CHAPTER ONE

INTRODUCTION, AIM AND OBJECTIVES OF THE STUDY

1.1. Introduction

The three-dimensional obturation of the root canals is the last step of root canal procedures following cleaning (disinfection) and shaping of canals (Barroso *et al.*, 2005). The aim of root canal obturation is to prevent the ingress of micro-organisms into the cleaned and disinfected root canal space. In addition, obturation helps in preventing recolonization (reinfection) and multiplication of bacteria in the root canal system. The obturation should provide an adequate seal to minimize the chance of root canal treatment (RCT) failure (Ford *et al.*, 2002, Guess *et al.*, 2003). The capability of root canal filling materials and techniques to ensure obturation and sealing of thin and irregular ramifications are an important clinical consideration (Venturi *et al.*, 2005). Leonard *et al.* (1996) stated that a complete seal of the canal system, however, cannot be accomplished with the available materials or techniques.

In general, obturation is composed of a core material such as gutta-percha (GP) and sealer (Farhad *et al.*, 2011). Worldwide, GP has been accepted as the "gold standard" root filling material (Monteiro *et al.*, 2011), against which most others are compared (Patel *et al.*, 2006, Stratton *et al.*, 2006). GP is used either in form of solid core or softened core in conjunction with different types of sealers (Kirkevang and Horsted-Bindslev, 2002). A good root canal obturation material should adapt well to the canal walls for the entire length of the canal and form a homogeneous mass. The quantity of core material should be maximized while the quantity of sealer should be minimized (Epley *et al.*, 2006, Hammad *et al.*, 2009). Although many techniques have been advocated for filling root canals, no one technique has been recognized as obviously superior (Collins *et al.*, 2006).

It is well accepted that the most favourable RCT outcomes are attained when the limit of root canal instrumentation and obturation is set at 1.0 mm short of the radiographic apex. This is because any material that was forced into the periapical tissues could lead

to a chronic inflammatory response (Schaeffer *et al.*, 2005, Gomes-Filho *et al.*, 2008). It is recommended that any extrusion beyond the apex must be avoided since it results in complications (Brkic *et al.*, 2009). An increased possibility of extrusion, however, is a characteristic of the warm vertical compaction techniques (Mente *et al.*, 2007).

The GP and root canal sealers have been used to obturate the root canals, despite their apparent incapability to accomplish a good seal along the dentinal walls of the root canal (Gesi *et al.*, 2005). GP appears to have a poor adhesion to the canal walls and to the sealer. This constitutes a weak point whereby a path for leakage might be created (Drukteinis *et al.*, 2009). Either zinc oxide-eugenol (Dorifill) or calcium hydroxide (Apexit) sealer failed to show adhesion to the canal wall or the GP (Khedmat and Sedaghati, 2006). A good root canal sealer should, to some extent, adhere firmly both to dentine and to GP (Lee *et al.*, 2002). Therefore, resin-based root canal sealers were introduced, which seemed to be more effective in sealing root canals than the zinc oxide-eugenol-based sealer (Adanir *et al.*, 2006). AH Plus, an epoxy based sealer, was shown to produce a high bond strength to the canal wall and an adequate long-term dimensional stability. However, its sealing ability still remains controversial since it did not bond to GP (Teixeira *et al.*, 2009). On the other hand, an initial report showed that EndoREZ® (ER) (Ultradent Products Inc., South Jordan, UT, USA), a urethane dimethacrylate resin based sealer, provides an effective seal (Zmener *et al.*, 2008). The hydrophilic properties of the resin (sealer) allow deep penetration into the root canal walls but not into GP (Zmener *et al.*, 2008). For this reason, another material called Resilon, a thermoplastic synthetic polymer-based root canal filling material, was introduced as an alternative to GP. Stratton *et al*. (2006) demonstrated that leakage in root canal filled with Resilon was significantly less compared to GP. However, another study pointed out that the adhesive potential of Resilon to radicular dentine was far from satisfactory or similar to GP (Nagas *et al.*, 2010). Alternatively, resin coated GP points,

the ER points, have been introduced (Drukteinis *et al.*, 2009). The ER sealer with ER point could be used to establish a continuous adhesion (uniblock or monoblock) between these materials (Pameijer and Zmener, 2010). In a study performed by Drukteinis *et al.* (2009), the ER point/ER sealer combination decreased the microbial leakage when compared with GP/AH Plus.

To date, studies that investigate the quality of root canal obturation using this resin-coated GP point are sparse.

1.2. Aim of study

The aim of this study was to evaluate and compare the quality of root canal obturation of different materials (ER Points/ER sealer and GP points/AH Plus® sealer) using different obturation techniques: cold lateral compaction (CLC), warm lateral compaction (WLC) and single cone (SC).

1.3. Objectives of study

The objectives of this study were to examine the obturation quality of ER Points/ER sealer and GP points/AH Plus® sealer in canals obturated with different techniques i.e. CLC, WLC and SC, with respect to:

1. Apical extrusion of obturation materials.
2. Percentage of canal area occupied by core filling materials versus sealer + voids.
3. Adaptation of obturation materials (core filling and sealer) to canal walls.

CHAPTER TWO

LITERATURE REVIEW

2.1. Root canal treatment

The aim of RCT is to preserve normal function of treated tooth and to eliminate root canal infection; thereby prevent or heal periapical inflammatory tissue lesions (De Chevigny *et al.*, 2008). In order to achieve complete healing, proper disinfection, three-dimensional obturation and adequate coronal seal are essential (Nawal *et al.*, 2011). Inadequate canal instrumentation and incomplete obturation of the canal space were considered as important causes of RCT failure (Rödig and Hülsmann, 2003).

2.1.1. Phases of RCT

There are three phases in RCT (Castellucci, 2004, Schwartz, 2005)

A. Diagnostic phase:

   Involve identification of the cause of the disease and develop a treatment plan that will solve or alleviate the problem.

B. Preparation phase:

   This phase includes removal of caries, defective restorations, debris, necrotic and infected tissues from the root canal space. The objective is to create an environment in which the body's immune system can produce healing of the periapical inflammatory tissue lesion.

C. Obturation phase:

   In this phase, the canal is filled with an inert material to obtain a good seal as close as possible to the cement-dentinal junction.

2.1.2. Rationale of RCT

The rationale for the RCT is to prevent microbial colonization and multiplication in the pulpal space, and subsequent symptomatic or nonsymptomatic presentations of apical periodontitis (Bergenholtz *et al.*, 2009c). Therefore, elimination or reduction of microorganisms in the pulp space followed by closure of the canal will result in healing (Spangberg and Haapasalo, 2002).

The outcome of the RCT is significantly influenced by the presence of bacteria in the root canals at the time of filling (Siqueira and Rôças, 2008). This indicates that persistent bacteria can live in treated canals and are able to induce or prolong periradicular tissue inflammation, thus the eradication of bacteria from the root canal system is the conclusive goal of the RCT of teeth with apical periodontitis (Siqueira and Rôças, 2008). This goal may be carried out in various ways, which include instrumentation and irrigation of the canal, and placement of medicaments, and, in some cases, surgery. No cases lend themselves to a successful treatment without a significant degree of debridement (Wiene, 2004) and complete sealing of the canal space to prevent ingress of bacteria (Jainaen *et al.*, 2007).

2.2. Preparation of root canal

Preparation of the root canal system is recognized as one of the important stages in RCT (Siqueira Jr, 2001, Hülsmann *et al.*, 2005). Schilder (1974) stated that effective cleaning and shaping of the root canal system is essential for achieving the biological and mechanical objectives of RCT. The objectives are to remove all the pulp tissue, bacteria and their by-products whilst providing adequate canal shape to receive a three-dimensional obturation of the entire root canal space (Yared, 2008). Shaping refers to maintaining the original canal configuration without creating any iatrogenic errors such as instrument fracture, external transportation, ledge, or perforation (Guelzow *et al.*,

2005, Aguiar *et al.*, 2009). In addition, canal shaping involves development of a continuously conical form of the root canal preparation which is wide coronally and narrows apically, with the apical foramen kept as small as is realistic (Beer *et al.*, 2000b).

2.2.1. Root canal preparation techniques

There are many different techniques to prepare the root canal system which includes:

a. Standardized technique

John Ingle was a member of the first committee to propose standardization of endodontic instruments and introduced the classic "standardized preparation." Standardized files were used sequentially to produce a canal preparation that assumed the same size and shape (taper) as the last instrument used. The canal could then be obturated with a filling material that of the same dimension. That is, the canal was made to fit the filling material (Johnson, 2002a). However, creating a true standardized tapered preparation is difficult in ideal situations and impossible in curved canals (Torabinejad and Walton, 2002). While the instrument sizes increase, the flexibility of the instrument decrease and can led to occurrence of iatrogenic errors in curved root canals. Other problems commonly encountered owing to compaction of dentine debris included ledging, zipping, elbow formation, perforation and loss of working length (Messing and Stock, 1988, Carrotte, 2004).

b. Step-back technique

The step-back technique has evolved from the step back or telescope technique first described in the 1960s. It replaced the standardized preparation techniques and aimed to reduce the transportation in the apical part (Harty and Ford, 2004).

The step-back techniques emphasize to maintain the apical preparation small, in its original position, and to produce a flared preparation. Briefly, this technique involves two phases of preparation: phase I is the preparation of apical constriction, and phase II involves preparation of remaining (coronal part) root canal system (Murgel *et al.*, 1991, Garg and Garg, 2007). In this technique the working length is established and then the first file that binds in the canal is set as the master apical file (MAF). Subsequently larger files are introduced at 1.0 mm shorter than the preceding size, thus a conical configuration of the root canal with an apical stop is achieved (Beer *et al.*, 2000b). Thus, a canal with greater tapers is produced compared with the standard technique and results in more dentine removal and cleaner canal walls (Johnson, 2002a). In curved canals, this technique can result in iatrogenic damage to the natural shape of the canal due to the inherent inflexibility of stainless steel files (Young *et al.*, 2007).

c. Step-down technique

This technique was first suggested by Schilder in 1974, and the technique was described in detail by Goerig in 1982 (Al Negrish, 2009). The principle of these techniques is that the coronal aspect of the root canal is widened and cleaned before the apical part (Carrotte, 2004, Al Negrish, 2009). It is important to introduce a fine, stainless steel taper 0.02 file first into the canal to determine if the canal is patent to the apical constriction, prior to cleaning and shaping (Ingle and Bakland, 2002a). Step-down technique involves preparation with larger instrument in the coronal part of the root canal and then work down (apically into) the root canal with progressively small files until the smaller file achieves the root apex (Beer *et al.*, 2006b). Then, the apical portion of the canal can be prepared at the working length. The final canal taper is accomplished using the step-back technique (Torabinejad and Walton, 2002).

The advantages of this technique presented by Goerig *et al.* (1982) are as follows:

1) During cleaning and shaping of the canal the majority of microorganisms and pulpal tissues are removed early, thus, the potential for introducing of microorganisms into the periapical area is reduced.
2) Unimpeded insertion of root canal instruments into the apical area once the coronal two-thirds are enlarged.
3) Complete shaping of the apical region quickly and efficiently.
4) Better irrigant penetration into the root canal system.

d. Balanced force technique

In 1985, Roane advocated an original preparation method for curved root canals, which produce a similar quality of preparation as is usually required and obtained for straight canals, while avoiding ledge formation, transportation of the foramen or perforation; it is called balanced force technique (Charles and Charles, 1998). In this technique the K-file is introduced into the canal without applying any force. Concurrently, the instrument is rotated one quarter turn clockwise. This movement advances the cutting blades of the instrument against the dentinal wall. Next, the file is given three-quarter turn counter clockwise while sustaining a light apical pressure to keep the file at the same level and prevent it from moving out of the canal. During this movement dentine is scraped from the canal wall. Then, the dentine shavings are picked up by half a turn clockwise and removed from the canal (Beer *et al.*, 2000b). Balanced force technique is specifically designed to operate K-type root canal instruments and should not be used with Broach type or Hedstrom type instruments, since neither possesses a left-hand cutting capacity (Garg and Garg, 2007). This technique has many advantages including good apical control of the file tip as the instrument does not cut over the complete length, good centring of the instrument because of the non-cutting safety tip, and pre-

curving of the instrument is unnecessary (Cohen and Burns, 2002, Hülsmann *et al.*, 2005). As stated by Ingle and Bakland (2002a), McKendry reported that the root canal debrided at the apical part by the balanced force technique was at least as adequate as the step-back technique. Moreover, significantly less debris was extruded apically using balanced force technique as compared to step-back technique (Beer *et al.*, 2000b).

e. Crown-down technique

Initially, Marshall and Pappin advocated a crown-down pressureless preparation in which Gates-glidden drills and larger files are first used in the coronal two thirds of the canal and then progressively smaller files are used from the crown downwards until the desired length is reached. This has become known as crown-down technique (Ingle and Bakland, 2002a). It has been reported to produce less apically extruded debris than the step-back technique (Kustarci *et al.*, 2008). The crown-down technique uses step-down concepts for shaping the entire canal length (De Leon Del Bello *et al.*, 2003). This technique has raised in popularity, especially among those using nickel-titanium (NiTi) instruments with varying tapers (Ingle and Bakland, 2002a). The NiTi rotary instruments were introduced in 1991 for root canal preparation (Shen and Haapasalo, 2008). The ability of NiTi rotary instruments to remove dentine and pulpal debris during canal shaping is related to designing features of the instrument, particularly cross-sectional profile and flutes (Kuzekanani *et al.*, 2009). These instruments enable clinicians to create more predictably and efficiently tapered preparations especially in curved canals (Shen and Haapasalo, 2008). These super-elastic instruments offer benefits over hand instrumentation for preparing curved root canals, including less transportation of the canal and reduced operating time (Kuzekanani *et al.*, 2009). Furthermore, finger fatigue is less because the hand piece is doing most of the work (Schäfer and Zapke, 2000). However, the main disadvantage of NiTi rotary instruments

is their tendency to fracture particularly if overused (Parashos *et al.*, 2004, Parashos and Messer, 2006). It was found that NiTi instruments because of their flexibility had deficiencies in preparation of oval root canals, since it was not possible to direct these flexible instruments into the recesses (Hülsmann *et al.*, 2005). Preliminary evidence indicates that debridement effectiveness with NiTi rotary instrument is comparable to that with hand instrumentation (Schäfer and Zapke, 2000). Conversely, a retrospective study of RCT outcome, showed that the NiTi rotary instruments had more success rate and lower procedural errors during root canal treatment than stainless steel hand files (Cheung and Liu, 2009). Another study by Taha *et al.* (2010), who investigated canal cleanliness especially in the apical third, found that the rotary instruments performed significantly better than hand instruments with regard to residual debris. In addition, engine-driven techniques produced less debris extrusion than the manual techniques (Ferraz *et al.*, 2001).

Over the past few years, the movement toward using NiTi rotary instruments for root canal preparation have resulted in a multitude of instrumentation systems in the market (Ingle and Bakland, 2002a). Examples are ProTaper, K3, RaCe and Mtwo rotary instruments.

In a study of the efficacy of rotary instrument in preparing an oval root canal, the Mtwo and ProTaper were found to be more efficient than NiTi hand files (Elayouti *et al.*, 2008). Mtwo instrument was found to be able to maintain the original canal curvature significantly better and was faster than K3, RaCe (Schäfer *et al.*, 2006a) or ProTaper instruments (Kuzekanani *et al.*, 2009).

2.3. Obturation of root canal

Obturation of the root canal is a significant stage in RCT. Presence of root canal filling facilitates in prevention of bacterial penetration and their products into the periradicular tissue (Yücel, 2006). In addition, obturation helps in prevention of contamination of pulp canal space that may occur from intercommunication with gingival sulcus or periodontal pocket (Ford *et al.*, 2002, Guess *et al.*, 2003). Teeth with inadequate obturation might need retreatment before coronal restoration (Hammad *et al.*, 2009).

2.3.1. Materials for root canal obturation

Root canal obturation is generally composed of core materials such as GP and sealer (Farhad *et al.*, 2011). These materials may be considered as true implants because they might be in contact with the tissues of the body (Tronstad, 2008). Therefore, they must possess several different properties relative to their functions and location. Some of the ideal properties include biocompatibility and sealiability (Orstavik, 2005).

Root canal filling materials may be divided into two distinct groups:

(1) Core materials

(2) Root canal sealers

(1) The core materials

a. Gutta-percha

GP is a polymer, primarily polyisoprene, which is extracted from a tropical tree. It is 60% crystalline at room temperature, while the rest of the mass has an amorphous structure (Tronstad, 2008). Crystalline GP may occur in two forms; alpha (α) or beta (β) phase. There are only minor differences in chemical behaviour and physical properties between the two. The α phase appears in nature; the β phase occurs during refining and manufacturing of the conventional GP that is used clinically (Orstavik, 2005). These

crystalline forms are interchangeable depending on the temperature. Upon heating the primary β form change to α form, and upon cooling it returns to its β form (Namazikhah *et al.*, 2000). At the room temperature α form is brittle, however, when heated it has adhesive characteristics, a low viscosity and highly flowable. On the other hand, the β form is more stable and flexible at the room temperature and when heated it is less adhesive and flowable than the α form (Tanomaru-Filho *et al.*, 2007a).

GP is the main filling material used in RCT, although it only forms about 20% of the composition of dental GP points. Zinc oxide is the main component (60-70%) of the point. The remaining 10% consists of a mixture of resins, waxes and metal sulphates (Ingle and Bakland, 2002b, Hauman and Love, 2003). GP points are available in standardized and non-standardized forms. The standardized forms match the same dimensions and uniformity of the files that are used in root canal preparation. The non-standardized forms, classified in some GP brands as medium, fine, medium-fine, or fine-fine, have greater tapers than standardized cones. They are often used in cold lateral compaction as accessory points and in techniques that involve vertical compaction of heat-softened GP (William, 2002).

