

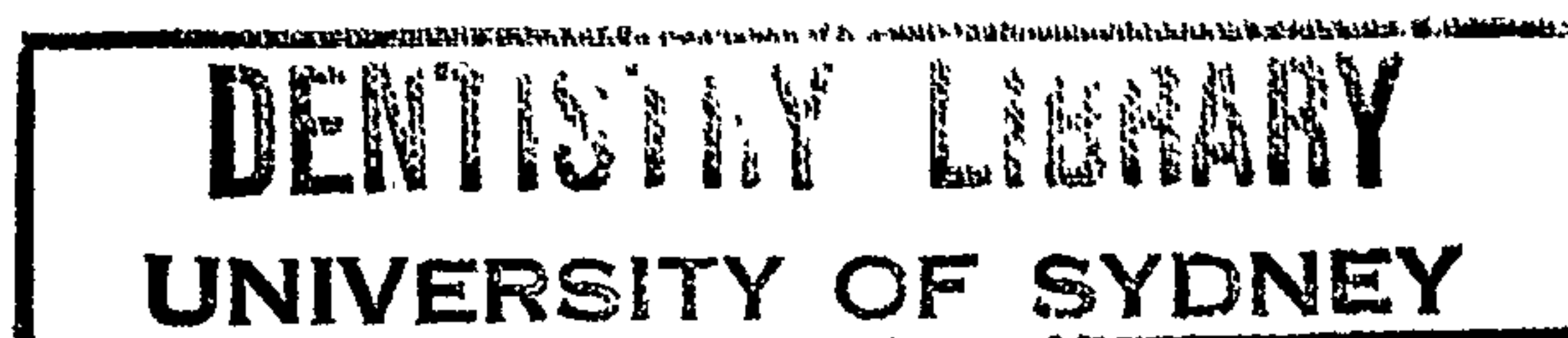
ENDODONTIC IRRIGANTS

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TABLE OF CONTENTS

<u>CHAPTER</u>		<u>Page</u>
<u>1</u>	<u>INTRODUCTION</u>	<u>1</u>
<u>2</u>	<u>HISTORY OF ENDODONTIC IRRIGATION - TO 1946</u>	<u>4</u>
2.1	INTRODUCTION	4
2.2	NINETEENTH CENTURY	5
2.3	TWENTIETH CENTURY	8
<u>3</u>	<u>IRRIGANTS AND MICROORGANISMS</u>	<u>14</u>
3.1	INTRODUCTION	14
3.2	ENDODONTIC MICROBIOLOGY	16
	3.2.1 Culturing and recognition	16
	3.2.2 Root canal flora	21
3.3	EFFECTS OF IRRIGANTS ON MICROORGANISMS	29
<u>4</u>	<u>IRRIGANTS, DENTINE AND THE SMEAR LAYER</u>	<u>40</u>
4.1	INTRODUCTION	40
4.2	DENTINE	40
	4.2.1 Structure of dentine	40
	4.2.2 Effects of irrigants on dentinal structure	43
4.3	SMEAR LAYER	44
	4.3.1 Coronal smear layer	44
	4.3.2 Effects of irrigants on coronal smear layer	48
	4.3.3 Radicular smear layer	48
	4.3.4 Effects of irrigants on radicular smear layer	53
<u>5</u>	<u>SODIUM HYPOCHLORITE</u>	<u>57</u>
5.1	INTRODUCTION	57
5.2	CHEMISTRY, MANUFACTURE AND STABILITY	58
	5.2.1 Influence of surface tension	63
5.3	SOLVENT ACTION	65
	5.3.1 Influence of temperature	68
	5.3.2 Influence of concentration	69
	5.3.3 Influence of volume	71
5.4	QUALITATIVE STUDIES	72
	5.4.1 Light microscopy	72
	5.4.2 Scanning electron microscopy	74

<u>6</u>	<u>CHELATING IRRIGANTS</u>	77
6.1	INTRODUCTION	77
6.2	EDTA	78
	6.2.1 Chemistry, concentration and pH	79
	6.2.2 Role in chemomechanical preparation	80
	6.2.3 Limitation of action	82
	6.2.4 Effect on dentine	84
	6.2.5 Effect on apical seal	86
	6.2.6 EDTA as a final flush	87
	6.2.7 RC-Prep	89
6.3	CITRIC ACID	93
<u>7</u>	<u>OTHER IRRIGANTS</u>	95
7.1	INTRODUCTION	95
7.2	HOT DISINFECTANTS	95
7.3	ENZYMES	96
7.4	UREA	97
7.5	UREA PEROXIDE	97
7.6	LOCAL ANAESTHETIC	99
7.7	CHLORAMINE-T AND AZOCHLORAMID	100
7.8	HYDROGEN PEROXIDE	102
7.9	HYDROGEN PEROXIDE/SODIUM HYPOCHLORITE	104
7.10	AMINOACRIDINE	106
7.11	QUATERNARY AMMONIUM COMPOUNDS	109
7.12	WESCODYNE AND IODOPAX/JODOPAX	115
7.13	GLUTARALDEHYDE AND POTENTIATED ACID 1,5 PENTANEDIAL	116
7.14	CHLORHEXIDINE	120
7.15	SALINE	124
7.16	WATER	127
7.17	POTASSIUM HYPOCHLORITE/HYDROGEN PEROXIDE	129
7.18	DECAL	129
7.19	POLYACRYLIC ACID	129
7.20	NELEX	130
7.21	SODIUM LAURYL SULFATE	131
7.22	DODECYLDIAMINOETHYL GLYCINE	132
7.23	ALCOHOL	132
7.24	TANNIC ACID	133

<u>8</u>	<u>IRRIGANTS AND ULTRASOUND</u>	134
8.1	INTRODUCTION	134
8.2	MECHANISMS OF ACTION	136
8.3	PRELIMINARY INVESTIGATIONS	138
8.4	SUBSEQUENT INVESTIGATIONS	141
	8.4.1 Debridement	141
	8.4.2 Curved canals	143
	8.4.3 Other studies	144
8.5	USE OF IRRIGANTS WITH ULTRASOUND	145
	8.5.1 Volume	145
	8.5.2 Time and flow	146
	8.5.3 Type of irrigant	148
<u>9</u>	<u>CYTOTOXICITY OF IRRIGANTS</u>	151
9.1	INTRODUCTION	151
9.2	ASSESSMENT OF CYTOTOXICITY	152
9.3	EARLY STUDIES	156
9.4	<i>IN VITRO</i> STUDIES	159
9.5	<i>IN VIVO</i> STUDIES	162
9.6	CLINICAL STUDIES	166
9.7	OTHER STUDIES	167
9.8	ACCIDENTS	168
<u>10</u>	<u>PRACTICAL ASPECTS</u>	171
10.1	INTRODUCTION	171
10.2	ACCESS AND CANAL SIZE	173
10.3	DELIVERY VOLUME	177
10.4	DELIVERY EQUIPMENT	180
10.5	IRRIGATION TECHNIQUES	189
10.6	IRRIGANT REMOVAL	192
10.7	CORROSIVE EFFECTS	193
<u>11</u>	<u>SUMMARY AND CONCLUSIONS</u>	198
	<u>BIBLIOGRAPHY</u>	201

LIST OF TABLES

	Page
Table 1. Recognized microbial genera and species in infected root canals.	22
Table 2. A current classification of <i>Bacteroides</i> <i>melaninogenicus</i> species.	28

CHAPTER 1

INTRODUCTION

Biomechanical preparation of the root canal combines two procedures : mechanical preparation by instruments, and the removal of debris from the pulp chamber by the process of irrigation (Ingle et al., 1985 p178).

Several authors have listed features considered essential for the "ideal" irrigant. Walton and Torabinejad (1989 pp205-6) suggested that the "ideal" irrigant should possess the following properties:-

1. *Tissue/debris solvent* - to dissolve or disrupt soft tissue and/or hard tissue remnants and permit their removal where inaccessible to instruments.
2. *Low toxicity* - the irrigant should not irritate the periapical tissues.
3. *Low surface tension* - to promote flow into inaccessible areas.
4. *Lubricant* - to aid in instrumentation and reduce the possibility of instrument fracture.
5. *Sterilization (or at least disinfection)*
6. *Removal of smear layer*
7. *Other factors* - including availability, moderate cost, adequate shelf life, and ease of storage.

Blackler (1981 p64) suggested properties of the ideal irrigant should also include:-

8. *Non-staining* to tooth structure and clothes.
9. *Should not leave a residue* within the prepared canal which could interfere with the canal wall/cement sealer interface.
10. *Finite span of chemical activity*.
11. *Non-corrosive* to endodontic instruments.

Salzgeber and Brilliant (1977) further included:-

12. The ability to act as a *bleaching agent*.

This treatise will examine these properties and review the contribution of irrigants to endodontic therapy. Because many current concepts about irrigants evolved in early times, a historical perspective of irrigants and the reasons for their introduction to endodontic treatment will be provided in Chapter 2.

The important roles of irrigants as disinfectants will be discussed in Chapter 3, and as tissue-debriding agents in Chapters 4 to 7. The utilisation of irrigants in lavage and lubrication to facilitate instrumentation will be examined in Chapter 10.

The recent introduction of purported work-saving devices, such as ultrasonic instrumentation units, have changed, and may further change, the approach to endodontics. Irrigants with ultrasound will be discussed in Chapter 8.

Issues such as safety, efficacy and toxicity are of utmost concern, and irrigants must be assessed from these perspectives. In Chapter 9, the cytotoxic potential of irrigants will be investigated.

It will become apparent during this treatise that irrigants and irrigation have a valid role in endodontics. Although few issues are fully resolved, there are convincing arguments that therapeutic benefits can be achieved with careful selection of irrigants.

CHAPTER 2

HISTORY OF ENDODONTIC IRRIGATION - TO 1946

2.1 INTRODUCTION

Inscriptions on excavated Ting dynasty ruins from 1400-1100 BC show the use of the Chinese character which means "caries". The design of this character reflects the concept of the era that caries was caused by the invasion of "worms" into teeth (Tsao, 1984). Similarly, around the time of the French revolution, extremely small "worms" with "a round head marked by a little black spot" were also considered responsible for dental disease (Fauchard, 1746 p130).

Whilst others were not convinced of the presence of "worms" (Fauchard, 1746 pp131-3), it was a well established view at that time that "acrid humour" was a major contributing factor to "fistulous abscesses". Thus, in an effort to remove this irritant, treatment generally involved trephination or drainage via the mucosa or root canal as well as application to the mucosa of various water-based preparations (Fauchard, 1746 p80). Leeches, poultices and cautery were employed, followed by gold foil root fillings extending to the apex. However, the concept of sterility was not as yet known, and it would appear that irrigation of root canals did not form part of the practise of dentistry at this time.

2.2 NINETEENTH CENTURY

In 1840, leading dental practitioners organized the American Society of Dental Surgeons and, in the same year, the General Assembly of Maryland chartered the Baltimore College of Dental Surgery as the first independent educational institution devoted exclusively to dentistry. Other dental colleges opened, but it was not until 1867 that the Harvard Corporation voted the establishment of the first dental school to be associated with a university and its medical school (Hapgood, 1930 pp4-7).

Concurrently in Britain, the London Institute for diseases of the Teeth opened in 1839, the first dental school was then founded in 1856, and in 1859 a Royal Charter was granted to the Royal College of Surgeons, allowing examinations in dental surgery (Harty, 1984). At this time, new developments such as general anaesthesia, rubber dam, gutta percha, barbed broaches and intracanal antiseptics were incorporated into dentistry.

In 1867, Joseph Lister had postulated that "germs" caused suppuration and putrefaction of wounds, and introduced antiseptic surgery by using a weak phenol solution in wounds (Grossman, 1976). Later research by Robert Koch, in 1876, showed that bacilli caused anthrax, and this was soon followed by development of Koch's postulates that were to change the approach to medicine and dentistry (Grossman, 1976). Further, in 1881, controlled laboratory studies by Koch showed that pure

cultures of bacteria could be destroyed by the use of hypochlorites (Dychdala, 1983 p157).

These developments had a direct effect on the practise of dentistry. It was widely believed that by wiping the root canal with disinfectants, and placing intracanal medicinal dressings, sterilization could be achieved. Thus medicaments and broaches became the stalwarts of root canal therapy, and the appearance of pericementitis after opening into a pulp was attributed to carelessness in attention to sterility by the operator (Miller, 1890 pp96-9). All authors of this time stressed the need for the use of rubber dam, an invention attributed to Sanford Barnum of New York in 1864 (Grossman, 1976).

The need for frequent syringing of the canal appears to have been first advocated by Taft (1859 p224). This recommendation was based upon the need to remove any irritants that may have been induced by the products of decomposition. As well, the use of irrigation was urged when arsenic trioxide, introduced by Shearjashub Spooner in 1836 (Grossman, 1987), was employed to destroy the pulp. Daily dressings with thorough canal irrigation (presumably with water) each visit were urged, and the use of "a deodorising agent, such as chlorid of sodium" was also recommended (Taft, 1859 p244).

One author advocated pulp extirpation under a pool of oil of eucalyptus, creosote or cloves, the rationale being that these oils would penetrate the canal as the

nerve was removed (Tomes, 1887 p439). Another suggested use for irrigation was to remove a firmly attached intra-canal cotton dressing (Ottolengui, 1892 pp193-4). However, barbs and broaches were assumed to be the chief "cleansers" of the canal, and when combined with hot air from a syringe it was considered possible to achieve complete canal debridement (Taft, 1883 p316).

The early literature describes various means of obtaining a clean canal using a variety of flushing agents and medicaments. One spectacular method involved the introduction of potassium and sodium metals into the canal with the resulting "explosion" effectively removing necrotic pulps (Schreier, 1893). As recently as 1948, this technique still had support from at least one author, who claimed it successful "in opening any multiple accessory apical foramenae present" (Waas, 1948).

To widen root canals, Callahan (1894) suggested a 20%-50% aqueous solution of sulphuric acid applied on a pledget of cotton and sealed in the tooth for 24-48 hours. A saturated solution of bicarbonate of soda was then introduced into the canal orifice producing an explosive effervescent reaction, thus purportedly neutralising the acid and forcing debris to the surface.

Another author recommended use of aqua regia (nitro-hydrochloric acid) which was observed to produce

an effervescence similar to hydrogen peroxide (Hays, 1900). This author rejected the use of sulphuric acid on the grounds that it corroded "every instrument with which it came in contact".

Treatment of abscesses via their fistulous openings was routinely recommended, one method involving injection of a solution of pure creosote and iodine into and through the fistula (Steele, 1892 p254). Following this, barbs and broaches ("Donaldson cleansers") carrying hydrogen peroxide were introduced into the canal (Steele, 1892 p40).

By the late nineteenth century, the introduction of local anaesthesia, as well as the discovery of radiographs, first used for dental purposes by Otto Walkhoff in 1895, further advanced the practice of endodontics (Grossman, 1976).

2.3 TWENTIETH CENTURY

Of great consequence to dentistry over the first part of the twentieth century was the theory of focal infection, introduced by Dr. William Hunter in 1910 and embraced by many dentists (Grossman, 1976). This theory initiated a period lasting some thirty years during which otherwise healthy teeth were extracted to supposedly cure systemic diseases. In fact, even by the middle 1940's this theory was still creating controversy (Lyons, 1946).

At the turn of the century, a textbook for dental students described a technique for draining fistulae in

which sodium peroxide was pumped (using a broach) through the canal until it appeared at the fistulous opening (Gorgas, 1901 pp72-5). Teeth affected by non-draining or "blind" abscesses were to be syringed with tepid water, dried, then injected with hydrogen peroxide until all bubbling ceased. Sulphuric acid or trichloroacetic acid, introduced into the canal by broaches and cotton, was then used to remove remaining filaments in the canal.

Following pulp removal, canals were "washed out" with 3% hydrogen peroxide, after which alcohol was pumped into the canal and allowed to dry. As asepsis was now considered of paramount importance, pumping a solution of sodium peroxide into the canals and leaving for a few minutes was recommended to "sterilize" the canal (Gorgas, 1901 pp72-5).

It was claimed that hydrogen peroxide had the added advantage of inducing haemostasis in the canal (Kirk, 1900 p460). As well, when accompanied by syringing with a saturated solution of sodium bicarbonate, the carbon dioxide produced was thought to drive any remaining canal debris into the pulp chamber.

"Septic" pulps could also be washed out with hot distilled water. Extremely tender teeth, too sensitive to manipulate, were treated by syringing with meditrina¹ and stirring with broaches. Routine and repeated syringing

1. Meditrina was described as "sea-water charged with electricity" and was said to possess antiseptic and deodorant properties (Gorgas, 1905 p457).

with 3% hydrogen peroxide was also recommended for "septic" pulps by Kirk (1900 p500).

Burchard (1904 p432) suggested irrigation of the coronal access cavity with hydrogen peroxide prior to pulp removal. Following instrumentation of the canal with moistened broaches carrying dry sodium hydrate¹ and hydrogen peroxide, a stream of water and a Donaldson cleanser were then used to free the canal of debris. The author stressed the need for the irrigant to flow through the apical foramen in order to destroy any debris forced into the periapical tissues during preparation. Previously, another author had noted that sodium hydrate (which is closely analogous with potassium hydrate), was highly soluble in water and very caustic (Wilson, 1886 p358).

With the need for a diligent aseptic approach in mind, Black (1908 pp363-81), a leading authority of the time, advocated flooding the root canal with medicaments such as oil of cloves or other "sedative" oils after completion of canal preparation. However, the use of "irritating germicides" to disinfect was also emphatically discouraged (Black, 1908 p381).

Prior to filling, the canal could be flooded with eucalyptol or oil of cajuput to remove any moisture from dentine by means of displacement, this means of drying

1. Sodium hydrate (NaOH) is also known as sodium hydroxide or "caustic soda" (Uvarov, Chapman and Isaacs, 1964 p277).

being seen as a superior method to hot air or hot instruments. As well, since the oils dissolved gutta percha slightly, it was thought any remaining oil served to "stick" the material to the walls of the canal (Black, 1908 p384).

Colyer (1910 pp535-46) suggested several applications for irrigants and irrigation. Perforations could be treated by syringing the canal with hydrogen peroxide to arrest haemorrhage. Frequent irrigation with warm water removed debris when performing immediate root fillings. Colyer also considered that the alternate use of sodium peroxide and hydrogen peroxide "sterilized" the canal, the rationale being that sodium peroxide in putrescent canals formed soap and thus destroyed the "fatty" contents of the canal. The rapid evolution of hydrogen peroxide would then eject the contents into the pulp chamber. In cases complicated by periodontitis, daily irrigation with antiseptics was advised.

The importance of removal of blood from the canal to avoid aesthetic problems was recognised, and irrigation with alcohol, or even better, with "Nature's greatest solvent, water" was advised (Buckley, 1911 p275). Freshly distilled, boiled or peppermint water (to which phenol had been added) was recommended. Sodium chloride could be included if desired.

The same solutions, or "any bland solution" (Buckley, 1911 p315; Thoma, 1916 p153) were recommended for an abscess with sinus, and vigorous irrigation via

the canal until forced through the sinus was advocated. However, the "common practice" of using a solution of hydrogen peroxide was denounced (Buckley, 1911 p315). A non-canal approach which involved incision of tissue and thorough irrigation of the wound with salt solution was suggested (Black, 1915 p354). The canal was then to be dressed with a medicament.

One enterprising dentist experimented with a variety of "irrigants" such as papain, an enzyme present in the leaves, fruit, milk and seeds of the pawpaw (Wilkinson, 1929). However, the utilisation of these solutions was only recommended as a substitute for unavoidable inadequate instrumentation.

In the first experimental work on irrigant solutions, Grossman and Meiman (1941) investigated the solvent action of several agents including enzymol (which contains gastric juice and proteolytic enzymes), galactonic lactone (a solvent of mucin), double strength chlorinated soda solution¹, potassium hydroxide (20%), sodium hydroxide and others. In this study, double strength chlorinated soda solution was found to be by

1. Grossman's (1943) formula for double strength chlorinated soda solution was:-

Monohydrated sodium carbonate	276grains	(18gm)
Chlorinated lime	398grains	(26gm)
Water	q.s. ad. 4 ounces.	

However, there appears to be some controversy in the literature surrounding the understanding of the term "double strength". While Svec and Harrison (1977) have described the term to have a requirement of greater than 5% available chlorine, Gordon, Damato and Christner (1981) specify 3% available chlorine.

far the most effective pulp tissue solvent. The solvent property of sodium hypochlorite (NaOCl) had been discussed much earlier by Dakin (1915) in an article on the treatment of infected wounds.

The paper by Grossman and Meiman in 1941 led to a landmark article (Grossman, 1943), which recommended the combined use of double strength NaOCl with hydrogen peroxide to wash out fragments of pulp tissue and dentinal shavings after mechanical instrumentation. The effervescence produced by this combination was claimed to "follow the line of least resistance namely, toward the mouth of the canal and into the pulp chamber". Because of this, dentists were advised to always irrigate finally with NaOCl to avoid buildup of pressure promoted by any remaining hydrogen peroxide. Grossman (1943a) additionally recognised that inorganic acids presented disadvantages such as periapical tissue irritation.

At about this time, the field of endodontics underwent an awakening, with the founding of the American Endodontic Society in February 1944, which established endodontics as a special field within dentistry. Two years later, in 1946, the Society started publishing its own journal, thus providing a vehicle for the development and spread of new ideas, along with a more standardised treatment approach. It is the research and ideas on endodontic irrigants originating since these times that this treatise will now discuss.

CHAPTER 3

IRRIGANTS AND MICROORGANISMS

3.1 INTRODUCTION

Although the presence of necrotic tissue (Shovelton and Sidaway, 1960) and degraded serum (Grossman, 1959; Ingle et al., 1985 p224) have been considered to be responsible for the initiation and maintenance of infection, there is now strong evidence that bacteria and their products are involved in the induction of pulpal and periapical pathology (Takehashi, Stanley and Fitzgerald, 1965; Möller et al., 1981; Fabricius et al., 1982, 1982a; Ringel et al., 1982; Pascon, Gremsky and Langeland, 1989).

Takehashi, Stanley and Fitzgerald (1965) used conventional and germ-free rats to examine the pathologic changes which resulted following experimentally induced and untreated pulpal exposures. While pulpal necrosis and periapical bone destruction developed in conventional rats, pulps of germ-free rats remained vital with dentinal bridging occurring after two weeks, even in the presence of gross food impaction.

Further confirmation of the important role of bacteria in endodontic infection was supplied by a study in which aseptically necrotised pulps of monkey teeth were exposed to the indigenous oral flora for seven

days, and then sealed (Möller et al., 1981). After six months all 52 exposed teeth had become infected by facultative anaerobic *streptococci*, coliform rods and obligate non-sporulating anaerobes. As well, the periapical region around all infected teeth showed strong inflammatory reactions. From these results the authors reasoned that infection was a prerequisite for the development of apical periodontitis, and that several indigenous oral bacteria have the ability to survive in the root canal and induce apical periodontitis.

Other investigations of monkey teeth over periods of three months, six months and three years showed that the relative proportion of obligate anaerobes in the sealed root canal increased over time, indicating that selective mechanisms exist which allow certain bacteria to survive and proliferate (Fabricius et al., 1982, 1982a).

Further, Ringel et al. (1982) used aerobic and anaerobic techniques to monitor asymptomatic teeth with necrotic pulps *in vivo*. "All 60 teeth yielded positive aerobic and anaerobic cultures after initial samples of the root canal contents were incubated. The significance of this finding is that bacteria may be present initially up to 100% of the time in teeth with necrotic pulps." In addition, a positive relationship has been found between the size of a periapical lesion and the number of species and microorganisms in the root canal (Byström et al., 1987).

Recently, a study was conducted in which experimentally induced pulp tissue lesions in baboon and monkey teeth were inoculated with dental plaque (Pascon, Gremsky and Langeland, 1989). Over a number of time periods up to 20 weeks, the animals were killed and the jaws and teeth sections examined. Bacteria were found at all levels of the root canal, in dentinal tubules and in periapical inflammatory tissue.

It appears, therefore, that there is an extremely strong link between the presence of microorganisms within the tooth and the development and maintenance of pulpal or periapical pathology. It is therefore reasonable to assume that the elimination of viable microorganisms, or at least a substantial reduction in their capacity to reproduce, is a major determinant of success in endodontic therapy.

3.2 ENDODONTIC MICROBIOLOGY

In this section, the current status of endodontic microbiology will be examined. A discussion on culturing and recognition has been included, to establish that the methods which have been used to determine the antibacterial effectiveness of irrigants clinically, provide limited information.

3.2.1 Culturing and recognition

Although advocated by some authors (Oliet and Sorin, 1969; Naidorf, 1985), the usefulness of routine

root canal culturing has been questioned by others, as the presence of a negative culture is not a determinant of canal sterility (Seltzer, Bender and Turkenkopf, 1963; Akpata, 1976; Eggink, 1982). Other questions, such as the accuracy of the culture in relationship to use of antibiotic medicaments, have also been raised (Bender and Seltzer, 1954). Spångberg (1973a) further commented that death of bacteria "designates only an irreversible loss of ability to proliferate" in a particular medium, and that "bacteria described as dead in one medium may therefore very well prove viable on transfer to a different medium".

Dupont, Brady and del Rio (1977) examined paper point samples from infected root canals under SEM, and correlated their data with results from other testing methods i.e. presence of exudate and status of bacteriological culturing. Under SEM, the presence of predominately red blood cells, a few white blood cells, as well as dentinal fragments and microorganisms was observed. Further, while recognition of particular bacteria by means of SEM "was not fruitful", SEM examination of paper point samples was considered to provide the most sensitive assessment of canal status.

Minor differences in culturing and examination procedures can greatly influence results (Fulghum, Wiggins and Mullaney, 1973; Zielke, Heggers and Harrison, 1976; Dupont, Brady and del Rio, 1977). These differences include degree of anaerobiosis within the media, size and

age of the inoculum, and whether pre-formed enzymes or growth of microorganisms are used for recognition.

Early studies utilised a range of sampling and incubation techniques, with results indicating that aerobic and facultatively anaerobic microorganisms predominated in the infected root canal (Lucas, 1920; Bibby, 1938; Cran, 1956; Blechman, 1957; Winkler and Van Amerongen, 1959; Sciaky and Sultzen, 1961; Seltzer, Bender and Turkenkopf, 1963). The bacterial growth media (broth) used in the studies were, in retrospect, not suitable for culturing these particular samples, as fast growing bacteria may have distorted results. Also, the presence of hydrogen peroxide and superoxide radicals in autoclaved anaerobic broth media may have actually killed anaerobes (Carlsson, Nyberg and Wrethén, 1978). In addition, gram staining techniques, while providing confirmation of the presence of bacteria, did not allow identification of species (Mjör, 1977). Under such circumstances, these studies found *streptococci* to be the most commonly isolated microorganism. An early study of 4186 cultures from over 1000 canals found up to 63% of all microorganisms to be facultatively anaerobic *streptococci* (Winkler and Van Amerongen, 1959).

More significantly though, anaerobic techniques were unavailable to early researchers, and although anaerobes can tolerate exposure to air when in media containing haemolysed blood or pus (Bartlett et al., 1976; Carlsson, Frölander and Sundqvist, 1977), since the

development of anaerobic collection and culturing techniques, it has been possible to further explore the role of anaerobes in periapical pathosis (Möller, 1966; Fulghum, 1971; Sundqvist, 1976).

Möller (1966) first described an aseptic and anaerobic technique to obtain samples from root canals. Ten years later, Sundqvist presented a landmark publication describing his studies of the microbiological flora of necrotic pulps in intact teeth (Sundqvist, 1976). Within this dissertation he described a meticulous approach to obtaining and culturing samples using an anaerobic glove box.

Concurrently, an anaerobic technique was also formulated at the Anaerobe Laboratory at the Virginia Polytechnic Institute and State University. In a study using this technique, anaerobic gram-negative rods were found in 75% of 40 intact teeth with necrotic pulps (Wittgow and Sabiston, 1975). Kantz and Henry (1974) utilised an anaerobic glove box which contained 85% nitrogen, 10% carbon dioxide and 5% hydrogen.

Anaerobic sampling techniques are now routinely used in the identification of the root canal flora (Byström and Sundqvist, 1981, 1983, 1985; Zavistocki et al., 1980; Yoshida et al., 1987).

However, anaerobic culturing has a number of practical problems, such as the need for 7-14 days to grow identifiable cultures of some microbial types, such as *Bacteroides*. It is time-consuming, needs sophisticated

equipment and thus is expensive. As well, certain organisms such as the black-pigmented *Bacteroides* species may be difficult to detect in the preliminary stages of pulpal pathosis when numbers may be low (Pantera et al., 1988).

Other alternatives to anaerobic culturing have been developed. Use of indirect immunofluorescence microscopy might offer a viable alternative to anaerobic culturing by providing advantages in time, expense and less need for technical expertise. As well, when Pantera et al. (1988) compared this technique to anaerobic culturing of *Bacteroides* species in root canals, it was found to be up to three times more sensitive in the detection of certain *Bacteroides* species.

Immunocytochemical methods of identification use antisera to identify bacteria in tissue specimens (Sundqvist and Reuterving, 1980; Happonen et al., 1985; Happonen, 1986), however availability of antisera to the entire range of species currently limits this technique.

Recently a further means of identification for *Bacteroides* and *Treponema* has been described in which these bacteria are identified using a rapid enzyme assay, which measures the trypsin-like activity of the microorganism on the colourless substrate N-benzoyl-DL-arginine-2-naphthylamide (BANA). Hydrolysis of BANA produces an orange-red colour (Breeland, Bergenholtz and Madison, 1989).

3.2.2 Root canal flora

There is convincing evidence that root canal infections are generally polymicrobial infections where anaerobes dominate (Bergenholtz, 1974; Kantz and Henry, 1974; Sabiston and Gold, 1974; Wittgow and Sabiston, 1975; Sabiston, Grigsby and Segerström, 1976; Sharkey, 1978; Dahlén and Bergenholtz, 1980; Zavistocki et al., 1980; Aderholt, Knothe and Frenkel, 1981; Brook, Grimm and Kielich, 1981; Fabricius et al., 1982a; Oguntebi et al., 1982; Williams, McCann and Schoenknecht, 1983; Sundqvist and Byström, 1983;1985; Van Winkelhoff, Carlee and de Graaff, 1985; Lewis, MacFarlane and McGowan, 1986; Nair, 1987; Sundqvist, Johansson and Sjögren, 1989; Yamamoto et al., 1989).

Prominent anaerobic species isolated from pulpal and periapical infections include *Bacteroides*, *Fusobacteria*, *Eubacteria* and *Peptostreptococci*. A summary of the species isolated from infected root canals is presented in Table 1. (p22).

Dark-field observation of the distribution of bacteria in extracted teeth with pulpal necrosis has revealed a significantly greater percentage of coccoids and rods in the coronal part than in the apical part (Thilo, Baehni and Holz, 1986). As well, more viable bacteria have been observed in the pulp canal, predentine and adjacent calcified dentine than in the deeper dentine (Bence et al., 1973). Haapasalo and Orstavik (1987) detected experimentally induced invasion of dentinal

Table 1.

