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In Vitro Evaluation of the Solubility of EndoSequence BC Sealer in Acidic Solutions

A thesis submitted in partial fulfillment of the requirements for the degree of Master of Science
in Dentistry at Virginia Commonwealth University.

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Abstract

IN VITRO EVALUATION OF THE SOLUBILITY OF ENDOSEQUENCE BC SEALER IN ACIDIC SOLUTIONS

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Thesis Advisor: Garry L. Myers, DDS

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Introduction: In recent years, bioceramic sealers (BCS), such as EndoSequence BC Sealer, have gained popularity in endodontic practices. Previous research has found BCS difficult to remove from canals during retreatments when compared with other endodontic sealers. Acidic solutions have been found to inhibit the set of BCS and increase their solubility. The purpose of this study was to evaluate the efficacy of acidic solutions in the removal of BCS as compared with a traditional retreatment solvent.

Methods: Roots of extracted human single rooted teeth (N=15) were prepared into 17 mm sections, then instrumented with ProTaper Gold (PTG) rotary files up to size F4. The canals were obturated 2 mm short of working length with gutta-percha (GP) and BCS using a single cone. The teeth were randomly assigned into 3 groups (N = 5/group) based on solution: chloroform, 5% acetic acid, or 37% phosphoric acid. GP was removed using ProTaper Universal

Retreatment files without solvent. The canals were filled with solution, then filed with PTG F1 to F4 to working length (WL). The time taken to regain WL and the ability to regain patency were evaluated. Differences in procedure time for the three different solutions were assessed using Kruskal-Wallis test.

Results: Patency was achieved for 100% of samples treated with acetic acid and phosphoric acid, and for 80% of teeth treated with chloroform. The time to reach WL for acetic acid was significantly faster than chloroform and phosphoric acid ($P < .05$).

Conclusions: Acetic acid significantly improved the ability to retreat canals filled using BCS.

Introduction

Success of non-surgical root canal therapy relies on the clinician's ability to thoroughly debride the root canal system, sterilize that system, and then completely obturate it to prevent reinfection (1). Treatment protocols should remove necrotic and inflamed tissue, microorganisms, and debris, while also preventing recontamination by sealing the root canal system with biocompatible materials (2). The primary goal of root canal obturation is complete three-dimensional filling of the entire root canal system with dimensionally stable materials (3). Although primary root canal therapy has a relatively high rate of success between 80% to 95%, failures still occur which may necessitate root canal retreatment (4,5). Reasons for treatment failure include inadequate aseptic control, poor access cavity design, missed canals, inadequate instrumentation and debridement, and leaking temporary or permanent restorations (6). Furthermore, apical periodontitis may persist due to persistent intraradicular infection in complex apical root canal anatomy, extraradicular infection, extruded root canal filling materials, accumulation of cholesterol crystals, and true cystic lesions (6).

The success of nonsurgical retreatment depends on the ability to remove previous root canal obturation materials, gain access to areas of the root canal that were untreated or have persistent infection, and effectively debride, disinfect, and obturate the entire root canal system (7). When the ability to debride the entire root canal system is hindered by the previous root

canal obturation, retreatment success diminishes significantly from 87% to 47% (8). Inability to remove prior root canal filling materials can potentially leave a residual barrier preventing the contact of irrigants and medicaments to root canal walls and dentinal tubules. Bacteria, including *E. faecalis*, are able to penetrate into dentinal tubules and those bacteria may contribute to refractory disease (9,10). Thus, if previous filling materials inhibit the contact of irrigants and medicaments with canal walls and dentinal tubules, greater concentrations of pathologic bacteria may remain in the root canal system and contribute to failure of secondary therapy. It is critical that obturation materials be fully retrievable by clinicians to allow for retreatment in scenarios where refractory disease persists after initial therapy.

