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An in Vivo Autoradiographic Study of the Relative Penetrating Abilities of Aqueous 2% Parachlorophenol and Camphorated 35% Parachlorophenol in Endodontics

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AN IN VIVO AUTORADIOGRAPHIC STUDY OF THE RELATIVE PENETRATING ABILITIES OF AQUEOUS 2% PARACHLOROPHENOL AND CAMPHORATED 35% PARACHLOROPHENOL IN ENDODONTICS

BY

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AUTOBIOGRAPHY

Gary N. Taylor was born in Detroit, Michigan on July 4, 1945, to Harry M. and Dorothy P. Taylor.

He was graduated from St. Jude Parochial School in Detroit and moved to Los Angeles, California in 1959 where he received his high school diploma from Bishop Alemany High School in 1963. A Bachelor of Science degree was earned in 1967 from Loyola University of Los Angeles before entering Loyola University School of Dentistry in Chicago. He received the degree of Doctor of Dental Surgery in June, 1971.

He continued his studies at Loyola as a resident in the Department of Endodontics and in the Department of Oral Biology, Graduate Division, to work toward the degree of Master of Science.

The author's wife is Mary Dawn, his two children are Dawn-Marie Dorothy and Joy Catherine.

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CHAPTER I

INTRODUCTION

Camphorated parachlorophenol has enjoyed over six decades as the medicament of choice during routine endodontic procedures. Numerous studies have reported the antimicrobial effectiveness of this drug as commercially prepared without investigating its pre-twentieth century formulation or range of diffusibility within the root canal system. It is clinically significant to the practicing dentist to understand the biological and physiological nature of the tissues undergoing treatment as well as the characteristics of the medications he is using. The utilization of any medicament in endodontic procedures without knowledge of its antimicrobial spectrum, toxicity, and diffusibility does not appear to be consistent with sound principles of chemotherapy.

Tissue reactivity to camphorated parachlorophenol has been adequately demonstrated *in vivo* as has its effects on those microorganisms commonly isolated from contaminated root canal systems and adjacent periapical regions. However, one dimension, namely its diffusibility to the root apex as well as into the dentinal tubules, has been conspicuously ignored. Sterilization of the internal aspect of the tooth requires the elimination of all microorganisms regardless of their location thus requiring effective dentinal tubule penetration by the intracanal medicament.

The purpose of this study will be to determine simultaneously the *in vivo* diffusibility of both camphorated and aqueous two percent parachlorophenol in the dog premolar teeth.

CHAPTER II

REVIEW OF THE LITERATURE

Few therapeutic agents in the history of dentistry have been utilized as extensively as camphorated parachlorophenol. This medicament, introduced in 1891 by Dr. Otto Wolkoff, is defined by The National Formulary as an eutectic solution of thirty-five percent parachlorophenol and sixty-five percent camphor, the latter theoretically maximizes the volitility of the drug. Its formulation, however, preceded the histologic confirmation of the pulp as a direct extension of normal periapical connective tissue. The impact of its clinical usage and effect on these tissues can be rationally assessed only when examined in light of its three major characteristics, namely, tissue toxicity, antimicrobial effectiveness and permeability.

A. TOXICITY

At the turn of the century, Dr. Green Vardiman Black insisted that antiseptic drugs be chosen on the basis of their immediate direct and latent residual effects on living tissue as well as their antimicrobial effectiveness. Blayney in the middle 1920's re-emphasized Black's early remarks by applying them to the endodontic situation. He proposed that induced chemical necrosis by highly

toxic drugs left in the root canals or forced beyond the apex was an invitation to clinical failure as well as acute post-operative symptomology.

No investigation before 1923 considered the toxicity of aqueous two percent parachlorophenol either alone or in its camphorated form. Price at this time reported that Wolkoff's preparation yielded less toxic results than many other drugs currently in clinical use (phenol, formalin and formocresol). Its tissue toxicity was not specifically mentioned in the dental literature again until Grossman, who in 1944, used an epidermal application technique to confirm Price's earlier work. Efforts to develop new and less damaging intracanal medicaments were begun by Casey and Gurney. Rubbo, Reich and Dixon in 1958, demonstrated the production of severe necrosis and ulceration with subcutaneous injections of camphorated parachlorophenol in rabbit's ears.

Schilder, in 1959, demonstrated the high toxicity of camphorated parachlorophenol by producing ocular inflammation experimentally in laboratory animals. Torneck's work, in 1961, yielded histologic data confirming the presence of severe tissue reactions but did not find the wide-spread necrosis as seen with other commonly used intracanal medicaments. Engstrom, in 1967, reported similar severe inflammatory responses associated with necrosis in the same animal species used by Torneck when he conducted a parallel experiment.

Finally, Harrison in 1971, reported conclusively "the severe toxic qualities of camphorated parachlorophenol as well as its ability to coagulate protein." Pronounced coagulation necrosis was evident in one to three day rabbit specimens after intradermal injection of 0.10 ml camphorated parachlorophenol. Although it is important to remember that this situation does not exactly conform to the usual clinical application, it does create speculation on potential tissue damage when using medications regardless of their historical association with dentistry. Interestingly, Harrison also tested one and two percent aqueous parachlorophenol preparations and found the inflammatory response to be of a mild nature.

B. ANTIMICROBIAL EFFECTIVENESS

The importance of microorganisms occupying the root canal space was brought to focus by W.D. Miller in 1888, when he stated that "each gangrenous tooth pulp may be in itself a center of infection." Medicaments of all forms including fixatives, caustics and antibiotics have been advocated in the attempt to render the canal environment sterile, camphorated parachlorophenol being most frequently popular.

An early study in 1948 by Ostrander showed the genus Streptococcus to be the dominant microorganism isolated from the root canals of infected teeth. Winkler and van Amerongen, in 1959, confirmed this work and concluded that not only did streptococci compose approximately sixty-three percent of the microorganisms in infected

root canals but that Streptococcus fecalis had the greatest frequency in pure culture. Crawford and Shankle in 1961 pointed out that current root canal culturing techniques fell far short of indicating the presence of intracanal infection and they were able to demonstrate histologically the presence of microorganisms following negative cultures.

Martin, Lasala and Michanowicz, in 1968, found that camphorated parachlorophenol did diffuse through the apices of extracted teeth and did provide inhibition of Group A streptococci on blood agar. This inhibition however was found to be significantly less than that produced by penicillin G or formocresol.

Harrison, in a Master's thesis in 1969, noted that not one study could be found in the dental literature which reports the antimicrobial spectrum of camphorated parachlorophenol. Likewise, the effective concentration of this drug has not been determined against the various microorganisms most frequently inhabiting the infected root canal. Harrison reported his in vitro findings in 1970 by stating that Streptococcus fecalis was the most resistant of the test microorganisms to aqueous two percent parachlorophenol while staphylococci and yeast were controlled at lower drug concentrations. The effective bactericidal concentration of aqueous two percent parachlorophenol against the most resistant microorganism was determined, by Harrison, to be 0.00125 gm/ml thus making a one percent aqueous solution of aqueous two percent parachlorophenol a ninefold increase beyond that

required for bacterial inhibition. He therefore suggested that the use of thirty-five percent parachlorophenol as it exists in camphorated parachlorophenol can no longer be justified due to the inherent toxicity of both camphor and parachlorophenol. These conclusions implied that the institution of the clinical usage of an aqueous one or two percent parachlorophenol preparation be considered a logical replacement for camphorated parachlorophenol.

Treanor and Goldman in 1972, showed in an in vitro study that camphorated parachlorophenol introduced into the pulp chamber of endodontically prepared and temporarily sealed teeth, could produce inhibitory effects on blood agar plates innoculated with the viridans group of streptococci. They also stated that the medicament was most effective at an interval of seventy-two hours rather than the forty-eight hour interval usually recommended for clinical usage.

Cwikla, also in 1972, showed that camphorated parachlorophenol sealed in a tooth similar to Treanor's method would produce zones of inhibition against Staphylococcus aureus but not to Streptococcus fecalis. Wantulok and Brown, in late 1972, independently duplicated Cwikla's study using Staphylococcus aureus and achieved the same results. These studies therefore serve to confirm in a situation more closely aligned to clinical endodontics, the results of Harrison showing Streptococcus fecalis to be the most resistant microorganism to camphorated parachlorophenol in vitro.

Wong, in a 1972 Master's thesis, showed the greatest effect of camphorated parachlorophenol on Streptococcus fecalis to be at seventy-two hours and that Streptococcus fecalis was more resistant than Staphylococcus aureus and Candida albicans to the medicament. An interesting aspect of his study which involved the exposure of Staphylococcus aureus to individual quantities of either camphor, parachlorophenol, or camphorated parachlorophenol, showed the volatile phase of each component to be inhibitory. Camphor was found to be the least effective and parachlorophenol to be the most effective component of the eutectic solution.

C. DIFFUSIBILITY

The method by which intracanal medicaments are dispersed within the root canal system has been until recently a matter of speculation. Clinical procedures have been therefore arrived at empirically rather than through the result of scientific research. Harlan, in 1891, was the first to advocate the use of medicaments that are capable of diffusion in the treatment of pulpless teeth. York showed in 1897 that carbolic acid was able to diffuse entirely through the dentin of extracted teeth.

Fish, in 1927, demonstrated the tubular nature of dentin by the infusion of India ink through dog teeth. Coolidge, in 1929, proposed that any chemical substance capable of coagulating protein

would have only shallow penetration in dentin regardless of its surface tension. Zander and Smith in 1945, conducted one of the last dye studies using silver nitrate. It is from that period that radioactive isotope studies dominated drug penetration research.

Hevesy, Holst and Krog in 1933, published the first paper on experimental work with radioactive isotopes in teeth. However no study, prior to Marshall's et al. report in 1960, was designed to investigate permeability from the pulpal side of the root canal. Using radioactive tracers, he showed that different drugs and treatment irrigants altered the permeability response of the dentinal tubules.

The first reference of the diffusibility of camphorated parachlorophenol in the dental literature as the result of experimental research is found in the textbook, Clinical Endodontics by Sommer, Ostrander and Crowley in 1961. Their reference is directed towards Stamp's work in 1953 when he claimed that camphorated parachlorophenol presented a moderate penetrating ability through dentin to a depth of 2.0 mm.

Avny in a 1970 Master's thesis began the first in vitro root canal study of parachlorophenol. He chose aqueous two percent parachlorophenol as the result of Harrison's work and by the use of autoradiography, Avny observed the movement of the tritiated parachlorophenol molecule through dentin. Some of his specimens, extracted human teeth, were intentionally and incompletely debrided to

consider Coolidge's postulate of protein coagulation. Avny's results showed that the medicament penetrated the dentin from the pulp chamber and root canal to the cemento-dentinal junction as well as diffused to the apex in fully debrided teeth. The effect of organic material left within the root canal was considered to have a limiting effect on the diffusion of the drug.

Heiman, repeating the study in 1971, substituted camphorated parachlorophenol for the aqueous two percent parachlorophenol preparation used by Avny and noted that diffusion was limited to a maximum penetration of 0.4 mm. into the dentin. He concluded, considering the results of Harrison and Avny, that a reevaluation of the choice of camphorated parachlorophenol as a root canal medicament should be considered.

The vaporization of camphorated parachlorophenol has been considered in conjunction with antimicrobial effectiveness studies. Treanor and Goldman showed that the bactericidal efficiency of a root canal medicament depends on its vaporization into the canals. According to their study, camphorated parachlorophenol was capable of diffusing through the apices of extracted teeth producing its greatest effect after seventy-two hours of incubation time. This would seem to indicate that the medicament is volatilized slowly and continuously. Cwikla considered the capillarity of all endodontic medicaments as insignificant but that vaporization of camphorated parachlorophenol did occur by evaluating the microbial inhibition of

innoculated culture media. Wantulok and Brown stated that when used in small quantities, camphorated parachlorophenol is capable of diffusing through the entire length of the root canal with gravity apparently having little or no effect. They also conclude that over-medication by the placement of endodontic medicaments with saturated paper points and flooding of canal is possibly contributing to periapical inflammation.

D. RADIOISOTOPE STUDIES

The selection of the proper radioactive isotope for any biologic study must be made on the basis of localization of the tagged molecule in order to prevent false labeling of neighboring tissue areas. Hughes, in a 1958 study, stated that a beta-ray has a maximum tissue penetrating distance of six microns with approximately fifty percent of the particles traveling less than two microns.