Recognized microbial genera and species in infected root canals*

gram positive bacteria		gram negative bacteria	
Aerobic and facult. anaerobic	Anaerobic	Aerobic and facult. anaerobic	Anaerobic
<u>Cocci</u>			
STREPTOCOCCUS	STREPTOCOCCUS		VEILLONELLA
<i>S. milleri</i>	<i>S. constellatus</i>		<i>V. parvula</i>
<i>S. mitior</i>	<i>S. intermedius</i>		
<i>S. mutans</i>	<i>S. morbillorum</i>		
<i>S. sanguis</i>			
<i>S. faecalis</i>	PEPTOSTREPTOCOCCUS		
	<i>P. anaerobius</i>		
	<i>P. magnus</i>		
	<i>P. micros</i>		
	<i>P. prevotil</i>		
<u>Rods</u>			
ACTINOMYCES	ACTINOMYCES		BACTEROIDES
<i>A. naeslundil</i>	<i>A. israelii</i>		<i>B. buccae</i>
<i>A. viscosus</i>	<i>A. meyeri</i>		<i>B. denticola</i>
	<i>A. odontolyticus</i>		<i>B. endodontalis</i>
	ARACHNIA		<i>B. gingivalis</i>
	<i>A. propionica</i>		<i>B. intermedius</i>
	EUBACTERIUM		<i>B. loeschei</i>
	<i>E. alactolyticum</i>		<i>B. oralis</i>
	<i>E. brachy</i>		<i>B. oris</i>
	<i>E. lentum</i>		<i>B. ureolyticus</i>
	<i>E. nodatum</i>		FUSOBACTERIUM
	<i>E. timidum</i>		<i>F. nucleatum</i>
		CAPNOCYTOPHAGA	
		<i>C. ochracea</i>	
		EIKONELLAS.	SELENOMONAS
		<i>E. corrodens</i>	<i>S. sputigena</i>
	LACTOBACILLUS		WOLINELLA
	<i>L. catenaforme</i>		<i>W. recta</i>
	<i>L. minutus</i>		<i>W. curva</i>
		CAMPYLOBACTER	
		<i>C. sputorum</i>	
	PROPRIONIBACTERIUM		
	<i>P. acnes</i>		

* from unpublished data by Göran Sundqvist, 1988.

tubules by *Enterococcus faecalis* under SEM and light microscopy. Further, when cementum was absent from the dentine samples, invasion was more easily instigated.

Another investigation has demonstrated differences in species distribution in different parts of the root canal system (Fabricius et al., 1982a). In this study of infected monkey teeth, apical areas were found to be dominated by slow-growing obligate anaerobes such as *Bacteroides melaninogenicus*, rather than the more rapidly growing facultative anaerobes such as *Escherichia coli*, which could be found in the coronal part of the canal.

While over 300 species of bacteria have been identified in the oral cavity (Moore et al., 1985), the number of species isolated in the infected root canal is considerably lower. Up to twelve species at any one time, and up to 10^7 in number, have been detected (Kantz and Henry, 1974; Dahlén and Bergenholtz, 1980; Zavistocki et al., 1980). Sundqvist, Johansson and Sjögren (1989) isolated 337 strains of bacterial cells from 72 teeth, the mean number of species per infected tooth being 8.5. Other studies have reported 4.5 (Williams, McCann and Schoenknecht, 1983), 4.9 (Brook, Grimm and Kielich, 1981) and less than 4 (Lewis, McFarlane and McGowan, 1986; Oguntebi et al., 1982) mean bacterial species from periapical abscesses.

Zavistocki et al. (1980) attempted to quantify the bacteria found in endodontic infections. Using a small sample size ($n=10$), paper points were weighed before and

after specimen collection. The mean concentration of bacteria obtained from non-vital canals was found to be 60 million viable organisms per paper point gram sample. The authors postulated that this is comparable to the bacterial concentration found at other infected anatomic sites.

Observations of a significant positive correlation between the percentage of spirochaetes (*Treponema*, *Leptospira* and *Borrelia*) in the apical one third of the canal, and the size of the periapical lesion on a pre-extraction radiograph have been made (Thilo, Baehni and Holz, 1986). Spirochaetes have been found in some endodontic abscesses (van Winkelhoff, Carlee and de Graaff, 1985) and, using electron microscopy, in necrotic pulp tissue (Nair, 1987).

However, other studies have not confirmed the existence of spirochaetes (Kantz and Henry, 1974; Williams, McCann and Schoenkrecht, 1983; Oguntebi et al., 1983), and it has been suggested that the presence of spirochaetes may indicate an abscess of periodontal origin (Farber and Seltzer, 1988; Trope et al., 1988).

It is important to recognise the dynamic nature of the flora within the infected root canal. Pulps contaminated by saliva initially contain a higher proportion of facultative anaerobes than strict anaerobes. However, over time, anaerobe strains have been observed to outnumber facultative anaerobes (Möller et al., 1981; Fabricius et al., 1982, 1982a). This is

thought to be determined by the development of diminished oxygen tension (Walden and Hentges, 1975) and a low reduction-oxidation potential (Loesche et al., 1983) which would favour anaerobic growth.

Other factors which influence the bacterial ecology involve availability of nutrients including disintegrating host tissue cells and tissue fluid (Loesche, 1968; Gillespie and Holt, 1987). As well, products of other bacteria, such as Vitamin K (Gibbons and MacDonald, 1960); haemin (Grenier and Mayrand, 1986) and succinate (Mayrand and McBride, 1980), provide nutrients which may be utilised by certain microbial species such as *Bacteroides* (Gibbons and MacDonald, 1960; McKee et al., 1986).

The bacterial ecology may be further controlled by other mechanisms which may provide protection against phagocytosis (Ingham et al., 1977). As well, the ecology is influenced by other factors, such as the production by bacteria of bacteriocins which may inhibit the growth of other species (Takazoe, Nakamura and Okuda, 1984; Hillman, Socransky and Shivers, 1985), and by the production of essential growth factors (Lev, Krudell and Milford, 1971; Mayrand and McBride, 1980).

Gram-negative endotoxin-producing bacteria include the *Bacteroides*, *Veillonella* and *Fusobacterium* species (Nygren, Dahlén and Nilsson, 1979; Dahlén and Bergenholtz, 1980). *Fusobacterium* are common isolates from infected root canals (Byström and Sundqvist, 1983).

Bacteroides species are obligately anaerobic, non-motile rods possessing certain surface components which behave as virulence factors. These components, which include fimbriae and capsular material, in addition to endotoxin, may act by participating in adherence and colonisation of microorganisms and subsequent tissue destruction (Sundqvist et al., 1982; Slots and Genco, 1984; Handley and Tipler, 1986).

An association has been suggested between the existence of clinical symptoms (defined as spontaneous pain, pain on percussion and presence of exudate) and anaerobes such as *Bacteroides melaninogenicus*, *Peptostreptococcus magnus* (Yoshida et al., 1987; Sundqvist, Johansson and Sjögren, 1989) and *Fusobacterium nucleatum* (Heimdahl et al., 1985). However, clinically asymptomatic teeth have also exhibited microflora containing *Bacteroides* species (Haapasalo, 1989; Sundqvist, Johansson and Sjögren, 1989).

More specifically, Haapasalo (1989) associated the incidence of acute symptoms with the presence of *Bacteroides gingivalis*, *Bacteroides endodontalis*, *Bacteroides buccae*, and further linked the presence of *Bacteroides melaninogenicus* with symptoms persisting beyond one week after commencement of treatment.

As observation techniques improve *Bacteroides melaninogenicus* as a species is proving to be a very heterogenous group, and final classification has not as yet been resolved. A current classification of

Bacteroides melaninogenicus (also known as "black pigmented *Bacteroides*") appears in Table 2. (p28). Although the role of bile-sensitive, non-pigmented *Bacteroides* in endodontic infections has yet to be established, many species have been identified such as *Bacteroides buccae*, *Bacteroides oris*, *Bacteroides buccalis* and *Bacteroides oralis*. A "*Bacteroides*-like" organism *Mitsuokella dentalis* has been observed by Haapasalo (1989).

As part of the *asaccharolyticus* subspecies, a current classification of *Bacteroides melaninogenicus* includes *Bacteroides endodontalis*, which was described by Van Steenberg et al. in 1984. Several strains have since been isolated. Features of *Bacteroides endodontalis* include slow growth, even on enriched media (Van Steenberg et al., 1984; Van Winkelhoff, Carlee and de Graaff, 1985), and probable high sensitivity to oxygen tension (Carlsson, Frölander and Sundqvist, 1977). Although *Bacteroides endodontalis* has been recovered from the tongues and supragingival plaque of patients with gingivitis, this bacteria is thought to be almost exclusively associated with endodontic abscesses (van Winkelhoff, Carlee and de Graaff, 1985).

Gram-positive bacteria found in non-vital root canals include species of *Streptococci*, *Lactobacilli*, and *Actinomyces*. *Streptococcus faecalis* has been cited as one of the most persistent and frequently isolated facultative anaerobes found in root canal cultures (Smith

Table 2. A current classification of *Bacteroides melaninogenicus* species*

<u>Subspecies saccharolyticus</u>	<u>Subspecies asaccharolyticus</u>
Fermentative species	<i>B. asaccharolyticus</i>
<i>B. melaninogenicus</i>	<i>B. endodontalis</i>
<i>B. melaninogenicus</i>	<i>B. gingivalis</i>
<i>B. loeschei</i>	
<i>B. denticola</i>	
Weakly fermentative species	
<i>B. intermedius</i>	
<i>B. corporis</i>	
<i>B. intermedius</i>	

* adapted from Farber and Seltzer, 1988; Haapasalo, 1989

and Wayman, 1986), while *Streptococcus milleri* has been identified in abscesses which have penetrated bone (Williams, McCann and Schoenknecht, 1983; Lewis, MacFarlane and McGowan, 1986).

Several case reports have associated *Actinomyces israelii* (Sundqvist and Reuterving, 1980; Weir and Buck, 1982; Happonen, 1986; O'Grady and Reade, 1988) and *Arachnia propionica* (Sjögren et al., 1988) with persistent endodontic infections.

3.3 EFFECTS OF IRRIGANTS ON MICROORGANISMS

"The antimicrobial activity of any compound cannot be viewed in isolation, but must be described with reference to pH, temperature, organic matter, and in-use dilution. Such factors significantly influence the usefulness of the preparation and manner of use" (Scott and Gorman, 1983 p76). In addition, variations in antimicrobial activity have been observed with the same disinfectant on surfaces of different porosity (Stedman, Kravitz and Bell, 1954).

As discussed in the previous chapter, the need to reduce or eliminate bacteria from the root canal has long been recognized. As well as offering an improved endodontic prognosis, the penetration of bacteria into the blood stream should be avoided for reasons such as prevention of infective endocarditis (Pogrel and Welsby, 1975).

In early papers, penicillin (Grossman, 1946) and

later, antibiotic mixtures (Grossman, 1951), were advocated as intracanal dressings to "sterilize" canals, but fell out of favour as questions on allergy and resistance emerged (Jolly and Sullivan, 1956).

Grossman (1945) found that the use of a concentrated sulfonamide solution as an intracanal dressing was not as effective as the application of other medicaments, such as camphorated monochlorophenol or beechwood creosote. However, in 1952, he suggested the utilization of a polyantibiotic paste to act upon *Streptococci viridans*, β -haemolytic *streptococci* and *Staphylococci aureus*. This paste contained a mixture of penicillin, bacitracin, streptomycin and sodium caprylate in a silicone base (Auerbach, 1953). Cran (1957) proposed an antibiotic preparation containing polymyxin, neomycin sulphate, bacitracin, methyl para hydroxy benzoate and propyl para hydroxy benzoate for use as a combined irrigant/intracanal medicament.

Grossman also stressed the need for the canal to be properly enlarged and irrigated, and suggested that attention to this regime, followed by a suitable antibiotic combination, should render the canal sterile in one to two treatments (Grossman, 1953).

By 1972, Grossman was continuing to endorse the use of antibiotics, despite recognising that the United States Food and Drug Administration had banned the use of antibiotic combinations as medicaments in endodontics. At this time, the bacteriological culture was still the

dominant means by which canal preparation was assessed. "With few exceptions, the culture method reflects the bacteriological status of the canal wall. Although a polyantibiotic paste is the most effective agent for attaining sterility of the root canal, other factors such as biomechanical cleansing and irrigation of the canal are equally important" (Grossman, 1972).

The conclusions reached in early studies sometimes used clinical and radiographic evidence (Blechman and Cohen, 1951; Grahnén, 1963), but usually the efficacy of irrigants was determined by assessment of bacteriological cultures (Auerbach, 1953; Stewart, 1955; Ingle and Zeldow, 1958; Stewart, Cobe and Rappaport, 1961; Grahnén and Krasse, 1963).

More recent research on antibacterial endodontic irrigants has focused primarily on NaOCl (Shih, Marshall and Rosen, 1970; Spångberg, Engström and Langeland, 1973; Cvek, Nord and Hollender, 1976; Ellerbruch and Murphy, 1977; Cunningham and Joseph, 1980; Harrison and Hand, 1981; Byström and Sundqvist, 1981; 1983; 1985; Buttler and Crawford, 1982), or used NaOCl as a standard for comparison with other irrigants (Ringel et al., 1982; Foley et al., 1983; Smith and Wayman, 1986; Nikolaus, Wayman and Encinas, 1988).

NaOCl yields hypochlorous acid which, in its undissociated form, is able to exert an antibacterial action, the germicidal efficacy of which is related to the ease and extent of liberation of the hypochlorous

acid (Harvey, 1980 pp973-4). The mechanisms of bactericidal action of NaOCl will be further discussed in Chapter 5.

Shih, Marshall and Rosen (1970) investigated the bactericidal action of 5.25% NaOCl on *Streptococcus faecalis* and *Staphylococcus aureus*. In serial tube dilution studies, NaOCl exerted germicidal activity against each organism within 30 seconds, even at dilutions of 1:1,000 but not 1:10,000. However, copious irrigation with full strength NaOCl was unable to prevent proliferation of organisms previously inoculated into the canals of instrumented root canals of extracted teeth. The authors stated that "the bactericidal efficiency of NaOCl in tube dilution studies is not related to the effect of NaOCl used in extracted human teeth".

A similar tube dilution study by Spångberg, Engström and Langeland (1973) concluded that 0.5% NaOCl possessed an antimicrobial effect against "all bacteria commonly present in necrotic cases", yet only *Streptococcus faecalis*, *Staphylococcus pyogenes aureus*, *Pseudomonas aeruginosa* and a fungus, *candida*, were included in the study.

Harrison and Hand (1981) also investigated the *in vitro* bactericidal properties of 5.25% NaOCl against *Streptococcus faecalis*. Whilst the presence of human serum albumin did not interfere with the action of NaOCl, dilution with water or hydrogen peroxide reduced its

bactericidal potency.

Cvek, Nord and Hollender (1976) examined the antibacterial effect on a mixed flora following use *in vivo* of 0.9% saline, 0.5% and 5% NaOCl as irrigants in mature and immature root canals. While saline had a minimal antibacterial effect, both concentrations of NaOCl were equally effective in providing an antibacterial effect more potent than saline. However, effectiveness in immature roots was less than in mature roots.

Ellerbruch and Murphy (1977) found that vapours produced by 5.25% NaOCl were bactericidal to selected microorganisms *in vitro* after 24 hours. In contrast, vapours from 2% glutaraldehyde (Cidex, Arbrog Inc., Arlington, Texas, USA) had a limited bacteriostatic effect. The microorganisms included in the study were *Streptococcus epidermis*, *Lactobacillus*, *a-streptococcus* and *Enterococcus*.

Morris had claimed, in 1958, that heating NaOCl would enhance its antiseptic properties. An *in vitro* study by Cunningham and Joseph (1980) compared the bactericidal effect of 2.6% NaOCl at room and body temperature (22°C and 37°C respectively) against *Staphylococcus aureus*, *Streptococcus sanguis*, *Escherichia coli*, *Proteus vulgaris* and *Bacillus subtilis* spores. Results from bacterial cultures indicated to the authors that use of a higher temperature solution produced a significant reduction in time needed to kill the

organisms. However, these results were in direct contrast with those of Raphael et al. (1981) who found no difference in culture reversals after seven days following use of NaOCl at three different temperatures (21°C, 29°C or 37°C).

Another study by Ringel et al. (1982) found 2.5% NaOCl to be more effective than 0.2% chlorhexidine in producing negative anaerobic cultures. Anaerobic and aerobic incubation techniques were used to evaluate *in vivo* the status throughout treatment of 60 asymptomatic single rooted teeth with necrotic pulps. Significantly more interappointment culture reversals (from negative at the end of one appointment to positive at the beginning of the next) were obtained following the use of chlorhexidine, thus indicating that NaOCl might provide a more prolonged bacteriostatic result. However neither irrigant was able to render canals bacteria free.

In a series of articles, Byström and Sundqvist (1981; 1983; 1985) followed the bacterial status of teeth undergoing endodontic treatment over several appointments. While numbers of bacteria immediately decreased following instrumentation, it was found that bacterial numbers increased between appointments when no intracanal dressing was used. While the first study (1981) used saline during instrumentation, the second (1983) compared saline and 0.5% NaOCl. Although there was no indication that specific bacteria were resistant to treatment, the authors commented that 0.5% NaOCl "seemed

to be more effective than saline in eliminating bacteria from the root canals". Even so, by the fifth appointment, no bacteria could be recovered from 8 of the 15 saline irrigated teeth, and 12 of the 15 NaOCl irrigated teeth.

The third study (Byström and Sundqvist, 1985) investigated the antibacterial action of 0.5% and 5% NaOCl, and 15% ethylene diamine tetraacetic acid (EDTA) in combination with 5% NaOCl. Although none of the three regimes were completely successful in producing a negative culture after three appointments, it was found that the combined use of EDTA and NaOCl was the most effective. In agreement with Cvek, Nord and Hollander (1976), no difference in antibacterial effect was observed between 0.5% and 5% NaOCl.

The results of these studies, and others, indicate that an interappointment antibacterial intracanal medicament should be routinely placed (Shih, Marshall and Rosen, 1970; Ringel et al., 1982; Sjögren and Sundqvist, 1987).

Buttler and Crawford (1982) investigated the concentrations of NaOCl required to detoxify two types of endotoxin *in vitro*, *Escherichia coli* w(0127:B8) and *Salmonella typhosa* w(0901) (Difco Laboratories, USA). While results indicated that 1 ml of 0.58%, 2.70% or 5.20% NaOCl was able to detoxify 100 mg of endotoxin in 1 ml of water, the methodology appeared to have critical problems. The volumes of endotoxin (up to 100 ng/ml) used in the study were considered by the authors to be

comparable to that encountered in the root canal system but no supporting evidence was supplied. In addition, the positive control (0.25N NaOH) produced identical results to the negative control (0.9% NaCl). Another variable was the need (for technical reasons) to add hydrochloric acid to NaOCl and the positive control, thus changing both chemistry and concentration of NaOCl and saline.

Linke and Chohayeb (1983) investigated the use of selected irrigants to sterilize fine gutta percha points (Mynol, Inc., Broomall, PA, USA). Irrigants included in the study were 3% hydrogen peroxide and 4.5% NaOCl. Gutta percha points were inoculated with gram-positive (including *Staphylococcus aureus* and a number of strains of *streptococci*) and gram-negative (enteric) microorganisms. The points were then immersed in the test solutions for five minutes, after which they were tested for bacterial growth. While NaOCl was bactericidal against the bacteria tested, hydrogen peroxide was found to have no effect on *Staphylococcus aureus* and *Penicillium sp.*.

In other studies, Senia et al. (1975) and Frank and Pelleu (1983) found a one minute immersion of gutta percha points in 5.25% NaOCl provided effective surface sterilization of gutta percha points contaminated with pure cultures of selected microorganisms (*Staphylococcus epidermis*, *Corynebacterium xerosi*, *Escherichia coli*, *Streptococcus faecalis* and *Bacillus subtilis* spores). Frank and Pelleu (1983) found a 2% glutaraldehyde

solution (Cidex 7; Johnson and Johnson Dental Products Division, East Windsor, NJ, USA) required 15 minutes to decontaminate gutta percha cones of *Bacillus subtilis* spores.

A further study by Stabholz et al. (1987) found a ten minute immersion in either 2% chlorhexidine or 5.25% NaOCl was able to decontaminate gutta percha cones previously contaminated by one of a number of bacterial strains (*Streptococcus mutans*, *Streptococcus sanguis*, *Streptococcus faecalis*, *Staphylococcus aureus*, *Escherichia coli*, *Bacillus subtilis*).

Cervone, Tronstad and Hammond (1989) used a controlled release system to deliver chlorhexidine to colonies of bacteria on blood agar plates. "Distinct zones of inhibition" were observed around the bacteria examined (*Actinobacillus actinomycetemcomitans*; *Actinomyces viscosus*; *Streptococcus mutans*; *Wolinella recta*; *Bacteroides gingivalis*; *Bacteroides intermedius*; *Eikenella corrodens*; *Pseudomonas aeruginosa*; *Enterobacter aerogenes* and *Enterobacter cloacae*). Heling et al. (1989) found 0.2% chlorhexidine had an antibacterial effect against *Streptococcus faecalis*.

Fischer and Huera (1984) suggested that use of an alkaline irrigant such as 5.25% NaOCl (pH 11) would inhibit aerobic flora growth. This was based on their findings that while growth of anaerobes was not pH-dependent, growth of aerobes had a definite relationship

to pH.

Smith and Wayman (1986) evaluated the *in vitro* effects of irrigants on *Streptococcus faecalis*, *Candida albicans* and a spore forming bacteria *Bacillus sp.* over 5 and 15 minute test periods. It was found that while 5.25% NaOCl was totally effective against all three microorganisms, sterile saline was totally ineffective. After 15 minutes, 50% citric acid destroyed 80% of *Streptococcus faecilis* but was ineffective in destroying *Candida albicans*. 25% citric acid had limited bactericidal effectiveness.

While most strains of *Bacteroides* found in the oral cavity are sensitive to penicillin (Baker et al., 1985; Haapasalo, 1989; Yamamoto et al., 1989), there is limited information available concerning the susceptibility of *Bacteroides* species to endodontic irrigants (Foley et al., 1983; Nikolaus, Wayman and Encinas, 1988).

An *in vitro* study by Foley et al. (1983) tested the effectiveness of selected irrigants in the elimination of *Bacteroides melaninogenicus* from the root canal system. It was found that while 5.25% NaOCl was highly effective, even when diluted up to 1:15,000, saline was ineffective. The authors concluded that *Bacteroides melaninogenicus* cannot be mechanically washed from the root canal system, and that it must be destroyed within the canal by an antibacterial irrigant such as NaOCl.

Nikolaus, Wayman and Encinas (1988) investigated the *in vitro* effects of 50% citric acid and 5.25% NaOCl

on four obligate anaerobic microorganisms (*Bacteroides melaninogenicus*, *Bacteroides fragilis*, *Clostridium perfringens* and *Peptostreptococcus anaerobius*) using 5 and 15 minute test times. All anaerobes were destroyed after 5 minute exposures to 50% citric acid and 5.25% NaOCl. However, volumes of irrigants used in this study were far in excess to that normally found in the apical third of the root canal.

CHAPTER 4

IRRIGANTS, DENTINE AND THE SMEAR LAYER

4.1 INTRODUCTION

The development of scanning electron microscopy (SEM) and transmission electron microscopy (TEM) has provided valuable tools for assessing the changes in dentine which occur after instrumentation and exposure to irrigants. The effects of irrigants on dentine morphology and the smear layer (also termed "smeared layer") have formed a substantial body of recent research on the efficacy of endodontic irrigants.

Because an interpretation of these studies relies upon observations of altered dentine morphology and examination the smear layer, this chapter will present a brief description of dentine morphology and a current understanding of the smear layer. The influence of irrigants on the coronal and endodontic smear layer will also be examined.

The influence of ultrasound on the smear layer will be discussed in Chapter 8.

4.2 DENTINE

4.2.1 Structure of dentine

Dentine is a hard tissue with tubules throughout its thickness. Vital dentine consists of 35% organic

matter (collagen, mucopolysaccharides) and water, and 65% inorganic material (hydroxyapatite) (Avery, 1986 p101). The normal morphology of dentine includes the highly mineralised, well defined ring of peritubular dentine forming the periphery of the dentinal tubules, in between which is found the more collagenous intertubular dentine comprising the bulk of dentine (Ten Cate, 1985 pp152-4).

At a level one millimetre from the pulp the number of coronal dentinal tubules varies from 30,000 to 40,000 tubules per mm^2 , each tubule having a diameter of 2.0-3.2 microns (Garberoglio and Brännström, 1976). However, the tubule diameter varies widely throughout the tooth, depending on distance from the pulp chamber and canal. The number of tubules per unit surface area is lower at the cementum side than at the pulpal side. In addition, there are fewer tubules in the apical region of the root than mid-root or coronal (Whittaker and Kneale, 1979; Carrigan et al., 1984; Tidmarsh and Arrowsmith, 1989). Tidmarsh and Arrowsmith (1989) found, on average, 27,000 tubules per mm^2 at a level 3 mm from the apex and half-way between the root canal and dentine-cementum junction. Near the dentine-cementum junction, the number averaged 13,000 tubules per mm^2 .

Dentine encountered in the wall of the root canal varies in morphology and structure. Primary dentine forms the bulk of the crown and root while secondary dentine, which has fewer dentine tubules per unit area, is laid down after root formation has been completed. Dentinal

tubules bend sharply as they pass from secondary to primary dentine. Predentine, which can be found lining the root canal adjacent to the pulp tissue, measures 2-6 microns in width and is not mineralised (Avery, 1986 p105).

Reparative dentine, which is produced as a defence mechanism in response to noxious stimuli, has fewer and more twisted dentine tubules than normal dentine, and varies markedly according to the different causative conditions (Mjör, 1972). Stewart (1986) commented that dense reparative dentine can frequently be found within the pulp chamber and root canal, especially in older teeth which have had restorative procedures or exposure to periodontal disease.

Sclerotic dentine, a form of reparative dentine, is composed of tubules completely occluded by deposition of calcium salts, and may be prevalent in older teeth (Nalbandian, Gonzales and Sognnaes, 1960; Avery, 1986 pp102-24). While Carrigan et al. (1984) found that dentinal tubule numbers decreased with age and proximity to the apex, Whittaker and Kneale (1979) did not find a similarly direct relationship. Tidmarsh and Arrowsmith (1989) found a high number of dentinal tubules in the apical regions of older teeth.

Because of its intrinsically porous structure, dentine is permeable to a range of substances (Pashley, 1986; Abbott, Hume and Heithersay, 1989). Regional variations in dentine produce differences in

permeability. Pashley et al. (1987) commented that the regional differences in coronal dentine may influence results in studies on dentine permeability. It can be speculated that the same observation may apply to studies on radicular dentine. Occlusal dentine is considered to be more permeable than dentine from the cervical one third of the root, and while the inner half of root dentine is more permeable than the outer half, both are much less permeable than coronal dentine (Pashley, 1988). Dentine is covered peripherally by cementum which would be expected to hinder block diffusion of substances (Pashley, 1986).

Adult dentine lining the canal wall may be blocked by atubular, irregular dentine, which is much less permeable than normal tubular dentine (Brännström, 1984; Pashley, 1989). In contradistinction, young and adult root canals might instead be lined by primary dentine into which bacteria and their products can deeply penetrate.

4.2.2 Effects of irrigants on dentinal structure

It is apparent that the variety of dentine which might be encountered by irrigants during endodontic procedures can enhance or hinder their effectiveness. In general, studies of the influence of irrigant solutions on dentine exposed after smear layer removal (see sections 4.3.2 and 4.3.4) do not take into consideration the type of dentine to which the irrigants are exposed.

Further, while some studies have restricted their tooth samples to juvenile premolars (Baumgartner and Mader, 1987) or third molars (Abbott, Hume and Heithersay, 1989), others have not specified the teeth used (Yamada et al., 1983). This last study presumably used teeth from a wide spectrum of patients, which while reflecting the typical patient population, introduces further variables to the experimental protocol. Young teeth would not be expected to have the secondary dentine routinely found in older teeth.

The removal of predentine by organic solvents such as 5.25% NaOCl has been observed under light microscopy (Rosenfeld, James and Burch, 1978) and SEM (Baumgartner and Mader, 1987). It has also been noted that under SEM, chelating agents appear to alter dentinal tubule morphology by opening tubule orifices (Baker et al., 1975; Goldberg and Abramovich, 1977; Koskinen, Meurman and Stenvall, 1980; Baumgartner and Mader, 1987). Ram (1980) believed that chelation occurs first in the intertubular dentine. Other authors have suggested that acid-etching results in loss of peritubular dentine, with a resultant widening of dentinal tubule apertures (Brännström, Glantz and Nordenvall, 1979; Pashley, Michelich and Kehl, 1981).

4.3. SMEAR LAYER

4.3.1 Coronal smear layer

Eick et al. (1970) identified a "smeared layer"

which he observed on cut surfaces of enamel under SEM. This was described as an organic film of approximately 0.5 microns thickness containing "tooth particles ranging in size from larger than 15 microns to less than 0.5 microns in length". Jones (1972) additionally noticed the presence of smear layers on instrumented calculus and cementum.

The smear layer was further defined by Cotton (1984) as "any debris produced iatrogenically by the cutting, not only of dentine, but also of enamel, cementum, and even the dentine of the root canal".

The type of instrument used during cutting, as well as presence or absence of coolant, influences the release of energy during cutting. Smearing and heat-coagulation of surface-dentine proteins during cutting of dentine is thought to contribute to the formation of the smear layer (Brännström, Glantz and Nordenvall, 1979).

A substantial body of research exists concerning the production of the smear layer during cutting procedures on coronal dentine, which includes Eick et al., 1970; Brännström and Johnson, 1974; Brännström, Nordenvall and Glantz, 1980; Gilboe et al., 1980; Boyer and Svare, 1981; Bowen et al., 1984; Pashley et al., 1978;1985; and Berry, von der Lehr and Herring, 1987. Research in this field has been stimulated by the expansion of adhesive restorative dentistry.

It has been noted that different methods of coronal dentine preparation produce morphologically different

smear layers (Eick et al., 1970; Brännström, Glantz and Nordenvall, 1979; Gilboe et al., 1980; Dippel, Borggreven and Hoppenbrouwers, 1984; Gwinnett, 1984). Gwinnett (1984) noted that preparation of coronal dentine by rotary burs with and without a water coolant produced a significant difference in morphology of the smear layer. Use of coarse diamond burs without coolant produced thicker and more widely spread deposits than with coolant. The depth also varied according to the conditions under which the dentine was cut.

The exact constituents of the intact smear layer are beyond the resolution of SEM (Pashley, 1984). However, following ultrasonification of the smear layer, globular particles measuring 0.05 to 0.1 microns in diameter have been identified under high resolution SEM (Pashley et al., 1988).

