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Comparative Evaluation of Root-End Preparation and Filling Techniques Used in Endodontic Microsurgery and Conventional Periapical Surgery: An In-Vitro Study

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ABSTRACT: Background:

Successful endodontic microsurgery depends on effective root-end cavity preparation and adequate adaptation of retrograde filling materials to achieve an optimal apical seal. The instrumentation technique and filling material may influence apical leakage, marginal adaptation, dentinal crack formation and the internal integrity of the restoration.

Aim:

To comparatively evaluate the influence of conventional bur and ultrasonic-assisted root-end cavity preparation techniques in combination with mineral trioxide aggregate (MTA) and Biodentine on apical sealing ability, marginal adaptation, dentinal crack formation and filling-material voids under in-vitro conditions.

Materials and Methods:

This in-vitro experimental study was conducted at a tertiary care centre from January to March 2026 and included 60 specimens divided equally into four experimental groups (n = 15). Group I received conventional bur preparation with MTA, Group II underwent conventional bur preparation with Biodentine, Group III received ultrasonic-assisted preparation with MTA, and Group IV underwent ultrasonic-assisted preparation with Biodentine. Apical dye penetration was measured in millimeters (mm), while marginal gaps were assessed in micrometres (μm). Dentinal crack formation and filling-material voids were recorded as present or absent. Continuous variables were analyzed using one-way analysis of variance (ANOVA), followed by Tukey's honestly significant difference (HSD) post-hoc test. Categorical variables were compared using the chi-square test.

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Statistical significance was established at probability (p) < 0.05.

Results:

The highest mean apical dye penetration was observed in the conventional bur + MTA group (2.84 ± 0.71 mm), whereas the ultrasonic + Biodentine group demonstrated the lowest value (1.39 ± 0.49 mm). Intergroup differences were statistically significant (F = 18.394, p < 0.001). Similarly, marginal gap measurements were greatest in the conventional bur + MTA group (1.92 ± 0.48 μm) and lowest in the ultrasonic + Biodentine group (0.86 ± 0.29 μm), with significant intergroup variation (F = 22.649, p < 0.001). Dentinal cracks were identified in 12 specimens (20.0%), while filling-material voids were detected in 10 specimens (16.7%). Neither dentinal crack formation (χ^2 = 4.167, p = 0.244) nor void occurrence (χ^2 = 2.400, p = 0.494) demonstrated a statistically significant association with experimental group allocation. Although the ultrasonic groups exhibited numerically fewer cracks and voids, these differences were not statistically significant.

Conclusion:

Ultrasonic-assisted root-end cavity preparation demonstrated significantly lower apical dye penetration and marginal gap measurements than conventional bur preparation, suggesting improved apical sealing and marginal adaptation. The ultrasonic + Biodentine combination exhibited the most favourable numerical outcomes across the evaluated parameters. However, differences in dentinal crack formation and filling-material void occurrence were not statistically significant. Further investigations involving larger sample sizes and advanced microscopic assessment are warranted to establish the clinical relevance of these findings.

INTRODUCTION

The primary objective of endodontic treatment is to eliminate microbial infection from the root canal system and establish a predictable apical seal to prevent reinfection of the periapical tissues [1]. Although conventional root canal treatment achieves favourable outcomes in most cases, certain teeth may continue to exhibit persistent periapical inflammation despite technically adequate endodontic procedures. Such conditions may arise due to complex root canal anatomy, inaccessible apical ramifications, persistent intraradicular microorganisms, extraradicular infection, or procedural complications [2]. When nonsurgical endodontic retreatment is considered unsuitable or fails to resolve the disease process, surgical endodontic intervention becomes an important treatment option. Periapical surgery aims to eliminate the pathological periapical tissue, manage the apical portion of the root, and establish an effective barrier against the passage of microorganisms and their by-products into the surrounding periapical tissues [3].

Conventional periapical surgery traditionally involves surgical access to the root apex, curettage of periapical pathological tissue, apical root resection, and preparation of a root-end cavity followed by placement of a retrograde filling material. The effectiveness of this procedure depends considerably on the quality of root-end preparation and the ability of the retrograde material to establish an adequate apical seal [4]. Conventional root-end preparation has commonly been performed using rotary instruments such as small round burs. However, these instruments may produce shallow or irregular root-end cavities and can increase the possibility of dentinal microcracks, excessive removal of root structure, and inadequate adaptation of the filling material. Furthermore, restricted surgical access and difficulty in maintaining the appropriate preparation angle may compromise the quality of the apical cavity [5].

Endodontic microsurgery has introduced significant improvements in the management of persistent periapical lesions through the application of magnification, illumination, ultrasonic root-end preparation, and contemporary biocompatible filling materials. The use of surgical operating microscopes and magnifying devices facilitates accurate identification of anatomical structures, improved visualization of the resected root surface, and more precise root-end preparation. Ultrasonic instruments allow the preparation of smaller and deeper root-end cavities along the long axis of the root, thereby preserving

additional radicular dentin and facilitating the placement of retrograde filling materials. These refinements are intended to improve the marginal adaptation of root-end fillings and reduce the possibility of apical leakage [6].

Root-end preparation represents a critical stage in periapical surgery because its configuration directly influences the adaptation and sealing performance of retrograde filling materials. Different preparation techniques, including conventional rotary bur preparation and ultrasonic preparation, may produce variations in cavity depth, marginal integrity, surface characteristics, and the occurrence of dentinal defects. Rotary burs are relatively straightforward to use but may create bevelled preparations and require greater removal of root structure. In comparison, ultrasonic tips facilitate more conservative and centrally aligned preparations, although excessive ultrasonic power or prolonged instrumentation may contribute to the development of dentinal microcracks. Therefore, evaluating the structural characteristics of root-end cavities prepared using different techniques is important for determining their suitability in surgical endodontic procedures [7].

