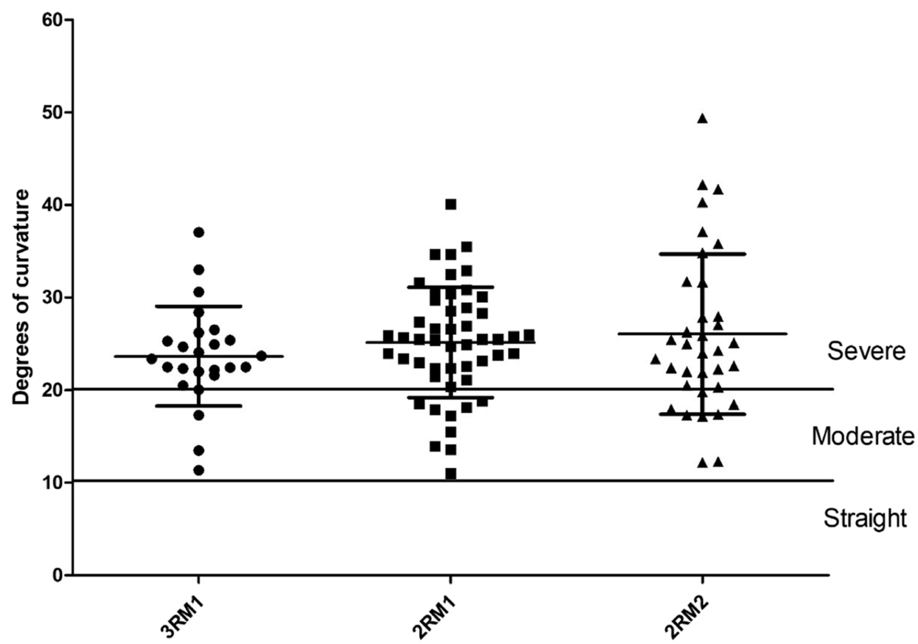

Fig. 3 Measurement results of canal curvatures in the mesial roots of mandibular molars (by Schneider method). No statistical difference was detected in canal curvature among the three tooth groups ($p > 0.05$). The error bar is SD

at each level. Generally, the mean depth of distal root concavities increased in the apical direction within the coronal third of the root, reaching a maximum at 2 mm below the furcation. Beyond this point, the depth gradually declining toward the apex. For mesial root concavities, the maximum depth was observed at 4 mm (3RM1), 3 mm (2RM1), and 2 mm (2RM2) below the furcation. The mean maximum depth of distal concavities was significantly greater (all $p < 0.05$) than that of mesial concavities at each level and for each tooth group. Figure 6 shows a representative 2RM1 with a root concavity located on the distal side of the mesial root, corresponding to the danger zone, while no root concavity was detected on the mesial side.

Discussion

The danger zone in the mesial roots of mandibular molars has garnered significant attention from researchers, as inadequate attention or excessive instrument in this area during root canal preparation can lead to strip perforation or increase the risk of root fractures [12–14]. Distal root concavities may cause an overestimation of distal canal wall thickness in conventional buccolingual radiographs [10, 21]. In contrast, in vivo cone beam computed tomography (CBCT) [22] and ex vivo micro-CT imaging [6] provide 3D visualization of the danger zone. Micro-CT, in particular, offers exceptionally high anatomical accuracy, enabling detailed visualization of internal root structures without damaging the specimen. This non-destructive method allows for thorough analyses of root canal systems and dentin thickness with submillimeter precision. Compared to traditional methods, micro-CT provides a more comprehensive and detailed mapping of anatomical features, enhancing the reliability and depth of the findings.

In this micro-CT study, the mesial root was digitally resliced along its long axis (Fig. 1) to measure canal wall thickness and depth of root concavities at different levels, providing a more reliable and accurate odontometric analysis. Previous studies, relying on tooth sections or micro-CT/CBCT scans, typically measured in a series of horizontal (axial) sections [6, 22, 23], which may overestimate the DWT and underestimate the MWT near the furcation level (Fig. 6). The presence of a 3RM1 is a significant racial characteristic in Mongolian populations, with prevalence rates ranging from 5 to 40%. In contrast, its frequency is typically below 5% in Black and White populations [6, 24]. Ethnicity also influences the presence of middle mesial (MM) canals in the mesial roots, with prevalence rates ranging from 1 to 23% and an overall prevalence of 7% observed across 15 countries worldwide [25].The MM canal is situated closer to the deepest point of the root concavities, and our previous study demonstrated that instrumentation of the MM canal with large diameter/taper files may result in thinner remaining canal wall thickness at the danger zone, potentially leading lead to root fragility [26].