GP is considered as the best material for root canal filling, regardless of the technique applied (Maniglia-Ferreira *et al.*, 2005). Nevertheless, leaching of zinc oxide, if any, from GP points has been shown to be cytotoxic *in vitro* (Pascon and Spångberg, 1990), tissue irritating *in vivo* and associated with an inflammatory reaction (Sjögren *et al.*, 1995). Tissue response to GP has been studied using subcutaneously implanted Teflon cages in which GP evoked two distinct types of tissue reaction. Large pieces of GP were well tolerated and encapsulated by collagen, with the surrounding tissue free of inflammation. In contrast, fine particles of GP evoked an intense, localized tissue response, characterized by the presence of macrophages and giant cells resulting in impairment in the healing of apical periodontitis (Sjögren *et al.*, 1995, Ingle *et al.*,

2008b). On the other hand, in an animal studies involving implanted pieces of GP, the material was considered to have acceptable biocompatibility with a low degree of toxicity (Hauman and Love, 2003, Bodrumlu, 2007). Another limitation of GP is that it has no adhesive properties to dentine of the root canal wall. Thus, it must be used with a sealer to fill the space between root canal wall and GP, as well as to fill the root canal irregularities (Pitt-Ford, 2004, Aydemir *et al.*, 2009). For this reason, resin bonded root canal filling materials have been recommended as an alternative to the traditional GP based system for a better seal (Stratton *et al.*, 2006).

b. Resin-based core filling materials

In 2004, a new resin based root filling material was marketed under the brand name, Resilon™ (Resilon Research LLC, Madison, CT, USA) which was stated as having the ability to overcome the limitations of GP (Shipper *et al.*, 2004b) and shown to have good biocompatibility (Garcia *et al.*, 2010). Resilon is a synthetic material based on polyester matrix with bioactive glass, bismuth and barium salts as fillers (Stratton *et al.*, 2006, Onay *et al.*, 2007). Resilon appears and manipulates like GP and is also called "resin-percha". The Resilon-Epiphany system consists of three parts: Resilon is considered as the major core-filling materal; Epiphany sealer is a resin-based composite that forms a bond to the dentine wall and to the core material and is dual cured; and Primer, which prepares the canal wall (Pawi Ska *et al.*, 2006, Wedding *et al.*, 2007). This combination forms a monoblock filling in the root canal system (Skidmore *et al.*, 2006). Proponents have shown that there was a decreased in leakage in canals filled with Resilon compared with GP using bacterial or fluid filtration models (Shipper *et al*., 2004b, Shipper *et al*., 2005, Stratton *et al*., 2006 and Wedding *et al.*, 2007). Although, there are many studies that mentioned advantages of Resilon over GP, Olga Onay *et al*. (2006) showed that the combination of GP and Epiphany had a higher (but not

significant) sealing ability than that of Resilon with Epiphany. A study by Epley *et al.* (2006) has shown that at 1, 3, and 5 mm levels (cross-sections) from the apex, there was no significant difference in the area occupied by voids between root canals obturated with lateral compaction or continuous wave obturation of Epiphany, and those with continuous wave obturation of GP.

Besides Resilon, there is another resin-based material called ER. ER point is polybutadiene-diisocyanate-methacrylate resin-coated gutta-percha. The ER sealer is a hydrophilic, dual-cured resin containing 30% urethane dimethacrylate (Lee *et al.*, 2008). While GP showed a weak bond to root canal sealers, resin-coated GP points (ER Points) have been introduced to overcome this problem (Zmener *et al.*, 2008). According to the manufacturers (Ultradent Products Inc., South Jordan, UT, USA), ER Points and ER sealer are designed to be used passively with a gentle "harpooning" motion which means, master point and accessory point are simple inserted as needed and then verify radiographically. ER Points and ER sealer may also be used with obturation techniques such as lateral or warm vertical compaction (Leonardo, 2009). Apical quality and adaptation of Resilon and ER root canal filling materials in combination with a non-compaction technique were evaluated radiographically by Herbert *et al.* (2009); there were no significant differences between the materials. Conversely, Hammad *et al.* (2009) found that root canals filled with GP points/TubliSeal sealer and GP points/GuttaFlow sealer showed less voids and gaps than those filled with ER points/ER sealer, RealSeal points/RealSeal sealer.

(2) Root canal sealers or sealing materials

Root canal sealers are generally unacceptable as root canal filling materials without a core material, because of shrinkage, solubility, or difficulties in removing the material. Providing a bacteria-tight seal of the canal that sufficiently prevents ingress and growth of bacteria is the most important requirement of a sealer. Other important properties include that it should not shrink and it should be biocompatible (Tronstad, 2008). In general, root canal sealers in its initial setting phases are cytotoxic and bacteriotoxic. Thus, extrusion of sealer material should be avoided (Bergenholtz *et al.*, 2009b). The sealer can be applied into the root canal by placing the sealer on the master cone and pumping the cone up and down in the canal, or placing the sealer on a file and spinning it counter clockwise. In addition, sealer can be placed using either a lentulo spiral, a syringe or an activated ultrasonic instrument (Bergenholtz *et al.*, 2009b). Some studies have shown that distribution of sealer in canal walls is not affected by the method of sealer placement (Wiemann and Wilcox, 1991). The ideal consistency of the sealer that is presented by Benatti and colleagues was achieved when the mixture can be held for 10 seconds on an inverted spatula without dropping off and will stretch between the slab and spatula for 2.0 cm before breaking. This ideal consistency permits sufficient clinical working time and minimal dimensional change (Ingle and Bakland, 2002b). There are many types of root canal sealers which include:

a. Zinc oxide and eugenol-based sealers

These sealers are zinc oxide-eugenol (ZnOE) cements modified for use in RCT. The liquid for these materials is eugenol whilst the powder contains finely sifted ZnO to improve the flow of the cement (Hauman and Love, 2003). It meets all the ideal requirements of sealer as proposed by Grossman (Ingle and Bakland, 2002b). ZnOE cements, however, cause tooth discoloration, which is produced via its silver content,

which was added for radiopacity. In 1958 Grossman recommended a non-staining ZnOE cement as a substitute for Rickert's formula (Ingle and Bakland, 2002b). In addition, ZnOE-based cement may adversely affect the adhesion to dentine and polymerization of the resin cements used for post cementation (Dias *et al.*, 2009). As was stated by Tronstad (2008), Serene *et al*. found that ZnOE sealers activated the complement system and thus initiated an inflammatory reaction. Furthermore, Guigand *et al.* (1999) confirmed the initial cytotoxicity of ZnOE during a period of 24, 72 and 168 hours. The toxicity is caused by free eugenol that is present in the freshly mixed material (Tronstad, 2008). On the other hand, ZnOE sealers have the advantage of being resorbed if extruded into the periradicular tissues (Johnson, 2002b). ZnOE sealers provide a bacteria-tight seal and, in general, do not shrink. Thus, it is able to fill the spaces between the GP points and the root canal wall (Tronstad, 2008).

b. Calcium hydroxide-based sealers

Calcium hydroxide [$Ca(OH)_2$] was introduced by Herman in 1920 for its pulp-repairing ability. It is mainly used for pulp capping procedures, as an intracanal medicament, for apexification, and as a component of root canal sealers (Desai and Chandler, 2009). In general, $Ca(OH)_2$-based products are different from one another, but they share two ingredients: $Ca(OH)_2$ and salicylate (Schmalz and Arenholt-Bindslev, 2009). $Ca(OH)_2$ has low solubility in water, an inherently high pH (approximately 12.5 - 12.8), and is insoluble in alcohol. Furthermore, it is antimicrobial via the release of hydroxyl ions that leads to a highly alkaline environment (Athanassiadis *et al.*, 2007). These sealers have been shown to have similar sealing ability to ZnOE sealer. Long-term exposure to tissue fluid, however, may lead to dissolution of the material as $Ca(OH)_2$ is capable to leach out (Pitt-Ford *et al.*, 2002). Several brands of $Ca(OH)_2$-based sealer, e.g. Sealapex, CRCS and Apexit have been marketed, which claim the benefits of the

biological effects of added $Ca(OH)_2$ and considered to be biocompatible (Hauman and Love, 2003). Conversely, a study by Bezerra Silva *et al.*, (1997) showed that the Sealapex, CRCS and Apexit sealers produce tissue necrosis.

c. Epoxy-based sealers

Several epoxy-based sealers are available in the market. AH26 (Dentsply DeTrey) is a product, either with silver or silver free, available as a powder-liquid formulation. AH Plus (Dentsply DeTrey) is a newer product (also marketed as Top Seal) available as paste-paste formulation (Schmalz and Arenholt-Bindslev, 2009). Kaplan *et al*. (2003) and Miletić *et al.* (2003) presented that epoxy resins are well established as an effective root canal sealer, displaying acceptable biocompatibility. Mcmichen *et al.* (2003) presented that they have shown insolubility and dimensional stability. In addition, Epoxy resin-based root canal sealers have shown apical sealing (Dultra *et al.*, 2006) and good adhesive and antibacterial properties (Pitt-Ford *et al.*, 2002). However, AH26 is very toxic when freshly prepared, which is attributed to the release of a very small amount of formaldehyde as a result of the chemical setting process (Hauman and Love, 2003). The amount of released formaldehyde is greater in AH26 than AH Plus (Leonardo and Da Silva, 1999). The former material was shown to be cytotoxic to fibroblasts that lasted for 1 week, whereas the cytotoxicity of AH Plus was in the early stage of mixing and was no longer detected 4 hours after mixing (Hauman and Love, 2003). AH Plus has been the sealer of choice for more than a decade (Drukteinis *et al.*, 2009) and the sealer against which new sealers and bondable root canal filling materials are compared (Onay *et al.*, 2010).

d. Glass ionomer-based sealers

Glass ionomer cements (GIC) have been developed and introduced as an endodontic sealer (e.g. Ketac-Endo). Glass ionomers in RCT was recommended as early as 1976 by Pitt-Ford in England (Ingle and Bakland, 2002b). The setting time of this material, however, is too fast and the material is difficult to remove in the event of re-treatment, as there is no known solvent for this material (Ingle and Bakland, 2002b). In addition, it is known to cause minor tissue irritation with low toxicity *in vitro* (Hauman and Love, 2003). Nevertheless, this material has the advantage of bonding to dentine, appears to provide an adequate apical and coronal seal, is biocompatible (Timpawat *et al.*, 2001b) and is antibacterial as a result of fluoride release and an acidic pH (Ogasawara *et al.*, 2003). The bonding properties are considered as an obvious advantage over the ZnOE type or epoxy resin-type sealer cements (Timpawat *et al.*, 2001b).

e. Methacrylates resin based sealers

EndoREZ (ER) (Ultradent Products Inc., South Jordan, UT, USA) is a hydrophilic, two-component (base and catalysts), dual-curing self-priming resin sealer (Pameijer and Zmener, 2010). ER sealer is urethane dimethacrylate (UDMA) that has a filler content of approximately 50% by weight of bismuth oxychloride, calcium lactate pentahydrate and silicon dioxide (Zmener and Pameijer, 2007). In addition, it contains zinc oxide and barium sulfate (Sousa *et al.*, 2006). It is the second generation of non-etching, bondable sealer, and does not require the adjunctive use of a dentine adhesive (Kim *et al.*, 2010). The manufacturer stated that the ER sealer has satisfactory sealing properties with an easy delivery system (Hammad *et al.*, 2007). It is designed to flow into accessory canals and dentinal tubules to facilitate resin tag formation for retention and seal after smear layer removal with sodium hypochlorite (NaOCl) and ethylene-diamine-tetra-acetic acid (EDTA) (Kim *et al.*, 2010). The resin tag formed by the hydrophilic nature of the sealer

enables the creation of an extensive network of 800 to 1200 μm after removal of smear layer (Ingle *et al.*, 2008d). It is highly radiopaque and has low viscosity, which produce a favourable handling property for the sealer. It is said to be applicable for both wide and narrow root canals and provides a good adaptation to the details of the dentinal walls (Zmener and Pameijer, 2007).

Urethane dimethacrylate resin has been used in orthopaedic surgery for many years as bone resin. Thus, it has a long track record of biocompatibility. This is very important since root canal sealer may be in direct contact with apical connective tissues for a long period of time, and has the potential to cause inflammatory degeneration and delaying periapical healing. Study on ER sealer has confirmed its biocompatibility, low cytotoxicity and antimicrobial properties (Leonardo, 2009). On the other hand, in bone tissue study that carried out by Zmener *et al.* (2005a), by means of quantitative analysis, they did not observe prolongation of an inflammatory reaction in the presence of ER. Conversely, ER sealer seems to cause a more intense reaction than that observed either in the AH Plus or the control group (EndoFill), after a period of 60 days (Scarparo *et al.*, 2009). However, this result disagreed with that of Zmener *et al.* (2010) who concluded that EndoREZ/Accelerator and RealSeal behaved similarly and were both well tolerated by the subcutaneous connective tissues of rats after 90 days of implantation. Therefore, elution of chemical components that are potential irritants decreased with time (Zmener *et al.*, 2010). In a microleakage study Zmener *et al.* (2005b), using CLC technique, found that teeth with ER sealer group showed less leakage than Grossman's cement group. ER sealer showed a better sealing ability of the root canal system, good penetration into dentinal tubules and did not permit the permeation of *Enterococcus faecalis* during the experimental period (30 days). On the other hand, AH Plus did not present the best result for intratubular penetration, although it was able to prevent the permeation of *Enterococcus faecalis* (Bortolini *et al.*, 2010).

RealSeal (SybronEndo, Orange, CA, USA) or Epiphany (Pentron Clinical Technologies,Wallingford, CT, USA) is considered the third generation of hydrophilic methacrylate resin-based material that includes a self-etching primer and sealer (Shrestha *et al.*, 2010). RealSeal sealer contains urethane dimethacrylate (UDMA), polyethylene glycol dimethacrylate (PEGDMA), ethoxylated bisphenol a dimethacrylate (PEPADMA), and bisphenyol A-dimethacrylates (Bis-GMA) resins which was designed for use with a polycaprolactone core material. Additionally, this sealer contains silane-treated barium borosilicate glass, barium sulphate, silica, calcium hydroxide, bismuth oxychloride with amines, peroxide, (a photo inhibitor), and pigments (Ingle *et al.*, 2008c). Early studies using Resilon and Epiphany showed less bacterial leakage and were associated with less periapical inflammation after coronal microbial inoculation in an animal (Shipper *et al.*, 2005). However, in a study of cytotoxicity, the findings demonstrated that in the freshly mixed state, the cytotoxicity of the Activ GP and RealSeal sealers is between the Kerr and AH26 sealers This cytotoxic nature can be attributed to the leaching of uncured monomers (Donadio *et al.*, 2009).

The major problem associated with endodontic bonding is the lack of relief of shrinkage stresses which are created in deep and narrow canals. Stress relief by resin flow is dependent upon cavity geometry and resin film thickness. For instance, in a class I box-like cavity there is five times more bonded surface area than unbonded surface area, giving a ratio of 5. This ratio of the bonded to the unbonded surface area is called the configuration factor or C-factor. During polymerization, the unbonded surface can move and flow, thereby relieving shrinkage stresses. As the unbonded surface area becomes small, as in a long narrow root canal, there is insufficient stress relief by flow and thus a high probability for one or more bonded areas to pull off or debond (Tay *et al.*, 2005c).

2.3.2. Methods of filling root canal

There are different methods in filling the root canal depending on the direction of the compaction, either lateral or vertical, and the temperature employed, either cold or warm (plasticized) (Bergenholtz *et al.*, 2009b). The followings are the various techniques:

a. Single cone technique

Single cone (SC) obturation was introduced in the 1960s with the ISO standardization for root canal instruments and filling points (Bergenholtz *et al.*, 2009b). The SC technique consists of matching a cone to the prepared canal, with the thickness of the sealer layer is depending on the fit of the cone to the canal walls (Wu *et al.*, 2009). With the introduction of NiTi rotary instruments greater-taper master cones that closely match the rotary instrument systems have been introduced to facilitate SC filling of root canals (Monticelli *et al.*, 2007). Although the technique is simple (Gordon *et al.*, 2005, Hörsted-Bindslev *et al.*, 2007), it has several disadvantages and cannot be considered as one that seals canals completely. Specifically, after preparation, root canals are seldom round throughout their length, except possibly for the apical 2.0 or 3.0 mm (Bergenholtz *et al.*, 2009b). On the other hand, Monticelli *et al.* (2007), using two SC techniques (ActiV GP and GuttaFlow), found that these root-filling techniques do not ensure a durable apical seal against bacterial leakage when compared to a warm vertical compaction technique. It has been shown that SC technique with a 0.06 tapered core filling was comparable to lateral compaction in the percentage of the GP occupying a prepared 0.06 tapered canal (Gordon *et al.*, 2005). Furthermore, SC fillings with RoekoRSA sealer which were used to obturate wide and straight canals have been shown to prevent fluid transport for one year (Wu *et al.*, 2006). In addition, Wu *et al.* (2009) presented that SC technique has similar quality to CLC technique in filling small curved root canals.

b. Lateral compaction technique

Cold lateral compaction (CLC) of GP is probably the most widely used method for root canal obturation (Mente *et al.*, 2007). It is the "gold standard" against which other techniques are compared (Wu *et al.*, 2009). In this technique, an ISO-standardized master GP point (with 0.02 taper), and numerous accessory GP points are used to fill the whole canal space (Heredia *et al.*, 2007). There are several important steps that have priority for this technique: spreader size determination, master and accessory points size determination (Ingle and Bakland, 2002b). The major advantages of CLC technique, it has a controlled placement of GP into the root canal (Lea *et al.*, 2005) and low cost (Peng *et al.*, 2007). However, this technique has been criticized for its lack of homogeneity, poor canal replication ability, very time and technique demanding. In addition, there is a high possibility of root fracture (Dadresanfar *et al.*, 2010) because of the compaction load that is created by the pressure of the spreader against the canal wall (Mente *et al.*, 2007). In addition, voids formation which are produced due to spaces found between individual GP cones and the root canal walls, can be resulted from poor root canal preparation, curved canals, inadequate lateral pressure during compaction, mismatches between GP points and the prepared root canal (Peng *et al.*, 2007), or a wrong spreader size (Beer *et al.*, 2000a).

c. Warm lateral compaction technique

Warm lateral compaction (WLC) was developed to improve GP flow while maintaining the ease of use of lateral compaction. WLC increases the homogeneity and density of the GP mass and maximized the advantages of both traditional lateral compaction and warm vertical compaction. Theoretically, maximized GP density leads to fewer voids and a better apical and coronal seal. In addition, the more GP in the canal will limit the space filled by a sealer (Collins *et al.*, 2006). In fact, WLC of GP is a modification of

CLC while, at the same time, thermoplasticizes the GP obturation mass. The principal difference between CLC and WLC is the use of a heated spreader in WLC versus a cold spreader in CLC (Kulild *et al.*, 2007). In general, with this technique a heated spreader is used to soften the GP point inside the canal, and then cold spreaders inserted to create sufficient space for accessory points. The process is repeated until the canal is properly filled (Ingle and Bakland, 2002b, Bergenholtz *et al.*, 2009b). The spreader may be heated by placing it in a hot bead sterilizer prior to insertion into the canal. Alternatively, the friction of ultrasonic vibration may be used to introduce heat into the root canal (Walmsley *et al.*, 2002). The use of a battery-operated Endotec thermal endodontic condenser has also been described (Nelson *et al.*, 2000). A radiographic evaluation study of sealing ability of different obturation techniques, showed that the best results was in WLC at apical and middle third where no voids were observed (Mittal *et al.*, 2002). In addition, it has been shown that the WLC result in a less leakage than CLC, thermo-mechanical compaction or low temperature injectable thermo-plasticized GP (Liewehr *et al.*, 1993, Nelson *et al.*, 2000).

d. Vertical compaction technique

Schilder in (1967) advocated vertical compaction with warm GP. He used heat to thermoplasticize GP and pluggers to pack the softened GP into the root canal. A proposed advantage of thermoplasticized vertical compaction technique is the ability to soften the GP which can thus be molded to various configurations within the root canal (Bowman and Baumgartner, 2002). This technique consists of fitting a GP point with a taper similar to the canal, short of the apical constriction and applying heat using a flame-heated carrier. Then, after the GP is softened, a plugger is used with apical pressure to produce a hydraulic force that moves the GP apically against the canal walls. Next, GP is added in small increments; each increment of GP is heated and softened and

packed vertically until the entire canal is filled (Walton and Torabinejad, 2002). There is another system described by Buchanan (1996) that used an electrically controlled heating system to ensure correct heating of the GP points after their placement within prepared root canals. This system combines the heating procedure with vertical compaction in one step, in order to obtain the correct pressure of the warm and soft GP to allow the homogenous distribution of the filling into the apical third of the root canal system. This device is called System-B Heat Source (Analytic Technology, Redmond,WA,USA) and its clinical application is related to a modified vertical compaction technique called continuous wave of condensation (Venturi *et al.*, 2002).

