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Comparative evaluation of efficacy of two Acid Solutions in the retreatment of endodontically treated teeth obturated with a Bioceramic sealer -An In Vitro Study

Abstract

Aims: This study aimed to assess the efficacy of 10 % Glycolic acid and 10 % Formic acid in the retreatment of endodontically treated teeth obturated with Bio C Sealer.

Methods and Material: Thirty straight single rooted, single canal premolars were decoronated and prepared with Protaper Gold rotary files up to size F3 followed by obturation with gutta percha and Bio C Sealer in single cone technique. The samples were then incubated at 37 degrees centigrade and 100 % humidity for 2 weeks. Gutta percha was removed using heat and rotary files. The samples were then randomly assigned into three groups according to the solvent used (n=10): Group 1 (10% Glycolic acid), Group 2 (10% Formic acid), and Group 3 (Saline control). Each sample received 0.3 ml of respective solvent for 5 min followed by passive ultrasonic activation. The samples were then longitudinally sectioned and observed under scanning electron microscope (SEM).

Results: 10 % Glycolic acid and 10 % Formic acid performed better than control group in the retreatment of endodontically treated teeth obturated with Bio C Sealer across all thirds of root canal.

Conclusions: 10 % Glycolic acid combined with ultrasonics demonstrated superior efficacy than 10 % Formic acid combined with ultrasonics in the retreatment of endodontically treated teeth obturated with bioceramic sealer even though it was not statistically significant.

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INTRODUCTION

Nonsurgical root canal retreatment is the primary treatment option for endodontic failure, with success rates comparable to those of primary treatments. The root canal system must be thoroughly cleaned, disinfected, and re-obtured after the removal of old obturation materials.^[1]

Bioceramic-based root canal sealers are a valuable advance in endodontics due to their superior physicochemical properties and biocompatibility.^[2] Bioceramic sealers, made from biocompatible ceramics, provide advantages such as little shrinkage, antibacterial characteristics, and bio integration.^[3]

Due to the strong chemical bonds formed between the bioceramic sealer and dentine hydroxyapatite, removal of these sealers often poses challenges during retreatment. Some authors have reported difficulties such as increased operatory time, inability to regain working length and apical patency, and incomplete removal of these types of sealers when compared to resin sealer/AH plus sealer.^[3] Gutta percha removal with heat/gates glidden is successful coronally, but sealer tags remain apically. Studies show 15-25% instrument fractures and 10% ledges, emphasizing chemical-mechanical synergy over files alone.^[4] Once set, bioceramic sealers form a hard, mineralised tissue that requires a solvent, such as a mild acid, to break down the sealer particles and make removal easier from the canal. Acids with strong and high concentrations should be avoided due to their biocompatibility.^[5]

Bio-C Sealer (Angelus, Londrina, PR, Brazil) is a pre-mixed bioceramic product that contains calcium silicates, calcium aluminate, and zirconium oxide. It releases calcium ions that help form mineralised tissue. When used in the single-cone obturation technique, the sealer volume in the canal is larger, which makes it harder to remove in complex anatomies. Research indicates that if the sealer removal is inadequate, working length is not re-established, and material residues persist following retreatment.^[6]

Glycolic acid, an alpha-hydroxy acid, removes smear layer and demineralizes dentin at concentrations of roughly 10%. It preserves dentin microhardness better than stronger chelators such as Ethylenediaminetetraacetic Acid (EDTA) while exhibiting dose-dependent cytotoxicity appropriate for short irrigation. Studies show that its smear layer dissolution rivals citric acid, making it a promising final rinse or retreatment additive.^[7] In addition, formic acid is a well-known corrosive solvent for hardened Portland cement, which has a chemical composition similar to bioceramic sealer. Also, in a recently published study, Garrib and Camilleri et al discussed how instrumentation along with 10% Formic acid effectively removed bioceramic sealer.^[8]

Till date, none of the solvents tested were able to completely remove bioceramic sealers, even though a large number of solvents have been evaluated. Hence combination of solvents along with ultrasonic activation in the retreatment of endodontically treated teeth obturated with a bioceramic sealers needs to be evaluated highlighting the importance of this study.