Stahl, Weiss and Tonna in 1969, designed a study on the auto-radiographic evaluation of periapical responses to pulpal injury. Their choice of tritium was based on the same principle that Zach, Topal and Cohen considered in 1969 when reporting on pulpal repair following operative procedures. Since the low beta-ray energy of tritium localizes the label closely on the radiosensitive emulsion and prevents excessive scatter to adjacent tissues, the tritiated molecule is feasible for histological studies.

Avny was the first, in 1970, to use a labeled parachlorophenol molecule. He was able to trace, by autoradiographic evaluation, the volatile components of an aqueous two percent parachlorophenol solution through the dentinal tubules to the cemento-dentinal junction after being sealed in the pulp chamber of extracted teeth and incubated for forty-eight hours. Heiman in 1971, using Avny's experiment design, was able to follow the movement of the tritiated parachlorophenol molecule in camphorated parachlorophenol and observed that dentinal penetration was limited to 0.40 mm.

The studies by both Avny and Heiman indicate the usefulness of tritium labeled studies. When considered on the basis of its low beta energy and suitable half-life, tritium allows the attainment of discrete autoradiographic results within the context of biological studies and histologic procedures necessary for tissue evaluation.

E. DOGS IN ENDODONTIC RESEARCH

The choice of dogs as experimental animals in endodontic research began with Grove in 1912 when he observed the histologic effects of formaldehyde on periapical tissues as the result of the "Buckley" method of treating gangrenous pulps. Coolidge, in 1932 and Orban in 1933, reported that root canal therapy could be successfully performed on dogs and that results obtained from such studies were applicable to human tissue responses.

Dixon and Rickert in 1938, compared the periapical tissue responses of both humans and dogs to root canal sealer and reported identical histologic responses. Gottlieb, Barron and Crook, in their 1950 text Endodontitia, reported the multiple apical foramina characteristic of dog teeth and concluded that if experimental root canal therapy could successfully control the tissue remaining in these foramena, then the methods can be recommended for human teeth without hesitation. Kukidome in 1957, reported on the healing endodontic procedures in infected root canals. Similar studies on periapical healing in dogs after treatment of both infected and sterile root canals were reported by Matsumiya and Kitamura in 1960.

A significant article by Barker and Lockett in 1971 advocated the utilization of the mandibular premolars of dogs for endodontic research. They recommended that the second, third and fourth premolars be used for evaluating pulp capping and root canal procedures. Again, like Orban, the complexity of the apical ramifications is considered by the authors as advantageous in evaluating endodontic procedures because it is thought to make the dog periapex more "sensitive" to root canal manipulation. Perforation of the apex with a rotary drill in an attempt to morphologically duplicate the human tooth apex was found to induce relatively severe apical reactions.

Scoralle, in a 1972 Master's thesis, states that when the root canal preparation is short of the apex in dog teeth, the apex becomes

inadvertantly packed with dentin filings which will inhibit the apical penetration of filling materials. The logical assumption from this statement in regard to endodontic medicaments would be that the volatile and non-volatile components of each will also be confined to the root canal itself and ineffective against periapical infection.

CHAPTER III

MATERIALS AND METHODS

This project was conducted with the intention of duplicating the clinical procedures associated with the routine practice of endodontics. The application of operating techniques and canal medication were strictly adhered to throughout the experiment in an attempt to standardize all collected data. The project began with a single pilot study utilizing the isotopes prepared for Avny's and Heiman's experiments. Procedural complications were realized and corrected while experimental techniques were standardized.

The components of the endodontic medicaments, camphor and paramonochlorophenol, were supplied to a commercial laboratory* for compounding and radiolabeling. The catalytic exchange method was used to prepare tritiated parachlorophenol with a specfic activity of 5 mCi/millimole. An eutectic solution of thirty-five percent parachlorophenol and sixty five percent camphor was compounded to form camphorated parachlorophenol while aqueous two percent parachlorophenol was formulated by maximum dissolution of the parachlorophenol crystals in distilled water yielding approximately aqueous two percent parachlorophenol in the supernatant. Both medicaments were equilibrated to yield the same specific activity.

*Amersham/Searle Corp.; Arlington Heights, Illinois

A total of fifty-four mandibular bi-rooted teeth in nine dogs were instrumented and examined in the study. The maxillary teeth were considered to be excessively difficult to radiograph as well as to histologically prepare in longitudinal sections and were therefore excluded in the study. The animals were selected at the animal research facility* on the basis of age, generally under one year, good coronal tooth condition, no physical abnormalities and availability. The weight range of the animals was from fourteen to twenty-three kilograms (Kg), the average weight being nineteen Kg. Each dog was numbered by collar tags for consistent identification and recorded on its appropriate work sheet, a copy of which is included in the appendix.

Before each scheduled day of laboratory work, all instruments were autoclaved and assembled in order of use. The operating room was completely prepared to receive the animal before attempting general anesthesia. Surgical masks and gowns were worn by those participating that day. The dogs were prepared in an adjacent weighing room designated for anesthesia induction by shaving the forearm and hind leg with shears exposing the large superficial veins.

General anesthesia was always administered by intravenous injection of one cubic centimeter (cc) of sodium pentobarbitol** for each two/Kg of body weight. One cc contained 320 milligrams (mg)

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*Animal Research Facility, Loyola University Medical Center, Maywood, Illinois.

**Holmes Serum Co. Inc., Springfield, Illinois.

of the barbiturate according to the manufacturer. The injection most consistently used was in the large saphonus vein (Fig. 4). The animal was immediately removed to the operating room (Fig. 5).

Pre-operative radiographs were taken prior to any operative procedure for evaluation of pulpal necrosis with obvious apical extension of all selected teeth as well as the patency of the root canal spaces. A portable X-ray unit* utilized at a setting of 90 KVP at 10 Ma for 0.5 seconds provided acceptable radiographs (Fig. 6). All radiographs were developed in a dark box allowing film evaluation within sixty seconds.

The maxillary and mandibular jaws were retracted by the use of a spring-loaded device that attached to the cuspid incisal tips of the side opposite that being instrumented. All work was confined to one side at a time. Originally it was planned to utilize the rubber dam over the teeth selected for instrumentation but experience in previous dog research revealed that salivary flow was almost non-existant while the effects of the anesthetic persisted. Large sterile cotton gauze was used to cover all oral structures except the teeth to prevent dispersement of debris and irrigating solutions. The teeth were swabbed with a seventy percent alcohol solution prior to preparation.

Access cavity preparations were standardized during the pilot study. The coronal portion of the tooth was reduced to within one

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*General Electric - 15 Ma portable unit.

millimeter (mm) of the gingival crest, with a large heatless stone exposing the central pulp horn (Figs. 7 and 8). A number four round bur removed the roof of the pulp chamber and created mechanical undercuts for retention of the temporary cement used at the end of the operation.

Each canal orifice was located with an endodontic explorer in the mesial and distal aspect of the tooth (Fig. 9). A number fifteen or twenty K-type nineteen mm file* was then placed in each canal and a working length radiograph taken (Fig. 10). Review of the radiograph indicated the location of the stop created by the apical morphology (Fig. 11). The final length was estimated by measuring the distance from the tip of the file to the radiographic apex. The canals were then enlarged incrementally until clean dentinal shavings were produced from the apical one-third of the tooth. Great care was taken to insure complete removal of all organic material within the root canal space and to prepare smooth dentinal walls. Diffuse irrigation with five percent sodium hypochloride and two percent hydrogen peroxide was consistently used. Final instrument sizes ranged from number fifty to seventy according to canal morphology and age. No canal enlargement ever extended beyond the natural apical stop prior to this time. The final instrument radiograph was taken and again evaluated before the canal was dried and prepared for apical perforation (Fig. 12).

*Union Broach Co., Inc.; Long Island City, New York.

The use of a long number two engine driven reamer provided simple apical perforation to the specified final length. The actual perforation of the reamer leaving the cemento-dentinal junction was always felt and every attempt to avoid damage to the periapical tissues was taken. A final radiograph was generally taken at the perforation length (Fig. 13), and the canal was then dried of any induced hemorrhage which incidentally occurred in only three of the treated teeth. The mesial root of the second bicuspid was never perforated to provide a histologic control for normal periapical tissue representation. The teeth were then considered ready for medication.

Early efforts to standardize the amount of medicament placed in each canal were found to be impractical due to the inherent problems of different canal size and delivery of micro-amounts of solution. The alternate method used was to pre-weight the cotton pellet and paper point segments so that the mean weight never varied more than ten percent. Introduction of the medicaments was in four different groups, for each medicament.

1. Cotton pellet saturated and then squeeze dried between sterile cotton gauze.

2. Cotton pellet saturated and then excess removed by blotting on sterile cotton gauze.

3. Paper point saturated and then squeeze dried between sterile cotton gauze.

4. Paper point saturated and then excess removed by blotting on sterile cotton gauze.

Different radioactive materials were handled with appropriately marked forceps to prevent contamination of control canals. Paper points were inserted to within three mm of the apical preparation while cotton pellets were placed only in the orifice of each canal. The access cavity was then sealed by a thick mix of zinc oxide and eugenol* and allowed to completely set before beginning the procedure on the opposite side. Table I indicates the teeth utilized in each quadrant and the drug dispensed. The second bicuspid was used as the control tooth for each quadrant receiving no medication.

It was routinely necessary to administer an additional does of barbiturate in order to finish the required procedures. The fastest and easiest method of administration was found to be on the ventral surface of the tongue which is abundant with superficial venous structures (Fig. 14). A maximum of two cc was always sufficient to induce the anesthesia required. After all procedures were finished the animal was returned to its cage and placed on a soft diet to prevent damage of the temporary seal until sacrifice.

The animals were sacrificed at pre-determined intervals of twelve, twenty-four and forty-eight hours. Death was induced by

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*Interstate Dental Company Inc., New York, New York.

TABLE I

TABLE OF EXPERIMENTAL PROCEDURES

SUBJECT	DRUG				VEHICLE				APPLICATION			
	RT.3	RT.4	LT.3	LT.4	RT.3	RT.4	LT.3	LT.4	RT.3	RT.4	LT.3	LT.4
1	C	C	C	C	1	1	2	2	D	D	W	W
2	C	C	C	C	2	2	1	1	D	D	W	W
3	C	C	C	C	1	2	1	2	W	W	D	D
4	P	P	P	P	1	1	2	2	D	D	W	W
5	P	P	P	P	2	2	1	1	D	D	W	W
6	P	P	P	P	1	2	1	2	W	W	D	D
7	P	C	P	C	1	1	2	2	D	D	W	W
8	P	C	P	C	1	1	2	2	D	D	W	W
9	P	C	P	C	1	1	2	2	D	D	W	W

*KEY:

Camphorated Parachlorophenol = C
Parachlorophenol = P
Cotton Pellet = 1

Paper Point = 2
Wet = W
Dry = D

using Totaltox,* a concentrated barbituate, first intravenously, then by intracardiac injection. Block sections were taken through the single rooted first bicuspid and bi-rooted first molar and placed in ten percent formalin within fifteen minutes of sacrifice. The inferior border of the mandible was removed through the center of the mandibular canal to expose the cancellous bone and insure diffusion of the fixative to all structures.

Fixation was continued for seven days with change of solution every twenty-four hours. Decalcification was achieved by a solution of formic acid and sodium citrate. The time interval was often three months before decalcification was complete. Each specimen was then cut into three unequal pieces through the bifurcation of the third and fourth bicuspids and reduced in all dimensions to facilitate imbedding and sectioning. (Figure I diagrammatically illustrates section areas.)

Autoradiography requires sections of a thickness approximating five microns. All sections prepared were from four to six microns depending on the size of the specimen. The sections selected were those demonstrating the greatest mesial-distal diameter of the root canal space (Fig. 2). Nuclear Fast Red, Indigo-Carmine Dye and Picric Acid provided counter staining of the specimen after autoradiographic development of the nuclear track emulsion for seven days.

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*Holmes Serum Co. Inc., Springfield, Illinois.

Collection of data was in two distinct groups: First, dentinal tubule penetration and second, periapical diffusion. A fifty micron grid under oil immersion was used to facilitate counting of the grains. The background or normal grain count was first established on the control tooth of each quadrant by ten random grain counts through different tissue areas and then compared with the background count on each slide.

The tubule penetration segment was conducted by starting at the pulpal wall of the canal and moving the grid one width at a time until no significant counts above the background were recorded. This procedure was repeated ten times at random points along vertical axis of the pulpal wall in each one-half of the remaining root to establish the planes of significant and non-significant penetration (Fig. 3).

The mean counts above background were determined for the various fields in each group. Statistical analysis of the results were carried out using the t-test for comparing two means.