This technique provides the homogeneous, three-dimensional obturation with a microfilm of sealer surrounding the bulk of homogeneous GP (Mente *et al.*, 2007). Conversely, there are some disadvantages. It takes a long time during obturation and may result in vertical root fractures if an inappropriate plugger fitting and excessive compaction forces are used. Excessive heat of the plugger may result in injury to the operator, patient's lip, or periodontal ligament. Extrusion of filling materials into the periapical tissues is considered as another risk with this technique (Mente *et al.*, 2007).

In a study that compared System B and Touch 'n Heat source, the System B produced an acceptable obturation, compared with Touch 'n Heat source (Silver *et al.*, 1999). Using warm vertical compaction was found to achieve 10% higher healing rates versus CLC for teeth with previous apical periodontitis (Farzaneh *et al.*, 2004).

e. Injection technique

Yee *et al.* (1977) introduced a concept of root canal obturation in which GP is thermoplasticized and ejected out of a needle into the root canal system (Ibarrola, 2002). The Obtura II (Obtura/Spartan, Fenton, MO.) is a high-temperature thermoplasticized GP system that heats the GP pellet to 150-200℃ prior to delivery into the root canal

(Rahimi *et al.*, 2010). The warmed GP is expressed through a needle as fine as 25 gauge (0.5mm diameter), and the GP leaves the needle at approximately 70^{o}C (Bergenholtz *et al.*, 2009b). This system can be used to backfill or to fill the entire root canal (Weine, 2004). The advantages of an injection technique are similar to those of warm vertical compaction. It is also useful in filling internal resorptive details (Bergenholtz *et al.*, 2009b), as well as wide and irregular canals (Pitt-Ford, 2004). There are some disadvantages, which include: difficulty in controlling the level of the root filling (Ingle and Bakland, 2002b), shrinkage of the GP during cooling that may result in voids. For this reason, a segmental filling technique where small portions are injected and compacted with pluggers has been advocated (Bergenholtz *et al.*, 2009b). However, Yilmaz *et al.* (2009a) examined the sealing efficacy of different obturation techniques using fluid conductance and found that the greatest amount of leakage was found in system B/obtura II group and BeeFill, compared with SC and CLC groups. Some other, *in vivo* and *in vitro* studies, however, showed that Obtura II had a good adaptation to the canal wall and did not cause periodontal tissue injury (Tani-Ishii and Teranaka, 2003).

There is another system, Ultrafil, (Ultrafil, Hygenic Corporation, Akron, OH) of low-temperature 70 ℃ thermoplasticized GP, that seems also able to replicate the details of the root canal system (Al-Dewani *et al*., 2000). In a study that performed by Al-Dewani *et al*. 2000) using a dye penetration method, they found a better sealability with low-temperature thermoplasticized GP than lateral compaction in canals prepared with NiTi rotary instruments. Conversely, a study performed by Pérez Heredia *et al.* (2007) who looked at the apical seal of curved canal, indicated that the Ultrafil system and CLC technique with 0.06 or 0.02 tapered master points were equally effective in providing a seal.

f. Core carrier technique

Johnson in (1978) described a technique for full-length root canal obturation using a metal file coated with GP. This technique of coating a metal carrier with thermoplasticized GP became available under the name Thermafil (Tulsa Dental Products, Tulsa, OK) (Behnia and Mcdonald, 2001). Nowadays, the metal carriers have all been replaced by thermoplastic polymers, polysulfone and Vectra (Lipski and Wozniak, 2003). This carrier method may be considered as a low-temperature technique with little risk of overheating the periapical tissues (Whitworth, 2005). Thermafil is sized and tapered to match the taper (0.04/0.06) of the root canal files. The central carrier is uniformly coated with a layer of α-phase GP (Ingle and Bakland, 2002b). The system includes metallic size-verifiers, which is tried in the prepared canal and helps in selecting the correct Thermafil core carrier. It also includes a Thermaprep Plus heating system to provide a controlled heating environment (Hegde, 2008).

Using the Thermafil system, root canals can be filled with GP quickly and with a reduced effort, in a single procedure. On the other hand, following canal preparation to size 25 there is a relatively high occurrence of underfilling and with canal preparation to size 35, filling reaches the apex of the canal, but some cases of overfilling may occur (Beer *et al.*, 2006a). Extrusion of filling material was much more frequent with Thermafil obturation than with CLC technique (Schäfer and Olthoff, 2002). However, clinically, the Thermafil or CLC technique seem to show no significant difference in the treatment outcome (Chu *et al.*, 2005). Gençoglu *et al.* (2007) reported for a leakage study comparing Thermafil with System B in the absence of the smear layer, that there were no significant differences in the short-term leakage pattern 10 hours or 24 days, whereas in a long term pattern, leakage at 67 days was greater in the Thermafil group.

2.4. Evaluation of quality of root canal obturation

Root canal obturation has been considered as one of the important steps which influence RCT outcome (Torabinejad and Walton, 2009). 60% of RCT failures were associated with signs of incomplete obturation of the root canal (Ingle and Bakland, 2002b).

Presence of microscopic gap between the root canal filling material and the tooth allows leakage of oral fluids and potential percolation, followed by bacterial penetration and growth which could contribute to treatment failure (Nawal *et al.*, 2011). The root canal filling should be a full, homogenous mass that completely fills the prepared canal (Zahed, 2008). There are a variety of methods *in vitro* for evaluation of the sealing ability of root canal materials (Olga Onay *et al.*, 2006) and techniques (Ai-Ghamdi and Wennberg, 1994). The followings are some of these methods:

1. Dyes penetration

Grossman in 1939 reported a method in which the teeth were immersed in various types of dyes (eosin, methylene blue, black India ink, procion brilliant blue and others). Dye penetration is considered as a widely used method because it is easy to perform (Ver ssimo and Vale, 2006). However, the ability of dyes to penetrate a void is determined by its particle or molecule size, concentration and pH (Souza *et al.*, 2009a). Although dye penetration has been the most common method *in vitro* for examining the root canal fillings, it typically generates data with large standard deviation, and its clinical relevance has been questioned (Karagen ç *et al.*, 2006). In general, this method for detecting microleakage involves bathing the tooth in a dye solution after the external surfaces have been coated with a layer of impermeable varnish. Following a certain time interval, the specimens are removed and washed (De Munck *et al.*, 2005). Next, they are sectioned either longitudinally or transversely, or cleared for visual examination to measure the extent of dye infiltration around the filling materials and the linear

penetration of the dye is recorded (Lucena-Martin *et al.*, 2002, De Munck *et al.*, 2005, Verís simo and Vale, 2006). While methylene blue was considered as the most frequently used dye tracer (Souza *et al.*, 2009a), there are some dental materials that can affect its color stability, and reducing its optical density. Wu *et al.* (1998) have found that mineral trioxide aggregate (MTA) can cause high rates of substance alteration, thus affecting the use of dye for leakage test. Moreover, methylene blue is water soluble and can be difficult to observe its maximum penetration point in some cases (Schäfer and Olthoff, 2002). Although dye penetration methods are simple and easy (Verís simo and Vale, 2006), they require sample sectioning during processing and do not permit evaluation of sealing ability over time (De Bruyne *et al.*, 2005).

2. Fluid filtration or transportation

Fluid filtration method was first used by Greenhill & Pashley in 1981 (cited by Wu *et al.*, 2003b, De Bruyne *et al.*, 2005). It was utilized for studying the physiology of dentine, as well as the effects of various restorative procedures on dentine permeability. In endodontics, it is used to evaluate the sealiability of root canal obturation techniques (Pommel and Camps, 2001, Reid *et al.*, 2003). This method has numerous advantages over dye penetration methods; such as non-destruction of samples. Therefore, evaluation of apical and coronal sealing over a period of time can be acquired. The method can provide quantitative measurements, thus avoiding operator errors; the results, are precise, as small volumes can be recorded (Wu and Wesselink, 1993). It is more sensitive than dye penetration in detecting empty spaces along the root canal and is highly reproducible (Da Silva Neto *et al.*, 2007). In addition, fluid transport values indicate the diameter and the length of the void while dye penetration method indicates the length of the void only (Wu *et al.*, 2003b). However, there has been no

standardization in the use of this technique as the pressures used ranged from 10 to 20 psi, and the measurement time ranged from 1 min to 3 hr (Pommel and Camps, 2001).

3. Bacterial leakage methodology

In 1989, Kersten and Moorer reported that assessment of leakage by bacteria is capable of providing a more clinically and biologically relevant information (Timpawat *et al.*, 2001a) than dyes penetration tests (De-Deus *et al.*, 2008). On the other hand, if the sealer possesses antimicrobial activity, the outcome will be affected with this method (Maltezos *et al.*, 2006). This method did not simulate any conditions found in the oral cavity such as temperature changes, dietary influences, and salivary flow (De-Deus *et al.*, 2008). Moreover, there can be difficulty in determining the quantity of bacteria that reach the apical foramen (Brosco *et al.*, 2010). Therefore, results of bacterial leakage studies have to be a qualitative rather than quantitative. If only one bacterium passes through the obturated root canal, it may multiply in the enriched broth and cause turbidity (Vijay and Indira, 2009). Different bacteria have been used to assess the seal ability of the root canal obturation; e.g., *Enterococcus faecalis* which is frequently used, because it is part of the normal mouth flora and is frequently found in root canal failure cases (Timpawat *et al.*, 2001a). Other bacteria, *Proteus vulgaris* and *Staphylococcus epidermidis*, were used by Torabinejad *et al*. (1990) for an apical recontamination study. Shipper *et al.* (2004b) investigated the penetration of *Streptococcus mutans* and *Enterococcus faecalis* in teeth obturated with Resilon and teeth filled with GP and found that Resilon resisted the bacterial penetration better than GP for a period of 30 days. Other than bacteria, Torabinejad *et al.* (1995) used endotoxin to evaluate its penetration into root filled canal based on the hypothesis that endotoxins can also cause inflammation distant from the site of bacterial infection. Endotoxins were seen to pass

through the root canal obturated with GP and sealer in 31.5% of teeth (Carrat *et al.*, 2002).

4. Radiographic evaluation

Radiographic evaluation is the only method applicable clinically to assess the adaptation of the root filling to the canal wall (Amditis *et al.*, 1992). In general, a root canal is considered to be optimally obturated when a continuous radiopaque mass that is free from voids, adapts well to the canal wall and slightly short of the apex is observed on the radiograph (Kersten *et al.*, 1987). However, radiographs are 2-dimensional representation of 3-dimensional structures, and certain clinical and biologic features might not be reflected in the radiographic image. In addition, morphologic variations, surrounding bone density, x-ray angulations, and radiographic contrast can influence radiographic interpretation (Estrela *et al.*, 2008). In a study by Tani-Ishii and Teranaka (2003), root canal obturation with Obtura II was evaluated clinically and radiographically, and found the treatment outcome for roots with apical periodontitis was not dependent on the level of root filling in relation to the root apex.

5. Scanning electron microscopy

Scanning electron microscopy (SEM) has been widely used in dentistry to investigate the surface and microstructure of enamel (Orellana *et al.*, 2008), the efficacy in removal of filling materials (retreatment) (Scelza *et al.*, 2008) and the interface of sealer and dentine (Pameijer and Zmener, 2010). In general, tooth is bisected longitudinally (Gurbuz *et al.*, 2008) or horizontally at a right angle to the long axis (Shipper *et al.*, 2004a) and investigated under the SEM. The marginal adaptation under the SEM provides information that could be used as an indicator of the sealiability of the filling material (Torabinejad *et al.*, 1995). In a study performed by Garcia *et al.* (2009) using

SEM to evaluate apical obturation quality, they found that Resilon/Epiphany and GP/Sealer 26 had similar behaviour with an adequate sealing of the apical region, and a minimum amount of sealer presented in the canal.

2.5. Assessment of quality of root canal obturation

Methods that have been used in assessment of root canal obturation quality were:

1. Sectioning of the tooth

Tooth sectioning has been used in many studies to assess the quality of root canal obturation by either horizontal (cross-section) tooth sectioning (Lyroudia *et al.*, 2000, Lucena-Martin *et al.*, 2002, Gulsahi *et al.*, 2007, Ozawa *et al.*, 2009) or longitudinal tooth sectioning (Limkangwalmongkol *et al.*, 1992, Ahlberg *et al.*, 1995).

a. Horizontal sections

A key of clinical success is complete closure of the materials-dentine interface, especially in the apical part to accomplish the best apical seal (Guigand *et al.*, 2005). The goal of various filling techniques is to maximize the amount of GP in the canal space and minimize the amount of sealer, which in turn decrease the potential for gaps that occur due to sealer contraction or dissolution (Souza *et al.*, 2009b). Shrinkage and solubility of the sealer can result in a leaky obturation that affects the long-term success of RCT (Tasdemir *et al.*, 2009); so, it is best to rely as little as possible on sealers and more on core filling material (Guigand *et al.*, 2005). Therefore, the reduced ratio of sealer to gutta-percha may improve the long term seal provided by fillings material (Masudi and Pameijer, 2011). Study by Ng *et al.* (2008) in assessing factors that affect the outcome of non-surgical RCT, they found that the success rates (secondary RCT) for roots with satisfactory pre-existing root fillings (primary RCT-absence of voids) were 6% lower than those with unsatisfactory pre-existing root fillings for the secondary

RCT. Satisfactory root fillings was found to be associated with significantly higher success rates than unsatisfactory root fillings (Farzaneh *et al.*, 2004). Another study, found that voids in root fillings at the middle or apical thirds had significantly worse outcome than those with voids in the coronal third or those without voids (Cheung and Chan 2003). Many investigators evaluated the quality of root canal filling by calculating the percentage area of GP using cross-section methods. For instance, the percentage of GP-filled canal area (PGCA) for teeth prepared by S-ApeX (FKG Dentaire, La Chaux-de-Fonds, Switzerland) and GT (Dentsply/Tulsa Dental, Tulsa, OK) files, and obturated using a lateral compaction (LC) technique or combination of lateral and vertical compaction (LCVC) technique was measured after cross-sectioning of a root canal at 2, 4, 6, and 8 mm from the apex. The average PGCA values were 97.6 % GT/LC, 99.3% S-ApeX/LCVC and 98.5% GT/LCVC groups (Van Der Borden *et al.*, 2010). Souza *et al.* (2009b) studied the effect of filling technique and root canal area on the percentage of GP in canals obturated with CLC technique, and found that PGCA was lower at the 3 mm level compared to the 6 mm level. Another study calculated the PGCA at 2 and 4 mm from the apex, and found that the warm GP group produced significantly higher PGCA than the cold GP group at 4 mm from the apex only (Wu *et al.*, 2001). Study by Carvalho-Sousa *et al.* (2010) used this method (cross-section) to evaluate filling of created lateral canals at 4, 7 and 10 mm from the roots apex, which obturated using; continuous wave of condensation, thermomechanical and lateral compaction techniques, indicated that the thermoplasticized GP obturation techniques were better for filling lateral canals than lateral compaction. This evaluation method was used in conjunction with dye penetration for examining leakage at 3, 6 and 9 mm from the apex (Paqué *et al.*, 2006). Serial, cross-sections were also used to investigate the extent of dye penetration (aided by a stereomicroscope at 40× magnification) at intervals of 1 mm for the presence or absence of dye (Roggendorf *et al.*, 2007).

b. Longitudinal section

The longitudinal sectioning is probably the most widely used method to assess dye leakage, with liner measurements expressed in millimetres (Pesce *et al.*, 2007). In this method, the roots are bisected through the apex and in a direction approximately parallel to the long axis of the root. Next, the linear dye penetration is blindly measured on either side of the samples, from the apex to the point where the dye no longer penetrated the interface between the root canal walls and the root canal filling (Wang *et al.*, 2005). The disadvantages of this method are (i) that only the visible dye is measured and the total leakage of the sample cannot be assessed from that section (Solano *et al.*, 2005), (ii) the random choice of the cut axis may have a very low probability of picking up the section with most extensive leakage and thus may result in underestimating the leakage and recording unreliable data (Ver ́ssimo and Vale, 2006), and (iii) only one plane of the root canal filling can be examined, and any cracks and lateral and secondary canals are difficult to detect. Furthermore, more than one cut may be needed especially in a curved root (Limkangwalmongkol *et al.*, 1992).

2. Tooth clearing technique

This technique provides a three dimensional view of the root canal space relative to the external surface of the tooth and allows a thorough examination of the pulp chambers and root canals (Omer *et al.*, 2004). Rendering the teeth transparent enables visual evaluation of any linear dye leakage and of the adaptation of filling material to the irregularities of the root canal wall (Kytridou *et al.*, 1999). However, the clearing technique remains useful only as a teaching/research means with little or no clinical applicability (Omer *et al.*, 2004). In general, this technique is performed by decalcifying the teeth using an acid such as hydrochloric acid, followed by ascending concentration of ethanol to dehydrate the tooth (Whitworth and Baco, 2005). Next, the teeth are made

transparent by immersing them in methylsalicylate (Ibarrola, 2002, Al-Qudah and Awawdeh, 2006). Subsequently, the cleared teeth can be examined under stereomicroscope (Venturi *et al.*, 2005). However, tooth clearing technique has been criticized as unacceptable because the adaptation of root filling materials cannot be observed directly, and it can only be inferred from the amount of leakage detected (Pathomvanich and Edmunds, 1996). In a study of leakage that was assessed with this technique, there was no significant difference in coronal leakage between SC and CLC techniques (Tidswell *et al.*, 1994).

CHAPTER THREE

MATERIALS AND METHODS

3.1. Teeth collection and storage

Two hundred extracted single-rooted mandibular premolar teeth were collected regardless of age and race. The teeth were stored in 0.5% Chloramine-T trihydrate solution for one week. Ultrasonic scaler (DENSPLY® CAVITRON® BOBCAT® PRO, USA) was used to remove both calculus deposits and residual periodontal tissues. Throughout this study, the teeth were kept hydrated in distilled water to maintain dentine permeability. The storage solution was changed regularly every one week and the teeth were stored at 4^{o}C.