The selection of an appropriate root-end filling material is equally important for achieving a predictable surgical outcome. An ideal retrograde filling material should demonstrate excellent sealing ability, dimensional stability, biocompatibility, resistance to moisture, adequate handling characteristics, and the ability to promote favourable tissue responses. Mineral trioxide aggregate (MTA) has been widely investigated because of its favourable biological properties, ability to set in the presence of moisture, and potential to support periapical tissue healing. However, certain limitations, including prolonged setting time, handling difficulties, and potential discoloration, have encouraged the development of alternative calcium silicate-based materials [8].

Biodentine and bioceramic root repair materials have gained considerable attention in contemporary endodontics because of their favourable physicochemical and biological characteristics. These materials demonstrate calcium ion release, bioactivity, and the potential to promote mineralized tissue formation. Their interaction with dentinal tissues may contribute to improved marginal adaptation and sealing performance. However, differences in material composition, consistency, setting characteristics, and interaction with the prepared root-end cavity may influence their overall performance. Consequently, comparative laboratory investigations are necessary to determine how different retrograde filling materials perform when placed in root-end cavities prepared using different instrumentation techniques.

An important consideration in evaluating root-end filling procedures is their ability to prevent apical microleakage. Microleakage may occur through gaps at the material–dentin interface, inadequate cavity preparation, material shrinkage, or incomplete adaptation of the filling material. Such leakage may permit the movement of bacteria, bacterial products, and tissue fluids between the root canal system and periapical tissues, potentially contributing to persistent inflammation or treatment failure. Dye penetration, fluid filtration, bacterial leakage, and microscopic examination are among the laboratory methods used to evaluate the sealing ability and marginal adaptation of root-end filling materials. These techniques provide valuable information regarding the influence of preparation methods and material selection on the quality of the apical seal [9].

Although endodontic microsurgery incorporates several technical refinements over conventional periapical surgery, the relative contribution of root-end preparation techniques and filling materials to the overall sealing performance requires further investigation. Differences in cavity geometry, dentinal surface characteristics, microcrack formation, and material adaptation may influence the effectiveness of the final retrograde restoration. An in-vitro experimental approach offers the opportunity to standardize specimen preparation, instrumentation procedures, material placement, and evaluation conditions, thereby facilitating a controlled comparison of different surgical techniques.

Therefore, the present in-vitro study was designed to comparatively evaluate root-end preparation and filling techniques used in endodontic microsurgery and conventional periapical surgery. The investigation focuses on assessing differences in root-end cavity characteristics, marginal adaptation, and sealing ability associated with the respective preparation and filling approaches. The findings may provide useful laboratory evidence regarding the influence of instrumentation techniques and retrograde filling materials on the quality of root-end restoration and may contribute to the selection of appropriate techniques for surgical endodontic management.

MATERIALS & METHODS

Study Design

The present investigation was designed as an in-vitro experimental study conducted at a tertiary care centre from January to March 2026 to compare the influence of different root-end cavity preparation techniques and retrograde filling materials on apical sealing ability, marginal adaptation, dentinal crack formation and filling-material voids. Two root-end preparation approaches, namely conventional bur instrumentation and ultrasonic-assisted preparation, were evaluated in combination with two retrograde filling materials, MTA and Biodentine. A total of 60 specimens were included and equally distributed among four experimental groups, with 15 specimens allocated to each group.

Group Allocation

The specimens were randomly or systematically allocated into the following four groups (allocation method to be confirmed):

- **Group I (n = 15):** Root-end cavity preparation using conventional burs, followed by retrograde filling with MTA
- **Group II (n = 15):** Root-end cavity preparation using conventional burs, followed by retrograde filling with Biodentine
- **Group III (n = 15):** Root-end cavity preparation using ultrasonic tips, followed by retrograde filling with MTA
- **Group IV (n = 15):** Root-end cavity preparation using ultrasonic tips, followed by retrograde filling with Biodentine

This four-group experimental arrangement enabled the comparative evaluation of preparation techniques and filling materials, both independently and in combination.

Root-End Cavity Preparation

Root-end cavity preparation was performed using two different instrumentation approaches. In Groups I and II, conventional rotary burs (Mani Inc., Tochigi, Japan) were used to prepare the root-end cavities. In Groups III and IV, ultrasonic-assisted cavity preparation was performed using an ultrasonic unit (Woodpecker, Guilin, China) equipped with appropriate endodontic ultrasonic tips. The root-end cavities were prepared according to the standardized laboratory protocol, maintaining consistency in cavity dimensions and preparation depth across the respective groups. Following cavity preparation, the specimens were subjected to retrograde filling using the assigned materials, namely MTA or Biodentine.

Retrograde Filling Procedures

Two calcium silicate-based root-end filling materials were evaluated: MTA and Biodentine. MTA was used in Groups I and III, whereas Biodentine was used in Groups II and IV. Following root-end cavity preparation, the respective materials were placed into the prepared cavities according to the manufacturer's recommendations and the standardized laboratory protocol. Details regarding material mixing, placement instruments, condensation procedures and setting conditions should be incorporated from the original experimental records.

Assessment of Apical Dye Penetration

Apical dye penetration was evaluated to compare the sealing performance of the four experimental groups. The extent of dye penetration was recorded in millimeters (mm) and used as an indicator of apical leakage. The measurements were tabulated separately for each group and expressed as mean and standard deviation (SD). Lower dye penetration measurements indicated comparatively better apical sealing performance. The specific dye employed, concentration, immersion duration, specimen coating procedure, sectioning method and measurement technique should be added according to the actual experimental protocol.

Evaluation of Marginal Gap

Marginal adaptation of the retrograde filling materials was assessed by measuring the gap between the root-end filling material and the surrounding dentinal walls. The measurements were recorded in micrometres (μm) for all specimens. The mean marginal gap and standard deviation were calculated for each experimental group. Smaller marginal gap measurements were considered indicative of closer adaptation of the filling material to the prepared cavity walls. The imaging system,

magnification level, number of measurement sites, and microscopic assessment procedure should be described in accordance with the actual laboratory protocol.

