6-1999

Effect of Obturation and / or Coronal Seal on the Success of Root Canal Therapy : An In Vivo Histologic Investigation

Douglas Snider

LOMA LINDA UNIVERSITY

GRADUATE SCHOOL

EFFECT OF OBTURATION AND / OR CORONAL SEAL ON THE SUCCESS OF ROOT CANAL THERAPY: AN IN VIVO HISTOLOGIC INVESTIGATION

By

Douglas Snider

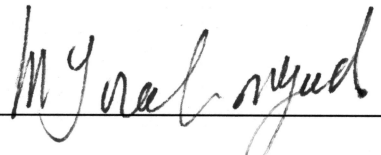
A Thesis in Partial Fulfillment

Of the Requirements for the Degree Master

Of Science in Endodontics

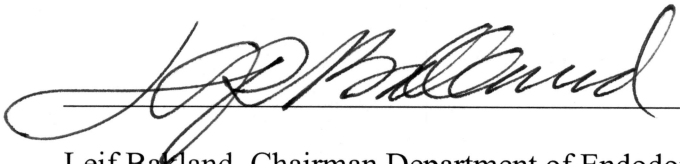
June, 1999

Each person whose signature appears below certifies that this thesis in their opinion is adequate, in scope and quality, as a thesis for the degree Master of Science.

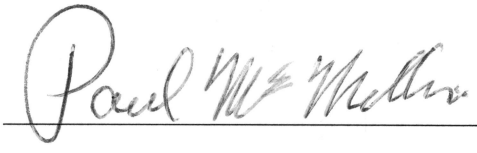


,Chairperson

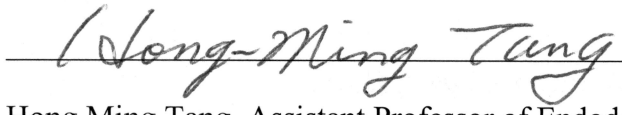
Mahmoud Torabinejad, Professor of Endodontics



Leif Bakland, Chairman Department of Endodontics



Paul Mc Millan, Professor of Anatomy



Hong Ming Tang, Assistant Professor of Endodontics

ACKNOWLEDGMENTS

I would like to express my appreciation to the individuals who helped me complete this study. I want to thank Dr. Bakland and Dr. Torabinejad for their advice and support. I am grateful to Dr. Tang for his hours of help with the animals. I wish to thank Mr. Hough for teaching histologic techniques and Dr. Mc Millan for help with the morphometric analysis.

American Association of Endodontists Foundation and Loma Linda University School of Dentistry supported this work.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABSTRACT

Effect of Obturation and / or Coronal Seal on the Success of Root Canal Therapy: An In Vivo Histologic Investigation

By Douglas Snider

Numerous studies have examined coronal leakage with in vitro protocols. A search of the literature showed the absence of a long term in vivo, histologic study, examining the effect of coronal and/or radicular leakage on periradicular health.

The purpose of this study was to evaluate the effect of obturation and / or coronal seal in preventing the formation of periradicular lesions in dogs over time. The study was conducted on six beagle dogs. Premolars were cleaned and shaped. Sixteen roots in each dog were randomly assigned to one of four treatment groups of equal size. Group one was obturated using warm vertical condensation with sealer, and had MTA placed into the upper two millimeters of the canal and filling the pulp chamber. The second group was obturated and had no coronal restoration placed. Group three was cleansed and shaped, not obturated and had an Intermediate Restorative Material (IRM) filling placed coronally. Group four was cleansed and shaped, not obturated and had Mineral Trioxide Aggregate placed as a coronal seal.

At 6, 12 and 24 weeks post-operatively, two dogs were euthanized at each time period. Six micron thick sagital sections through the roots were taken, and evaluated for the presence of periradicular lesions.

The size of any bony lesion and the degree of inflammation were evaluated microscopically and measured with morphometric analysis. Analysis of variance, with four treatments and three time periods was used.

There was a statistically significant difference (95% confidence level) in the degree of inflammation and the size of the lesions between the two treatments with well sealed coronal access (MTA) and the two groups without permanently sealed accesses. These results confirm the need for, and emphasize the importance of proper restoration of endodontically treated teeth.

LITERATURE REVIEW

ETIOLOGY OF PERIRADICULAR LESIONS

The goal of endodontic therapy has been defined as the elimination and future exclusion of bacteria from the root canal system (Cohen and Burns, 1994). Periradicular inflammatory lesions develop as a sequella to pulpal infection. The elimination or reduction of bacteria, and any antigenic or toxic material, to a physiologically tolerable level will result in healing of the periradicular lesion (Moller et al. 1981). The establishment of an effective barrier to prevent microorganisms or their products from reaching the periradicular tissues is important. The coronal seal of the root canal system, and by extension the periradicular tissue, has been implicated (Moller et al. 1981) as a factor in the long term success of endodontic treatment. Evidence suggests that the exclusion of bacteria is the most important factor in prevention of the periradicular lesions (Kakehashi et al. 1965). Leakage of bacteria occurs clinically, because of a fracture in the tooth, decay or a defective temporary or permanent restoration. Also, important are bacterial metabolites, toxins and degradation products, which are smaller than bacteria and penetrate faster than bacteria. These may also cause pathosis of the periradicular tissues. Leakage and recontamination results in failure of the root canal treatment, necessitating retreatment or periapical surgery (Gish et al. 1994). Weine (1976) has indicated that improper restoration leads to loss of more endodontically treated teeth than actual failure of endodontic therapy.

CORONAL LEAKAGE

The placement of a proper final restoration and its effect on success has been evaluated retrospectively. Swartz et al. (1983) found that the presence of a proper restoration, at recall, revealed a highly significant increase in success ($p = .005$) compared to those not restored (1007 cases). Rapp et al. (1991) reviewed 715 endodontic surgery cases and found that teeth with permanent restorations, at the recall, showed over 69% completely healed. This is significantly better than the 49% of teeth lacking a permanent restoration. A retrospective examination of 1010 endodontically treated teeth radiographically demonstrated that the technical quality of the coronal restoration was significantly more important than the technical quality of the endodontic treatment for apical periodontal health (Ray & Trope 1995). Bergenholtz et al. (1982) determined that oral bacteria penetrate under restorations, resulting in the inflammation of subjacent pulpal tissue. White et al. (1992 & 1994) used in-vitro and in-vivo studies to show that all crown cements tested leaked at the tooth-cement interface within six months.

Most coronal restoration materials have come to endodontics by way of restorative dentistry and, historically, have not been placed by endodontists but by the general dentist. Testing to determine the protection offered by permanent restorations is important. Cox et al. (1987) tested the biocompatibility of surface sealed dental materials such as silicate cement, zinc phosphate cement, amalgam and composites. They concluded that the components of the restorative materials were less significant to

pulpal biocompatibility than its ability to seal out bacteria around the restoration margins.

A report by Chong (1995) presents a case in which undiagnosed coronal leakage resulted in failure in the endodontic management of a tooth. Coronal leakage occurred during root canal treatment as a result of the presence of deficient composite resin fillings and secondary caries. Despite repeated visits of cleaning and dressing, the canal continued to be contaminated and symptoms persisted. On referral, the reason for treatment failure was diagnosed. The tooth was successfully treated by the replacement of the deficient fillings, after the elimination of underlying caries. Symptoms resolved enabling the completion of the root canal treatment.

Dental amalgam continues to be evaluated in an attempt to find a material that will reduce microleakage at the tooth/amalgam interface. Conventional amalgam alloys exhibit leakage at the interface of the cavity margins and the restoration. Copal varnish was recommended prior to placement of the amalgam to prevent initial leakage and was expected to be replaced by amalgam corrosion products and become self-sealing. High copper alloys are now primarily in use and do not readily produce corrosion products. Wright et al. (1991) clinically compared high copper amalgams unlined with those lined with Copalite, using calcium hydroxide as the microleakage detection agent. Copal varnish did not eliminate microleakage or prevent bacterial infiltration.

Dental adhesives have been used to reduce marginal leakage around coronal restorations. The heterogeneous structure of dentin makes dentin bonding difficult. Etching removes the smear layer and improves mechanical retention in two ways.