3.2. Selection of specimens

Following examinations and evaluations, a total of ninety six (96) out of the two hundred teeth was selected for the following inclusion and exclusion criteria:

The inclusion criteria were:

- Single root canal.
- Relatively straight root canal.
- Well developed root and completely formed canals.
- Patent foramen.
- Teeth with canal diameter smaller or equal to a size 15 K-file at 4-5 mm from the radiographic apex. In addition, an attempt was made to standardize the size and shape of the selected teeth (diameters at cervix were 5.0-5.5 mm mesiodistally and 7.5-8.0 mm buccolingually).

The exclusion criteria were:

- Teeth with more than one canal.
- Obstruction within the canal system.
- Internal or apical resorption.
- Teeth with cracks, or open apices.

- Teeth with pulp stone.
- Tooth with previous RCT.
- Teeth with canal diameter larger than a size 15 K-file at 4-5 mm from the radiographic apex.

The examination and evaluation procedures were done under 10× magnification using a stereomicroscope (OLYMPUS szx7, Olympus Corp., Tokyo, Japan) (Fig. 3.1). The teeth were then decoronated with a sectioning machine (METKON®- MICRACUT® 125 Low Speed Precision Cutter) (Fig. 3.2) at the level of 16 mm from the apex perpendicular to the long axis of the root canal to relatively standardize the root canal length in all specimens. With size 15 K-file (Colorinox, Dentsply Maillefer) in the root canal, at the apical foramen, a buccolingual and a mesiodistal radiograph were taken for each specimen.

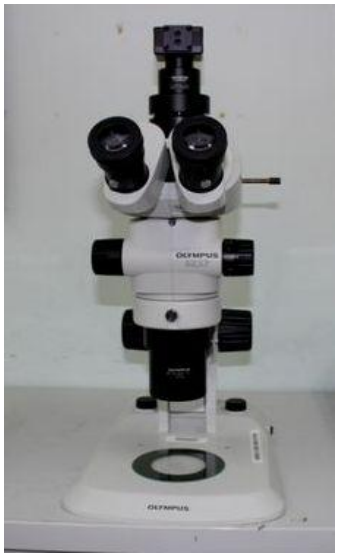


Figure 3.1: Stereomicroscope (OLYMPUS szx7, Olympus Corp., Tokyo, Japan)

Figure 3.2: Sectioning machine METKON®- MICRACUT® 125 Low Speed Precision Cutter

3.3. Preparation of root canal

Each tooth was mounted in an impression compound (Kemdent, Associated, Dental Products, Swindon, UK) except the apical part, to facilitate handling of specimen during root canal preparation and obturation procedures.

The working length was determined by insertion of size 15 K-file (Colorinox, Dentsply Maillefer) into the root canal until it was visible at the apical foramen, and then the rubber stop was adjusted to the coronal reference point. The file was then removed from the canal and the working length of each canal was calculated to 1.0 mm short of that position. All root canals were prepared with K-files (Colorinox) and Mtwo® NiTi rotary instrument (VDW, München, Germany) (Fig. 3.3).

The size 15 K-file was used to start the preparation, then the Mtwo® NiTi rotary instruments (VDW, München, Germany) were used according to the manufacturer's instructions to complete the root canal preparation. All canals were prepared up to size 35/0.04 as the master apical file (MAF). The Mtwo® NiTi rotary instruments were coated with Filecare® EDTA NiTi rotary files lubricant (VDW, München, Germany) before introduction into the root canal. All rotary instruments were used to the full working length of the canals, employing a cyclical in-out brushing motion. Each instrument was removed from the canal, when it could rotate freely at the apical terminus. Irrigation of the canal was performed after each instrument with 2.0 ml of 5.25% sodium hypochlorite (NaOCl) (Clorox™, Sdn. Bhd., Malaysia) delivered via a 5 ml syringe (Ultradent Products Inc., South Jordan, UT, USA) with a 27-gauge needle (Ultradent). Each individual instrument was used to prepare five root canals and then discarded. Apical patency was established throughout the preparation by passing a size 10 K-file, 1.0 mm beyond the apical foramen.

Once the root canal preparation was completed, each canal was then irrigated in the following sequence:

1) 3.0 ml 5.25% NaOCl (Clorox™, Sdn. Bhd., Malaysia) for one minute.
2) 3.0 ml 18% ethylene-diamine-tetra-acetic acid (EDTA) (Ultradent Products Inc., South Jordan, UT, USA) for one minute to remove the smear layer.
3) 3.0 ml distilled water for one minute to ensure complete removal of NaOCl and EDTA from the root canal.

The final size of the canal was confirmed by inserting a file, one size larger than the MAF (#35) to approximately 1.0 mm shorter than the working length.

All specimens were then stored in distilled water to prevent dehydration of specimens prior to obturation.

All preparations were completed by a single operator (to avoid inter-operator variability).

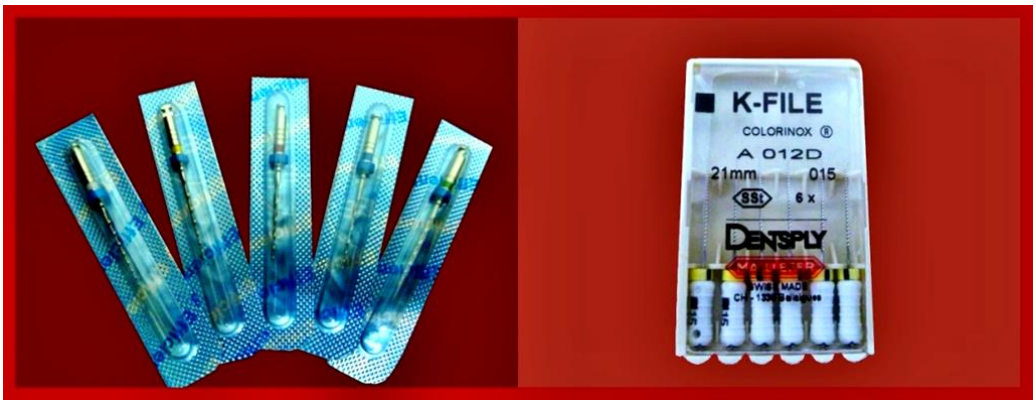


Figure 3.3: Mtwo® NiTi rotary instrument (VDW, München/Germany) and size 15 K-file (Colorinox, Dentsply Maillefer).

3.4. Compatibility between finger spreader and accessory points (GP and ER)

ISO standardized size 25 and 30 finger spreaders (VDW, München, Germany), size 25/0.02 GP accessory points (VDW, München, Germany) and size 25/0.02 ER accessory points (Ultradent Products Inc., South Jordan, UT, USA) (Fig. 3.4) were selected for the CLC and WLC techniques. The compatibility between the finger spreaders and accessory points were measured at 1mm (D1), 3mm (D2) and 6mm (D3) from their tip using OLYMPUS szx7 Stereomicroscope (at 10× magnification) and Cell^ D software (OLYMPUS Soft Imaging Solutions GmbH, 2008, Munster).

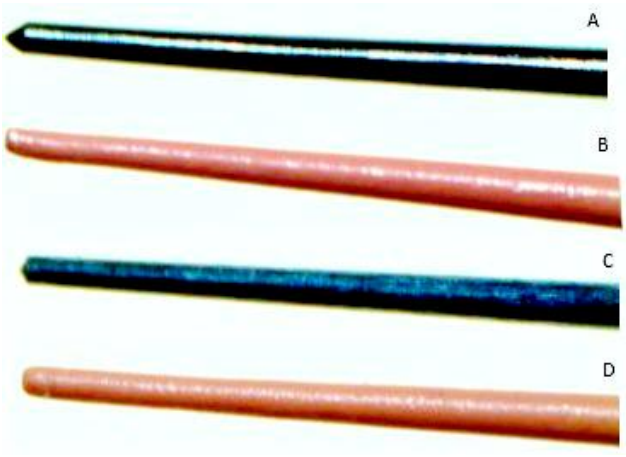


Figure 3.4: (A) Finger spreader size 30, (B) Size 25/0.02 GP accessory point, (C) Finger spreader size 25 and (D) Size 25/0.02 ER accessory point.

3.5. Obturation of root canal

The total sample of 96 single-rooted mandibular premolars was randomly divided into two groups of 48 (2×48 teeth). One group represented canals filled with GP points (VDW, München, Germany) (Fig. 3.5) and AH Plus® sealer (Dentsply DeTrey, Konstanz, Germany) (Fig. 3.6), whereas the second group was filled with ER points (Fig. 3.7) and ER sealer (Fig. 3.8) (Ultradent Products Inc., South Jordan, UT, USA). Each group was further divided into three subgroups of sixteen teeth, according to the techniques that were used, namely, CLC, WLC and SC. A summary of the groups and subgroups of canal obturation materials and techniques is illustrated as in Fig. 3.9.

Figure 3.5: Sizes 25/0.02, 35/0.02 and 35/0.04 GP points (VDW, München. Germany).

Figure 3.6: AH Plus® sealer (Dentsply DeTrey, Konstanz, Germany).

Figure 3.7: Sizes 25/0.02, 35/0.02 and 35/0.04 ER points (Ultradent Products Inc., South Jordan, UT, USA).

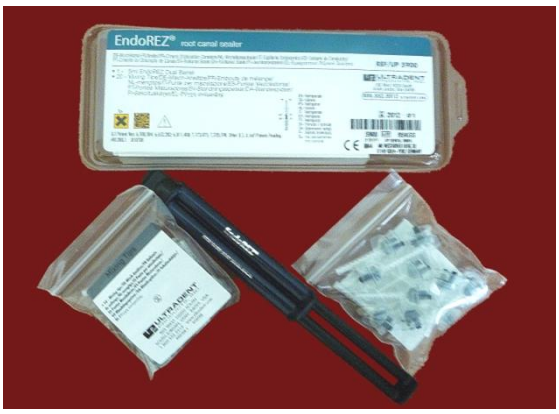


Figure 3.8: ER sealer (Ultradent Products Inc., South Jordan, UT, USA).

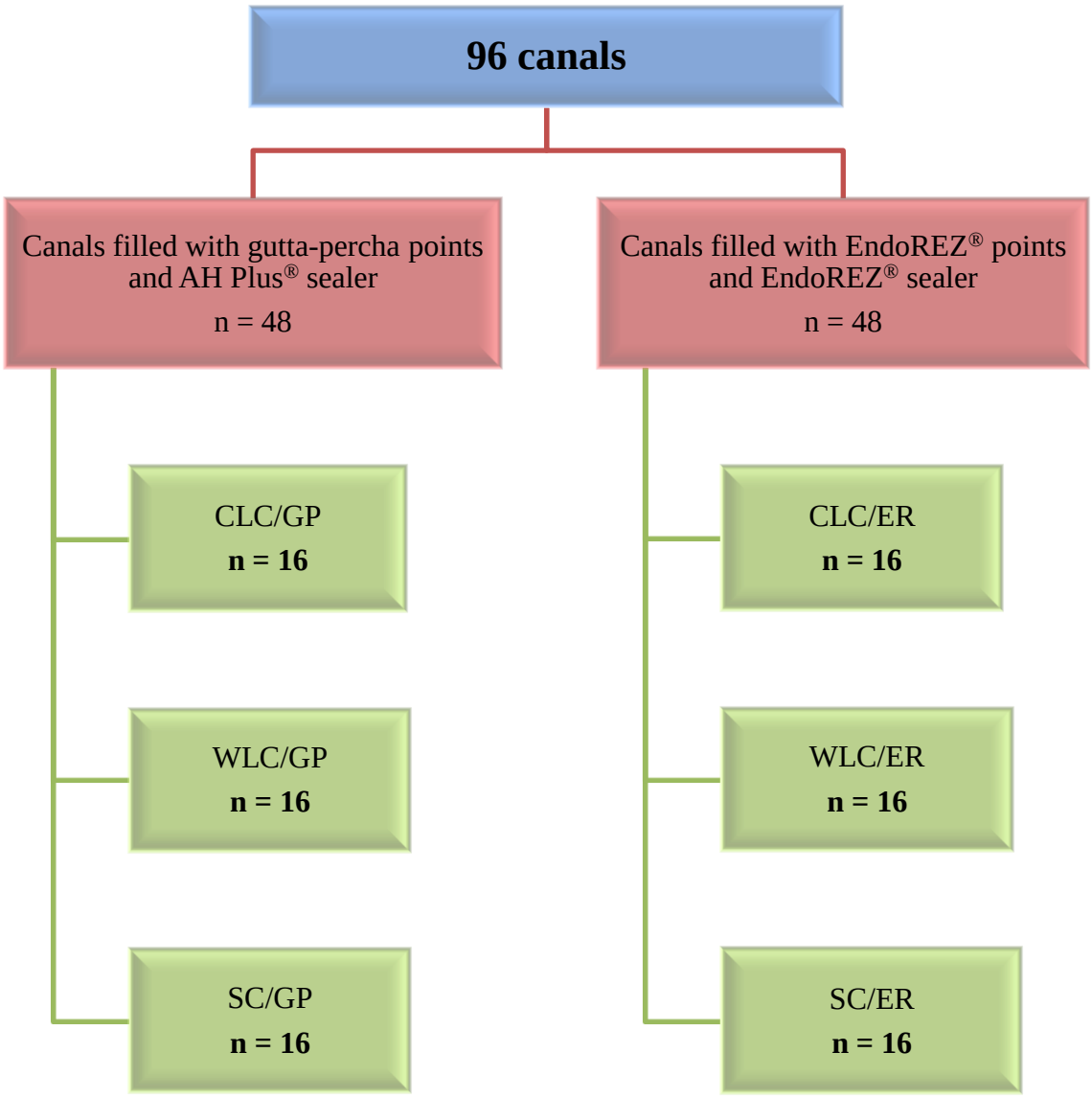


Figure 3.9: Summary of groups and subgroups of canal obturation materials and techniques.

3.5.1. Cold lateral compaction/gutta-percha subgroup (CLC/GP)

1. Each canal was dried with paper points (VDW, München, Germany) prior to the application of sealer and obturation materials.
2. An ISO standardized GP point size 35/0.02 (VDW, München, Germany) which fitted to the working length and evidenced by "tug back" was chosen as the master cone. A pair of locking tweezers was used to hold the GP point to the working length in the canal.
3. Once the master cone was selected, a finger spreader, either size 25 or 30 (VDW, München, Germany) that passed down to within 1.0 mm of the end point of preparation was selected.
4. A paper point, size 35, was then coated with AH Plus® sealer (Dentsply DeTrey, Konstanz, Germany) and inserted to the working length and smeared onto the canal walls. The paper point was then removed.
5. The master cone (from step 2) was then inserted to the working length.
6. The selected finger spreader was inserted alongside the master cone with controlled apical pressure until it reached to within 1.0 mm from the working length. Apical pressure was applied for 10 seconds in a constant manner and the master cone was laterally compacted against the wall. Following this, the finger spreader was removed by turning it 180 º to prevent dislodgement of the compacted cone.
7. The first GP accessory point size 25/0.02 was held with locking tweezers at the point corresponding to the length of the spreader that was inserted into the canal. The GP accessory point was then inserted into the space created by the spreader. If the accessory point cannot pass to that length, a larger finger spreader size 30 was used to permit the insertion of the previously selected accessory point.

8. The selected spreader was then cleaned with a piece of gauze and reinserted into the canal as described in step 6, but to 1.0 mm shorter than the previous length.
9. The sequence of spreader application and GP accessory point insertion continued, until the spreader could only penetrate 2-3 mm coronally.
10. The heat plugger standard of BeeFill® 2-in-1 device (VDW, München, Germany) (Fig. 3.10) was used to sear off the points from the canal orifice. Following this, the GP in the coronal portion was vertically compacted using a root canal plugger, size 4.
11. The access cavity was then restored with IRM (Dentsply Caulk, Milford, USA).

Figure 3.10: Heat plugger standard of BeeFill® 2-in-1 device (VDW, München, Germany)

3.5.2. Cold lateral compaction/EndoREZ® subgroup (CLC/ER)

1. Each canal was dried with paper points (VDW, München, Germany) prior to the application of sealer and obturation materials.
2. An ISO standardized ER 0.02 point size 35/0.02 (Ultradent Products Inc., South Jordan, UT, USA) which fitted to the working length and evidenced by "tug back" was chosen as the master cone. A pair of locking tweezers was used to hold the ER point to the working length in the canal.
3. Once the master cone was selected, both finger spreader either size 25 or 30 (VDW, München, Germany) that passed down to within 1.0 mm of the end point of preparation was selected.
4. The ER root canal sealer (Ultradent) was applied in the root canal according to the manufacturer's instruction by inserting delivery NaviTip (Ultradent) into root canal to within 2-4 mm short of the apex. The tip was withdrawn slowly while expressing ER sealer coronally to the canal orifice.
5. A paper point, size 35, was then inserted to the working length and smeared onto the canal walls to remove excess sealer from the canal, and then the paper point was removed.
6. The master cone (from step 2) was then inserted to the working length.
7. The subsequent procedures were the same as those in section 3.5.1 (step 6-10), except that ER accessory points were used instead of GP accessory points.
8. The coronal surface was then light cured for 40 seconds, following the manufacturer's instructions, to produce an immediate coronal seal.
9. The access cavity was then restored with IRM (Dentsply Caulk, Milford, USA).

3.5.3. Warm lateral compaction/gutta-percha subgroup (WLC/GP)

1. Each canal was dried with paper points (VDW, München, Germany) prior to the application of sealer and obturation materials.
2. An ISO standardized GP point size 35/0.02 (VDW, München, Germany) which fitted to the working length and evidenced by "tug back" was chosen as the master cone. A pair of locking tweezers was used to hold the GP point to the working length in the canal.
3. Once the master cone was selected, a finger spreader, either size 25 or 30 (VDW, München, Germany) that passed down to within 1.0 mm of the end point of preparation was selected.
4. The heat plugger standard of BeeFill® 2-in-1 device (VDW, München, Germany) that passed down to within 2.0 mm of working length was selected and the rubber stop was used for depth control.
5. A paper point, size 35, was coated with AH Plus® sealer (Dentsply DeTrey, Konstanz, Germany) and then inserted to the working length and smeared onto the canal walls. The paper point was then removed.
6. The master cone (from step 2) was then inserted to the working length.
7. The selected heat plugger standard of BeeFill® 2-in-1 device was activated to 100^{o}C (the lowest power setting).
8. The heat plugger is gently forced apically and laterally into the canal alongside the master cone, at 2.0 mm short of the working length.
9. The heat plugger was then gently withdrawn.
10. The selected finger spreader was then inserted alongside the thermoplasticized master cone with controlled apical pressure until it reached to within 1.0 mm from the end point of obturation. Apical pressure was applied for 10 seconds in a constant manner and the master cone was laterally compacted against the wall.

Then, the finger spreader was removed by turning it 180^{o} to prevent dislodgement of the compacted cone as the thermoplasticized master cone cooled down.