Current techniques to obturate root canals mostly involve the use of gutta percha and sealer (11). The most common chemical types of sealers used today include: zinc-oxide eugenol, resin, glass ionomer, silicone, calcium hydroxide, and most recently calcium silicate (12,13). The primary role of sealer is to seal against ingrowth of bacteria from the oral cavity, entomb any remaining microorganisms in the root canal, and complete obturation at a microscopic level to prevent accumulation of fluid which may serve as a nutrient source for bacteria (12). Root canal sealers serve to help obturate canal irregularities and fill voids between canal filling materials and canal walls. Sealers have been shown to be critical in reducing microleakage after obturation when compared to the use of gutta percha alone (14,15). The judicious use of sealer is necessary during obturation to ensure a complete, three-dimensional seal of the entire root canal system, whereas gutta percha alone is insufficient to create an appropriate seal.

According to Grossman, the ideal properties of a root canal sealer include: ability to create a hermetic seal laterally and apically, radiopaque, minimal shrinkage upon setting, non-staining, bacteriostatic, insoluble in tissue fluids, well tolerated by periapical tissues, and soluble

in common endodontic solvents (retreatable) (16). Manufacturers have synthesized various types of sealers in the pursuit of one product that satisfies all of these criteria. However, no sealer developed so far has been able to meet all these standards and each have their own shortcomings. Furthermore, current techniques and solvents used in retreatment do not allow for complete removal of all previous filling materials (17). While some sealers, like epoxy resin based ones, result in greater bond strength to dentin, they also leave greater volumes of residual materials upon removal (18). Most sealers are irritating to periapical tissues, especially during their setting reactions (19). Sealers also generally shrink significantly when setting and wash out in the presence of tissue fluids (20,21). Traditional obturation methods utilize maximum amounts of gutta percha and minimize the thickness of sealer in order to minimize the negative effects of sealer shrinkage and resorption (13). This paradigm has shifted with the relatively recent advent of bioceramics sealers.

Bioceramics are materials that typically contain alumina and zirconia, bioactive glass, glass ceramics, hydroxyapatites, and calcium phosphates (22). The introduction of bioceramics in endodontics started with the release of mineral trioxide aggregate (MTA), which is a combination of medical grade Portland cement and bismuth oxide. MTA was initially used as a root repair material to seal perforations and to create apical barriers to prevent overextensions of root fillings. Bioceramics have many favorable properties for use in dentistry and endodontics. Primarily, bioceramics are biocompatible and are readily accepted by body tissues with minimal inflammation and with favorable healing around the materials (23–25). They are not sensitive to moisture or blood contamination (26,27). They are dimensionally stable and expand when setting, resulting in an excellent seal (28,29). During their setting reaction, calcium hydroxide is released, resulting in a high pH and an antimicrobial environment (30). Dental bioceramics are

considered bioactive when set as they are able to interact and react with body tissues in a favorable manner to synthesize hard tissue. When in contact with tissue fluids, calcium hydroxide is released, which interacts with phosphates in the fluids to form a hydroxyapatite precipitant (31).

While bioceramics have numerous favorable properties that are desired in sealers, the adaptation of MTA into a sealer met some hindrances. MTA was initially formulated with a large particle size due to the technology available at its advent. Furthermore, it was hard to apply in narrow canals. Attempts to create an MTA with a more liquid consistency were always found to negatively change the material's favorable mechanical and physical characteristics (32). However, more recently, a new pre-mixed bioceramic sealer, EndoSequence® BC Sealer™ (BC Sealer), was released with a chemical composition including calcium silicates, zirconium oxide, tantalum oxide, calcium phosphate monobasic, and some fillers (22). BC Sealer has greatly improved handling properties when compared to MTA, but maintains and improves upon MTA's desired properties that were discussed previously (22).

BC Sealer has many properties that are desired in an ideal sealer. It is highly hydrophilic and favors the moisture in root canals and dentinal tubules during its setting reaction (22). When unset, it has a high pH around 12, with similar antimicrobial properties as calcium hydroxide (29,30). It is insoluble in tissue fluids and expands when setting instead of shrinking like most sealers (29,33). When used with compatible gutta percha cores, BC Sealer will bond to the core and minimize any gap formation between the two materials (22). BC Sealer has also been found to bond to dentin, minimizing any gaps between the sealer and tooth structure (34). This bonding is achieved by diffusion of sealer particles into dentinal tubules, infiltration of minerals

into intertubular dentin which results in a mineral infiltration zone, and formation of hydroxyapatite along this mineral infiltration zone (30,35,36).