First, acid etching removes the pellicle, demineralizes the dentin surface and exposes collagen fibers that otherwise are surrounded by hydroxyapatite. The resulting web of exposed collagen can be infiltrated with a primer and bonding agent to form a mechanical bond. Second, acid etching opens and widens dentin tubules and their branches, which, with resin permeation, retains the restoration. Dentin bonding is technique sensitive and no long term results greater than three years are currently available (Soderholm 1997).

Berry et al. evaluated, in vitro, the microleakage of Class I amalgams lined with dentin adhesives without etching (1994) and when using dentin bonding system primers (1996). Copalite was unable to eliminate marginal leakage. The dentin bonding agents alone significantly reduced leakage. However, all etched restorations were significantly more effective in reducing microleakage than the non-etched, no liner or Copalite lined groups.

Barkhordar et al. (1997) compared in vitro sealing performance of a chemically cured and a photopolymerization composite for access cavity restorations in endodontically treated teeth. In addition, the effects of four dentin pretreatments were evaluated. One hundred twenty-eight extracted human incisors were cleansed, shaped and obturated with sealer and gutta percha. The access was temporarily sealed with Cavit and placed in 100% humidity and 37 deg. C for seven days to simulate interappointment conditions. Half of the access cavities within each group were filled with a chemically cured microhybrid composite. The other half were filled with a photopolymerized composite. The teeth were sectioned and evaluated for linear

photochemical dye penetration. The results show that for all cavity treatments the chemically cured composite exhibited less dye penetration than the photopolymerized composite. This result is probably due to the fact that chemical cured composites tend to have less polymerization shrinkage than those photochemically cured. The best barrier was created with All-Bond 2 and Scotchbond Multipurpose. Glass ionomer dentin treatments had significantly more microleakage.

Coronal leakage studies have been pursued with in vitro dye leakage, to evaluate the protection afforded by temporary fillings between appointments and before a permanent restoration can be placed. Two studies, Melton et al. (1990) and Noguera & McDonald (1990), had conflicting findings. The first showed that Cavit sealed more consistently than Term (Temporary Endodontic Restorative Material - a composite like product of urethane dimethacrylate polymer), and the second reported that Term exhibited the least leakage compared to Cavit or IRM or Dentemp an unreinforced ZOE material available over the counter. Materials tested were not consistent in reducing leakage. Uranga et al. (1999) compared temporary filling materials to bonded composite and recommended placing restorative materials and not using temporary filling materials due to coronal leakage

Mineral Trioxide Aggregate (MTA) has recently been developed at Loma Linda University to seal off pathways of communication between the root canal system and the external surface of the tooth. MTA has the following properties: pH of 12.5, mineral ions, long setting time (3 hr.) and 67 mPa compressive strength at 21 days (Abedi & Ingle 1995). The sealing properties of MTA have been evaluated using SEM

on replicas showing better marginal adaptation than amalgam, IRM, or Super EBA (Torabinejad et al. 1995). With in-vitro dye penetration and bacterial invasion studies, Torabinejad et al. (1993,1994,1995) showed that MTA had better sealing ability than amalgam, Super EBA, and other materials.

OBTURATION

The root canal filling is the next victim of coronal bacterial penetration. The advance of infective agents down canals filled with sealer and gutta percha has been measured. Swanson & Madison (1987) evaluated the time that obturated canals could be exposed to artificial saliva before compromising the seal's integrity. After exposing obturated roots to artificial saliva for 7 days, they found that ink penetrated from 79% to 85% of the canal length in all exposed specimens after a 48 hour ink bath. There was no leakage in samples not exposed to the artificial saliva. Madison et al. (1987) ran the same experiment with three different sealers and lateral condensation. They showed that AH26 demonstrated more leakage than Sealapex and Roth's sealers. Madison & Wilcox (1988) then exposed obturated roots in 64 teeth of monkeys to their oral environment for 1 week. Following tooth removal, the teeth were placed in dye and then cleared to visualize penetration. All samples showed presence of leakage, regardless of the type of sealer. In two samples the dye penetrated the entire length of the canals.

Lateral and vertical condensations were tested with dye and bacterial leakage. The techniques leaked at the same rate, between the filling material and the dentin wall

(Khayat et al. 1993). Magura et al. (1991) soaked obturated anterior teeth in whole human saliva. At three months both dye penetration, and stained histologic sections showed salivary penetration that was considered to be clinically significant. Torabinejad et al. (1990) showed that bacterial leakage in 45 obturated canals allowed *Staph. epidermidis* to completely contaminate 50% of the canals in 19 days, and *Proteus vulgaris* in 42 days in vitro.

Endotoxin has been used as a clinically relevant marker. To test for leakage along sealed gutta percha in coronally unsealed endodontically treated teeth in vitro, Trope (1995) tested 16 obturated roots. After 21 days, a gel coagulation and light transmission technique showed that endotoxin had penetrated completely through five of the samples.

Gish et al. (1994) prepared post spaces in extracted teeth leaving only 4 - 5 mm. of sealer and gutta percha. Bacterial leakage was evident in 17 of 20 specimens within the 90 day period. The speed of penetration varies from 1 to 90 days so there may be no safe length of time that obturation materials can remain exposed to oral flora without ill effects.

One source of endodontic failure is necrotic debris being left in the root canal system. A primary goal of endodontic therapy is to chemomechanically debride all pulp tissue, bacteria, and necrotic debris from the root canal system. This cleaning and shaping should be completed while maintaining the original canal configuration (Schilder 1974). Histologic studies have shown that root canal systems can be very irregular and complex (Hess 1925; Vertucci 1984). Alterations in canal space due to

age, operative procedures, decay, trauma, periodontal disease and treatment have been observed (Hess 1925, Davis et al. 1972). Limitations in current cleaning and shaping techniques can leave ramifications, such as accessory canals, fins, and deltas. These are left uncleaned even following aggressive mechanical procedures (McDonald and Hovland, 1996). These areas may allow viable colonies of bacteria to remain and may contribute to pathosis of the periradicular tissues.

The smear layer, as a result of mechanically instrumenting the canal walls, may be a barrier to bacteria, but may decrease the endodontic sealer's ability to prevent leakage. The objective of a study by Behrend et al. (1996) was to determine the effect of removal of the smear layer on canal obturation. They measured penetration of bacteria from a coronal direction. Using 54 extracted human roots, they removed the smear layer with 17% EDTA and 5.25% NaOCl prior to obturation. Using *Proteus vulgaris*, the frequency of bacterial penetration through teeth obturated with intact smear layer (70%) was significantly greater than that of teeth from which the smear layer had been removed (30%) $p<0.05$.

Fouad, Walton and Rittman (1993) used 24 ferret canine teeth to evaluate the healing of induced periapical lesions after root canal treatment as opposed to when infected debris was sealed within the canal. Most treated teeth showed a new cementum layer containing cementocytes and the lesions becoming very vascular. Early inflammatory infiltrate was primarily macrophages and plasma cells. Plump fibroblasts and osteoblasts were found with only rare osteoclasts. No epithelial rests or bacteria were found in any of the periapical lesions. The infected sealed controls

showed variable degrees of cementum resorption and a mild chronic inflammatory infiltrate with macrophages predominant.

INCOMPLETE ROOT FILLING

For decades procedures and materials have evolved to accomplish three dimensionally filled root canals. The concept is valid and its significance has been evaluated.

In retrospective radiographic success and failure studies, under-filled obturations have not been found to statistically decrease success and sometimes have better healing than those less than 2 mm. to the apex or beyond the apex (Storms 1969, Swartz et al., 1983, Szajkis & Tagger, 1983, Lin et al., 1992). Davis, Joseph and Butcher (1971) filled fourteen canals in four dogs, short by at least 3 mm. Periapical healing around under-filled canals compared favorably with that around canals obturated to the apex. In some cases, the unobturated canal space became filled with viable tissue in the form of a complete and apparently functional attachment apparatus that was continuous with the periodontium.

Allard et al. (1979) introduced micro-organisms into unfilled root canals of dogs. They were able to recover these same organisms from other unfilled previously uninfected root canals in the same jaw. They did not do blood sample testing to confirm a bacteremia. They concluded that the bacteria could not have reached the unfilled canals in any way other than via the bloodstream.