Table 3 Measurement results of the mesial and distal minimum canal wall thickness of the MB and ML canals of mandibular molars at different root levels (mean ± SD, mm)

Tooth Type	Level (mm below FL)	*n*	MB canals			ML canals			
			MWT	DWT	DWT < 1 mm *n* (%)	*n*	MWT	DWT	DWT < 1 mm *n* (%)
3RM1	0 mm	21	1.63 ± 0.27^{A}	$1.16\pm0.21^{**a}$	4 (19.0%)	21	1.68 ± 0.27^{A}	$1.31\pm0.25^{**b}$	1 (4.8%)
	1 mm	22	1.31 ± 0.15^{A}	$1.02\pm0.19^{**a}$	10 (45.5%)	22	1.36 ± 0.16^{A}	$1.05\pm0.16^{**a}$	9 (40.9%)
	2 mm	21	1.16 ± 0.11^{A}	$0.93\pm0.22^{**a}$	13 (61.9%)	21	1.23 ± 0.14^{B}	$0.98\pm0.19^{**a}$	13 (61.9%)
	3 mm	20	1.04 ± 0.14^{A}	$0.90\pm0.17^{**a}$	16 (80.0%)	20	1.07 ± 0.13^{A}	$0.88\pm0.21^{**a}$	14 (70.0%)
	4 mm	20	0.92 ± 0.15^{A}	0.88 ± 0.12^{a}	16 (80.0%)	20	0.93 ± 0.18^{A}	0.85 ± 0.17^{a}	16 (80.0%)
	5 mm	16	0.86 ± 0.16^{A}	0.85 ± 0.12^{a}	15 (93.8%)	16	0.87 ± 0.13^{A}	0.86 ± 0.14^{a}	13 (81.3%)
	6 mm	6	0.88 ± 0.06^{A}	0.89 ± 0.16^{a}	4 (66.7%)	6	0.90 ± 0.13^{A}	0.82 ± 0.20^{a}	5 (83.3%)
	7 mm	6	0.84 ± 0.06^{A}	0.87 ± 0.16^{a}	5 (83.3%)	6	0.81 ± 0.13^{A}	0.83 ± 0.21^{a}	5 (83.3%)
	8 mm	3	0.87 ± 0.14^{A}	0.79 ± 0.04^{a}	3 (100.0%)	3	0.78 ± 0.07^{A}	0.79 ± 0.04^{a}	3 (100.0%)
2RM1	0 mm	44	1.66 ± 0.23^{A}	$1.26\pm0.23^{**a}$	5(11.4%)	44	1.74 ± 0.21^{A}	$1.42\pm0.18^{**b}$	0 (0.0%)
	1 mm	44	1.39 ± 0.16^{A}	$1.03\pm0.19^{**a}$	19 (43.2%)	44	1.52 ± 0.18^{B}	$1.11\pm0.19^{**b}$	10 (22.7%)
	2 mm	44	1.22 ± 0.14^{A}	$0.95\pm0.19^{**a}$	30 (68.2%)	44	1.31 ± 0.15^{B}	$1.01\pm0.19^{**a}$	19 (43.2%)
	3 mm	45	1.11 ± 0.16^{A}	$0.96\pm0.21^{**a}$	28 (62.2%)	45	1.20 ± 0.15^{B}	$0.94\pm0.20^{**a}$	28 (62.2%)
	4 mm	44	1.04 ± 0.14^{A}	$0.91\pm0.19^{**a}$	34 (77.3%)	44	1.09 ± 0.17^{B}	$0.91\pm0.21^{**a}$	31 (70.5%)
	5 mm	39	0.99 ± 0.15^{A}	0.95 ± 0.15^{a}	25 (64.1%)	39	1.03 ± 0.16^{A}	$0.92\pm0.26^{**a}$	29 (74.4%)
	6 mm	34	0.92 ± 0.15^{A}	0.95 ± 0.20^{a}	22(64.7%)	34	0.95 ± 0.20^{A}	$0.83\pm0.19^{**b}$	27 (79.4%)
	7 mm	22	0.86 ± 0.11^{A}	0.90 ± 0.16^{a}	16 (72.7%)	23	0.86 ± 0.18^{A}	0.79 ± 0.27^{a}	18 (78.3%)
	8 mm	16	0.71 ± 0.17^{A}	0.77 ± 0.13^{a}	15 (93.8%)	13	0.80 ± 0.20^{A}	$0.68\pm0.18^{*a}$	13 (100.0%)
	9 mm	12	0.71 ± 0.23^{A}	0.61 ± 0.16^{a}	12 (100.0%)	11	0.73 ± 0.23^{A}	0.57 ± 0.19^{a}	11 (100.0%)
	10 mm	7	0.62 ± 0.20^{A}	0.44 ± 0.17^{a}	7 (100.0%)	6	0.64 ± 0.15^{A}	0.46 ± 0.17^{a}	6 (100.0%)
	11 mm	2	0.60 ± 0.05^{A}	0.20 ± 0.08^{a}	2(100.0%)	1	0.43^{A}	0.16^{a}	1 (100.0%)
2RM2	0 mm	13	1.47 ± 0.22^{A}	$1.11\pm0.20^{**a}$	4 (30.8%)	13	1.65 ± 0.20^{A}	$1.23\pm0.15^{**a}$	0 (0.0%)
	1 mm	17	1.38 ± 0.24^{A}	$1.03\pm0.16^{**a}$	4 (23.5%)	17	1.45 ± 0.23^{A}	$1.07\pm0.18^{**a}$	5 (29.4%)
	2 mm	17	1.24 ± 0.13^{A}	$0.99\pm0.17^{**a}$	9 (52.9%)	17	1.29 ± 0.15^{A}	$0.95\pm0.20^{**a}$	9 (52.9%)
	3 mm	15	1.14 ± 0.18^{A}	$0.92\pm0.19^{**a}$	11 (73.3%)	15	1.17 ± 0.17^{A}	$0.88\pm0.1^{**a}$	12 (80.0%)
	4 mm	15	1.00 ± 0.13^{A}	$0.83\pm0.14^{**a}$	12 (80.0%)	15	1.06 ± 0.19^{A}	$0.84\pm0.17^{**a}$	13 (86.7%)
	5 mm	8	0.99 ± 0.24^{A}	0.85 ± 0.25^{a}	5 (62.5%)	8	1.03 ± 0.22^{A}	0.87 ± 0.3^{a}	7 (87.5%)
	6 mm	4	0.85 ± 0.12^{A}	1.02 ± 0.37^{a}	3 (75.0%)	4	1.01 ± 0.21^{A}	0.92 ± 0.2^{a}	3 (75.0%)
	7 mm	2	0.99 ± 0.09^{A}	0.92 ± 0.19^{a}	1 (50.0%)	2	0.89 ± 0.23^{A}	0.86 ± 0.1^{a}	2 (100.0%)