Proponents of BC Sealer have declared a paradigm shift in canal obturation. Since BC Sealer appears to mitigate the negative properties of most traditional sealers, namely setting shrinkage and solubility in tissue fluids, minimizing the thickness of sealer and maximizing the gutta percha core may no longer be necessary (22). Instead, the gutta percha core may be used as a delivery vehicle that allows hydraulic condensation of BC Sealer into the three-dimensional anatomy of the root canal system and allow the sealer to be the main filling component (22). Although BC Sealer meets many of the ideal criteria that Grossman defined for sealers, the primary concern regarding its use is its retreatability and solubility in endodontic solvents. Treatment using BC Sealer and gutta percha as the root canal filling has been shown to have a success rate of 91%, indicating that there will be many instances where retreatment of these cases is indicated (37).

Removal of BC Sealer from canal walls and ramifications has been found to be difficult using traditional mechanical and chemical methods, including heat, rotary instruments, and chloroform. When BC Sealer was used as the sole material to fill the apical 2 mm of root canals, it was found that full working length (WL) and apical patency were more difficult to reestablish than when gutta percha was fit to full WL or when AH Plus sealer was used (38). Furthermore, significant amounts of BC Sealer were found to be left on canal walls after retreatment (39). When evaluated under micro-CT, canals obturated with gutta percha and BC Sealer were found to have significantly greater residual material left after retreatment than canals filled with AH Plus (40). While it has been evident that retreatment of canals using a variety of sealers tended to leave residual materials inside the canals, the paradigm of filling more canal volume with BC

Sealer rather than gutta percha may increase the overall amount of residual sealer inside canals, especially in isthmuses, ramifications, and tubules (18). Thus, a solvent that dissolves set BC Sealer would be beneficial to increase the efficacy in its removal, especially in areas where mechanical instrumentation is not possible.

One potential class of solvents for the retreatment of BC Sealer is weak acids, as the sealer is highly sensitive to pH. BC Sealer has a high pH prior to setting, and its hydration reaction creates calcium hydroxide as a byproduct (30). This alkaline environment is important in BC Sealer's setting reaction, so disruption of the environment with an acid may negatively affect the set of the material. Furthermore, acidic solutions have been shown to be effective in dissolving bioceramic materials. Acidic solutions, including maleic acid and HEDP, have been shown to increase the solubility of bioceramics (41). Carbonic acid and acetic acid were both found to reduce the surface microhardness of bioceramic materials, with acetic acid being more effective in this regard (42). It appears that acidic solutions may be beneficial in decreasing the amount of residual BC Sealer left after retreatment, especially in isthmuses and tubules where mechanical instrumentation is ineffective.

Phosphoric acid is a weak acid that is commonly used in dentistry as an etchant during bonding procedures for composite resins. Composite resins are often used to restore endodontically treated teeth and to bond fiber posts in endodontically treated teeth with significant loss of coronal tooth structure. Some sealers, such as those containing eugenol, have been found to significantly decrease composite polymerization and bond strength (43,44). As BC Sealer has been found to penetrate dentinal tubules, it may also impair the micro-mechanical bonding of composite resins (45). The effect of phosphoric acid on BC Sealer has not yet been determined. If phosphoric acid increases the solubility of BC Sealer and allows its removal in

greater amounts, it may be an effective agent in root canal retreatment. Furthermore, it may also help mitigate any negative effects that BC Sealer may have on composite bonding.

As BC Sealer is a relatively new material used in root canal therapy, there remains a lack of evidence determining the effectiveness of various solutions in its removal. The aim of this study was to compare the effectiveness of acidic solutions and a traditional endodontic retreatment solvent in the removal of BC Sealer when used to fill root canals with gutta percha in a single cone technique. The effectiveness of various solutions was evaluated by comparing the time to instrument the canal to the original working length and the ability to regain apical patency. The null hypothesis was that there is no significant difference in the effectiveness of acidic solutions and a traditional root canal retreatment solvent in the removal of BC Sealer from root canal systems.