Delivanis and Fan (1981) however, showed that, despite weekly intravenous injections of saline suspensions of *Strep. sanguis* over a two month period, instrumented but unfilled feline canines did not become infected by an induced bacteremia. Infection of the unfilled canal was possible only when the periapical tissues were intentionally traumatized with a file and bleeding into the canal was induced during the course of bacteremia.

ECOLOGY OF THE ROOT CANAL

The root canal represents an enclosed environment in which selective pressure and competition for space results in the establishment of restricted populations of oral flora. Serial communities of mixed bacteria proceed until obligate anaerobes ultimately dominate the bacterial mix. Bacterial interrelationships, the nutritional supply and an increase in waste are key factors in determining the outcome of the infection. Endodontic treatment, apart from directly eliminating bacteria can completely disrupt the delicate ecology and deprive persisting bacteria of their nutritional source. However, it has been shown that anaerobic bacteria, which have survived the biomechanical treatment, may multiply in high numbers in the canal if no intracanal dressing or filling material is used. After the canal is sealed the anaerobiosis is restored and, in one author's opinion, an influx of tissue fluid into the canal can support a regrowth of bacteria (Sundqvist 1992).

Sjogren and Sundqvist (1987) agreed that since tissue fluid could act as a substrate for micro-organisms, which in many instances are left in the root canal after

thorough root canal debridement, the apical seal is important in preventing microcirculation between root canal and periradicular tissues

INTRODUCTION

The elimination or reduction of bacteria, and any antigenic or toxic material, to a physiologically tolerable level will result in healing of a periradicular lesion (Moller et al. 1981). Weine (1976) has indicated that improper restoration leads to loss of more endodontically treated teeth than actual failure of endodontic therapy. For decades procedures and materials have evolved to accomplish three dimensionally filled root canals. Evidence suggests that the exclusion of bacteria is the most important factor in preventing periapical lesions (Kakehashi et al. 1965).

The coronal seal of the root canal system has been implicated as a factor in the long term success of endodontic treatment. The placement of a proper final restoration has a highly significant effect on success (Swartz et al. 1983). Rapp et al. (1991) concurred, showing over 69% of teeth healed after endodontic treatment when a permanent restoration was placed compared to only 49% when a permanent restoration had not been placed. Another retrospective examination of 1010 endodontically treated teeth radiographically demonstrated that "the technical quality of the coronal restoration was significantly more important than the technical quality of the endodontic treatment for apical periodontal health" (Ray & Trope 1995).

The defense against bacterial ingress and infection of periradicular tissues has relied on a double seal: coronal restoration and radicular obturation. The advance of infective agents down the length of an obturated canal has been measured. Swanson & Madison (1987) evaluated the time that obturated canals could be exposed to artificial saliva before compromising the seal's integrity. They found that ink penetrated from

79% to 85% of the canal length in all exposed specimens. Madison & Wilcox (1988) exposed obturated roots in monkeys to their oral environment for one week. All samples showed presence of leakage, regardless of the type of sealer. Lateral and vertical condensations were tested with dye and bacterial leakage showing leakage at the same rate, between the filling material and the dentin wall (Khayat et al. 1993). Torabinejad et al. (1990) showed that obturated canals allowed *Staph. epidermidis* to completely contaminate 50% of the canals in nineteen days.

In 1997 Friedman et al. used a beagle dog model to assess the functional efficacy of endodontic fillings in vivo. At periods up to fourteen weeks they graded periradicular inflammation around canals treated in four different ways: 1.-Obturated with sealer and gutta percha, 2.-Obturated with just gutta percha, 3.-Obturated with just sealer, 4.-Unobturated roots. They inoculated each canal with plaque. The inflammation with each type of obturation was not significantly different but the unfilled canals all produced rarefying osteitis as early as three weeks with severe inflammation.

Temporary filling materials tested were not consistent in reducing leakage (Melton et al. 1990, Noguera & McDonald 1990, Uranga et al. 1999).

Mineral Trioxide Aggregate (MTA) was developed at Loma Linda University to seal off pathways of communication between the root canal system and the external surfaces of the tooth. The sealing properties of MTA have been evaluated using SEM, showing better marginal adaptation and superior sealing ability than amalgam, IRM, or Super EBA (Torabinejad et al. 1995). With in-vitro dye penetration and bacterial

invasion studies, Torabinejad et al. (1993,1994,1995) showed that MTA leaked significantly less than amalgam, Super EBA, and other materials.

The purpose of this study was to evaluate the effect of obturation and / or coronal seal in preventing the formation of periradicular lesions in dogs.

MATERIALS AND METHODS

This study was conducted on six female beagle dogs, one and a half years old, from the same litter. Their roots were fully formed. The dogs were randomly assigned to three evaluation periods, 6 weeks, 12 weeks, and 24 weeks. For every treatment, the dogs were anesthetized by an intravenous induction dose of thiopental (10-20 mg / kg), followed by inhalation anesthesia using 1% Halothane and 1-2 liters per minute of oxygen. This was supplemented with a local infiltration of 2% lidocaine with 1/100,000 epinephrine.

Root Canal Treatments

Maxillary first premolars and mandibular second, third, and fourth premolars were selected for this study. These teeth were isolated with a rubber dam, swabbed with iodine, then cleaned and shaped with balanced forces technique (Roane, 1985) copious 5.25% NaOCl irrigation. Ninety-six roots were randomly assigned to one of four treatment groups. Group one was obturated using warm vertical condensation with gutta percha and sealer and had MTA (Tulsa/Dentsply) mixed with sterile water placed into the upper 2 mm. of the canal and filling the pulp chamber. The second group was obturated in a similar manner, but had no coronal restoration placed. Group three was cleansed and shaped, not obturated and had an Intermediate Restorative Material (Caulk/Dentsply) filling placed coronally. Group four was cleansed and shaped, not obturated and had MTA placed as a coronal seal (see Table 1).

Animal Sacrifice

At 6, 12 and 24 weeks post-operatively, two dogs were euthanized with an overdose of intravenous Nembutal and perfused with 10% formalin. Block sections of the mandibles were removed, and fixed in 10% formalin. The specimens were decalcified in 10% formic acid. They were then embedded in butylmethacrylate and paraffin mix, and sagitally sectioned in six micron thick slices. Sections through the apical region of the roots were stained with hematoxylin and eosin and evaluated for the presence of periradicular lesions.

Evaluation

Microscopic evaluation of the degree of inflammation was subjectively ranked "none (0), mild (1)", "moderate (2)", or "severe (3)" on an ordinal scale. The size of any bony lesion including normal periodontal ligament space for statistical purposes and the percent density of inflammation were measured with morphometric analysis as described by Weibel (1979). Using the Image-Pro Plus Version 1.2 (Media Cybernetics 1994) each mounted specimen's image was captured digitally. Evaluating the last millimeter of the root, the area between the root surface and the bone was manually delineated. The program calculated the enclosed area. The program also calculated the percentage of area covered by nuclei stained with hematoxylin. This serves as an indicator of the density of inflammatory cells.

The data was evaluated statistically using analysis of variance, of four treatments and three time periods (Tables 3,4,5,and 6).

RESULTS

Table 2 shows that four samples were lost from the six week group due to technical errors in histologic preparation.

There was a statistically significant difference (95% confidence level) in the degree of inflammation between the two treatments with well-sealed coronal access (MTA) and the two groups without a permanently sealed access. There was no statistical difference between group 1 with obturated canals and a sealed access and group 4, not obturated but cleansed and shaped with a well sealed coronal access to prevent bacterial recontamination. There was also no difference between group 2 without a coronal seal and group 3 with an IRM temporary filling (Table 3). The only significant difference in the area of lesions was between the temporary filling group and the experimental group with MTA seal (Table 4).

The ordinal data from the microscopic evaluation and morphometric evaluation is contained in the APPENDIX. The correlation between the subjective visual microscopic evaluation and the morphometric analysis was extremely good (0.7887, $p < 0.001$) compared to the degree of inflammation and fairly well correlated to the area of the lesion (0.5642, $p < 0.01$).

Analysis of variance examined time as the independent variable on the dependent variables of area and density (Tables 5 and 6). Time is positively correlated to the density of inflammatory cells ($f=6.87$, $p=0.0018$). However, ANOVA found that time does not relate to the area of lesions ($f=0.83$, $p=0.44$). Using an addition model there was no interaction between time and treatment in any case.