It has been estimated, from *in vitro* research on prepared coronal dentine surfaces, that removal of the smear layer increases the permeability of dentine by 10% to 40% (Pashley et al., 1978; Dippel, Hoppenbrouwers and Borggreven, 1981; Pashley, Michelich and Kehl, 1981; Dippel, Borggreven and Hoppenbrouwers, 1984; Pashley and Galloway, 1985). Furthermore, smear layer removal increases the dentinal tubule area available for diffusion (Pashley et al., 1978; Boyer and Svare, 1981; Dippel, Borggreven and Hoppenbrouwer, 1984), filtration (Boyer and Svare, 1981), and hydraulic conductance (Pashley et al., 1978; Reeder et al., 1978). It has been

reported that patients have experienced increased sensitivity following smear layer removal from unanaesthetised dentine (Johnson and Brännström, 1974).

Brännström and Nyborg (1973) found that bacteria could survive in the smear layer under restorations, and postulated that these microorganisms could obtain nourishment from both the smear layer and dentinal fluid. Suggestions have been made that bacterial products may diffuse through the smear layer, infiltrate the coronal dentinal tubules and enter the underlying pulp (Brännström and Nyborg, 1973; Bergenholtz, 1977; Bergenholtz and Reit, 1980; Brännström, 1984). Pashley et al. (1985) further speculated that filtration or bulk fluid movement of bacterial products might occur in response to masticatory forces. Williams and Goldman (1985) found that the presence of the smear layer delayed, but did not prevent, the *in vitro* passage of bacteria through dentine.

Conversely, it has been suggested that the smear layer produced during restorative procedures may act as a diffusion barrier, thereby preventing passage of bacteria into tubules (Pashley, Michelich and Kehl, 1981; Diamond and Carrel, 1984). *In vitro* studies have shown that removal of the smear layer from coronal dentine allows penetration of bacteria into the tubules (Michelich, Schuster and Pashley, 1980; Williams and Goldman, 1985; Meryon, Jakeman and Browne, 1986). Retention of the smear layer, however, does not prevent the passage of fluids,

and thus potentially, the diffusion of bacterial toxins through dentinal tubules (Bergenholtz, 1977; Michelich, Schuster and Pashley, 1980; Pashley, Michelich and Kehl, 1981).

It has also been suggested that the depth of tubule plugging, rather than the extent of surface coverage by the smear layer, is responsible for the degree of permeability of dentinal tubules (Dippel, Borggreven and Hoppenbrouwers, 1984).

4.3.2 Effects of irrigants on coronal smear layer

The smear layer can be removed from coronal dentine by application of demineralising agents such as 50% citric acid, 40% and 25% polyacrylic acid, 37% phosphoric acid, 20% and 50% lactic acid, and various concentrations of EDTA (Brännström and Johnson, 1974; Leidal and Eriksen, 1979; Brännström, Nordenvall and Glantz, 1980; Jodaikin and Austin, 1981; Pashley, Michelich and Kehl, 1981; Ando, 1985a; Berry, von der Lehr and Herring, 1987; Meryon, Tobias and Jakeman, 1987; Meryon, Tobias and Johnson, 1988).

The coronal smear layer cannot be removed by hydrogen peroxide, NaOCl or water (Brännström, Glantz and Nordenvall, 1979; Leidal and Eriksen, 1979; Brännström, 1984; Berry, von der Lehr and Herring, 1987).

4.3.3 Radicular smear layer

Most investigations examining the smear layer have

been performed on coronal dentine following cutting procedures (Eick et al., 1970; Bowen et al., 1984; Gwinnet, 1984), thus producing a smear layer potentially different to that found following endodontic procedures. However, Brännström (1984) has hypothesised that the smear layers produced are likely to be morphologically similar.

The presence of the smear layer on the surface of endodontically instrumented radicular dentine has been observed under SEM by many researchers including Baker et al., 1975; McComb, Smith and Beagrie, 1976; Lester and Boyde, 1977; Rubin et al., 1979; Bolanos and Jensen, 1980; Goldman et al., 1982; Cymerman, 1983; Baumgartner et al., 1984; Cameron, 1987; Baumgartner and Mader, 1987; Krell and Dang, 1987; and Farouz, Delzangles and Laurent, 1988a. Based upon coronal smear layer research, as well as the fact that it has not been observed on uninstrumented root canal walls (Lester and Boyde, 1977; Moodnik et al., 1976; Goldman et al., 1981; Mader, Baumgartner and Peters, 1984), the existence of a radicular smear layer can be assumed to be a result of instrumentation.

The radicular smear layer produced by K-type files has been described by Mader, Baumgartner and Peters (1984) to consist of "two confluent components; the smeared layer on the surface of the canal wall and the smeared layer which had been packed into some of the dentinal tubules." The superficial layer on the surface

of the canal wall was typically 1-2 microns thick, and the component packed into the dentinal tubules, from a few up to 40 microns in depth, with a surface appearance which is "typically amorphous, irregular and granular".

The radicular smear layer is thought to be primarily inorganic in nature with organic components (Lester and Boyde, 1977; Goldman et al., 1981; Baumgartner et al., 1984; Baumgartner and Mader, 1987). Goldman et al. (1981) suggested that the smear layer was mostly calcific and "probably a slurry of dentine filings".

The biologic half-life of the smear layer is unknown, but obviously would be dependent upon the immediate environment. An experimentally produced smear layer left on dentine exposed to the oral environment will disappear after two days (Brännström, 1984). It has been postulated that under a well condensed root filling, the smear layer may remain intact for many years (Pashley, 1989).

In endodontics, the clinical significance of the presence of the smear layer has yet to be determined (Bolanos and Jensen, 1980; Goldberg and Abramovich, 1977; Goldman et al., 1981; 1982; Berg et al., 1986; Alaçam, 1987; Walton and Torabinejad, 1989 p206). The necessity for its removal has been questioned by some authors (Yamada et al., 1983; Dow, 1984; Callis and Sabatini, 1985), while others have suggested that its removal may allow better penetration of intracanal medicaments,

sealers and gutta percha (Goldberg and Abramovich, 1977; Yamada et al., 1983; White, Goldman and Sun Lin, 1984, 1987; Buchanan, Buchanan and Ryder, 1985; Cergneux et al., 1987; Abbott, Heithersay and Hume, 1988; Farouz, Delzangles and Laurent, 1988; Gettleman, Messer and ElDeeb, 1989; Abbott, Hume and Heithersay, 1989a). It has also been postulated that the smear layer may obliterate lateral or accessory canals (Rubin et al., 1979).

Brännström (1984) believed that while it was desirable to remove smear plugs in the root canal to allow medicaments to reach microorganisms in the dentinal tubules, if the coronal seal was poor, then an increased risk of reinfection via widened and open tubules existed. Even so, if coronal seal is poor, it is feasible that organic acids produced by invading bacteria may etch away the smear layer (Pashley and Galloway, 1985).

Suggestions have been made that bacteria and necrotic pulp tissue may become incorporated into the smear layer (Byström and Sundqvist, 1985; Rome, Doran and Walker, 1985). Safavi, Spångberg and Langeland (1989) demonstrated *in vitro* that facilitation of passive penetration of *Streptococcus faecium* into root dentine resulted after removal of the smear layer. Gutiérrez et al. (1982) found that radicular dentine became more permeable to bacteria after removal of the smear layer, indicating that the presence of the smear layer inhibited penetration of bacteria into radicular dentine.

The incidence of external inflammatory root

resorption has been associated with bacteria and their products entering dentinal tubules via the root canal (Andreason, 1981; Hammarström et al., 1986; 1986a; 1986b; Lindskog, Berg and Pierce, 1986). While removal of the smear layer would allow improved diffusion of medicaments (Abbott, Hume and Heithersay, 1989a), its retention could provide protection against bacterial penetration into dentinal tubules, though not against bacterial products.

Some *in vitro* studies have found that the absence of a smear layer provides no benefits to the integrity of the apical seal (Biesterfeld and Taintor, 1980; Madison and Krell, 1984; Goldberg et al., 1985; Evans and Simons, 1986). Conversely, other authors have demonstrated that elimination of the smear layer provides significantly reduced apical leakage *in vitro* (Kennedy, Walker and Gough, 1986; Cergneux et al., 1987).

Lester and Boyde (1977) advised that if the smear layer was to be removed, then sealer particle size needed to be sufficiently small to allow sealer to enter the tubules. It has been shown *in vitro* that removal of the smear layer from root canal dentine allows improved adaptation of bis-GMA unfilled resin to dentinal tubules (Goldman, DeVitre and Pier, 1984; Goldman et al., 1984).

Callis and Sabatini (1985) histologically observed migration of polymorphonuclear leucocytes into patent dentinal tubule openings of endodontically treated teeth implanted into rats and guinea pigs. They hypothesised that a "possible polymorphonuclear leucocyte-mediated

chronic irritation of the periapical tissues" could occur. For this reason they questioned the necessity to remove smear layer, and suggested that opened tubules should be sealed with the smear layer or root canal filler.

Removal of the smear layer may allow better penetration of antibiotic medicaments (Hammarström et al. 1986; 1986a; 1986b; Abbott, Hume and Heithersay, 1989), though possibly not calcium hydroxide (Wu et al., 1989). Kennedy, Walker and Gough (1986) observed that sclerotic radicular dentine was relatively impervious to the penetration of dye regardless of smear layer.

4.3.4 The effects of irrigants on radicular smear layer

The presence or absence of the smear layer is frequently used to assess irrigant efficacy. While no single irrigant has been found to remove both the smear layer and organic debris, the ability of chelating irrigants to remove the smear layer has been shown (McComb and Smith, 1975; McComb, Smith and Beagrie, 1976; Goldberg and Abramovich, 1977; Goldman et al., 1981; Goldberg and Spielberg, 1982; Yamada et al., 1983; Baumgartner et al., 1984). SEM studies have revealed the effectiveness of EDTA and citric acid in both removing the smear layer and increasing the opening of dentine tubules (Tidmarsh, 1978; Goldman et al., 1981; Baumgartner et al., 1984; Dippel, Borggreven and Hoppenbrouwers, 1984; Baumgartner and Mader, 1987; Berg

et al., 1986).

McComb and Smith (1975) found, *in vitro*, that sealing REDTA¹ (Roth Drug Company, Chicago, Ill., USA) in the canal for 24 hours, followed by subsequent irrigation with water, resulted in an "exceptionally clean canal" with smear layer "entirely removed" to reveal the dentinal tubules. A subsequent investigation by McComb, Smith and Beagrie (1976) examined root canals treated with different irrigating solutions immediately prior to extraction. While it was found that 6% EDTA (pH 8) with cetrimide most consistently produced the cleanest canal walls, none of the irrigating regimens were able to remove both smear layer and superficial debris completely.

Prolonged exposures (3 hours) to 5.25% NaOCl and to an ampholytic soap (1% dodecyldiaminoethyl glycine) have been unsuccessful in removal of the radicular smear layer (Goldman et al., 1981). Other authors have found that NaOCl does not remove the smear layer from radicular dentine (Yamada et al., 1983; Rome, Doran and Walker, 1985; Baumgartner and Mader, 1987; Ciucchi, Khettabi and Holz, 1989).

Because no single irrigant has been found to remove both organic and inorganic debris from instrumented canals, the need for separate solutions to remove each

1. REDTA contains 17.00 gm EDTA, 0.84 gm cetrimide, 5N sodium hydroxide solution 9.25 ml and distilled water 100.00 ml (McComb and Smith, 1975).

component has been suggested (Loel, 1975; Wayman et al., 1979; Koskinen, Meurman and Stenvall, 1980; Koskinen, 1981; Goldman et al., 1981; Yamada et al., 1983; Baumgartner and Mader, 1987). A high volume final flush with both NaOCl and EDTA has been proposed (Goldman et al., 1982; Yamada et al., 1983). The alternate use, during preparation, of 15% EDTA and 5.25% NaOCl (Baumgartner and Mader, 1987), 50% citric acid and 5% NaOCl (Loel, 1975) or 10% citric acid and 2.5% NaOCl (Wayman et al., 1979) has also been recommended.

Goldman et al. (1981) observed that REDTA removed the smear layer but not soft debris, and suggested that a combination of irrigants be investigated. A later study (Goldman et al., 1982) examined the effect of a final flush with REDTA followed by 5.25% NaOCl, and vice versa. The former regimen was found to be superior. It was speculated that this resulted from the organic component of the smear layer being exposed after use of REDTA thus allowing the organic solvent NaOCl to act upon it.

The third part of this study (Yamada et al., 1983) confirmed the effectiveness of a final flush with 17% EDTA followed by 5.25% NaOCl, but the reverse combination was not part of the experimental protocol. However, when the solutions were used alone they were less effective for removal of superficial debris and smear layer, respectively.

Instrumentation with 10% citric acid and 2.5% NaOCl followed by a final flush of citric acid was found to

consistently result in clean canal walls, although crystalline deposits were noted on the dentine surfaces following use of citric acid (Wayman et al., 1979). Baumgartner et al. (1984) found that the sequence of use of citric acid and NaOCl was insignificant in removal of superficial debris and smear layer.

Baumgartner and Mader (1987) instrumented one half of individual canals, leaving the other half uninstrumented. It was found that the combination of 15% EDTA and 5.25% NaOCl was able to remove organic debris and predentine from the uninstrumented half, as well as the smear layer from the instrumented half.

CHAPTER 5

SODIUM HYPOCHLORITE

5.1 INTRODUCTION

Sodium hypochlorite is probably the endodontic irrigant most commonly in use today (Ingle et al., 1985 p180). It has been attributed with three properties essential to an ideal irrigant; namely, tissue solvent capability, antibacterial properties and, when used appropriately, tissue tolerance.

The introduction of NaOCl to medicine is credited to Dakin (1915) who reported the use of a 0.5%-0.6% NaOCl solution ("Dakin's solution") for the irrigation of wounds incurred by soldiers during World War I. This solution was prepared by dissolving sodium carbonate in tap water and adding chlorinated lime. Boric acid was then added to the resultant filtrate, producing a solution with a one week shelf life. Too much free alkali was to be avoided because of its purported irritant effects. Dakin's solution was a standard product of the British Pharmacopeia up to 1968 (Dychdala, 1983 p158).

The earliest use of NaOCl in endodontics has been attributed to AB Crane in 1920 (Cunningham and Joseph, 1980). In 1936, Walker recounted his clinical observations following use of a double strength chlorinated soda solution (see p12) during endodontic

treatment, a regimen purportedly suggested by Dr Blass of New York University. Based on his clinical experience, Walker described this solution as a powerful germicide, as well as a "very satisfactory" organic solvent. The solvent properties of double strength chlorinated soda solution were later confirmed by Grossman and Meiman (1941).

Stewart (1955) believed that repeated irrigation using NaOCl could help in removal of necrotic tissue from the root canal, while Masterton (1965) claimed good post-operative results (clinically and radiographically) with chlorinated soda. Sommer, Ostrander and Crowley (1966 p181) recommended the use of 4%-6% NaOCl with tincture of green soap. Gresham (1983) maintained that "freshly made electrolytic NaOCl" was the "ideal solution for endodontic therapy".

This chapter will discuss the properties and characteristics of NaOCl which have led to its recommendation, in several current endodontic texts, as the irrigant of choice (Weine, 1982 p318; Ingle et al., 1985 p185; Gutmann and Dumsha, 1987 p157; Grossman, Oliet and del Rio, 1988 p188; Walton and Torabinejad, 1989 p206).

5.2 CHEMISTRY, MANUFACTURE AND STABILITY

"Sodium hypochlorite solution is one of a number of preparations (known as chlorophers) which release chlorine in the form of hypochlorous acid (HOCl). These

agents are potent germicides and bleaching agents, and they readily dissolve blood clots and necrotic debris." (Deuben, 1980 pp660-1).

Chlorine exerts its action in both the elemental state and as undissociated HOCl, which is formed by the hydrolysis of chlorine (Harvey, 1980 p973). The germicidal efficacy of a chloropher is directly associated with the ease and degree of liberation of HOCl (Penick and Osetek, 1970; Harvey, 1980 p974).

"Available chlorine" is an indicator of oxidizing capacity. Penick and Osetek (1970) stated that a 5% NaOCl solution yielded approximately 1% available chlorine. Chlorine has an affinity for electrons thus making it a strong oxidizing agent. Once the chlorine atom is reduced to chloride it ceases to possess disinfectant properties. Reactions with inorganic substances are rapid, but with organic substances, are generally slow and dependent on the concentration of the available chlorine (Dychdala, 1983 p158). Reactions of NaOCl rely heavily upon covalent bonding or on formation of strong ionic attachments, and are not usually dependent on hydrogen bonding and van der Waals forces (Myers and Yagiela, 1980 p19).

Because chlorine is very reactive and bound by organic debris, the formation of HOCl can be blocked, and thus bactericidal activity reduced (Penick and Osetek, 1970; Harvey, 1980 p973). Therefore, it has been stressed that "the full germicidal potential of NaOCl cannot be realised until thorough debridement has been

accomplished" (Penick and Osetek, 1970). Cotter et al. (1985) noted that reactions of NaOCl with proteins and other cellular debris resulted in depleted antimicrobial activity. Consequently, for continuation of maximum antimicrobial activity, the reapplication of NaOCl was necessary.

The bactericidal action exerted by chlorine was described by Knox et al. (1948) as resulting from the oxidation of sulfhydryl enzymes essential to the glycolysis process. Spångberg (1973a) further defined this action as oxidation of enzymatic proteins containing cysteine (which has a side-chain terminating in a sulfhydryl group). As these enzymes can only function if the sulfhydryl group remains reduced and free, the effective binding of these sulfhydryl groups by halogens, such as chlorine, has a very destructive effect on the cell. The sulfhydryl groups of coenzyme A (a carrier of acyl groups) can also be oxidized by chlorine (Spångberg, 1973a).

The pH level greatly influences the concentration of undissociated HOCl, and thus the antimicrobial activity of chlorine in solution. Lowering the pH increases the biocidal activity of NaOCl (Bloomfield and Miles, 1979; Harvey, 1980 p973), while increasing the pH significantly decreases this activity (Dychdala, 1983 p163). However, when NaOCl solutions are formulated at a lower pH they are less stable (Cotter et al., 1985).

Pashley et al. (1985) described a 5.25% solution

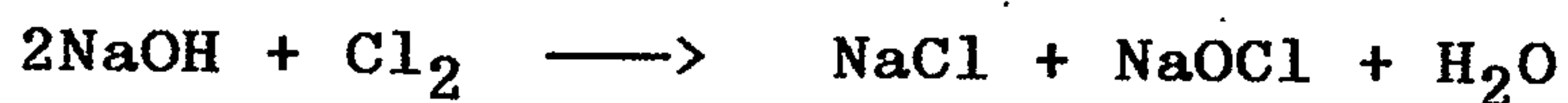
of NaOCl as containing about 5% NaCl in a very alkaline solution (pH 11) of NaOH, thus making the solution very hypertonic (≈ 2800 mOsmol/kg). These authors maintained that the clinical efficacy of the solution resulted from its ability to oxidize, hydrolyse and, to some extent, osmotically draw fluids out of tissues.

A source of 5.25% NaOCl, commercially known as Clorox (The Clorox Company, Oakland, California, USA), was introduced by Lewis in 1954 (Trepagnier, Madden and Lazzari, 1977). Shih, Marshall and Rosen (1970) detailed Clorox (pH 11.0-11.5) as containing:-

NaOCl	5.25%
Sodium carbonate	0.20%
Sodium chloride	4.00%
Free sodium hydroxide	0.005-0.015%

References to "Zonite", a commercially available 5% NaOCl solution, can be found in earlier dental literature (Penick and Osetek, 1970). A currently utilised NaOCl preparation is Milton solution (Richardson-Vicks Ltd, Egham, Surrey, England), which contains either 1% or 2% (w/v) NaOCl and 16.5% (w/v) sodium chloride (Lee, Miles and Perry, 1985). A 1% Milton solution has 0.12% available chlorine (Lester and Boyde, 1977). Newman (1983) questioned the use of 2% Milton solution. He was "astounded to see the degree of crystalline encrustation" in the dental suction unit after use of this product.

The means by which NaOCl is now manufactured (in NSW) was described by Cameron (1988). The different commercially available NaOCl solutions result from variations in diluting, buffering, stabilising and packaging of the stock solution. Preparation of the stock solution involves passing chlorine gas over a column of sodium hydroxide, which produces a solution containing 12.5% available chlorine and 0.1% to 0.5% sodium hydroxide:-



The presence of sodium hydroxide raises the pH to 12, thus increasing stability of the solution. Reduction of pH is possible by dilution of the solution or by addition of sodium chloride. Increases in stability at low concentrations have an exponential value, with the shelf life of 12%, 5% and 3% NaOCl solutions being approximately one, four and ten years respectively.

Two reactions occur during the decomposition of NaOCl, the second of which is representative of 85% of the decomposition reaction (Cameron, 1988):-



Grossman (1943) claimed that storage of double strength chlorinated soda solution in a cool place would allow a shelf life of three months, and storage in brown glass bottles was recommended by Moorer and Wesselink

(1982) to allow stability for at least one month. Velvart (1987a) found that the concentration of test solutions of 0.5%, 1% and 2% NaOCl did not change over an observation period of two to six months when stored in dark brown bottles at either of two temperatures, 4°C and room temperature. A diluted Milton solution was found to be stable for at least one year at room temperature (Morris, Kingsley and Chinwah, 1980).

Cunningham and Balekjian (1980) tested the stability of NaOCl maintained at 37°C. After 24 hours, 5% and 2.5% NaOCl showed a decrease in content of available chlorine of 9.5% and 4% respectively.

The use of 5% thiosulphate solution to inactivate NaOCl was suggested by Möller (1966). Cameron (1987a) suggested that irrigation with local anaesthetic solution arrested the solvent activity of NaOCl.

5.2.1 Influence of surface tension

"An open surface of a *liquid* is under a state of tension, causing a tendency for the portions of the surface to separate from each other; the surface thus shows properties similar to those of a stretched elastic film over the liquid. The tension is an effect of the forces of attraction existing between the *molecules* of a liquid" (Uvarov, Chapman and Isaacs 1964 p294).

A surfactant is a substance introduced into a liquid in order to influence properties of the liquid which depend on surface tension, such as spreading and

wetting. It has been suggested that "low surface tension and good penetrating ability are desirable" in an irrigant (Hakala, Koskinen and Närvänen, 1976).

Efforts have been made to improve the flow of NaOCl by the addition of surfactants, with the aim of decreasing surface tension thereby enabling better penetration and cleansing of narrow, confined portions of the root canal system (Lehman, Bell and Gerstein, 1981; Abou-Rass and Patonai, 1982; Cunningham, Cole and Balekjian, 1982; Cameron, 1986a).

Lehman, Bell and Gerstein (1981), using a light microscope to view the canal walls, found that the addition of a surfactant, sodium lauryl sulfate (SLS), to NaOCl did not induce a more debris-free surface. In fact, 5.25% NaOCl alone removed more debris than when combined with SLS. In addition, combinations of 1% SLS and 0.525% NaOCl, as well as 0.1% SLS and 5.25% NaOCl formed precipitates attributed to pH imbalances and could not be used. It was noted though, that those solutions containing SLS were "easier to work with than those that did not", which was attributed to the lubricating ability of the solution.

In another study ethanol was added to an unspecified concentration of NaOCl (Cunningham, Cole and Balekjian, 1982). While a solution containing equal parts of NaOCl and ethanol showed a significantly increased ability to spread, the solution was almost completely depleted of available chlorine within 15 minutes, and was

therefore seen as inappropriate for clinical use. The authors recommended a freshly mixed combination of seven parts NaOCl and three parts ethanol.

An evaluation was made of the effect of reducing the surface tension by addition of a surfactant (Polysorbate 80) to 2.6% and 5% NaOCl (Abou-Rass and Patonai, 1982). The authors asserted that this reduced surface tension by between 15% and 20%, producing enhanced flow properties over the control.

Cameron (1986a) added a fluorochemical surfactant Fluorad FC99 (3M, St Paul, MN, USA) to 4% NaOCl. Although reduction in surface tension (from 70mN/m to 27mN/m) was maintained over a one month period, by the second month, stability of the test solution had declined.

5.3 SOLVENT ACTION

Two early studies investigated the solvent action and behaviour of Dakin's solution (Taylor and Austin, 1918; Austin and Taylor, 1918). While Dakin's solution was found to be an effective solvent of liver emulsion (Taylor and Austin, 1918), another investigation observed intense inflammation of previously healthy rabbit ears after their immersion in Dakin's solution for seven days (Austin and Taylor, 1918).

Using different experimental methods, samples, and volumes of NaOCl, several authors have observed a more effective solvent action of NaOCl at higher concentrations (Hand, Smith and Harrison, 1978; Thé,

1979; Cunningham and Balekjian, 1980; Koskinen, Meurman and Stenvall, 1980; Koskinen, Stenvall and Uitto, 1980; Abou-Rass and Oglesby, 1981; Nakamura et al., 1985) and higher temperature (Morris, 1958; Cunningham and Balekjian, 1980; Cunningham and Joseph 1980; Nakamura et al., 1985). In contrast, other studies have found variations in temperature (Litton and Schilder, 1989) and concentration (Gordon, Damato and Christner, 1981) exert no or inconsistent control over solvent action.

It would appear that differences in tissue type, size, and volume of solution significantly affect results of *in vitro* studies. In one study, bovine pulps and tendon collagen were more easily dissolved than bovine gingiva by NaOCl (Nakamura et al., 1985). In addition, while Gordon, Damato and Christner (1981) found necrotic tissue dissolved more readily than fresh tissue in NaOCl, other authors have demonstrated the reverse (Thé, 1979; Abou-Rass and Oglesby, 1981). However, fixed tissue¹ appears to require the longest time for dissolution (Thé, 1979). Gordon, Damato and Christner (1981) commented that necrotic pulp produced *in vitro* is likely to be quite different from that occurring *in vivo*, where products of the immune system would be expected to be encountered.

The solvent action of NaOCl was formerly assessed

1. A common practice exists in Europe where necrotic pulp tissue is fixed with parachlorophenol or formaldehyde "to avoid transient bacteraemia during mechanical preparation". However, the tissue is apparently subsequently difficult to dissolve (Thé, 1979).

macroscopically, but now sophisticated assays are used where the hydroxyproline content of the used irrigant can be measured, thus reflecting the amount of collagen dissolution¹. However, Koskinen (1981) commented that measurements of hydroxyproline content may give misleading results as the collagen or hydroxyproline that is broken down or altered beyond recognition is not taken into account. This would be of greater significance when higher concentrations of NaOCl are used.

Hydroxyproline has been detected in used irrigating solutions of 2.5% and 5% NaOCl by several authors (Trepagnier, Madden and Lazzari, 1977; Wayman et al., 1979; Zach and Kaufman, 1983). In contrast, Koskinen, Stenvall and Uitto (1980) found that although loss of hydroxyproline from tissue samples was measurable, hydroxyproline was unable to be detected in irrigant extracts.

A recent study found that pretreatment of necrotic porcine muscular tissue with calcium hydroxide enhanced the tissue dissolving effect of 0.5% NaOCl (Hasselgren, Olsson and Cvek, 1988). The authors suggested that the tissue became more accessible to NaOCl after the hydroxide component of calcium hydroxide caused

1. The presence of collagen in pulp tissue is well established (Avery, 1986 p143). Uitto and Ranta (1973) found that collagen forms approximately 37% of the protein content of pulps of intact premolars with complete root formation, Gordon, Damato and Christner (1981) described pulp tissue as containing 15% collagen of which 13% is hydroxyproline.

denaturation and hydrolization. However, in this study, volumes of solutions were larger than normally would be encountered clinically.

5.3.1 Influence of temperature

Cunningham and Balekjian (1980) compared the collagen-dissolving ability of 2.6% and 5.2% NaOCl at room temperature (21°C) and at body temperature (37°C). They found 2.6% NaOCl heated to 37°C to be equally effective a tissue dissolving agent as 5.25% NaOCl at either room or body temperature.

Another study placed samples of fresh, fixed, and necrotic rat connective tissue (1-2 mm thick and 8mm diameter) in large volumes (30 ml) of either 2.6% or 5.25% NaOCl (Abou-Rass and Oglesby, 1981). Temperature was maintained at either 22°C or 60°C (a temperature considered clinically tolerable). Complete dissolution of samples was most rapidly attained at the higher concentration and temperature for all tissue types.

Nakamura et al. (1985) included 2%, 5% and 10% NaOCl at temperatures of 4°C, 22°C and 37°C in their *in vitro* study on the solvent action of 1 ml NaOCl on 10 mg of bovine tissue. While 10% NaOCl at 37°C was the most effective dissolving regimen, there was no difference between 2% and 5% at any of the temperatures. Dissolution was assessed by measurement of hydroxyproline content in used solution samples.

Litton and Schilder (1989) found that NaOCl heated

to 60°C "rapidly comes to thermal equilibrium with the tooth upon irrigation" *in vivo*, and that this final temperature was close to body temperature.

5.3.2 Influence of concentration

Over the years a variety of concentrations of NaOCl have been suggested for endodontic use. A 5% solution has been recommended by some authors (Grossman, 1943; Leubke, 1967; Blackler, 1981; Weine, 1982 p318), 1% by Martin (1979), at least 2.5% by Mentz (1982), 3% by Thé (1979), and 0.5% by Spängberg (1973). Sampeck (1967) recommended 4%-6% while Hand, Smith and Harrison (1978) suggested 5.25%. Trepagnier, Madden and Lazzari (1977) advised 2.5% or 5% while Moorer and Wesselink (1982) recommended between 0.5% and 2%. Aron (1987a) recommended a concentration no higher than 0.5%. A 2% (Cameron, 1988b) or 5% (Crabb, 1982) NaOCl solution activated by ultrasound has also been suggested.

A comparison of 2.5% and 5.25% NaOCl following application to previously instrumented teeth over a 5 minute period, showed no significant difference in the amount of hydroxyproline content in the used irrigants. However, 0.5% NaOCl was found to have little solvent action (Trepagnier, Madden and Lazzari, 1977).

The effect of dilution of NaOCl on its solvent properties was also examined by Hand, Smith and Harrison (1978). These authors investigated the ability of different concentrations of NaOCl to dissolve necrotic

tissue (epithelium and connective tissue of rats). By calculating the change in weight of specimens after a 7 minute exposure to various concentrations of NaOCl (5.25%, 2.5%, 1%, 0.5%), it was found that dilution of NaOCl produced a significant decrease in its ability to dissolve necrotic tissue.

In a qualitative study, Thé (1979) performed experiments on necrotic abdominal wall tissue of the rat. A 3% NaOCl solution was found to be more effective than 1% or 2% NaOCl, especially when employed over a longer tissue contact time. Thé believed that the use of a 3% NaOCl solution could result in "adequate" dissolution of necrotic tissue.

Gordon, Damato and Christner (1981) subjected fresh and necrotic foetal bovine pulp tissue to several concentrations of NaOCl *in vitro*. The authors concluded that 3% NaOCl was a more effective tissue solvent than 1% NaOCl. It was found that 3% and 5% NaOCl were equally effective in dissolving fresh tissue after two minutes exposure, and after five minutes, 1%, 3% and 5% NaOCl were equally effective in dissolving 90% of necrotic tissue. These results, however, have not been reproduced by any other researchers, and may have been influenced by the large volumes of solutions used (10 ml) relative to specimen size (4 mg).