CHAPTER IV

RESULTS

Grain counting of experimental tissue sections was accomplished with the use of a 50 micron square ocular grid for oil immersion (1000x). Background grain counts were recorded and computed for the control tooth of each quadrant and the crestal bone on each experimental slide to insure an accurate estimation of the number of grains not attributable to the radiolabeled medicament.

Grain counting always began just past the pulpo-dentinal junction and proceeded until two lines paralleling the pulpal wall could be established that represented a significant and non-significant level of radiation as the result of medicament penetration. The t-test for comparing two means was used to establish the statistically significant level of radiation above background. The results are tabulated in tables II and III for the camphorated parachlorophenol group and the aqueous two percent parachlorophenol group respectively.

Camphorated parachlorophenol when used in clinical dosages by application of a dried cotton pellet or paper point was found to have limited penetrating ability to a maximum distance of 1.0 mm in one sample. The mean distance of all samples in the cotton pellet group was .53 mm beyond the pulpo-dentinal junction while the mean

distance of the paper point group was .65 mm. Camphorated parachlorophenol when used in quantities above clinical dosages, i.e. the wetted and then blotted cotton pellet and paper point, was found to penetrate to the cemento-dentinal junction in every specimen. The level of significance of dentinal penetration was $P < 0.05$. Accumulated data which is included in the appendix illustrates individual P values and penetration distances. Observation of grain densities in the periodontal ligament and surrounding alveolar bone sections showing diffusion to the cemento-dentinal junction demonstrated grain counts higher than in the adjacent dentin suggesting accumulation of the labeled material after leaving the tooth proper. The design of the experiment did not allow statistical analysis of the periapical diffusion of either medicament. However, distances with grain counts above the mean background count were recorded for each specimen. These results show the camphorated parachlorophenol group to have a mean penetrating distance of .40 mm beyond the apical cementum.

Aqueous two percent parachlorophenol applied by cotton pellet and paper point in either a wet or dried state was consistently found to penetrate to the cemento-dentinal junction at a $P \leq 0.05$ level of significance. Periodontal ligament and alveolar bone grain counts above background were noted as they were in the camphorated parachlorophenol specimens. Apical penetration for the

TABLE II

DIFFUSIBILITY OF CAMPHORATED PARACHLOROPHENOL

CORONAL ONE-HALF

	COTTON PELLET		PAPER POINT	
	DRY	WET	DRY	WET
T	2.19 - 5.07	4.14 - 6.41	2.24 - 3.97	3.22 - 11.32
P	<0.05 - <0.01	<0.01	<0.05 - <0.01	<0.01
MEAN DENTIN PENETRATION	.53 mm	PDL	.65 mm	PDL

APICAL ONE-HALF

T	2.42 - 3.33	2.36 - 6.88	2.17 - 3.97	2.59 - 7.05
P	<0.05 - <0.01	<0.05 - <0.01	<0.05 - <0.01	<0.05 - <0.01
MEAN DENTIN PENETRATION	.52 mm	PDL	.65 mm	PDL

TABLE III

DENTIN DIFFUSIBILITY OF AQUEOUS TWO PERCENT PARACHLOROPHENOL

CORONAL ONE-HALF

	COTTON PELLET		PAPER POINT	
	DRY	WET	DRY	WET
T	2.10 - 4.59	4.13 - 10.00	2.94 - 3.75	2.11 - 7.37
P	<0.05 - <0.01	<0.01	<0.01	<0.05 - <0.01
MEAN DENTIN PENETRATION	PDL	PDL	PDL	PDL

APICAL ONE-HALF

T	2.25 - 3.83	2.20 - 4.01	2.48 - 4.30	2.14 - 6.60
P	<0.05 - <0.01	<0.05 - <0.01	<0.02 - <0.01	<0.05 - <0.01
MEAN DENTIN PENETRATION	PDL	PDL	PDL	PDL

parachlorophenol group was determined to be approximately .42 mm beyond the apical cementum. Table IV represents these findings for both medicaments.

Grain counts along the length of the periodontal ligament appeared to be greater in the parachlorophenol group than in the camphorated parachlorophenol group. Both groups however did show accumulations of grains in the ligament space greater than in the adjacent dentinal tubules. Alveolar bone penetration except for periapical tissue was considered to be too varied to be significant.

A comparison of means demonstrates that the wet sample groups were generally of a greater magnitude than the dry sample groups with resultant P values in the wet group approaching the $P < 0.01$ level of significance for either medicament.

The existence of a qualitative relationship between undebrided organic tissue remaining in the instrumented root canal and both drugs was confirmed. Accumulations of grains in uncountable numbers were noted in all uninstrumented areas of the canal as well as free tissue remnants. Grain counting occasionally revealed the presence of an accessory canal cut tangentially in sectioning with vital tissue remaining. Examination revealed large quantities of medicament within the organic substance of the canal.

The inflammatory responses of the periapical tissue at the area of perforation appeared to be dependent upon the time of sacrifice

TABLE IV

MEAN APICAL PENETRATION OF CAMPHORATED PARACHLOROPHENOL AND AQUEOUS TWO PERCENT PARACHLOROPHENOL

Camphorated Parachlorophenol

Cotton Pellet		Paper Point	
DRY	WET	DRY	WET
.41 mm	.39 mm	.36 mm	.44 mm

Parachlorophenol

Cotton Pellet		Paper Point	
DRY	WET	DRY	WET
.39 mm	.45 mm	.34 mm	.50 mm

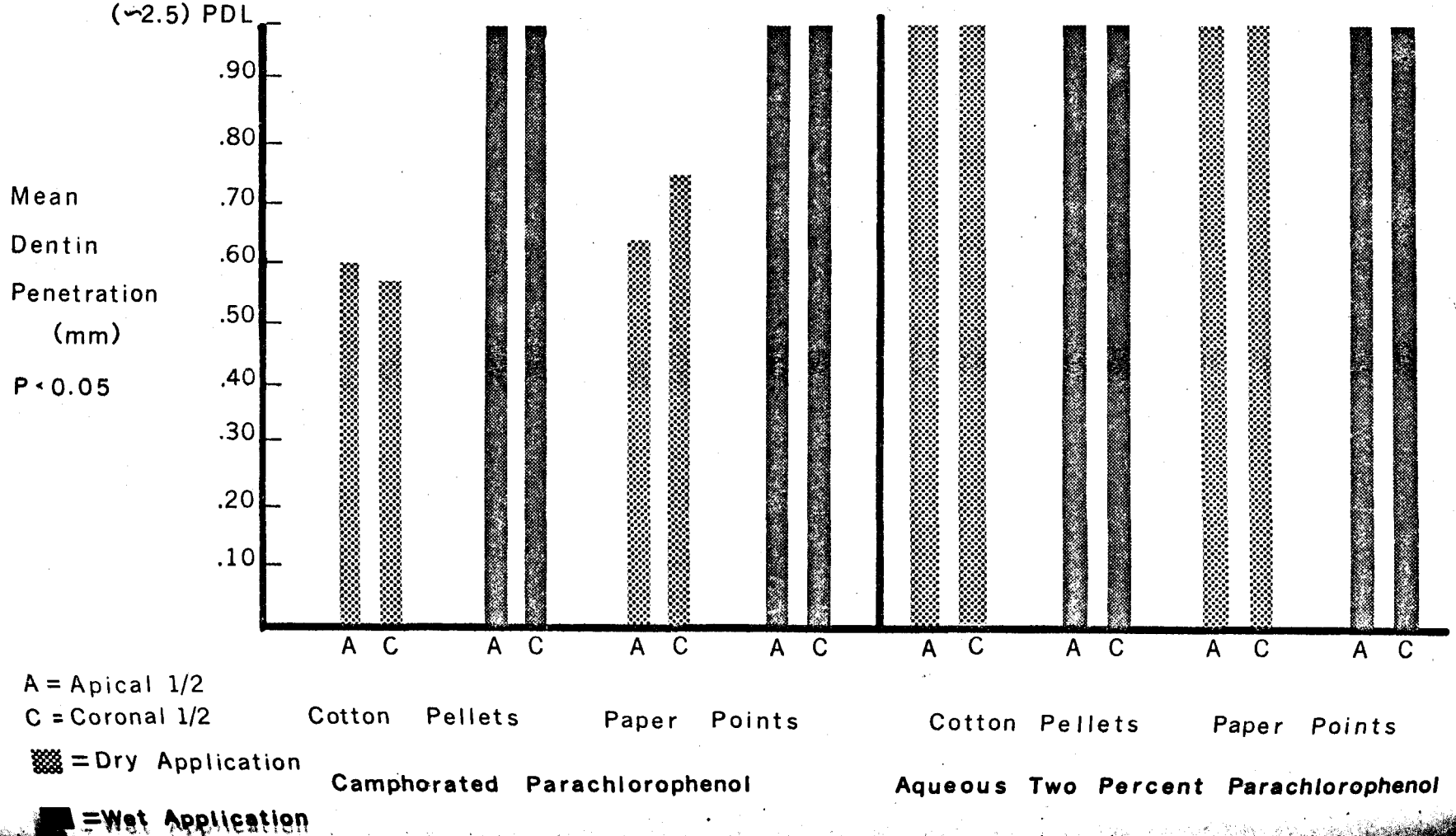
and length of perforation. The presence of acute inflammatory cells was routinely seen along with extravasated blood cells especially in those teeth where hemorrhage had been previously noted. Generally no blood was found within the confines of the prepared root canal above the perforation.

The relationship of time and penetrability was not statistically evident since diffusion to the periodontal ligament occurred in as little as twelve hours in those groups demonstrating transdentin penetration. No quantitative difference could be illustrated between the three intervals of sacrifice. Those groups showing limited penetrating ability varied insignificantly among themselves and the timed groups.

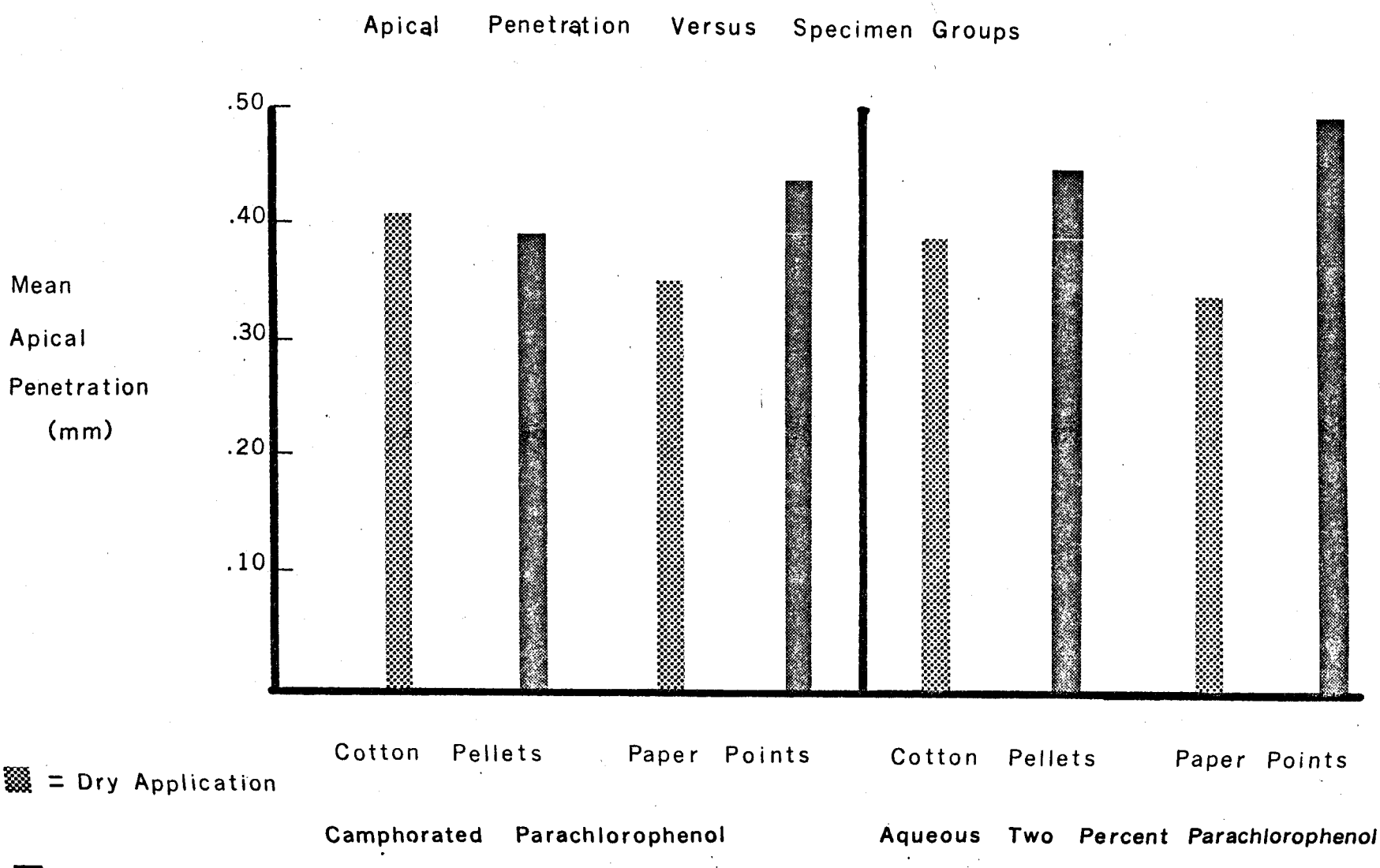
Graphs 1 and 2 illustrate medicament penetration in dentinal tubule and periapical areas, respectively.