Methods

Specimen Preparation

Fifteen recently extracted, single rooted, human anterior teeth with root curvature less than 20 degrees were selected. Only teeth with fully formed apices and single root canals were selected after radiographic assessment with buccal and proximal radiographs. Teeth were divided into 3 groups of 5 teeth each:

Group 1: Canals obturated with GP/BC Sealer and retreated using chloroform

Group 2: Canals obturated with GP/BC Sealer and retreated using 5% acetic acid

Group 3: Canals obturated with GP/BC Sealer and retreated using 37% phosphoric acid

The crown of each tooth was removed to a standardized length of 17 mm each using a tapered diamond bur (Kerr Corporation, Orange, CA, USA). Canals were accessed and canal patency and working length were determined using a 15 k-file. The file was inserted into each canal until just visible at the apical foramen, then 1 mm was subtracted and recorded as the working length. Teeth were instrumented with ProTaper Gold (Dentsply, Tulsa Dental Specialties, Tulsa, OK, USA) to a final size of F4 at the working length using an electric motor following the manufacturer's protocol. Canals were filled with 6% NaOCl during instrumentation and were irrigated with 0.5 mL of NaOCl after each file. Each canal was irrigated with a final rinse of 3 mL 6% NaOCl, then 1 mL 17% EDTA for 1 minute, then 3mL

6% NaOCl, and finally 1 mL sterile saline 1 mm from the established working length. Canal patency was reconfirmed with a 15 k-file after irrigation was complete.

Canal Obturation

All canals were obturated using GP and BC Sealer using a single cone technique. This obturation technique was confirmed radiographically and clinically to be effective in creating a solid apical plug of BC Sealer in the terminal 3 mm of the root canal during a pilot study. Size F4 ProTaper Gold gutta percha (Dentsply) were fit to full working length in each canal, then 2 mm from the apex of each cone was removed using an 11 blade (Figure 1). This allowed for a solid plug of BC Sealer to fill the apical 3 mm of the root canal. Root apices were covered in a ball of clear utility wax to prevent overexpression of sealer. EndoSequence BC Sealer (Brassler USA) was introduced directly into the coronal third of each canal using tips included by the manufacturer. A size 30 lentulo spiral (Dentsply) was spun in each canal at 5000 RPM to a length 2 mm short of working length to ensure full coating of canal walls with sealer. The previously fit and trimmed gutta percha was then coated in a thin layer of BC Sealer, placed into each canal, seared off at the level of the orifice, then condensed with pluggers (Figure 2). All samples were stored at 37°C in 100% humidity for 7 days to allow sealer to fully set.

Figure 1: Master Cone PA radiograph confirming gutta percha fit 2mm short of WL and 3mm short of the root apex.

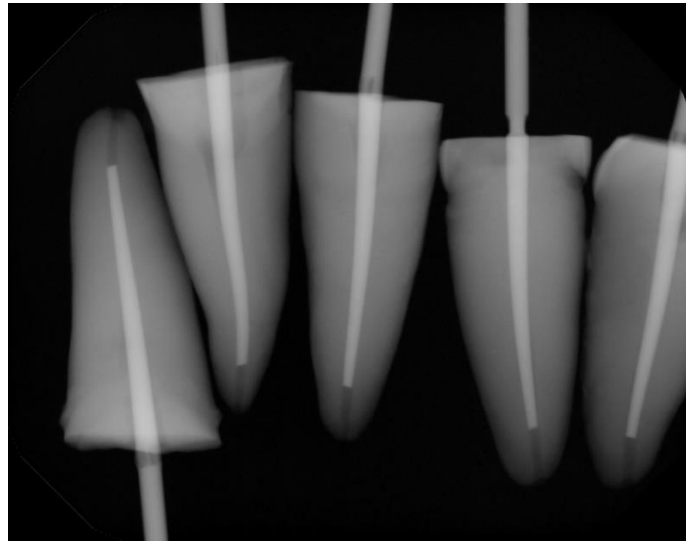
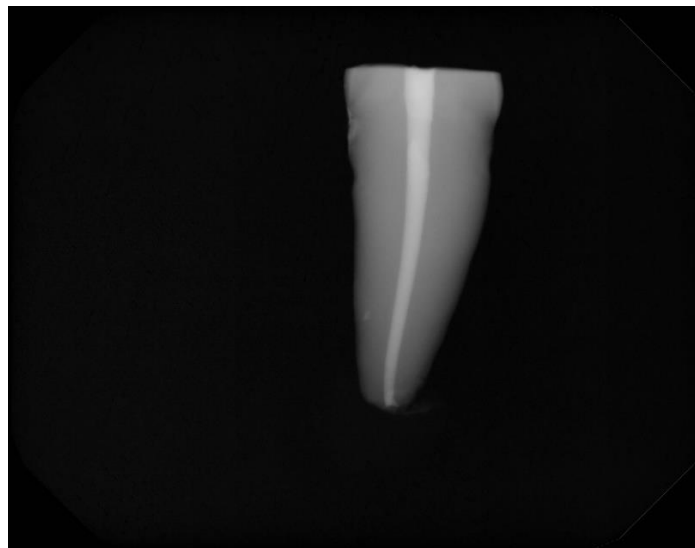