MB is mesio-buccal canal, ML is mesio-lingual canal, M-single is a single mesial canal, 3RM1 is 3-rooted mandibular first molar, 2RM1 and 2RM2 are 2-rooted mandibular first and second molar, respectively. MWT and DWT is the minimum mesial and distal wall thickness, respectively. *n* is number of slices. * means statistically significant difference between mesial and distal sides (paired *t*-test, $p < 0.05$), and ** means $p < 0.01$. The values with the different superscript uppercase (MWT) or lowercase (DWT) letters along the same line means statistically significant difference between MB and ML canals (paired *t*-test, $p < 0.05$)

Regarding the incidence of double/multiple canals, root length and canal curvature, no statistical difference was found between 2RM1s and 3RM1s (all $p > 0.05$) (Tables 1 and 2), suggesting that the presence of the DL root has limited impact on the anatomy and odontometric parameters of the mesial root. The mesial root of 2RM2s was approximately 1 mm shorter than that of the 2RM1s ($p < 0.05$), likely due to the smaller size of the second molars. The Schneider's angle of the mesial canals averaged 25.3 degrees, with the majority (81.8%) exhibiting severe curvatures (Fig. 3). This mean value aligns closely with the 24.9 degrees reported by Lim et al. [19] and 26.8 degrees by Jungman et al. [27], suggesting that canal curvature in mesial roots is intrinsic and consistent across different populations, necessitating anti-curvature measures during preparation of the mesial canals. Curvatures in the cervical and middle thirds of mesial roots are typically inherent and consistently directed towards the distal side, leading to excessive dentine removal on the inner curve wall (furcation side) during root canal instrumentation. In contrast, apical curvatures are more variable and unpredictable in both directions and severity, often resulting in complications such as ledge or elbow formation, canal zipping, and apical transportation.