Koskinen, Stenvall and Uitto (1980) measured the dissolution of uninfected minced and freeze-thawed bovine pulp tissue (100 mg samples) in NaOCl (3 ml). Measurement

of loss of collagen and change in dry weight of specimens was used to assess dissolution. NaOCl in 5% and 2.5% concentrations was found to be effective, but not 0.5%. From these results, the authors extrapolated that clinically, 1% NaOCl was too weak for use in dissolution of necrotic pulp tissue.

A later study by Koskinen (1981) found 0.5% NaOCl to be ineffective, but 2.5% and 5% NaOCl were effective in dissolving the organic component of powdered root dentine after a ten minute application. This study was also able to measure a drop in the pH of NaOCl solutions after exposure to powdered dentine.

In vitro studies by Litton and Schilder (1989) indicated that 2%, 3%, 4% and 5% NaOCl at 37°C had a similar solvent action over time on human dura tissue.

5.3.3 Influence of volume

An association between the amount of exposed surface area and dissolution by NaOCl has been made by several authors (Shih, Marshall and Rosen, 1970; Senia, Marshall and Rosen, 1971; Hand, Smith and Harrison, 1978; Thé, 1979; Koskinen, Stenvall and Uitto, 1980; Gordon, Damato and Christner, 1981). Thé (1979) asserted that the solvent action of NaOCl depended on the ratio of the amount of tissue to the quantity of solution. Hasselgren, Olsson and Cvek (1988) noted that continuous exchange of fresh solution enhanced the capacity of 0.5%

NaOCl to dissolve tissue.

Moorer and Wesselink (1982) presented a comprehensive paper investigating the interaction of three different concentrations (0.6%, 1.2% and 3%) of NaOCl with various concentrations of protein hydrolysate. It was found that when NaOCl was in excess, most of its activity was retained. However, when organic matter was in excess, the activity of NaOCl was rapidly depleted, and the pH drastically lowered during the first moments of the reaction. Thus, the authors concluded that the frequency of irrigation, as well as mechanical agitation of the irrigant during preparation, was more important than the concentration of the solution.

5.4 QUALITATIVE STUDIES

5.4.1 Light microscopy

Under a binocular microscope, Gutiérrez and Garcia (1968) observed a heavy precipitation of salts on canal walls which they attributed to oversaturation of irrigating solutions such as saline, EDTA and NaOCl (concentration not provided). The authors found no difference in effectiveness of debridement between saline and NaOCl. Nicholls (1984 p146) recommended use of these irrigants to be immediately followed by irrigation with sterile water to remove any crystals.

Senia, Marshall and Rosen (1971) instrumented the mesial canals of extracted lower first molars to size 30, and then applied 5.25% NaOCl claiming "one drop from the

syringe filled the entire canal". This was then stirred and "carried apically every 5 minutes by means of a number 10 reamer". No difference was found between a 15 or a 30 minute application. In this study, the authors used photomicrographs and concluded that 5.25% NaOCl was not an effective tissue solvent in small canals, and further, that cleansing of the apical 5 mm was inadequate using this technique. However, the canals were prepared using saline as an irrigant, and NaOCl was added only after canal preparation was complete. In addition, as will be discussed in Chapter 10, their method of irrigation was unlikely to be adequate.

Moodnik et al. (1976) took 1,100 photomicrographs of 25 canals which had been flushed during instrumentation with either saline or 2.5% NaOCl. No difference in cleanliness was found at different levels of the canal when using either irrigant.

A light microscope study of the effect of 5.25% NaOCl on vital pulps of noninstrumented young premolars demonstrated that destruction of predentine was the most consistent finding (Rosenfeld, James and Burch, 1978). While NaOCl was found to exert a "nonspecific, noncoagulating digestive effect on vital, young healthy human pulp tissue", this was limited by the access and diameter of the canal. The authors stressed that 5.25% NaOCl had limited capability to dissolve the confined deeper pulpal tissue.

5.4.2 Scanning electron microscopy

Studies which have employed SEM to evaluate the effects of a range of irrigants or combinations of irrigants, have generally found NaOCl to be effective in the removal of superficial debris (Goldman et al., 1981; 1982; Baumgartner et al., 1984; Yamada et al., 1983; Madison and Krell, 1984; Berg et al., 1986). Other studies have found that the use of NaOCl as an irrigant does not provide a superior cleaning result over other irrigants (Baker et al., 1975; Rubin et al., 1979; Bolanos and Jensen, 1980).

Baker et al. (1975) concluded that debridement was a function of quantity rather than type of irrigant (however in this study 1% NaOCl was the only concentration included). Rubin et al. (1979) considered that instrumentation was the crucial component of biomechanical preparation rather than type of irrigant, though 2.5% NaOCl was described as "an excellent pulpal and predentine solvent" of exposed pulps of split teeth.

An *in vitro* study found that liberal use of either 1% or 6% NaOCl as the sole irrigant was the most efficient irrigant for removal of debris, although 1% was considered to be less effective (McComb and Smith, 1975). A later investigation, in which teeth were instrumented just prior to extraction, examined irrigants including 1% and 2.5% NaOCl and water (McComb, Smith and Beagrie, 1976). Under SEM there was evidence of canal walls with smear layer intact but less superficial debris than with

water.

Another paper concluded that use of 5.25% NaOCl solution produced greater tissue dissolution than 50% aqueous citric acid after 5 minutes, but a less clean canal wall (Kopp, Lazzari and Madden, 1977). Wayman et al. (1979) similarly found that, although 5.25% NaOCl provided significantly greater organic tissue dissolution than either lactic or citric acid, debris could still be detected. Baumgartner et al. (1984) found 5.25% NaOCl to be superior to citric acid in removal of superficial debris from the apical third of the canal.

After instrumented canal walls were treated with 5% NaOCl for three days, Lester and Boyde (1977) observed loss of remaining predentine, and the presence of areas of localised intact calcospherites existing alongside larger tracts of prepared root canal wall. Lester and Boyde (1977) could offer "no overwhelming argument" for the use of NaOCl as an irrigant, but felt that features such as antibacterial and debriding effects justified its continued use.

Koskinen, Meurman and Stenvall (1980) observed that application of 2.5% and 5% NaOCl "dissolved most of the predentine, exposing the globular appearance of the mineralising front". The authors immersed young split premolars, with pulps stripped away, in 0.5%, 2.5% or 5% NaOCl for ten minutes. It was also noted that the effects of NaOCl were most evident with 5% NaOCl and least

apparent apically and when using 0.5% NaOCl.

Baumgartner and Mader (1987) instrumented one half of individual intact root canals while using a variety of irrigants. When 5.25% NaOCl was used the surfaces of the uninstrumented halves were described as "composed entirely of fields of exposed calcospherites, with no evidence of pulpal remnants or predentine". In contrast, saline irrigation resulted in uninstrumented halves showing "some residual superficial pulpal fibres, the collagen fibrils of the predentine, and the orifices of the dentinal tubules". Following use of NaOCl or saline on instrumented halves of the canal, there was little evidence of superficial debris.

Berg et al. (1986) found that following the use of saline "clearly evident" debris was present, while 5.25% NaOCl resulted in "minimal debris". The large apical size (size 70) could explain the lack of difference in the amount of debris detected in the apical third.

CHAPTER 6

CHELATING IRRIGANTS

6.1 INTRODUCTION

Chelation has been defined as "the ability of certain chemicals to donate atoms - usually oxygen, nitrogen or sulphur - which will form co-ordinate bonds with polyvalent metallic ions : the result is a stable, water-soluble, non-ionic metallic ring in which the metal is firmly held" (British Medical Journal, 1953).

A chelating agent is a chemical which unites with metallic ions to form chelates (Gurney, 1967). Examples of chelating agents include ethylene diamine tetraacetic acid (EDTA), lactic acid and citric acid (British Medical Journal, 1953). Demineralisation of dentine occurs by the action of the chelating agent seeking the calcium metal ion in hydroxyapatite crystals (Nikiforuk and Sreebny, 1953).

In endodontics, chelating agents are employed to assist in negotiation and enlargement of severely constricted or obstructed root canals. The use of EDTA to replace inorganic acids during endodontic therapy was first suggested by Östby in 1957, who believed it could assist in overcoming the "insurmountable difficulties" encountered in narrow or obstructed canals, as well as save time during instrumentation. Other authors

recommended EDTA in order to soften pulp stones (Chesney, 1968), and help prevent instrument breakage when negotiating difficult narrow canals (Grossman, 1969).

Lactic and citric acid have also been investigated for use as chelating irrigants. It has been suggested that since they occur "naturally"¹, these solutions may be more acceptable biologically than other irrigants (Loel, 1975; Wayman et al., 1979).

Another solution which has been suggested as a chelating agent in endodontics is aminoquinaldinium diacetate, known commercially as Salvizol (Ravensburg GMBH, Konstanz, W Germany) or Solvidont (DeTrey, Dentsply, Zurich, Switzerland), an irrigant first described by Kaufman et al. (1977). This irrigant will be discussed in Chapter 7.

Interpretation of many SEM studies on chelating irrigants relies upon examination of the smear layer which has been discussed in Chapter 4. Research on cytotoxicity of chelating agents will be included in Chapter 9. This chapter will address other research on chelating agents.

6.2 EDTA

In 1970, Penick and Osetek speculated that EDTA was the chelating agent most commonly utilised during

1. Lactic acid occurs during vigorous muscular activity under anaerobic conditions (Stryer, 1981 p352), while citric acid is a component of the citric acid cycle (Stryer, 1981 p283).

endodontic treatment.

6.2.1 Chemistry, concentration and pH

EDTA ($C_{10}H_{16}N_2O_8$) is an odourless, insoluble, crystalline, white powder with a molecular weight of 372.10 (Obst, 1970). It contains four acetic groups attached to ethylene diamine (Seidberg and Schilder, 1974). Various sodium salts of the acid are soluble in water (British Medical Journal, 1953). The sodium salts are noncolloidal agents capable of forming soluble non-ionic chelates with a large number of metallic ions such as calcium (Nikiforuk and Sreebny, 1953).

Preliminary research on EDTA was performed by Nikiforuk and Sreebny (1951; 1953), who investigated the use of EDTA to decalcify hard tissue undergoing histologic and histo-chemical examination. It was considered that there were advantages in using EDTA over other demineralising solutions (such as nitric acid) because it acted at a neutral or alkaline pH, and thus avoided subjecting tissue specimens to the protein-denaturing effect of a low pH. From their research on the demineralising effect of EDTA on rat mandibular bone, Nikiforuk and Sreebny (1953) showed that the optimal pH range for demineralisation was between 7 and 8. Below this, the chelating effect of EDTA dropped markedly. The authors therefore concluded that, when large amounts of calcium were to be chelated, EDTA solutions needed to be buffered within that range.

Cury, Bragotto and Valdrighi (1981) examined the demineralising effects of EDTA (0.3M) on powdered dentine using EDTA solutions of varying pH. From measurements of the amount of phosphorus liberated, it was shown that a pH range of between 5 and 6 provided the most efficient demineralisation of powdered dentine.

Other studies which examined the effect of EDTA on bone (Hunter and Nikiforuk, 1954) and powdered dentine (Ando, 1985), found that shorter decalcification times were required with a stronger concentration of EDTA. In addition, it was shown that elevating the temperature of EDTA produced faster demineralisation of bone specimens (Hunter and Nikiforuk, 1954).

6.2.2 Role in chemomechanical preparation

Östby (1957) recommended a 15% concentration of EDTA for endodontic use. His formula (known as "Östby's formula") which produced a solution of pH 7.3 was:-

disodium salt of EDTA	17 gm
distilled water	100 ml
5N sodium hydroxide	9.25 ml

In 1957, Östby claimed that EDTA was "neither bactericidal nor bacteriostatic, but it inhibits the growth of, and will eventually destroy, bacteria by the process of starvation." Further, because metallic ions necessary for bacterial growth were chelated by EDTA, solutions containing EDTA would therefore be "self

sterilizing."

Cetrimide¹, a quaternary ammonium bromide was later added to EDTA in an effort to render the solution bactericidal, as well as lower the surface tension (Hill, 1959; Östby, 1961; von der Fehr and Östby, 1963; Goldberg and Abramovich, 1977; Ando, 1985a; Ciucchi, Khettabi and Holz, 1989). This addition resulted in the products known as EDTAC (Laboratories Farmadental, Buenos Aires, Argentina)² and REDTA (see p54). Hill (1959) found EDTAC to be "most satisfactory" in softening canal walls within three to five minutes, while Weinreb and Meier (1965) commented that the addition of cetrimide "does not seem to be of much value". Ando (1985) found that the chelating effect of EDTA was not reduced by the addition of the quaternary ammonium compound.

The clinical efficacy of EDTA was questioned by some authors (Wandelt, 1961; 1965; Fromme, Guttzeit and Riedel, 1970; Dow, 1984), who doubted that a sufficient volume of solution could be placed into fine canals. Wandelt (1965) considered that canals unable to be hand instrumented would also be incapable of dilation with the use of chelating solutions. He concluded that EDTA was not suitable as an adjuvant in root canal treatment. In

1. Cetrimide (cetyl trimethylene ammonium bromide) is frequently referred to as Cetavlon (Imperial Chemical Industries Limited, England) (Foulkes, 1973).

2. EDTAC is a 15% EDTA solution which is formed by the addition of 0.84 gm cetrimide to Östby's formula (von der Fehr and Östby, 1963; Golberg and Abramovich, 1977).

1984, Dow asserted that "a fine, sharp flexible file in experienced hands may have more effect in negotiating fine tortuous canals than relying on an indeterminate chemical reaction".

Glantz and Hansson (1972) showed that EDTA had a poor wetting capacity, and formed a lens on dentine. Abou-Rass and Patonai (1982) found that the addition of polysorbate 80 to EDTA reduced surface tension by between 15 and 20%. EDTA concentrations were not identified in either study.

Under x40 magnification, Gutiérrez and Garcia (1968) observed "a heavy precipitation of salts" on prepared canal walls following use of EDTA, which was attributed to oversaturation of the irrigant solution.

Obst (1970) recommended storage of EDTA in a plastic container, and its clinical use in a plastic syringe. Contact with glass was discouraged to avoid exhausting the chelating power of EDTA (Penick and Osetek, 1970). Barnett, Godick and Tronstad (1985) found that the cleaning effect of 15% EDTA was "good" when used with the sonic Endostar 5 (Syntex Dental Products, Valley Forge, PA). However, technical problems associated with its use included crystallisation of EDTA in the holding tank, tubing and handpiece as well as corrosion of the handpiece.

6.2.3 Limitation of action

Östby (1957) stated that EDTA would "demineralise a

certain, distinctly limited, zone of the dentine surrounding the root canal when used during reaming or deposited in the canal". These comments were endorsed by von der Fehr and Östby (1963) who found that application of EDTAC to dentine for five minutes produced a demineralised layer 20 microns-30 microns thick, while exposures of up to 48 hours resulted in complete removal of mineral salts to a depth of 50 microns. The demineralising effect was rapid, but this was soon "exhausted", depending upon the quantity of reagent present.

However, several authors questioned Östby's statement (Patterson, 1963; Nicholson et al., 1968; Biesterfeld and Taintor, 1980). Patterson (1963) reported that the action of EDTA was not immediately self-limiting, but could remain active in the canal for as long as five days, until all available EDTA formed a complex with the calcium salts of dentine. A five-day waiting period prior to final instrumentation and obturation of the root canal was therefore advocated.

In contrast, Seidberg and Schilder (1974) examined the effects of EDTA on powdered dentine over a nine hour period, and found it to be self-limiting within that period. The rate of reaction was most rapid within the first hour of application, with equilibrium being approached by seven hours. The rate of reaction was directly related to the surface area exposed.

Despite the self-limiting action of EDTA, caution

was required by the clinician to avoid chelating a perforation in sharply curved canals (Orlay, 1962; Seidberg and Schilder, 1974).

There is little information in the literature concerning the minimum required application time for EDTA. Weinreb and Meier (1965) recommended renewal of EDTA every three minutes. Goldberg and Abramovich (1977) observed the effects of a 30 minute application of EDTA under SEM. In a later study, Goldberg and Spielberg (1982) found that EDTA needed to be in contact with dentine for 15 minutes for maximum effect, although some effects were observable after five minutes. The authors suggested the optimum working time of EDTAC to be 15 minutes, after which the solution needed renewal.

For clinical use, chemicals which have been proposed to limit the action of EDTA have included calcium hypochlorite (Gurney, 1967), 10% calcium chloride solution (Gutiérrez et al., 1982), NaOCl (Weine, 1982 p323) and, with "careful" use, 10% hydrogen peroxide (Patterson, 1963).

6.2.4 Effect on dentine

Patterson (1963) reported that 10% EDTA markedly lowered the microhardness of dentine after a 24 hour exposure, while Fraser (1974) observed cervical and midroot, but not apical, dentine softening following a 15

minute application of Largal Ultra¹.

Two early radioautogram studies on the effect of EDTA on dentine permeability, which are frequently referenced in the literature, were presented by Marshall, Massler and Dute (1960) and Hampson and Atkinson (1964). However, questions arise concerning the methodologies employed. Both studies used uninstrumented controls and instrumented test groups, thereby introducing two significant variables - a larger surface area in the test group and a different type of surface on which the solutions could act.

Marshall, Massler and Dute (1960) found that assessment of the effect of EDTA on dentine permeability was dependent on the radioisotope used. In this study, root canals were treated with various chemicals, then one of four radioisotopes placed in each canal, radioautograms taken, and dentine permeability assessed. Although EDTA reduced the permeability of dentine to one isotope, ³⁵S, "this decrease was not significant."

Hampson and Atkinson (1964) used radioopaque dyes and radioautograms to compare canals after a five minute exposure to various chemicals, including EDTA. Their findings indicated that of the solutions tested, an organic solvent (5% chloramine-T - see p100) increased dentine permeability most. A slight increase in isotope

1. Largal Ultra (Septodent, Paris, France), pH 7.4, contains 15% EDTA and 0.75% cetrimide (Koskinen, Meurman and Stenvall, 1980).

penetration was observed after placing EDTA in mechanically enlarged canals for five minutes.

Heling, Shapiro and Sciaky (1965) measured the amount of calcium in used irrigants following mechanical preparation of extracted teeth. No significant difference was found between the amount of calcium removed by an inorganic acid, 20% hydrochloric acid and 15% EDTA, thus indicating that "EDTA is at least as effective as 20% hydrochloric acid in widening root canals".

Another study found EDTAC to be significantly more effective in mineral removal than 50% sulphuric acid (Weinreb and Meier, 1965). However, details of data collection were not clear.

6.2.5 Effect on apical seal

Goldberg et al. (1985) attempted to assess the effects of either EDTAC or distilled water on the apical seal of obturated root canals by performing bacterial penetration and viability analyses, as well as SEM examination. Although the study appeared to be fraught with technical difficulties, the authors felt their results showed that there was no significant difference between the two groups, thus indicating that the use of EDTAC does not interfere with the quality of the apical seal.

An evaluation of the effect of irrigation solutions on the filling of lateral and accessory canals was made in a later study by Goldberg, Massone and Spielberg

(1986). After obturation of 66 recently extracted single-rooted human teeth which had been instrumented using:- (1) 15% EDTA; (2) 5% NaOCl; or (3) distilled water, the number of filled lateral canals was counted from post-obturation radiographs. It was asserted that canals prepared with EDTA showed a "larger number of filled lateral root canals" than the other two groups. The authors concluded that use of EDTA as an irrigant "increases the likelihood of filled lateral and accessory canals".

In two other studies obturated teeth were immersed in methylene blue dye following mechanical preparation using different irrigating regimens. Madison and Krell (1984) found no significant difference in apical penetration of dye following the use of either:- (1) 2.5% NaOCl and REDTA; or (2) 2.5% NaOCl alone. Cergneux et al. (1987) showed that less apical infiltration of dye occurred following the use of 15% EDTA than after 3% NaOCl with ultrasound.

6.2.6 EDTA as a final flush

Several *in vitro* SEM studies have qualitatively assessed the efficacy of irrigants used as a high volume final flush following completion of instrumentation (Goldman et al., 1981; 1982; Yamada et al., 1983; White, Goldman and Sun Lin, 1984; 1987).

Examination under SEM by Goldman et al. (1981) showed that a final flush with REDTA removed the smear

layer, but did not remove pulp tissue and predentine from uninstrumented areas. In the same study, split root specimens soaked in REDTA for 3 hours showed no appreciable difference from those flushed only.

A later SEM study examined the efficacy of various combinations of REDTA and 5.25% NaOCl for the removal of debris during and after instrumentation (Goldman et al., 1982). While 5.25% NaOCl was found to be the most effective irrigant during instrumentation, 10 ml REDTA followed by 10 ml 5.25% NaOCl was found to be the most effective final flush regimen. However, as there was no uniformity of canal preparation during this study, the results might have been misleading.

Yamada et al. (1983) similarly examined various solutions and combinations of solutions. Their results indicated that a final flush of 10 ml 17% EDTA followed by 10 ml 5.25% NaOCl obtained the best removal of both organic and inorganic debris. 17% EDTA removed the smear layer but consistently left superficial debris. 8.5% EDTA was "almost equally effective" as 17% EDTA. The authors concluded that a high volume final flush required the combination of a chelating agent and a tissue solvent.

Scanning electron microscopy was used to examine the penetration of the plastic filling materials, Hydron (National Patent Development, New York, NY, USA), and Endo-fill (Lee Pharmaceuticals, South El Monte, CA, USA) following a final flush of either:- (1) 20 ml 5.25% NaOCl; or (2) 10 ml 17% EDTA followed by 10 ml 5.25%

NaOCl. Penetration of filling material was superior following EDTA use (White, Goldman and Sun Lin, 1984). In a following paper, (1987) the authors also included laterally condensed gutta percha sealed with either zinc oxide/eugenol (Roth 801; Roth Drug Co., Chicago, IL, USA) or epoxy resin (AH26; DeTrey, Zurich, Switzerland). All four materials were able to penetrate dentinal tubules after a final flush of 10 ml 17% EDTA followed by 10 ml 5.25% NaOCl.

6.2.7 RC-Prep¹

The combination of 15% EDTA and 10% urea peroxide was introduced as a chelating agent in endodontics by Stewart, Kapsimalas and Rappaport in 1969. They believed that after using this preparation there was a greater potential for penetration of intracanal antibacterial medicaments into dentinal tubules. These authors found that greater radioisotope dye penetration into dentine occurred following instrumentation with RC-Prep than without RC-Prep. However, a different irrigant was used in the experimental roots (NaOCl) than that used in the control roots (distilled water).

Cohen, Stewart and Laster (1970) examined the

1. The commercial preparation, RC-Prep (Premier Dental Products, Philadelphia, USA), was described by Cohen, Stewart and Laster (1970) as a mixture of 15% EDTA and 10% urea peroxide in a water-soluble base of Carbowax. Carbowax is a stable base of polyethylene glycol (Fraser and Laws, 1976). Its addition to RC-Prep was necessary to prevent oxidation of EDTA by urea peroxide (Stewart, Kapsimalas and Rappaport, 1969).

effects of a number of irrigating solutions on dentine permeability. These solutions included organic solvents, inorganic acids and EDTAC. Permeability was assessed by measuring the depth of methylene blue dye penetration into dentine following immersion of the root in the dye for 30 minutes. Results indicated that the alternate use of RC-Prep and 5% NaOCl produced increased dentine tubule permeability in the apical and middle thirds of the root.

Applying a 60 gm load with a Tukon hardness tester, Fraser (1974) observed cervical and midroot, but not apical, dentine softening following a 15 minute application of RC-Prep. In a later article, Fraser and Laws (1976) reported that RC-Prep reduced the diffusion of phloxine dye into the root canal. The decreased dye penetration was attributed to possible occlusion of the tubules by "CaEDTA sludge" and to the large dye molecule of phloxine. While these results contradicted those of Stewart, Kapsimalas and Rappaport (1969) and Cohen, Stewart and Laster (1970), it must be noted that all three investigations used different methodologies.

Zurbriggen, del Rio and Brady (1975) biomechanically prepared 40 freshly extracted single rooted teeth using RC-Prep radioisotopically labelled with carbon-14 and 2.5% NaOCl. Subsequent collection of carbon-14 showed that, on average, 3.8% of the original quantity of RC-Prep applied remained in the canals. These results were not affected by increasing instrumentation time and volume of irrigation.

A further study which employed RC-Prep during mechanical preparation, measured radioactive iodine uptake to observe periapical leakage after canal obturation (Cooke, Grower and del Rio, 1976). It was found that significantly increased periapical leakage was associated with teeth which had employed RC-Prep versus controls which had not. The authors suggested that "some component in the EDTA-urea peroxide-Carbowax mixture may remain in the canal and adversely affect the seal of the root canal filling".

It has been suggested by Biesterfeld and Taintor (1980) that studies using different radioisotopes cannot be directly compared, as each isotope will show different degrees of leakage depending on the type of material in contact with the tooth. Biesterfeld and Taintor (1980) asserted that a poorer apical seal was obtained after using RC-Prep with obturation one week later rather than immediately, leading them to question the "self-limiting quality of RC-Prep".

Gutiérrez et al. (1982) investigated the ability of selected bacteria to penetrate dentine following root canal preparation which had utilised one of the following proprietary chelating agents:- (1) Östby's formulation; (2) Largal Ultra ; or (3) RC-Prep (pH 2.70), all containing 15% EDTA. After preparation, during which the chelating agents remained in contact with radicular dentine for up to 10 minutes, the entire tooth was soaked in trypticase soy broth previously seeded with cultures

of various microorganisms and then examined under SEM. The authors concluded that, of the three chelating agents, RC-Prep produced dentine tubules that were the most dilated and permeable to microorganisms. Bacterial penetration into dentine was least when Östby's formulation was used. This study prompted a comment from Stewart (1983), who suggested that there were no normal clinical techniques which would deliberately inoculate or contaminate prepared root canals. In response, Gutiérrez (1983) defended his paper by citing leaky temporary restorations as a common cause of root canal contamination after preparation.

In several SEM studies disadvantages, or at least, no advantages, have been reported using RC-Prep (McComb and Smith, 1975; Rubin et al., 1979; Bolanos and Jensen, 1980; Blackler, 1981; Cameron, 1984). McComb and Smith (1975), in examining the effects of various irrigants on root canal walls, found RC-Prep in combination with 6% NaOCl resulted in a smeared surface with more superficial debris than that resulting from instrumentation with water alone. Blackler (1981) observed that RC-Prep used in combination with 1% NaOCl produced "extensively smeared canal walls". Using different combinations of saline and 1% NaOCl, with or without RC-Prep, no significant differences in canal cleanliness were found in another SEM study (Bolanos and Jensen, 1980). Rubin et al. (1979) found the use of RC-Prep had no effect on either pulp tissue or dentine after a 30 minute

application. Cameron (1984) found that the debridging action of 3% NaOCl and ultrasound was not enhanced by RC-Prep.

6.3 CITRIC ACID

Using SEM, Brännström and Johnson (1974) showed that demineralising agents such as citric and lactic acid opened and widened dentinal tubule apertures, resulting in a "clean surface" when applied to coronal dentine.

Loel (1975) first suggested citric acid as a root canal cleanser. It was claimed that 50% citric acid (EpoxyLite 9060 Cavity Cleanser, Lee Pharmaceuticals, California, USA) combined with NaOCl produced a foaming reaction "that assists in the removal of debris within the canal".

As part of an *in vitro* investigation, Loel (1975) applied 50% citric acid to dentine which was then subjected to malachite green dye penetration tests. Examination under SEM and light microscopy showed deeper penetration of dye into treated dentine than into untreated dentine (Loel, 1975).

Loel (1975) also compared histologically the solvent effects of 50% citric acid and 5% NaOCl on pulps of extracted teeth, observing greater destruction by citric acid. However, as the teeth had been extracted for unspecified and perhaps variable lengths of time, these observations have limited usefulness. As well, the author reported a "bland response" to 5% NaOCl, a comment which

has not been endorsed by other studies.

Another SEM study showed that root canals flooded with 50% citric acid during mechanical preparation were "generally free of smeared debris" (Tidmarsh, 1978). However, to avoid residual crystals (probably calcium citrate) remaining on the canal wall, "copious irrigation" was required after using citric acid, although no particular irrigant was suggested.

Wayman et al. (1979) compared the efficacy of various irrigants, including 50% lactic acid and three concentrations of citric acid (10%, 25% and 50%). While citric and lactic acid were found to dissolve more calcium than 5.25% NaOCl or saline, NaOCl provided greater organic tissue dissolution. Under SEM lactic acid was shown to leave a "film" on the dentine surface. The authors recommended the use of 10% citric acid as part of an irrigating regimen which also needed to include NaOCl.

Baumgartner et al. (1984) found that 50% citric acid was effective in removal of the smear layer. Combinations of 5.25% NaOCl and 50% citric acid, irrespective of order, were capable of removing both superficial debris and smear layer. A final flush with distilled water was recommended to avoid precipitation of crystals.

It would appear that while citric acid is a satisfactory chelating agent, it provides no advantages over EDTA.

CHAPTER 7

OTHER IRRIGANTS

7.1 INTRODUCTION

Over the years a variety of solutions have been proposed for use as endodontic irrigants. Many of these agents, recommended on the strength of anecdotes and personal clinical experience, have not been subjected to comprehensive inquiry. Other solutions, whilst being examined more thoroughly, have been investigated by only a few researchers and require further independent assessment before routine use can be endorsed.

While many of the irrigants to be discussed might possess distinct beneficial properties, none have proved capable of fulfilling the requirements of the "ideal irrigant". It will become apparent in this loosely chronological review that none of these irrigants provide significant advantages over NaOCl or EDTA.

7.2 HOT DISINFECTANTS

Irrigation with hot (60°C-70°C) disinfectant solutions of mercury phenylborate, invert soap, organic mercury or monomolecular mercury were recommended by Prader (1948). The author claimed that the higher the temperature of the solution, the shorter would be the

time required to obtain sterility.

7.3 ENZYMES

The rationale behind the use of enzymes in solution as irrigants was to enhance the natural defensive response of the host. Examples were Tryptar (The Armour Laboratories, Chicago, USA) and Varidase (Corderle Laboratories Division, American Cyanamid Company, NY, USA).

Based on eight cases, Auslander and Roth (1953) recommended Tryptar as an irrigant and medicament, and a safe and effective necrotic tissue solvent. The authors described Tryptar as a "highly purified crystalline trypsin derived from mammalian pancreas". Indication for use included "selective physiologic debridement of necrotic tissue". Although one patient experienced an allergic reaction following application, the authors felt that this could be obviated by prior administration of an antihistamine agent.

Varidase, a combination of streptokinase and streptodornase, both extracellular enzymes elaborated by β -haemolytic *streptococci*, was endorsed as an irrigant and medicament by Blechman (1953). He asserted that streptokinase lysed clotted blood, while streptodornase catalysed the breakdown of deoxyribonucleoprotein, which was speculated to constitute 30%-70% of the sediment of thick purulent exudates. Golden and Musgrave (1954) claimed good clinical results with Varidase, which they

asserted would enhance and complement the natural defence mechanisms of the surrounding tissues. No allergic reactions occurred during 13 cases.