SUBJECTS AND METHODS:

Sample selection

Thirty straight single-rooted, single canal, mandibular and maxillary premolar teeth extracted for the purpose of orthodontic and periodontal reasons were collected. Teeth with extensive caries, fractures, resorption, multiple canals, calcifications were excluded. Sample size was calculated based on Chhabra and Parolia et al (2023) using G*Power software at 95% confidence and 80% power (effect size = 0.613), yielding a total of 30 specimens.^[15]

Specimen preparation

All the teeth were cleaned and stored in distilled water until used. Samples were decoronated at the cemento-enamel junction to standardize the remaining root length to 16mm ±1. No 8 K files were used to negotiate the canal until visualized at the apical foramen, and hand filing was done up to 20 size K file. Then, pro taper gold files (Dentsply, Switzerland) were used to prepare the canal up to size F3 in a crown-down technique and irrigated with distilled water followed by 5 ml of 3.0 % Naocl (Prime Dental Products Pvt Ltd, India) and 17% EDTA (Anabond Stedman, Desmar, India) to remove the smear layer. Final rinsing was done with distilled water. Canals were dried with paper points. All samples were obturated with gutta percha points 1mm short of the apex, and temporary sealing of the coronal portion with temporary filling material (Cavit G, 3M ESPE, Germany) was done. The samples were covered with a damp gauze and soaked in saline and stored in an incubator at 100% humidity at 37 degrees centigrade for 2 weeks.

Retreatment procedure

After 2 weeks, the temporary filling material was removed with a round tungsten carbide bur and gutta percha in the coronal third was softened and removed using heat. K files were used to remove gutta percha followed by H files and pro taper universal retreatment files (Dentsply, USA) to 1 mm short of working length. The specimens were randomly divided into three groups (n=10).

Group 1 (Glycolic Acid) - 0.3 ml of 10% glycolic acid (Kanton Laboratories, India) placed in the canal for five minutes followed by passive ultrasonic activation.

Group 2 (Formic Acid) - 0.3 ml of 10% formic acid (Kanton Laboratories, India) placed in the canal for five minutes followed by passive ultrasonic activation.

Group 3 (Control) - 0.3 ml of saline placed in the canal for five minutes followed by passive ultrasonic activation. After 5 minutes, the sealer was removed with an ultrasonic tip (Ultra X, Eighteenth Medical Technology Co., Ltd., Changzhou, Jiangsu, China) followed by flushing the canal using 1 ml of acid solutions and the ultrasonic tip was activated for two cycles of twenty seconds each. Final rinse of all the canals was done with distilled water and dried using paper point.

Evaluation

All the specimens were longitudinally sectioned into two halves and observed under a scanning electron microscope. The residual obturation material was scored

and tabulated according to the following criteria by Somma et al:

Score 0: 0-25 % of obturation material covering the dentinal surface

Score 1: > 50 % of obturation material covering the dentinal surface

Score 2: 50-75 % of obturation material covering the dentinal surface

Score 3: 75-100 % of obturation material covering the dentinal surface

The SEM images were assessed by an independent evaluator, and the results were tabulated according to the scoring criteria mentioned above.

Statistical analysis

Data were coded in Microsoft Excel and analysed using IBM SPSS Statistics version 27. The Kruskal–Wallis test was used to assess differences across the three groups for each canal third. Bonferroni-adjusted post hoc comparisons were applied following a significant Kruskal–Wallis result. A p-value of less than 0.05 was considered statistically significant.

RESULTS:

Statistically significant differences were observed for all the three groups across the coronal, middle and apical thirds of the root canal ($p < 0.001$ for all thirds). All SEM scores are presented in table 1 and 2

Table 1: Distribution of SEM scores for residual Bio-C Sealer across root canal thirds in different solvent groups

Group	Coronal third n (%)				Middle third n (%)				Apical third n (%)			
	S0	S1	S2	S3	S0	S1	S2	S3	S0	S1	S2	S3
Group I – GA†	6 (60)	4 (40)	0	0	6 (60)	4 (40)	0	0	4 (40)	2 (20)	4 (40)	0
Group II – FA‡	4 (40)	2 (20)	4 (40)	0	2 (20)	4 (40)	0	4 (40)	2 (20)	0	6 (60)	2 (20)
Group III – Control§	0	0	6 (60)	4 (40)	0	0	4 (40)	6 (60)	0	0	1 (10)	9 (90)

S0 = Score 0 (0–25% residual obturation material); S1 = Score 1 (25–50%); S2 = Score 2 (50–75%); S3 = Score 3 (75–100%), based on scoring criteria of Somma et al.