Table 4 Measurement of the mesial and distal minimum canal wall thickness of the single canals in the mesial roots of mandibular molars at different root levels (mean ± SD, mm)

Level (mm below FL)	Mandibular first molars				Mandibular second molars			
	n	MWT	DWT	DWT < 1 mm *n* (%)	*n*	MWT	DWT	DWT < 1 mm *n* (%)
0 mm	10	1.55 ± 0.22	1.09 ± 0.28**	3 (30.0%)	22	1.68 ± 0.26	1.09 ± 0.25**	6 (27.3%)
1 mm	9	1.34 ± 0.18	0.88 ± 0.18**	7 (77.8%)	18	1.41 ± 0.15	0.94 ± 0.22**	13 (72.2%)
2 mm	10	1.16 ± 0.22	0.78 ± 0.22**	8 (80.0%)	18	1.23 ± 0.20	0.81 ± 0.20**	16 (88.9%)
3 mm	9	1.04 ± 0.25	0.73 ± 0.22**	8 (88.9%)	20	1.10 ± 0.19	0.81 ± 0.19**	17 (85.5%)
4 mm	10	0.94 ± 0.23	0.71 ± 0.24**	8(80.0%)	17	1.04 ± 0.16	0.75 ± 0.22**	14 (82.4%)
5 mm	13	0.96 ± 0.17	0.68 ± 0.24**	11 (84.6%)	17	0.97 ± 0.19	0.75 ± 0.18**	15 (88.2%)
6 mm	12	0.94 ± 0.21	0.80 ± 0.21	10 (83.3%)	12	0.94 ± 0.18	0.78 ± 0.21	11 (91.7%)
7 mm	9	0.85 ± 0.32	0.70 ± 0.20	9 (100.0%)	5	0.96 ± 0.17	0.60 ± 0.16*	5 (100.0%)
8 mm	6	0.82 ± 0.32	0.72 ± 0.29	6 (100.0%)	2	0.93 ± 0.21	0.64 ± 0.11	2 (100.0%)
9 mm	4	0.76 ± 0.17	0.70 ± 0.32	3 (75.0%)	-	-	-	-
10 mm	1	0.73	0.67	1 (100.0%)	-	-	-	-
11 mm	1	0.62	0.53	1 (100.0%)	-	-	-	-

MWT and DWT were the minimum mesial and distal wall thickness, respectively. *n* is number of slices. * means statistically significant difference between mesial and distal sides (paired *t*-test, $p < 0.05$) and ** means $p < 0.01$

Since the mean length of the mesial roots was 8.9, 9.2, or 9.7 mm across different tooth group (Table 2), the root portion from 0–3 mm below the furcation was classified as the cervical third, 4–6 mm below the furcation as the middle third, and the apical 0–3 mm or ≥ 7 mm below the furcation as the apical third of the root. Measurements of MWT and DWT indicated that, in the majority of cases, the DWT was smaller than the MWT, with statistically significant difference (all $p < 0.05$) observed in the coronal and/or middle root thirds [at 0–3 mm (MB and ML of 3RM1), 0–4 mm (MB of 2RM1, and MB and ML of 2RM2) and 0–5 mm (single mesial canals of mandibular first and second molars), and 0–6 mm (ML of 2RM1) below furcation]. As shown in Table 3 and Fig. 4, generally, both MWT and DWT decreased along the root length due to the tapering of the root. The mean DWT decreased to less than 1.0 mm at levels ≥ 2 mm (MB and ML canals in 3RM1 and 2RM2, MB canals of 2RM1), ≥ 3 mm (ML canals of 2RM1), or ≥ 1 mm (all single mesial canals) below the furcation, whereas the mean MWT decreased to less than 1.0 mm at levels ≥ 4 mm below the furcation. These data corroborate previous reports [12, 27–29], indicating that the canal in the mesial roots is not centrally located and, in most cases, is positioned closer to the distal side. However, our findings also suggest that the vertical location of the danger zone cannot be determined solely by the DWT value, as the smallest value consistently occurs at the apical side due to root tapering.