Table 1 – Four experimental groups

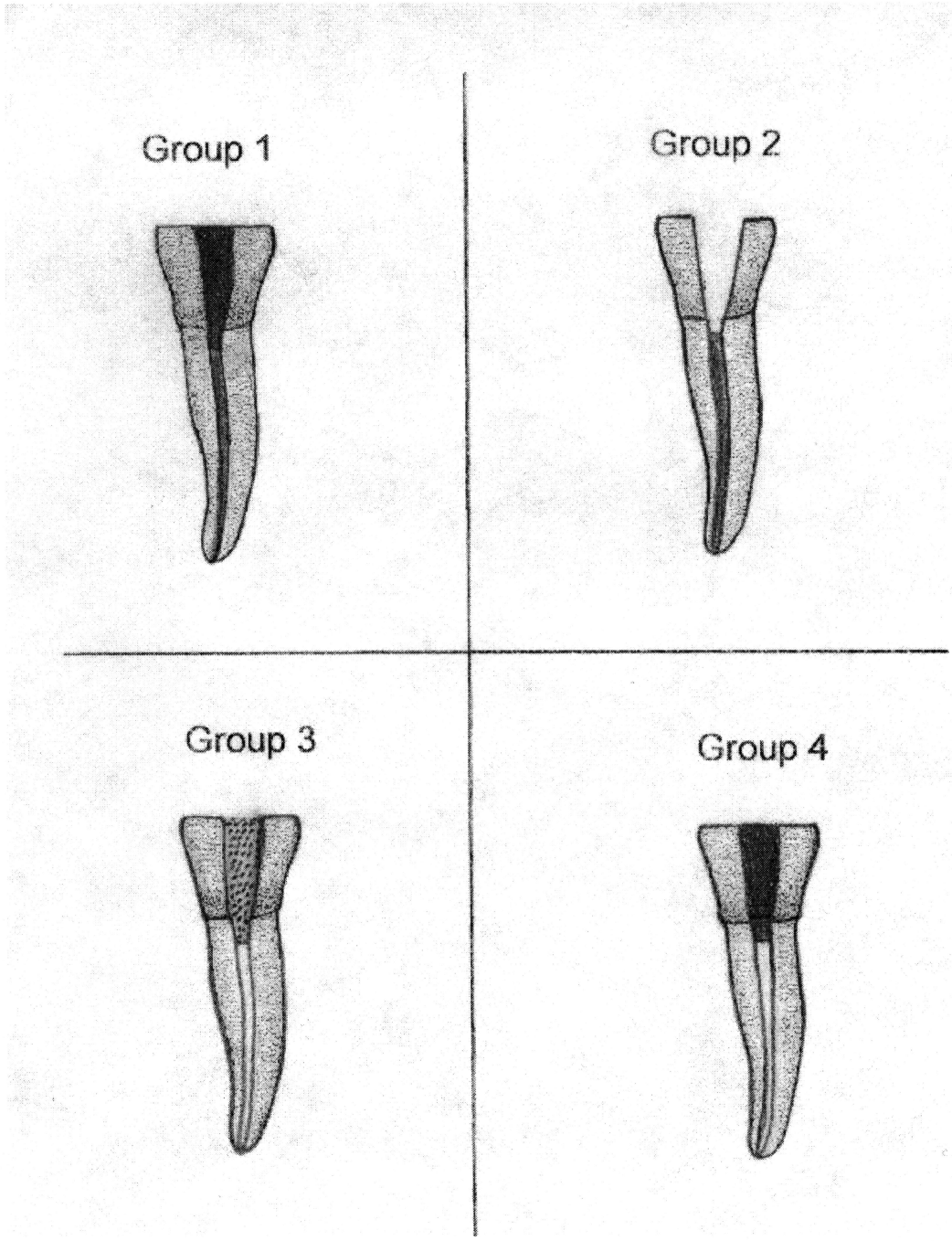


Table 2. Number of samples in each group by time period

	Group 1	Group 2	Group 3	Group 4
6 weeks	6	8	6	8
12 weeks	8	8	8	8
24 weeks	8	8	8	8

Table 3. Morphometric analysis of inflammatory cell density (as implied by stained nuclei)

Treatment Group	Mean %	Std. Deviation	Samples
Group 1	0.974	1.897	22
Group 2	5.017	4.726	24
Group 3	4.864	5.565	22
Group 4	0.579	0.455	24

Table 4. Morphometric analysis of apical area

Treatment Group	Mean Sq. mm.	Std. Deviation	Samples
Group 1	0.363	0.354	22
Group 2	0.543	0.418	24
Group 3	0.611	0.516	22
Group 4	0.273	0.118	24

Table 5. Morphometric analysis of inflammatory cell density with regards to time (as implied by stained nuclei)

Inflammatory cell density as a percentage.	Samples	Mean	Std. Deviation.	Min.	Max
6 weeks	28	0.976	0.971	0	3.6
12 weeks	32	4.241	5.524	0.1	22.6
24 weeks	32	3.116	4.070	0	17.5

Table 6. Morphometric analysis of apical area with regards to time

Area in Sq. mm.	Samples	Mean	Std. Deviation.	Min.	Max
6 weeks	28	0.386	0.313	0.08	1.60
12 weeks	32	0.431	0.357	0.12	1.97
24 weeks	32	0.514	0.489	0.05	1.90

DISCUSSION

Three of the treatment groups were designed to represent patient situations that occur frequently and one situation that is experimental.

Group one (Figures 1 and 2) is the current standard of care. Thorough cleansing and shaping with complete obturation with sealer and gutta percha, followed by permanent coronal restoration, prevents bacterial invasion. The low amount of inflammation found in this group supports the findings of Kakehashi et al. (1965). If bacterial access to the apical tissue is reduced or prevented the apical tissue will stay healthy.

Group two (Figures 3 and 4) represents patients who do not follow through obtaining the recommended final restoration and whose temporary restoration leaks or comes out altogether. This group had a high level of inflammation and supports the in vitro research by Swanson and Madison (1987), Magura et al. (1991) and Torabinejad et al. (1990). Obturation with gutta percha and sealer alone does little to slow the rapid progress of bacteria down the length of a canal. In addition you need a coronal seal.

Group three (Figures 5 and 6) creates the situation of a patient treated with a pulpectomy. They have no pain and neglect to return to finish the endodontic procedure. This group also had a high level of inflammation and is consistent with the in vitro studies by Melton et al. (1990), Noguera and McDonald (1990) and Uranga et al. (1999). These in vitro dye leakage studies show that temporary filling materials provide an inconsistent and unpredictable seal.

Group four (Figures 7 and 8) is an experimental category that controls the coronal access of bacteria but does not prevent the ingress of fluids, or material from the apical tissues which are presumably sterile. The low inflammation found in this group agrees with the retrospective studies by Swartz et al. (1983), Rapp et al. (19991) and Ray, Trope (1995). The presence and adequacy of the permanent coronal restoration is of utmost importance. The best root canal will fail if the coronal seal does not protect it.

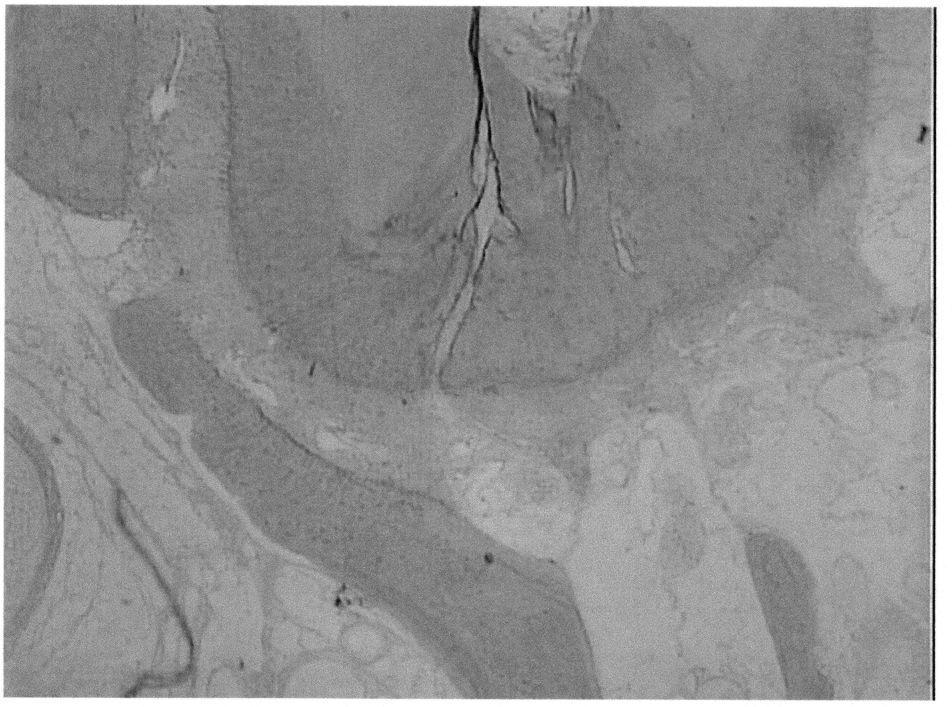


Figure 1 - Group One, 6 Weeks, Specimen 28d (4X)