†GA: 10% Glycolic acid + passive ultrasonic activation; ‡FA: 10% Formic acid + passive ultrasonic activation; §Control: Normal saline + passive ultrasonic activation.

Each group: n = 10. Values represent number of samples (percentage). SEM: Scanning electron microscope

Table 2: Statistical comparison of solvent efficacy across root canal thirds using Kruskal-Wallis and Bonferroni-adjusted pairwise tests

Canal third	Kruskal-Wallis H	P value*	Group I vs III†	P value*	Group II vs III‡	P value*	Group I vs II§	P value*
Coronal	17.974	<0.001	15.600	<0.001	10.800	0.012	–4.800	0.609 (NS)
Middle	15.791	<0.001	15.000	<0.001	6.600	0.243 (NS)	–8.400	0.079 (NS)
Apical	17.537	<0.001	15.400	<0.001	9.800	0.025	–5.600	0.398 (NS)

* $P < 0.05$ considered statistically significant. Bonferroni-adjusted post hoc test applied following a significant Kruskal–Wallis test.

†Group I (GA) vs Group III (Control); ‡Group II (FA) vs Group III (Control); §Group I (GA) vs Group II (FA).

NS: Not significant. GA: 10% Glycolic acid + passive ultrasonic activation; FA: 10% Formic acid + passive ultrasonic activation.

Statistical analysis performed using IBM SPSS Statistics, version 27

In the coronal third, 60% of samples in the glycolic acid group showed Score 0, whereas 40% showed Score 1 (Figure 1a). 40% of samples in the formic acid group had a Score of 0, 20% had a Score of 1 (Figure 2a), and 40% had a moderate amount of sealer left on them (Score 2). The majority of samples in the control group (60%) had moderate remains (Score 2), whereas 40% (Figure 3a) had

substantial residual material (Score 3). Pair wise comparisons showed significant differences between the control and glycolic acid groups ($p < 0.001$) and between the control and formic acid groups ($p = 0.012$). No significant difference was found between the glycolic and formic acid groups ($p = 0.609$).

Glycolic acid continued to work well in the middle third, which followed a similar pattern. 10% Glycolic Acid outperformed the other groups consistently, scoring 0 for six samples and score 1 for four samples (Figure 1b). Results with 10% formic acid were inconsistent, with 20% receiving a score of 0 and 40% receiving a score of 1 (Figure 2b). Nevertheless, considerable sealer residues were still present in 40% of the samples in this group (Score 3). In the control group, 60 % of the samples received a score of 3 where most of the remnants were left behind (Figure 3b). Pairwise comparisons demonstrated a significant difference between the control and glycolic acid groups ($p < 0.001$); No significant difference was observed between the two acid groups ($p = 0.079$).

Across all groups, the apical third turned out to be the most challenging area for sealer removal. With 40% of samples attaining Score 0 and 20% achieving Score 1,

10% glycolic acid continued to be the most effective agent, especially in the apical area (Figure 1c). Merely 40% displayed moderate residues (Score 2). The efficacy of 10 % Formic acid significantly decreased in the apical region, where 20% of samples had Score 0, 60% had moderate remnants (Score 2), and 20% had considerable remnants (Score 3) (Figure 2c). 90% of samples in the control group had a score of 3, indicating an almost complete failure to remove the sealer in the apical third (Figure 3c). Significant pairwise differences were found between the control and glycolic acid groups ($p < 0.001$) and between the control and formic acid groups ($p = 0.025$); the difference between the two acid groups was not statistically significant ($p = 0.398$). SEM images revealed greater dentinal surface cleanliness in the glycolic acid group, with more open dentinal tubules visible across all three thirds.

