In Vitro Comparison of the Push-Out Bond Strength of Three Endodontic Sealers With and Without Amoxicillin

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IN VITRO COMPARISON OF THE PUSH-OUT BOND STRENGTH OF THREE ENDODONTIC SEALERS WITH AND WIHOUT AMOXICILLIN

by

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ABSTRACT

IN VITRO COMPARISON OF THE PUSH-OUT BOND STRENGTH OF THREE ENDODONTIC SEALERS WITH AND WIHOUT AMOXICILLIN

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INTRODUCTION: The purpose of this in vitro study was to compare the push-out bond strengths of three endodontic sealers with and without amoxicillin. **METHODS**: Thirty single-rooted extracted human teeth were used for this study. Each tooth was instrumented and irrigated with 5.25% NaOCl and 17% EDTA. The teeth were then divided into six test groups. Group 1-gutta percha (GP)/AH Plus®, group 1a-GP/AH Plus® with 10% by weight amoxicillin, group 2-GP/Pulp canal sealer EWT™, group 2a-GP/Pulp canal sealer EWT™ with amoxicillin, group 3-Resilon® /RealSeal SE™, and group 3a-Resilon® /RealSeal SE™ with amoxicillin. After the sealer was set the entire root was sectioned into 1mm thick slices. A push-out bond strength test was preformed using a universal testing machine. The Mann Whiney and Student's t-test were used to compare the sealer bond strength within the specific sealer test groups overall and within each sealer at apical, middle and coronal root levels. The film thickness, dimensional change and the solubility of each sealer were also tested according to ISO standards. **RESULTS**: There was no significant difference between test groups within each sealer, Group 1 vs. Group 1a ($p=.85$), Group 2 vs. Group 2a ($p=.59$) or Group 3 vs. Group 3a ($p=.52$). There was no significant difference ($p>.05$) in push-out bond strength within each sealer with or without amoxicillin at the same root level. Significant differences were seen in film thickness and dimensional change when amoxicillin was added to the sealers. **CONCLUSION**: The addition of 10% by weight of amoxicillin does not significantly ($p>.05$) change the overall push-out bond strength of GP/AH Plus®, GP/Pulp canal sealer EWT™ and Resilon® /RealSeal SE™ or when compared at the apical, middle and coronal tooth level.

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Introduction

The primary objective of non-surgical endodontic therapy is the thorough chemomechanical cleaning and shaping of the canal system followed by complete three-dimensional obturation (1). Endodontic failure is primarily caused by the persistence of bacteria and bacterial byproducts in the root canal system (2-3). Bacteria may be present from the incomplete cleaning and shaping of the system or due to leakage from the oral cavity. Three-dimensional obturation of the radicular space is essential to long-term success (4). This prevents both the leakage of bacteria and their byproducts into the periapical tissues and entombs any bacteria and byproducts not removed during cleaning and shaping of the canal system. Traditionally obturation has been accomplished with the use of an endodontic sealer and gutta-percha. More recently resin-based sealers and resin-based obturation materials have been introduced as an alternative for endodontic obturation (5-6). Regardless of the obturation system used, failures do occur due to bacteria remaining in the root canal system (7). One recently introduced strategy to address the bacterial etiology of endodontic failure is the use of endodontic sealers with an added antibiotic (8-9).

An ideal endodontic sealer would exhibit antimicrobial properties (10). Baer et al (9) showed that the addition of amoxicillin to endodontic sealers is effective in eradicating *E. faecalis* for at least 7 days after mixing. The addition of amoxicillin to endodontic sealers could change their ability to seal the canal system. One model used to determine the sealing ability of an endodontic sealer is to test the micro push-out bond strength (11-14).

The purpose of this in vitro study was to compare the push-out bond strengths of three endodontic sealers with and without embedded amoxicillin. The null hypothesis tested was that there is no significant difference in the push-out bond strength within specific sealers with and without amoxicillin overall or when compared at the same tooth level (apical, middle, coronal). The alternative hypothesis is that the addition of amoxicillin will decrease the push-out bond strength of the tested sealers. Additional observations about film thickness, dimensional change and solubility were made based on ISO testing.

Materials and Methods

The study was granted exempt status by the Marquette University Institutional Review Board. Thirty single-rooted extracted human teeth with curvatures less than 20° were used in this study. All teeth were initially disinfected by immersion in 5.25% sodium hypochlorite for 6 hours followed by storage in sterile saline. Residual debris was removed from the root surfaces with the use of periodontal curettes. Each tooth was decoronated with a high-speed carbide bur with water spray. Working length was established by passing a 15 K-file (Densply Maillefer, Tulsa, OK) to the apex as observed under 4.8X magnification and subtracting 1mm. A glide path was created using hand files up to a 20(.02). The canals were instrumented using RaCe (Brasseler, Savannah, GA) 35 (.08) and 40 (.10) files in the coronal and middle thirds and EndoSequence nickel-titanium rotary instruments (Brasseler, Savannah, GA) in the apical third. The canals were instrumented to a final apical size of 40(.06) using no less than 6mL of 5.25% sodium hypochlorite to irrigate. A final rinse of 6mL of 17% EDTA over a 1 minute time period was accomplished. The canal was then dried with paper points and the teeth randomly assigned to one of the six obturation groups.

Group 1: gutta-percha and AH Plus® (AHP).

Group 1a: gutta-percha and AH Plus® with the addition of amoxicillin.

Group 2: gutta percha and Pulp Canal Sealer EWT™ (EWT).

Group 2a: gutta percha and Pulp Canal Sealer EWT™ with the addition of amoxicillin.

Group 3: Resilon® and RealSeal SE™ (RS) sealer.

Group 3a: Resilon® and RealSeal SE™ with the addition of amoxicillin.

Each obturation group consisted of five roots obturated using the specified gutta-percha or Resilon and sealer combination, using a warm vertical compaction technique. Each plain sealer was mixed and used according to the manufacturer's instructions. The sealers with amoxicillin were mixed initially according to manufacturer's instructions followed by the addition of 10% by weight of crushed amoxicillin (TEVA Pharmaceuticals, Sellersville, PA) as described by Baer (9).

In the teeth obturated with gutta-percha the master cone was coated with sealer and placed into the canal at working length with tug back. The cone was removed, more sealer applied and the cone placed to working length again. In order to ensure adequate sealer was present in the canal, this was repeated until a small extension of sealer was visualized at the apex. A System B (Analytic, Redmond, WA) heated plugger, set at 200^0 C was used to burn down the master cone leaving 4mm of gutta-percha and sealer at the apex. A paper point was used to coat the canal walls with sealer and the canal was backfilled with warm gutta-percha dispensed from an Obtura II (Obtura Spartan, Fenton, MO). Firm apical pressure was placed on the coronal gutta-percha with a large plugger as it was allowed to cool in the canal. The teeth obtuated with Resilon and Real Seal SE were completed using the same procedure as above, with the exception that the System B was set at 150^0 C and the Obtura II was set to 125^0 C as specified by the manufacture.

The coronal aspect of each tooth was sealed with 1mm of Fuji IX (GC Corp, Tokyo, Japan) glass ionomer.