Graph 1

Dentin Penetration Versus Specimen Groups



Graph 2


Apical Penetration Versus Specimen Groups
Mean Apical Penetration (mm)
.50
.40
.30
.20
.10
Cotton Pellets
Paper Points
Cotton Pellets
Paper Points
= Dry Application
Camphorated Parachlorophenol
Aqueous Two Percent Parachlorophenol
= Wet Application

CHAPTER V

DISCUSSION

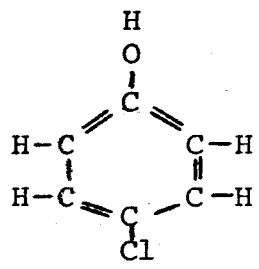
The utilization of chemical medicaments as an adjunct to any clinical discipline must be justified on the basis of its therapeutic value, toxicity to the host and knowledge of its exact mode of action. Camphorated parachlorophenol has been routinely accepted and employed in the management of endodontic cases for almost three-quarters of a century without satisfying one of these requirements. Poorly designed investigations on the diffusion characteristics of this drug have seemingly been universally accepted as well as misunderstood so that its clinical usage is based on what could be described as intellectual ignorance. Avny, in 1970, and Heiman in 1971, initiated the first valid studies on the dentinal penetration of parachlorophenol in both aqueous and camphorated forms respectively. It was the purpose of this study to attempt the first *in vivo* observation of the diffusibility of this commonly used medicament.

One aspect of most endodontic therapy is the biomechanical cleansing of the dentin walls in conjunction with canal enlargement. This procedure is designed to eliminate not only organic debris but also to reduce the bacterial content of the canal if present. It is upon this basis that the current trend of eliminating the culture appointment is founded. It is generally agreed that the attainment

of canal sterilization is seldom if ever achieved and dependence on antiseptic agents is normally used in conjunction with canal preparation to reduce the infected state to a level of clinical insignificance. Inter-appointment reinfection of the root canal space may occur by two mechanisms, excluding the mishap of temporary seal leakage, first, the migration of microorganisms from the periapical area to the instrumented canal and second, failure to remove or destroy those microorganisms present within the length of the dentinal tubules and irregularities of the root canal system.

Harrison, in 1969, reported the severe toxic qualities of camphorated parachlorophenol which was essentially due to the effect of the camphor base. He was then led to investigate the antimicrobial effectiveness of parachlorophenol since it alone was reportedly the active agent of the drug. He found that a one percent aqueous solution of parachlorophenol represented a ninefold increase beyond that required for bacterial inhibition. The elimination of camphor, being the purported vehicle for diffusion of camphorated parachlorophenol, necessitated investigation to determine if dentinal tubule permeability of the parachlorophenol molecule in a water base did occur and to what extent when compared to a camphor base.

The structural formula of parachlorophenol is shown below.



Commercial preparation of the test solutions was accomplished by the addition of the tritiated parachlorophenol molecule to either camphor or distilled water so as to yield the identical specific activity per millimole for each medication. This was considered essential to the evaluation of results since the exact volume of fluid used in each canal was impossible to quantitate. Pilot studies attempting the use of a micro-pipette proved to be unsuccessful and it was decided to standardize volumes by the use of pre-weighted paper points and cotton pellets which were moistened in the identical manner for all specimens. Although this was not theoretically perfect, the technique was considered practical and closely related to clinical practice.

The number of specimens for each medicament category was decided on the basis of clinical implication. The use of squeezed dried cotton pellets was considered most applicable and was examined in twenty-four roots of twelve dogs. On the basis of Heiman's study in 1971 in which he showed limited penetrating ability of camphorated parachlorophenol, a maximum dose of each medicament on a paper point was also introduced into twenty-four roots in twelve dogs to study penetration following overmedication. Each of the four remaining categories included six samples each. Great care was taken to control radioactive contamination in all phases of the experiment. The first six animals were medicated with one or the other medicament but not both. The last three animals were medicated with each medicament.

Decalcification was done in separate containers as was the final exposure and developing of the tissue specimens for autoradiography. Sacrifice to developing time was impossible to maintain at a standardized length due to differing rates of decalcification which apparently varied with the age of the animal. Autoradiography was fixed however at exactly eighty-four hours.

The results of this study indicate that at clinical dosages using either a paper point or cotton pellet, aqueous two percent parachlorophenol has the ability to penetrate the entire distance of the dentinal tubules. Levels of significance often approximating and exceeding $P < 0.01$ were consistently observed just pulpalward of the cemento-dentinal junction. Camphorated parachlorophenol on the other hand, is limited to a calculated mean distance of approximately .60 mm from the pulpal wall or less than one-fifth the entire mean dentinal width of the tooth.

The obvious clinical impact of these findings is on the validity of the use of camphorated parachlorophenol especially if canal disinfection and sterilization are to remain criteria for final obturation of the root canal. Its role in the ultimate success or failure of the effected tooth is a widely debated topic but few clinicians would challenge the advisability of controlling the bacteriologic status of any endodontically involved tooth if possible. The statement can therefore be made that if greater dentin penetration is

possible by a less toxic and equally effective antiseptic agent than the one presently available, the use of the latter should be discontinued.

When the volume of medicament is increased to overdosage levels by application of wet paper points and cotton pellets, a change in penetration distances occurs. In overdose specimens, the camphorated form as well as the aqueous form was found at significant levels near the cemento-dentinal junction to $P < 0.01$. The significance of this data is in the manner of interpretation and not the obvious statistical figures. Since a smaller dosage of the aqueous form was found to penetrate to the cemento-dentinal junction, it is not surprising that the overdose specimens demonstrate the same results. It would then appear that larger dosages of camphorated parachlorophenol increases its diffusibility possibly through the effects of a concentration gradient and availability factor. It must be remembered however that camphorated parachlorophenol is not a compound but rather an eutectic solution, the definition of which requires that the components be only mutually soluble in each other. No study had ever shown that the known volatility of camphor can in any way mediate the diffusion of parachlorophenol.

The in vivo circumstances of this experiment in conjunction with the presence of an artificial apical communication of the root canal space and periapical tissues suggests the liklihood of an increase of intracanal moisture. Since free parachlorophenol molecules are

available from the eutectic solution, an aqueous form of parachlorophenol may have formed and could be responsible for the penetration observed in those samples where camphorated parachlorophenol was in excess. It is impossible to validate this hypothesis from the results of this study due to the fact that the camphor segment of camphorated parachlorophenol is unmarked and undetectable by any histologic evaluation.

The results of this study closely parallel those described by Avny and Heiman. Their in vitro application of medicaments at close to clinically acceptable doses showed limited movement of camphorated parachlorophenol to only .40 mm and transdentinal movement of aqueous two percent parachlorophenol. The only discrepancy between their techniques was a higher specific activity used by Avny which may have been thought to cause greater apparent diffusion than seen by Heiman. In this study however, both medicaments were equilibrated so that increased grain counts represented increased parachlorophenol penetration. Heiman also stated that the camphorated parachlorophenol appears to stay predominantly in the area where it is placed. His samples however, did not have the available moisture content as the specimens of this study and represented diffusion in a dry environment. An experiment designed to investigate the reaction of camphorated parachlorophenol and different concentration levels of humidity would prove valuable in the evaluation of parachlorophenol diffusion.

Another aspect of diffusibility is the movement of the medicament to and through the apical foramen. Several studies have shown that *in vitro* volatilization of camphorated parachlorophenol does occur but no *in vivo* study has ever confirmed this fact. This experiment was not designed so that statistical data could confirm or reject such phenomenon but did yield evidence indicating the occurrence of limited apical diffusion. The inherent problem of gathering statistical information for periapical grain counting was in the histologic preparation of the specimens. All teeth were taken in block section before placement in formalin. The most representative sections for studying dentin penetration of the medicament were considered to be those at the greatest mesial-distal diameter of the root canal. Since the apical perforation seldom coincided with the long axis of the root canal, it was decided that a true representation of the level apical penetration was not available on the same slide. Several sections showed what appeared to be the result of an accurately aligned tissue block on the microtome when examining the canal length and diameter but failed to show the apical perforation. It must be remembered that the instrument used for perforation was small, a number 20 reamer, as well as the fact that there were usually more than one root on each slide. Since no two roots were ever in the same exact vertical plane, the possibility of attaining representative sections of necessary root canals and associated perforations was highly unlikely. Therefore, rather than report statistical analysis

of invalid data, it was decided that too few slides demonstrated periapical tissue directly below the sight of perforation and only a comparison of grain counts at different distances from the apical cementum with the mean background count would be reported. The greatest consistent depth of penetration was at a mean distance of .50 mm from the apical cementum and were in the specimen group medicated with paper points after wetting with aqueous two percent parachlorophenol. The other groups showed distances ranging from .34 mm to .45 mm. The relationship of parachlorophenol and the apical vasculature is unknown as regards its localization and removal into the peripheral circulation. When comparing the numbers of grains needed for statistical significance in dentin penetration, apical penetration was not demonstrable within the limits of this study. Clinical application of this fact is based on the question of whether or not apical diffusion is desirable. Assuming normal immunological responses, it is seldom necessary to deliver the medicament out of the tooth regardless of its toxicity. Its prime function must be the maintenance of the bacteriological environment in the root canal system below clinical significance. The management of any infection whose virulence is beyond the natural defenses of the host should not be dependent upon possible diffusion of an intracanal drug but treated directly by the topical application or systemic medication.

Avny and Heiman both observed an obvious accumulation of grains in association with remaining pulpal tissue. Avny felt that the

parachlorophenol had a definite affinity for protein which is consistent with the properties of most phenol derivatives. Such a characteristic must be considered to produce a limited penetrating capacity of the medicament if the canal contents are not thoroughly removed. The present study observed that when either medicament was available to organic tissue whether in the main canal (Fig. 15), an accessory canal or the periodontal ligament tissue (Fig. 16), there appeared to be a localization of the activated grains. Two significant observations may be derived accordingly. First, it was found that whenever an accessory canal was found with intact vital tissue, large numbers of grains were apparent to the level of statistical significance. The implication of this is the possibility of some antiseptic effect on accessory canals unavailable to instrumentation and irrigation which may provide an additional margin of clinical success in infected cases. Second, greater average grain counts were observed along the length of the periodontal ligament than in the adjacent dentin. The significance of this observation is in the fact that the higher grain counts were observed in those specimens representing aqueous two percent parachlorophenol medication rather than camphorated parachlorophenol. This seems to be another indication of the superior penetrating ability of the aqueous form. Localization is probably due to protein precipitation by the phenolic ring.

Histologic evaluation of the periapical inflammatory response disclosed varying degrees of acute cellular infiltration. Control

specimens having been only instrumented and not perforated showed responses limited to the contents of the multiple apical foramena (Fig. 17). Perforated specimens demonstrated inflammation which varied with the length of penetration into the alveolar bone and the time interval before sacrifice (Fig. 18). No correlation could be made between the medicament used and the degree of inflammation present. The only valid observation which could be made was the occurrence of high grain counts in the tissue of the multiple apical foramena. The effect of peripheral edema and increased apical tissue pressure due to perforation may account for the absence of high levels of activated grains indicating periapical diffusion. It is unlikely that the periapical inflammation, regardless of its nature, had any effect on dentinal tubule penetration.

The implications of this study are:

1. The diffusibility of aqueous two percent parachlorophenol is significantly greater than camphorated parachlorophenol when used in volumes approximating clinical dosages.

2. The production of an aqueous form of parachlorophenol within the root canal space medicated with camphorated parachlorophenol is a theoretical possibility and may be responsible for the clinical effectiveness of the commercial drug.