Figures 5 and 6 indicate that the distal root concavities were more pronounced than the mesial ones, as evidenced by the greater depth and higher occurrence rate of the distal concavities. The deepest point (button) of the distal concavity was, on average, located 2 mm below the furcation. Considering that the mean canal length in the root trunk (from the orifice to the furcation level) is 2.5 mm, the deepest point of the distal concavity is expected to be located, on average, 4.5 mm below the orifice. This finding is in consistent with the report by Estrela et al. [13], which identified the danger zone as being located 4–6 mm below the canal chamber orifice. Several other studies [20, 25] have arbitrarily defined the distal furcal root dentine, 2 mm below the furcation in the mesial roots of mandibular molars, as the danger zone, with average thickness ranging from 0.78 to 1.27 mm [22, 30–33]. In this study, the mean distal thickness at this level was 0.79–0.99 mm; and the mean depth of the distal root concavities was 0.56–0.63 mm (Fig. 5), which is less than the 0.7 mm reported by Bower [10], also lower than the 0.86–1.04 mm reported by Sauáia et al. [30]. The discrepancy in canal wall thickness may be attributed to differences in the ethnicity and age of the subjects, as well as variations in experimental protocols, definitions, and the determination of the danger zone. Sauáia et al. [30] also reported that the danger zone was associated with the root length, and the deepest concavities, along with the thinnest canal wall in the distal side of the mesial roots, were found in the longest roots. De-Deus et al. [15] analyzed 50 mesial roots of mandibular molars in a Brazilian population using micro-CT, which allowed for the generation of a full 3D mapping of canal wall thickness for each root. Their study demonstrated