The obturated roots were stored at 37^{o}C in 100% humidity for 14 days to allow for sealer set. The roots were then sectioned perpendicular to the long axis into 1mm thick slices by using an Isomet saw (Buehler Ltd, Lake Bluff, IL) with water lubrication. At this time the apical side of the disk was marked to ensure the plunger of the push-out test pushed from the apical to coronal direction, to avoid any interference owing to root canal taper. Slices from all six groups were collected from the apical, middle and coronal thirds. Slices containing filling material of a noncircular shape were discarded to avoid non-uniform stress distribution during testing and inaccurate measurements. The thickness of each disk, at the filling material dentin interface, was determined using a digital caliper to the nearest 0.01mm. The diameter of the filling material within each disk was determined using a digital caliper under 4.8x magnification. One of five stainless steel cylindrical plungers that most closely matched the diameter of the filling material without contacting dentin was connected to a universal testing machine (Instron Corp, Norwood, MA). A vertical load was applied to the obturation material in an apical to coronal direction at the rate of 0.5mm/min. A load versus time curve was plotted in real time by a software program connected to the universal testing machine. Failure of bond was determined when a sharp decline was observed on the graph. On completion of push-out testing each specimen was examined under 4.8x magnification. If any evidence of the plunger scrapping dentin was observed, the specimen was discarded and the data collected was not included in further analysis. The bond strength, expressed in MPa, was calculated by dividing the maximum load in Newtons by the area of the bonded interface. The area of the bonded interface was calculated using the formula, area = $2\pi r$ X h, where π is the constant 3.14 and r and h are the measured radius and height in millimeters of the filling material. Figure 1. shows photographs of a. the prepared slices ready for push-out testing, b. the push-out apparatus used to test bond strengths, and c. an example of a specimen after push-out testing was completed

Statistical analysis was used to determine if there was an overall difference in bond strength between matched obturation groups (Group 1 and 1a, Group 2 and 2a, and Group 3 and 3a) with and without amoxicillin, regardless of tooth level. Analysis was also completed to compare matched obturation groups with and without amoxicillin at the apical, middle and coronal levels of the root. The data was first analyzed using the Shapiro- Wilk test to determine if the data was normally distributed. If the data was normally distributed the independent samples t-test was used to compare the two groups. If the data was not normally distributed the nonparametric alternative to the t-test, the Mann-Whitney test was employed to compare the groups.

Additional observations were made based on ISO testing of the three sealers with and without amoxicillin. Testing was conducted to compare film thickness, dimensional change and solubility of the three endodontic sealers with and without amoxicillin. The study was conducted according to the procedures outlined in ISO standards 6876-7.5, 6876-7.6 and 6876-7.7.

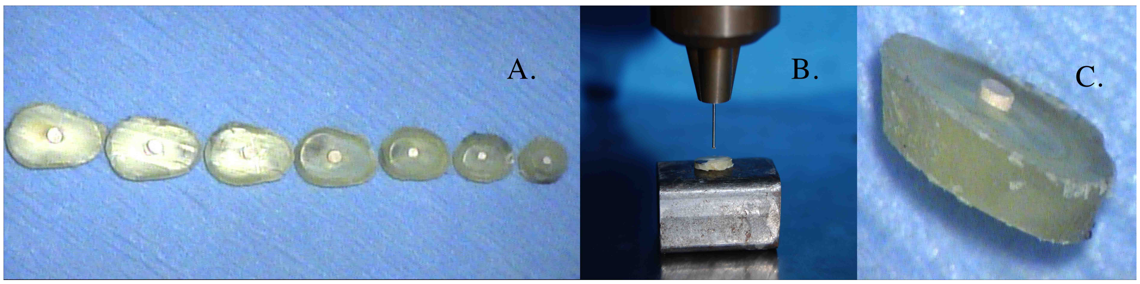

Figure 1. Push-out bond strength testing. (A) 1mm thick specimens (B) Apparatus used to test push-out bond strength (C) Specimen after push-out testing.

Film thickness was tested with two 14mm X 14mm square glass plates with uniform thickness of 2mm. The initial thickness of the two glass plates was measured to the nearest 1 μm using a digital caliper. Each sealer was mixed according to manufacturer's instructions. A portion of each sealer was deposited in the center of one of the glass plates. The other glass plate was placed centrally on the sealer. One hundred eighty seconds after the start of mixing the glass plates were placed in a loading device and a load of one hundred fifty Newtons was delivered vertically on the top of the two plates. Ten minutes after the start of mixing the sealer, the load was removed and the thickness of the plates was measured to the nearest 1 μm using a digital caliper. This procedure was repeated five times for each sealer test group. The film thickness was calculated by determining the difference between the plates with and without sealer.

The dimensional change was tested by using five brass split cylindrical moulds, having internal diameter of 6mm and height of 12mm were used to prepare five samples of each sealer test group for testing. Each sealer test group was prepared as described above. Each mould was slightly overfilled with the sealer to be tested and a polyethylene sheet was placed over the mould. A glass plate was placed over the polyethylene sheet and the mould and plate were held firmly together with a C-clamp. Five minutes after the start of mixing the sealer, the mould with sealer, glass plate and clamp was placed in 100% relative humidity at 37^0 C for 24 hours to allow for sealer set. After the set of the sealer specimens was confirmed, the specimens were removed from the moulds and the ends of each specimen ground down using fresh 600 grit wet sandpaper to ensure a flat surface for measurement. The distance between the flat ends was measured to an accuracy of 1μm. The specimens were then stored in distilled water at 37^0C for thirty days. After thirty days, the specimens were remeasured to an accuracy of 10μm. The change in length was calculated as a percentage of the original length.

Solubility was tested by using four split ring Teflon moulds with internal diameter of 20mm and height of 1.5mm to prepare six specimens of each sealer test group. Each sealer test group was prepared as described above. The mould was placed on a glass plate and filled to slight excess with the sealer to be tested. Another glass plate faced with a sheet of plastic was placed on top of the sealer to create a uniformly flat surface. The glass plate was removed, and the moulds with sealer were stored at 100% relative humidity at 37^0 C for 24 hours. Sealer set was confirmed and specimens removed from the moulds. Two specimens of each sealer test group were placed in pre-weighed (accurate to the nearest .001g) plastic dishes so that they did not touch. Fifty mL of deionized water was added to the dishes. The dishes were covered and stored at 37^0C for twenty-four hours. The specimens were then removed and washed with 3mL of fresh deionized water, recovering the washings in the dish. The specimens were discarded and the water in the dishes was evaporated. Each dish was then re-weighed, accurate to the nearest .001g. The difference between the original mass of the dish and the final mass was recorded to the nearest 0.001g. The difference in mass was calculated as a percentage of the original combined mass of the two specimens to the nearest 0.1%. The test was completed three times for each test group.

Results of the ISO testing were analyzed using the independent sample t-test to compare sealers with and without amoxicillin. It was also determined if the sealers did or did not meet the stated ISO standard.

Results

Five teeth were prepared for each test group. Approximately 9 slices were obtained from each tooth. After elimination of slices due to irregular canal shape or impingement of the dentin by the plunger a total of 39 slices for group 1, 45 slices for group 1a, 42 slices for group 2, 41 slices for group 2a, 39 slices for group 3 and 44 slices for group 3a were obtained for analysis. Push-out bond strength was recorded in MPa. When comparing the match test groups it was found that the data in every case was not normally distributed, thus the Mann-Whitney test was used to make comparison. The mean push-out bond strengths of each group were: Group 1- 4.66±3.22 MPa, Group 1a- 4.34±2.50 MPa, Group 2- 2.13±2.01 MPa, Group 2a- 2.00±2.03 MPa, Group 3-2.49±2.25 MPa and Group 3a-2.18±2.08 MPa. There was no significant difference between test groups within each sealer, Group 1 vs. Group 1a ($p=.85$), Group 2 vs. Group 2a ($p=.59$) or Group 3 vs. Group 3a ($p=.52$).