3. Localization of parachlorophenol has a qualitative relationship to the effectiveness of mechanical debridement of the root canal system. Incomplete canal preparation leaving organic tissue remnants

may result in a limitation of the maximum penetrating ability of the medicament.

4. Transdentinal penetration of parachlorophenol is possible within twelve hours of application when the same dose is allowed to remain in the closed tooth.

5. Accumulation of parachlorophenol appears to occur in the periodontal ligament. Higher average grain counts in the periodontal ligament adjacent to the cemento-dentinal junction in those groups medicated with aqueous two percent parachlorophenol suggest more effective penetration.

6. The techniques used in the studies by Avny and Heiman are reproducible and their results have been verified in vivo.

Considering the research of the past five years beginning with Harrison, it is my suggestion that the use of aqueous two percent parachlorophenol be instituted as a superior replacement for camphorated parachlorophenol. As aspects of this drug, aqueous two percent parachlorophenol, particularly its toxicity, antimicrobial effectiveness and diffusibility have been shown to be more effective with fewer detrimental side effects than camphorated parachlorophenol, the present standard drug of choice. In light of these findings, three quarters of a century of indiscriminate use of camphorated parachlorophenol should be terminated.

CHAPTER VI

SUMMARY

The *in vivo* diffusibility of camphorated parachlorophenol and aqueous two percent parachlorophenol was compared by autoradiographic evaluation. Aqueous two percent parachlorophenol demonstrated complete dentin penetration at clinically equivalent dosages while camphorated thirty-five percent parachlorophenol was limited to a mean dentinal tubule distance of 0.58 mm. The results of this study substantiate the findings of previous *in vitro* investigations and suggest that the use of camphorated parachlorophenol be discontinued and replaced by aqueous two percent parachlorophenol in routine endodontic therapy.

FOOTNOTES

1. Amersham/Searle Corp., Arlington Heights, Illinois.
2. Animal Research Facility, Loyola University Medical Center, Maywood, Illinois.
3. Holmes Serum Co. Inc., Springfield, Illinois.
4. General Electric - 15 Ma Portable Unit.
5. Union Broach Co. Inc., Long Island City, New York.
6. Interstate Dental Company Inc., New York, New York.
7. Holmes Serum Co. Inc., Springfield, Illinois.

CHAPTER VII

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CHAPTER VIII

APPENDIX

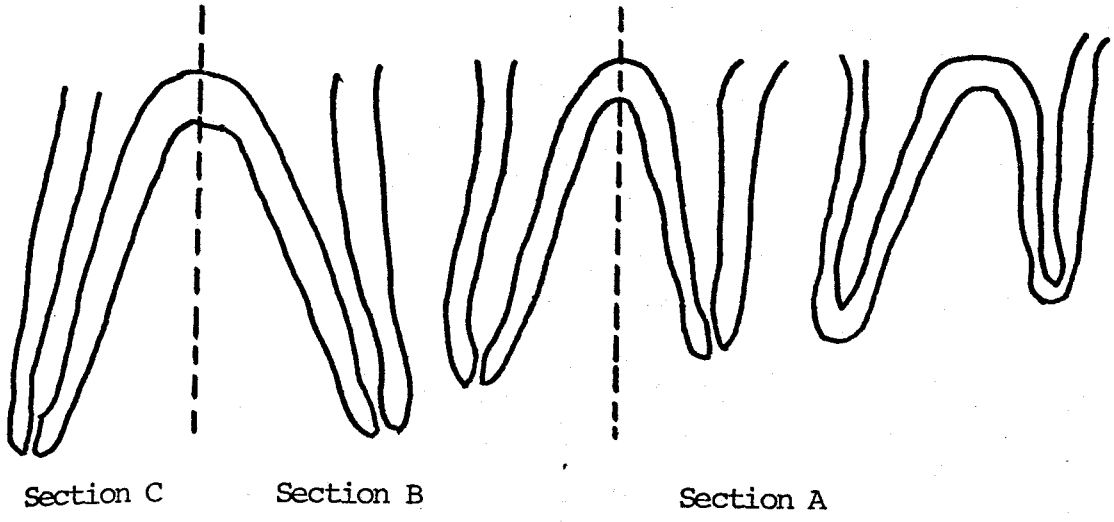
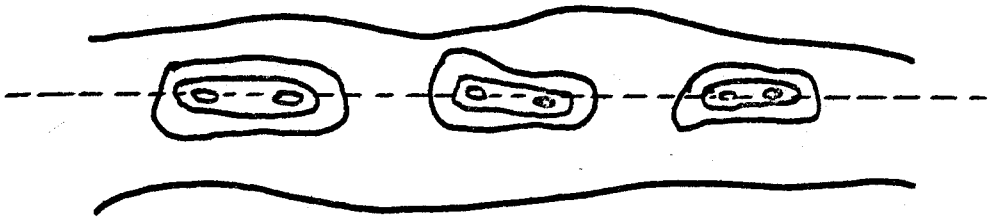
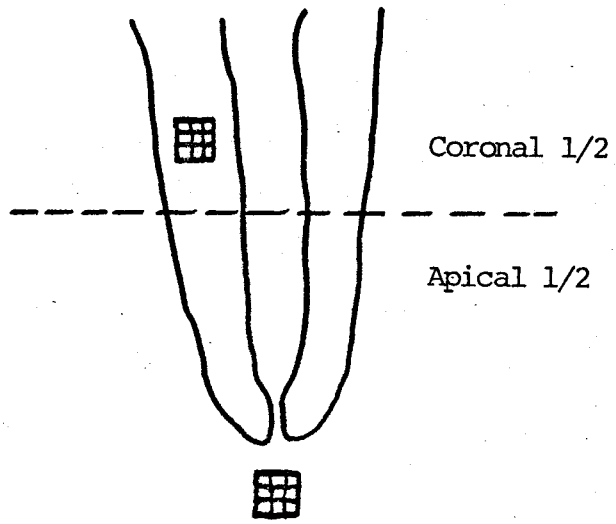