Assessment of Dentinal Crack Formation

All specimens were examined for the presence or absence of dentinal cracks following the experimental procedures. Crack occurrence was recorded separately for each group. The number of specimens exhibiting dentinal cracks and those without detectable cracks was documented. The results were expressed as absolute frequencies and percentages to facilitate comparisons between conventional bur and ultrasonic preparation techniques. The microscopic examination method and criteria used to identify dentinal cracks should be detailed based on the actual assessment protocol.

Evaluation of Filling-Material Voids

The specimens were also evaluated for the presence of voids within the retrograde filling materials. Each specimen was classified according to whether voids were present or absent. The frequency and percentage of specimens demonstrating filling-material voids were calculated for all four experimental groups. This assessment was performed to compare the quality and internal homogeneity of the retrograde fillings obtained using the two preparation techniques and filling materials. The imaging and examination criteria used to identify filling-material voids should be specified according to the actual laboratory protocol.

Outcome Measures

The following parameters were considered for comparative evaluation:

1. **Apical dye penetration:** Measured in mm to assess the apical sealing ability of the retrograde filling
2. **Marginal gap:** Measured in micrometres (μm) to evaluate the adaptation of the filling material to the cavity walls
3. **Dentinal crack formation:** Recorded as present or absent for each specimen
4. **Filling-material voids:** Documented as present or absent to evaluate the internal quality of the retrograde restoration

Statistical Analysis

The collected data were compiled, tabulated and statistically analyzed using International Business Machines (IBM) Statistical Package for the Social Sciences (SPSS) Statistics for Windows, Version 29.0 (IBM Corp., Armonk, NY, USA). Continuous variables, including apical dye penetration and marginal gap measurements, were expressed as mean ± SD. Intergroup comparisons were performed using one-way analysis of variance (ANOVA). Tukey's honestly significant difference (HSD) post-hoc test was subsequently applied for pairwise comparisons to identify differences between individual experimental groups. Categorical variables, including dentinal crack formation and filling-material void occurrence, were presented as frequencies and percentages. Comparisons among the four experimental groups were performed using the chi-square test. A probability (p)-value of less than 0.05 was considered indicative of statistical significance for all analyses.

RESULTS

Table 1 & Figure 1 demonstrate the allocation of the 60 specimens into four experimental groups, comprising 15 specimens per group. Group I involved root-end cavity preparation using conventional burs, followed by MTA as the retrograde filling material. In Group II, conventional bur instrumentation was performed, and the prepared cavities were filled with Biodentine. Group III underwent ultrasonic-assisted root-end cavity preparation, followed by MTA placement, whereas Group IV received ultrasonic root-end preparation with Biodentine as the filling material. This grouping facilitated the comparative assessment of the two root-end preparation techniques in combination with the two retrograde filling materials.

Table 1: Group allocation

Group	Root-end preparation	Root-end filling	Sample size
I	Conventional bur	MTA	15

II	Conventional bur	Biodentine	15
III	Ultrasonic tip	MTA	15
IV	Ultrasonic tip	Biodentine	15

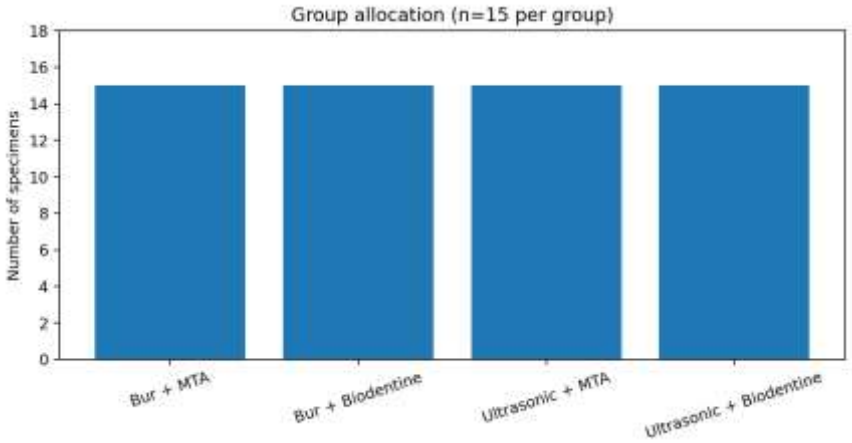

Figure 1: Distribution of specimens among the four experimental groups according to root-end preparation technique and retrograde filling material

Table 2 & Figure 2 demonstrate the intergroup comparison of apical dye penetration among the four experimental groups. The conventional bur + MTA group exhibited the highest mean dye penetration (2.84 ± 0.71 mm), followed by the conventional bur + Biodentine group (2.51 ± 0.66 mm), ultrasonic + MTA group (1.72 ± 0.55 mm), and ultrasonic + Biodentine group (1.39 ± 0.49 mm). Table 3 presents ANOVA revealed a statistically significant difference among the four groups ($F = 18.394$, $p < 0.001$).

Table 4 presents Tukey HSD post-hoc analysis demonstrated significantly lower dye penetration in both ultrasonic preparation groups compared with their corresponding conventional bur groups. However, no statistically significant difference was observed between MTA and Biodentine when used with the same preparation technique ($p = 0.4535$). Overall, the findings indicate that ultrasonic root-end preparation resulted in reduced apical dye penetration compared with conventional bur preparation, irrespective of the filling material used.