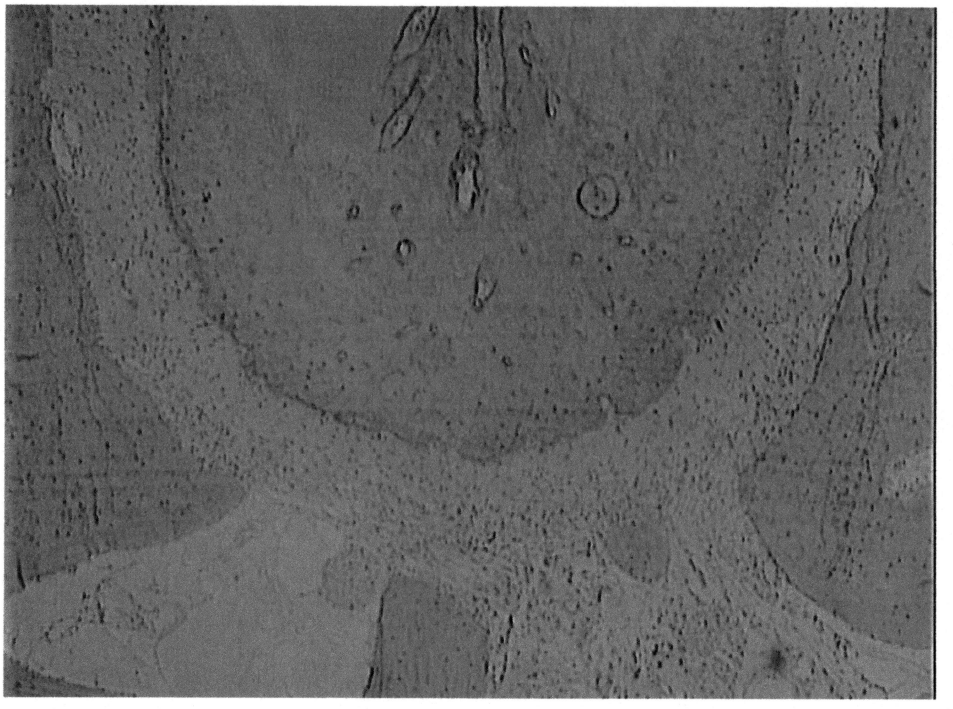


Figure 2 - Group One, 24 Weeks, Specimen 07d (4X)

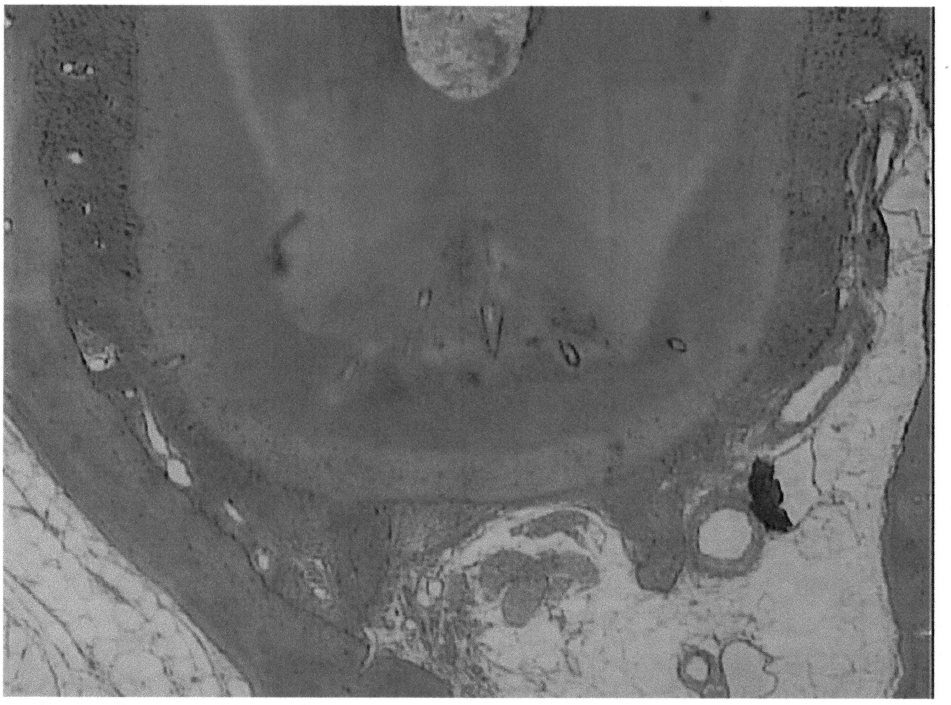


Figure 3 - Group Two, 6 Weeks, Specimen 31m (4X)

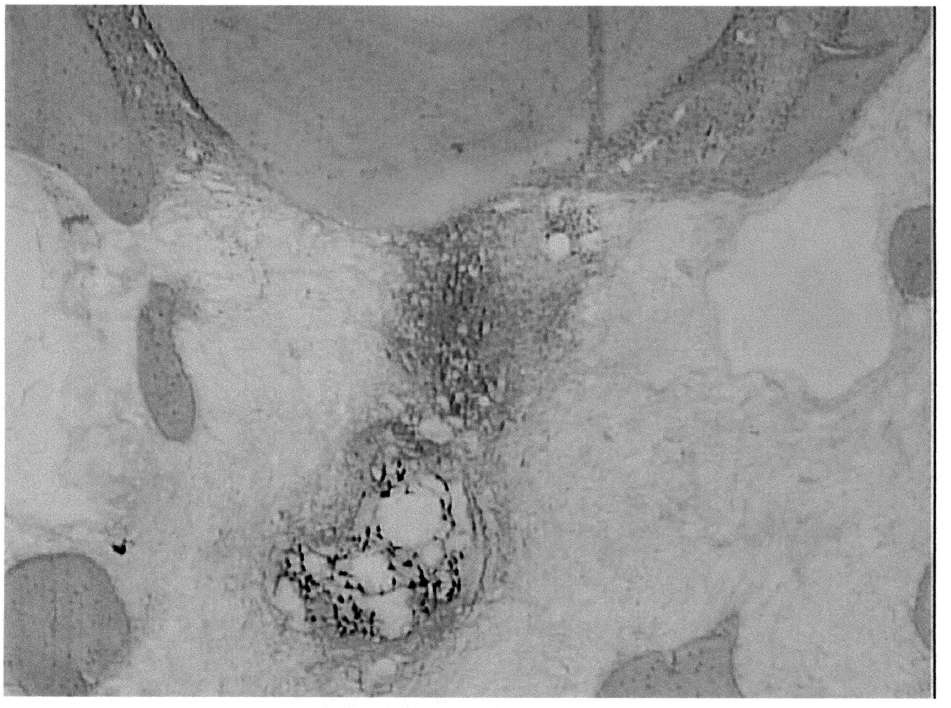


Figure 4 - Group Two, 12 Weeks, Specimen 35m (4X)