Figure 2: Post-op PA radiograph confirming complete obturation of the root canal with BC Sealer filling the terminal 3mm.



Retreatment

Teeth were divided into 3 groups depending on the solution used during retreatment; group 1 was treated using chloroform, group 2 was treated using 5% acetic acid, and group 3 was treated using 37% phosphoric acid. Gutta percha was removed using ProTaper Retreatment Files (Dentsply) size D1 to a length of 13 mm with gentle pecking motions using an electric motor at

the manufacturer's recommended settings. After gutta percha was removed, canals were filled in entirety with group-specific solutions. A 21 mm length, size F1 ProTaper Gold rotary file (Dentsply) was then used with gentle pecking motions to prepare through the apical plug of BC Sealer to the original working length. The file was removed upon meeting resistance or after 5 amplitudes of movement. The canal was refilled with the test solution prior to each cycle of instrumentation. This process was repeated until a size F4 ProTaper Gold file was able to reach the original working length. Time elapsed from the start of retreatment to reaching working length with a size F4 ProTaper Gold file was recorded. After working length was reached with rotary instruments, the canal was filled again with the test solution, and a 15 k-file was used with watch-winding motions until patency was achieved, or until 5 minutes elapsed. Ability to reobtain patency was recorded.

Statistical Analysis

Ability to achieve patency was summarized using counts and percentages. Differences in standardized procedure time for the three different irrigants were assessed using Kruskal-Wallis test. Post hoc pairwise comparisons were performed using Dwass, Steel, Chrtchlow-Flinger method. Significance level was set at 0.05.

Results

A total of 5 teeth were treated with each of the three solutions. The WL was re-established in 100% of samples in all 3 groups. Patency was achieved for 100% of samples treated with both acetic acid and phosphoric acid. Patency was not achieved for one tooth treated with chloroform (n=1, 20%) (Table 1).

Table 1: Summary of the ability to regain WL and Patency

	Group 1 (%)	Group 2 (%)	Group 3 (%)
WL Regained	100	100	100
Patency Regained	80	100	100