Table 2: Intergroup comparison of apical dye penetration

Experimental group	**n**	**Mean (mm)**	**SD**
Conventional bur + MTA	15	2.84	0.71
Conventional bur + Biodentine	15	2.51	0.66
Ultrasonic + MTA	15	1.72	0.55
Ultrasonic + Biodentine	15	1.39	0.49

Table 3: One-way ANOVA for apical dye penetration

Source	**SS**	**df**	**MS**	**F**	**p-value**
Between groups	20.4495	3	6.8165	18.394	<0.001
Within groups	20.7522	56	0.3706		
Total	41.2017	59			

Table 4: Tukey HSD post-hoc comparisons for apical dye penetration

Pairwise comparison	Mean difference (mm)	95% CI	P value	Inference
Conventional bur + MTA vs Conventional bur + Biodentine	0.33	-0.26 to 0.92	0.4535	Not significant
Conventional bur + MTA vs Ultrasonic + MTA	1.12	0.53 to 1.71	<0.0001	Significant
Conventional bur + MTA vs Ultrasonic + Biodentine	1.45	0.86 to 2.04	<0.0001	Significant
Conventional bur + Biodentine vs Ultrasonic + MTA	0.79	0.20 to 1.38	0.0042	Significant
Conventional bur + Biodentine vs Ultrasonic + Biodentine	1.12	0.53 to 1.71	<0.0001	Significant
Ultrasonic + MTA vs Ultrasonic + Biodentine	0.33	-0.26 to 0.92	0.4535	Not significant

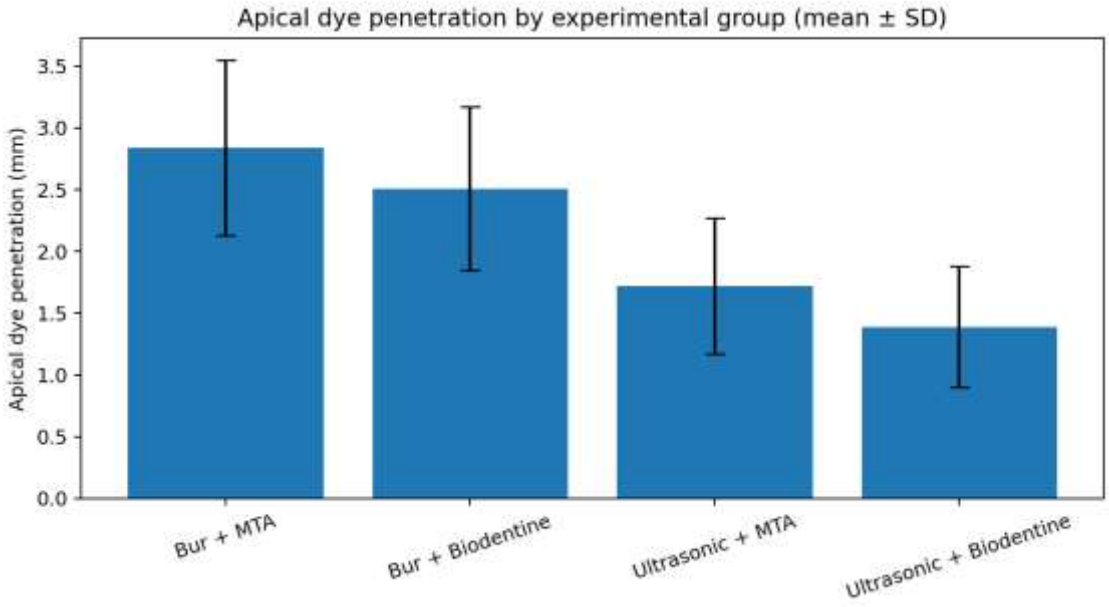

Figure 2: Comparative mean apical dye penetration among the four experimental groups (mm)

Tables 5 & Figure 3 illustrate the comparison of marginal gap measurements across the four experimental groups. The conventional bur + MTA group recorded the highest mean marginal gap (1.92 ± 0.48 μm), followed by the conventional bur + Biodentine group (1.61 ± 0.43 μm) and the ultrasonic + MTA group (1.08 ± 0.35 μm). The lowest mean marginal gap was observed in the ultrasonic + Biodentine group (0.86 ± 0.29 μm).

Table 6 presents one-way ANOVA demonstrated a statistically significant difference in marginal gap measurements among the four groups ($F = 22.649$, $p < 0.001$).Table 7 presents Tukey HSD post-hoc analysis revealed significantly smaller marginal gaps in both ultrasonic preparation groups compared with their corresponding conventional bur groups. A significant difference was also identified between the conventional bur + MTA and ultrasonic + Biodentine groups (mean difference = 1.06 μm, $p < 0.001$). However, comparisons between MTA and Biodentine within the same preparation technique did not demonstrate statistically significant differences (conventional bur: $p = 0.1492$; ultrasonic: $p = 0.4279$).

Overall, the findings indicate that ultrasonic root-end cavity preparation was associated with better marginal adaptation, as evidenced by reduced marginal gap measurements, compared with conventional bur preparation, irrespective of the retrograde filling material employed.

Table 5: Intergroup comparison of marginal gap

Experimental group	n	Mean (µm)	SD
Conventional bur + MTA	15	1.92	0.48
Conventional bur + Biodentine	15	1.61	0.43
Ultrasonic + MTA	15	1.08	0.35
Ultrasonic + Biodentine	15	0.86	0.29

Table 6: One-way ANOVA for marginal gap

Source	SS	df	MS	F	p-value
Between groups	10.5641	3	3.5214	22.649	<0.001
Within groups	8.7066	56	0.1555		
Total	19.2707	59			

Table 7: Tukey HSD post-hoc comparisons for marginal gap

Pairwise comparison	Mean difference (µm)	95% CI	Adjusted p	Inference
Conventional bur + MTA vs Conventional bur + Biodentine	0.31	-0.07 to 0.69	0.1492	Not significant
Conventional bur + MTA vs Ultrasonic + MTA	0.84	0.46 to 1.22	<0.0001	Significant
Conventional bur + MTA vs Ultrasonic + Biodentine	1.06	0.68 to 1.44	<0.0001	Significant
Conventional bur + Biodentine vs Ultrasonic + MTA	0.53	0.15 to 0.91	0.0029	Significant
Conventional bur + Biodentine vs Ultrasonic + Biodentine	0.75	0.37 to 1.13	<0.0001	Significant
Ultrasonic + MTA vs Ultrasonic + Biodentine	0.22	-0.16 to 0.60	0.4279	Not significant