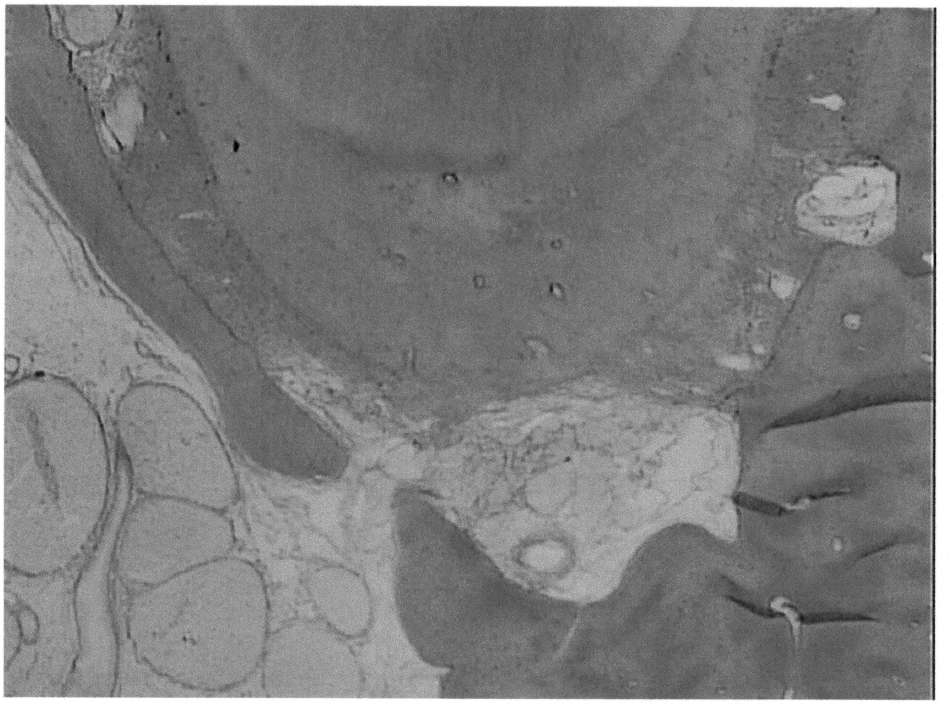


Figure 5 - Group Three, 6 Weeks, Specimen 32m (4x)

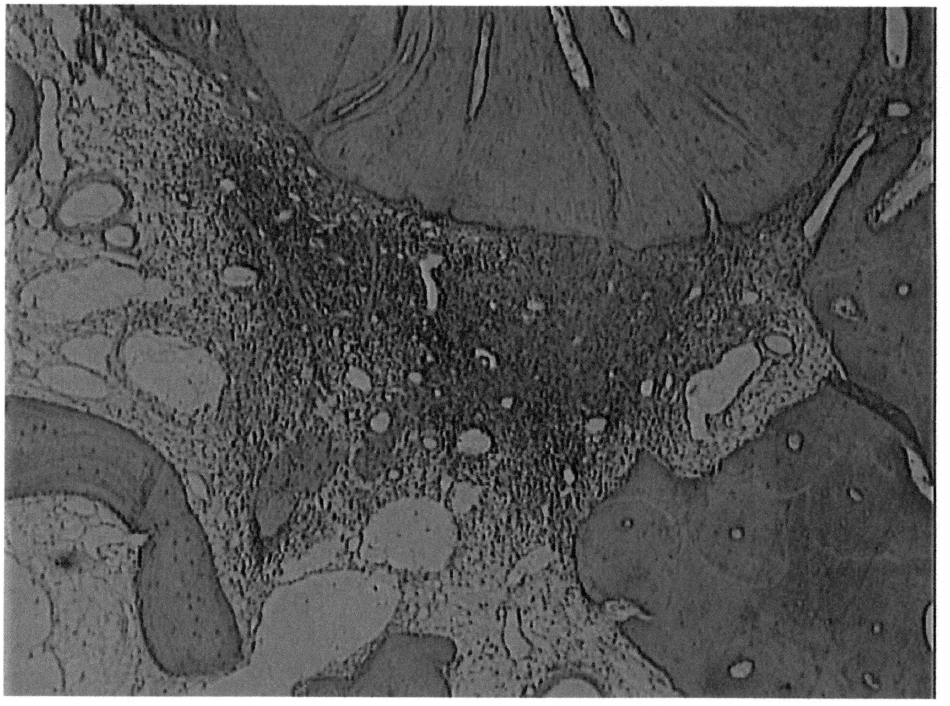


Figure 6 - Group Three, 12 Weeks, Specimen 46d (4X)

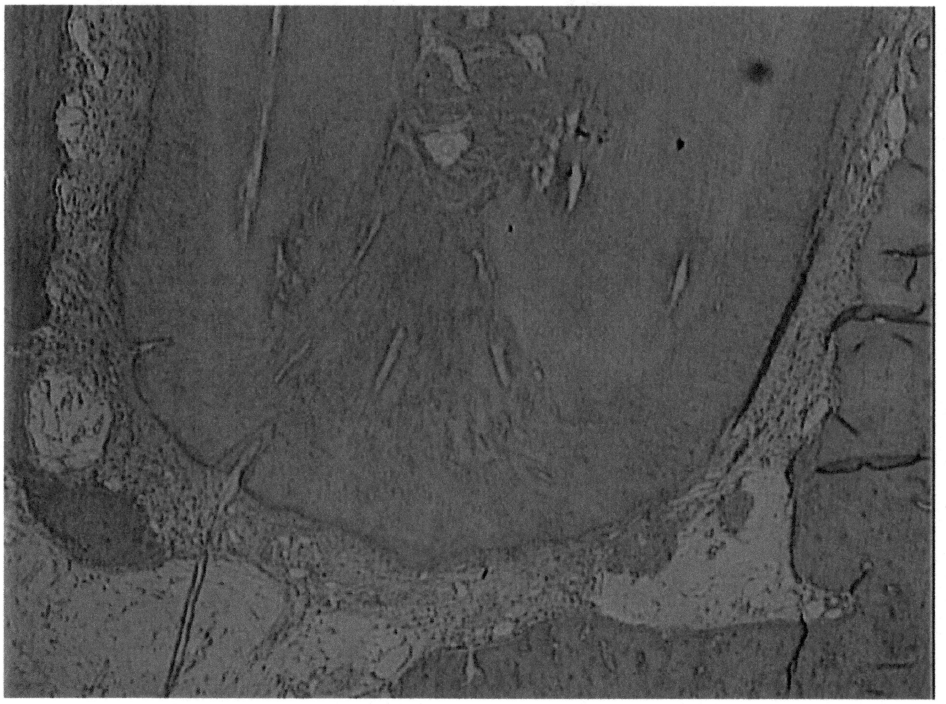


Figure 7 - Group Four, 12 weeks, Specimen 43m (4X)

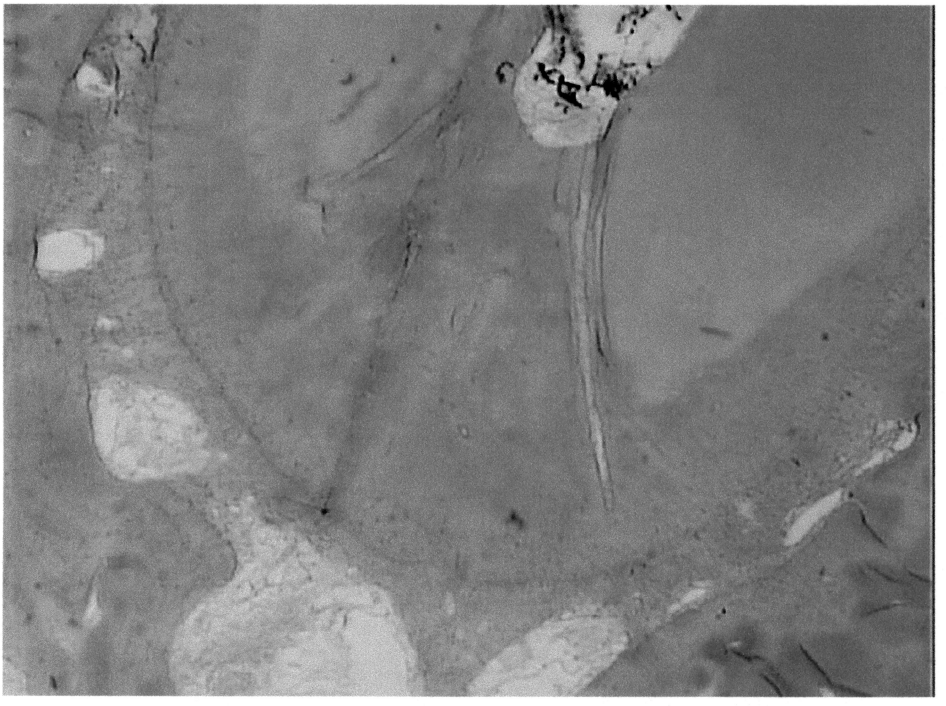


Figure 8 - Group Four, 24 Weeks, Specimen 5d (4X)

Beagle dogs with mature apices were used based on previous investigation that identified them as suitable models for the histologic evaluation of periradicular inflammation after experimental root canal treatments (Friedman et al. 1997). In our study, lesions were not induced to evaluate healing of apical tissue. It is more reliable to detect a change from no signal to evidence of a signal (i.e. inflammation or bone loss) than it is to detect a change in an existing signal (i.e. less or more inflammation or bone loss). Tissue, healthy at the start of treatment, was evaluated for any deviation from normal.