There was no significant difference ($p>.05$) in push-out bond strength within each sealer with or without amoxicillin when comparison was made at the same root level. The data for AHP at the apical level was normally distributed and the independent sample t-test analysis showed no significant difference between specimens with or without amoxicillin ($p=.16$). The data for AHP at the middle level was normally distributed and the independent sample t-test analysis showed no significant difference between specimens with or without amoxicillin ($p=.21$). The data for AHP at the coronal level was normally distributed and the independent sample t-test analysis showed no significant difference between specimens with or without amoxicillin (p=.86). The data for EWT at the apical level was not normally distributed and the Mann-Whitney test showed no significant difference between specimens with or without amoxicillin (p=.91). The data for EWT at the middle level was not normally distributed and the Mann-Whitney test showed no significant difference between specimens with or without amoxicillin (p=.29) The data for EWT at the coronal level was not normally distributed and the Mann-Whitney test showed no significant difference between specimens with or without amoxicillin (p=.45). The data for RS at the apical level was not normally distributed and the Mann-Whitney test showed no significant difference between specimens with or without amoxicillin (p=.06). The data for RS at the middle level was normally distributed and the independent sample t-test analysis showed no significant difference between specimens with or without amoxicillin (p=.06). The data for RS at the coronal level was not normally distributed and the Mann-Whitney test showed no significant difference between specimens with or without amoxicillin (p=.74). Figures 2-4. show graphs comparing push-out bond strengths of the three sealer at the apical, middle and coronal sections with and without amoxicillin.

Results of the ISO testing are reported in Table 1. The difference in mean film thickness of AHP (31µm) and AHP with amoxicillin (65µm) was found to be significantly different (p=.01). The difference in mean film thickness of EWT (53µm) and EWT with amoxicillin (55µm) was found not to be significantly different (p=1). The difference in mean film thickness of RS (35µm) and RS with amoxicillin (62µm) was found to be significantly different (p=.01).

The difference of dimensional change of AHP was found to be .28% expansion without amoxicillin and .32% expansion with amoxicillin. This was found not to be statistically significant (p=.14). The difference of dimensional change of EWT was found to be .66% shrinkage without amoxicillin and .24% shrinkage with amoxicillin. This was found to be statistically significant (p=.01). The difference of dimensional change of RS was found to be 1.59% expansion without amoxicillin and 2.98% expansion with amoxicillin. This was found to be statistically significant (p=.01).

The solubility of AHP was found to be 0.25% without amoxicillin and 0.14% with amoxicillin. This was found not to be significantly different (p=.08). The solubility of EWT was found to be 0.97% without amoxicillin and 0.66% with amoxicillin. This was found not to be significantly different (p=.66). The solubility of RS was found to be 0.08% without amoxicillin and 0.26% with amoxicillin. This was found not to be significantly different (p=.19).

The ISO standard for film thickness is 50µm. AHP met the standard before the addition of amoxicillin, but did not meet the standard after amoxicillin was added. EWT did not meet the standard with our without amoxicillin. RS met the standard before the addition of amoxicillin, but not after amoxicillin was added.

The ISO standard for dimensional change is less than 1.0% shrinkage or less than 0.1% expansion. AHP did not meet this standard with or without amoxicillin. EWT met this standard both with and without amoxicillin. RS did not meet this standard with or without amoxicillin.

The ISO standard for solubility is less than 3% mass fraction. All of the samples tested met the ISO standard. Examples of tested specimens are found in Figure 5.

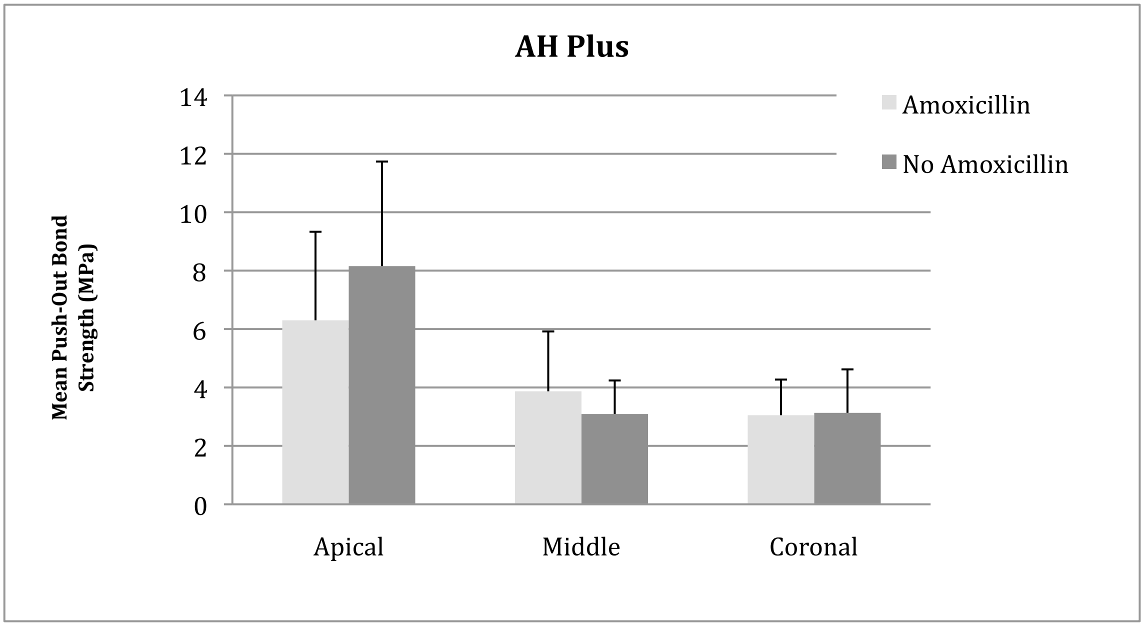

Figure 2. The effects of the addition of amoxicillin to AH Plus® sealer on the mean push-out bond strength at the apical, middle and coronal root sections.

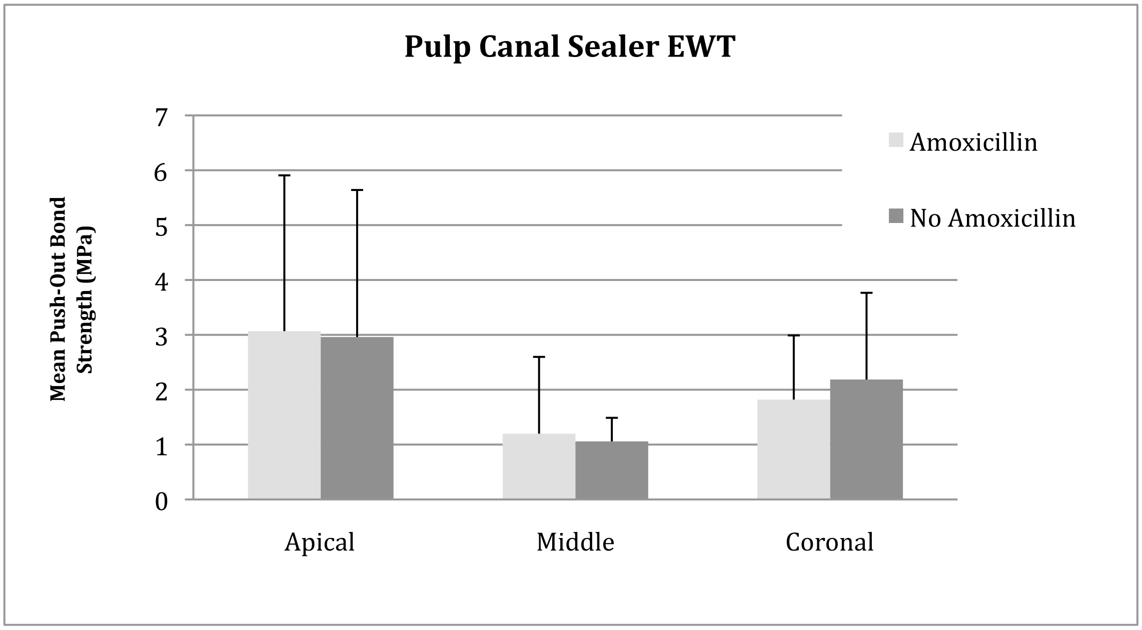

Figure 3. The effects of the addition of amoxicillin to Kerr Pulp Canal Sealer EWT™ sealer on the mean push-out bond strength at the apical, middle and coronal root sections.