Figure 1

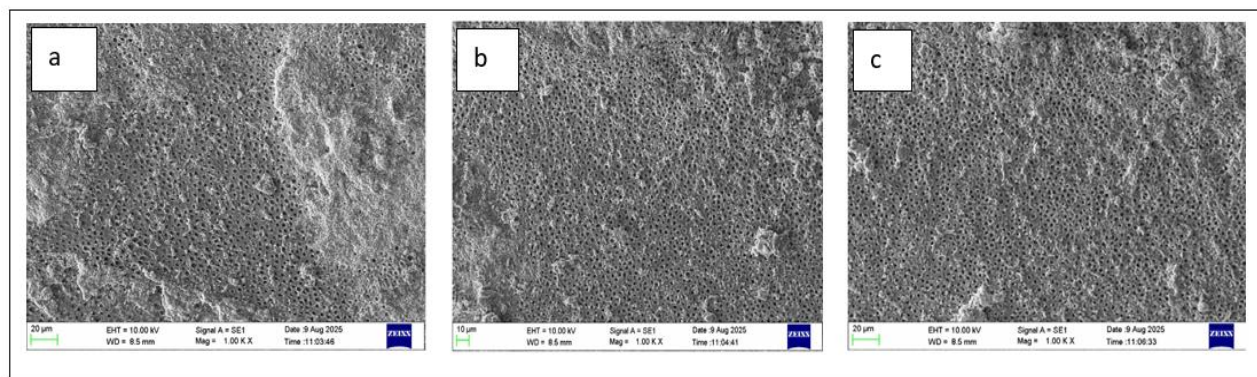


Figure 1a: Glycolic acid group (Coronal third) Figure 1b: Glycolic acid group (Middle third) Figure 1c: Glycolic acid group (Apical third)

Figure 2

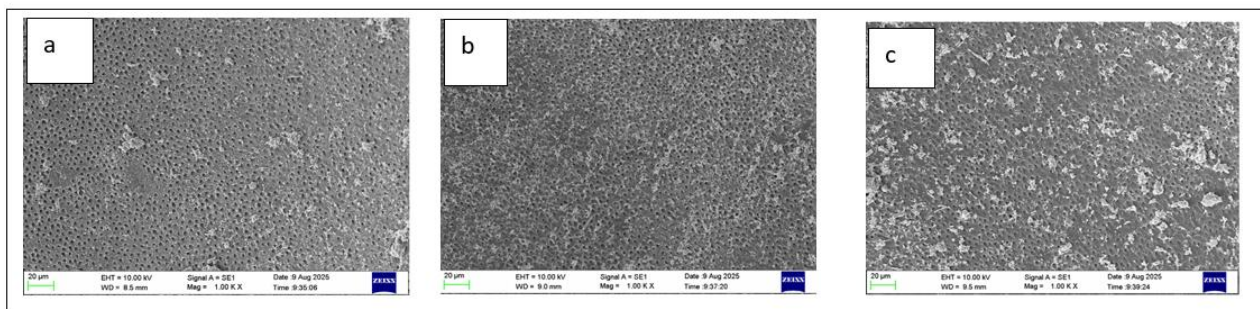


Figure 2a: Formic acid group (Coronal third) Figure 2b: Formic acid group (Middle third) Figure 2c: Formic acid group (Apical third)

Figure 3

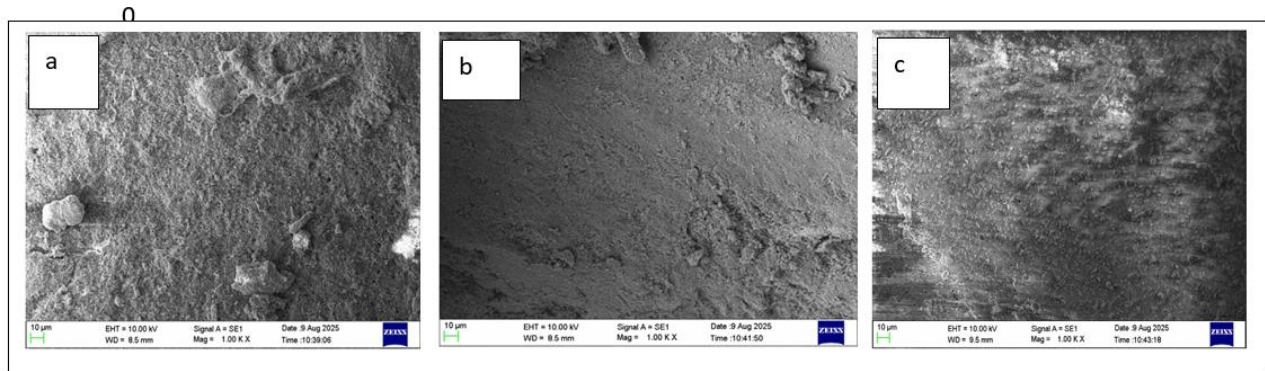


Figure 3a: Control group (Coronal third) Figure 3b: Control group (Middle third) Figure 3c: Control group (Apical third)