Some of our results were similar to those found by Friedman et al. (1997). Both studies have demonstrated that when intraoral bacteria were allowed access to the root canal system, periradicular periodontitis results. In their study Friedman et al. resealed the access cavities coronally after their treatment procedures. In contrast, after finishing our procedures the dogs were allowed to return to a normal kennel diet. This experimental design reproduces more closely clinically relevant conditions, including the effects of mastication and the washing out of a temporary filling or sealer. The positive controls with open canal spaces and bacterial contamination were not included in our study because of its predictability as noted in the study by Friedman et al.

Friedman et al. (1997) were limited in their ability to demonstrate significant differences between the experimental procedures by using only two to three samples per group and a fourteen week time period. Our study obtained additional power by using eight sample roots per experimental group and the observation period went to twenty-four weeks.

These two studies used different materials for coronal seal, glass ionomer and MTA. Three of five of the unfilled non-infected roots protected by glass ionomer, in the Friedman research (1997), became inflamed apically. This delayed and inconsistent development of periradicular periodontitis could have resulted from inadvertent bacterial ingress due to inadequate coronal seal, a risk that would equally affect all the groups. Their explanation was that other groups, also not inoculated, did not develop inflammation thus the risk of contamination was not significant. The effects of coronal leakage may have been delayed by the obturation, and become evident over a longer experimental period.

As has been shown previously MTA provides an excellent seal (Torabinejad et al. 1993, 1994, 1995). The reason for the presence of inflammation in two roots of group one was a lack of seal. Breakdown of the MTA was visible in both samples. As suggested by Torabinejad et al. because of low compressive strength MTA can not be used for a final restoration.

A curious finding was noted in that the length of time was positively correlated to the density of inflammation but not to the size of the lesion. There may be periods of rapid increase in lesion size followed by periods where inflammation density increases but the area of the lesion reaches a plateau.

The distinctly poorer results from the groups with no permanent filling may be a factor in the decision to complete endodontic treatment in one appointment and place an access restoration immediately.

These results confirm the importance of the permanent restoration in the success of endodontic therapy. If bacteria are excluded tissues remain healthy. Our most diligent effort needs to be applied to materials research and clinical techniques to prevent recontamination of root canals with oral flora, in order to give our patients the greatest opportunity for health.

CONCLUSION

Providing a good seal coronally prevents recontamination of the root canal system and the formation of periradicular lesions, with and without obturation.

APPENDIX

Twenty four weeks data

Specimen	Treatment Group	Microscopic Evaluation	Morphometric Area (sq. mm.)	Morphometric % inflammation
01M	3	0	0.20	0.0
D	3	3	0.49	5.4
02M	3	3	1.90	1.9
D	3	1	0.13	0.3
03M	2	0	0.34	2.5
D	2	0	0.24	0.8
04M	2	0	0.18	3.3
D	2	2	0.46	3.9
05M	4	0	0.30	0.5
D	4	0	0.30	0.1
06M	4	0	0.37	0.6
D	4	0	0.21	0.8
07M	1	0	0.05	0.4
D	1	0	0.26	0.5
08M	1	0	1.03	1.8
D	1	0	0.53	0.8
09M	4	0	0.07	0.1

D	**4**	**0**	**0.08**	**0.3**
10M	**3**	**3**	**0.511**	**2.3**
D	**3**	**3**	**0.42**	**7.8**
11M	**4**	**0**	**0.13**	**0.1**
D	**4**	**0**	**0.20**	**0.2**
12M	**3**	**0**	**1.16**	**6.0**
D	**3**	**2**	**0.53**	**7.2**
13M	**2**	**3**	**0.93**	**7.3**
D	**2**	**3**	**1.75**	**17.5**
14M	**2**	**3**	**1.60**	**6.2**
D	**2**	**2**	**0.80**	**7.0**
15M	**1**	**0**	**0.15**	**1.0**
D	**1**	**0**	**0.2**	**0.1**
16M	**1**	**0**	**0.38**	**1.7**
D	**1**	**1**	**0.55**	**1.3**

Six weeks data

17M	**1**	**0**	**0.13**	**0.0**
D	**1**	**0**	**0.08**	**0.0**
18M	**2**	**0**	**0.40**	**0.1**

D	**2**	**1**	**0.36**	**2.9**
19M	**3**	**0**	**0.18**	**0.3**
D	**3**	**0**	**0.38**	**1.3**
20M	**4**	**0**	**0.37**	**0.7**
D	**4**	**0**	**0.28**	**0.3**
21M	**4**	**0**	**0.23**	**0.9**
D	**4**	**0**	**0.31**	**1.1**
22M	**1**	**missing**		
D	**1**	**1**	**0.15**	**1.2**
23M	**2**	**0**	**0.18**	**0.1**
D	**2**	**0**	**0.58**	**1.5**
24M	**3**	**1**	**0.94**	**1.8**
D	**3**	**missing**		
25M	**1**	**0**	**0.18**	**0.0**
D	**1**	**0**	**1.60**	**0.4**
26M	**2**	**0**	**0.19**	**2.3**
D	**2**	**0**	**0.20**	**2.4**
27M	**3**	**0**	**0.54**	**2.4**

D	**3**	**0**	**0.25**	**0.2**
28M	**1**	**missing**		
D	**1**	**0**	**0.22**	**0.2**
29M	**4**	**0**	**0.36**	**0.5**
D	**4**	**0**	**0.35**	**0.6**
30M	**4**	**0**	**0.43**	**0.8**
D	**4**	**0**	**0.31**	**0.8**
31M	**2**	**0**	**0.26**	**0.6**
D	**2**	**1**	**0.90**	**3.6**
32M	**3**	**1**	**0.46**	**0.3**
D	**3**	**Missing**		

Twelve week data

33M	**1**	**0**	**0.32**	**0.1**
D	**1**	**0**	**0.17**	**0.2**
34M	**3**	**1**	**0.22**	**1.6**
D	**3**	**0**	**0.19**	**0.5**
35M	**2**	**3**	**0.74**	**12.7**
D	**2**	**3**	**0.48**	**7.1**
36M	**4**	**2**	**0.58**	**1.9**

D	**4**	**1**	**0.38**	**1.6**
37M	**3**	**2**	**0.62**	**8.9**
D	**3**	**2**	**0.62**	**10.6**
38M	**2**	**0**	**0.19**	**0.6**
D	**2**	**1**	**0.43**	**3.0**
39M	**1**	**0**	**0.18**	**0.3**
D	**1**	**0**	**0.15**	**0.5**
40M	**4**	**0**	**0.17**	**0.4**
D	**4**	**0**	**0.21**	**0.3**
41M	**4**	**0**	**0.12**	**0.2**
D	**4**	**0**	**0.31**	**0.2**
42M	**1**	**0**	**0.58**	**0.5**
D	**1**	**0**	**0.43**	**0.1**
43M	**4**	**0**	**0.28**	**0.4**
D	**4**	**0**	**0.20**	**0.5**
44M	**2**	**3**	**0.38**	**7.9**
D	**2**	**2**	**0.25**	**2.9**
45M	**3**	**0**	**0.13**	**0.1**

D	**3**	**3**	**1.97**	**6.8**
46M	**3**	**3**	**0.50**	**8.7**
D	**3**	**3**	**1.12**	**2.6**
47M	**2**	**2**	**0.52**	**15.1**
D	**2**	**2**	**0.68**	**9.1**
48M	**1**	**2**	**0.41**	**9.1**
D	**1**	**1**	**0.27**	**1.2**

BIBLIOGRAPHY

Abedi H, Ingle J. Mineral Trioxide Aggregate: a review of a new cement. Cal. Dent. J. 36 - 39 Dec. 1995.