Fig. 1



Plane of Histologic Sectioning

Fig. 2



Division of Root in Equal Segments

Fig. 3

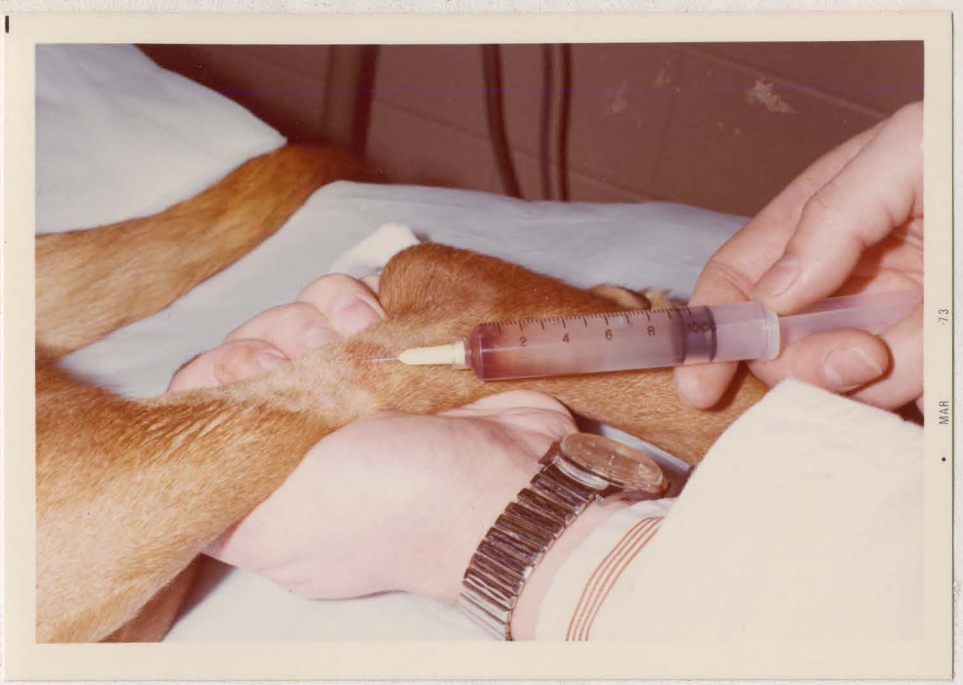


Fig. 4 Induction of General Anesthesia

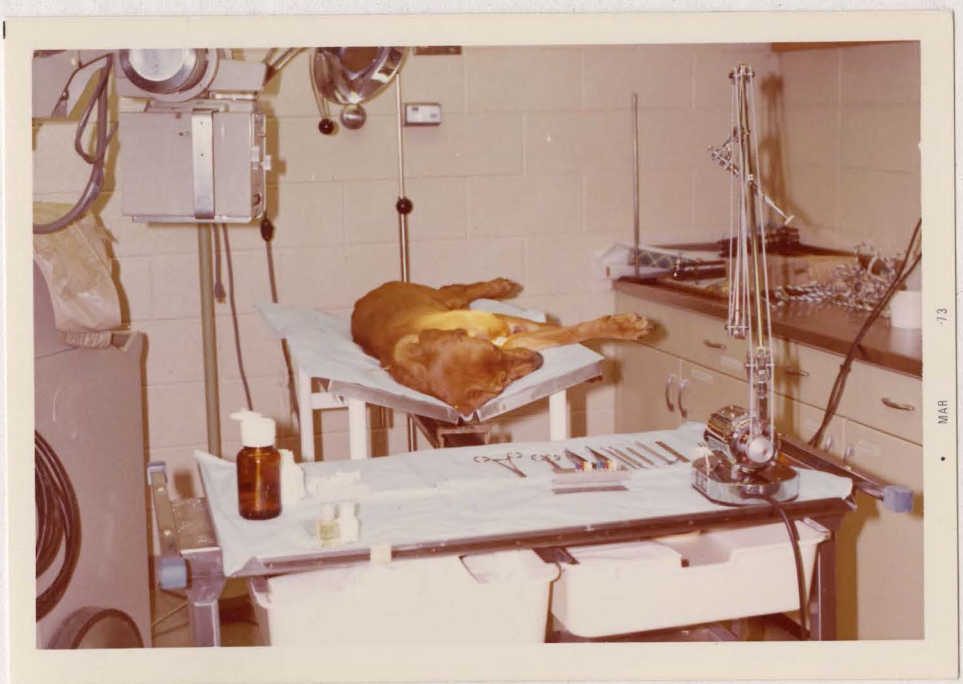


Fig. 5 Animal Prepared for Procedure

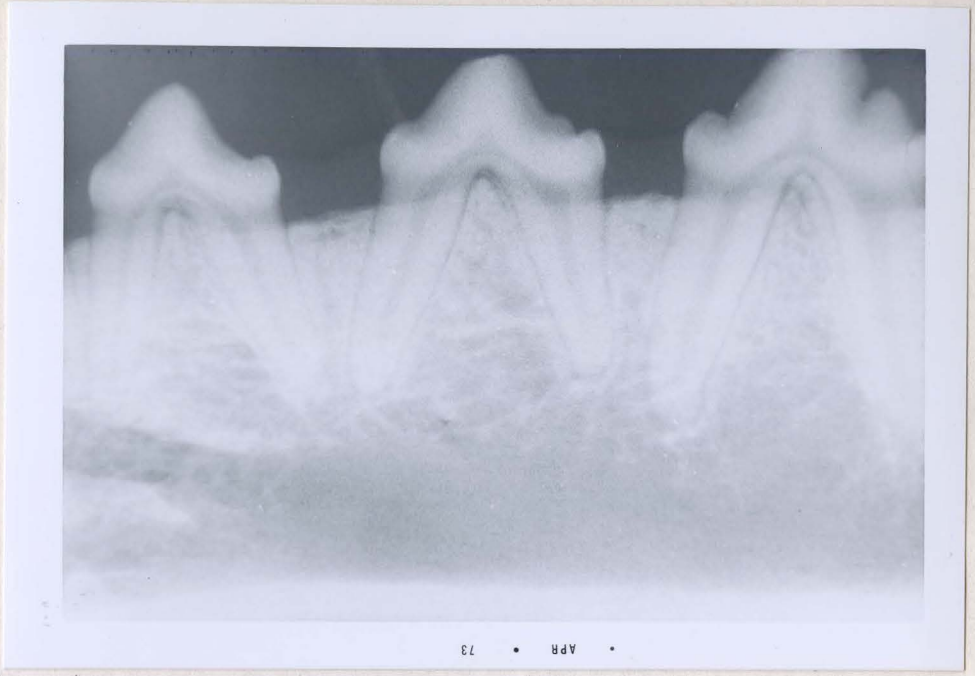


Fig. 6 Pre-Operative Radiograph

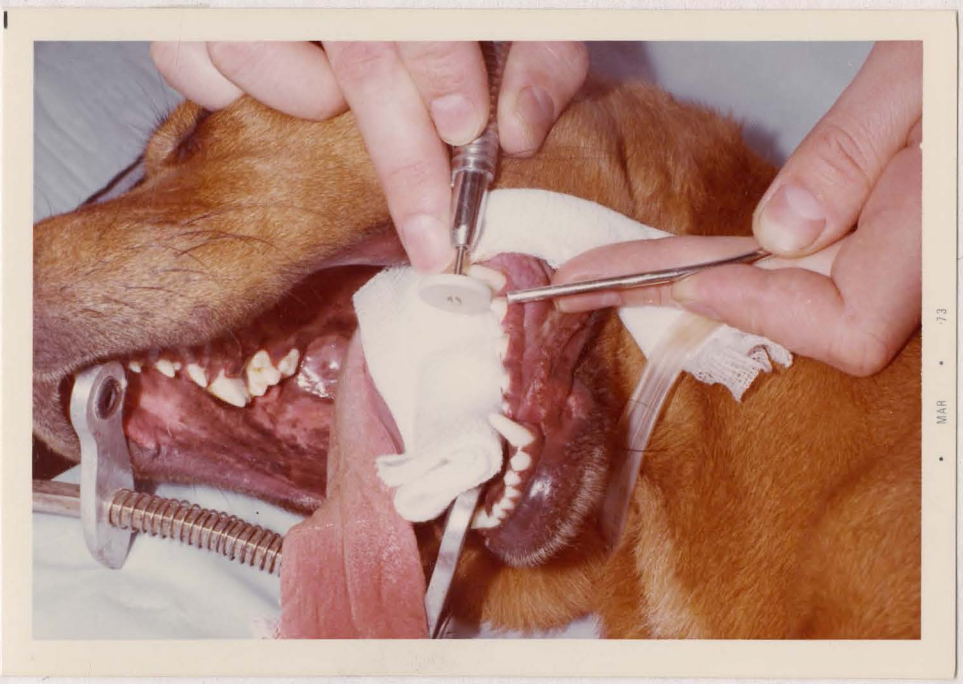


Fig. 7 Access Cavity Preparation

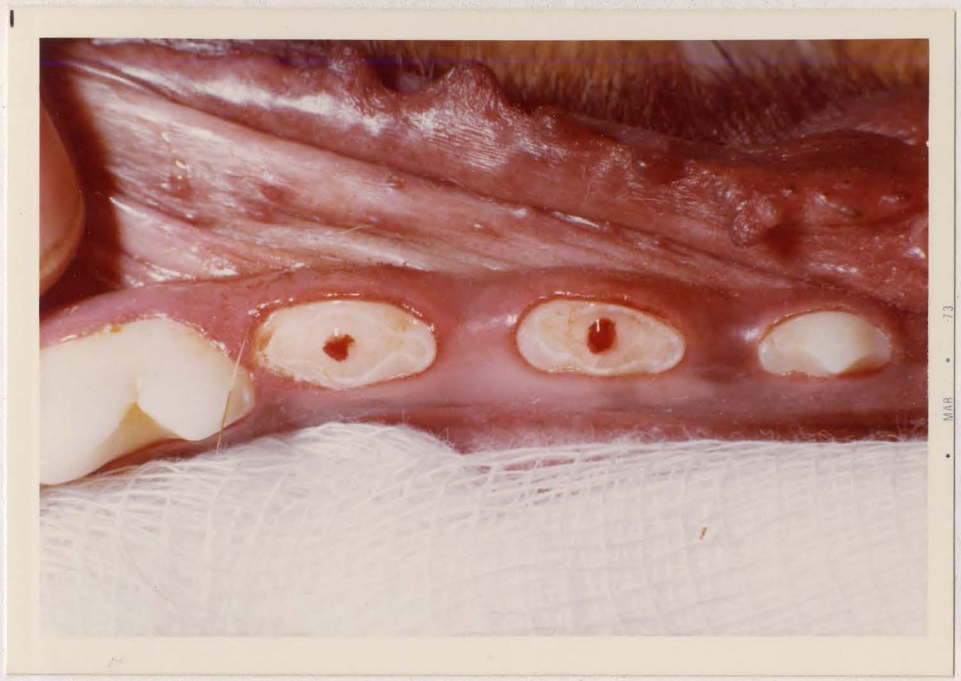


Fig. 8 Central Pulp Horn Exposure

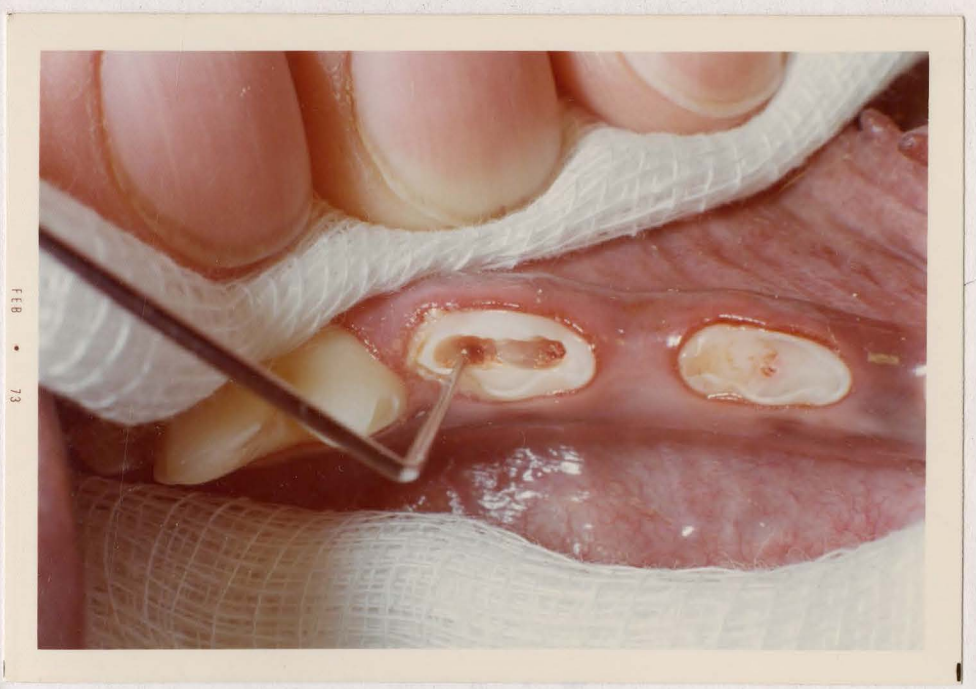


Fig. 9 Canal Isolation

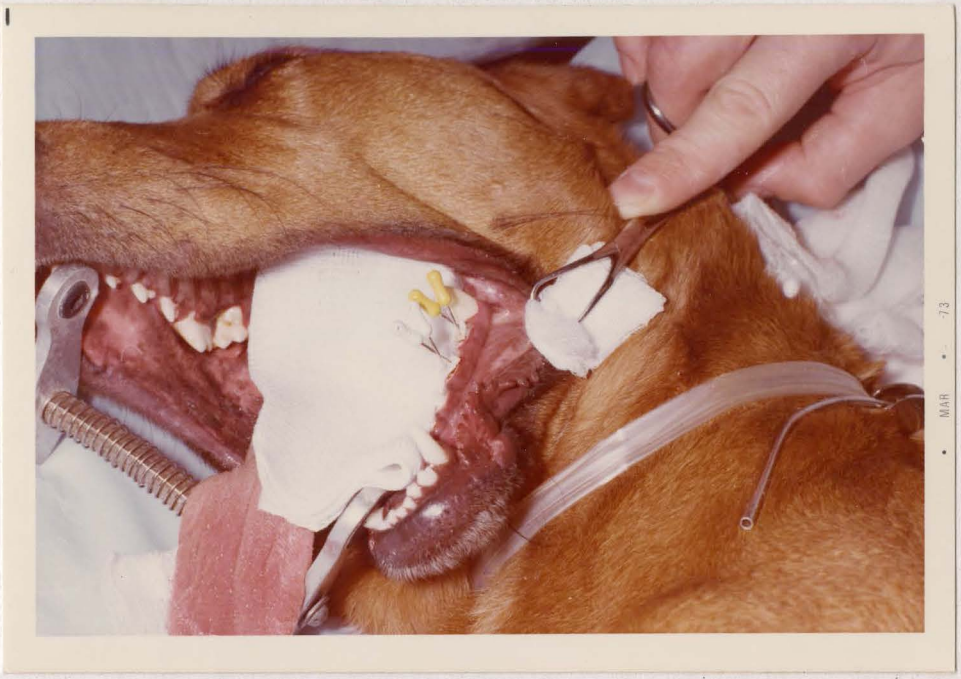


Fig. 10 Initial Instruments in Place

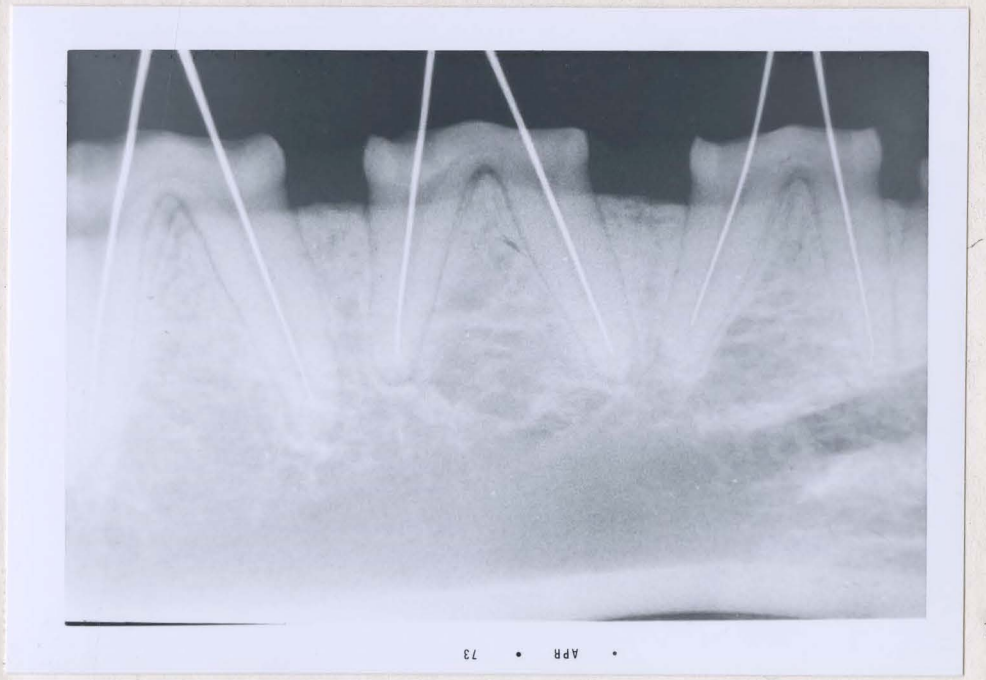


Fig. 11 Initial Instrument Radiograph Showing Location of Natural Dentin Stop

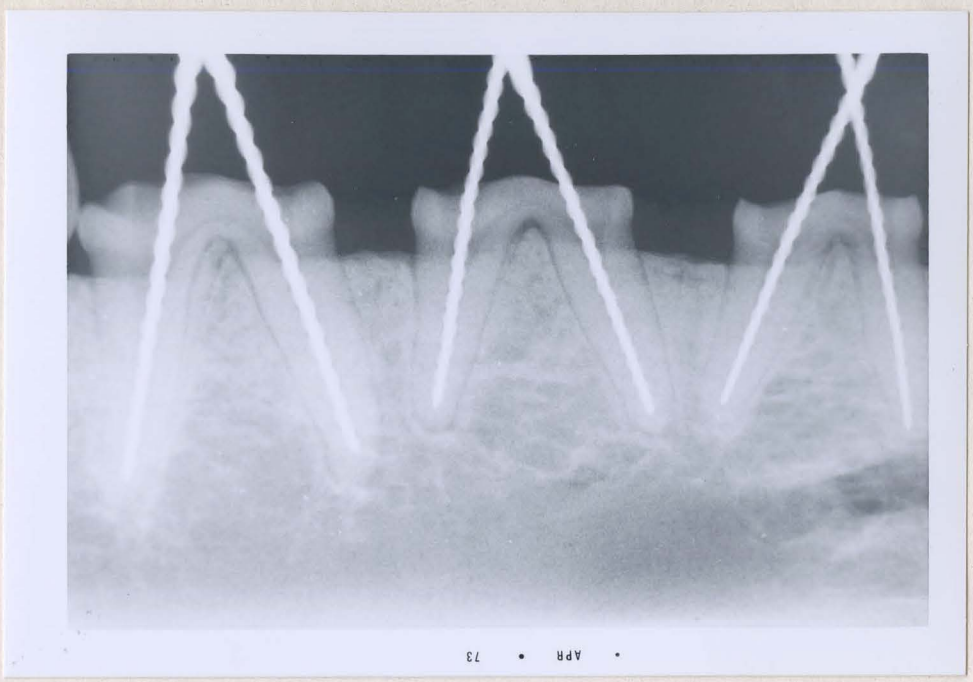


Fig. 12 Final Instrument Radiograph Showing Preparation Short of Apex