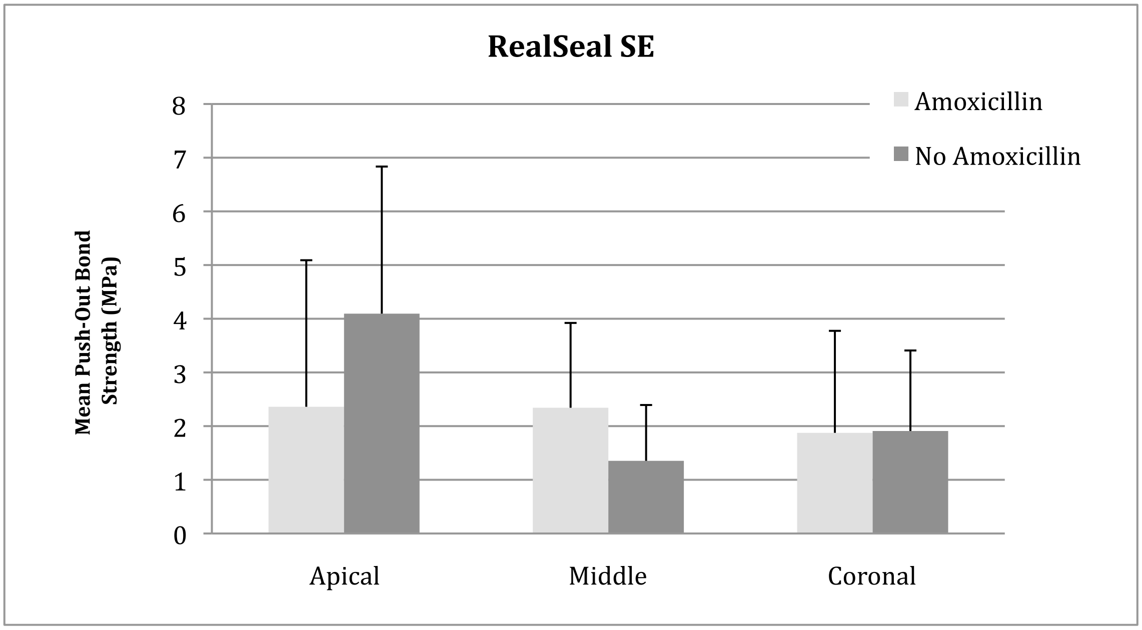

Figure 4. The effects of the addition of amoxicillin to RealSeal SE™ sealer on the mean push-out bond strength at the apical, middle and coronal root sections.

	FILM THICKNESS	**DIMENSIONAL CHANGE**	**SOLUBILITY**
ISO STANDARD	<50µm	<1% Shrinkage (-) and <0.1% Expansion (+)	<3% Mass Fraction
AH Plus	31µm*	+0.28%	0.25%
AH Plus and Amoxicillin	65µm *	+0.32%	0.14%
EWT	53µm	-0.66%*	0.97%
EWT and Amoxicillin	55µm	-0.24%*	0.66%
RS	35µm *	+1.59%*	0.08%
RS and Amoxicillin	62µm *	+2.98%*	0.26%

Table 1. Results of ISO testing. * signifies the matched pair is statistically significantly different. Red cells indicate the measured mean value does not meet the ISO standard.

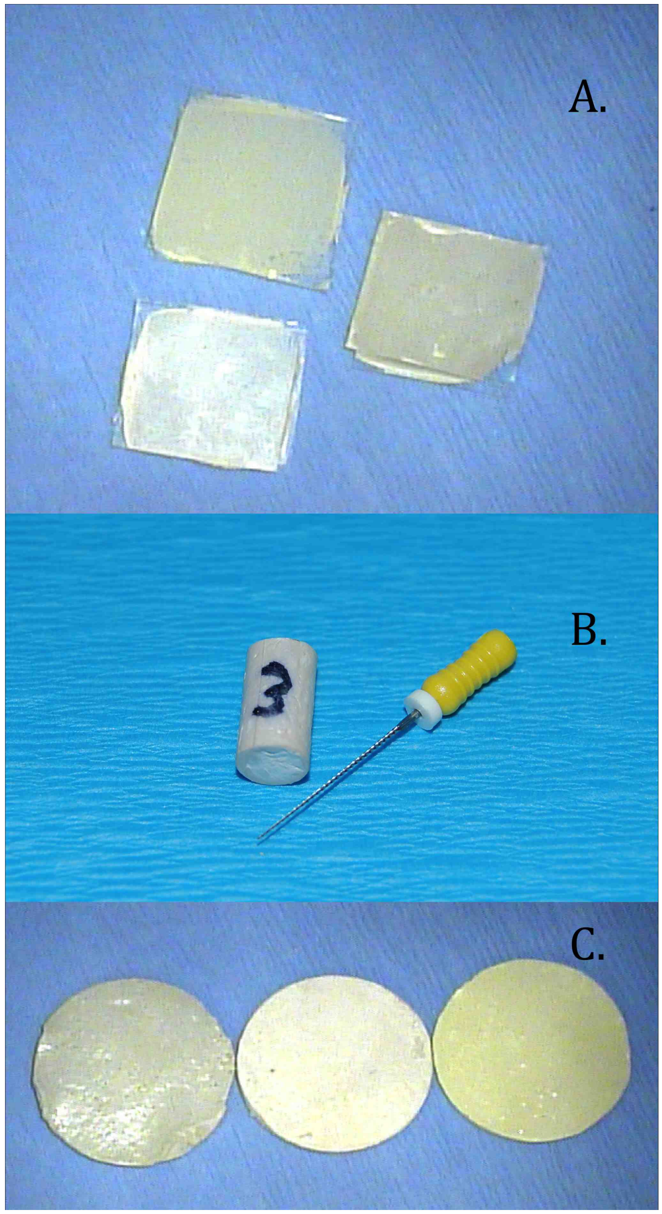

Figure 5. ISO Testing. (A) Film Thickness specimens after testing. (B) Dimensional Change specimen with 21.0mm file for size comparison. (C) Solubility specimens.

Discussion

Antibiotics are commonly used systemically in endodontics(15). The local application of antibiotics to the root canal system and periapical tissues may be a more effective method of delivery (16). A few studies (8,9) have expounded on the concept of endodontic sealers for local antibiotic delivery. Baer et al (9) showed that endodontic sealers with added amoxicillin not only demonstrated inhibition of bacterial cell growth initially, but also demonstrated inhibition after seven days of sealer set. However, the addition of amoxicillin may change the properties of the endodontic sealers. A pilot study was conducted to compare film thickness, dimensional change and solubility of AHP, EWT and RS with and without amoxicillin according to ISO standards. The results of this pilot study suggested further testing of sealers with added amoxicillin should be conducted to more closely mimic the clinical environment.

The push-out bond strength test is one way to evaluate the effectiveness of an endodontic obturation material or technique. Other methods of testing include bacterial leakage, fluid filtration and dye penetration testing (17). While each method of *in vitro* testing seeks to replicate the clinical environment the correlation between leakage studies and clinical success has been questioned (17-21). The push-out model has been used extensively to evaluate the dentin obturation interface, but its relevance has also been called into question (22). There is no evidence that any of these testing methods is the best for measuring the clinical effectiveness of an endodontic obturation material or technique.

The results of this study are in agreement with the results of many other studies comparing the bond strength of gutta-percha and Resilon® to dentin using different sealers (11-14). Generally these studies have shown gutta-percha and AHP to have higher bond strengths than Resilon® and gutta-percha EWT obturation systems. The current study made no attempt to compare push-out bond strengths between different sealers with amoxicillin. The well-established differences in the bond strengths between plain sealers (11-14) suggest any differences in push-out bond strength found between different sealers with added amoxicillin would be due to the inherent differences in the plain sealers push-out bond strength.