11. The first accessory GP point size 25/0.02 was held with locking tweezers at a point corresponding to the length of the spreader that was inserted into the canal. Then, it was inserted into the space created by the spreader. If the accessory point cannot pass to that length, a larger finger spreader size 30 was used to permit the insertion of the previously selected accessory point.
12. The heat plugger standard of BeeFill® 2-in-1 device and selected finger spreader were then cleaned with a piece of gauze and reinserted into the canal as described in step 7 to 10, but to 1.0 mm shorter than the previous length.
13. The sequence of heat plugger and spreader application (heat and cold) and GP accessory point insertion continued, until the finger spreader could only penetrate 2-3 mm coronally.
14. The heat plugger standard of BeeFill® 2-in-1 device (VDW, München, Germany) was used to sear off the points from the canal orifice. Following this, the GP in the coronal portion was vertically compacted using a root canal plugger, size 4.
15. The access cavity was then restored with IRM (Dentsply Caulk, Milford, USA).

3.5.4. Warm lateral compaction/EndoREZ® subgroup (WLC/ER)

1. Similar sequence of procedures in section 3.5.3 (step 1-4) were followed except that in step 2 ER master cone (size 35/0.02) was used instead of GP master cone.
2. Then, ER root canal sealer (Ultradent) was applied in the root canal according to the manufacturer's instruction by inserting delivery NaviTip (Ultradent) into root canal to within 2-4 mm short of the apex. The tip was withdrawn slowly while expressing ER sealer coronally to the canal orifice.

3. A paper point, size 35, was then inserted to the working length and smeared onto the canal walls to remove excess sealer from the canal, and then the paper point was removed.
4. The ER master cone was then inserted to the working length.
5. The subsequent procedures were the same as those in section 3.5.3 (step 7-14), except that the ER accessory points were used instead of GP accessory points.
6. The coronal surface was then light cured for 40 seconds, following the manufacturer's instructions, to produce an immediate coronal seal.
7. The access cavity was then restored with IRM (Dentsply Caulk, Milford, USA).

3.5.5. Single cone technique/gutta-percha subgroup (SC/GP)

1. Each canal was dried with paper points (VDW, München, Germany) prior to the application of sealer and obturation materials.
2. An ISO standardized GP point size 35/0.04 (VDW, München, Germany) which fitted to the working length and evidenced by "tug back" was chosen as the master cone. A pair of locking tweezers was used to hold the GP point to the working length in the canal.
3. A paper point, size 35, was coated with AH Plus® sealer (Dentsply DeTrey, Konstanz, Germany) and then inserted immediately to the working length and smeared onto the canal walls. The paper point was then removed.
4. The master cone was then inserted to the working length.
5. The GP accessory points size 25/0.02 were inserted passively alongside the master cone.
6. The heat plugger standard of BeeFill® 2-in-1 device (VDW, München, Germany) was used to sear off the GP points from the canal orifice. Then, the GP in the coronal portion was vertically compacted using a root canal plugger, size 4.

7. The access cavity was then restored with IRM (Dentsply Caulk, Milford, USA).

3.5.6. Single cone technique/EndoREZ® subgroup (SC/ER)

1. Similar sequences of procedures in section 3.5.5 (step 1-2) were followed except that ER master cone was used instead of GP master cone.
2. The ER root canal sealer (Ultradent) was applied in the root canal according to the manufacturer's instruction by inserting delivery NaviTip (Ultradent) into root canal to within 2-4 mm short of the apex. The tip was withdrawn slowly while expressing EndoREZ® coronally to the canal orifice.
3. A paper point, size 35, was then inserted immediately to the working length and smeared onto the canal walls to remove excess sealer from the canal, and then the paper point was removed.
4. The subsequent procedures were the same as those in section 3.5.5 (step 4-6), except that ER accessory points used instead of GP accessory points.
5. The coronal surface was light cured for 40 seconds, following the manufacturer's instruction, to produce an immediate coronal seal.
6. The access cavity was then restored with IRM (Dentsply Caulk, Milford, USA).

3.6. Evaluation of obturation quality of GP and ER

3.6.1. Assessment of extrusion of filling materials

Under stereomicroscope at 10× magnification, any extruded root filling material through the apical foramen was assessed and scored as either Yes or No.

Each specimen was then removed from the impression compound. Next, a buccal and a mesial radiograph were taken for all specimens and placed on an X-ray viewer and viewed without magnification i.e. only naked eyes were used. The obturation was considered acceptable, if it appeared dense and without noticeable voids. If it was deemed unacceptable, the specimen was discarded and replaced with a new one.

All specimens were kept in an incubator (Memmert GmbH, Schwabach, Germany) at 37^{o}C in 100% humidity for one week, to allow complete setting of the sealer.

3.6.2. Assessment of percentage of core filling material and sealer + voids

Initially, each specimen was fixed in a plastic cuvette by using baseplate wax, and then the specimens were embedded in epoxy resin (Mirapox; Miracon, Malaysia). The epoxy resin was allowed to set for 24 hrs. Next, the specimens were ground from the root tip with a grinding machine (Twin Wheel Grinding/Polishing Machine, Coventry, England) (Fig. 3.11) until the obturation material was visible.

Figure 3.11: Twin Wheel Grinding/Polishing Machine, Coventry, England

Fifteen specimens in each subgroup were then sectioned serially in horizontal direction perpendicular to the long axis in a sectioning machine (METKON®- MICRACUT® 125 Low Speed Precision Cutter) using a diamond rotary blade with copious coolant irrigation at four levels; L1 (1 mm), L3 (3 mm), L6 (6 mm) and L9 (9 mm) from the obturator terminus. The thickness of the blade is 0.3 mm. Thus, the first cut was made 1.3 mm from the obturator terminus. Consequently, the other three cuts were made at 3.3 mm, 6.3 mm and 9.3 mm from the obturator terminus (Fig. 3.12). A large holder was used to hold the diamond blade to reduce as much as possible the movement of the diamond blade during specimen sectioning.

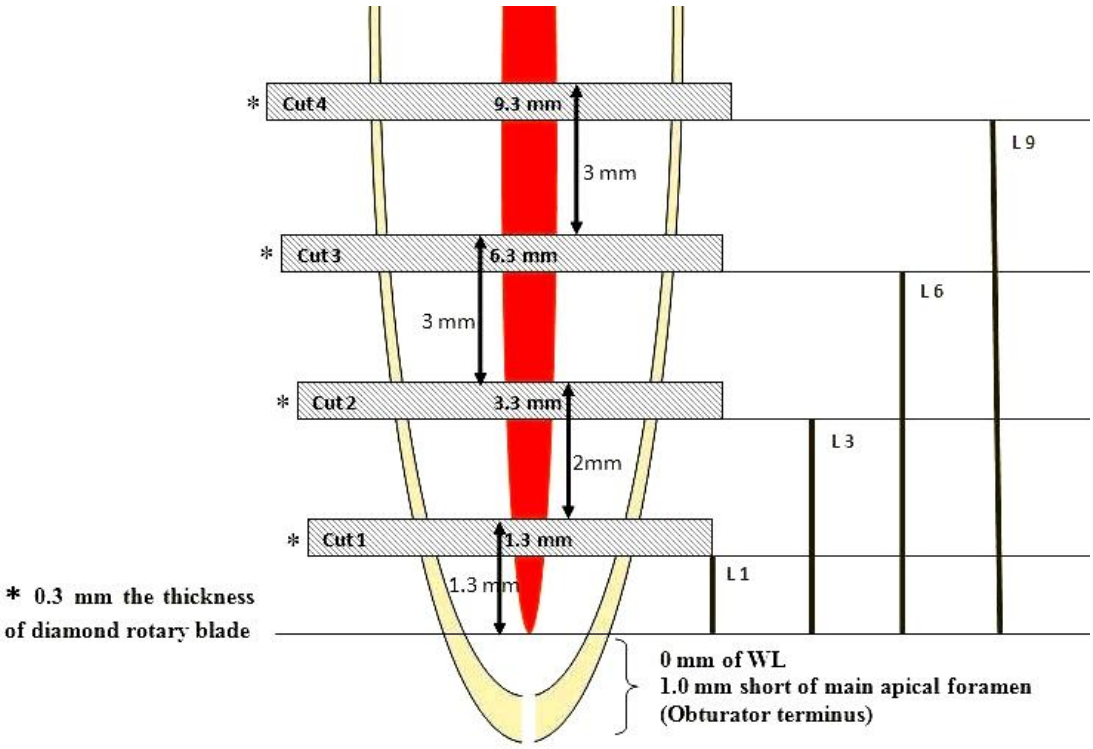


Figure 3.12: Diagram illustrates serial sections in each specimen at 4 different levels: L1, L3, L6 and L9

All sections were then viewed under the stereomicroscope (OLYMPUS szx7) at 40× magnification and micrographs were obtained.

The area occupied by the core filling material (GP or ER) was determined using Cell^ D software (OLYMPUS Soft Imaging Solutions GmbH, 2008, Munster).

The measurement of the area occupied by filling material (GP or ER) for each image was measured by drawing a line around the area of interest by clicking the left button of the mouse which was considered as the first point of the line. The mouse was then dragged around the area of interest until the last point meets up the first. The left button of the mouse was then released and the right button was clicked to form a closed loop and, at the same time, provide the measurement in (mm^2) of the area of interest in GP group (Fig. 3.13) and ER group (Fig. 3.14). This process of measurement was repeated three times, and the mean measurement was calculated. Then, the ratio of sealer + voids to root canal area was calculated for each section. Next the percentages of core filling material and sealer + void filled areas were calculated as a measure of obturation quality.

All horizontal sections were stored in distilled water in between measurements.

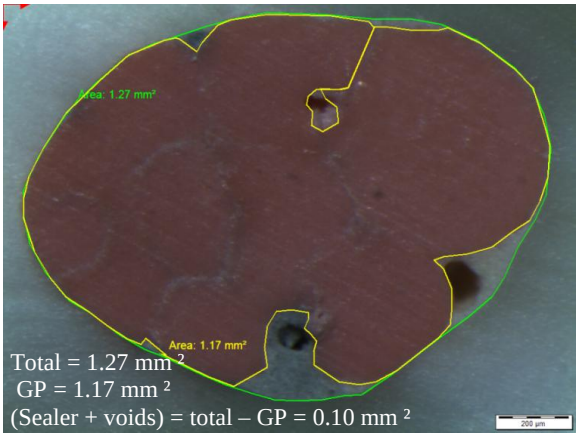


Figure 3.13: Cross-section at L9 of CLC/GP. Green line represents total canal area, whereas yellow line represents area of GP.

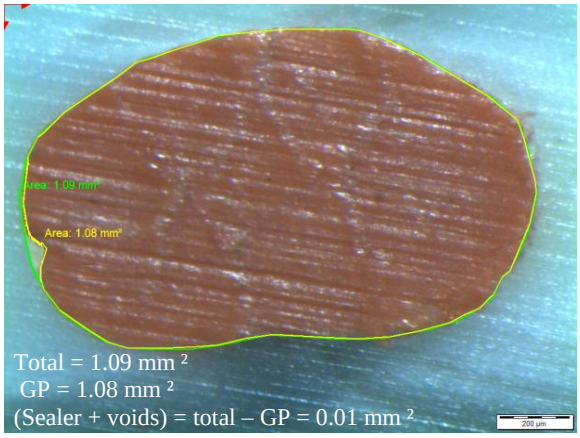


Figure 3.14: Cross-section at L9 of CLC/ER. Green line represents total canal area, whereas yellow line represents area of ER points.

3.6.3. Reliability test

Intra-examiner reliability was assessed by re-evaluating randomly 10 % of all horizontal sections on two occasions at one week intervals and without the knowledge of the previous readings. The canal area occupied by core filling material (GP or ER) in 36 randomly selected horizontal sections was measured and the percentage of core filling material (GP or ER) was calculated. Data collected was then statistically analysed. Based on the variance of the mean, the variation for each repeated measurement was calculated and then the total variations were obtained. The mean of percentage of canal area occupied by core filling material in GP and ER groups is presented in [Table F (Appendix II) pg 135]. The reliability test [Table G (Appendix II) pg 136] has shown that there was a correlation between the first and second measurements. Variations in measurements of the canal area occupied by core filling material (GP or ER) were negligible.

3.6.4. Assessment of adaptation

Scanning electron microscope (Field-emission gun scanning electron microscope) (FESEM) (Low Vacuum Operating Mode) (Fig. 3.15) was used to assess adaptation of the filling materials to the root canal wall based on formation of gap, if any.

One sample from each technique was selected randomly. A central line was drawn along the longitudinal axis of the specimen that was embedded in epoxy resin. The specimen was then sectioned longitudinally. Next, the specimen was mounted on an aluminum stub using carbon tape as an adhesive. Subsequently, the specimen was mounted on the specimen stage inside the chamber. The specimen was not coated prior to analysis. Imaging was done under 10 and 15 KV accelerating voltage at magnification between 200× to 4000× using back-scattered and secondary electron signals.

Figure 3.15: Field-emission gun scanning electron microscope (FESEM).

3.7. Data analysis

Data were analysed using SPSS, version 12 and the tests chosen were as follow:

3.7.1. Extrusion of core filling material through the apical foramen

A chi-square test was used to compare extrusion between obturation materials (GP and ER) in each obturation technique.

3.7.2. Percentage of canal area occupied by core filling material (GP or ER) and sealer (AH Plus® or EndoREZ®) + voids

- Two-way repeated measure was conducted to explore the impact of two different materials (GP and ER) in each technique (CLC, WLC and SC), as measured by the percentage of core filling material and (sealer + voids) at different levels (L1, L3, L6 and L9).

- Multiple comparisons were done using Mann-Whitney test to identify any difference in the percentage of core filling material and (sealer + voids) at each level between materials (GP and ER) in each technique (CLC, WLC or SC).
- Multiple comparisons between percentages of core filling material at different levels in each technique (CLC, WLC or SC) for GP group were done using Wilcoxon Signed Ranks test.
- Multiple comparisons between percentages of core filling material at different levels in each technique (CLC, WLC or SC) for ER group were done using Wilcoxon Signed Ranks test.
- Multiple comparisons between percentages of (sealer + voids) at different levels in each technique (CLC, WLC or SC) for GP group were done using Wilcoxon Signed Ranks test.
- Multiple comparisons between percentages of (sealer + voids) at different levels in each technique (CLC, WLC or SC) for ER group were done using Wilcoxon Signed Ranks test.

The significance level was set at $P < 0.05$ for all the above tests.

CHAPTER FOUR

RESULTS

4.1. Compatibility between finger spreaders and accessory points (GP and ER)

The raw data on the diameter measurements for twelve finger spreaders, size 25 and 30, GP accessory points 25/0.02 (VDW, München, Germany) and ER accessory points 25/0.02 (Ultradent Products Inc., South Jordan, UT, USA) is available in [Table A (Appendix II) pg 130].

Table 4.1 displayed the mean diameter at 1.0 mm (D1), 3.0 mm (D2) and 6 mm (D3) from the tip of finger spreader sizes, 25 and 30, and accessory points size 25/0.02 (GP and ER).

Table 4.1: Mean diameter of finger spreaders and accessory points

	D1	D2	D3
Finger spreader size 25	0.25	0.29	0.34
Finger spreader size 30	0.29	0.33	0.39
GP accessory points size 25	0.28	0.32	0.38
ER accessory points size 25	0.23	0.28	0.36

At all levels, mean diameter of GP accessory points 25/0.02 appeared slightly smaller than the mean diameter of finger spreader size 30, and slightly larger than the mean diameter of finger spreader size 25. Whereas, the mean diameter of ER accessory points 25/0.02 appeared smaller than the mean diameter of both finger spreaders, sizes 25 and 30, at all levels except at D3.

4.2. Post-obturation radiographic evaluation

In this study, from radiographic evaluation, it was found that two specimens from each group were unacceptable and were replaced by two new specimens that obturated using respective materials and techniques accordingly.

4.3. Assessment of extrusion of core filling material (GP or ER) through the apical foramen

4.3.1. Assessment of extrusion of filling material in CLC technique

The raw data on the assessment of extrusion of filling material for each specimen is shown in the [Table B (Appendix II) pg 131].

There were two out of sixteen canals showing extrusion in the GP group, whereas one out of sixteen canals showed extrusion in the ER group (Table 4.2). Fisher's exact test was used instead of chi-square test due to the assumption of expected count not being met, and the number of expected cell size was less than five. There was no significant difference between extrusion of filling material and type of materials used (*P*=1.000).

Table 4.2: Assessment of extrusion of filling material in CLC technique

			Extrusion		Total	*P* value*
			Yes	No		
Materials	GP	Count	2	14	16	1.000
		% within materials	12.5%	87.5%	100%	
	ER	Count	1	15	16	
		% within materials	6.3%	93.8%	100%	
Total		Count	3	29	32	
		% within materials	9.4%	90.6%	100%	

*Fisher's Exact test

4.3.2. Assessment of extrusion of filling material in WLC technique

The raw data on the assessment of extrusion of filling material for each specimen is shown in the [Table B (Appendix II) pg 131].

There were three out of sixteen canals showing extrusion in the GP group, whereas two out of sixteen canals showed extrusion in the ER group (Table 4.3). Fisher's exact test was used instead of chi-square test due to the assumption of expected count not met and the number of expected cell size was less than five. There was no significant difference between extrusion of filling material and type of materials used (*P*=1.000).

Table 4.3: Assessment of extrusion of filling material in WLC technique

			Extrusion		Total	*P* value*
			Yes	No		
Materials	GP	Count	3	13	16	1.000
		%within materials	18.8%	81.3%	100%	
	ER	Count	2	14	16	
		% within materials	12.5%	87.5%	100%	
Total		Count	5	27	32	
		% within materials	15.6	84.4%	100%	

*Fisher's Exact test

4.3.3. Assessment of extrusion of filling material in SC technique:

The raw data on the assessment of extrusion of filling material for each specimen is shown in the [Table B (Appendix II) pg 131].

With the SC technique no apical extrusion was found for both materials, GP and ER.

4.4. Percentage of canal area occupied by core material (GP or ER) and sealer (AH Plus® or ER) + voids using different obturation techniques

4.4.1. Percentage of the core filling material in CLC technique

The data of percentage of core filling material for GP and ER groups at different levels (L1, L3, L6 and L9) are shown in [Table C (Appendix II) pg 132]. Initially, two-way repeated measure test was conducted to explore the impact of the two materials (GP and ER), as measured by the percentage area of core filling material at L1, L3, L6 and L9. There was statistically significant interaction between the materials and levels (*P*=0.032). Therefore, the univariate one-way repeated measure was used. However, the assumption was not normal [(Fig. 1 and 2) Appendix II pages 137 and 138]. Thus, non-parametric Friedman test was used to identify any difference in the percentage area of core filling material (GP or ER) between levels (Table 4.4).