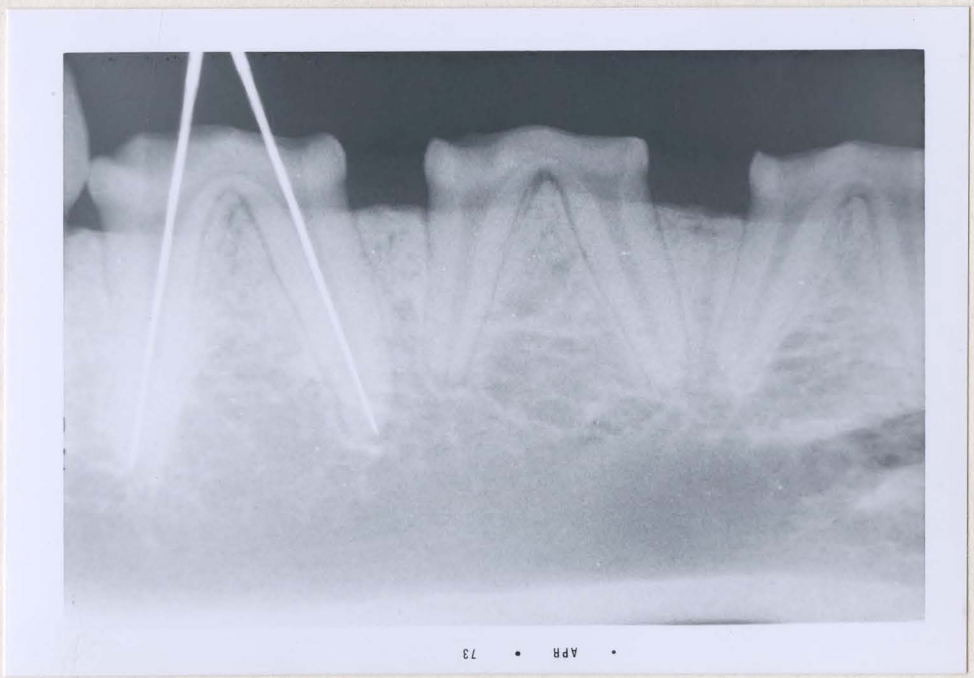


Fig. 13 Apical Perforation Radiograph

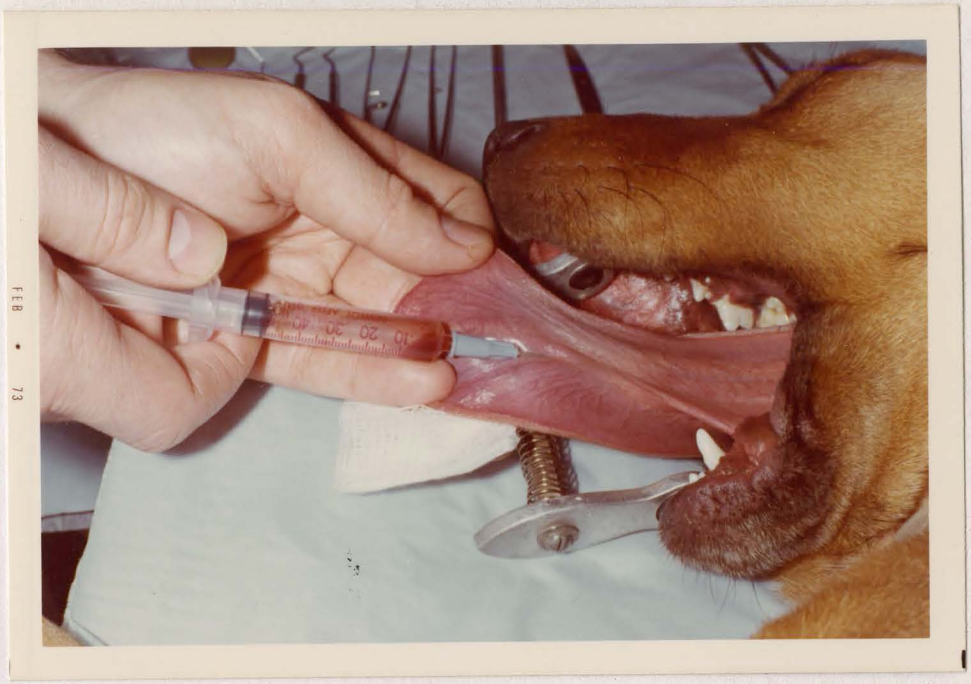


Fig. 14 Ventral Tongue Injection

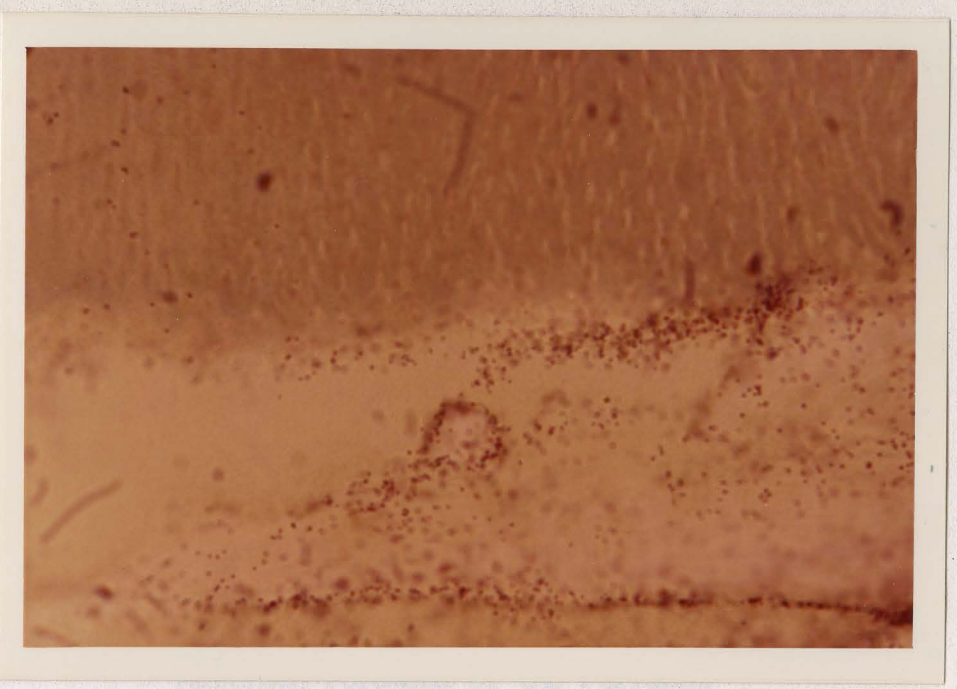


Fig. 15 Localization of Activated Grains in Remaining Organic Tissue Found in the Main Canal

Fig. 16 Localization of Activated Grains in the Periodontal Ligament

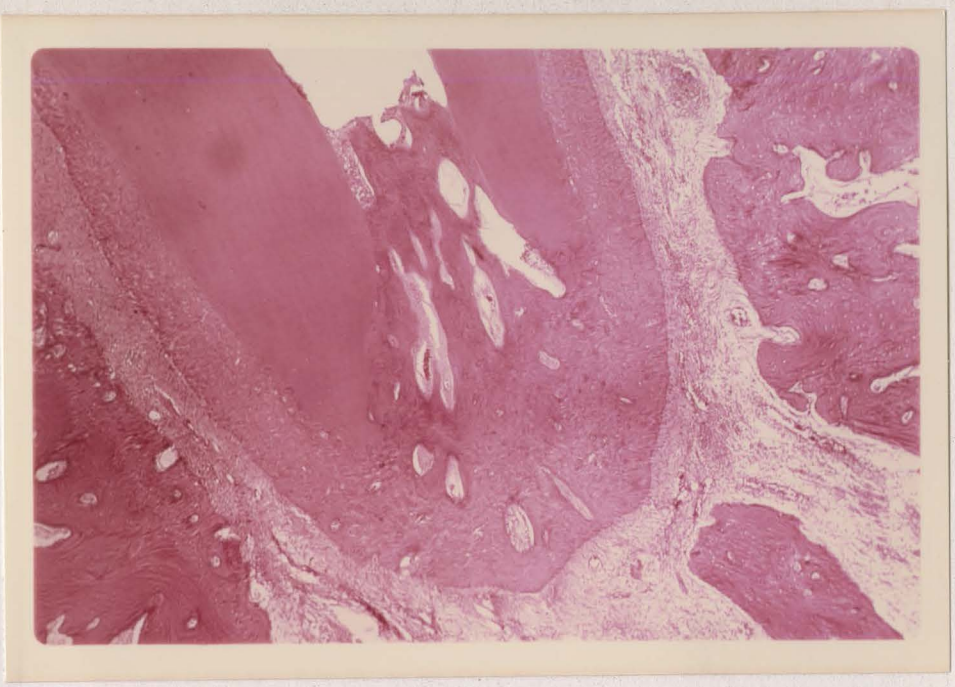


Fig. 17 Multiple Foramena of the Periapex of the Dog Bicuspid

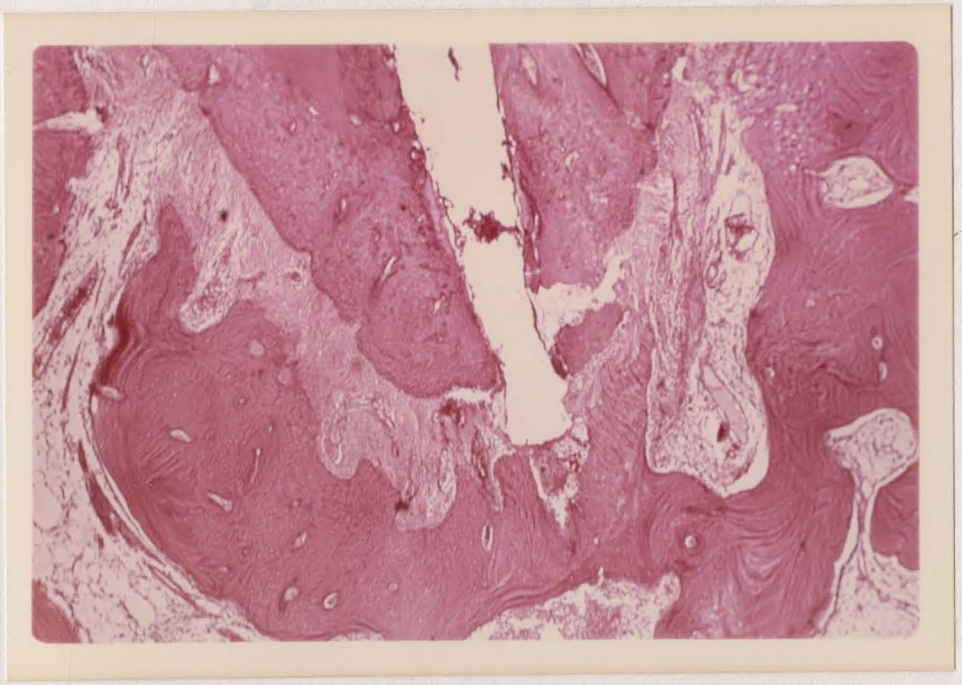


Fig. 18 Perforation Periapex of the Dog Bicuspid

LEGEND:

$\overline{BC}$	=	Mean Background Grain Count
$\overline{GC}$	=	Mean Sample Grain Count
T	=	T value
P	=	Level of Statistical Significance
D	=	Dentin Penetration in Millimeters
PDL	=	Periodontal Ligament
$\overline{AP}$	=	Mean Apical Penetration
S	=	Significant
NS	=	Nonsignificant