Tukey HSD adjusted p-values. Mean difference is first group minus second group

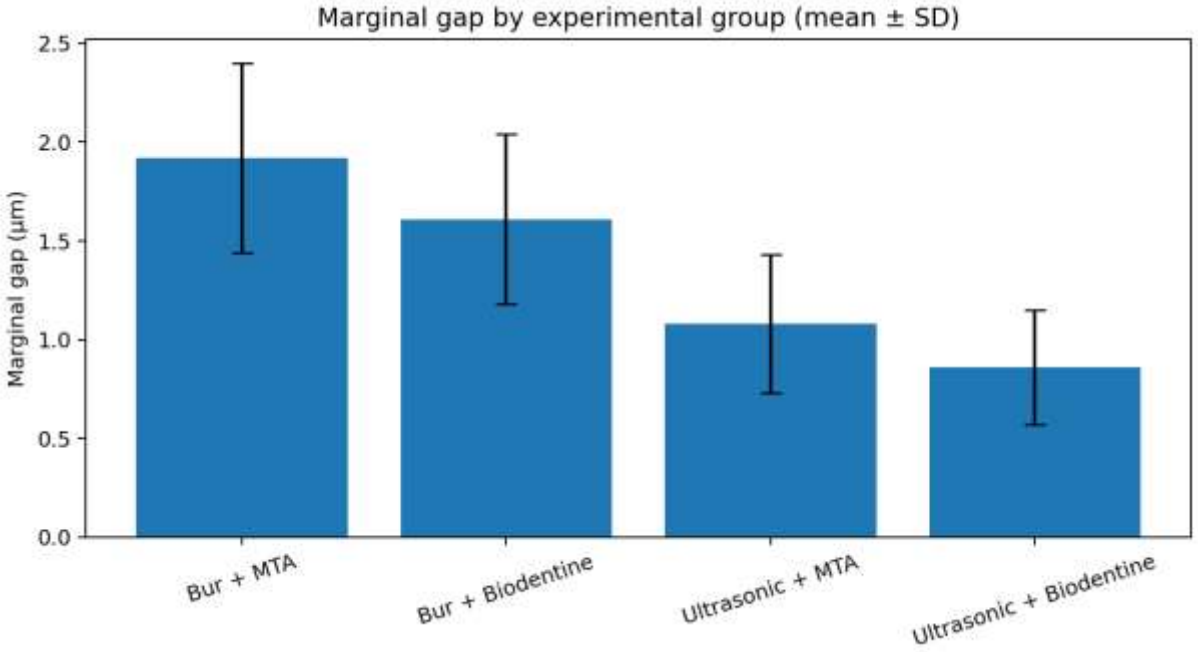

Figure 3: Comparative evaluation of mean marginal gap measurements among the four experimental groups (µm)

Table 8 & Figure 4 illustrate the distribution of dentinal crack formation across the four experimental groups. The highest incidence of cracks was recorded in the conventional bur + MTA group (5/15; 33.3%), followed by the conventional bur + Biodentine group (4/15; 26.7%). Comparatively, fewer cracks were identified in the ultrasonic + MTA group (2/15; 13.3%) and the ultrasonic + Biodentine group (1/15; 6.7%).

Across all 60 specimens, dentinal cracks were detected in 12 (20.0%), whereas the remaining 48 specimens (80.0%) showed no evidence of crack formation. Chi-square analysis revealed no statistically significant association between the experimental groups and dentinal crack occurrence ($\chi^2 = 4.167$, $p = 0.244$). Although the ultrasonic preparation groups exhibited comparatively lower crack frequencies than the conventional bur groups, this difference did not reach statistical significance.

Table 8: Dentinal cracks by experimental group

Experimental group	Cracks, n (%)	No cracks, n (%)	Chi Square value	P value
Conventional bur + MTA	5 (33.3%)	10 (66.7%)	4.167	0.244
Conventional bur + Biodentine	4 (26.7%)	11 (73.3%)		
Ultrasonic + MTA	2 (13.3%)	13 (86.7%)		
Ultrasonic + Biodentine	1 (6.7%)	14 (93.3%)		
Total	12 (20.0%)	48 (80.0%)		

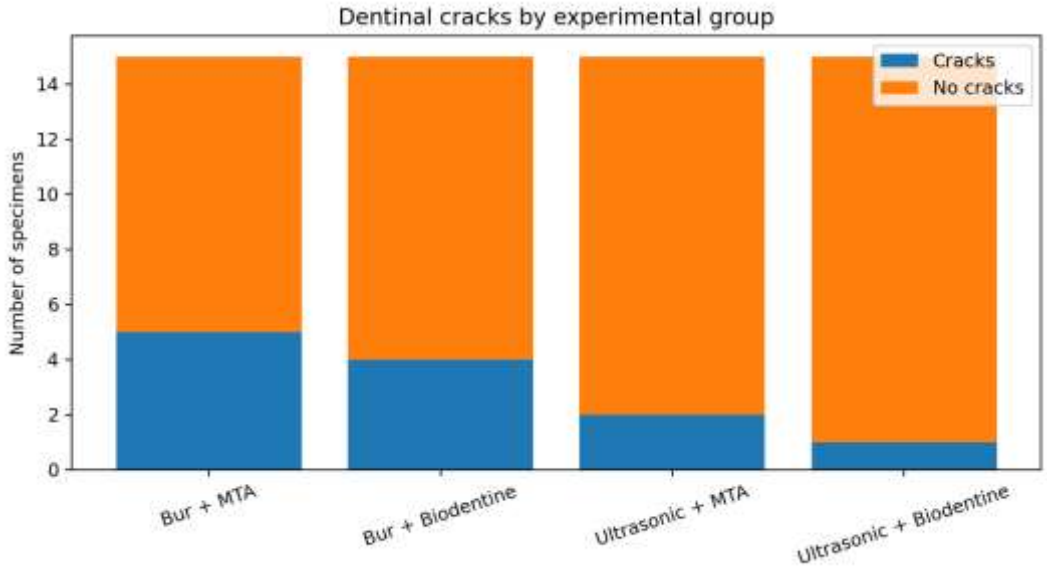

Figure 4: Comparative distribution of dentinal crack occurrence among the four experimental groups