Table 4.4: Mean percentage of core filling material at different levels in CLC technique

Materials	Levels	N	Mean (SD)	*P* value[a]
GP	L1	15	79.45 (10.741)	**0.000**
	L3	15	91.80 (5.254)	
	L6	15	91.57 (6.805)	
	L9	15	89.87 (6.812)	
ER	L1	15	91.32 (8.582)	0.068
	L3	15	95.90 (6.054)	
	L6	15	95.40 (5.369)	
	L9	15	94.60 (4.239)	

a-Friedman test

Table 4.4 shows that there was a significant difference in the mean percentage of GP core filling material between different levels **(*P*=0.000)**. On the other hand, there was no significant difference in the mean percentage of ER core filling material between different levels (*P*=0.068).

Therefore, Wilcoxon test was used to identify where the significant difference lies between different levels in GP group (Table 4.5).

Table 4.5: Multiple comparisons between percentages of core filling material at different levels in CLC/GP

	Levels	Difference between levels	*P* value[a]
GP	L3-L1	12.35	**0.012**
	L6-L1	12.12	**0.012**
	L9-L1	10.42	**0.036**
	L6-L3	-0.23	3.744
	L9-L3	-1.93	1.392
	L9-L6	-1.7	1.182

a-Wilcoxon test used with *P* value after Bonferroni correction was done
Level of significant set at *P*<0.05

Table 4.5 shows that the mean percentage of GP core filling material at L3, L6 and L9 levels were significantly higher than L1 **(*P*=0.012)**, **(*P*=0.012)** and **(*P*=0.036)** respectively. The difference between the more coronal levels (L3, L6 and L9) was not significant.

In order to identify any difference between the two materials, Mann-Whitney test was performed (Table 4.6).

Table 4.6: Multiple comparisons of percentage of core filling material between GP and ER groups using CLC

Dependent variables	GP median (IQR)	ER median (IQR)	*P* value[a]
L1	83 (19)	94 (16)	**0.005**
L3	93 (10)	100 (6)	**0.023**
L6	93 (15)	97 (8)	0.110
L9	92 (10)	94 (7)	0.051

a-Mann Whitney test
Level of significant set at *P*<0.05
IQR= Interquartile range

Table 4.6 shows that the percentage of core filling material was higher for ER group at all levels than GP group with a significant difference lies at L1 and L3 levels **(*P*=0.005)** and **(*P*=0.023)** respectively.

4.4.2. Percentage of the core filling material in WLC technique

The percentage of core filling material for GP and ER groups at different levels (L1, L3, L6 and L9) is shown in [Table D (Appendix II) pg 133]. Initially, two-way repeated measure was conducted to explore the impact of the two materials (GP and ER), as measured by the percentage area of core filling material at L1, L3, L6 and L9. There was no statistically significant interaction between the materials and levels (*P*=0.108). However, the assumption was not normal [Fig. 3 (Appendix II) pg 139]. Therefore, univariate one-way repeated measure was conducted. The assumption for residuals was not normal [Fig. 4 and 5 (Appendix II), pg 140 and 141]. Thus, non-parametric Friedman test was used to identify any difference in the percentage area of core filling material (GP or ER) between different levels (Table 4.7).

Table 4.7: Mean percentage of core filling material at different levels in WLC technique

Materials	Levels	N	Mean (SD)	*P* value
GP	L1	15	88.46 (9.001)	**0.014**
	L3	15	96.13 (4.809)	
	L6	15	98.50 (1.955)	
	L9	15	96.73 (2.789)	
ER	L1	15	95.56 (6.621)	**0.027**
	L3	15	99.53 (1.246)	
	L6	15	99.60 (1.056)	
	L9	15	99.13 (1.246)	

a-Friedman test

Table 4.7 shows that there was a significant difference in the mean percentage of GP core filling material between different levels **(*P*=0.014)**. In addition, there was a significant difference in the mean percentage of ER core filling material between different levels **(*P*=0.027)**.

Therefore, Wilcoxon test was used to identify where the significant difference lies between different levels in GP group (Table 4.8) and ER group (Table 4.9).

Table 4.8: Multiple comparisons between percentages of core filling material at different levels in WLC/GP

	Levels	Difference between levels	*P* value[a]
GP	L3-L1	7.67	0.084
	L6-L1	10.04	**0.018**
	L9-L1	8.27	0.06
	L6-L3	2.37	0.444
	L9-L3	0.6	5.574
	L9-L6	-1.77	0.378

a-Wilcoxon test used with *P* value after Bonferroni correction was done
Level of significant set at *P*<0.05

Table 4.8 shows that the mean percentage of GP core filling material at L6 was significantly higher than L1 **(*P*=0.018)**. However, the comparisons for other levels were not significant.

Table 4.9: Multiple comparisons between percentages of core filling material at different levels in WLC/ER

	Levels	Difference between levels	*P* value[a]
ER	L3-L1	3.97	0.168
	L6-L1	4.04	0.168
	L9-L1	3.57	0.216
	L6-L3	0.07	4.71
	L9-L3	-0.4	1.818
	L9-L6	-0.47	0.396

a-Wilcoxon test used with *P* value after Bonferroni correction was done
Level of significant set at *P*<0.05

Table 4.9 shows that the mean percentage of ER core filling material was not statistically significant between different levels.

In order to identify any difference between the two materials, Mann-Whitney test was performed (Table 4.10).

Table 4.10: Multiple comparisons of percentage of core filling material between GP and ER groups using WLC

Dependent variables	GP median (IQR)	ER median (IQR)	*P* value[a]
L1	88 (18)	100 (9)	**0.029**
L3	97 (6)	100 (0)	**0.006**
L6	100 (3)	100 (0)	0.055
L9	97 (4)	100 (2)	**0.007**

a-Mann Whitney test
Level of significant set at $P<0.05$
IQR= Interquartile range

Table 4.10 shows that the percentage of core filling material was higher for ER group at L1, L3 and L9 than GP group with a significant differences lies at L1, L3 and L9 **(*P*= 0.029), (*P*=0.006)** and **(*P*=0.007)** respectively.

4.4.3. Percentage of the core filling material in SC technique

The percentage of core filling material for GP and ER groups at different levels (L1, L3, L6 and L9) is shown in [Table E (Appendix II) pg 134]. Initially, two-way repeated measure was conducted to explore the impact of the two materials (GP and ER), as measured by the percentage area of core filling material at L1, L3, L6 and L9. There was no statistically significant interaction between the materials and levels (P=0.51). However, the assumption was not normal [Fig. 6 (Appendix II) pg 142]. Therefore, univariate one-way repeated measure was conducted. The assumption for residuals was not normal [Fig. 7 and 8 (Appendix II), pg 143 and 144]. Thus, non-parametric Friedman test was used to identify the significant difference in the percentage area of core filling material (GP or ER) between different levels (Table 4.11).

Table 4.11: Mean percentage of core filling material at different levels in SC technique

Materials	Levels	N	Mean (SD)	*P* value[a]
GP	L1	15	58.39 (11.219)	**0.000**
	L3	15	67.18 (7.158)	
	L6	15	78.26 (7.714)	
	L9	15	82.75 (3.911)	
ER	L1	15	75.38 (10.683)	**0.000**
	L3	15	87.76 (6.242)	
	L6	15	89.60 (7.453)	
	L9	15	93.67 (4.624)	

a-Friedman test

Table 4.11 shows that there was a significant difference between the mean percentage of GP core filling material within different levels **(*P*=0.000)**. In addition, there was a significant difference between the mean percentage of ER core filling material within different levels **(*P*=0.000)**.

Therefore, Wilcoxon test was used to identify where the significant difference lies between different levels in GP group (Table 4.12) and ER group (Table 4.13).

Table 4.12: Multiple comparisons between percentages of core filling material at different levels in SC/GP

	Levels	Difference between levels	*P* value[a]
GP	L3-L1	8.79	0.156
	L6-L1	19.87	**0.006**
	L9-L1	24.36	**0.006**
	L6-L3	11.08	**0.006**
	L9-L3	15.57	**0.006**
	L9-L6	4.49	0.414

a-Wilcoxon test used with *P* value after Bonferroni correction was done
Level of significant set at *P*<0.05

Table 4.12 shows that the mean percentage of GP core filling material at L6 and L9 was significantly higher than L1 and L3 where ***P*=0.006** for all of them. However, the other comparisons were not significant.

Table 4.13: Multiple comparisons between percentages of core filling material at different levels in SC/ER

	Levels	Difference between levels	*P* value[a]
ER	L3-L1	12.38	**0.024**
	L6-L1	14.22	**0.024**
	L9-L1	18.29	**0.006**
	L6-L3	1.84	1.146
	L9-L3	5.91	**0.03**
	L9-L6	4.07	0.558

a-Wilcoxon test used with *P* value after Bonferroni correction was done
Level of significant set at *P*<0.05

Table 4.13 shows that the mean percentage of ER core filling material was significantly high between (L3-L1), (L6-L1), (L9-L1) and (L9-L3) where ***P*=0.024**, ***P*=0.024**, ***P*=0.006** and ***P*=0.03** respectively. However, the comparisons for other levels were not significant.

In order to identify any difference between the two materials, Mann-Whitney test was performed (Table 4.14).

Table 4.14: Multiple comparisons of percentage of core filling material between GP and ER groups using SC

Dependent variables	Gutta-percha median (IQR)	EndoREZ® median (IQR)	*P* value[a]
L1	57 (22)	74 (17)	**0.001**
L3	66.23 (14)	90 (6)	**0.000**
L6	78 (14)	91 (8)	**0.000**
L9	82 (6)	94 (3)	**0.000**

a-Mann Whitney test
Level of significant set at *P*<0.05
IQR= Interquartile range

Table 4.14 shows that the percentage of core filling material was higher for ER group at all levels than GP group with a significant difference present for all levels L1, L3, L6 and L9 where ***P*=0.001**, ***P*=0.000**, ***P*=0.000** and ***P*=0.000** respectively.

4.4.4. Percentage of sealer + voids in CLC, WLC and SC techniques

In this section, no further analysis was performed since by definition the percentage area of sealer + voids is equal to the whole area minus the percentage area occupied by the core filling material (i.e. they are inversely related). As for an example, the results of percentage of sealer + voids in each obturation technique (CLC, WLC and SC) for GP and ER groups were the same as those results mentioned in the previous sections 4.4.1, 4.4.2 and 4.4.3 respectively.

4.5. Quality of obturation

4.5.1. Cold lateral compaction of gutta-percha (CLC/GP)

The stereomicroscopic images [Fig. 9 (Appendix II) pg 145 and 146] show the cross-sections of CLC/GP. The best cross-section was from a sample 8-L9 (Fig. 4.1) which appeared to be smooth and homogenous. Whereas, the poorest one was from a sample no. 9-L9 (Fig. 4.2) which appeared with large amount of sealer and voids. The sealer was seen in the contact areas between the core filling materials in this subgroup. Voids and sealer were seen along the canal walls in many cross-sections.

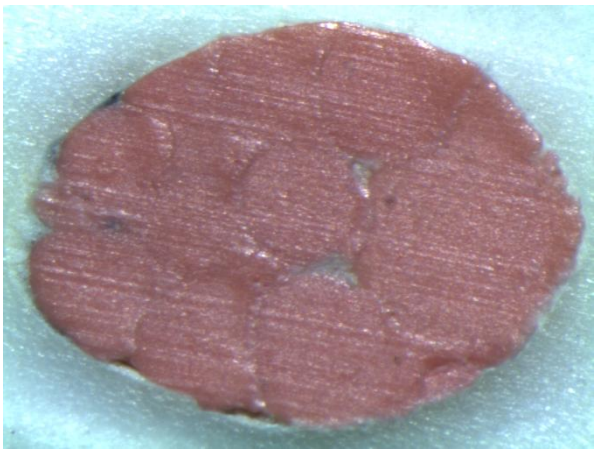


Figure 4.1: Sample no.8 section L9 CLC/GP

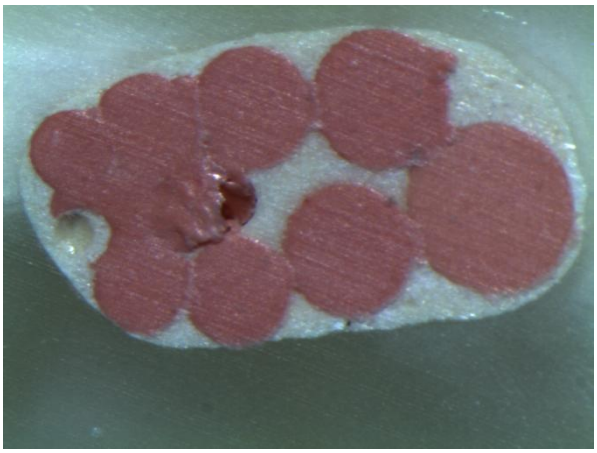


Figure 4.2: Sample no.9 section L9 CLC/GP

4.5.2. Cold lateral compaction of EndoREZ® (CLC/ER)

The stereomicroscopic images [Fig. 10 (Appendix II) pg 147 and 148] show the cross-sections of CLC/ER. The best cross-section was from a sample no. 5-L9 (Fig.4.3) which appeared to be homogenous. Whereas, the poorest one was from a sample no.1-L6 (Fig.4.4) which appeared irregular and the sealer + voids showed at one side of the canal wall. The sealer was not seen in between ER core filling materials in the majority of this subgroup.

Figure 4.3: Sample no.5 section L9 CLC/ER

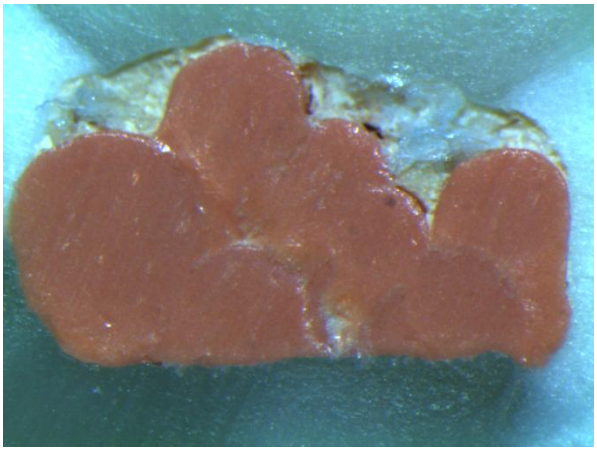


Figure 4.4: Sample no.1 section L6 CLC/ER

4.5.3. Warm lateral compaction of gutta-percha (WLC/GP)

The stereomicroscopic images [Fig. 11 (Appendix II) pg 149 and 150] show the cross-sections of WLC/GP. The best cross-section was from a sample no. 1-L9 (Fig.4.5) which presented with a good adaptation of the warm GP core filling material to the canal walls. The poorest one was from a sample no. 2-L1 (Fig.4.6) which appeared to be irregular with sealer and voids, and the warm GP failed to compact correctly to the canal walls. The sealer and voids were seen in the periphery of the canal walls in the majority of samples, especially at L1 and L9. At L6, the majority of warm GP was presented with smooth and homogenous mass that adapted to the canal walls.

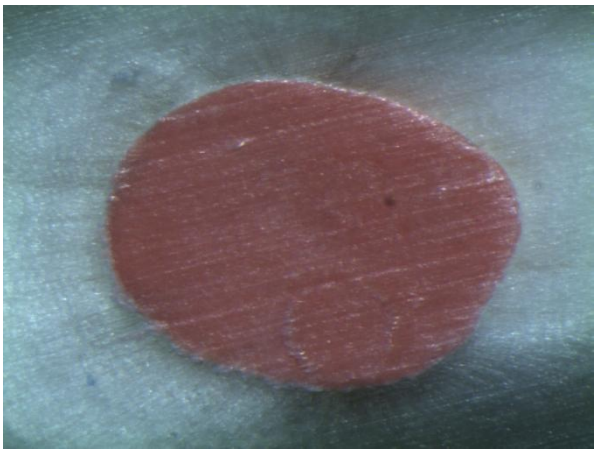


Figure 4.5: Sample no.1 section L9 WLC/GP

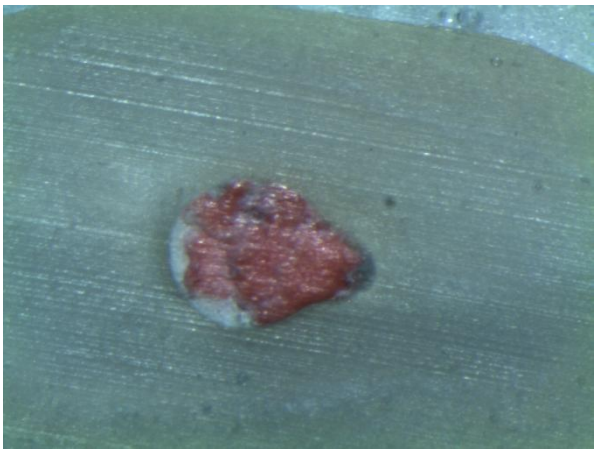


Figure 4.6: Sample no.2 section L1 WLC/GP

4.5.4. Warm lateral compaction of EndoREZ® (WLC/ER)

The stereomicroscopic images [Fig. 12 (Appendix II) pg 151 and 152] show the cross-sections of CLC/ER. The best cross-section was from a sample no. 3-L6 (Fig.4.7) which appeared to produce close adaptation of the warmed ER core filling material to the canal walls like a mono-block. The warmed ER core filling material was appeared homogenous at the majority of the cross-sections. The poorest cross-section was from a sample no. 11-L1 (Fig.4.8) which appeared irregular with sealer at one side of the canal walls.

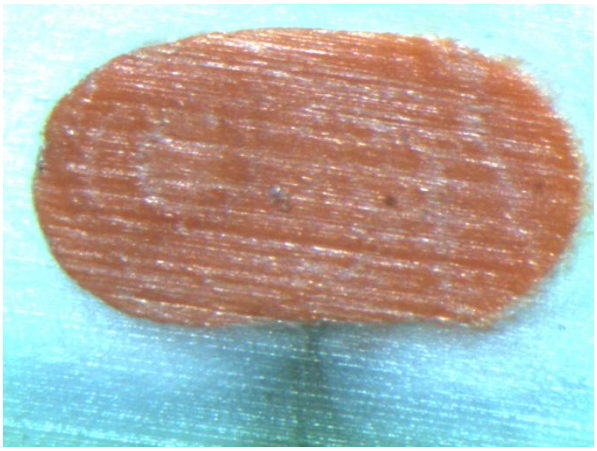


Figure 4.7: Sample no.3 section L6 WLC/ER

Figure 4.8: Sample no.11 section L1 WLC/ER

4.5.5. Single cone technique of gutta-percha (SC/GP)

The stereomicroscopic images [Fig. 13 (Appendix II) pg 153 and 154] show the cross-sections of SC/GP. The best cross-section was from a sample no. 10-L3 (Fig.4.9) which had a round shape and appeared to be filled with the GP core filling material and small amount of sealer surrounding the GP in the canal. The poorest one was from a sample no. 2-L6 (Fig.4.10) had an irregular canal shape and appeared to be filled with a large amount of sealer and voids. The sealer was seen around and in between the GP core filling materials in this subgroup.