7.4 UREA

A 30% urea solution was recommended as an "excellent root canal wash, following vital pulp removal with haemorrhage" (Blechman and Cohen, 1951). It was considered to be non-toxic and non-irritating to the periapical tissues. Further, it was claimed that 30% urea removed clotted blood from the canal walls, in addition to deodorising putrescent suppurating canals (Blechman, 1953).

Penick and Osetek (1970) described a 30% saturated solution of urea as a non-toxic, mild solvent of necrotic tissue and pus, with mild antiseptic properties, which was not as effective as 5% NaOCl.

7.5 UREA PEROXIDE

The crystalline powder urea peroxide is composed of urea and hydrogen peroxide. It is unstable in aqueous form but becomes stable when combined with an anhydrous glycerol vehicle, which then allows dissociation into urea and hydrogen peroxide at a slower rate (Penick and Osetek, 1970).

In an effort to find an antiseptic compound more stable than 3% hydrogen peroxide, Stewart, Cobe and Rappaport (1961) used the presence of growth-free

cultures to assess the germicidal properties of Gly-Oxide (International Pharmaceuticals Corporation, Philadelphia) compared to aqueous hydrogen peroxide. The authors believed that Gly-Oxide, a solution of 10% urea peroxide in a vehicle of anhydrous glycerol, was a more stable antiseptic compound than hydrogen peroxide, and also had lubricant properties.

A previous study by one of the authors (Cobe, 1960) had attempted to assess the action of 3% Gly-Oxide compared to 3% hydrogen peroxide, by measuring the release time and height of foam formed when these agents were exposed to citrated human blood. Cobe reported that evolution of bubbles was immediate with 3% hydrogen peroxide, whereas Gly-Oxide produced more "compact" foam which developed gradually and lasted longer. Cobe suggested that the longer the period of oxygen release, the greater the chance for bactericidal action to occur.

Stewart, Cobe and Rappaport (1961) suggested the combined use of Gly-Oxide and 5% NaOCl to increase effervescence and bactericidal effect. However, Cohen, Stewart and Laster (1970) found this combination increased dentine permeability to methylene blue dye. Brown and Doran (1975) found that the combination of Gly-Oxide and 5% NaOCl was able to reduce the height of dentine particles in a simulated canal when the needle tip was positioned 2 mm from the dentine debris.

Weine (1982 p319) claimed that because of its low toxicity, Gly-Oxide was an "excellent irrigant" for teeth

with wide apices, where a risk existed of escape of solution into the periapical area. Its lubricant properties were also considered effective in narrow or curved canals.

An *in vitro* study by Foley et al. (1983) tested the effectiveness of Gly-Oxide in the elimination of *Bacteroides melaninogenicus* from the root canal system. Gly-Oxide was effective, but not when diluted.

Using SEM, Rome, Doran and Walker (1985) observed that Gly-Oxide in conjunction with NaOCl did not remove the smear layer. Further, there was no difference in smear layer formation using NaOCl alone or in combination with Gly-Oxide.

7.6 LOCAL ANAESTHETIC

Although there appears to be no published research on the efficacy of local anaesthetic as an irrigant, there has been reference to it as a bland, sterile flushing agent (Martin, 1979; Abou-Rass and Piccinino, 1982). Topical anaesthetic has been suggested as a lubricant to aid mechanical preparation (Walton and Torabinejad, 1989 p208).

Mayne (1959) recommended sterile isotonic solution, such as local anaesthetic warmed in hot water, as an irrigant during the removal of vital pulps. It was speculated that a non-isotonic solution could cause the rupture of red blood cells, thereby allowing the breakdown products of haemoglobin to enter the dentinal

tubules, and result in subsequent tooth discolouration.

Local anaesthetic should not be used as an irrigant with the Neosono (Amadent, Inc., Cherry Hill, NJ, USA) apex locator as, according to the manufacturer, it might interfere with measurement readings (Gutmann and Dumsha, 1987 p167).

7.7 CHLORAMINE-T AND AZOCHLORAMID

The organic chloramines are produced by the reaction of HOCl with an amine, amide or imide (Harvey, 1980 p974; Dychdala, 1983 p173). They are not stable in water and release chlorine slowly. Chloramines have been in existence for many years as general skin and wound disinfectants (Austin and Taylor, 1918; Taylor and Austin, 1918). Because chloramines contain a variable N-chloro substitute, there are several chloramines available, one of which is chloramine-T. However, the dental literature frequently refers to "chloramine-T" and "chloramine" interchangeably.

Chloramine-T (sodium p-toluene sulfonchloramide) is a white crystalline powder which contains approximately 25% available chlorine (Dychdala, 1983 p173). It is formed by the combination of sodium 4-toluene sulfonamide and HOCl (Deuben, 1980 pp660-1).

Although, like hypochlorites, chloramine-T hydrolyses to form HOCl, Penick and Osetek (1970) considered it to have a considerably slower bactericidal action than the hypochlorites. Nicholls (1962) found no

difference in bactericidal effectiveness between chloramine-T and hydrogen peroxide/NaOCl.

Luebke (1967) claimed that 4% chloramine-T was a more stable and less irritating, but less effective, solution than NaOCl. However, another writer believed that chloramine-T was more germicidal, more stable and less irritating than NaOCl but offered no supporting evidence (Hampson, 1964).

Attala and Calvert (1969) provided a formula for preparation of chloramine-T irrigant:-

chloramine	134.4 gm
sodium chloride	26 gm
water	3,300 ml

These authors found that chloramine-T produced a mild inflammatory response when applied to dog's eyes and injected into guinea pig subcutaneous tissue. In contrast, Wennberg (1980) found 5% chloramine-T produced severe tissue reactions in thigh muscle of rabbits.

Martin (1979) claimed that 3%-5% chloramine-T possessed antiseptic activity and lower toxicity than NaOCl, but a poorer solvent activity. Spängberg (1985 p570) believed chloramine-T possessed "excellent antimicrobial qualities", and a long shelf life if stored in a cold place and away from light. Ingle et al. (1985 p183) commented that its solvent ability was poor.

Reichardt (1973) advocated the use of 1%-2% chloramine-T heated to 60°C when irrigating lesions of

periodontal and endodontic origins.

The combination of 10% chloramine-T with EDTA produced precipitation of toluene-sulfamide, which were colourless crystals and "probably innocuous" (Östby, 1957).

Chloroazodin, or Azochloramid, is a chlorine compound (N,N' -dichloroazodicarbonamidine) which is slightly soluble in water (Dychdala, 1983 p176). It is light sensitive and can exert a mild but sustained germicidal activity, however, it is less reactive with organic matter than other chlorine compounds such as chloramine-T and NaOCl. Contact with metals accelerates its decomposition.

Ursini (1947) described Azochloramid as a compound which slowly liberated chlorine and suggested it as a dressing or irrigant. Azochloramid was later shown to exhibit a very high irritant potential (Schilder and Amsterdam, 1959).

7.8 HYDROGEN PEROXIDE

Hydrogen peroxide is an oxidising agent which is rapidly decomposed to molecular oxygen and water by tissue catalase (Penick and Osetek, 1970; Molinari, 1988 p79). Dakin (1915) regarded hydrogen peroxide as an antiseptic of slight value - "the mechanical detergent action connected with the rapid disengagement of oxygen gas on infected surfaces is probably of greater value than any antiseptic action exerted by the hydrogen

peroxide".

Low concentrations of hydrogen peroxide will inhibit bacteria in culture (Molinari, 1988 p79). Weine (1982 p318) suggested that the liberation of oxygen from hydrogen peroxide in contact with tissue would destroy strict anaerobes.

During endodontic procedures, hydrogen peroxide has generally been used in a 3% concentration and in combination with NaOCl (Luebke, 1967). Alone, it does not have the ability to dissolve necrotic tissue or organic debris (Penick and Osetek, 1970; Hand, Smith and Harrison, 1978). Penick and Osetek (1970) maintained that the activity of hydrogen peroxide was brief and ineffective.

Cartin (1955) endorsed the use of hydrogen peroxide, and suggested that its foaming action would force debris out of the canal. However, another author believed the use of hydrogen peroxide was dangerous, because of the possibility of oxygen release into the periapical tissues (Masterton, 1965). Weine (1982 p319) endorsed hydrogen peroxide as the irrigant of choice for perforations or in cases of severe pericementitis surrounding an apical constriction.

Three percent hydrogen peroxide produced severe and prolonged reactions when applied to dog's eyes, while injections into guinea pig subcutaneous tissue resulted in mild inflammation (Attalla and Calvert, 1969).

7.9 HYDROGEN PEROXIDE/SODIUM HYPOCHLORITE

Grossman first suggested the use of double strength chlorinated soda (see p12) in combination with 3% hydrogen peroxide in 1943. It was considered that the effervescence produced by combining the two solutions would liberate nascent oxygen, which could then follow the line of least resistance towards the opening of the canal. Grossman advised the use of double strength chlorinated soda (a reducing solution) as the final irrigant in order to avoid the build-up of gas pressure from any remaining hydrogen peroxide (an oxidising solution). The combination has since been endorsed by other authors (Stewart, 1955; Morris, 1958; Mayne, 1959; Curson, 1966; Weine, 1982 p191, p318). Sommer, Ostrander and Crowley (1966 p548) believed it to be "helpful" in large canals.

Stewart (1955) recommended an irrigation technique which used 0.5 ml 3% hydrogen peroxide, followed by 0.5 ml NaOCl after each instrument change, claiming his results showed that 94% of canals were rendered sterile after this procedure.

Repeated alternate irrigation with hot NaOCl and room temperature hydrogen peroxide, finishing with NaOCl, was recommended by Morris (1958). The use of 6% hydrogen peroxide and double strength chlorinated soda was recommended during treatment of teeth with non-vital pulps (Mayne, 1959).

Marshall, Massler and Dute (1960) found increased

permeability of radicular dentine to ^{35}S , after treatment of root canals with 3% hydrogen peroxide and 5.25% NaOCl. The combination was more effective than either solution used alone.

Using a light microscope, Svec and Harrison (1977) observed that the combination of 5.25% NaOCl and 3% hydrogen peroxide was more effective than saline in debridement of the apical 3 mm of the canal. At the 5 mm level, there was no significant difference between the control and experimental groups. Later research compared the combination with NaOCl alone (Svec and Harrison, 1981). No significant difference was found between the two groups at the apical 3 mm of the canal. The authors concluded that the effervescence which occurs when NaOCl is combined with hydrogen peroxide "may not offer a decided advantage in terms of increased debridement of the apical portions of root canals in single-rooted teeth". Further, Harrison and Hand (1981) found that the combination of 3% hydrogen peroxide and 5.25% NaOCl had no antibacterial effect *in vitro* against *Streptococcus faecalis* while 5.25% NaOCl alone was effective in 45 seconds.

Other studies have observed that the combined use of hydrogen peroxide and NaOCl does not provide superior irrigating properties (Brown and Doran, 1975; Thé, 1979; Rubin et al., 1979; Blackler, 1981; Abou-Rass and Piccinino, 1982).

A study by Thé (1979) found that the combination of

2% or 5% NaOCl and 3% hydrogen peroxide was less effective than NaOCl alone for dissolving fixed or unfixed necrotic tissue. Further, the solvent action of NaOCl was likely to be diminished when hydrogen peroxide was added to the solution. Thé commented that no proof existed for the assumption that "the release of nascent oxygen of hydrogen peroxide can bubble out debris from the root canal".

Brown and Doran (1975) observed dentine particles in simulated canals were "agitated" when the combination was used within 2 mm of the debris, but this was followed by settling of the particles. Baumgartner and Ibay (1987) found that the chemical combination of hydrogen peroxide and NaOCl produced no detectable chlorine gas.

It would appear that there is no benefit in the alternate use of NaOCl and hydrogen peroxide.

7.10 AMINOACRIDINE

Acridine is the nitrogen compound benzquinolone which is structurally related to the antimalarial quinolines (Rossmore, 1983 p300). Quaternarization of acridine increases bacteriostatic activity, as the most highly ionized cationic compounds are the most active. At 37° C and pH 7.3, 9-aminoacridine is in 99% cationic form. The early literature (Lyell, 1952; Jolly and Sullivan, 1956) refers to 5-aminoacridine, which was commonly used as a mucous membrane antiseptic (Jurecko, 1974).

A 1:1000 solution of Monacrin (5-amino acridine hydrochloride), described as being active in the presence of serum proteins and pus, was recommended as an irrigant by Lyell (1952). As a result of the inability of this agent to act effectively on *Candida albicans* and *Pseudomonas pyocyanea*, Jolly and Sullivan (1956) added other agents to form the following mixture:-

Monacrin (5-aminoacridine hydrochloride - Bayer) 1:500
 Cetavlon (alkyl trimethyl-ammonium bromide - ICI) 1:500
 Methyl NIPA ester (methyl parahydroxybenzoate) 1:500
 Propyl NIPA ester (propyl parahydroxybenzoate) 1:3000
 Titrated to pH 6.0 with N/10 NaOH.

This combination formed a solution which was used routinely in 1956, both as an irrigant and an intracanal medicament by staff and students at the University of Sydney Dental School. To increase the bactericidal effect of the mixture, the addition of 1:500 chlorhexidine digluconate was suggested by Atkinson and Hampson (1964).

Jurecko (1974) examined the effectiveness of 9-aminoacridine alone, and in combination with benzalkonium chloride (a quaternary ammonium compound - see p113). It was found that 9-aminoacridine had antibacterial activity against *Staphylococcus aureus*, *Staphylococcus epidermis*, *Streptococcus faecalis* and *Candida albicans*, but not against *Streptococcus mitis*. Application of the solution to rabbit eyes showed a mild response to 9-aminoacridine when compared with eugenol and parachlorophenol. Jurecko

believed that the antibacterial action and apparent low toxicity of 9-aminoacridine could provide the appropriate properties required for an irrigant, and its routine use for lavage prior to obturation was also advocated. The addition of benzalkonium chloride was thought to provide greater antibacterial action against *staphylococci* and *Candida albicans* and, potentially, slightly increased eye tissue irritation.

Pressure irrigation of periapical tissues using 9-aminoacridine via the root canal, was suggested by Breese (1950).

Schmitz (1980) recommended the routine use of 9-aminoacridine as an endodontic irrigant. He believed it resisted inactivation by pus, secretions and body fluids and had a low order of toxicity. However, its mechanism of antimicrobial action, which purportedly involved inhibition of bacterial cell protein and DNA synthesis by frame-shift mutation, was extrapolated to be non-carcinogenic from studies performed on a "similar acridine derivative proflavine". This might not be a valid assumption.

9-amino acridine is inactivated by soap, and stains carious dentine brown while intensifying the yellow colour of surrounding dentine. This could create aesthetic problems following irrigation of anterior teeth (Schmitz, 1980).

7.11 QUATERNARY AMMONIUM COMPOUNDS

Quaternary ammonium compounds are cationic surface active agents characterised by structural balance between one or more hydrophilic and hydrophobic centres (Harvey, 1980 p978). They might possess antibacterial properties via proteins, metabolic reactions and cell permeability. However their mode of action has not yet been fully elucidated (Petrocci, 1983 p325).

In low concentrations, quaternary ammonium compounds might be bactericidal by causing changes in permeability of the cell membrane. It has been suggested that when these agents accumulate on the surface of the cell wall, they break down the protoplasmic membrane of the bacterial cell by osmosis, after which cell regulation of the entry and release of essential substance is lost (Spängberg, 1973).

Certain chemicals are incompatible with these cationic agents. Anionic surface-active agents such as soaps, in addition to tissue constituents and pus, might inhibit the bactericidal effects (Harvey, 1983 p978).

Strindberg (1957) first advocated the use of a "non-toxic bactericidal quaternary ammonium compound" 0.1% Biosept ¹ in preference to sulphuric acid, which was described as both toxic and corrosive. A previous paper by this author (1956) had suggested that no significant differences existed in "cleansing effect" or "risk of

1. Biosept (Recip, Stockholm, Sweden) contains cetyl pyrimidinium chloride (Engström and Spängberg, 1969).

exacerbation" between the two solutions.

Grahnén and Krasse (1963) compared 0.1% Biosept with a polyantibiotic, Nebacetin (containing neomycin and bacitracin) in water. No obvious difference in efficacy was found from bacteriologic culturing and assessment of "clinical results" (tenderness, pain, swelling, and presence of exudate). A later study by Engström and Spångberg (1969) found 1% Biosept had a "marked antimicrobial effect" *in vitro* against *Staphylococcus aureus*, *Streptococcus faecalis* and *candida*, but not against *Pseudomonas aeruginosa*. However, the authors claimed that the potent toxic effect of 1% Biosept against HeLa cells could not justify its use as an irrigant.

Using cultures to assess effectiveness, Shapiro, Heling and Erb (1966) examined another quaternary ammonium compound, Zephiran¹ as an irrigant. However, the authors could only endorse it in conjunction with paramonochlorophenol as an intracanal medicament. Zephiran was described as relatively nonirritating to tissue (Schilder and Amsterdam, 1959). Others observed that cationic detergents, such as Biosept and benzalkonium chloride, had a detrimental effect on tensile strength of healing skin wounds in rats (Rydberg

1. Zephiran (Winthrop Laboratories, USA), also referred to as "Zephiran chloride", is benzalkonium chloride available as a 0.133% solution, tincture and tincture spray (Harvey, 1980 p978).

and Zederfeldt, 1968).

Kuttler (1964) recommended storage of sterilised endodontic instruments in Zephiran. Linke and Chohayeb (1983) found the solution was bactericidal to selected microorganisms.

A comparison of the wetting characteristics of several irrigant solutions found 1% benzalkonium chloride had favourable wetting properties on dentine (Glantz and Hansson, 1972). Other solutions such as EDTA, hydrogen peroxide and saline were found to exhibit different contact angles with dentine. The authors considered that the high wetting capability was preferable to allow enhanced penetration of solution.

Since 1977, reports on another quaternary ammonium compound, Salvizol¹, and more recently Solvidont (DeTrey, Dentsply, Zurich, Switzerland), have appeared in the dental literature.

Kaufman et al. (1977) described Salvizol as a "new chelating solution" which possessed both inorganic solvent properties (due to its acetate group) and possible organic solvent properties, thereby providing a superior irrigating solution to EDTA.

In a later article by Kaufman et al. (1978), assertions were made that Salvizol had definite organic

1. Salvizol (Ravensburg GMBH, Konstanz, W Germany), also referred to as "bis-dequalinium acetate" or "BDA", is the proprietary name for the quaternary ammonium compound, N¹-N¹ - decamethylene- N⁴-N⁴ - decamethylene-bis-4-aminoquinaldinium-diacetate 0.5% in propylene glycol, pH 6.6 (Koskinen, Meurman and Stenvall, 1980).

and inorganic solvent properties, as well as low toxicity, wide ranging bactericidal qualities, lubricant and surfactant features plus biological compatibility and a neutral pH. From an SEM study, the authors claimed the superiority of Salvizol over EDTAC in cleaning the apical one third of the canal. However, Zach and Kaufman (1983) found that root canal dentine collagen was not dissolved by Salvizol, and other SEM studies found that Salvizol failed to remove the smear layer (Berg et al., 1986) or effectively remove debris (Crabb, 1982).

In an investigation on prepared and unprepared root canal surfaces, it was suggested that Salvizol had a superior chelating effect than EDTA and RC Prep on unprepared surfaces (Ram, 1980). However, only the coronal 5 to 7 mm of the root was instrumented, and comparisons appeared to have been made between micrographs of varying, and at times, unspecified magnification. Koskinen, Meurman and Stenvall (1980) found Salvizol did not interfere with surface morphology of uninstrumented root canal dentine of young premolars.

Solvidont was developed as an "endodontic kit" based on Salvizol, and consisted of BDA in three different concentrations and viscosities:- (1) 0.5% BDA working solution; (2) 0.05% BDA irrigation solution; and (3) 0.5% BDA intracanal medication paste. Kaufman (1981a; 1983; 1983a) presented case reports describing healing of periapical pathosis in a "relatively short time" after use of BDA. Claims were made that periapical lesions

unable to be accessed because of calcified or obstructed canals, would be healed with the use of this material. In addition, it was asserted that, even in the event of a lost temporary filling, the material would continue to act as an effective bactericidal agent in the canal. However, Velvart (1987) found "the cleaning efficacy of Solvidont was unsatisfactory", and Griffiths and Stock (1986) found NaOCl resulted in significantly less debris than Solvidont.

Kaufman and Greenberg (1986) compared the cleansing efficacy of Solvidont irrigant and 5.25% NaOCl using saline as a control. Canals were instrumented, then silicone impressions taken, and examined under a binocular microscope for imprints of canal surface irregularities. It was observed that "most of the gross debris was captured by the silicone impression material". It was asserted that Solvidont was "more effective" in the apical one third, but this was not statistically confirmed.

Spängberg et al. (1978) claimed that Salvizol was "remarkably less toxic" than EDTAC in enhanced vascular permeability tests on rats, and recommended it as an irrigant in preference to EDTAC. However, Spängberg et al. (1988) asserted that data based on the 1978 article had shown Salvizol in aqueous solution was a product "not indicated for endodontic use". Yet in another article (Spängberg et al., 1988a) it was said that Salvizol "has biocompatibility and cleansing properties that makes it

potentially useful in clinical endodontics". It would appear that some confusion exists about this product.

Aron (1986; 1987; 1987a) indicated that Salvizol was less cytotoxic to human cell cultures than NaOCl, and recommended 0.6% Salvizol as a root canal irrigant.

Spängberg et al. performed *in vivo* (1988) and *in vitro* (1988a) studies with 0.125% Solvidont. This irrigant was compared *in vivo* to 1% neutral buffered NaOCl, using enhanced vascular permeability tests on Sprague-Dawley rats (Spängberg et al., 1988). It was found that greater dilution of 1% NaOCl (1:64) than Solvidont (1:8) was required to achieve an equal level of dye leakage. It was concluded that Solvidont was less irritating than 1% NaOCl.

The antimicrobial and toxic effects of the same solutions were examined *in vitro* (Spängberg et al., 1988a). Cytotoxicity tests were performed using labelled L929 mouse fibroblast cells and measured radiochromium release. In addition, the antimicrobial activity of 1% neutral buffered NaOCl and 0.125% Solvidont against *Streptococcus faecalis*, *Escherichia coli* and *Candida albicans* was measured. From this data a cytotoxicity-antimicrobial quotient was obtained which indicated that while the antimicrobial activity of Solvidont was superior to NaOCl, its cytotoxicity was higher. The authors concluded that 0.125% Solvidont did "not provide a better alternative" to 1% NaOCl.

It would appear that studies on Salvizol/Solvidont

have been dominated by one or two authors and claims of superior clinical and long term results (Kaufman, 1981a; 1983; 1983a) have not been obtained by means of controlled experiments. Other research, although also limited, has been unable to substantiate the claims of Kaufman and coworkers. Thus more investigations are required before the routine use of these solutions can be promoted.

7.12 WESCODYNE¹ AND IODOPAX²

Similarly to chlorine, iodine binds the sulfhydryl groups of cysteine in enzymes, thus having a destructive effect on cells (Spängberg, 1973). While iodine is a powerful antiseptic, drawbacks such as hypersensitivity, corrosiveness and staining properties have precluded its use as an irrigant. However organic iodine compounds, or iodophors, such as Iodopax and Wescodyne, provide iodine held in dissociable complexes. Molinari, (1988 pp77-8)

1. The composition of Wescodyne (West Chemical Products, Inc., Long Island City, NY, USA) is:-

Polyethoxy polypropoxy polyethoxy ethanol-iodine complex	9.10%
Nonylphenoxypoly (ethyleneoxy) ethanol-iodine complex	8.74%
Inert ingredients	82.16%

Provides 1.6% min. iodine

Diluted to 0.45% (0.0072% iodine) in distilled water for clinical use (Spängberg, Engström and Langeland, 1973).

2. Iodopax (Ferrosan, Sweden), also known as "Jodopax", is an aqueous solution of 5% iodine dissolved in octylphenoxy polyglycol ether. It is diluted to 0.80% (0.04% iodine) in distilled water for clinical use (Spängberg, Engström and Langeland, 1973).

described iodophors as non-staining, having a prolonged activity and as being significantly less irritating than iodine, while retaining a broad spectrum antimicrobial action. The carriers of the iodine are neutral polymers exhibiting surface-active properties which improve the wetting ability of the solutions (Gottardi, 1983 pp188-9).

From *in vitro* investigations, Engström and Spängberg (1969) found a low ratio of antimicrobial to cytotoxic effect when using Iodopax, and indicated that the potential for toxicity should determine the concentration of solution to be used.

An antimicrobial and cytotoxicity study by Spängberg, Engström and Langeland (1973) found Wescodyne had no antimicrobial effect on *Staphylococcus pyogenes aureus*, *Streptococcus faecalis*, *Pseudomonas aeruginosa* and *candida*, while Iodopax acted only on *Streptococcus faecalis*. Both of these agents were considerably more cytotoxic to L-cells and HeLa cells (human epithelial cells isolated from cancer tissue) than bactericidal to the microorganisms tested. Nonetheless, Spängberg (1985 p571) described iodophors as low surface tension organic solutions of iodine which are excellent aids for cleaning root canals without causing allergic reactions.

7.13 GLUTARALDEHYDE AND POTENTIATED ACID 1,5

PENTANEDIAL

Glutaraldehyde is an antimicrobial agent which has

a broad spectrum of activity, and rapid kill rate against viruses, spores and the cell constituents of vegetative bacteria. It is a saturated five-carbon dialdehyde (1,5 pentanedial) which reacts readily with protein, principally amino groups, to form stable glutaraldehyde-protein crosslinks. The presence of free aldehyde groups is a prerequisite for good biocidal activity. Increases in temperature can produce more free aldehyde in acid solution, while in alkaline solution, temperature increase leads to loss of reactive aldehyde (Scott and Gorman, 1983 pp65-80).

The biocidal activity of glutaraldehyde is highly dependent upon pH, and the rate of reaction increases considerably over pH 4-9 (Scott and Gorman, 1983 p67). At a lower pH, kill rate is decreased. However, the stability of alkaline solutions is considerably less than acid solutions, because of polymerisation reactions at the higher pH, which result in a corresponding loss of antimicrobial activity. Lowering the pH has the effect of prolonging the shelf life of the solution by reducing the rate of glutaraldehyde polymerisation.

In practice, glutaraldehyde is generally available as a 2% solution to which an "activator" is added, bringing the pH to approximately 8 before use (Scott and Gorman, 1983 pp75-6). To maintain the antimicrobial effect, a surfactant is added.

Endodontic application of 2% glutaraldehyde, as an irrigant which "fixes" necrotic pulp contents, has been

suggested (Wemes et al., 1982; 1982a; Wemes, Veldkamp and Purdell-Lewis, 1983; Wemes and Arends, 1984). Wemes, Veldkamp and Purdell-Lewis (1983) hypothesised that, since lateral canals were unable to be reached during standard instrumentation, glutaraldehyde could be used to "irreversibly" and "instantaneously" fix organic material. It was claimed that, as the canal would be "sterilised" instantly by fixation of bacteria, the need for other medicaments routinely used during endodontic procedures could be avoided. These authors claimed high success rates using glutaraldehyde as an irrigant, in a pilot study of patients undergoing single appointment endodontics.

In an *in vivo* comparison with another aldehyde (formocresol), glutaraldehyde was found to be significantly less irritating to periapical tissues (Wemes et al., 1982a). A further study indicated that, while formocresol readily diffused through dentinal tubules, glutaraldehyde showed no evidence of diffusion after 72 hours (Wemes et al., 1982). Assertions that glutaraldehyde "softened" dentine were unsupported (Wemes et al., 1982).

Wemes and Arends (1984) investigated the claims of dentine softening following use of 2% glutaraldehyde (pH 2.7-3.7) in a later study. Bovine dentine samples were immersed in glutaraldehyde for either 10 or 20 minutes, and subsequently tested for differences in microhardness, both immediately and 48 hours later. Results indicated an

immediate softening of dentine followed by a subsequent increase in microhardness of up to 15% more than originally. While the authors claimed their results were "significant", details of statistical analysis were not included. The increase in microhardness after 48 hours was considered to result from cross-linking of collagen originally freed from mineralised dentine by glutaraldehyde.

Attempts have been made to potentiate the antimicrobial activity of glutaraldehyde by the addition of surface active agents (cationic, nonionic, anionic, amphoteric and nonionic ethoxylates of isomeric linear alcohols) (Martin, 1975). It was speculated that this would increase the wettability of bacterial cell walls, and thus allow faster penetration of the medicament (Martin, 1975). Martin compared the bactericidal effects of 2% potentiated acid 1,5 pentanediol, pH 5.8 (Sonacide, Wave Energy Systems, Inc. NY, USA), with those of 5.5% NaOCl. Both agents were shown to be bactericidal against *Streptococcus faecalis*, *Streptococcus mitis*, *Staphylococcus aureus* and *Escherichia coli*. However, 5.5% NaOCl was found to be less effective in the presence of serum proteins, and for this reason, Martin suggested that potentiated acid 1,5 pentanediol had "potential" as an endodontic irrigant.

A later study (Martin, 1978) showed that a 2% solution caused toxic reactions when applied to rabbit eyes and rat connective tissue, while a 1% solution was

less toxic.

7.14 CHLORHEXIDINE

Chlorhexidine is a cationic bisbiguanide with antibacterial properties. It is a strong base, which is most stable in the digluconate form of its salts (Foulkes, 1973). The preparation, Chlorhexidine Gluconate BP, is a 20% w/v aqueous solution from which several formulations containing 0.02%-1.0% w/v chlorhexidine are prepared (Gardner and Gray, 1983 pp251-4). The gluconate solution is used in alcohol solutions with alcohol concentration ranging from 60%-90%. Most research on chlorhexidine, for dental use, has been based on its role in the control of dental plaque (Hennessey, 1973). However, in 1962, Savlon¹ was introduced as an irrigant, either alone, or as an adjunct to EDTAC at the Adelaide Dental School (Heithersay, 1984).

Chlorhexidine is incompatible with some anions including natural soap, synthetic detergents, and inorganic anions such as chloride and phosphate (Gardner and Gray, 1983 p252). These latter precipitate chlorhexidine as insoluble salts, thereby diminishing the availability of its active form. The optimum pH for antibacterial activity of chlorhexidine is 5.5 to 7.0. Above pH 8.0 the chlorhexidine base is precipitated, and at a low pH, the activity of the solution gradually

1. Savlon (ICI, Australia) solution contains 0.03% chlorhexidine and 0.3% cetrimide (Heithersay, 1984).

deteriorates.

Chlorhexidine is inactive against spores at room temperature, but is biocidal to vegetative gram-positive and gram-negative bacteria (Molinari, 1988 p79). It has also been described as having a broad spectrum antibacterial action with greater activity against gram-positive than gram-negative microorganisms (Hennessey, 1973).

Emilson (1977) examined the susceptibility of a range of aerobic, facultative anaerobic and anaerobic microorganisms to chlorhexidine *in vitro*. Specimens were gathered from wounds, saliva and dental plaque. Anaerobes such as *Propionibacterium* were found to be susceptible to chlorhexidine, while another anaerobe, *Veillonella* was one of the least susceptible. There was a broad range of response to chlorhexidine among gram-positive and gram-negative microorganisms. *Bacteroides* species, which appear to play a significant role in endodontic infections (see Chapter 4) were not included in this study.