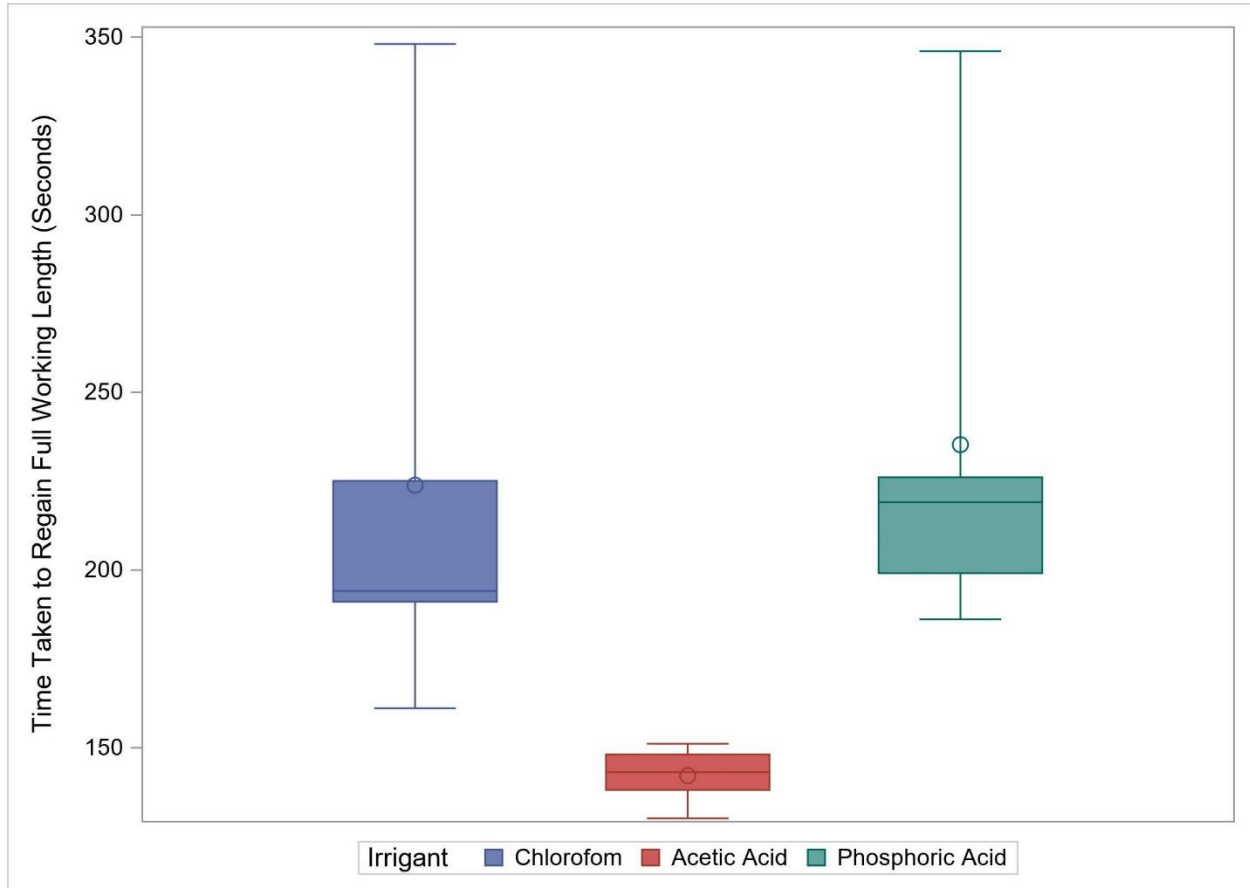
Table 2: Summary of time taken to regain full WL

	Group 1 (sec)	Group 2 (sec)	Group 3 (sec)
1	225	151	219
2	194	148	186
3	348	138	226
4	161	130	346
5	191	143	199

The times required for retreating each sample are summarized in Table 2. The median procedure time was 143 seconds for acetic acid, 194 seconds for chloroform, and 219 seconds for phosphoric acid. Statistically, the differences in procedure time were significantly different across the three irrigants (p-value=0.0087) (Figure 3). The procedure time for acetic acid was significantly faster than both chloroform (p-value=0.0245) and phosphoric acid (p-

value=0.0245). Procedure time was not significantly different between chloroform and phosphoric acid (p-value=0.8605).

Figure 3: Boxplot displaying differences in mean and median times to reach WL



Discussion

This study aimed to evaluate and compare the effectiveness of acidic solutions and chloroform on the solubility and removal of set BC Sealer. The results of this study demonstrated the effectiveness of acetic acid in the removal of EndoSequence BC Sealer during non-surgical root canal retreatment. When BC Sealer was used to create a 3 mm apical plug in combination with a single cone gutta percha technique, 5% acetic acid allowed the reestablishment of the original working length in significantly less time than chloroform or 37% phosphoric acid ($p\text{-value} < 0.05$). Furthermore, 5% acetic acid allowed for greater consistency in achieving WL, demonstrated by the small variance and nearly normal distribution of data. While the effect of 5% acetic acid on the solubility and hardness of set BC Sealer was not directly measured in this study, the solvent allowed for the operator to remove BC Sealer with subjectively less apical force and fewer cycles of instrumentation, and thus with less stress on rotary instrumentation and less removal of dentin.