TABLE I

STATISTICAL DATA FOR CAMPHORATED PARACHLOROPHENOL
UTILIZING THE COTTON PELLET IN A DRY APPLICATION STATE

CORONAL ONE-HALF

	1		2		3		4		5		6		7		8		9		10		11		12	
	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS
$\overline{BC}$	4.3		4.3		4.3		4.3		4.7		4.7		6.0		6.0		4.8		4.8		4.9		4.9	
$\overline{GC}$	7.1	6.0	7.6	4.9	7.8	5.9	8.9	5.3	10.0	5.3	9.4	5.2	10.1	5.4	11.4	6.3	11.2	5.3	12.8	6.4	11.2	6.1	12.4	6.1
T	2.46	1.36	4.78	.66	5.07	2.01	2.74	1.12	2.93	.48	2.17	.49	2.20	.6	2.56	.32	2.19	.4	2.99	1.31	2.79	1.03	4.09	.94
P	0.05		0.01		0.01		0.02		0.01		0.05		0.05		0.02		0.05		0.01		0.02		0.01	
D	.5	.7	.3	.4	.5	.6	.5	.6	.6	.7	.8	1.0	.4	.5	.4	.5	.6	.8	.6	.8	.6	.7	.6	.7

APICAL ONE-HALF

$\overline{BC}$	4.3		4.3		4.3		4.3		4.7		4.7		6.0		6.0		4.8		4.8		4.9		4.9	
$\overline{GC}$	7.3	3.9	6.3	4.5	7.7	5.3	7.9	5.9	9.7	4.9	9.1	5.4	11.7	5.7	12.3	6.3	11.7	5.8	12.2	6.5	9.9	6.7	11.4	5.9
T	2.80	.40	2.94	.29	2.78	1.45	2.88	1.73	2.42	.14	2.95	.48	3.24	.32	3.02	1.15	4.05	.74	3.33	1.51	3.01	1.34	2.91	.78
P	0.02		0.01		0.02		0.01		0.05		0.01		0.01		0.01		0.01		0.01		0.01		0.01	
D	.5	.7	.3	.4	.4	.5	.4	.6	.6	.7	.8	1.0	.4	.5	.4	.5	.6	.8	.6	.8	.6	.7	.6	.7
$\overline{AP}$	.15		---		.40		.50		.25		.50		.20		.50		.45		.50		.50		.50	

TABLE II

STATISTICAL DATA FOR CAMPHORATED PARACHLOROPHENOL UTILIZING THE COTTON PELLET IN A WET APPLICATION STATE

CORONAL ONE-HALF

	1		2		3		4		5		6	
	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS
$\overline{BC}$	4.7		4.7		4.7		4.7		5.5		5.5	
$\overline{GC}$	26.7		32.4		33.8		33.3		35.6		29.3	
T	4.29		6.41		5.96		5.35		4.14		4.34	
P	0.01		0.01		0.01		0.01		0.01		0.01	
D	PDL		PDL		PDL		PDL		PDL		PDL	

APICAL ONE-HALF

$\overline{BC}$	4.7	4.7	4.7	4.7	5.5	5.5
$\overline{GC}$	2.10	25.0	29.1	30.4	27.5	26.3
T	0.05	0.01	0.01	0.01	0.01	0.02
P	PDL	PDL	PDL	PDL	PDL	PDL
D	---	.35	.30	.50	.40	.40

TABLE III

STATISTICAL DATA FOR CAMPHORATED PARACHLOROPHENOL UTILIZING THE PAPER POINT IN A DRY APPLICATION STATE

CORONAL ONE-HALF

	1		2		3		4		5		6	
	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS
$\overline{BC}$	5.8		5.8		5.8		5.8		4.7		4.7	
$\overline{GC}$	14.5	5.2	9.2	5.2	9.5	6.0	10.2	6.6	9.4	5.2	10.1	4.8
T	3.97	.36	3.43	.79	2.24	2.9	3.41	.96	2.17	.49	2.61	.10
P	0.01		0.01		0.05		0.01		0.05		0.02	
D	.6	.7	.5	.7	.6	.7	.6	.8	.8	1.0	.8	.6

APICAL ONE-HALF

$\overline{BC}$	5.8		5.8		5.8		5.8		4.7		4.7	
$\overline{GC}$	13.1	6.4	7.9	4.6	8.6	5.2	11.1	6.1	9.1	5.4	8.5	5.5
T	3.23	.21	3.81	2.07	2.14	.74	2.83	.36	2.95	.48	4.52	1.92
P	0.01		0.01		0.05		0.02		0.01		0.01	
D	.6		.5		.6		.6		.6		.8	

TABLE IV

STATISTICAL DATA FOR CAMPHORATED PARACHLOROPHENOL UTILIZING THE PAPER POINT IN A WET APPLICATION STATE

CORONAL ONE-HALF

	1		2		3		4		5		6		7		8		9		10		11		12	
	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS
$\overline{BC}$	4.7		4.7		4.7		4.7		5.5		5.5		5.2		5.2		5.8		5.8		4.7		4.7	
$\overline{GC}$	18.1		16.5		19.9		17.9		39.0		37.3		9.2		11.4		14.5		14.4		13.6		10.9	
T	10.1		6.86		11.34		9.85		4.53		5.14		3.22		5.74		3.23		4.09		3.85		4.30	
P	0.01		0.01		0.01		0.01		0.01		0.01		0.01		0.01		0.01		0.01		0.01		0.01	
D	PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL	

APICAL ONE-HALF

$\overline{BC}$	4.7		4.7		4.7		4.7		5.5		5.5		5.2		5.2		5.8		5.8		4.7		4.7	
$\overline{GC}$	10.9		14.9		14.0		11.6		32.5		30.8		9.2		9.6		10.3		12.1		10.1		11.2	
T	5.64		5.26		7.05		4.13		2.85		3.48		2.59		3.01		2.31		3.36		2.61		3.93	
P	0.01		0.01		0.01		0.01		0.02		0.01		0.02		0.01		0.05		0.01		0.02		0.01	
D	PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL	
$\overline{AP}$	.50		.35		.50		.45		.50		---		---		.25		.35		.50		.50		.45	

TABLE V

STATISTICAL DATA FOR PARACHLOROPHENOL
UTILIZING THE COTTON PELLET IN A DRY APPLICATION STATE

CORONAL ONE-HALF

	1		2		3		4		5		6		7		8		9		10		11		12	
	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS
$\overline{BC}$	5.5		5.5		5.5		5.5		4.9		4.9		6.0		6.0		4.8		4.8		4.9		4.9	
$\overline{GC}$	13.7		9.4		12.6		15.1		11.2		12.9		13.2		13.9		15.3		11.3		9.6		10.7	
T	3.09		2.10		4.38		4.59		2.89		2.95		2.55		2.52		3.99		2.71		2.30		3.16	
P	0.01		0.05		0.01		0.01		0.01		0.01		0.02		0.02		0.01		0.02		0.05		0.01	
D	PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL	

APICAL ONE-HALF

$\overline{BC}$	5.5		5.5		5.5		5.5		4.9		4.9		6.0		6.0		4.8		4.8		4.9		4.9	
$\overline{GC}$	12.6		10.1		12.6		11.6		9.7		11.6		12.4		11.7		10.4		8.8		10.0		8.6	
T	3.51		3.65		3.51		2.36		2.57		2.49		3.83		3.74		2.56		3.85		2.25		2.47	
P	0.01		0.01		0.01		0.05		0.02		0.05		0.01		0.05		0.05		0.01		0.05		0.05	
D	PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL	
$\overline{AP}$	.30		.40		.25		.30		.45		.50		.35		.30		.50		.50		.50		.40	

TABLE VI

STATISTICAL DATA FOR PARACHLOROPHENOL
UTILIZING THE COTTON PELLET IN A WET APPLICATION STATE

CORONAL ONE-HALF

	1		2		3		4		5		6	
	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS
$\overline{BC}$	5.2		5.2		----		5.2		5.3		5.3	
$\overline{GC}$	17.4		15.8		----		15.1		19.6		16.9	
T	10.0		6.27		----		4.13		5.39		5.85	
P	0.01		0.01		----		0.01		0.01		0.01	
D	PDL		PDL		----		PDL		PDL		PDL	

APICAL ONE-HALF

$\overline{BC}$	5.2		5.2		----		5.2		5.3		5.3	
$\overline{GC}$	10.7		11.1		----		8.90		13.0		10.1	
T	2.51		4.01		----		3.93		2.75		2.20	
P	0.02		0.01		----		0.01		0.02		0.05	
D	PDL		PDL		----		PDL		PDL		PDL	
$\overline{AP}$	.50		.50		----		.50		.50		.50	

TABLE VII

STATISTICAL DATA FOR PARACHLOROPHENOL
UTILIZING THE PAPER POINT IN A DRY APPLICATION STATE

CORONAL ONE-HALF

	1		2		3		4		5		6	
	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS
$\overline{BC}$	5.5		5.5		5.5		----		6.3		6.3	
$\overline{GC}$	12.0		14.1		15.5		----		17.3		16.4	
T	2.94		3.16		3.75		----		3.43		3.01	
P	0.01		0.01		0.01		----		0.01		0.01	
D	PDL		PDL		PDL		----		PDL		PDL	

APICAL ONE-HALF

$\overline{BC}$	5.5		5.5		5.5		----		6.3		6.3	
$\overline{GC}$	13.3		14.1		13.6		----		14.1		17.4	
T	2.48		4.23		4.30		----		2.98		3.11	
P	0.02		0.01		0.01		----		0.01		0.01	
D	PDL		PDL		PDL		----		PDL		PDL	
$\overline{AP}$	.50		.30		----		----		.50		.50	

TABLE VIII

STATISTICAL DATA FOR PARACHLOROPHENOL
UTILIZING THE PAPER POINT IN A WET APPLICATION STATE

CORONAL ONE-HALF

	1		2		3		4		5		6		7		8		9		10		11		12	
	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS	S	NS
$\overline{BC}$	6.9		6.9		6.9		6.9		5.3		5.3		5.2		5.2		5.8		5.8		4.7		4.7	
$\overline{GC}$	17.4		14.5		14.5		16.3		18.2		17.1		11.9		14.4		14.9		10.8		11.4		11.2	
T	4.01		2.72		2.11		3.67		7.37		6.1		3.41		4.27		2.87		2.20		3.17		3.71	
P	0.01		0.01		0.05		0.01		0.01		0.01		0.01		0.01		0.02		0.05		0.01		0.01	
D	PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL	

APICAL ONE-HALF

$\overline{BC}$	6.9		6.9		6.9		6.9		5.3		5.3		5.2		5.2		5.8		5.8		4.7		4.7	
$\overline{GC}$	15.7		14.6		17.3		17.4		14.7		15.8		9.7		12.0		11.0		11.3		9.2		10.1	
T	2.40		4.07		2.78		6.60		2.67		6.36		2.50		2.78		2.18		2.68		3.43		2.14	
P	0.05		0.01		0.02		0.01		0.02		0.01		0.05		0.02		0.05		0.02		0.01		0.05	
D	PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL		PDL	
$\overline{AP}$	.30		.35		----		----		.30		.35		.20		.25		.50		.50		.40		.20	

DATA SHEET NO. _____

DOG NUMBER _____

DOG WEIGHT _____

START DATE _____

INNOCULATION TIME _____

SACRIFICE TIME _____

AGENT USED _____

DELIVERY MODE: COTTON PELLET WET _____ DRY _____

PAPER POINT WET _____ DRY _____

PROJECTED SECTION DATE _____

1. RIGHT MANDIBLE

		Instrument Number	Working Length	Final Length
2nd Bicuspid	Mesial	_____F	_____mm	_____mm
	Distal	_____F	_____mm	_____mm
3rd Bicuspid	Mesial	_____F	_____mm	_____mm
	Distal	_____F	_____mm	_____mm

2. LEFT MANDIBLE

2nd Bicuspid	Mesial	_____F	_____mm	_____mm
	Distal	_____F	_____mm	_____mm
3rd Bicuspid	Mesial	_____F	_____mm	_____mm
	Distal	_____F	_____mm	_____mm
4th Bicuspid	Mesial	_____F	_____mm	_____mm
	Distal	_____F	_____mm	_____mm

*Denotes Hemorrhage During Instrumentation

APPROVAL SHEET

The thesis submitted by Dr. Gary N. Taylor has been read and approved by three members of the Graduate School faculty.

The final copies have been examined by the director of the thesis and the signature which appears below verifies the fact that any necessary changes have been incorporated, and that the thesis is now given final approval with references to content, form and mechanical accuracy.

The thesis is therefore accepted in partial fulfillment of the requirements for the degree of Master of Science.

May 21, 1973
Date

John V. Madonia
Signature of Advisor