Other *in vitro* studies have shown that chlorhexidine can be adsorbed to hydroxyapatite (Rölla, Löe and Schiött, 1970; Nordbö, 1972; Emilson et al., 1973), and when concentration of chlorhexidine in the immediate environment is low, adsorbed chlorhexidine is released. Further investigations are required to determine the action of chlorhexidine on the necrotic and contaminated contents of a root canal.

Reports of adverse toxicity following use of chlorhexidine are few. Foulkes (1973) reported that inadvertent intravenous administration of up to one litre of 1 in 5000 (0.02% w/v) chlorhexidine resulted in haemolysis from the ensuing hypotonicity rather than from a direct effect of the chlorhexidine, and following exchange transfusion, patient recovery was complete. Concentrations greater than 2% may lead to dermal discomfort and up to 0.2% are tolerated by the eye (Gardner and Gray, 1983 p259). However, Spångberg, Engström and Langeland (1973) found the toxic effects of 0.05% chlorhexidine on L-cells and HeLa cells to be ten times greater than the bactericidal effect. Biological evaluation of 0.2% chlorhexidine, *in vitro* and *in vivo*, revealed that it exerted a less toxic effect on HeLa cells and rabbit muscle tissue than did 5% chloramine-T and a similar effect to that produced by Biosept, Iodopax and 0.5% NaOCl (Wennberg, 1980).

Using SEM, the efficacy of a solution containing 0.1% chlorhexidine was investigated by Tucker, Mizrahi and Seltzer (1976). Their results were compared with those from a previous SEM study by Baker et al. (1975), in which many irrigants was assessed. Results indicated that no canal was free of debris regardless of which irrigant was used.

Parsons et al. (1980) investigated the adsorption

of Hibitane¹ and 1% chlorhexidine to bovine pulp and dentine specimens following 20 minute and 40 minute contact times. Release of chlorhexidine from the specimens was measured either immediately or one week later. Antibacterial action to *Streptococcus faecalis* was also assessed. It was found that pulp and dentine specimens both took up and released chlorhexidine. In addition, antibacterial properties were acquired which were unchanged one week later. Changes in concentration and time of exposure did not make a significant difference to chlorhexidine uptake, or the acquisition of antibacterial properties by the specimens.

It would appear that while chlorhexidine may be a superior irrigant to saline (Delany et al., 1981), it is not as effective as 2.5% NaOCl (Ringel et al., 1982). Delaney et al. (1981) compared the *in vitro* antibacterial effect of 0.2% chlorhexidine compared to 0.9% sterile saline. Although results indicated that chlorhexidine significantly reduced the number of viable microorganisms, the experimental methodology indicated that the use of an intracanal medicament might have influenced the results.

Ringel et al. (1982) performed an *in vivo* comparison between 0.2% chlorhexidine and 2.5% NaOCl. Anaerobic culturing of 60 teeth with necrotic pulps and positive cultures at the commencement of treatment,

1. Hibitane (Imperial Chemical Industries, England) contains 0.2% chlorhexidine (Parsons et al., 1980).

showed that 2.5% NaOCl was a more effective antibacterial agent than 0.2% chlorhexidine. The hypothesis that chlorhexidine was likely to be a superior agent over time, because of its ability to be adsorbed and released by dental tissues, thus providing a prolonged effect, was not endorsed. The prevalence of positive cultures and culture reversals over three appointments indicated that chlorhexidine did not have a prolonged bactericidal effect.

More recent studies have indicated that chlorhexidine, both in solution and when delivered via a controlled release system, might be able to exert an antibacterial effect *in vitro* against a variety of bacteria (Cervone, Tronstad and Hammond, 1989; Heling et al., 1989).

7.15 SALINE

Sodium chloride (NaCl) has a molecular weight of 58.44 and is a highly water-soluble white crystal. Its mode of bacterial inhibition is primarily by plasmolysis. Other bactericidal effects include dehydration, interference with enzymes, oxygen removal, and toxicity when either of its ion components is in high concentration. There is great variation in the tolerance of bacteria to saline, and it has been suggested that the lethal effect of saline is associated with loss of Mg^{2+} from the bacterial cells (Busta and Foegeding, 1983 p661).

The most salt-sensitive bacteria are mesophilic gram-negative rods and psychrotrophic¹ bacteria which tolerate no more than 4% to 10% salt (Busta and Foegeding, 1983 p661). However, physiologic sterile saline (0.9%), which is the concentration generally employed for endodontic irrigation, is unlikely to have a bactericidal effect, and in fact, might provide nourishment for bacteria.

According to the dental literature, saline has been mostly employed as a negative control irrigant (Schilder and Amsterdam, 1959; Grahén and Grasse, 1963; Kopp, Lazzari and Madden, 1977; Trepagnier, Madden and Lazzari, 1977; Svec and Harrison, 1977; Hand, Smith and Harrison, 1978; Martin, 1978; Rosenfeld, James and Burch, 1978; Bolanos and Jensen, 1980; Yamada et al., 1983; Berg et al., 1986; Kaufman and Greenberg, 1986; Baumgartner and Mader, 1987).

However, the routine use of saline has been advocated as the sole irrigant and to control bleeding (Morse, 1974, pp453-6). It has also been proposed as a final flush to reduce irritation from other irrigants (Curson, 1966), to avoid the irritation potential of other irrigants (Matsumiya and Kitamura, 1960; Martin, 1979; Sinanan, Marshall and Quinton-Fox, 1983) and because no other irrigant provides superior qualities

1. Psychrotrophic bacteria grow over a temperature range of 0°C to 30°C (Hammond, 1988 p57).

(Baker et al., 1975; Tucker, Mizrahi and Seltzer, 1976).

Gutiérrez and Garcia (1968) suggested that a "heavy crystalline deposit" was produced by oversaturation of canals with saline solution, which then made subsequent canal obturation more difficult. Jolly and Sullivan (1956) noted that Cetavlon should not be used with saline irrigation, as the inorganic ions could inhibit the cationic quaternary ammonium compound .

Senia, Marshall and Rosen (1971) found no significant difference in the debridement effect of 5.25% NaOCl and saline when viewed at x100 magnification. However, NaOCl was used only after completion of canal preparation, which had used saline solution. Svec and Harrison (1977) found no difference between saline and the combined use of hydrogen peroxide and NaOCl at the level of 5 mm from the apex under a light microscope, while in a SEM study, Bolanos and Jensen (1980) observed that saline irrigation produced similar results to NaOCl and RC-Prep.

In vivo studies established that saline irrigation during mechanical preparation, without subsequent application of an intracanal medicament, resulted in increased numbers of microorganisms at following appointments (Byström and Sundqvist, 1981; 1983). However, it was found that there were fewer bacterial cells at the end of each appointment than at the beginning, which was attributed to the mechanical flushing effect of the solution. Mentz (1982) suggested

that sterile saline was a useful flushing agent, but ineffective as a bactericidal agent.

Saline does not remove the smear layer (Baumgartner et al., 1984; Berg et al., 1986; Baumgartner and Mader, 1987), nor provide chelating action (Wayman et al., 1979), or effective tissue solvent properties (Kopp, Lazzari and Madden, 1977; Hand, Smith and Harrison, 1978; Wayman et al., 1979; Baumgartner and Mader, 1987). It would appear that far better irrigating solutions than saline exist, and the routine use of saline is inappropriate.

7.16 WATER

References to the use of tap and distilled water as irrigants appear in the literature.

Sterile distilled water has been described as a bland solution useful for its flushing action (Ingle and Zeldow, 1958; Nicholls, 1962; Martin, 1979; Mentz, 1982; Sinanan, Marshall and Quinton-Fox, 1983), but it has not been satisfactorily assessed. Several studies have used distilled water as a control irrigant (Cohen, Stewart and Laster, 1970; Abou-Rass and Patonai, 1982; Berg et al., 1986; Goldberg, Massone and Spielberg, 1986).

Auerbach (1953) attempted to show that reaming and filing using 5.25% NaOCl, followed by irrigation with hot distilled water, produced a sterile canal, thus negating the need for an antibiotic intracanal dressing. Using this technique, this author claimed a 75% success rate in

obtaining sterile bacteriological cultures of previously contaminated canals. However, it is probable that any bactericidal effect was because of NaOCl.

Others found distilled water did not have an antibacterial effect, and suggested that mechanical instrumentation using water might only temporarily reduce the numbers of microorganisms in the canal (Ingle and Zeldow, 1958; Shih, Marshall and Rosen, 1970).

Distilled water irrigation was found to be as effective as the alternate use of 3% hydrogen peroxide and 6% NaOCl (McComb and Smith, 1975), but neither was effective in removing both smear layer and superficial debris from canal surfaces (McComb, Smith and Beagrie, 1976).

In an SEM study, Rubin et al. (1979) indicated that tap water was as effective an irrigant as the more conventional chemical solutions. Regardless of which irrigant was used, organic and inorganic debris remained in the canal. Hand, Smith and Harrison (1978) found that distilled water was an ineffective necrotic tissue solvent.

Other authors have recommended final irrigation with sterile water to prevent any precipitate formation following use of saline, NaOCl, EDTA or citric acid, as well as terminate any solvent action of irrigants previously used (Baumgartner et al., 1984; Baumgartner and Mader, 1987).

7.17 POTASSIUM HYPOCHLORITE/HYDROGEN PEROXIDE

According to Schroeder (1981), this combination was recommended by Redtenbacher in 1972. Potassium hypochlorite or Pulpolyt (Heilmittelwerke, Vienna, Austria) provided 7% effective chlorine, while Perhydrol (Heilmittelwerke, Vienna, Austria) provided 30% hydrogen peroxide. It was claimed that the "spreading" effect of Pulpolyt in combination with Perhydrol lead to an "explosion-like cleaning" of the canal "in a coronal direction" (Schroeder, 1981).

7.18 DECAL

Fraser (1974) observed cervical and midroot, but not apical, dentine softening following a 15-minute application of Decal (Glover Laboratories, Melbourne, Australia), which contained 5.3% oxyacetic acid, 4.6% ammonium oxyacetate, and 0.06% cetyl trimethyl ammonium bromide at pH 3.4.

Koskinen, Meurman and Stenvall (1980) examined the effect of Decal on specimens from uninstrumented root canal wall surfaces of young premolars. While Decal was found to have no effect on organic tissue, it did cause decalcification of exposed mineralised dentine.

7.19 POLYACRYLIC ACID

Using SEM, McComb and Smith (1975) concluded that, on instrumented root canal surfaces, 20% polyacrylic acid irrigation, was inferior to REDTA. This was thought

possibly to be caused by the higher viscosity, and thus slower movement, of polyacrylic acid.

Less viscous solutions of polyacrylic acid (5% and 10%) were examined in a later SEM study with "disappointing" results (McComb, Smith and Beagrie, 1976). Accessible areas of the canal were etched by the solutions and the smear layer removed, but more apically, the solution tended to coat the canal.

7.20 NELEX

Hakala, Koskinen and Närvänen (1976) investigated the effectiveness of the irrigant Nelex (BYK-Gulden, Lomberg, GMBH, Konstanz, West Germany), a condensate of m-cresolsulphonic acid and formaldehyde (pH 0.6). The authors claimed that Nelex was well tolerated by tissues, with "a tendency to dissolve necrotic tissue selectively, leaving normal tissue unaffected". It was also asserted that while dissolution of inorganic material was dependent upon pH, dissolution of organic necrotic matter was dependent upon the concentration of the solution. Nelex was found to be seven times more effective than EDTAC in the demineralisation of powdered dentine. However, the authors cautioned that the strong acid action of Nelex had the capacity to etch enamel, and inactivate other medicaments. A later study (Koskinen, Meurman and Stenvall, 1980) also found that 5% and 20% Nelex exerted a powerful demineralising effect on uninstrumented root canal dentine, which extended inside

the tubules and resulted in funnelling of tubule orifices.

7.21 SODIUM LAURYL SULPHATE

Sodium lauryl sulphate is an anionic surface-active agent, which is able to exert antibacterial activity against gram-positive and, to a lesser extent, gram-negative organisms. Although the mechanism of action is not known, it might involve protein denaturation, inactivation of enzymes essential to cell metabolism and/or disruption of cell membrane permeability. It is stable, possesses good detergent qualities, and is rapidly active and effective against a wide range of microorganisms, but is effective only at acid pH (Dychdala, 1983a pp330-1).

One article appears in the dental literature investigating this compound as an irrigant. Lehman, Bell and Gerstein (1981) examined the debridement ability of sodium lauryl sulphate (SLS) alone, in various proportions with 5.25% NaOCl, and compared to 5.25% NaOCl. Mechanical preparation using the irrigants was performed on 45 mandibular molars, then root sections were examined at x100 magnification under a binocular microscope for evaluation of remaining debris. The inclusion of SLS was not found to improve the ability of solutions to remove debris, and in fact, 5.25% NaOCl alone provided the best debridement. However, the comparisons of solutions may not be valid as the canals

were not filed to a standard lumen size. In fact the comment was made that "the larger the diameter the more effective the cleaning action".

7.22 DODECYLDIAMINOETHYL GLYCINE

Dodecyldiaminoethyl glycine or TEGO (Goldschmidt Products Corporation, White Plains, NY, USA) has been described as an ampholytic soap which forms both cations and anions, thus potentially having both the bactericidal activity of cations and the solvent action and surface tension reduction properties of anions. Ampholytic soaps in combination with protein cause little or no precipitation (Hesselgren, Dahl and Needlick, 1973).

The efficacy of smear layer removal by a 1% solution of TEGO was compared to REDTA and 5.25% NaOCl using SEM (Goldman et al., 1981). TEGO did not remove the smear layer and its detergent action was not found to be useful. Bactericidal activity was not investigated in this study, however evidence of bactericidal activity of dodecylaminoethyl glycine against *Streptococcus faecalis*, *Escherichia coli* and *Candida albicans* has been observed previously (Hesselgren, Dahl and Needlick, 1973).

7.23 ALCOHOL

The use of 70%-90% methanol or ethanol (0.5 ml to 1 ml) has been suggested as a final irrigant to dry and dessicate the canal, as well as remove any traces of remaining chemicals (Walton and Torabinejad, 1989 p208),

although no evidence exists to support the effectiveness of the regimen. Penick and Osetek (1970) suggested that irrigation with alcohol should follow chloroform or Xylene when removing gutta percha.

7.24 TANNIC ACID

Using SEM, Bitter (1989) examined the effectiveness of 25% tannic acid as an irrigant to remove the smear layer. It was speculated that tannic acid could precipitate protein, as well as affect the inorganic component of dentine. Bitter asserted that tannic acid produced a "cleaner dentinal surface", however, he compared it with the combined use of NaOCl and hydrogen peroxide, a regimen which McComb and Smith (1975) had shown did not remove the smear layer. Further, tannic acid was not actually used to irrigate the canal, but was "placed in the canal for 30 seconds followed by irrigation with water". Additional studies are required before tannic acid can be endorsed as an endodontic irrigant.

CHAPTER 8

IRRIGANTS AND ULTRASOUND

8.1 INTRODUCTION

The use of irrigants in conjunction with ultrasonic instrumentation features increasingly in current literature. It has been heralded as the answer to the long standing endodontic concern of totally effective root canal debridement. Up to this time, no single instrument design or technique has satisfied this demand (Ahmad, Pitt Ford and Crum, 1987).

Ultrasound encompasses vibratory waves with frequencies higher than the human hearing range (Walmsley, 1987). It has been employed in science and industry in the fields of diagnosis, machining, cutting and smoothing. In dentistry, ultrasonically activated instruments have been used for many years to remove calculus from tooth surfaces.

The application of ultrasonic energy to endodontics (frequently termed "endosonics") was first suggested by Richman in 1957, who described the use of the Cavitron Ultrasonic Dental Unit (Cavitron Equipment Corporation, Long Island, NY, USA) during canal preparation, and in bone and tooth removal during apicectomy procedures. Richman also suggested drying the root canal by running the machine at very low power without a coolant present.

Endodontic ultrasonic instrumentation systems use files vibrating at approximately 25 kHz, coupled to an ultrasonic transducer which causes the file to vibrate in either transverse or longitudinal nodes (Lumley, Walmsley and Laird, 1988; Williams, 1987). The vibration results in abrasion of the canal wall, and is supplemented by cooling liquid (which can be a disinfectant) that is carried along the length of the file. Two commercially available ultrasonic units are the Cavi-Endo (LD Caulk, Division of Dentsply Intl., Milford, DE, USA) and the Enac (Osada Electric Co., Tokyo, Japan).

As opposed to ultrasonic systems, sonic systems "produce vibrations within the audible frequency range by means of compressed air, which activates a rotor-shaft assembly as a source of vibration" (Heuer and Miserendino, 1987 p431). Sonic instrumentation systems, such as the Endostar-5 (Syntex Dental Products, Inc., Valley Forge, PA, USA) and Endosonic 3000 (Medidenta International Inc., Woodside, NY, USA), utilise modified air-driven handpieces. The irrigant used with sonic systems is limited to filtered tap water, delivered via the dental unit (Fairbourn, McWalter and Montgomery, 1987; Heuer and Miserendino, 1987 p431).

This chapter will discuss the efficacy of ultrasonics in endodontics, with particular emphasis on irrigants associated with their use.

8.2 MECHANISMS OF ACTION

The chief mechanisms attributed to the effect of ultrasound in endodontics, are cavitation and acoustic streaming (Ahmad, Pitt Ford and Crum, 1987), although this is controversial. The involvement of heating, stirring and agitation has also been proposed (Cunningham, Martin and Forrest, 1982).

Cavitation has been defined as "the growth and subsequent violent collapse of a small gas-filled preexisting inhomogeneity in the bulk fluid" (Ahmad, Pitt Ford and Crum, 1987). A cavitating liquid is considered to be capable of inducing certain chemical processes which contribute to the destruction of bacteria, such as oxidation and degradation of polysaccharides, proteins, nucleic acids and glycosaminoglycans (Pedicord, Eldeeb and Messer, 1986).

Martin (1976) suggested that the key physical mechanism of ultrasonics in endodontics was provided by cavitation within the irrigating solution. Moorer and Wesselink (1982) further suggested that cavitation enhanced the tissue dissolving capabilities of NaOCl.

However, Ahmad, Pitt Ford and Crum (1987), in a study using the Cavi-Endo ultrasonic unit, found no evidence of the phenomenon of cavitation, and suggested that the displacement amplitude of the file in this system was not great enough to generate the appropriate negative acoustic pressure amplitude necessary to induce cavitation. These researchers reasoned that if, as Martin

(1976) suggested, cavitation was the essential feature in the mechanism of ultrasonics, then equally clean canals should result with water or NaOCl as the irrigant, as the cavitation threshold of water is similar to that of NaOCl. This was not observed.

In a later study, Ahmad et al. (1988) found that it was necessary to enlarge a canal to size 40 before an oscillating size 15 file could induce cavitation. Under SEM, they then compared canal walls which had, or had not, been subjected to cavitation. No difference in cleanliness between the two groups was detected.

Another author noted that the geometrical limitations placed upon the design of endodontic ultrasonic instruments meant that cavitational activity was unlikely to occur to any great extent (Williams, 1987). Lumsley, Walmsley and Laird (1988) found that although cavitational activity could be detected with the Cavi-Endo, it did not occur along the entire length of the file.

Acoustic streaming, described as "the rapid movement of particles of fluid in a vortex-like motion about a vibrating object" (Ahmad, Pitt Ford and Crum, 1987), appears to play a more important role in the mechanism of ultrasonic debridement, by disaggregating bacteria and removing debris from the walls of the canal (Walmsley, 1987). However, when following the manufacturer's instructions for the Cavi-Endo ultrasonic unit, insufficient acoustic streaming may be produced to

displace debris (Ahmad, Pitt Ford and Crum, 1987). Using smaller files on higher power settings might generate more acoustic streaming (Ahmad, Pitt Ford and Crum, 1987a).

Ultrasonic activation of irrigants increases the temperature of the solution (Moorer and Wesselink, 1982; Cameron, 1988a). This is postulated to result from the absorption of acoustic energy by the liquid (Pedicord, Eldeeb and Messer, 1986), and to heat production resulting from frictional contact between the canal wall and oscillating file (Martin, Cunningham and Forrest, 1982).

8.3 PRELIMINARY INVESTIGATIONS

Ultrasonic lavage was seen as an advancement over its forerunner, pulsed mechanical lavage (Martin, 1976). The advantages of pulsating lavage over syringe irrigation were shown in a study on inflicted wounds in dogs (Stewart et al., 1971). A pulsed mechanical lavage system was able to deliver irrigating solution at a higher peak pressure than rubber bulb syringes, and in doing so was more effective in the removal of bacteria from wounds.

It was research by Martin, Cunningham and co-workers (1976-1982), using a prototype model ultrasonic unit (Litton model LT: 200; Litton Dental Products, Olathe, KS, USA), that led to the introduction, in 1982, of the Cavi-Endo, a commercial machine specifically

designed for endodontic use. It is of note that these early studies contained several unsubstantiated statements. In addition, the Cavi-Endo does not have the aspiration feature of the Litton prototype.

Martin (1976) examined the activity of ultrasonics with and without bactericidal irrigation solutions (NaOCl and potentiated acid 1,5 pentanediol). He claimed that while ultrasonics used with a neutral phosphate buffer solution had a "reducing effect" on four bacteria (*Streptococcus faecalis*, *Streptococcus mitis*, *Staphylococcus aureus* and *Escherichia coli*), "coupling it with a biocidal agent led to a more efficient bactericidal synergism". Takagi (1978) also asserted that ultrasound enhanced the "bactericidal action" of solutions, such as 10% NaOCl and 3% hydrogen peroxide.

From calculations of the percentage weight reduction of prepared 4 mm root sections, it was found that greater dentine removal could be obtained by ultrasonic filing than by conventional hand filing methods (Martin et al., 1980; Martin, Cunningham and Norris, 1980).

In a later study, which used eleven pairs of extracted teeth, Cunningham, Martin and Forrest (1982) hand instrumented one tooth of each pair to an unspecified size and compared these with the partner tooth instrumented ultrasonically to size 15. The irrigant in each case was 2.5% NaOCl and instrumentation time for each tooth, regardless of method used, was three

minutes. Light microscopy observation demonstrated the superior cleaning ability of ultrasonic irrigation to the authors. This was attributed partly to the flow-through irrigation feature, which formed part of the ultrasonic system, and allowed constant replenishment of the irrigating solution, and potentially, elimination of any protein inactivated solution.

The amount of material extruded beyond the apex using either hand or ultrasonic instrumentation was compared by weighing (Martin and Cunningham, 1982). Results indicated that ultrasonic techniques produced less debris regardless of whether the preparation had extended through the apex or remained within the canal. The authors noted that the ultrasonic technique employed in their experiment had used a higher volume of irrigant than the hand technique. In addition, the aspiration feature, which was an integral component of the prototype ultrasonic unit they used, provided a means of debris removal.

A survey conducted on 328 patients undergoing endodontic therapy, half of whom had conventional instrumentation and half ultrasonic instrumentation, reported no significant difference in the incidence of post-operative pain (Martin and Cunningham, 1982a).

Martin (1984) maintained that ultrasonic instrumentation provided many advantages over conventional hand instrumentation i.e. reduced operator fatigue, less chance of ledging, a shorter required

preparation time and the ability to create a significantly cleaner canal through continuous irrigation. They claimed that because a high volume of irrigant was able to pass along the file into the canal, debridement and disinfection could occur simultaneously.

8.3 SUBSEQUENT INVESTIGATIONS

Many studies have been executed to verify the claims of earlier research, with a variety of methods and results.

8.3.1 Debridement

Reinforcing the claims of the superior debridging potential of ultrasound in endodontics are Weller, Brady and Bernier, 1980; Crabb, 1982; Cameron, 1982; 1983; 1986; 1987; 1988; Goodman et al., 1985; Nehammer and Stock, 1985; Alaçam, 1987; Stamos et al., 1987; Teplitsky et al., 1987; Yamaguchi et al., 1988 and Haidet et al., 1989. It has also been suggested that ultrasonic preparation might eliminate inflammatory root resorption (Lindskog, Berg and Pierce, 1986).

However, several authors have been unable to demonstrate significant benefits in use of ultrasonic instrumentation over hand instrumentation (Cymerman, 1983; Tauber et al., 1983; Langeland, Liao and Pascon, 1985; Ahmad, Pitt Ford and Crum, 1987; Lev et al., 1987; Reynolds et al., 1987; Goldman et al., 1988; Baker et al., 1988; Biffi and Rodrigues, 1989; Ciucchi, Khettabi

and Holz, 1989; Walker and del Rio, 1989). Velvart (1987) commented that "for proper instrumentation of the root canal the use of hand instruments is unavoidable".

Controversy exists over whether ultrasonic instrumentation can eliminate bacteria from the root canal more effectively than hand instrumentation. While some studies indicated that ultrasonification was able to more efficiently eliminate bacteria from experimentally infected canals (Martin, 1976; Cunningham et al., 1982), and during clinical trials (Sjögren and Sundqvist, 1987), other studies have not found this in regards to the elimination of a mixed flora (Barnett et al., 1985), *Bacteroides intermedius* (Ahmad, 1989) or *Serratia marcescens* (DeNunzio et al., 1989).

Sundqvist, Johansson and Sjögren (1989) observed that less post treatment symptoms occurred after ultrasonic instrumentation than after hand instrumentation. However, this data came from a small sample and was not statistically analysed. The authors hypothesised that hand instrumentation was more likely than ultrasonic instrumentation to pump bacteria through the apical foramen.

While Sjögren and Sundqvist (1987) suggested that ultrasound might enhance the bactericidal potential of NaOCl, DeNunzio et al. (1989) concluded that "selection of the Cavi-Endo or hand filing method of instrumentation should be dependent on factors other than the ability to

remove bacteria from the root canal system".

8.4.2 Curved canals

The effect of ultrasonic instrumentation on narrow, curved canals has been examined, again, with varying results.

Chenail and Teplitsky (1985) found that more than 94% of curved canals (defined as possessing 10-110 degrees curvature) were not straightened following size 15 file ultrasonic instrumentation, and canals appeared to be enlarged uniformly around the entire file. However, in a later study under the same conditions, larger and less flexible size 20 and 25 ultrasonic files were used, with canals being straightened in all cases (Chenail and Teplitsky, 1988).

Yamaguchi et al. (1988) noted that when apical preparation reached size 60 in canals with curvatures up to 30 degrees, ledging occurred, which the authors suggested could be avoided by precurving ultrasonic files. Regardless, the requirement for size 60 apical preparation in curved canals is debatable.

Other studies have compared ultrasonic with hand instrumentation to assess efficacy (Langeland, Liao and Pascon, 1985; Reynolds et al., 1987; Ahmad and Pitt Ford, 1989; Tang and Stock, 1989).

Langeland, Liao and Pascon (1985) showed that both techniques thoroughly cleaned straight, circular canals. However, after observing that neither technique totally

cleaned curved or irregular canals, it was concluded that root canal anatomy and pathosis were the vital factors in determining full debridement.

In another study, which compared hand and ultrasonic techniques on moderately curved canals (10-30 degrees), it appeared that a greater proportion of the canal wall was planed with hand instrumentation (Reynolds et al., 1987).

Ahmad and Pitt Ford (1989) observed that "severely" curved canals were prepared in a "similar manner" using either hand or ultrasonic instrumentation, while in contrast, Tang and Stock (1989) indicated that ultrasonic instrumentation produced a lower incidence of apical zipping and elbow formation in severely curved canals. Differences in operator technique might explain this apparent discrepancy.

8.4.3 Other studies

Claims of a shorter required working time using ultrasonic preparation (Martin et al., 1980; Martin, Cunningham and Norris, 1980) were not supported by Pedicord, Eldeeb and Messer (1986). In a trial which attempted to simulate clinical conditions more closely, the time taken to complete hand instrumentation was found to be significantly shorter than the time required for ultrasonic instrumentation.

Using the Cavi-Endo and Enac ultrasonic units, Yahya and ElDeeb (1989) also found that hand

instrumentation, as well as producing the "best shape at the coronal level", was faster than ultrasonic instrumentation.

A clinical evaluation of the Cavi-Endo over a ten month period reported lowered operator fatigue (Stamos et al., 1985). However, care needed to be taken to avoid using too large a file or leaving the file in the canal for too long, or "zipping" occurred. Nehammer and Stock (1985) commented that the only disadvantage with ultrasonic instrumentation lay in the lack of "feel" when cutting dentine, which could potentially result in perforation of curved canals. Calhoun and Montgomery (1988) also warned that care was required when using the Enac ultrasonic instrument, because of its very aggressive cutting action.

Other clinicians have reported inconvenience with ultrasonic instrumentation because of the inability to place length determining rubber stoppers, which would block the flow of irrigant (Chenail and Teplitsky, 1985; Pedicord, Eldeeb and Messer, 1986). However, this problem has since been circumvented, as both the Cavi-Endo and Enac units now utilise a section of plastic tubing, which acts as a moveable marker around the shaft of the instrument (Heuer and Miserendino, 1987 p431).

8.5 USE OF IRRIGANTS WITH ULTRASOUND

8.5.1 Volume

Early proponents of the ultrasonic system suggested

it was a superior technique partly because of its ability to provide a high volume, constant flow irrigation system (Martin and Cunningham, 1982). However, these claims were not supported by Baker et al. (1988) who measured the volumes of irrigant employed to prepare canals using either hand or ultrasonic instrumentation. On average, ultrasonic instrumentation utilised 141 ml of irrigant per tooth, while hand instrumentation used 35 ml. Despite this volume disparity, no significant difference in overall canal cleanliness was observed under SEM.

A study which utilised high volume constant flow irrigation for both hand and hand/ultrasonic preparation of canals, found that neither technique cleaned the canal completely, although hand/ultrasonic preparation produced significantly cleaner canals at the 1 mm apical level and at isthmuses (Goodman et al., 1985). However, in a similar study, Lev et al. (1987) found differences only at isthmuses.

8.5.2 Time and flow

It has not been unequivocally established whether ultrasound enhances the flow of irrigant solution.

Teplitsky et al. (1987) used radioopaque dyes and radiographs to determine the degree of irrigant penetration along the length of simulated canals during syringe and ultrasonic irrigation. Greater dye penetration was observed following ultrasonic irrigation. However, in the same study, syringe irrigation with a 30-

gauge needle was found to be more effective in natural teeth once apical preparation reached size 35. The authors proposed that because it was difficult to repeatedly place a small needle to the desired length, conventional syringe irrigation techniques presented problems for clinicians. This factor, coupled with the fragility of a 30-gauge needle provided, in their opinion, support for the use of ultrasonics.

A study using fabricated resin canals found that the flow of irrigants, used with oscillating ultrasonic diamond files (size 35 and 45), was limited to 1 mm beyond the tip of the file (Krell and Johnson, 1988). The authors concluded that ultrasonic delivery could not provide apical irrigation.

Other research (Krell, Johnson and Madison, 1988), also found that freely vibrating files had "nodes", described as crossover points on the file, "where the energy of the amplitude changes from the positive side of the deflection to the negative side of the deflection". It was shown that, when a size 15 ultrasonic file was bound by a canal obstruction, there was no irrigant penetration beyond the first node, and only after the instruments were freed could the irrigant penetrate the canal completely. In addition, ultrasonic files required a minimum activation period to remove coronal obstructions, especially in small canal spaces. The authors suggested that current controversies about ultrasonic debridement were a reflection of operator

variables, such as length of time of file activation.