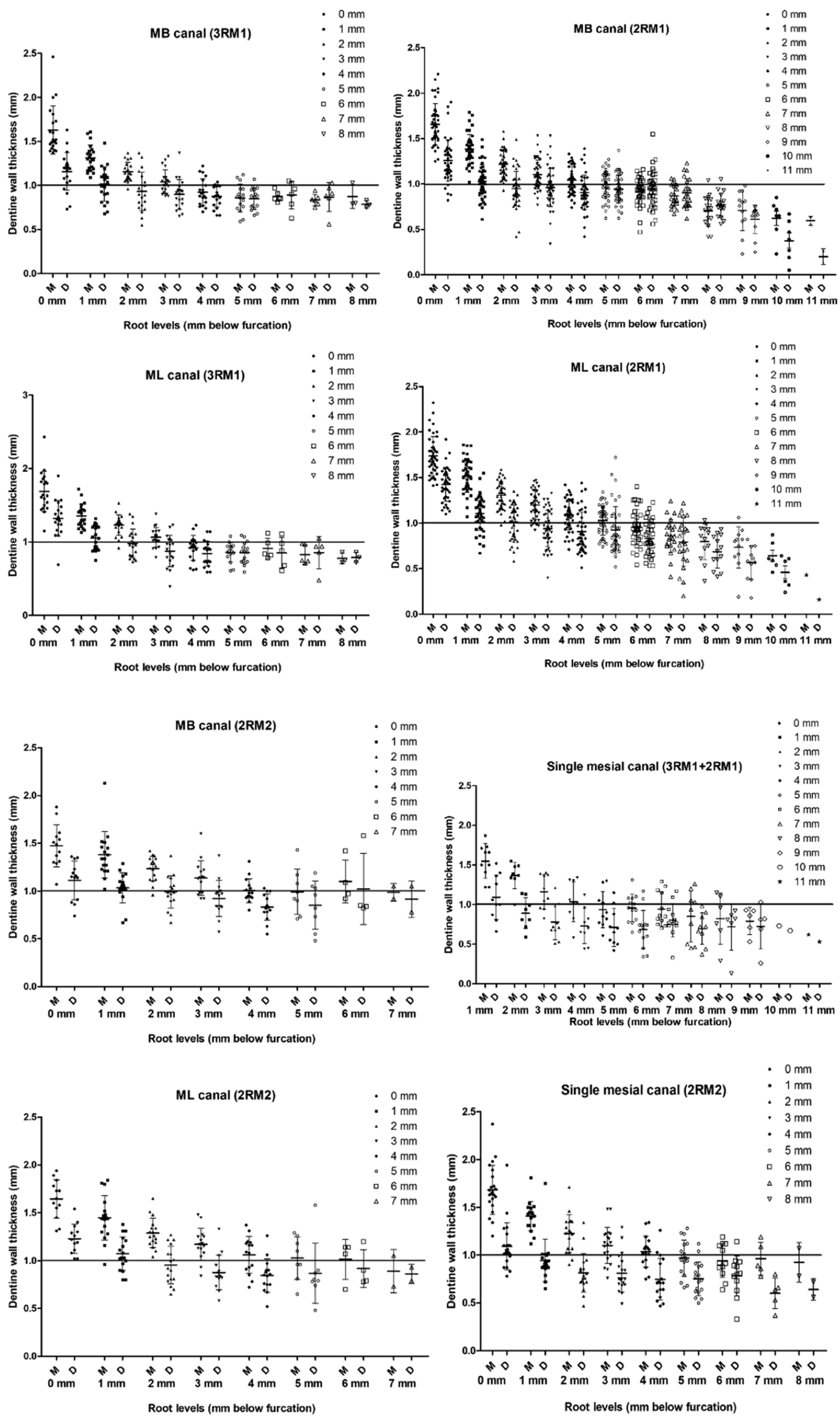

Fig. 4 Distribution plots of mesial and distal canal wall thickness of mesial roots of mandibular molars at different root levels. The error bar is SD

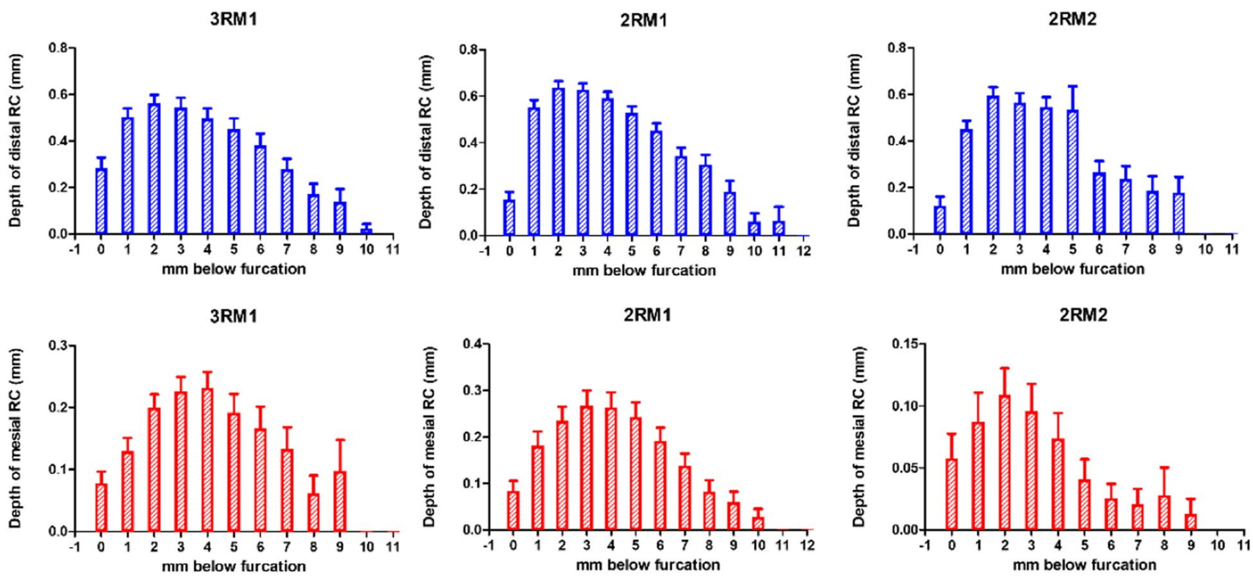

Fig. 5 Measurement results of the depth of distal and mesial root concavities in the mesial roots of mandibular molars at different levels. 3RM1 is three-rooted mandibular first molar; 2RM1 is two-rooted mandibular first molar; 2RM2 is two-rooted mandibular second molars; RC is root concavity. The error bar is SE

that the thinnest dentine was located on the mesial side in approximately 40% of the canals, while the overall vertical location of the danger zone was found to be at the middle third of the root (4.37 ± 1.68 mm below the furcation). However, even in slices where the minimum canal wall thickness was on the mesial side, it remains true that the mesial area is on the outer side of the curvature and is often minimally instrumented or left unprepared by endodontic instruments. In contrast, the distal wall, located on the inner side of the curvature, is consistently subjected to maximum dentin removal. Therefore, in our opinion, the danger zone is always located on the distal aspect (furcation side). Based on CBCT images, Bolbolian et al. [34] measured the dentin thickness and depth of distal concavity of the mesial roots from the furcation to 5 mm below. The area with the greatest concavity depth was used to calculate the minimum dentin thickness. The danger zone was most observed in the range of 0 to 1 mm below the furcation, and was seen in the range of 0 to 3 mm below the furcation with a probability of 93.4% [34].