Table 9 & Figure 5 demonstrate the distribution of filling-material voids among the four experimental groups. The greatest proportion of voids was observed in the conventional bur + MTA group (4/15; 26.7%), followed by the conventional bur + Biodentine group (3/15; 20.0%). The ultrasonic + MTA group showed a lower incidence of voids (2/15; 13.3%), while the ultrasonic + Biodentine group recorded the lowest frequency (1/15; 6.7%).

Among the 60 specimens evaluated, filling-material voids were identified in 10 specimens (16.7%), whereas 50 specimens (83.3%) exhibited no detectable voids. Chi-square analysis demonstrated no statistically significant association between the experimental groups and the occurrence of filling-material voids ($\chi^2 = 2.400$, $p = 0.494$). Although the ultrasonic preparation groups showed numerically fewer voids than the conventional bur groups, the observed differences were not statistically significant.

Table 9: Filling-material voids by experimental group

Experimental group	Voids, n (%)	No voids, n (%)	Chi Square value	P value
Conventional bur + MTA	4 (26.7%)	11 (73.3%)		
Conventional bur + Biodentine	3 (20.0%)	12 (80.0%)		
Ultrasonic + MTA	2 (13.3%)	13 (86.7%)	2.400	0.494
Ultrasonic + Biodentine	1 (6.7%)	14 (93.3%)		
Total	10 (16.7%)	50 (83.3%)		

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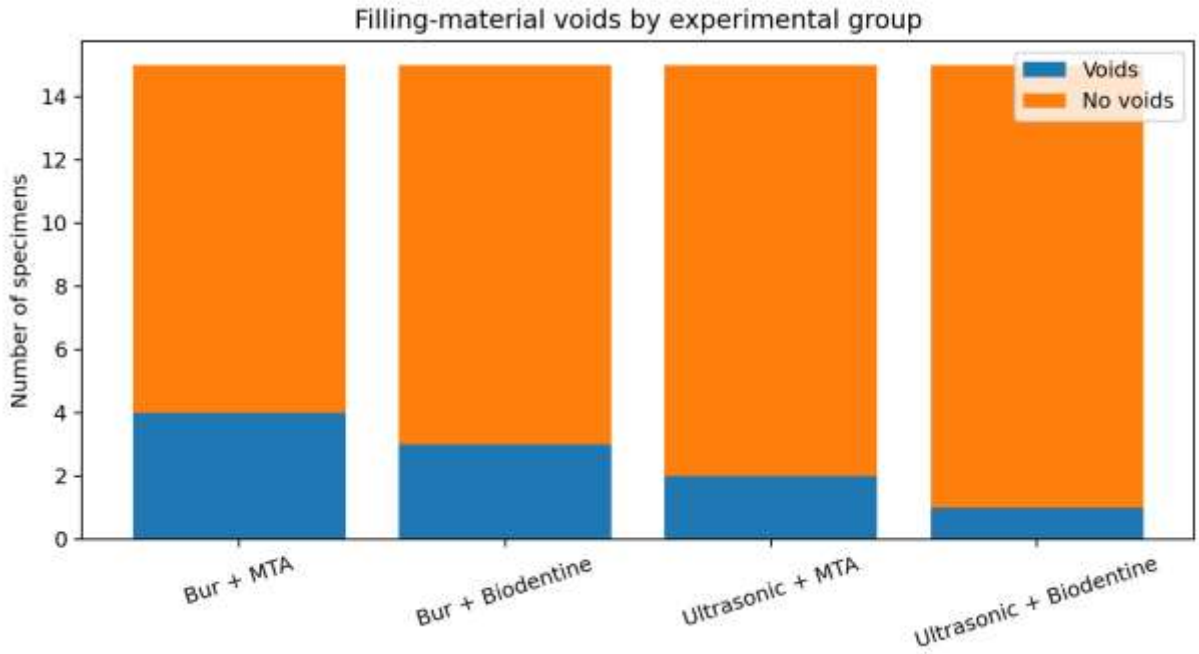

Figure 10: Comparative distribution of filling-material voids among the four experimental groups

DISCUSSION

The success of surgical endodontic treatment depends on several factors, including adequate root-end preparation, effective removal of apical irritants, appropriate selection of retrograde filling materials, and the establishment of a reliable apical seal. Conventional periapical surgery and contemporary endodontic microsurgery differ primarily in their instrumentation techniques, visualization methods, and approaches to root-end cavity preparation. The introduction of ultrasonic instrumentation, surgical magnification, and biocompatible calcium silicate-based materials has contributed to improvements in the precision of surgical procedures and the quality of root-end restorations. The present study focuses on comparing root-end preparation and filling techniques used in conventional periapical surgery and endodontic microsurgery, with particular emphasis on cavity characteristics, marginal adaptation, and apical sealing ability.