The present study demonstrated that the highest mean bond strengths were found at the apical segment of each test group. This could be due to pooling of sealer at the apical segment as evident by the extrusion of sealer through the apex during obturation. Each tooth was prepared so tug-back was felt when placing the master cone. This demonstrates a tight fit that could influence push-out bond strength.

The apical segments exhibited the highest standard deviation. This may be due to the small diameter of the obturation material found near the apex. Attempts were made to match the diameter of the plunger used in the push out testing to the diameter of the filling material to eliminate the plunger touching the wall of the canal. All the slices were examined under 4.8X magnification after push-out. If evidence was found that the canal wall was touched the sample was discarded. It is possible that errors in sample elimination occurred. The middle and coronal slices were more easily aligned with the push-out plungers minimizing the need to discard slices. The overall standard deviations in this study ranged from 57% to 101% of the mean push out bond strength. This is comparable to other push-out studies (11-14) that demonstrate standard deviations that range from 31% - 127% of the mean push-out bond strength.

The ISO testing showed mixed positive and negative results when adding amoxicillin to the three sealers. There were multiple instances where the addition of amoxicillin significantly changed the film thickness and dimensional change, but only the addition of amoxicillin to both AHP and RS negatively affected the film thickness to the point that AHP and RS with amoxicillin did not meet the ISO standard. EWT was positively affected by adding amoxicillin. The shrinkage of EWT with amoxicillin was significantly less than the shrinkage of EWT without amoxicillin. There were no changes in solubility when amoxicillin was added to the three sealers. The results of the push-out bond strength tests suggest that the changes in film thickness and dimensional change do not negatively affect the dentin/sealer interface.

This study represents a step in the process of evaluating the effectiveness of endodontic sealers as a vehicle for the local delivery of antibiotics. While this study demonstrates the push-out bond strength is not significantly ($p > 0.05$) decreased by adding amoxicillin further clinical testing should be completed to explore the clinical effectiveness of sealers with added amoxicillin. Is the dose of amoxicillin high enough to evoke an allergic response? Can the long-term use of sealer/antibiotic combinations lead to bacterial resistance? How long will the sealer maintain it's antimicrobial properties when used clinically? What are the implications of a sealer/antibiotic "puff" in the periapical tissues? These and other questions must be answered before the clinical use of a sealer with added antibiotic can be advised.

In conclusion, this study demonstrated that the addition of 10% by weight of amoxicillin does not significantly ($p>.05$) change the overall push-out bond strength of GP/AHP, GP/EWT and Resilon® /RS or when compared at the apical, middle and coronal tooth level. Therefore the null hypothesis is accepted and the alternative hypothesis is rejected.

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APPENDIX

Push-out bond strength data:

TOOTH	REGION	SLICE	SEALER	Amox?	WIDTH	DIAM. (MM)	LOAD kgf	BOND STRENGTH (NEWTONS)
1	APICAL	1	REAL SEAL	N	1.35	0.75	1.271	3.920
1	APICAL	2	REAL SEAL	N	0.96	0.8	0.027	0.110
1	APICAL	3	REAL SEAL	N	1.28	0.85	1.72	4.937
1	MIDDLE	4	REAL SEAL	N	1.02	0.65	0.0028	0.013
1	MIDDLE	5	REAL SEAL	N	1.01	0.75	0.0236	0.097
1	MIDDLE	6	REAL SEAL	N	1.07	1	0.194	0.566
1	CORONAL	7	REAL SEAL	N	1.08	1	0.826	2.388
1	CORONAL	8	REAL SEAL	N	1.11	1.05	0.041	0.110
1	CORONAL	9	REAL SEAL	N	1.05	1.2	1.864	4.620
2	APICAL	1	REAL SEAL	N	1.13	0.8	1.57	5.424
2	APICAL	2	REAL SEAL	N	0.99	0.6	1.43	7.518
2	APICAL	3	REAL SEAL	N	1.09	0.5	0.2747	1.574
2	MIDDLE	4	REAL SEAL	N	1.11	0.75	0.482	1.808
2	MIDDLE	5	REAL SEAL	N	1.07	0.8	0.773	2.820
2	MIDDLE	6	REAL SEAL	N	1.06	0.9	0.849	2.779
2	CORONAL	7	REAL SEAL	N	1	1	1.007	3.145
2	CORONAL	8	REAL SEAL	N	1.08	1.05	1.107	3.049
2	CORONAL	9	REAL SEAL	N	1.05	1.2	0.715	1.772
3	APICAL	1	REAL SEAL	N	1.02	0.5	1.294	7.924
3	APICAL	2	REAL SEAL	N	1.02	0.6	1.8	9.185
3	APICAL	3	REAL SEAL	N	1.16	0.7	1.12	4.307
3	MIDDLE	5	REAL SEAL	N	1.06	0.8	0.1168	0.430
3	MIDDLE	6	REAL SEAL	N	1.02	0.9	0.1612	0.548
3	MIDDLE	7	REAL SEAL	N	1.01	1	0.278	0.860
3	CORONAL	8	REAL SEAL	N	1.11	1.1	0.296	0.757
3	CORONAL	9	REAL SEAL	N	1.03	1.2	0.169	0.427
4	APICAL	1	REAL SEAL	N	1.09	0.6	0.345	1.647
4	APICAL	2	REAL SEAL	N	1.05	0.75	0.819	3.248
4	APICAL	3	REAL SEAL	N	1.03	0.9	0.548	1.846
4	MIDDLE	4	REAL SEAL	N	1.1	0.9	0.685	2.161
4	MIDDLE	5	REAL SEAL	N	1.05	1	0.804	2.391
4	CORONAL	6	REAL SEAL	N	1.1	1.2	0.078	0.185
4	CORONAL	7	REAL SEAL	N	1.04	1.225	1.33	3.260

5	APICAL	1	REAL SEAL	N	1.02	0.5	0.744	4.556
5	APICAL	2	REAL SEAL	N	1.07	0.75	0.29	1.129
5	MIDDLE	4	REAL SEAL	N	1.1	0.85	0.758	2.532
5	MIDDLE	5	REAL SEAL	N	1	0.9	0.435	1.509
5	MIDDLE	6	REAL SEAL	N	1.01	1	0.139	0.430
5	CORONAL	7	REAL SEAL	N	1.11	1.25	0.571	1.285
6	APICAL	1	EWT	N	1.18	0.6	0.768	3.388
6	APICAL	2	EWT	N	1	0.7	0.379	1.691
6	APICAL	3	EWT	N	1.06	0.75	0.373	1.465
6	MIDDLE	4	EWT	N	1.05	0.75	0.162	0.642
6	MIDDLE	5	EWT	N	1.08	0.8	0.244	0.882
6	MIDDLE	6	EWT	N	1.08	1	0.2518	0.728
6	CORONAL	7	EWT	N	1.07	1	0.428	1.249
6	CORONAL	8	EWT	N	1.05	1.1	1.303	3.523
7	APICAL	1	EWT	N	1.13	0.5	0.36	1.990
7	APICAL	2	EWT	N	1.08	0.6	0.454	2.188
7	APICAL	3	EWT	N	1.12	0.75	0.336	1.249
7	MIDDLE	4	EWT	N	1	0.85	0.341	1.253
7	MIDDLE	5	EWT	N	1.03	1	0.175	0.531
7	CORONAL	6	EWT	N	1.08	1.2	0.891	2.147
7	CORONAL	7	EWT	N	1.05	1.3	0.426	0.975
7	CORONAL	8	EWT	N	1.04	1.5	0.515	1.031
8	APICAL	1	EWT	N	1.32	0.5	1.62	7.665
8	APICAL	2	EWT	N	1.06	0.55	0.507	2.716
8	APICAL	3	EWT	N	1.07	0.6	0.864	4.203
8	APICAL	4	EWT	N	1.01	0.8	0.594	2.296
8	MIDDLE	5	EWT	N	1.06	0.8	0.482	1.775
8	MIDDLE	6	EWT	N	1.06	0.9	0.426	1.395
8	MIDDLE	7	EWT	N	0.92	1	0.471	1.599
8	CORONAL	8	EWT	N	1.05	1.1	0.195	0.527
8	CORONAL	9	EWT	N	1.09	1.25	1.73	3.965
9	APICAL	1	EWT	N	1.18	0.5	1.385	9.302
9	APICAL	2	EWT	N	1.06	0.55	0.146	0.782
9	APICAL	3	EWT	N	1.06	0.7	0.167	0.703
9	MIDDLE	4	EWT	N	1.09	0.8	0.311	1.114
9	MIDDLE	5	EWT	N	1.07	0.9	0.505	1.638
9	CORONAL	6	EWT	N	1.04	1.1	0.342	0.934
9	CORONAL	7	EWT	N	1.05	1.2	0.654	1.621