Allard V, Nord C, Sjoberg L, Stromberg T. Experimental injections in the jaws of dogs. Oral Surg 48:454, 1979.

Barkhordar R, Kempler D. Microleakage of endodontic access cavities restored with composites. CDA Jour. 25;3:215-218, 1997.

Bergenholtz G. Cox C, Loesche W, Syed S. Bacterial leakage around dental restorations: its effect on the dental pulp. J Oral Path 11:439-450,1982.

Berry F, Tjan A. Microleakage of amalgam restorations lined with dentin adhesives. Am J Dent 7;6:333-336,1994.

Berry F, Parker S, Rice D, Munoz C. Microleakage of amalgam restoration using dentin bonding system primers. Am J Dent 9;4:174-178,1996.

Chong B. Coronal leakage and treatment failure. J Endod 21;3:159-160,1995.

Clem W. Endodontics the adolescent patient. Dent Clin North Am 13:483, 1969.

Cohen S, Burns R. Pathways of the pulp. Ed 6, p 179, St.Louis, 1994, Mosby.

Cox C, Keall C, Ostro E, Bergenholtz G. Biocompatibility of surface-sealed dental materials against exposed pulps. J Prosth Dent. 57:1-8, 1987.

Davis M, Joseph S, Butcher J. Periapical and intracanal healing following incomplete root canal fillings in dogs. Oral Surg 31;5:662-675,1971.

Davis SR, Brayton SM and Goldman M. The Morphology of the Prepared Root Canal. Oral Surg;34:642-648,1972.

Delivarris PD and Fan VSC. The localization of blood born bacteria in instrumented unfilled and overinstrumented canals. J Endod 10:521,1984.

Fouad A, Walton R, Rittman B. Healing of induced periapical lesions in ferret canines. J Endod 19;3:123-129,1993.

Friedman S, Torneck C, Komorowski R, Ouzounian Z, Syrtash P, Kaufman A. In vivo model for assessing the functional efficacy of endodontic filling materials and techniques. J Endod 23;9:557-561, 1997.

Gish S, Drake D, Walton R, Wilcox L. Coronal leakage: bacterial penetration through obturated root canals following post preparations. JADA 125;10:1369-1372,1994.

Hess W. The Anatomy of the Root Canals of the Teeth of the Permanent Dentition: Part I. New York: Winwood & Co.;1-47.

Jansson L, Ehnevid H, Lindskog, S Blomlof L. Development of periapical lesions. Swed Dent J 17:85-93,1993.

Kakehashi S, Stanley H, Fitzgerald R The effects of surgical exposures of dental pulps in germ-free and conventional laboratory rats. O.S., O.M. & O.P. 20;3:340-349, 1965.

Khayat A, Lee S, Torabinejad M.
Human saliva penetration of coronally unsealed obturated root canals. J Endod 19;9:458-461, 1993.

Leonardo M, Almeida W, Bezerra da Silva L, Utrilla L. Histopathological observations of periapical repair in teeth with radiolucent areas submitted to two different methods of root canal treatment. J Endod 21;3:137-141,1995.

Lin L, et al. Factors associated with endodontic treatment failures. J Endod 18;12:625-627,1992.

Madison S, Swanson K, Chiles S. An evaluation of coronal microleakage in endodontically treated teeth. Part II Sealer types. J Endod 13;3:109-112, 1987.

Madison S, Wilcox L. An evaluation of coronal microleakage in endodontically treated teeth. Part III In Vivo study. J Endod 14;9:455-458, 1988.

Magura M, Kafrawy A, Brown C, Newton C. Human saliva coronal microleakage in obturated root canals: An in vitro study. J Endod 17;7:324-331, 1991.

McDonald N, Hovland E. Surgical endodontics; a conservative approach.
Principles and Practice of Endodontics; Walton & Torabinejad. Phil.PA, WB Sanders Co. 401-422, 1996.

Melton D, Cobb S, Krell K.
A comparison of two temporary restorations: light-cured resin versus a self-polymerizing temporary restoration. O.S., O.M., O.P. 70;2:221-225, 1990.

Moller AJR, Fabricius L, Feiglin B,Anderson L, Lindskog S. Influence on periapical tissues of indigenous oral bacteria and necrotic pulp tissue in monkeys. Scand J Dent Res 89:475-481, 1981.

Noguera A, McDonald N. Comparative in vitro coronal microleakage study of new endodontic restorative materials. J Endod 16;11:523-527, 1990.

Rapp E, Brown C, Newton C. An analysis of success and failure of apicoectomies. J Endod 17;10:508-511,1991.

Ray H., Trope M. Periapical status of endodontically treated teeth in relation to the technical quality of the root filling and the coronal restoration. Intern. Endo. Jour. 28:12-18, 1995.

Roane J, Sabala C, Duncanson M. The "Balanced Force" concept for instrumentation of curved canals. J Endod 11;5:203-211, 1985.

Schilder H. Cleaning and shaping the root canal. Dent Clin North Am 18:269-296,1974.

Sjogren U, Sundqvist G. Bacteriologic evaluation of ultrasonic root canal instrumentation. OS, OM, OP 63:366-370,1987.

Soderholm K. Technique sensitivity of dentin bonding. Gen Dent May-June 1997.

Storms J. Factors that influence the success of endodontic treatment. J. Canad. Dent. Ass 32;2, 1969.

Sundqvist G. Ecology of the root canal flora. J Endod 18;9:427-430,1992.

Swanson K, Madison S.
An evaluation of coronal microleakage in endodontically treated teeth. Part I Time periods. J Endod 13: 56-59, 1987.

Swartz D, Skidmore A, Griffin J. Twenty years of endodontic success and failure. J Endod 9;5:198-202, 1983.

Szajkis S, Tragger M. Periapical healing in spite of incomplete root canal debridement and filling. J Endod 9;5:203-209, 1983.

Torabinejad M, Ung B, Kettering J. In vitro bacterial penetration of coronally unsealed endodontically treated teeth. J Endod 16:566, 1990.

Torabinejad M, Watson T, Pitt Ford T. Sealing ability of a mineral trioxide aggregate when used as a root end filling material. J Endod 19:591-595, 1993.

Torabinejad, M, Higa R, McKendry D, Pitt Ford T. Dye leakage of four root end filling materials: effects of blood contamination. J Endod 20:159-163, 1994.

Torabinejad M, Falah R, Kettering J, Pitt Ford T. Bacterial leakage of mineral trioxide aggregate as a root-end filling material. J Endod 21;3:109-112, 1995.

Torabinejad M, Wilder Smith P, Pitt Ford T. Comparative investigation of marginal adaptation of mineral trioxide aggregate and other commonly used root end filling materials.J Endod 21:295-299, 1995.

Trope M, Chow E, Nissan R. In vitro endotoxin penetration of coronally unsealed endodontically treated teeth. Endod Dent Traumatol 11:90-94,1995.

Uranga A, Blum J, Esber S, Parahy E, Prado C. A comparative study of four coronal obturation materials in endodontic treatment. J Endod 25:178-180.

Vertucci F. Root Canal Anatomy of the Human Permanent Teeth. Oral Surg; 58:589-599, 1984.

Weibel E Stereological Methods Vol. 1. Practical methods for biological morphometry. London: Academic Press 106 - 108, 1979.

Weine F. Endodontic therapy. 2nd ed.CV Mosby, St. Louis 1976.

White S, Sorensen J Kang S, Caputo A. Microleakage of new crown and fixed partial denture luting agents. J Pros Dent 67:156-161,1992.

White S, Yu Z, Tom J, Sangsurasak S. In-vivo microleakage of luting cements for cast crowns. J Pros Dent 71:333-338,1994.

Wright W, Mazer R, Teixeira L, et al. Clinical microleakage evaluation of a cavity varnish. Am J Dent 5:263-265,1991