Kim et al. (2016) [10] conducted a randomized controlled clinical study comparing mineral trioxide aggregate (MTA) with super ethoxybenzoic acid (Super-EBA) as root-end filling materials in endodontic microsurgery. Their investigation evaluated the long-term clinical outcomes associated with the use of these materials and provided evidence regarding the importance of retrograde filling material selection in determining surgical success. MTA has gained considerable attention because of its favourable biological properties, moisture tolerance, and ability to support periapical tissue healing. In comparison, Super-EBA has traditionally been used because of its handling characteristics and sealing properties. Their findings highlight that the selection of a suitable root-end filling material is an important consideration in achieving predictable outcomes following endodontic microsurgery. The relevance of this investigation to the present study lies in evaluating the performance of different retrograde materials and understanding their contribution to the quality of the apical seal.

Lindeboom et al. (2005) [11] performed a prospective randomized clinical investigation comparing MTA and intermediate restorative material (IRM) as root-end filling materials in single-rooted teeth undergoing endodontic surgery. Their study emphasized the importance of material selection in determining the clinical outcome of periapical surgical procedures. MTA offers favourable biological compatibility and the ability to establish a stable interface with surrounding dentinal tissues, whereas IRM represents a conventional reinforced zinc oxide-eugenol (ZOE) -based restorative material. The comparison of these materials provides an important background for evaluating contemporary retrograde filling alternatives. In the context of the present investigation, differences in material composition and physical properties may influence the adaptation of the filling material to the prepared root-end cavity and consequently affect its sealing performance.

Ishikawa et al. (2003) [12] evaluated root-end cavity preparation using ultrasonic retrotips and investigated their application in surgical endodontics. Ultrasonic instrumentation has become an important component of modern endodontic microsurgery because it allows the preparation of relatively conservative root-end cavities along the longitudinal axis of the root. Compared with conventional rotary instrumentation, ultrasonic retrotips offer improved access to the apical region and facilitate the preparation of cavities with appropriate depth and configuration. The preservation of radicular dentin is particularly important because excessive removal of root structure may compromise the mechanical integrity of the remaining root. Their investigation supports the importance of evaluating instrumentation techniques independently of filling material selection. These considerations are particularly relevant to the present study, in which the characteristics of root-end cavities prepared using different techniques are important determinants of the final restoration.

Saunders et al. (1994) [13] investigated microleakage associated with EBA root-end fillings following ultrasonic root-end preparation. Their work contributed to the understanding of the relationship between cavity preparation techniques and the sealing performance of retrograde filling materials. An effective root-end filling should establish close adaptation with the surrounding dentinal walls to minimize the movement of microorganisms and their by-products through the apical region. The quality of cavity preparation, surface characteristics, and adaptation of the filling material may influence the development of marginal gaps and subsequent leakage. Their findings provide an important experimental basis for evaluating the sealing ability of root-end restorations prepared using ultrasonic instrumentation. The present investigation similarly considers apical sealing performance as an important parameter for comparing conventional and microsurgical root-end procedures.

Naik et al. (2015) [14] conducted an in-vitro investigation to assess the apical sealing ability of MTA and Biodentine following irrigation of the root-end cavity with MTAD. Their study explored the influence of cavity irrigation and the selection of retrograde filling materials on the quality of the apical seal. MTA and Biodentine belong to the calcium silicate-based material group and possess favourable physicochemical and biological characteristics. Biodentine has attracted attention because of its handling properties, relatively shorter setting time, and potential to support mineralized tissue formation. MTA, on the other hand, has an established role in surgical endodontics because of its biocompatibility and sealing characteristics. Their investigation highlights that the performance of root-end filling materials may also be influenced by the condition of the prepared cavity and the irrigation protocol used before material placement. This is relevant to the present study because variations in cavity preparation and material adaptation may influence the final sealing outcome.

The study by Singh et al. (2013) [15] compared the microleakage of different root-end filling materials using an in-vitro experimental approach. Their investigation addressed an important aspect of retrograde restoration, namely the ability of different materials to prevent leakage at the root-end interface. Microleakage remains a significant concern in surgical endodontics because inadequate sealing may permit the passage of microorganisms, bacterial products, and tissue fluids between the root canal system and surrounding periapical tissues. Differences in material composition, consistency, setting characteristics, and adaptation to dentinal surfaces may influence the extent of leakage. Their work supports the importance of laboratory-based comparisons for determining the relative sealing performance of different retrograde filling materials. The present study extends this area of investigation by considering the contribution of root-end preparation techniques alongside filling material selection.

Collectively, the available evidence highlights the interdependence of root-end cavity preparation and retrograde filling material in achieving an effective apical seal. Preparation techniques influence cavity geometry, dentin preservation, and marginal characteristics, while the properties of the filling material contribute to adaptation and resistance to microleakage. Evaluating these factors together is therefore essential for understanding the quality and reliability of root-end restorations.

The comparative assessment of rotary and ultrasonic techniques may provide valuable insights into their ability to produce appropriately shaped cavities while preserving the structural integrity of the root. Similarly, investigating MTA, Biodentine, and other bioceramic materials may help clarify differences in their sealing performance and suitability for retrograde application. Although in-vitro studies cannot fully replicate the biological responses and functional stresses encountered clinically, they provide a controlled environment for identifying technique- and material-related variations in restoration quality. Parameters such as cavity dimensions, marginal adaptation, and microleakage are particularly relevant for assessing these differences.

The present investigation is particularly relevant because it considers root-end preparation and filling techniques as interrelated components rather than evaluating the filling material in isolation. An effective retrograde restoration requires not only a material with favourable sealing properties but also a suitably prepared cavity that permits adequate material placement and adaptation. The combined evaluation of these factors may help provide a more comprehensive understanding of the technical considerations involved in conventional periapical surgery and endodontic microsurgery. Further clinical investigations with longer follow-up periods would be valuable to establish whether differences observed under laboratory conditions translate into meaningful improvements in periapical healing and long-term tooth retention.