Previous studies have evaluated the retreatment of canals filled with gutta percha and BC Sealer by measuring the ability to achieve apical patency and the time and ability to reach the original instrumented working length. Hess et al. compared the retreatment of canals filled with AH Plus and EndoSequence BC Sealer with gutta percha placed to or 2 mm short of the instrumented WL, using chloroform, hand and rotary files, and heat. Their results differ from this study as WL was not achieved in 70% of specimen with the master cone placed short of WL,

and patency was not established in 20% and 70% of specimen with the master cone placed to and short of WL respectively (38). In contrast, Agrafioti et al. found that patency and original WL were achieved in all cases obturated and retreated in a similar manner to Hess et al. (39). However, time to attain WL was significantly longer in all groups filled with BC Sealer, particularly in those groups with gutta percha placed short of WL. These differences in achieving WL and patency may be due to differences in the root canal anatomy of the teeth treated. Hess et al. used the mesiobuccal roots of mandibular molars, which exhibit more complex anatomy that may make it more difficult to attain patency. The teeth treated in this study were all single rooted, anterior teeth with minimal curvature.

Chloroform is a common solvent used during retreatment that is safe and effective in dissolving gutta percha and some traditional sealers (18,46,47). Its effectiveness on the removal of BC Sealer is still equivocal. One study found that the use of chloroform did not increase the removal of BC Sealer (39). However, Oltra et al. found that the use of chloroform resulted in less residual BC Sealer in the coronal third of canals and allowed patency to be regained in a significantly greater percentage of teeth when compared to teeth in which chloroform was not used (40). In this study, the use of chloroform allowed instrumentation to the original WL in 100% of cases, and patency to be achieved in 80% of cases. However, original WL and patency were achieved in 100% of cases in the other two groups. Thus, chloroform is not more effective than acetic acid or phosphoric acid in removing BC Sealer. In fact, acetic acid allowed for significantly faster instrumentation to the original WL than chloroform.

Phosphoric acid is commonly used as an etchant prior to bonding procedures. It was included in this study to determine whether it is effective in removing set BC Sealer prior to composite resin bonding to root dentin after root canal treatment, such as when placing a fiber

post. Although further research is necessary, the data in this study suggest that phosphoric acid is not more effective than acetic acid or chloroform in removing BC Sealer. As BC Sealer is able to penetrate up to 2 mm into dentinal tubules and it may not be effectively removed by phosphoric acid, its use may inhibit bonding of composite resin restorations and posts (45).

No published study at this time has directly assessed the effectiveness of 5% acetic acid in the removal of BC Sealer during root canal retreatment. While these data demonstrate the effectiveness of the solution, there are limitations in the research design. Firstly, this study demonstrated the effectiveness of 5% acetic acid in regaining WL and apical patency, but did not assess the total remaining filling material after retreatment. Furthermore, the effectiveness of 5% acetic acid was only assessed in areas where hand and rotary files were able to reach, but not in dentinal tubules, lateral canals, isthmuses, and ramifications. Finally, this study did not assess the effect acetic acid has on the physical properties of root dentin, nor which concentration of acetic acid is most effective.

Future research on 5% acetic acid should assess its effectiveness in the total removal of BC Sealer both in the main canal and in areas that rotary instruments are not effective through the use of sectioning and light microscopy or SEM, or the use of micro-CT imaging. The concentration of acetic acid which is most effective should be assessed, along with the effect of acetic acid on root dentin. Finally, teeth with multiple roots and more complex anatomy than single rooted anterior teeth should be evaluated.

Conclusion

This study demonstrated that 5% acetic acid allowed for the re-establishment of WL and patency significantly faster and more consistently than chloroform or 37% phosphoric acid during the retreatment of canals filled with BC Sealer. 5% acetic acid is a readily available, cost effective solution that has the potential for use in the retreatment of canals obturated using gutta percha and BC Sealer. Further research on the effect of acetic acid on the physical properties of root dentin and the ideal concentration for use during retreatment is necessary prior to use *in vivo*.

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