Coronal flaring can help eliminate cervical interferences during the preparation of the apical third of the root canal. To prevent strip perforations at the danger zone, coronal flaring should be limited and directed towards the mesial and thicker aspects of the canal walls [12]. However, formal evidence is still lacking regarding the minimum cervical enlargement necessary for effective irrigation and apical filling [11]. To avoid perforation or vertical root fracture, Lim et al. [19] proposed an arbitrary minimum canal wall thickness of 0.3 mm after instrumentation. In recent decades, several novel nickel-titanium (NiTi) instruments have been developed to improve root canal shaping, with changes in geometrics, alloy composition, and kinetics. When addressing the danger zone in the mesial roots of mandibular molars, clinicians must consider multiple factors, particularly the distribution of canal curvatures and the locations of root concavities, in selecting appropriate instruments. The ideal enlargement of the root canal space should be based on preoperative anatomical dimensions [35, 36]. Theoretically, selecting instruments with small tapers and small sizes, especially for coronal flaring or master apical files, may offer a safer approach. Keles et al. [37] conducted ex vivo preparation experiments on the mesial roots of mandibular molars with a MM canal using ProTaper Next (PTN) X2 (#25,0.06) and X3 (#30,0.07) instruments (Dentsply Sirona, USA). The study found that the pre- and postoperative dentine thickness of the MM canal walls, in both mesial and distal aspects of the root, were significantly thinner than those of MB and ML canals; files with small tapers should be preferred for preparing mesial canals in mandibular molars [37]. Another study [38] compared the shaping ability of the XP-endo Shaper (XPS) (FKG Dentaire SA, La Chaux-de-Fonds, Switzerland) and PTN systems (both with snake-like movements and final sizes of #30) in double-canaled mandibular premolars with radicular grooves. The thin area in the canal wall of the radicular groove was also

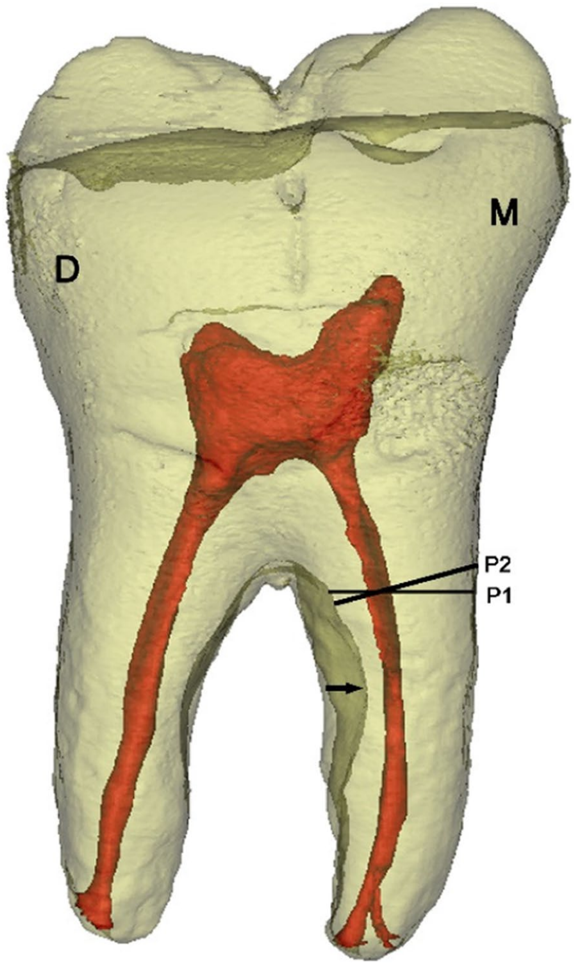

Fig. 6 A deep root concavity is located at the distal side of the mesial root of a mandibular first molar, which corresponds to the minimum canal wall thickness (arrow indicates the bottom of the root concavity). P1 indicates the axial plane, and P2 indicates the transverse plane of the mesial root which is perpendicular to the root canal

considered a danger zone. The study found that canal preparation using XPS was more conservative compared to PTN, likely due to its unique geometric design (a smaller mass and adaptive core, a fixed taper of 0.07 from D1 to D3, followed by a decreasing tapered design) and alloy type. The XPS' unique alloy properties allow it to change its cross-sectional shape during rotation, better adapting to the natural morphology of the root canal. This adaptability helps reduce the risk of canal deviation and strip perforation.