The degree of toluene dye displacement from simulated resin canals (prepared up to size 25 apically), following conventional and ultrasonic instrumentation, was investigated by Druttman and Stock (1989). While it was observed that replacement of irrigant became "less effective towards the apex regardless of the method of irrigation", effectiveness of ultrasonic irrigation depended on time of activation, and not on flow rate. Furthermore, conventional syringe and 30-gauge needle delivery was better able to clean the apical level of the canal, although complete clearance of dye was not achieved. However, the authors felt that the clinical disadvantages associated with the use of a 30-gauge needle, such as the risk of deposition of irrigant through the apex, precluded its use.

8.5.3 Type of irrigant

Several authors have recommended using NaOCl with ultrasonics (Crabb, 1982; Alaçam, 1987; Stock, 1987; Cameron, 1988b; Lilley, 1988; Nehammer and Stock, 1985; Wilcox, 1989). Crabb (1982) suggested using 5% NaOCl, Nehammer and Stock (1985) recommended a 2% concentration, while Cameron (1987; 1988b) indicated that 2% NaOCl was the optimal concentration for removal of the smear layer. Cameron (1987a) suggested that "ultrasound did not seem to alter the mode of action of NaOCl, but merely speeded up the reaction, so that one minute of sodium

hypochlorite activated by ultrasound was as effective as 3 minutes of static sodium hypochlorite".

Observations have been made that NaOCl is superior to water in removal of smear layer (Ahmad, Pitt Ford and Crum, 1987), debris (Griffiths and Stock, 1986; Stock, 1987) and bacteria (Lilley, 1988). However, Krell and Dang (1987) found that neither NaOCl nor saline used with ultrasonics affected smear layer thickness.

Cameron described a technique of ultrasonic irrigation involving activation of an ultrasonic tip which had been placed in the middle one third of a prepared canal filled with NaOCl (Cameron, 1982; 1983; 1986; 1987a; 1988b). It was speculated that removal of the loosely attached component of the smear layer, leaving the dentinal tubules closed, would ultimately assist in adherence of root canal sealer to the canal wall (Cameron, 1983). Cameron (1983) postulated that this could be achieved by reducing the time allotted to removal of most of the smear layer from three minutes to one minute. In contrast, to debride immature root canals, ultrasonic activation of 4% NaOCl for three minutes was recommended (Cameron, 1987a).

Using SEM, Alaçam (1987) compared the effects of various irrigating systems and irrigants on straight canals, observing that a combination of three minutes of ultrasound with 5% NaOCl resulted in canals entirely free of smear layer and debris. In contrast, Ciucchi, Khettabi and Holz (1989) found ultrasonic irrigation to be of

little value in apical debridement of curved canals.

Reports exist that using NaOCl with ultrasonic instrumentation produces a fine spray which some operators have found irritating to the eyes (Cameron, 1982; Tronstad et al., 1985). Griffiths and Stock (1986) suggested that a disadvantage of NaOCl with ultrasonics was the need for the patient to be protected with an apron.

The combination of 15% EDTA with ultrasound was investigated by Ciucchi, Khettabi and Holz (1989), who found no significant difference in the ability of EDTA to remove the smear layer, with or without ultrasound. However, it was noted, that because the efficacy of EDTA is related to the duration of its exposure, the 3 minute exposure time allowed by the investigation might not have allowed chelation to occur. The authors speculated that using EDTA and ultrasound could even produce a new smear layer, from demineralised dentine combining with an oscillating file.

The utilisation of ultrasonics with either chloroform or NaOCl in final reinstrumentation during removal of gutta percha/sealer, has been suggested by Wilcox (1989).

CHAPTER 9

CYTOTOXICITY OF IRRIGANTS

9.1 INTRODUCTION

At the Fifth International Conference on Endodontics in 1973, Trowbridge commented that "unfortunately, many effective antimicrobial agents are quite toxic and inflict damage on the tissues of the host at the same time that they are destroying invading bacteria".

It is understood that chemicals and drugs cause damage by acting on some vital cellular function, such as membrane permeability, osmotic homeostasis or the integrity of an enzyme or cofactor (Robbins, Angell and Kumar, 1981 p6). Torneck (1961) described seven factors that could influence tissue reactions to solutions used during endodontic treatment:-

- (1) the type, concentration and physical form of the drug;
- (2) the volume of drug;
- (3) the manner in which the drug is placed and sealed in the canal;
- (4) the size of the apical foramen;
- (5) the histologic status of the periodontium;
- (6) the length of time the tissues are exposed to the drug;

(7) the individual patient's susceptibility to injury.

During endodontic treatment, irrigants can contact dentine, cementum, bone, periodontal ligament and associated cells. Following endodontic treatment, changes which occur at the apex might be a response to the materials employed, chemomechanical instrumentation, residual microorganisms or the variable host response (Murphy, 1988). This chapter will discuss cell and tissue reaction research of the solutions recommended as root canal irrigants.

9.2 ASSESSMENT OF CYTOTOXICITY

It has been remarked "that any dental material can be shown to be toxic or non-toxic, depending on the test conditions" (Hensten-Pettersen, 1988). Hensten-Pettersen further suggested that even though this makes assessment of cell culture studies difficult, it may in fact reflect the sensitivity and variability of the clinical population.

In 1988, Murphy asserted that "the development and use of medicaments has tended to be entirely empirical with little research *in vitro* to support them." *In vitro* methods of testing provide the advantages of being cheap and rapid, and as well have the ability to be strictly controlled and theoretically highly reproducible (Browne, 1988). Spångberg (1978) felt that advantages of *in vitro* techniques included the potential for an increased level of objectivity, as well as a greater degree of control

over the experimental situation.

In contrast, *in vivo* animal studies present practical limitations, introduce many biological variables, and are expensive, in addition to being socially questionable. Further, the results of non-human *in vivo* studies may not necessarily reflect human responses. Spångberg (1978) believed the use of *in vivo* implantation techniques were "poorly suited for the evaluation of medicaments because the immediate reaction to medicaments is most important".

Early *in vivo* evaluation of irrigants involved histologic examination of tissues following implantation (Coolidge, 1932; Torneck, 1961). A major inconsistency arising from this approach included the inability to ascertain whether the inflammation was caused by surgical trauma, or by the actual irrigant. As well, difficulties arose in attempting to quantify and interpret inflammation histologically. More recently, another method of evaluation *in vivo* was developed, which involved examining enhanced vascular permeability of dye following administration of test solutions (Rutberg, Spångberg and Spångberg, 1977). Despite limitations imposed by the methodology, the authors believed this technique provided a useful means of assessing cytotoxicity.

The use of HeLa cells (human epithelial cells isolated from cancer tissue) for *in vitro* examination of cytotoxicity of endodontic irrigants was first reported

by Engström and Spångberg (1967; 1967a). Previously this technique had been utilised in assessment of endodontic filling materials (Rappaport et al., 1964).

Cultivated cell lines of fibroblasts and lymphoblasts have since been used to study cytotoxicity of irrigants (Koskinen, Rahkamo and Tuompo, 1981). Spångberg (1978) postulated that fibroblasts, regardless of their origin, rapidly dedifferentiated in culture and thus "no significant difference" existed between, for example, pulp fibroblasts and skin fibroblasts when used in investigations evaluating toxicity.

In vitro tests assessing the cytotoxicity of irrigants include the millipore filter (Wennberg, 1988) and the chromium release (Spångberg, Engström and Langeland, 1973) methods. Cytotoxic effects are monitored by parameters such as inhibition of cell growth, cytolysis, changes in membrane permeability and alterations in metabolic activity (Hensten-Pettersen, 1988). Trowbridge (1973) commented that, in general, cells with a high metabolic rate and high oxygen requirement, for example, odontoblasts, were likely to be more susceptible to injury by toxic chemicals than cells with a low metabolic rate, for example, undifferentiated mesenchymal cells.

It would appear that *in vitro* tests are extremely sensitive, perhaps because greater contact can be obtained between solution and test cells, which might not reflect the clinical situation. The influence of

different stages of the cell cycle might also introduce other variables. In 1988, Spångberg and Pascon demonstrated that variations in material preparation for cytotoxicity tests achieved differing results, underlining the extreme sensitivity of these procedures to experimental method.

In vitro assay systems are susceptible to artifacts (Browne, 1988). Many culture methods for *in vitro* studies use foetal or neonatal calf serum as a culture medium, a solution which is likely to vary in composition according to batch, and which has not been fully defined chemically (Browne, 1988).

Further, despite the existence of official documents on cell culture methods (ADA 1979, BSI 1980, FDI 1980, ISO 1984, AFNOR 1985, DIN 1986), the basis for selection of cytotoxicity tests is not clear. However, Hensten-Pettersen (1988) asserted that, while variations exist in sensitivity between cell types when two or more cell type assay systems are used, "materials which are markedly toxic are toxic in all systems".

Results of *in vitro* and *in vivo* studies are often not comparable. *In vitro* studies using single cell types do not have access to the "normal" *in vivo* metabolic, inflammatory and immunological responses. In addition, a problem which exists when comparing the results of different studies is on determining the identity and concentration of substances to which the cells were exposed. Time, temperature, surface area, growth media

and mode of cell-material contact also vary (Hensten-Pettersen, 1988).

Walton and Torabinejad (1989 p206) commented that "the difference between *in vitro* and *in vivo* studies may be that in the clinical situation the irrigating solutions have very limited surface contact and also may be quickly buffered and neutralized. In the canal, irrigants would not have extensive and intimate contact with all areas of tissue".

It is also important to note that irrigants may only be potentially cytotoxic when introduced beyond the apex, and not if they are confined within the canal. Although the dynamics of fluid movement from lumen to the periodontal ligament are not well understood, a dramatic reduction in the concentration of solutions would help explain differences between *in vitro* and *in vivo* studies (Abbott, Heithersay and Hume, 1988; Abbott, Hume and Heithersay, 1989). This might be clarified by further investigations into diffusion across radicular dentine.

Ingle et al. (1985a p233), while reviewing cytotoxicity tests on endodontic sealers, made a comment also applicable to the examination of irrigants - "one is struck by the sad conclusion that all the products tested are tissue toxic, and it is only a matter of degree - which is the most and which the least".

9.3 EARLY STUDIES

Coolidge (1932) examined the effects of 4%

chloramine-T and 5% NaOCl on healthy periapical connective tissue following vital pulp removal and sealing of medicaments in the root canals of dogs. While NaOCl was found to produce little effect on the periapical tissue, chloramine-T resulted in an extensive leukocytic infiltration.

Schilder and Amsterdam (1959) investigated the potential of solutions such as NaOCl, hydrogen peroxide, Zephiran chloride, and Azochloramid to induce inflammation in rabbit eyes and abdominal integument. Azochloramid produced a severe inflammatory reaction three to four hours after application, which was still present one day later, while application of NaOCl and hydrogen peroxide generated moderate inflammation that continued into the next day. Zephiran chloride induced slight inflammation with complete resolution one day later. Abdominal integument reactions to NaOCl and Azochloramid were severe after one day, while hydrogen peroxide and Zephiran chloride produced no inflammatory response.

Östby (1957) performed histological investigations on the effects of EDTA on periapical tissues. One to fourteen months after completion of endodontic treatment utilising EDTA, teeth and their "surrounding structures" were removed surgically, sectioned and examined. The author stated that "EDTA had caused no deleterious effect on the periapical tissues". In a later study (1961), Östby discussed the ability of EDTA to act as an

anticoagulant, by chelation of calcium from the blood, thereby interfering with the clotting process. However, he considered the effect of EDTA on the periapical tissues to be insignificant.

Torneck (1961) noted that normal healing of hamster tissue occurred after implantation of EDTAC. However, Patterson (1963) observed extreme tissue irritation following implantation and intramuscular injections of 15% EDTA during cytotoxicity studies on rats. Intramuscular injections of 10% EDTA into albino rats produced a severe ulcerating inflammatory reaction. Differences in methodology could explain this disparity.

Patterson (1963) carried out a clinical trial using 1 ml of 10% EDTA at each instrument change. Assessment of cytotoxicity was made according to the incidence of clinical symptoms and status of bacteriological cultures. Clinically, EDTA was found to induce no deleterious effects.

A clinical and histological study on dogs and guinea pigs investigated various irrigants, including 3% hydrogen peroxide, 15% EDTA, chloramine-T and 5% NaOCl (Attala and Calvert, 1969). One drop of solution was deposited into the conjunctival sac of dogs' eyes. It was found that EDTA and chloramine-T produced an inflammatory response still present five days later, but not as severe as that produced by either 3% hydrogen peroxide or NaOCl. Five days after injection of 0.2 ml of NaOCl or EDTA into guinea pig abdominal subcutaneous connective

tissue, histological observation showed signs of severe inflammation, while chloramine-T and 3% hydrogen peroxide produced a mild inflammatory response.

9.4 IN VITRO STUDIES

A number of studies have investigated the effects of various irrigants on cell cultures (Engström and Spängberg, 1967; 1967a; Spängberg, Engström and Langeland, 1973; Wennberg, 1980; Koskinen, Rahkamo and Tuompo, 1981; Massillamoni, Kettering and Torabinejad, 1981).

Using the cytotoxic effect of phenol on HeLa cells as a reference, a study by Engström and Spängberg (1967) found 1% Biosept, to be more cytotoxic than 0.8% phenol. Other solutions, such as 5% chloramine-T, Dakin's solution, and 0.1% Biosept, were found to be less cytotoxic. Using the same techniques, the cytotoxic effects of EDTA and 50% sulphuric acid were also investigated in a later study (Engström and Spängberg, 1967a). EDTA was found to be four times less toxic than sulphuric acid. However, a variety of growth conditions and concentrations were used in these studies, and unsupported assumptions were made about HeLa cells. Engström and Spängberg (1969) later recommended Dakin's solution as an effective antibacterial agent with low toxicity.

Spängberg, Engström and Langeland (1973) investigated the toxicity of solutions used during root

canal therapy on ^{51}Cr labelled L-cells and HeLa cells. Methodology was based on a technique described by Spångberg (1973). Solutions tested included:- (1) 0.05% chlorhexidine; (2) 0.08% Iodopax; (3) 5% and 0.5% NaOCl; and (4) 0.45% Wescodyne. Test tubes of cells and solutions were incubated for periods of 4 and 24 hours at 37°C (5% NaOCl for four hours only). Assessment of toxicity was made by registering the amount of ^{51}Cr released during cell lysis. The antimicrobial effectiveness of the solutions on *Staphylococcus pyogenes aureus*, *Streptococcus faecalis*, *Pseudomonas aeruginosa* and *candida* was also measured. After four hours it was found that the toxic effects of 5% NaOCl and chlorhexidine were much greater than their antimicrobial effects. The recommendation was made that 0.5% NaOCl would be the solution most likely to combine a reasonably high antimicrobial effect with a low toxicity.

An evaluation of the ^{51}Cr release method as a screening test for a number of endodontic irrigants was made by Koskinen, Rahkamo and Tuompo (1981). Whereas Spångberg (1973) had used 70% ^{51}Cr release as an indicator of total cell lysis, this study adopted a "more cautious" approach by using 50% ^{51}Cr release as an indicator. Cell types used were human lymphoblasts and fibroblasts, the latter being found to have a better average tolerance to the test solutions than lymphoblasts. However, 5% NaOCl was found to be a "potent cell killer even at dilutions of 1:1000-1:10,000", and

lymphoblasts appeared to exhibit greater tolerance than fibroblasts. Interestingly, at concentrations exceeding 0.2%, chlorhexidine was observed to cause precipitation of the cell-free growth medium, Ham's F-10 medium. These precipitates were found to contain protein. The authors concluded that while this test was extremely sensitive, both unforeseen cell-dependent variations and methodologic errors occurred, which reduced its suitability as a sole method of testing cytotoxicity of solutions.

Recovery from damage induced by antiseptics was investigated by Wennberg (1980). The response of HeLa cells exposed to test solutions for one hour was examined. The test solutions were (1) 0.04% Iodopax; (2) 0.1% Biosept; (3) 0.1% chlorhexidine; (4) 5% chloramine-T; and (5) 0.5% NaOCl. Responses were examined either immediately or after four hours maintenance in growth medium by registration of tritium-labelled thymidine uptake, an indicator of the rate of cellular DNA synthesis. Chloramine-T was found to be the most toxic solution, with the remaining antiseptics displaying similar toxicity. The four hour "recovery" period indicated a greater ability of the cells to overcome damage induced by Iodopax, chloramine-T and NaOCl, than by chlorhexidine or Biosept.

Massillamoni, Kettering and Torabinejad (1981) exposed human lung fibroblasts to various concentrations of solutions for 18 to 24 hours, after which H^3 -thymidine

was added. It was postulated that cells not killed by the cytotoxic action of the medicament, would incorporate the labelled thymidine. In addition, the antibacterial effect of the solutions was tested using three different microorganisms (*Streptococcus mitis*, *Streptococcus faecalis* and *Staphylococcus aureus*) by scoring either growth or no growth. From this data the authors established a "biocompatibility index" which represented "the degree of non-toxicity at the greatest dilution of medicament which is still capable of preventing growth of test bacteria". Data was not statistically analysed, but indicated that 17% EDTA was likely to be a more tissue compatible irrigant than 5.25% NaOCl. However, the authors suggested that "currently used concentrations" of irrigants were cytotoxic, and needed to be diluted for clinical use.

9.5 IN VIVO STUDIES

Rutberg, Spångberg and Spångberg (1977) used Evan's blue dye, which purportedly binds plasma proteins, to assess the inflammatory potential of endodontic irrigants in enhanced vascular permeability tests. The solutions studied were:- (1) EDTAC; (2) 0.1% Zephiran chloride; (3) 1% NaOCl; and (4) 0.04% Iodopax, and were delivered via intradermal injection into the backs of Sprague-Dawley rats. The dye was injected one hour later and could be observed around the site of injection to varying degrees depending on the irrigant. The greatest irritant effect

was found to be produced by EDTAC, followed by Zephiran chloride and 1% NaOCl. The least irritating by far, was the iodophor, Iodopax.

Wennberg (1980) inserted teflon implants into thigh muscle tissue of rabbits. Six weeks later the resultant indentations were subjected to various test solutions for 15 minutes. Test solutions were (1) 0.04% Iodopax; (2) 0.1% Biosept; (3) 0.1% chlorhexidine; (4) 5% chloramine-T; and (5) 0.5% NaOCl. Tissue sections were then examined microscopically for evidence of inhibition of succinate dehydrogenase¹ activity. No difference was found between solutions after a 15 minute contact period, with the exception of chloramine-T, which produced a more severe zone of inhibition. After 45 minutes, the best tissue recovery was observed in tissue subjected to chlorhexidine and Biosept.

Martin (1978) applied 2% acid glutaraldehyde and 5.25% NaOCl to rabbit's eyes. After deposition of one drop of 2% acid glutaraldehyde, a severe necrotising conjunctivitis was observed histologically 72 hours later. As part of the same study, injection of 0.05 ml of 2% acid glutaraldehyde into subcutaneous connective tissue of rats resulted in a severe and extensive necrotic reaction, presumed to be due to the fixative nature of the solution. Less severe reactions were

1. This is an enzyme involved in the oxidation of succinate to fumarate during the final stages of the citric acid cycle (Stryer, 1981 p287).

observed following use of 1% acid glutaraldehyde and 5.25% NaOCl.

After intraperitoneal injections of EDTA into rats (30mg/100mg body weight dose - a seemingly very large amount), increased numbers of osteoclasts were noted in trabecular rib bone after 10 minutes. After three hours, numbers returned to normal (Hedlund, Hulth and Johnell, 1982).

Lamers, van Mullem and Simon (1980) mechanically prepared 61 anterior teeth of 21 *Macaca mulatta* monkeys using copious irrigation with either sterile physiological saline or 1% NaOCl. Access cavities were then sealed with Cavit. After 2, 7 and 42 days, the animals were killed, and blocks of bone containing teeth *in situ* were dissected and subsequently examined histologically for evidence of inflammation. The periapical response after all periods was mild and mainly resorptive, with macrophages being the predominate cell type. At 42 days, a reparative reaction was observed where resorption had occurred. No statistical difference in the induction of noxious effects was found between saline and 1% NaOCl.

Another study used 5% NaOCl during endodontic preparation of incisors and premolars of beagle dogs, which were killed 1 to 30 days after treatment. The periapical tissues around the treated teeth were then evaluated histologically for the presence or absence of inflammation (Hwang et al., 1980). No significant

difference in the incidence of inflammation was found between normal saline or 5% NaOCl.

Polyethylene tubes containing sterile physiological saline, and different concentrations (0.9%, 2.1%, 4.1% and 8.4% (all pH 12)) and volumes (0.005 ml or 0.02 ml) of NaOCl, were implanted in the backs of guinea pigs (Thé, Maltha and Plasschaert, 1980). At either seven or fourteen days, the animals were killed, and sections of tissue surrounding the implants were examined microscopically for evidence of inflammation. While volume appeared to have no effect, there were differences between the two time periods. After seven days moderate reactions were observed regardless of concentration, but after fourteen days the response was reduced. The authors concluded that all the tested concentrations of NaOCl were well tolerated. From this data, it was extrapolated that decisions concerning irrigant concentration of NaOCl should be based on solvent and bactericidal requirements, and not on toxicity.

In contrast, Pashley et al. (1985) stated that "even the suggestion that sodium hypochlorite, at some dilution will only affect necrotic tissue should be abandoned". Pashley et al. (1985) splashed 5.25% and 0.52% NaOCl into rabbit's eyes, and observed moderately severe conjunctival palpebral oedema and hyperaemia within 30 minutes, which peaked in severity after two hours. Intradermal injections into rats induced immediate haemorrhage, while elevation of NaOCl concentration

increased haemolysis of red blood cells. At a 1:1000 dilution of 5.25% NaOCl, total haemolysis of red blood cells occurred. Pashley et al. issued a strong warning that NaOCl was a very caustic, nonspecific agent, whose action was not limited to necrotic tissue, but was cytotoxic to all cells except heavily keratinised epithelia.

Others have indicated that prolonged ingestion of various concentrations of NaOCl was not carcinogenic in F344 rats and B6C3F1 mice (Hasegawa et al., 1986; Kurokawa et al., 1986).

9.6 CLINICAL STUDIES

Any irrigant used during endodontic therapy may reach the periapical area and thus be potentially toxic to the periapical connective tissue. While it is desirable to restrict irrigants to the confines of the root canal, this does not always occur (Schilder and Amsterdam, 1959). A study using autoradiographic tracings showed that Ca^{45} -labelled EDTA was deposited in the periapical area following application of EDTA and irrigation by saline, although details of canal preparation were not included (Nicholson et al., 1968).

Svec and Harrison (1977) stated that there was no evidence that 5.25% NaOCl had any effect on post-operative discomfort. Using the hypothesis that a toxic irrigant should be associated with interappointment pain, a clinical trial was conducted to determine the

relationship between interappointment pain, and irrigant type (Harrison, Svec and Baumgartner, 1978). The irrigants included in the study were:- (1) saline; (2) 5.25% NaOCl; and (3) 3% hydrogen peroxide/5.25% NaOCl. While no relationship was found between interappointment pain and the types of irrigants used during mechanical preparation, this study relied upon patient reports, and interpretations of "pain" may have varied widely.

A later study used similar methods to determine the effect of the combination of irrigants and intracanal medicaments on interappointment pain (Harrison, Baumgartner and Zielke, 1981). The irrigation procedure used 0.5 ml 3% hydrogen peroxide followed by 0.5 ml 5.25% NaOCl at each instrument change. The authors concluded that 5.25% NaOCl alone, in combination with 3% hydrogen peroxide, or when used with intracanal medicaments, was not associated with interappointment pain.

9.7 OTHER STUDIES

Investigations by Baumgartner and Ibay (1987) examined the chemical reactions occurring when 5.25% NaOCl was mixed with other root canal irrigating solutions (3% hydrogen peroxide, 50% citric acid, 15% EDTA). The authors were concerned about the potentially

hazardous effects of inhalation of chlorine gas¹ by operators using combinations of irrigants. Significantly more chlorine gas was evolved when NaOCl was mixed with citric acid (3.5 ppm) than with EDTA (<0.5 ppm). NaOCl combined with hydrogen peroxide produced no detectable concentration of chlorine gas. The authors therefore recommended using EDTA rather than citric acid with NaOCl.

Reports of allergic hand dermatitis following contact with 1% NaOCl have appeared in the literature (Habets et al., 1986; Van Joost et al., 1987). It would appear that the use of gloves when handling this solution is advisable.

9.8 ACCIDENTS

A number of case reports describing misuse of irrigants are found in the literature. While some describe the events following the mishandling of hydrogen peroxide (Shovelton, 1957; Bhat, 1974; Kaufman, 1981), more recent reports have concerned NaOCl (Barbas et al., 1987; Reeh and Messer, 1989; Sabala and Powell, 1989).

A warning to avoid using hydrogen peroxide under pressure, without availability of an escape vent, was made by Bhat (1974), who described a case in which a

1. An environmental concentration of 3.5 ppm of chlorine was identified as the concentration at which an odour could be detected, 4 ppm the maximum concentration breathable for one hour without incurring damage and 15 ppm the level at which throat irritation occurred (Baumgartner and Ibay, 1987).

lateral perforation was irrigated under pressure with hydrogen peroxide. Sudden pain and swelling occurred, which was considered to result from hydrogen peroxide coming into contact with blood and tissue protein, resulting in rapid liberation of oxygen.

Another case report described the sequelae of hydrogen peroxide being forced through the apex during routine endodontic treatment (Kaufman, 1981). The resulting emphysema lasted for three days, and was accompanied by sharp pain during the first day, which lasted for several hours. More recently, Patterson and McLundie (1989) reported a similar event with swelling not resolving until twelve days after the event.

After accidental introduction of 0.5 ml of 5.25% NaOCl through the apical foramen, immediate severe pain, swelling and haematoma occurred (Becker, Cohen and Borer, 1974). One month elapsed before complete healing occurred. Similar events have been described recently (Reeh and Messer, 1989; Sabala and Powell, 1989). Reeh and Messer (1989) further reported long-term paraesthesia after introduction of 5.25% NaOCl through a midroot perforation.

Walker (1975) discouraged the technique of utilising a used local anaesthesia cartridge as a vehicle for irrigants, because of the potential danger of mistakenly injecting the irrigant as a local anaesthetic. Herrmann and Heicht (1979) recounted a case where a patient accidentally received 1.8 ml of 5.25% NaOCl

instead of local anaesthesia, for a mandibular block injection. Severe pain and extensive swelling resulted in the necessity for the patient's admittance to an intensive care unit for observation of an airway obstruction. After two weeks, complete resolution was obtained, following treatment for pain and prevention of infection.

Another case report, based on circumstantial evidence, described a fatality following endodontic treatment to a right maxillary tooth during which NaOCl had been used as an irrigant (Barbas et al., 1987). The report listed a chain of events leading to a fatal intracerebral haemorrhage - "sudden unexpected fifth nerve stimulation caused by hypochlorite, pain, elevation of blood pressure, and haemorrhage from unprotected arterioles in the left cerebral hemisphere".

CHAPTER 10

PRACTICAL ASPECTS

10.1 INTRODUCTION

The process of irrigation is performed to remove from the root canal a diverse mixture of debris, which might include microorganisms, dentinal shavings, blood, pus, exudate and arguably, smear layer. This process is influenced by factors such as limited surface contact, volume of solution and solution exchange (Senia, Marshall and Rosen, 1971).

Irrigation can only be effective when the irrigant reaches the apex, and creates a current or force to carry away debris (Chow, 1983). Walton and Torabinejad (1989 p205) suggested that irrigant delivery was possibly more important than irrigating solution *per se*.

During the establishment and development of endodontics the need to irrigate has been consistently advocated by many authorities, including Buckley, 1911 p315; Grossman, 1943; Godwin, 1957; Sampeck, 1967; Schilder, 1974; Moser and Heuer, 1982; Ingle et al., 1985 p179; and Walton and Torabinejad, 1989 p205.

In 1943 Grossman lamented the reliance by the clinician upon drug therapy, rather than upon removal of debris using irrigants. "Mechanical instrumentation should be followed by irrigation of the canal in order to

wash out fragments of pulp tissue and dentinal shavings" (Grossman, 1943). However, the difficulty in clinically evaluating irrigation was noted by Godwin (1957), who commented that "drainage which appears clear to the naked eye, may contain particles of microscopic size".

Sampeck (1967) believed that "irrigation of the canals will allow the solution to dissolve whatever debris cannot be reached with the hand instruments. Filings have a tendency to float and can readily be removed. Tissue debris, which may block a dry canal, will be loosened by the wetting agent, allowing the file to pass." Luebke (1967) described irrigation as "lavage of the pulp chamber and root canals", and recommended its frequent use during mechanical preparation.

Grossman (1969) maintained that keeping the canal moist with irrigant decreased the chance of instrument breakage. Schilder (1974) urged that "dentin shavings must be kept in loose suspension in the irrigating solution to prevent accumulation and compaction of dentin mud".

Morse (1974 p456) declared that it was "better to over-irrigate than under-irrigate", while Salzgeber and Brilliant (1977) believed that to ensure maximum debridement efficacy, the irrigant solution "should be exchanged throughout the canal with new solution".

Schroeder (1981) contended that irrigation would float up and rinse out necrotic debris and for this reason dry instrumentation of necrotic canals was to be

avoided. Moser and Heuer (1982) maintained that "the objective of irrigation is to remove the debris that cannot be removed by instrumentation".

Current texts support the view that thorough and copious flushing at each instrument change, with appropriate irrigants, is an essential part of chemomechanical preparation (Ingle et al., 1985 p179; Gutmann and Dumsha, 1987 p157; Walton and Torabinejad, 1989 p205). Grossman, Oliet and del Rio (1988 pp187-8) have recommended that canals be full of fresh solution during mechanical preparation.

In this chapter, the spectrum of working aspects associated with the use of irrigants will be examined, in addition to the various irrigant delivery systems and techniques which have been discussed in the dental literature.

10.2 ACCESS AND CANAL SIZE

The need for adequate enlargement of the root canal to allow irrigation was recognised by Grossman (1943), who postulated that unless enlarged sufficiently, only the mouth of the canal was likely to be irrigated. Leeb (1983) indicated that without adequate canal orifice enlargement, chemomechanical preparation would be restricted.

The existence of irregularities in the root canal system has been established (Skidmore and Bjorndal, 1971; Fromme and Riedel, 1972; Pineda and Kuttler, 1972).

Further, studies investigating the effect of chemomechanical preparation on root canals have shown that portions of the canal are routinely left untouched (Gutiérrez and Garcia, 1968; Haga, 1968; Senia, Marshall and Rosen, 1971; Davis, Brayton and Goldman, 1972; Coffae and Brilliant, 1975; Klayman and Brilliant, 1975; Mizrahi, Tucker and Seltzer, 1975; Walton, 1976; Littman, 1977; Svec and Harrison, 1977; 1981; Abou-Rass and Jastrab, 1982; Sinanan, Marshall and Quinton-Cox, 1983; Montgomery, 1985; Reynolds et al., 1987; Goldman et al., 1988; Haidet et al., 1989).