9	CORONAL	8	EWT	N	1.07	1.3	2.04	4.580
10	APICAL	1	EWT	N	1.14	0.52	0.0769	0.405
10	APICAL	2	EWT	N	1.13	0.51	1.226	6.644
10	APICAL	3	EWT	N	1.13	0.6	0.147	0.677
10	MIDDLE	4	EWT	N	1.03	0.65	0.152	0.709
10	MIDDLE	5	EWT	N	1.05	0.8	0.1888	0.702
10	MIDDLE	6	EWT	N	1.06	1	0.2659	0.783
10	CORONAL	7	EWT	N	1.1	1	0.023	0.065
10	CORONAL	8	EWT	N	1.03	1.1	1.236	3.407
10	CORONAL	9	EWT	N	0.93	1.2	1.57	4.393
11	APICAL	1	AH	N	1.12	0.51	1.994	10.902
11	APICAL	2	AH	N	1.01	0.7	1.33	5.875
11	MIDDLE	3	AH	N	1.02	0.75	0.1698	0.693
11	MIDDLE	4	AH	N	1.1	0.81	1.316	4.613
11	MIDDLE	5	AH	N	1.04	1	0.695	2.087
11	CORONAL	6	AH	N	1.02	1.15	1.165	3.102
11	CORONAL	7	AH	N	1.08	1.25	1.015	2.348
12	APICAL	1	AH	N	1.1	0.45	2.15	13.564
12	APICAL	2	AH	N	1.09	0.5	2.629	15.065
12	APICAL	3	AH	N	1.13	0.6	1.549	7.135
12	MIDDLE	4	AH	N	1.06	0.7	0.792	3.333
12	MIDDLE	5	AH	N	1.06	0.95	1.019	3.160
12	MIDDLE	6	AH	N	1.04	0.75	0.881	3.527
12	CORONAL	7	AH	N	1	1.1	1.61	4.571
12	CORONAL	8	AH	N	1.09	1.15	1.925	4.796
13	APICAL	1	AH	N	1.09	0.5	1.56	8.939
13	APICAL	2	AH	N	1.06	0.7	1.1	4.630
13	APICAL	3	AH	N	1.14	0.8	1.074	3.678
13	MIDDLE	4	AH	N	1.01	0.8	1.087	4.201
13	MIDDLE	5	AH	N	1.04	1	0.7865	2.362
13	MIDDLE	6	AH	N	1.15	1.2	1.718	3.888
13	CORONAL	7	AH	N	1	1.15	0.73	1.982
13	CORONAL	9	AH	N	1.19	2.5	0.759	0.797
14	APICAL	1	AH	N	1.11	0.52	1.671	9.041
14	APICAL	2	AH	N	1.05	0.7	1.017	4.321
14	MIDDLE	3	AH	N	0.95	0.75	0.4066	1.782
14	MIDDLE	4	AH	N	1.13	0.85	1.208	3.928
14	MIDDLE	5	AH	N	1.1	0.9	1.14	3.596

14	CORONAL	6	AH	N	1.02	1	1.37	4.195
14	CORONAL	7	AH	N	1.06	1.05	1.55	4.349
14	CORONAL	8	AH	N	0.91	1.2	1.266	3.621
15	APICAL	1	AH	N	1.02	0.5	1.25	7.654
15	APICAL	3	AH	N	1.09	0.6	1.477	7.053
15	MIDDLE	4	AH	N	1.04	0.7	1.0776	4.623
15	MIDDLE	5	AH	N	1.09	0.8	0.778	2.786
15	MIDDLE	6	AH	N	1.1	0.9	0.5599	1.766
15	CORONAL	6	AH	N	1.05	1	0.507	1.508
15	CORONAL	7	AH	N	0.95	1.1	0.447	1.336
15	CORONAL	8	AH	N	1.03	1.45	2.37	4.956
16	APICAL	1	REAL SEAL	Y	1.08	0.5	0.0675	0.390
16	APICAL	3	REAL SEAL	Y	0.98	0.52	0.0967	0.593
16	APICAL	3	REAL SEAL	Y	1.07	0.7	0.014	0.058
16	MIDDLE	4	REAL SEAL	Y	1.1	0.8	0.071	0.252
16	MIDDLE	5	REAL SEAL	Y	1.06	0.95	1.199	3.718
16	MIDDLE	6	REAL SEAL	Y	1.05	1.1	0.992	2.682
16	CORONAL	7	REAL SEAL	Y	1.01	1.2	0.179	0.461
16	CORONAL	8	REAL SEAL	Y	1.04	1.25	0.2006	0.482
16	CORONAL	9	REAL SEAL	Y	1.07	1.25	0.346	0.808
16	CORONAL	10	REAL SEAL	Y	1.11	1.3	0.269	0.582
17	APICAL	1	REAL SEAL	Y	0.96	0.5	0.571	3.715
17	APICAL	2	REAL SEAL	Y	1.08	0.6	0.268	1.292
17	APICAL	3	REAL SEAL	Y	1.11	0.7	0.0195	0.078
17	APICAL	4	REAL SEAL	Y	1.15	0.85	0.0132	0.042
17	MIDDLE	5	REAL SEAL	Y	1	0.89	0.245	0.860
17	MIDDLE	7	REAL SEAL	Y	1	1	1.407	4.394
17	CORONAL	8	REAL SEAL	Y	1.11	1	0.093	0.262
17	CORONAL	9	REAL SEAL	Y	1.04	1.05	0.0408	0.117
17	CORONAL	10	REAL SEAL	Y	1.05	1.1	2.105	5.692
18	APICAL	1	REAL SEAL	Y	1.09	0.49	0.763	4.461
18	APICAL	2	REAL SEAL	Y	1.08	0.6	0.592	2.853
18	APICAL	3	REAL SEAL	Y	1.09	0.75	0.425	1.624
18	MIDDLE	4	REAL SEAL	Y	1.01	0.75	0.73	3.010
18	MIDDLE	5	REAL SEAL	Y	1.03	0.9	0.875	2.948
18	MIDDLE	6	REAL SEAL	Y	1.1	1.1	0.8387	2.165
18	CORONAL	7	REAL SEAL	Y	1.1	1.2	0.037	0.088
18	CORONAL	8	REAL SEAL	Y	1	1.25	0.0379	0.095
18	CORONAL	9	REAL SEAL	Y	0.96	2.5	1.85	2.407