In summary, there is no consensus on the exact location of the danger zone, as the concept is largely empirical and based on clinical experience [12]. Current data suggest that canal wall thickness should not be considered in isolation when determining the danger zone. It must be evaluated alongside other factors, such as the distribution and severity of canal curvature, root concavity, and the parameters of the instruments used, as these factors may influence the probability and site of strip perforation. Theoretically, the more severe and more coronally located the curvature, the greater the internal bending stress experienced by the instrument. This stress results in increased dentin removal from the inner wall of the canal curvature (furcation side) during instrumentation, particularly at the coronal levels. This trend is more pronounced with longer roots and when using instruments with larger tapers or sizes. Conversely, dentin wall thickness generally increases coronally due to root tapering, which can counteract these factors and lead to a reverse trend towards the apical side in determining potential strip perforation sites. The deepest point of the distal root concavity is typically located 2 mm below the furcation, complicating the prediction of perforation sites. Further exploration, through either ex vivo laboratory experiments or in vivo clinical studies, of how these factors interact and influence outcomes would enhance the analysis and provide a more comprehensive understanding. In general, utilizing more flexible NiTi instruments with smaller tapers and sizes is an effective clinical strategy for minimizing the removal of peri-cervical dentin and avoiding strip perforation. Clinicians must balance the thorough removal of infected dentin and the preservation of healthy tooth structure, paying particular attention to the proper management of the danger zone.

In terms of the strengths of this study, the use of micro-CT technology stands out as a significant advantage, providing high anatomical accuracy. This advanced imaging technique enabled a more detailed analysis than previous studies have achieved. Additionally, the study's comprehensive anatomical mapping and its consistency with existing literature further underscore its robustness and reliability. However, this study also has several limitations. First, all the sample teeth were collected from a Chinese population, and since ethnicity may influence tooth dimension and root/canal variation [10], further studies involving other ethnic populations are necessary to verify the generalizability of our conclusions. Second, the current study used extracted teeth, and potential influences of age and gender were not considered, which may introduce bias into the findings. Future in vivo CBCT studies with a large sample size could address this limitation. Finally, the analysis of canal curvatures was primarily conducted in the bucco-lingual view. In the mesial root, the root canal configuration may exhibit greater variability and unpredictability in the proximal view. Although this issue was not the focus of the current study, it is also a significant factor contributing to iatrogenic errors and warrants further investigation.

Conclusions

The mesial roots of mandibular first and second molars frequently exhibit severe curvature towards the distal side, with a mean Schneider's angle of 25.3 degrees, and the thinnest dentin wall is typically on the distal aspect. Distal root concavities are significantly deeper than the mesial ones, with the deepest point generally located 2 mm below the furcation. In determining the location of the danger zone, factors such as DWT should not be considered in isolation. Canal curvature, distal root concavities, and the type of instrument used are also critical factors influencing the likelihood and location of strip perforation, though their precise roles warrant further investigations.

Abbreviations

2RM1	Two-rooted mandibular first molar
2RM2	Two-rooted mandibular second molar
3RM1	Three-rooted mandibular first molar
3D	Three-dimensional
ANOVA	One-way analysis of variance
DL	Distolingual
DWT	Distal wall thickness
LSD test	Fisher's least significant difference test
micro-CT	Micro-computed tomography
MWT	Mesial wall thickness
MB	Mesiobuccal
ML	Mesiolingual
MM canal	Middle mesial canal
nickel-titanium	NiTi

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Authors' contributions

Y.G. contributed to the study's conception and design. Data collection was performed by Y.T., Y.Q., P.Z., L.W. and J.F.; J.F.,W.Z. and J.L. contributed to the analysis or interpretation of data. Statistical analysis was performed by Y.T. and Y.G. The main manuscript text was written by Y.T. and Y.Q. All authors reviewed the manuscript.

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Data availability

All the datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

The human mandibular first and second molars used in this study were obtained from the Department of Dentistry, Ninth People's Hospital of Suzhou. All methods have been performed in accordance with the Declaration of Helsinki and have been approved by the Ethics Committee of Ninth People's Hospital of Suzhou with the approval number # KY2022-089–01. Informed consent was obtained from all subjects and/or their legal guardian(s).

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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