LIMITATIONS

The present in-vitro investigation provides useful insights into the influence of root-end cavity preparation techniques and retrograde filling materials on apical sealing ability, marginal adaptation, dentinal crack formation and filling-material voids. Nevertheless, certain limitations should be acknowledged when interpreting the findings and considering their applicability to clinical endodontic practice.

1. In-vitro study design: The investigation was conducted under controlled laboratory conditions, which cannot completely reproduce the complex biological and mechanical environment encountered during clinical endodontic microsurgery. Factors such as periodontal tissue response, periapical inflammation, tissue healing, blood contamination and variations in the oral environment were not incorporated into the experimental model. Consequently, the observed outcomes may not directly represent the clinical performance of the evaluated techniques and materials.

2. Limited sample size: The study included 60 specimens distributed equally among four experimental groups, with 15 specimens in each group. Although this allocation permitted comparative evaluation, the relatively small sample size may have limited the statistical power to identify differences in categorical outcomes, particularly dentinal crack formation and filling-material void occurrence. The absence of statistically significant findings for these parameters should therefore be interpreted cautiously.

3. Restricted selection of root-end preparation techniques: Only two instrumentation approaches, namely conventional rotary bur preparation and ultrasonic-assisted preparation, were investigated. Other commercially available root-end preparation systems and advanced instrumentation techniques were not included. Differences in instrument design, cutting efficiency, vibration characteristics and operating parameters may influence cavity morphology, dentinal integrity and the quality of the prepared root-end surface. Therefore, the findings cannot be generalized to all available root-end preparation systems.

4. Limited selection of retrograde filling materials: The investigation evaluated only two calcium silicate-based materials, MTA and Biodentine. Although both materials are widely used in endodontic procedures, several alternative retrograde filling materials with different physical, chemical and handling characteristics were not assessed. Materials such as bioceramic putties, Super EBA and other contemporary root-end restorative formulations may demonstrate different sealing and adaptation properties. Their exclusion restricts the comparative scope of the present investigation.

5. Assessment of apical sealing using dye penetration: Apical dye penetration was used as an indicator of sealing effectiveness. Although this method is commonly employed in laboratory investigations, dye penetration does not completely reproduce the movement of microorganisms, bacterial products or fluids through the root-end interface. The results may also be influenced by dye characteristics, immersion conditions, specimen preparation and measurement procedures. Therefore, dye penetration findings should not be considered a direct substitute for clinical leakage or long-term sealing performance.

6. **Limitations associated with marginal gap assessment:** Marginal adaptation was evaluated through measurements of the gap between the retrograde filling material and the surrounding dentinal walls. However, marginal characteristics may vary across different regions of the root-end cavity. A limited number of measurement locations may not adequately represent the complete material–dentin interface. Furthermore, microscopic measurements provide information about selected sections rather than the entire three-dimensional (3D) adaptation of the filling material.

7. **Limitations in dentinal crack detection:** Dentinal crack formation was documented according to the presence or absence of detectable cracks. This binary classification did not differentiate between crack dimensions, depth, orientation, severity or anatomical location. Additionally, pre-existing structural defects and newly developed cracks may be difficult to

distinguish without detailed baseline imaging. More advanced techniques, including micro-computed tomography (micro-CT), could provide improved characterization of dentinal defects before and after root-end preparation.

8. **Qualitative assessment of filling-material voids:** Filling-material voids were recorded as present or absent rather than being quantified according to their volume, dimensions, distribution or location within the retrograde restoration. Consequently, minor and extensive voids were classified under the same category, potentially overlooking clinically relevant differences in the internal quality of the fillings. 3D imaging and volumetric analysis could provide a more comprehensive evaluation of internal void formation.

9. **Absence of long-term evaluation:** The investigation focused on laboratory-based assessments and did not examine the long-term stability of the retrograde filling materials. Factors such as dimensional changes, material dissolution, degradation, marginal deterioration and resistance to prolonged moisture exposure were not evaluated. Long-term observations would be necessary to determine whether the initial sealing and adaptation characteristics remain stable over time.

10. Limited evaluation of material handling and biological characteristics: The study primarily concentrated on physical and structural parameters. Important material-related properties, including biocompatibility, antimicrobial activity, bioactivity, calcium ion release, setting behaviour and interaction with periapical tissues, were not investigated. These characteristics may influence the overall suitability of retrograde filling materials in clinical applications.

11. Potential influence of operator-related factors: Root-end cavity preparation and material placement may be influenced by operator experience, instrumentation pressure, working angulation, vibration intensity, preparation duration and condensation technique. Although standardized laboratory procedures were intended to maintain consistency, minor variations in these factors could have influenced the experimental outcomes. Future investigations should incorporate detailed operator calibration and reproducibility assessments.

12. Limited generalizability of the findings: The results were obtained using a specific experimental arrangement involving two preparation approaches and two filling materials. Variations in tooth morphology, root anatomy, dentinal thickness, canal configuration and clinical accessibility were not comprehensively represented. Therefore, the findings should be interpreted within the boundaries of the experimental conditions rather than extrapolated directly to all clinical endodontic microsurgical situations.

Future investigations should consider larger sample sizes, advanced 3D imaging techniques, quantitative evaluation of dentinal defects and void distribution, and longer observation periods. Incorporating additional retrograde filling materials and clinically relevant experimental conditions may further clarify the relationship between root-end preparation techniques and restoration performance.

CONCLUSION

The present in-vitro study demonstrated that ultrasonic-assisted root-end cavity preparation resulted in significantly lower apical dye penetration and marginal gap measurements compared with conventional bur preparation, indicating improved apical sealing and marginal adaptation. The ultrasonic + Biodentine combination exhibited the most favourable numerical outcomes among the four experimental groups. However, differences in dentinal crack formation and filling-material void occurrence were not statistically significant. Overall, ultrasonic instrumentation appears to be a promising approach for root-end cavity preparation, although further studies are required to validate its long-term clinical effectiveness.

Conflicts of Interest: Nil

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