19	APICAL	1	REAL SEAL	Y	1.11	0.51	0.306	1.688
19	APICAL	2	REAL SEAL	Y	1.09	0.7	0.967	3.958
19	MIDDLE	3	REAL SEAL	Y	1.04	0.75	1.25	5.005
19	MIDDLE	4	REAL SEAL	Y	1.02	0.85	1.025	3.692
19	MIDDLE	5	REAL SEAL	Y	1.08	1	0.277	0.801
19	CORONAL	6	REAL SEAL	Y	1.1	1.25	1.154	2.621
19	CORONAL	7	REAL SEAL	Y	1.16	1.3	2.06	4.266
19	CORONAL	8	REAL SEAL	Y	0.96	1.4	1.87	4.345
20	APICAL	1	REAL SEAL	Y	1.1	0.5	1.808	10.266
20	APICAL	2	REAL SEAL	Y	1.14	0.6	0.443	2.023
20	MIDDLE	3	REAL SEAL	Y	1.01	0.8	0.00711	0.027
20	MIDDLE	4	REAL SEAL	Y	1.1	0.95	0.3021	0.903
20	MIDDLE	5	REAL SEAL	Y	1.1	1	0.821	2.331
20	CORONAL	6	REAL SEAL	Y	1.05	1.1	1.08	2.920
20	CORONAL	7	REAL SEAL	Y	1.06	1.5	2.16	4.242
20	CORONAL	8	REAL SEAL	Y	1	2.5	0.486	0.607
21	APICAL	1	EWT	Y	1.12	0.5	0.232	1.294
21	APICAL	2	EWT	Y	1.1	0.65	0.323	1.411
21	MIDDLE	3	EWT	Y	1.04	0.75	0.938	3.756
21	MIDDLE	4	EWT	Y	1.06	0.9	1.269	4.154
21	MIDDLE	5	EWT	Y	1.03	1	1.054	3.196
21	CORONAL	6	EWT	Y	1.08	1.2	1.035	2.494
21	CORONAL	7	EWT	Y	1.06	1.4	0.902	1.898
21	CORONAL	8	EWT	Y	0.86	1.45	1.29	3.231
22	APICAL	1	EWT	Y	1.07	0.5	1.836	10.717
22	APICAL	2	EWT	Y	1.08	0.6	0.6215	2.995
22	APICAL	3	EWT	Y	1.12	0.75	0.05178	0.193
22	MIDDLE	4	EWT	Y	0.97	0.75	0.0503	0.216
22	MIDDLE	5	EWT	Y	1.04	0.85	0.0728	0.257
22	MIDDLE	6	EWT	Y	1.03	1	0.207	0.628
22	CORONAL	7	EWT	Y	1.05	1.1	0.264	0.714
22	CORONAL	8	EWT	Y	1.11	1.2	0.621	1.456
22	CORONAL	9	EWT	Y	0.72	1.2	0.32	1.157
23	APICAL	1	EWT	Y	1.1	0.55	0.983	5.074
23	APICAL	2	EWT	Y	1.11	0.65	0.315	1.363
23	MIDDLE	3	EWT	Y	1.05	0.75	0.055	0.218
23	MIDDLE	4	EWT	Y	1.07	0.8	0.032	0.117

23	MIDDLE	5	EWT	Y	1.03	0.9	0.0296	0.100
23	CORONAL	6	EWT	Y	1.04	1	0.307	0.922
23	CORONAL	7	EWT	Y	1.13	1.15	0.183	0.440
24	APICAL	1	EWT	Y	1.05	0.49	0.897	5.445
24	APICAL	2	EWT	Y	1.08	0.51	0.836	4.740
24	APICAL	3	EWT	Y	1.01	0.6	0.2137	1.101
24	MIDDLE	4	EWT	Y	1.08	0.75	0.233	0.898
24	MIDDLE	5	EWT	Y	1.11	0.85	0.289	0.957
24	MIDDLE	6	EWT	Y	1.03	1	0.243	0.737
24	CORONAL	7	EWT	Y	1.12	1.1	0.197	0.499
24	CORONAL	8	EWT	Y	0.92	1.3	1.31	3.421
24	CORONAL	9	EWT	Y	1.22	2.25	2.01	2.287
25	APICAL	1	EWT	Y	1.15	0.45	0.3888	2.346
25	APICAL	2	EWT	Y	1.2	0.5	0.2467	1.284
25	APICAL	3	EWT	Y	1.09	0.6	0.3999	1.910
25	MIDDLE	4	EWT	Y	1.09	0.7	0.2011	0.823
25	MIDDLE	5	EWT	Y	1.04	0.8	0.192	0.721
25	CORONAL	7	EWT	Y	1.08	1.2	1.298	3.128
25	CORONAL	8	EWT	Y	1.03	1.25	0.16	0.388
25	CORONAL	9	EWT	Y	1.04	1.5	1.72	3.443
26	APICAL	1	AH	Y	1.12	0.55	1.123	5.693
26	APICAL	2	AH	Y	1.07	0.65	1.137	5.105
26	MIDDLE	3	AH	Y	1.06	0.7	0.56	2.357
26	MIDDLE	4	AH	Y	1.03	0.85	0.358	1.277
26	MIDDLE	5	AH	Y	1.02	1	0.707	2.165
26	CORONAL	6	AH	Y	1.04	1.1	0.699	1.908
26	CORONAL	7	AH	Y	1.06	1.15	0.815	2.088
27	APICAL	1	AH	Y	1.15	0.48	0.235	1.330
27	APICAL	2	AH	Y	1.06	0.5	0.738	4.349
27	APICAL	3	AH	Y	1.05	0.75	0.9857	3.909
27	MIDDLE	4	AH	Y	1.03	0.9	1.769	5.960
27	MIDDLE	5	AH	Y	1.03	1	0.466	1.413
27	MIDDLE	6	AH	Y	1.09	1.1	2.357	6.139
27	CORONAL	7	AH	Y	1.11	1.2	1.19	2.790
27	CORONAL	8	AH	Y	1.07	1.4	1.77	3.690
27	CORONAL	9	AH	Y	0.96	1.8	2.02	3.651
28	APICAL	1	AH	Y	1.14	0.5	0.332	1.819
28	APICAL	2	AH	Y	0.97	0.52	1.81	11.206

28	APICAL	3	AH	Y	0.99	0.7	1.384	6.237
28	MIDDLE	4	AH	Y	1.11	0.75	2.063	7.739
28	MIDDLE	5	AH	Y	1.09	0.85	1.959	6.603
28	MIDDLE	6	AH	Y	1.04	1	1.52	4.564
28	CORONAL	7	AH	Y	1.03	1.2	0.596	1.506
28	CORONAL	8	AH	Y	1.03	1.3	1.63	3.802
28	CORONAL	9	AH	Y	1.09	1.4	1.2	2.456
28	CORONAL	9	AH	Y	1.07	1.7	1.7	2.919
29	APICAL	1	AH	Y	1.02	0.49	1.318	8.235
29	APICAL	2	AH	Y	1.09	0.6	1.3699	6.541
29	APICAL	3	AH	Y	1.16	0.75	2.037	7.312
29	MIDDLE	4	AH	Y	0.97	0.8	1.14	4.588
29	MIDDLE	5	AH	Y	1.02	1	1.12	3.429
29	MIDDLE	6	AH	Y	1.05	1.15	1.07	2.767
29	CORONAL	7	AH	Y	1.05	1.5	0.8	1.586
29	CORONAL	8	AH	Y	0.97	1.6	1.25	2.515
29	CORONAL	9	AH	Y	1.22	1.65	2.25	3.491
30	APICAL	1	AH	Y	1.09	0.45	0.96	6.112
30	APICAL	2	AH	Y	1.1	0.5	2.042	11.595
30	APICAL	3	AH	Y	1.03	0.65	1.878	8.760
30	MIDDLE	4	AH	Y	1.01	0.7	1.03	4.550
30	MIDDLE	5	AH	Y	1.05	0.75	0.391	1.551
30	MIDDLE	6	AH	Y	1.09	0.9	0.922	2.935
30	CORONAL	7	AH	Y	1.05	1	1.33	3.956
30	CORONAL	8	AH	Y	1.03	1.1	1.74	4.796
30	CORONAL	9	AH	Y	1.07	1.4	1.9	3.961
30	CORONAL	10	AH	Y	1.06	1.7	2.13	3.691