DISCUSSION:

The present study compared 10% glycolic acid and 10% formic acid, both combined with passive ultrasonic activation, as chemical adjuncts in the retreatment of endodontically treated teeth obturated with Bio-C Sealer, with saline as a control. The experimental results of this investigation, as determined by SEM score (0-3), demonstrated substantial differences in removal efficacy amongst the three solvents over the coronal, middle, and apical thirds of the root canal. The wider canal width in the coronal region allow for a larger volume of fresh acid to be supplied and activated, which accounts for the increased success rate.

The SEM was particularly chosen for this study, as it provides high-definition surface morphology and topographical information of the dentin-sealer interface. If metallic or high density material like bioceramic sealers incorporating zirconium oxide is present, CBCT scans have a high propensity for "beam hardening" artefacts, leading to an inability to visualize the canal walls and inaccurate assessment of sealer residues. Although CBCT is widely used for clinical diagnosis, its resolution is typically measured in hundreds of microns, which is insufficient for detecting fine layers of residual bioceramic sealer. Micro-CT is great for non-destructive 3D volumetric investigation of filling materials, but even high-resolution Micro-CT scans find it difficult to reliably recreate the level of detail that SEM provides for determining if dentinal tubules are open or occluded by sealer residues.

Recent research on Bio-C Sealer (Angelus, Brazil) demonstrates that it has a lesser retreatability than other bioceramic formulations. In comparative studies using micro-computed tomography, Bio-C Sealer was found to leave most residual material, particularly in the apical area, even when supplemental approaches such as passive ultrasonic irrigation were used. Its great flowability and small particle size allow it to penetrate deeply into anatomical imperfections, providing a "monoblock" effect that is surprisingly resistant to typical retreatment processes.^[9] This is one of the reasons why Bio-C Sealer has been evaluated in this study. One important feature that affects the retrievability of the set calcium silicate sealer (CSS) is its physical strength. The retrievability of different brands of CSSs varies, primarily due to differences in their composition. Compared to hard-set

sealers, soft-set sealers are easier to remove and hard set sealers may cause additional challenges during the retreatment process and may require the use of solvents in addition to mechanical debridement. Carillo et al. evaluated the retreatability of teeth obturated with EndoSequence BC sealerTM, EdgeBioceramicTM and NeoSEALERFloTM and found that NeoSEALERFloTM-filled canals were found to have a greater success rate in restoring patency than the other sealers. The authors discovered that the type of CSS employed could have an impact on restoring patency and that NeoSEALERFloTM might be classified as a soft-setting CSS.^[10-12]

The use of 10% formic acid for retreatment of Bio C Sealer-obturated teeth makes use of its documented corrosive activity against Portland cement precursors found in bioceramic formulations. Formic acid, a low-molecular-weight organic acid, dissolves sealers at a 10% concentration through proton-mediated dissolution, releasing hydrogen ions, which primarily targets calcium ions in the calcium silicate hydrate (C-S-H) matrix. This ion exchange affects the gel's structural cohesiveness, causing calcium depletion, surface pitting, and microcrack propagation, thereby increasing susceptibility to mechanical fragmentation by ultrasonic energy.^[9,12] Garrib and Camilleri demonstrated that 10% formic acid applied for 5 minutes achieves over 95% removal of hydraulic calcium silicate sealer while preserving radicular dentin integrity.^[8,13]

Glycolic acid is an alpha-hydroxy acid with a low molecular weight that allows for extensive penetration into the complex anatomy of the root canal and mineral infiltration zone. Unlike simple acids, glycolic acid has a twofold action mechanism. The carboxylic group gives off the needed protons for the initial breakdown of the minerals, and the nearby hydroxyl group chelates the calcium ions. This inhibits the reprecipitation of dissolved minerals, which would otherwise restrict the canal space.^[1,14] Chhabra et al demonstrated that 10% glycolic acid combined with ultrasonic activation produced significantly less residual OrthoMTA than 5% glycolic acid, 10% citric acid, or distilled water controls, with SEM revealing superior chelating activity against calcium silicate matrices when ultrasonically activated.^[15] Because OrthoMTA and Bio-C Sealer share identical tricalcium silicate hydration mechanisms producing

resistant calcium silicate hydrate gels, these findings are directly relevant to the present study.