However, it is not unknown to observe periapical healing despite inability to obtain ideal debridement and filling (Szajkis and Tagger, 1983), and one author has noted that removal of all the diseased tissue is not essential, but a sufficient reduction is enough for endodontic success (Seltzer, 1988 p209). In addition, Littman (1977) has suggested that the final canal shape applicable to successful endodontic treatment has not been demonstrated, and that, in fact, many obturation techniques allow conformation of gutta percha to the existing shape.

Methods of instrumentation employing step-back techniques and coronal flaring might facilitate irrigation. It has been postulated that the progressive taper produced using these techniques allows a greater degree of instrument and irrigant contact with the surfaces of the canal walls, in addition to easier access

to the apical area and an adequate coronal escape way (Schilder, 1974; Coffae and Brilliant, 1975; Hession, 1977; Ram, 1977; Bolanos and Jensen, 1980; Abou-Rass and Jastrab, 1982; Goerig, Michelich and Schultz, 1982; Weine, 1982 p321; Fairbourn, McWalter and Montgomery, 1987).

Chapman (1971) has shown that both reaming and filing are capable of transmitting contaminated material through the apical foramen. This material may contain necrotic debris, dentin, pulp fragments, microorganisms and irrigants, and could feasibly induce an inflammatory reaction (Seltzer, 1988 p260). Therefore, it would seem preferable to prevent escape of the canal contents into the periapical tissues.

Preceding apical preparation by coronal preparation, as in step-down techniques (Goerig, Michelich and Schultz, 1982), double flared techniques (Fava, 1983) and crown-down pressureless techniques (Morgan and Montgomery, 1984), might allow better penetration of irrigants, while avoiding extrusion of debris through the apical foramen.

However, Ruiz-Hubard, Gutmann and Wagner (1987) found that using the step-back technique and crown-down technique in simulated canals produced extrusion of material through the apex. Fairbourn, McWalter and Montgomery (1987) measured apically extruded debris with four preparation techniques. Ranked in order of amount of debris extruded, the techniques were (from highest to

lowest):- (1) step-back technique; (2) ultrasonic technique; (3) cervical flaring; and (4) sonic. However, the quantities were statistically significant only between techniques (1) and (4), although different volumes of irrigant (tap water) were used with each technique.

A time-motion study by Mykleby and Krell (1985) investigated various irrigants or combinations of irrigants using a step-back technique. Results indicated that no specific irrigant was able to decrease the time required for mechanical preparation.

In vitro investigations by VandeVisse and Brilliant (1975) demonstrated apical debris extrusion with irrigation, but no detectable debris extrusion from teeth mechanically prepared without irrigation. It was postulated that a file acted like a plunger during chemomechanical preparation, forcing debris through the apex, and the authors questioned whether the presence of a column of moisture in the root canal would "facilitate the extrusion of material through the apical foramen".

In spite of their results, VandeVisse and Brilliant (1975) considered that preparation of canals without irrigants was not justified, because of the risk of instrument binding and breakage in "dry" canals, in addition to a tendency for "dry" canals to become plugged with "sludge". The authors did not comment whether the presence of "sludge" during their study might have prevented the extrusion of material in dry canals.

Baker et al. (1975) estimated that approximately 70% more debris remained in canals instrumented without an irrigant. Goldman et al. (1979) showed that a 20 ml final flush was more effective in debris removal than no final flush.

Two studies which utilised a radiopaque material, Hypaque¹, to qualitatively assess irrigation efficacy, are frequently referenced (Salzgeber and Brilliant, 1977; Ram, 1977). However, in both studies there were uncontrolled variables in the methods employed. An *in vivo* study by Salzgeber and Brilliant (1977) indicated that the irrigating solution does not contact the entire length of the canal until at least a size 35 file has been used.

An *in vitro* study by Ram (1977) indicated that canals needed to be enlarged to at least size 40 before "effective irrigation" with a 25-gauge needle could occur in the apical 5 mm of the canal. It was speculated that cleansing was a function of canal diameter rather than type of irrigant, and that increasing irrigation time and pressure was not critical.

10.3 DELIVERY VOLUME

At the most fundamental level, volume determines,

1. Hypaque (Winthrop Laboratories, New York, USA), or 50% sodium diatrizoate, is a polyiodide compound commonly used in medical radiology. It has a comparable specific gravity to NaOCl, although a lower surface tension (Salzgeber and Brilliant, 1977).

in part, the difference between a medicament and an irrigant. Whereas a medicament is generally used in small quantities and left in the canal between appointments, an irrigant is used during the clinical appointment, and in greater volume.

Stewart (1948) measured the volume of root canals and was struck by "the minute volumes of drugs with which endodontists must work". As a result, he stressed the need to enlarge the canals adequately to produce a large enough canal size for placement of a sufficient volume of medicament to destroy bacteria. In a later study, the mean volume of a maxillary central incisor pulp cavity was estimated to be 12.4 mm^3 and a maxillary first molar 68.2 mm^3 (Fanibunda, 1986). The mean volume of one drop of distilled water from a 1 ml syringe, delivered by a 21 gauge needle was 12.2 mm^3 .

There is no standard volume of irrigating solution recommended for use during endodontic procedures. Obviously, the size, shape and number of canals will influence the volume of solution used. However, it is important to note that the deposition of a sufficient volume of solution at the working area of the instrument, especially in fine or tortuous canals, might not be possible (Heuer and Miserendino, 1987 p410).

Nonetheless, several articles refer to volumes of irrigants. Grossman (1960 p213) recommended using 0.5 ml irrigant at a time, up to 2.0 ml of solution, or until debris was no longer present in the used solution.

Luebke (1967) suggested that canals could be cleaned using 0.5 ml-1.0 ml of irrigant. The technique employed by Schilder (1974) averaged 39 ml NaOCl per patient visit. A pilot study by Baumgartner and Ibay (1987) showed that clinicians used up to 30 ml of irrigant(s) over a 15 minute appointment.

After discussion with several endodontists, Brown and Doran (1975) calculated that the average delivery rate of irrigating solution, using a 25-gauge needle, was 2 ml every 15 seconds. Moser and Heuer (1982) estimated that one minute was the average time required to empty a 2 ml-3 ml syringe, using "approximations of the forces used in clinical situations". However, Chow (1983) found that the average time taken by various operators to inject 5 ml of saline solution via a 23-gauge needle was 18.3 seconds.

It has been postulated that it is the flushing action of a volume of fluid which determines debris removal, and not the type of irrigant (Baker et al., 1975; Tucker, Mizrahi and Seltzer, 1976; Lehman, Bell and Gerstein, 1981).

The results of a SEM study by Baker et al. (1975), in which various irrigants were investigated, indicated that the type of irrigant was unrelated to quantity of debris removed. A greater (2.5 ml and 5.0 ml), rather than a lesser (0.5 ml and 1.0 ml), volume of irrigant resulted in less debris. In an extension of this SEM study, Tucker, Mizrahi and Seltzer (1976) observed no

difference in effectiveness of irrigation with either 2.5 ml or 5 ml of solution between each instrument change. However, in both of these studies the total volume of irrigant solution varied for each specimen. Completion of mechanical preparation was determined by the presence of clean dentine shavings.

Lehman, Bell and Gerstein (1981) found no difference in debris removal 1 mm and 3 mm from the apex, but at the 5 mm level, where canal diameter was larger and access for a greater volume of irrigant potentially possible, superior cleansing was evident under binocular microscope examination.

10.4 DELIVERY EQUIPMENT

A variety of irrigation equipment has been presented in the dental literature. Buckley (1911 p315) recommended "a good hypodermic syringe with a long straight needle for anteriors and a curved needle for posterior teeth". The armamentarium for irrigation recommended by Grossman (1943) was an all-glass 1.5 ml syringe and a 1.25 inch stainless steel 25-gauge needle with the bevel removed.

Using a suction and irrigating device the author claimed had been successful for 17 years, it was possible to flush and aspirate one litre of liquid into a root canal within ten minutes (Seidner, 1946, 1952). Liquids such as water were suggested, and for putrescent pulps a

soft soap solution was recommended.

Alternatively, a hot water and steam apparatus used electrically heated points inserted into a canal flooded with tap water. The water would then turn into steam, the author claiming that this technique would result in a "fairly sterile pulp chamber" (Davis, 1948).

Barandun and Boitel (1962) recommended a technique to treat inflammation and infectious processes, which involved the combined use of ozone¹ gas and the Barandun vacuum-suction irrigator.

Another apparatus, consisting of a handpiece to which two glass ampoules were attached, used "weakly concentrated" bactericidal solutions, preheated to a temperature of 60°C-70°C (Zerosi, 1960). An increase of 10°C was claimed to double the bactericidal effect of the solution.

A variety of needle gauges have been utilised and recommended for intracanal irrigant delivery:-

22-gauge (Heuer, 1963; Schilder and Yee, 1984 p179)

23-gauge (Svec and Harrison, 1977; Fairbourn, McWalter and Montgomery, 1987)

25-gauge (Penick and Osetek, 1970; Kahn et al., 1973; Svec and Harrison, 1981; Weine, 1982 p319; Nicholls 1984 p145)

26-gauge (Penick and Osetek, 1970; Senia, Marshall and

1. Ozone (O₃) is described as a stronger oxidising agent than O₂ (Brown and LeMay, 1985 p652).

Rosen, 1971)

27-gauge (Walton, 1976; Abou-Rass and Piccinino, 1982; Ingle et al., 1985 p184; Walton and Torabinejad, 1989 p206)

28-gauge (Bolanos and Jensen, 1980)

30-gauge (Abou-Rass and Piccinino, 1982; Chow, 1983)

The largest sized needle to reach the apex of various sized canals was investigated by Chow (1983). He estimated that a 21-gauge needle could reach the apex of a size 80 canal, a 23-gauge needle to size 50, a 25-gauge needle to size 45, and a 30-gauge needle to size 20.

Chow (1983) suggested that a stronger current was created by an irrigant delivered via a narrower than a larger gauge needle. It was hypothesised therefore, that irrigation would be most effective in large canals using narrow gauge needles, as better currents could be produced. It was also postulated that there would be more resistance to fluid leaving the canal with larger gauge needles.

Senia, Marshall and Rosen (1971) speculated that the use of NaOCl with "a needle with a very fine lumen", was not clinically practical, as crystallisation of the NaOCl solution in the lumen could occur during the course of treatment.

Although a 30-gauge needle was found to penetrate well into small canals, Druttman and Stock (1989) postulated that it was more likely than larger gauge needles to force irrigant through the apex, and they

therefore could not recommend its clinical use.

An irrigant enables the clinician to obtain a cleaner canal than would otherwise be attained (Baker et al., 1975). However, studies have indicated that needle and syringe irrigation (frequently referred to as "conventional") does not routinely clean the apical one third of the canal or curved canals (Senia, Marshall and Rosen, 1971; Baker et al., 1975; McComb, Smith and Beagrie, 1976; Ram, 1977; Ciucchi, Khettabi and Holz, 1989).

As there appears to be little flushing action beyond the tip of the needle, the extent of the effectiveness of irrigation is, in part, determined by the depth of insertion. This in turn is dependent upon needle gauge, and a small gauge needle would appear to be appropriate (Chow, 1983), although a large force is then required to push the irrigant through the needle (Moser and Heuer, 1982). However, Druttman and Stock (1989) observed that "irrigant replacement became less effective towards the apex of the canal, regardless of the method of irrigation or the needle size" .

It has been recommended that the needle be bent to an obtuse angle (Ingle et al., 1985 p184; Grossman, Oliet and del Rio, 1988 p190), or "approximately thirty degrees" (Weine, 1982 p319) to allow easier access to canals.

Gutmann and Dumsha (1987 p158) advised avoiding needle placement near the apex, because of the risk of

deposition of irrigant into the periapical tissues.

However, it is considered that the proximity of the needle to the apex plays an important role in debris removal (Brown and Doran, 1975; Abou-Rass and Piccinino, 1982; Chow, 1983).

Mullaney (1979) commented that "regardless of the type of irrigant used, the liquid cannot adequately flush the dentine chips from the canals in the smaller sizes 10 to 20." It is unlikely that conventional irrigant delivery could deposit irrigant at the apex of a severely curved canal (Sinanan, Marshall and Quinton-Fox, 1983).

As part of a study by Brown and Doran (1975), the apical 3 mm of simulated canals (apical size 30) were filled with dentine particles. With the point of a 25-gauge needle positioned at three different depths (15 mm, 10 mm and 5mm from the apex), various irrigating solutions were delivered into the canal. Efficacy of irrigation was assessed by measuring the change in particle column height. Results indicated that the proximity of the delivery needle to the debris was more critical to success in debris removal than the actual solutions used.

Several authors have stressed that the needle should not bind in the canal, otherwise irrigant solution might be forced through the apex, and the return flow of irrigant impeded (Grossman, 1943; Abou-Rass and Piccinino, 1982; Mentz, 1982; Nicholls, 1984 p145; Gutmann and Dumsha, 1987 p157; Grossman, Oliet and del

Rio, 1988 p189).

When no apical stop is present, or when the foramen opens directly into the maxillary sinus, the need to avoid needle binding is more critical (Walton and Torabinejad, 1989 p206). By ensuring careful insertion, slight withdrawal after binding, or by utilising a "slight pumping action during irrigation", it is thought that binding can be avoided (Walton and Torabinejad, 1989 p206). Nicholls (1984 p145) recommended grinding the tip of the needle to prevent it from binding to the root canal wall.

Chow (1983) observed little fluid exchange and displacement of particles beyond the tip of the needle in simulated canals. In addition, the presence of air bubbles prevented the passage of the irrigant beyond it. He therefore questioned the effectiveness of irrigation when the needle is positioned in the coronal third of the canal, and concluded that "effectiveness of irrigation appears to be a function of the depth of the needle".

Over the years, irrigating needles specifically designed for endodontic use have been investigated (Goldman et al., 1976, 1979; Drobotij et al., 1980; Sinanan, Marshall and Quinton-Fox, 1983; Heuer and Miserendino, 1987 p410).

A perforated needle (National Patent Development Corp., New York, USA) was introduced by Goldman et al. (1976). It was described as 31 mm in length, closed at one end, with ten perforations along its shaft from 2 mm

to 12 mm from the tip. The first hole was positioned not less than 2 mm from the tip. To enable the needle to be inserted to within 1 mm of the apex, as recommended by the authors, the needle was available in several gauges (24,26,28,30) and was designed to fit a Luer-Lock syringe (Burron Medical Products, Inc., Bethlehem, Penn, USA). The rationale behind a closed end was to prevent solution exiting into the periapical tissues.

Goldman et al. (1976) postulated that the perforated needle was able to develop a "hydraulic force, which, when directed laterally, forces material off the canal wall". It was claimed that this would then make larger amounts of debris available for degradation by the irrigating solution, as well as safely permit the use of high volumes of irrigants. Because the solution was able to exit in many directions along the sides of the needle further assertions were made that washing the entire 360° of the canal could occur.

In an *in vitro* investigation, Goldman et al. (1976) compared root sections irrigated with Toluidine blue dye using the perforated needle and a conventional 23-gauge needle. Comparisons were made under incident light with a dissecting microscope. Subjective analysis of results indicated that the perforated needle produced a more widespread and uniform distribution of the dye in the canal, and therefore potentially a "more complete irrigation with this technique".

In a later paper, Goldman et al. (1979) described

another *in vitro* comparison of the efficacy of the perforated needle versus a 23-gauge conventional needle. Qualitative SEM examination of irrigated canal surfaces indicated that the perforated needle resulted in accumulation of less dentine debris. However, canal diameter was variable as the final file used was dependent upon operator assessment of cleanliness.

Drobotij et al. (1980) examined the efficacy of four types of needles in delivering 5 ml saline to remove water-soluble ^{125}I -albumin gel from 50 root canals prepared to size 55. The four needles were:- (1) a 26-gauge perforated needle as described by Goldman et al. (1976); (2) a 23-gauge open-ended endodontic needle (Becton-Dickinson - manufacturer's details not provided); (3) A 26-gauge standard hypodermic needle of length one and five eighth inches, open at one end and with a tapered point; and (4) a prototype endodontic needle made by cutting off the pointed tip of a standard 26-gauge hypodermic needle, sealing the end with solder, and placing two perforations on the shaft. Remaining radioactive labelled material was measured by a gamma scintillation radiation counter. Overall, irrigation was effective in the removal of 32% to 46% of the labelled material, however, the authors indicated that there was no difference between the four needles in the removal of labelled material.

Further, Drobotij et al. (1980) observed more material in the apical 4 mm and 8 mm sections of the

canal after irrigation than before irrigation. The authors postulated that "the lateral pressure of irrigation forces ^{125}I -albumin into the dentinal tubules and accessory canals at a greater magnitude than it does into the main root canal space". Differences in needle placement depth and the method of removal of excess solution from the canals may have influenced results.

The Endovage system (Burron Medical Products. Inc. Bethlehem, Pa, USA), is designed to perform both irrigation and aspiration, and consists of a syringe to which is attached a special needle (IMAX, Stram Inc. Northbrook, Ill, USA). The needle has a side slit positioned 1 mm from the tip. It has been noted that the Endovage system, particularly when used with a narrow gauge (27, 30) needle, requires "excessive" force to empty, thus limiting its clinical usefulness (Moser and Heuer, 1982; Sinanan, Marshall and Quinton-Fox, 1983).

Moser and Heuer (1982) showed that a larger volume of solution could be delivered via a 23-gauge blunted open-ended needle (Monoject; Sherwood Industries Inc., St Louis, Mo, USA) than via a perforated needle or the Endovage system. These authors also found the most effective removal of debris, from size 100 simulated canals, occurred using irrigation with the Monoject needle inserted to working length.

In contrast, a comparison of similar needles by Sinanan, Marshall and Quinton-Fox (1983) found the Endovage system to be superior in cleansing the apical 1

mm of canals instrumented to a considerably smaller size 40. It was also observed that, while all irrigating devices cleaned equally well at levels 3 mm from the apex, many canals remained unclean.

Several authors have noted that specially designed needles have a tendency to break at the perforations (Drobotij et al., 1980; Moser and Heuer, 1982; Sinanan, Marshall and Quinton-Fox, 1983).

Grossman, Oliet and del Rio (1988 p190) also commented that the perforated needle bent out of shape, and they endorsed instead a "notched needle" (Monoject-Winthrop Laboratories, New York) claiming that its use "lessens the pressure from the forceful ejection of the irrigating solution". Heuer and Miserendino (1987 p410) recommended a specially designed disposable irrigating needle (details of manufacturer not provided) which was described as "blunted and slit for 4 to 5 mm along one side from the tip toward the hub to provide an escape for fluids should the needle inadvertently bind in a canal".

10.5 IRRIGATION TECHNIQUES

Penick and Osetek (1970) described an irrigation technique for use with NaOCl: "A 2.5 cc sterile syringe with a blunted 25 or 26-gauge needle is filled with NaOCl, and the needle is placed *loosely* in the canal. The solution is then deposited *very slowly*. Extreme care must be exercised to avoid forcing this caustic material into the delicate periapical tissue. If the needle fits

loosely in the canal without binding and if the solution is deposited slowly, there is little likelihood that the periapical tissues will be embarrassed." This procedure needed to be repeated "at least as often as instruments are changed".

Most authors in current texts advocate this "conventional" technique or slight variations of it, using either plastic or glass syringes (Nicholls, 1984 p145; Ingle et al., 1985 p184; Heuer and Miserendino, 1987 p410; Grossman, Oliet and del Rio, 1988 p189; Walton and Torabinejad, 1989 p206).

However, while syringe and needle are most frequently used, alternative irrigation techniques have been described (Sampeck, 1967; Thé, 1979a; Mentz, 1982; Weine, 1982 p319).

Sampeck (1967) expressed concern that debris could be forced into apical tissue using conventional irrigation techniques. Therefore, particularly for maxillary teeth and with the use of NaOCl, Sampeck suggested placement of a NaOCl-saturated paper point in the canal. It was postulated that the paper point acted as a "wick" to carry the solution to the apical areas of the canal.

Thé (1979a) believed that a cotton-wrapped file soaked in NaOCl solution was advantageous over syringe and needle irrigation. Smaller volumes of solution were required, there was no danger of penetrating the apical foramen, and application of suction was not required. As

well, he considered that the pressure of the cotton against the walls allowed better mechanical cleaning.

Weine (1982 p319) believed that it was the action of the endodontic instruments, and not the injection process, that distributed the irrigant to the "nooks and crannies of the canal". For posterior teeth and narrow canals, he suggested placement of the solution in the pulp chamber and distribution of the irrigant by files, claiming the "capillary action of the narrow canal diameter will retain much of the solution". Mentz (1982) recommended this technique for narrow canals in particular. Sommer, Ostrander and Crowley (1966 p548) speculated that when this technique was used with NaOCl, organic material would be "drawn out of the canal in a semi-gelatinous state".

Druttman and Stock (1989) also recommended frequent mixing of canal contents with files. The authors speculated that this allowed improved irrigant replacement, which they indicated was a more important factor than "copious irrigation" when removing dye from simulated canals.

Injection of irrigant solution under pressure is to be avoided. Slow injection of solution may circumvent extrusion of solution and debris through the apex into the periapical tissue (Grossman, 1943; Mentz, 1982; Nicholls, 1984 p145; Heuer and Miserendino, 1987 p410).

A criterion for completion of debridement has traditionally included the presence of clean irrigating

solution (Grossman, 1960 p213; Baker et al., 1975), however Walton and Torabinejad (1989 p198) have commented that this is inaccurate and can only serve "as a crude indicator".

10.6 IRRIGANT REMOVAL

It is important to remove irrigant solution from the canal prior to obturation or placement of a medicament (Grossman, 1943; Sommer, Ostrander and Crowley, 1966 p548; Nicholls, 1984 p145). Grossman, Oliet and del Rio (1988 p190) described three stages of drying the root canal:- (1) catching return flow of solution on gauze sponge or by aspiration; (2) aspirating the residual solution by withdrawal of the plunger of the syringe while the needle is still in the canal; and (3) final drying with absorbent paper points.

Devices and techniques have been described to aid in the removal of irrigant. These methods range from attaching a drainage tube to a modified rubber dam frame (Godwin, 1957) or to a saliva ejector (Hill, 1959; Kahn et al., 1973), to the use of an aspirating tip (Penick and Osetek, 1970; Weine 1982 p320), or cotton rolls and gauze (Grossman, 1943; Curson, 1966; Penick and Osetek, 1970). Complicated and expensive aspirating machinery has also been described (Rowe, 1976). Residual solution can additionally be removed with absorbent sterile paper points (Grossman, 1943; Weine, 1982 p320; Grossman, Oliet and del Rio, 1988 p190). Irrigation with alcohol to

dessicate the canal and "remove traces of other chemicals" has been suggested by Walton and Torabinejad (1989 p208).

The use of compressed air to remove excess irrigant is not recommended. A side effect of blowing compressed air into a canal was documented following the "embarrassment" experienced by one dentist when this approach caused emphysema around the patient's eye, which then took two weeks to subside (Salvas, 1931). In an animal study it was shown that prolonged application of pressurised air into the root canals of dogs, resulted in the death of four of the seven dogs from air embolisms in the right ventricles (Rickles and Joshi, 1963). In this paper, the authors also referred to a possible death following compressed air use during root canal therapy. Other papers have also reported adverse effects following use of compressed air to dry canals (Shovelton, 1957; Pearson, 1958; Harris, 1971).

10.7 CORROSIVE EFFECTS OF IRRIGANTS ON INSTRUMENTS

Mueller (1982) defined corrosion as the "deterioration of a metal by chemical or an electrochemical reaction with its environment". Intracanal instruments are repeatedly exposed to irrigants, and the ability of irrigants to corrode instruments has been long recognised (Hays, 1900; Heling, Shapiro and Sciaky, 1965; Grossman, 1969). In addition, damage to instruments stored in a high humidity saline

environment has been reported (Aker, Serowski and Bailey, 1981).

Carbon steel files are composed of more than 99% iron (Mueller, 1982). Eichner et al. (1976) postulated that the corrosion products of carbon steel instruments were likely to be oxides of iron, and expressed concern that these products may be deposited in the canal and periapical area.

The deterioration of carbon steel endodontic instruments during mechanical preparation was investigated by Gutiérrez, Gigoux and Sanhueza (1969). Following exposure to an unspecified concentration of NaOCl (pH 9.4) for an average time of 18 seconds, "rust blisters" on Kerr reamers were evident under a binocular microscope. To produce a similar oxidative effect, hydrogen peroxide (pH 3.36) required six minutes and EDTA (pH 7.3) 56 minutes. Three hours contact with NaOCl resulted in complete destruction of reamer flutes.

Partly for this reason, the routine use of stainless steel files, in preference to carbon steel files, has been recommended (Grossman, 1969; Eichner et al. 1976).

Eichner et al. (1976) examined the effects of a ten minute exposure of 1% and 5% NaOCl (with and without purified bovine collagen) on carbon and stainless steel endodontic instruments (Kerr Manufacturing Co., Romulus, Michigan, USA). Neither type of instrument displayed evidence of corrosion when immersed in the control water

(with and without collagen). Stainless steel instruments did not exhibit any corrosion when subjected to NaOCl, with or without collagen. In contrast, carbon steel instruments showed evidence of corrosion when exposed to 1% NaOCl with collagen (light, uniform coating of corrosion), 5% NaOCl (small amounts of corrosion randomly distributed) and 5% NaOCl with collagen (heavy, uniform coating of corrosion).

Oliet and Sorin (1978) observed the corrosive effects of 5% NaOCl with or without "crushed-tooth material" on carbon steel instruments, which was not evident at x20 magnification on stainless steel instruments. It was found that most corrosion took place within the first five minutes and increased with higher temperature. As well, corrosion was increased in the presence of crushed tooth material, though the mechanism for this was not proposed.

Gallegos and Bertolotti (1981) considered that it was "very advantageous that inadvertently broken instruments, lodged in a canal, can be more readily rusted away if made of carbon steel than of stainless steel". These authors studied the effects of 2.5% NaOCl on the fracture strength of carbon steel instruments over various time periods. Despite observing visible corrosion after 15 minutes, the authors concluded that immersions of carbon steel instruments in 2.5% NaOCl would not affect instrument strength up to 8 hours immersion. Further claims were made that most of the corrosion did

not occur on instrument surface and cutting edges, however, this was not substantiated or examined. The authors concluded that carbon steel instruments should not be discarded because of the presence of corrosion products.

In attempts to inhibit corrosion of carbon steel instruments, proposed corrosion inhibitors such as benzotriazole, sodium silicate, sodium benzoate, mercaptobenzothiazole and gelatin were added to NaOCl solutions (Oliet and Sorin, 1978). Addition of benzotriazole to NaOCl resulted in inhibition of corrosion. When a surfactant (sodium alkyl arylsulfonate) was added to NaOCl, increased corrosion occurred. However, addition of both benzotriazole and the surfactant, provided the most effective inhibition. No "inhibitor" was able to offer "complete protection" against corrosion.

In contrast, Mueller (1982) found that corrosion of carbon steel instruments was effectively inhibited when 3% sodium silicate was added to NaOCl, saline and EDTA-Na solutions. The author claimed no toxicological problems would be presented by this inhibitor, though this assertion was not substantiated. The different results obtained by Mueller can perhaps be explained by his more sensitive assessment methods, which utilised electrokinetic potentiodynamic anodic polarization traces.

Neal, Craig and Powers (1983) studied the cutting

effectiveness of K-type size 30 stainless steel files after immersion and agitation in one of four irrigating solutions:- (1) 0.9% saline; (2) 5.25% NaOCl; (3) 3% hydrogen peroxide; and (4) RC-Prep (applied with a camel hair brush). Cutting ability was assessed both before and after immersion, and the experiment was performed daily for ten days. Repeated ten minute exposures to all solutions, with the exception of saline, produced significant reductions in cutting ability of the files. Exposure to NaOCl resulted in the files being covered with a brown corrosion product, while no visually observable effect was noted with the other irrigants. The authors urged that instruments should be cleaned immediately after use of NaOCl, hydrogen peroxide and EDTA-urea peroxide solutions.

In a study which subjected ultrasonic diamond files and stainless steel K-files to twenty minutes usage under copious flow of 2.6% NaOCl, Scott and Walton (1986) found no deleterious effects on files.

After immersion of "the thinnest Kerr reamers and Hedströms files" (presumably carbon steel) in EDTA, Östby (1957) subjected the instruments to various tests. "The exposed files showed no differences from the new ones in the various bending and torsion tests." However, the use of EDTA with a sonic instrumentation system, the Endostar 5, produced corrosion of the handpieces (Barnett, Godick and Tronstad, 1985).

CHAPTER 11

SUMMARY AND CONCLUSIONS

Historically, the concept of irrigation was closely linked to the understanding that apical healing required thorough debridement of the canal. Irrigants are now employed to physically debride the root canal, and, with interappointment medicament use, to contribute to the disinfection of the dentinal tubules.

Irrigation aids in debridement of tissue remnants and viable bacteria during endodontic treatment. However, as is apparent from this review, neither the process of irrigation, nor the action of irrigants, can reliably eliminate debris and microorganisms from all areas of the root canal system.

There is no "perfect" irrigant delivery system, but the following regimen would appear to provide the most reliable and consistent irrigant delivery system:- (1) a sterile 27-gauge needle, bent for ease of access; (2) a sterile plastic syringe; (3) a non-forceful injection technique; (4) avoidance of needle binding; and (5) utilisation of a greater rather than lesser volume of irrigant.

Controversy exists over the synergistic effects of irrigants used with ultrasound. While ultrasound might enhance the bactericidal and debriding action of

irrigants *in vitro*, its effectiveness has yet to be conclusively established *in vivo*. Further research should clarify the efficacy of the combination, as well as the optimal type, temperature and concentration of irrigant.

There is confirmation that 2.5% to 5% NaOCl, of the large number of irrigants available, most closely fulfils the requirements of an ideal irrigant. It has also been established that 10%-15% EDTA solution is an effective chelating agent. Irrigation with NaOCl, in conjunction with EDTA, might best provide organic and inorganic dissolution, as well as removal of debris and smear layer.

While most bacteriologic studies of irrigants have involved facultative anaerobes, there are indications that NaOCl is effective against strict anaerobes *in vitro*. Further *in vivo* research on the effects of irrigants on anaerobes is required. In addition, because of the potentially complex interactions between the various microorganisms in root canals, only *in vivo* studies involving full and serial microbial sampling are likely to provide definitive data on the bactericidal effectiveness of various irrigating regimens.

A potential for irrigant toxicity exists. However, because *in vitro* and animal studies have limited practical applicability, clinical studies might be of greater value. Further research which investigates the combined biocompatibility and antibacterial properties of irrigants will probably be of still greater relevance.

It has yet to be conclusively established whether smear layer removal improves the prognosis of endodontic therapy. However, the effects of irrigants on uninstrumented dentine, and on dentine following smear layer removal, is of considerable interest. Further research could investigate the extent of diffusion of irrigants through radicular dentine, especially in relation to cytotoxic effects. In addition, the permeability of different types of radicular dentine to irrigants should be examined.

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