ISO testing data:

Film Thickness:

Sealer	Amox?	Specimen#	Initial Thickness (mm)	Final Thickness (mm)	Change (mm)	Average Film Thickness (mm)
AH Plus	N	1	1.98	2.01	0.03	
AH Plus	N	2	1.963	1.996	0.033	
AH Plus	N	3	1.982	2.022	0.04	
AH Plus	N	4	1.973	1.997	0.024	
AH Plus	N	5	1.974	2.004	0.03	0.0314
AH Plus	Y	1	2.002	2.062	0.06	
AH Plus	Y	2	2.002	2.067	0.065	
AH Plus	Y	3	2.017	2.08	0.063	
AH Plus	Y	4	1.99	2.059	0.069	
AH Plus	Y	5	1.979	2.048	0.069	0.0652
EWT	N	1	2.007	2.045	0.038	
EWT	N	2	1.984	2.042	0.058	
EWT	N	3	1.997	2.056	0.059	
EWT	N	4	1.983	2.051	0.068	
EWT	N	5	1.973	2.017	0.044	0.0534
EWT	Y	1	1.98	2.034	0.054	
EWT	Y	2	1.992	2.062	0.07	
EWT	Y	3	1.987	2.024	0.037	
EWT	Y	4	1.962	2.014	0.052	
EWT	Y	5	1.963	2.029	0.066	0.0558
Real Seal	N	1	1.999	2.023	0.024	
Real Seal	N	2	1.995	2.011	0.016	
Real Seal	N	3	1.973	2.054	0.081	
Real Seal	N	4	1.983	2.007	0.024	
Real Seal	N	5	1.982	2.01	0.028	0.0346
Real Seal	Y	1	1.992	2.064	0.072	
Real Seal	Y	2	1.979	2.037	0.058	
Real Seal	Y	3	1.971	2.032	0.061	
Real Seal	Y	4	1.996	2.049	0.053	
Real Seal	Y	5	1.997	2.064	0.067	0.0622

Dimensional Change:

Sealer	**Amox?**	**Specimen #**	**Orig. Length**	**30 Day Length**	**Dimensional Change(mm)**	**Dimensional Change (%)**	**Average Dimensional Change (%)**
AH Plus	N	1	11.624	11.717	0.093	0.800068823	
AH Plus	N	2	11.807	11.829	0.022	0.186330143	
AH Plus	N	3	11.802	11.825	0.023	0.194882223	
AH Plus	N	4	11.733	11.742	0.009	0.076706725	
AH Plus	N	5	11.957	11.979	0.022	0.18399264	0.288396111
AH Plus	Y	1	12.009	12.047	0.038	0.316429345	
AH Plus	Y	2	11.904	11.942	0.038	0.31922043	
AH Plus	Y	3	11.603	11.632	0.029	0.249935362	
AH Plus	Y	4	11.987	12.021	0.034	0.283640611	
AH Plus	Y	5	9.051	9.09	0.039	0.430891614	0.320023472
EWT	N	1	12.002	11.905	-0.097	-0.808198634	
EWT	N	2	12.013	11.954	-0.059	-0.491134604	
EWT	N	3	12.024	11.97	-0.054	-0.449101796	
EWT	N	4	12.019	11.965	-0.054	-0.449288626	
EWT	N	5	12.007	11.872	-0.135	-1.124344133	-0.664413559
EWT	Y	1	12.054	12.036	-0.018	-0.149328024	
EWT	Y	2	12.033	12.009	-0.024	-0.199451508	
EWT	Y	3	12.028	12.017	-0.011	-0.091453276	
EWT	Y	4	11.993	11.945	-0.048	-0.40023347	
EWT	Y	5	12.035	11.99	-0.045	-0.373909431	-0.242875142
Real Seal	N	1	11.628	11.794	0.166	1.427588579	
Real Seal	N	2	11.671	11.883	0.212	1.816468169	
Real Seal	N	3	11.678	11.87	0.192	1.644117143	
Real Seal	N	4	11.652	11.851	0.199	1.707861311	
Real Seal	N	5	11.665	11.824	0.159	1.363051865	1.591817414
Real Seal	Y	1	11.685	11.994	0.309	2.644415918	
Real Seal	Y	2	11.912	12.284	0.372	3.122901276	
Real Seal	Y	3	11.525	11.892	0.367	3.184381779	
Real Seal	Y	4	11.654	12.025	0.371	3.183456324	
Real Seal	Y	5	11.307	11.622	0.315	2.78588485	2.984208029

Solubility:

Sealer	Amox ?	Specimen #	Sample Weight	Empty container	Container + Solute	Solute Weight	% Sealer Lost	Avg. % Lost
AH Plus	N	1	3.420	18.707	18.717	0.010	0.284	
AH Plus	N	2	3.137	14.209	14.217	0.008	0.258	
AH Plus	N	3	3.069	14.179	14.186	0.007	0.227	0.256
AH Plus	Y	1	3.034	14.234	14.238	0.004	0.138	
AH Plus	Y	2	3.157	14.172	14.178	0.007	0.215	
AH Plus	Y	3	3.112	14.212	14.214	0.003	0.080	0.145
EWT	N	1	2.174	149.102	149.134	0.032	1.476	
EWT	N	2	1.794	149.808	149.810	0.002	0.089	
EWT	N	3	2.046	149.675	149.702	0.028	1.365	0.977
EWT	Y	1	0.779	19.067	19.078	0.011	1.360	
EWT	Y	2	2.075	14.185	14.191	0.006	0.275	
EWT	Y	3	2.327	19.647	19.655	0.008	0.360	0.665
Real Seal	N	1	2.436	14.032	14.032	0.001	0.026	
Real Seal	N	2	2.583	14.348	14.354	0.006	0.232	
Real Seal	N	3	2.390	14.370	14.370	0.000	0.012	0.090
Real Seal	Y	1	3.050	14.394	14.401	0.006	0.207	
Real Seal	Y	2	2.876	14.480	14.489	0.008	0.291	
Real Seal	Y	3	2.528	14.167	14.175	0.008	0.312	0.270

Institutional Review board summary:

OFFICE OF RESEARCH COMPLIANCE

MARQUETTE
UNIVERSITY

December 16, 2009

Mr. Brian Kleinman
Dentistry

Dear Mr. Kleinman:

Thank you for submitting your protocol titled, "*Push out bond strength of three endodontic sealers embedded with anoxicillin,*" protocol number HR-1942. On December 14, 2009, the Marquette University Institutional Review Board granted exempt status for this protocol under Exemption Category #4: Collection or Study of Existing Data.

You may proceed with your research. Your protocol has been granted exempt status as submitted. Any changes to your protocol affecting participant risk must be requested in writing by submitting an IRB Protocol Amendment Form (http://www.marquette.edu/researchcompliance/research/irbforms.shtml). These changes must receive IRB review before being initiated, except when necessary to eliminate apparent immediate hazards to the human subjects. If there are any adverse events, please notify the Marquette University IRB immediately.

If you have any questions or concerns, please do not hesitate to contact me. Thank you for your time and cooperation.

Sincerely,

Elizabeth M. McDonough
Research Compliance Officer – Human Subjects and Radiation Safety

cc: Dr. Rebecca Bardwell, IRB Chair
Dr. Jim Bahcall, Dentistry
Ms. Erin Fox, Graduate School

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