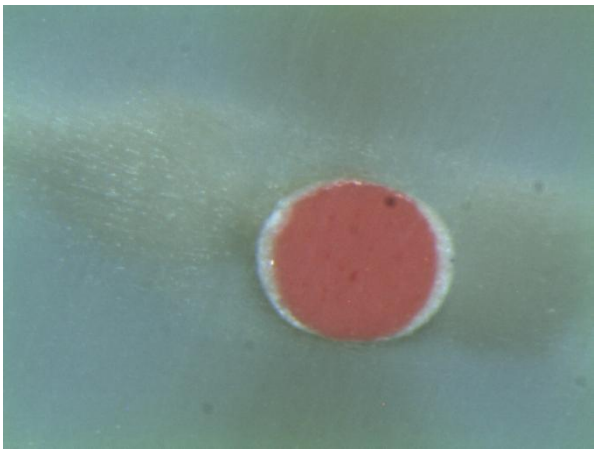


Figure 4.9: Sample no.10 section L3 SC/GP

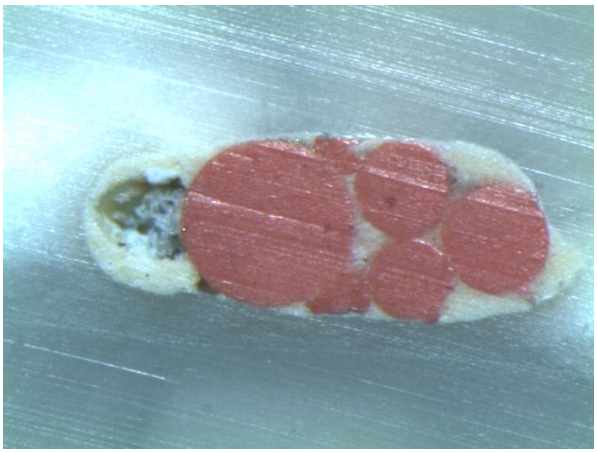


Figure 4.10: Sample no.2 section L6 SC/GP

4.5.6. Single cone technique of EndoREZ® (SC/ER)

The stereomicroscopic images [Fig. 14 (Appendix II) pg 155 and 156] show the cross-sections of SC/ER. The best cross-section was from a sample 7-L9 (Fig.4.11) which appeared to be filled very well with the core filling material and accessory points. The poorest cross-section was from a sample no.13-L9 (Fig.4.12) which had an irregular canal shape. It was appeared to be irregularly filled and with the presence of an amount of sealer and voids.

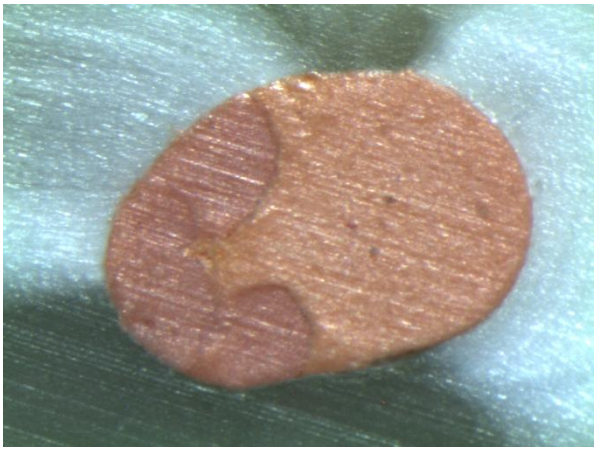


Figure 4.11: Sample no.7 section L9 SC/ER

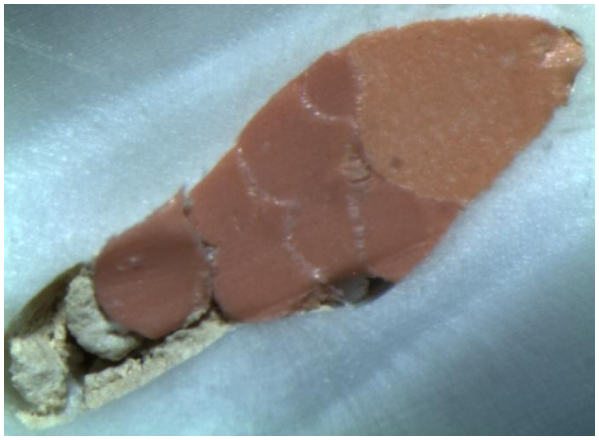


Figure 4.12: Sample no.13 section L9 SC/ER

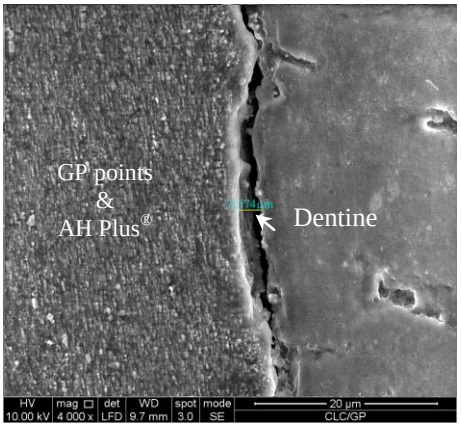
4.6. Scanning electron microscopy

One specimen from each group was selected randomly and examined under a field-emission gun scanning electron microscope (FESEM) at apical regions (1 - 5 mm from the apex) for the adapting of obturation material (core filling and sealer) to dentine.

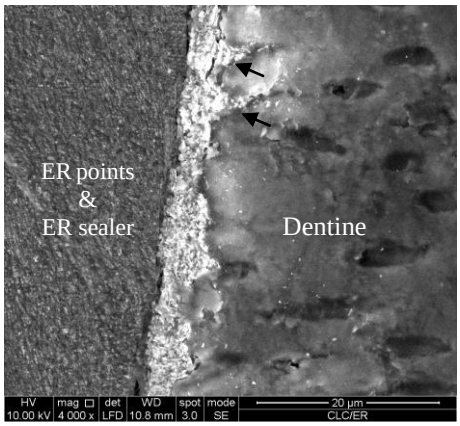
The SEM micrographs of GP and ER groups at various magnifications: 200× [Fig. 15 (Appendix II) pg 157], 600× [Fig. 16 (Appendix II) pg 158) and 1000× [Fig. 17 (Appendix II) page 159], provided an overview of apical region of the specimen.

For the GP group, at 4000× magnification, the gaps were identified at the obturation material-dentine interface for all techniques. The widest gap observed in the CLC technique was about 2.58 μm wide (Fig. 4.13 A), whereas in the WLC technique the widest gap was about 4.66 μm and the sealer was not clearly visible (Fig. 4.13 C). In the SC technique, the gap was found but to be smaller in comparison to the other techniques with a large amount of AH Plus® present (Fig. 4.13 E).

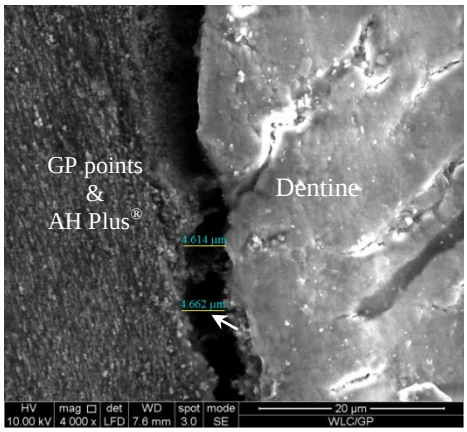
For the ER group, at 4000× magnification, there was hardly gap identified at the obturation material-dentine interface for both CLC and WLC techniques (Fig. 4.13 B and D). In the SC technique, no gap was identified, but a large amount of ER sealer was present (Fig. 4.13 F) than the other two techniques. Resin tags were clearly visible especially for WLC and SC techniques with ER material (Fig. 4.13 D and F).



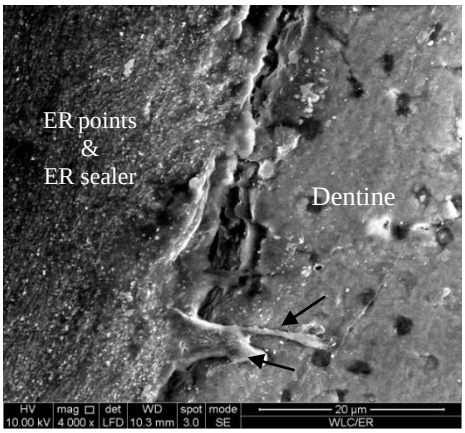
A. CLC/GP. Gap (white arrow) is evident between GP points/AH Plus® sealer and dentine.



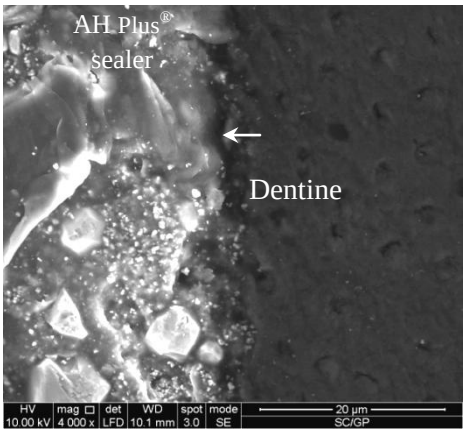
B. CLC/ER. Resin tags (black arrows) are evident in the dentine area.



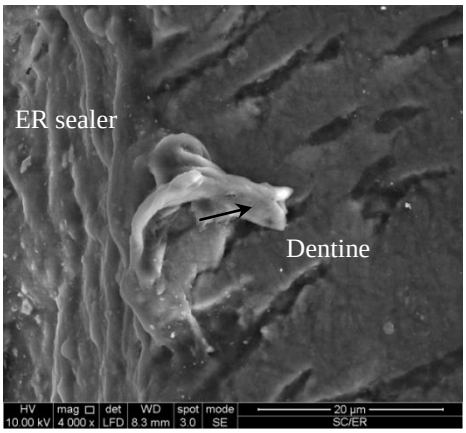
C. WLC/GP. Gap (white arrow) is evident between GP points/AH Plus® and dentine.



D. WLC/ER. Resin tags (black arrows) are evident in the dentine area.



E. SC/GP. Gap (white arrow) is evident at between AH Plus® sealer and dentine. No resin tags can be identified.



F. SC/ER. Resin tag (black arrow) is evident in the dentine area.

Figure 4.13 (A-F): High power SEM (4000×) micrographs of a longitudinal section of root filled with GP point/AH Plus® and ER point/ER sealers using CLC, WLC and SC techniques.

CHAPTER FIVE

DISCUSSION

5.1. Methodology

5.1.1. Collection and storage of teeth

Many chemical solutions have been used as a storage medium to keep extracted teeth. Examples are 10% formalin (Attam *et al.*, 2009), 2% thymol (Ezzie *et al.*, 2006), chloramines T- trihydrate (Kimyai *et al.*, 2010) and saline with 0.2% sodium azide (Zhang *et al.*, 2009). In this study, the teeth were stored in 0.5% Chloramine-T trihydrate solution for one week (ISO/TS 11405/2003). Throughout the experiment, the teeth were kept hydrated in distilled water as this does not seem to alter dentine permeability (Goodis *et al.*, 1993, Komabayashi *et al.*, 2009). The storage solution was changed regularly every week, and the teeth were stored at 4^{o}C (ISO/TS 11405/2003).

5.1.2. Selection of teeth

Mandibular premolars were used as the study sample. This tooth type is known to have variation in its root canal morphology with some possessing an oval canal shape (Paqué *et al.*, 2010). Oval or ribbon-shaped canals occur in approximately 25% of teeth (Ozawa *et al.*, 2009). Oval canal is defined as having a maximum diameter of up to 2 times greater than the minimum diameter in cross-section, and "long oval" or ribbon shaped as having a maximum diameter of 2 to 4 times greater than the minimum diameter (Paqué *et al.*, 2010). Preparation and obturation of these canals have been considered to pose a challenge (Ozawa *et al.*, 2009). Premolars with relatively straight roots were used in this study, as these teeth usually have only one canal (for mandibular premolars) and are easy to collect (from extraction for orthodontic purposes).

Human teeth were used in this study to simulate the clinical situation. Teeth with similar root morphology, size, and shape were selected in an attempt to standardize the specimens (Bachicha *et al.*, 1998). They were decoronated to a standard length of 16 mm. Those with buccolingual and mesiodestal dimensions (at the CEJ) of 7.5-8.0 mm

and 5.0-5.5 mm, respectively, were included for further standardization. Only teeth which allowed a size 15 K-file to fit in the apical 4-5 mm from the apex were selected. If the size 15 K-file was lose at the apex that meant the canal was wide, thus the specimen was discarded. These were done in order to reduce as much as possible the anatomical variations among specimens.

5.1.3. Root canal preparation

5.1.3.1. Root canal instrumentation

The Mtwo rotary instruments were used in this study to prepare the root canals. All canals were prepared according to the manufacturer's instructions; the instruments were used in a single-length technique. Mtwo rotary instrument was found to maintain the original canal curvature significantly better and faster than K3, RaCe (Schäfer *et al.*, 2006b) or standard hand K-file instruments (Safi *et al.*, 2008). Furthermore, in a study that assessed the efficacy of rotary instrument in preparing an oval root canal, Mtwo and ProTaper (with greater taper) were found to be more efficient than NiTi hand files (Elayouti *et al.*, 2008). It is noted that, after preparation, uninstrumented recesses may be left in oval canals, regardless of the instrumentation technique, thus leaving a debris and unprepared root canal walls behind (Grande *et al.*, 2008).

Determination of the working length is one of the important procedural steps. Instrumentation short of the canal risks leaving inflamed tissue and infectious elements in the root canal space. On the other hand, instrumentation beyond the apical foramen may force infected debris into the periapical tissue resulting in periapical inflammation, foreign body reaction or incomplete regeneration of the supporting tissues. Thus, it is believed that the apical termination of the root canal instrumentation should be at the apical constriction (Bergenholtz *et al.*, 2009a). The apical constriction is the apical

portion of the root canal having the narrowest diameter. Its position may vary but is usually between 0.5-1.0 mm short of the centre of the apical foramen (Ingle *et al.*, 2008a). Therefore, the working length was adjusted to 1.0 mm short of the apical foremen in this study.

Size 35, 0.04 taper, was selected as the master apical size. A minimal apical enlargement has been suggested to preserve tooth structure and limit extrusion of filling material (Fornari *et al.*, 2010). Conversely, strong emphasis on reducing the number of instruments and limiting apical preparations to small sizes does not produce clean apical preparations in infected teeth. Thus, larger instrumentation was beneficial in reducing the debris in the apical third of the canal (Baugh and Wallace, 2005). In addition, it promote the removal of infected dentine and allow the irrigating solution to reach the apex (Siqueira Jr *et al.*, 1999, Hecker *et al.*, 2010). On the other hand, over preparation can result in more procedural errors or weaken the root structure (Hecker *et al.*, 2010). Preparation of the root canal to three sizes larger than the first file that binds at the working length was and still is used as a rule (Mickel *et al.*, 2007). However, there is no evidence that the instrument that binds first in the working length does actually reflect the diameter of the canal in the apical region. Furthermore, since many canals are oval in shape it is doubtful whether removing dentine from the wall of the recesses is always possible. In fact, cleaning in these recessed areas of the canal may have to rely on irrigation (Wu *et al.*, 2002).

Creation and subsequent maintenance of a smooth glide-path from the canal orifice to the apical foramen using 2% taper stainless steel hand files is an essential preparatory step before commencing NiTi instrumentation in order to reduce the risk of iatrogenic

errors such as ledge and fracture of instrument (Young *et al.*, 2007). This was practiced in this study, by the use of a size 15 K-file.

Plotino *et al.* (2006), who studied the fatigue life of NiTi rotary instruments, demonstrated that each Mtwo rotary instrument can be used safely in a simulated clinical situation up to 10 times in oval canals. To eliminate the chance of errors during root canal preparation, each Mtwo rotary instrument and K-file in this experiment were used up to 5 times only.

Apical patency is a technique in which the apical portion of the canal is maintained free of debris by recapitulation with a small file through the apical foramen (Tsesis *et al.*, 2008, Arias *et al.*, 2009). According to the technique described by Buchanan in 1989, apical patency can be achieved by moving a small file (size 08 or 10) passively 0.5-1.0 mm through the apical constriction (Goldberg and Massone, 2002, Bellido *et al.*, 2010). Thus, apical blockage and interference with the working length could be eliminated (Souza, 2006). Again, this was practiced during root canal preparation with a size 10 K-file in this study.

5.1.3.2. Root canal irrigation

During root canal preparation, a combination of organic and inorganic debris (smear layer) is formed on the root canal wall that may also contain bacteria and their by-products (Violich and Chandler, 2009). It has been demonstrated that approximately 70% more debris appeared to stay in the root canals when no irrigant is used during instrumentation compared with irrigated canals (Siqueira Jr *et al.*, 2002). NaOCl has been the most widely used root canal irrigant and is currently available in different concentrations (Mônika and Fröner, 2006). Full strength of NaOCl is recommended

over lower concentrations because of its superior antimicrobial and tissue dissolution properties (Carson *et al.*, 2005). Ayhan *et al.* (1999) studied the antimicrobial effects of various root canal irrigants on selected microorganisms and observed that 5.25% NaOCl was superior to 0.5% in its antimicrobial abilities. Therefore, in this study, 5.25% NaOCl was used to disinfect the root canal system and to simulate the clinical scenario.

NaOCl, however, cannot dissolve the smear layer (Zehnder, 2006), which layer can act as a reservoir or substrate for microorganisms (Onay *et al.*, 2010). In addition, the smear layer has been shown to hinder diffusion of intracanal medication or the penetration of sealer into lateral canals and dentinal tubules, which my compromise the seal of root canal filling material (Mancini *et al.*, 2009, Onay *et al.*, 2010). Thus, demineralizing agents such as EDTA and citric acid have been recommended as an adjunct in RCT (Zehnder, 2006). The alternate use of EDTA and NaOCl has been recommended for the removal of the smear layer (Hashem *et al.*, 2009). In previous research, no significant difference on the smear layer removal was seen with different concentrations of EDTA solution (1%, 5%, 10% and 15%) (Sen *et al.*, 2009), whereas, another in vitro study showed that EDTA 18.6% and 24% groups were superior compared with 15% and the control groups (without demineralizing agent) (Putzer *et al.*, 2008). In a study of microbial leakage evaluation for teeth obturated with GP/AH Plus and ER points/ ER sealer, EDTA 18% was used as a canal irrigation (Drukteinis *et al.*, 2009). Therefore, in the current study, EDTA 18% was used to remove the smear layer from the root canal system.