Ultrasonic energy helps to remove sealer by generating high-velocity fluid vortices and cavitation, which is the implosion of micro-bubbles that releases energy against the sealer surface. This shows a clear synergy: the 10% acids chemically "soften" the Bio C Sealer by lowering its microhardness, making the set material susceptible to the hydrodynamic forces of ultrasonic tips. Without this chemical pre-treatment, as demonstrated in the saline group, mechanical forces of ultrasonics alone are inadequate to break the chemical bond of set bioceramic sealer. This is corroborated by Saleh and Alkhalidi et al, who found that citric acid supplemented with ultrasonic activation significantly improved bioceramic sealer removal over mechanical retreatment alone, with ultrasonic activation proving particularly beneficial in the apical third.^[16]

Several related investigations support these findings. Hanson et al. reported that Endo Sealer Solvent and EDTA outperformed chloroform and sodium hypochlorite in dissolving EndoSequence BC Sealer, achieving 100% patency restoration and superior smear layer removal, indicating that chelating agents are more effective than conventional solvents against inorganic bioceramic matrices.^[17] Johnson et al found that xylene and 10% citric acid were more effective than chloroform in the dissolution of bioceramic sealers.^[18] Alamoudi et al demonstrated that bioceramic sealers consistently showed greater residual material than AH Plus regardless of solvent type — D-limonene, 10% formic acid, or 20% hydrochloric acid — underscoring the inherent difficulty of retreating bioceramic-obtured teeth.^[19] Almnea et al found that an experimental dual-action Deep Eutectic Solvent was superior to Endosolv R and GuttaClear in terms of sealer removal efficacy achieving over 92% removal in the apical third of the canal. This was due to the dual action of softening gutta-percha and enzymatically dissolving bioceramic sealers.^[20]

17% EDTA decreases root dentin microhardness by 20–31% after 3–5 minutes due to calcium chelation with hydroxyapatite crystals. In contrast, 10% glycolic acid maintains flexural strength relatively close to control values because of its alpha-hydroxy structure that allows superficial demineralization with minimal disruption to the collagen fibers.^[21] Rezaei et al demonstrated that a 10% formic acid resulted in less dentin microhardness reduction than a 20% hydrochloric acid, even though the latter had the fastest rate of patency, with a more intense degree of dentinal tubule deterioration.^[8] This supports the safety assessment of a 10% concentration of both acids when used for a short period of time, i.e., 5 minutes, as in the present study.

The clinical significance of this study lies in proving how 10% glycolic acid with ultrasonic activation is a potentially useful technique for the retreatment of endodontically treated teeth obturated with a bioceramic sealer, with particular benefits in the apical third where conventional solvents tend to fail. Nonetheless, this study has inherent limitations. The results of this in vitro study may not completely simulate the complex physiological conditions of the oral cavity, such as the presence of periapical tissues or intricate natural biofilm. This study may not fully reflect the state of a bioceramic sealer that

has been ageing in a canal for several years, as it may become more resistant to breakdown over time. Only straight single-rooted teeth were considered, and the results may not be applicable to curved, calcified, and/or multi-rooted anatomy. The effect on the long-term fracture resistance of dentin was not tested, and the evaluation was restricted to the Bio-C Sealer; the results may vary with different calcium silicate-based sealer materials because of their different compositions. Although the SEM can provide excellent surface magnification, it cannot detect the presence of the sealer that might be retained in the lateral canals and dentinal tubules.

Future studies should also include complex anatomy, including curved canals, isthmuses, and oval canals, to better simulate clinical scenarios. The efficacy of glycolic acid may also be assessed in conjunction with laser, as well as multisonic systems such as the GentleWave system. The efficacy of glycolic acid may also be assessed in relation to emerging solvents, like Endo Sealer Solvent and deep eutectic solvents. The use of micro-CT scans, along with Vickers hardness tests, would also give insight into the long-term integrity of the dentin after exposure to the acid.

CONCLUSION

10 % Glycolic acid combined with ultrasonics demonstrated superior efficacy than 10 % Formic acid combined with ultrasonics in the retreatment of endodontically treated teeth obturated with bioceramic sealer even though it was not statistically significant. This study will be of value to clinicians as the retreatment of endodontically treated teeth obturated with bioceramic sealer is challenging.

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