All canals were irrigated with NaOCl which was followed by EDTA due to its efficiency to minimize the residual effect of NaOCl on free radical polymerization of resin (Jainaen *et al.*, 2007). Some recent studies have related this interference mainly to the oxidizing effect of NaOCl (Nagpal *et al.*, 2007), as resin does not polymerize when

in contact with air (Pameijer and Zmener, 2010). Finally, canals were rinsed with distilled water to remove any remnant of the irrigating solutions (Jainaen *et al.*, 2007).

5.1.4. Pilot study

A pilot study was carried out to determine the possibility and difficulty of various obturation techniques to fill oval-shaped canals. Root canal preparation and obturation techniques that have been used in the current study were practiced comprehensively earlier. In addition, all specimens were prepared and obturated by one operator to reduce interoperator variability.

In lateral compaction technique, a closely matching finger spreader with accessory GP points would be an ideal and should allow obturation with fewer voids (Hartwell *et al.*, 1991). Some disagreement exists regarding whether spreader size should be equal or larger than the accessory point size (Gound *et al.*, 2001). However, lack of uniformity between the accessory points and finger spreaders from different manufacturers could cause an unfilled space (Marroquín *et al.*, 2001). Incompatibility will result in the accessory point hang up before full insertion, resulting in either a cement pool at best or a void at worst (Whitworth, 2005). Thus, ISO standardized size 25 finger spreaders (VDW, München, Germany), size 25/0.02 GP accessory point (VDW, München, Germany) and size 25/0.02 ER accessory point (Ultradent Products Inc., South Jordan, UT, USA) were measured prior to use in this study. The result showed that the diameters of both accessory (GP and ER) point size 25/0.02 at 1, 3 and 6 mm short of the tip were variable; some showed a same size, some slightly smaller and some larger than the diameter of a size 25 finger spreader at the same points. Therefore, a size 30 finger spreader (VDW, München, Germany) was selected, which has a same size or slightly larger than the diameter of both accessory point (GP and ER) size 25/0.02.

Therefore, in the current study, two finger spreaders size 25 and 30 were used due to the variations in diameter of accessory points found in the same box of both GP and ER.

In the pilot study, with WLC technique, the heat plugger of BeeFill® 2-in-1 device at 100°C was inserted to 1.0 versus 2.0 mm short of the working length. Extrusion of obturation materials was obviously noticeable in the 1.0 mm group. Therefore, in the current study, and according to Collins *et al.* (2006), the heat plugger was inserted 2.0 mm short of the working length. High temperatures on the root surface are believed to cause damage to the periodontal attachment apparatus of the tooth (Lee *et al.*, 1998). Thus, lowering the temperature of the plugger tip would appear to be appropriate (Jung *et al.*, 2003). Accordingly, in the current study, the temperature of the heat plugger of BeeFill® 2in1 device was set to 100°C as that was the lowest temperature that could be set with this device.

SC is considered as a faster and easier technique for obturation root canal system (Gordon *et al.*, 2005, Hörsted-Bindslev *et al.*, 2007). Such a "harpooning" technique is recommended for reducing sealer volume in larger oval-shaped canals (Tay *et al.*, 2005a, Hiraishi *et al.*, 2006). Therefore, in this study, SC technique was performed by applying the sealer (AH Plus® or ER) to the canal walls, then a matching master cone size 35/.04 (GP or ER) to the prepared canal was selected and inserted into the canal, followed by passive placement of multiple accessory points (GP or ER) size 25/0.02. Insertion of accessory points size 25/0.02 beside the master cone was performed passively without using a finger spreader, because the lateral compaction techniques (cold and warm) were already included in this study. In fact, the passive insertion of accessory points is recommended by the manufacturer of ER.

5.1.5. Evaluation method

In general, one goal of various filling techniques is to maximize the amount of GP and minimize the amount of sealer. Shrinkage, solubility and breakdown of sealer can potentially affect the success of RCT (Tasdemir *et al.*, 2009, Abedi *et al.*, 2010). Quantifying the content of the obturated canal can be assessed by sectioning the obturated canals and microscopic evaluation (James *et al.*, 2007). A number of authors have been advocating the assessment of the quality of a root canal obturation by cross-sectional analysis of the space occupied by GP, sealer and voids (Lyroudia *et al.*, 2000, Lucena-Martin *et al.*, 2002, Gordon *et al.*, 2005, Gulsahi *et al.*, 2007, Ozawa *et al.*, 2009). The percentage of GP filled canal area in various cross-sections has been used as a measure of the quality of the root filling (Souza *et al.*, 2009b). This method was employed in this study to assess the quality of the root canal obturation. A high percentage of core filling material with a low percentage of sealer + voids would indicate a good obturation quality (Wu *et al.*, 2001).

It is widely known that the anatomy of the apical region of the root canal is extremely complex and remaining pulp tissue and debris are usually confined in areas that remain untouched by root canal instruments (De Instrumentos, 2002). Baumgartner and Mader (1987) also reported that the apical and middle third of the root canal are considered the most difficult areas to be cleaned. It follows that, if the canal is not clean, it cannot be filled properly (Lendini *et al.*, 2005). In this study, the canal length was 16 mm, thus sections at 1 and 3 mm represented the apical third and sections at 6 and 9 mm represented the middle third of the root canal. The cross-sections were obtained by using a low-speed diamond disc under copious water irrigation (coolant) so that the heat generated during sectioning would not cause smearing and distortion of the filling material (Wu *et al.*, 2001).

In general, good obturation should be consisted of the maximum amount of core material and minimal amount of sealer. This is because sealer will be dissolved with time, thus creating spaces which can act as venue for potential leakage. The amount of voids should be minimized as large amount of voids can contribute to a poor RCT outcome. Initially, in this study, an attempt was made to quantify the root canal contents i.e. core filling material, sealer and voids as separate entities. However, during the measurement using the software (Cell^ D), it was found that the amount of voids presence was very small, compared with the area of the core material (GP or ER). Thus, in this study, sealer and voids were pooled for the calculation.

Evaluation of the penetration of the root canal sealers into dentinal tubules by SEM was done by many researchers before (Sevimay and Dalat, 2003, Stevens *et al.*, 2006, Yang *et al.*, 2007). The image created by the electron array from SEM is able to show us a three-dimensional view, exhibiting the outline of the dentinal walls to be observed and the root canal sealer. Therefore, all surfaces of the dentinal wall can be scanned and examined in details, and the results can be characterized, measured and interpreted (De-Deus *et al.*, 2004). Examination of the marginal adaptation under SEM could provide information that could be used as an indicator in the sealability of obturation materials (Torabinejad *et al.*, 1995).

The use of SEM in low-vacuum mode permits observation of specimens, which have not been coated with a conductive material such as gold or carbon, therefore, saving of preparation time and effort. Low-vacuum can be used to view specimens, which are difficult to image with a conventional SEM and this method eliminates surface charging problems experienced in a high vacuum SEM (Sammons and Marquis, 1997). Low vacuum SEM is a relatively inexpensive alternative, which has a number of potentially useful applications in the biomaterials research field. Back-scattered imaging was used

in a low-vacuum SEM to improve contrast, thereby facilitating measurement (Sammons and Marquis, 1997, Shipper *et al.*, 2004a). This method was used to evaluate the adaptation of the obturation materials to the canal walls.

5.2. Results

5.2.1. Evaluation of obturation quality of GP and ER

5.2.1.1. Extrusion of root filling material

The ideal length for preparation and filling of a root canal is between 1and 2 mm short of the root apex (Scarano *et al.*, 2007). Any extrusion of filling material beyond the root apex might cause a mechanical or chemical irritation to the periradicular tissues, which can then result in an inflammatory response. This response may, in turn, lead to a RCT failure (Brki *et al.*, 2009).

In this study, extrusion of filling material was noted slightly higher in the WLC technique as compare to the CLC technique, both in GP and ER groups. In addition, GP group showed slightly higher occurrences of extrusion than the ER group, which findings, however, were not significantly different. The finding of this study is similar to the studies performed by Da Silva *et al.,* (2002) and Gilhooly *et al.,* (2001) who found that the extrusion of obturation material in CLC was less than the thermoplasticized GP techniques. It is said that CLC technique is capable of ensuring a good length-control of the root filling (Lea *et al.*, 2005, Bergenholtz *et al.*, 2009b, Sadeghi and Sadeghi, 2009). As for the SC group, no extrusion was found in both materials (GP and ER), which finding may be attributed to the passive insertion of accessory points during the obturation procedure.

5.2.1.2. Percentage of core filling material and sealer + voids

The ideal root canal filling material should be adapted well to the canal walls and its irregularities. The entire root canal space should be densely compacted with a homogenous mass of the filling material. GP has good physical and biological properties. However, it fails to adhere to the canal walls (Shipper *et al.*, 2004b). It has been shown that GP frequently separates from the root canal sealer, resulting in gap formation (Tay *et al.*, 2005b). To address this problem, resin-coated GP points (namely, ER points) and sealer (namely, ER sealer) were introduced. In this study, using different obturation techniques i.e. CLC, WLC and SC, the percentage of core filling material and sealer + voids were quantified and compared between GP and ER groups.

1. CLC technique

In the current study, using CLC technique, GP group produced different percentage of core filling material (mean=79.45-91.80%; Table 4.4 pg 62) occupying the canal space at different canal levels namely; L1, L3, L6 and L9. In particular, the percentage of GP was significantly lower at L1 (i.e. the most apical canal area in this study) compared to other levels. In the ER group, however, the percentage of core filling material (mean=91.32-95.90%; Table 4.4 pg 62) was not significantly different between different levels. This finding shows that it was rather much more difficult to place accessory points at the most apical level in the GP group (Wu *et al.*, 2003a). Thus, using CLC technique, resin coated GP (ER group) is capable of producing more densely, compacted and homogeneous mass of root filling for the entire root canal space than GP group in this study.

When the GP and ER groups were compared, it was found that using CLC technique, the percentage of ER core filling materials was higher than the percentage of GP core

filling material. The different were significant especially at L1 (*P*=0.005; Table 4.6, pg 63) and L3 (*P*=0.023; Table 4.6, pg 63). Inversely, the percentage of sealer + voids was significantly lower in the ER group than the GP group at L1 and L3.

In this study, ER group was capable of producing better obturation quality, compared to the GP group, especially at the apical third of the canal area. This might be due to the fact that during obturation procedure the penetration depth of the finger spreader in the ER group was greater than the GP group. This was in agreement with the study by Nielsen and Baumgartner (2006) who found that spreader penetration was better in Resilon than GP. Improved penetration of spreader would allow better placement of accessory points. A microleakage study of GP/AH Plus and ER points/ER sealer group, performed by Drukteinis *et al.,* (2009), found that leakage was lower in ER points/ER sealer than GP/AH Plus group, indicating that using CLC technique, the ER point/ER sealer can produce better obturation quality than GP/AH Plus®.

One of the important factors that influenced the results of this study is the original canal morphology, which varies among different tooth groups and, even, for the same tooth group. It is recognized that root canals are not always round in shape but may exhibit broader bucco-lingual dimension appearing as oval to a ribbon-shaped configuration. A good cleaning and subsequently better apical seal is believed to be readily achieved in round canal (Iqbal *et al.*, 2007). In this study, with CLC technique, for both GP and ER groups, the mean percentage of core filling material at L3 was higher than the other levels (L6, L9 and L1). This might be due to a round shape of the prepared canals at the 3 mm level as compared to other levels. Thus, a dense core filling material was obtained at L3. This was in agreement with a study performed by Iqbal *et al.* (2007), who evaluated the apical obturation quality after root canal preparation with GT, Lightspeed,

and Profile instruments, and they found that at 3 mm the round shape canals were observed and these resulted in better obturation quality.

Although the canals appeared to be obturated by a large number of accessory points, they were not obturated sufficiently. Wu *et al.* (2001) found that the quality of obturation with CLC in oval canals was less reliable and the percentage of core filling material (GP) ranged from 70% to 100%. The discrepancy may be attributed to several factors, including the irregular shape of the canal and the operator's ability in performing the technique (Wu *et al.*, 2001).

2. WLC technique

In this study, using WLC technique, both GP and ER group resulted in difference percentage of core filling material. In GP group, the mean percentage of GP core filling material (mean=88.46-98.50%; Table 4.7, pg 64) was higher at L3, L6 and L9 levels than L1 with a significant difference lies between L6-L1 (P=0.018; Table 4.8, pg 65). In ER group, the percentage of ER core filling material (mean=95.56-99.60%; Table 4.7, pg 64) was not significantly different after Bonferroni correction. Thus, with the WLC technique, ER point/ER sealer resulted in a more mass of material in the root canal space than the GP group.

When the GP and ER groups were compared, it was found that using WLC technique, the percentage of ER core filling material was higher than the percentage of GP core filling material. The significant difference lies at L1 (P=0.029; Table 4.10, pg 66), L3 (P=0.006; Table 4.10, pg 66) and L9 (P=0.007; Table 4.10, pg 66). Inversely, the percentage of sealer + voids was significantly lower in ER than GP group at the same levels (L1, L3 and L9).

In this study, the ER group yielded a better obturation quality than the GP group. This might be associated to better plasticity and flow of the softened core filling material of ER group as compared to that of GP group. This was to some extent in agreement with a study performed by Karabucak *et al.* (2008), who evaluated the filling of lateral canals, and found that the penetration of filling material is a function of viscoelastic properties of the filling material. Gurgel-Filho *et al.* (2006) assessed ability of different brands of GP to fill simulated lateral canals, and found that the brand of GP points influenced the length of filling within lateral canals. This may be a reflection of the formulation of dental GP points that affect the thermoplasticity behaviour of the material. Brands with higher amounts of GP in their composition showed better results while filling simulated lateral canals (Gurgel-Filho *et al.*, 2006). A study performed by Tanomaru-Filho *et al.* (2007b), who evaluated the thermoplasticity of different GP brands and Resilon, found that the Resilon had a higher thermoplasticity than various GP brands. However, there is no report that evaluated the thermoplastic property of ER point.

3. SC technique

In this study, using SC technique, the percentage of core filling material occupying the root canal space was significantly different between levels in both the GP and ER group. In GP group, the mean percentage of core filling material (mean=58.39-82.75%; Table 4. 11, pg 67) was significantly higher at L6 and L9 than L1 (P=0.006; Table 4.12, pg 67) and also higher at L6 and L9 than L3 with same P value. In ER group, the mean percentage of core filling material (mean=75.38-93.67%; Table 4.11, pg 67) was significantly higher at L3, L6 and L9 than L1 (P=0.024, P=0.024 and P=0.006) respectively (Table 4.13, pg 68) and between L9-L3 (P=0.03; Table 4.13, pg 68). This difference between the levels in both materials (GP and ER) might be attributed to that the SC technique relies on the configuration of the root canal (Gordon *et al.*, 2005).

However, in this study, using SC technique, the mean percentage of core filling material was the highest at L9, followed by L6, L3 and L1 levels, regardless of the material (GP or ER) used. In other words, SC technique was capable of producing good coronal obturation only.

When the GP and ER groups were compared, it was found that using SC technique, the percentage of core filling material was higher in ER group than in GP group with a significant differences lies at all levels L1 (*P*=0.001; Table 4.14, pg 69), L3, L6 and L9 (*P*=0.000; Table 4.14, pg 69). Inversely, the percentage of sealer + voids was lower in ER group than in GP group at all levels. This is possibly due to the "mono-block" which is created by bonding property between ER points and ER sealer (Pameijer and Zmener, 2010).

In this study, the mean percentage of core filling material in SC was lower and, inversely, the mean percentage of sealer + voids was higher than the other techniques for both materials (GP and ER) at different levels. This result is in agreement with a study performed by Wu *et al.* (2000), who evaluated the percentage of sealer in the apical and middle regions of canals filled by SC, CLC and vertical compaction techniques. They found that SC resulted in a significantly higher percentage of sealer than the other groups. Conversely, this finding is in disagreement with a study performed by Uppal and Kaur (2011) of the apical sealability of CLC and SC techniques, and they found that SC technique with tapered GP point resulted in a significantly higher percentage of GP filled area than CLC technique, however, this evaluation was done using simulated root canal in a resin blocks with 30^{o} curvature.

Mandibular premolars exhibit a high degree of complex anatomy with fine ribbon shaped canal system; which are difficult to access, clean and obturate. These irregularities create a challenge for the clinician and influence in the outcome of the RCT (Iyer *et al.*, 2006). These teeth are difficult to treat endodontically and show a high failure rate, possibly due to the extreme variations in the root canal morphology (Sandhya *et al.*, 2010). Moreover, Ingle stated that the canal anatomy of this tooth might account for a greater increase in RCT failure (Moayedi and Lata, 2004).

Oval shaped and large-diameter root canals would require excessive preparation for cold obturation technique to be effective. However, the use of warm compaction technique with matched-taper core material would ensure adequate adaptation to canal irregularities (Gordon *et al.*, 2005). In this study, the use of WLC with either material produced a higher percentage of filling core materials than the CLC and SC at all levels where sealer or voids were rarely seen, especially at L3 and L6. Sealer had probably been pushed away during compaction of the thermoplasticized core filling material. This result is in agreement with Wu *et al.* (2000), who reported that sealer may be removed from the canal wall by compaction procedure, regardless of the sealer placement method. In addition, this is may be due to the fact that the WLC technique allows a better flow of core filling material in the root canal space. This result was in agreement with a study performed by Nelson *et al.* (2000) to increase the density of GP, and found that the WLC technique, system B in low-heat, resulted in denser GP than CLC technique. Furthermore, Zhu *et al.* (1994) compared the apical sealability of WLC and CLC techniques and found that the WLC with sealer resulted in the best sealability.

5.2.1.3. Scanning electron microscopy

Owing to a lack of chemical union between GP (a polyisoprene) and various sealers, including ZnOE, epoxy resin, or glass ionomer-based sealers, a "mono-block" formation in the root canal has not been possible (Ko *et al.*, 2008). In contrast, a resin-coated GP enables the polyisoprene to be chemically coupled to methacrylate based resin sealers (Tay *et al.*, 2005a). The combination of ER sealer and ER points establish the so-called "mono-block" and is the reason for the superior sealing properties of the system (Pameijer and Zmener, 2010). In this study, SEM revealed a good union of ER points to ER sealer, despite the presence of some gap-containing regions between the obturation material and dentine. It is possible that the gaps may be a result of polymerization shrinkage of the methacrylate-based sealer with a high C-factor that is present in long, narrow root canals (Tay *et al.*, 2005c) and, possibly, an incomplete removal of the smear layer in some areas of the root canal (Tay *et al.*, 2005a). Resin tag formation was evident in some gap-free regions in ER group. This result is in agreement with a study performed by Tay *et al.* (2005a), who evaluated the effectiveness of resin-coated GP point materials and a dual-curing ER in obturated root canals, and found that resin tags were impregnating the canal walls, but gaps and silver leakage were also present. Conversely, fissures and gaps were observed between the GP/AH Plus® and dentine, which may attributed to the GP being pulled away from AH Plus® and also AH Plus® pulled away from the dentinal tubules. In the current study, however, no resin tags were seen in the GP group. This result is in agreement with a study performed by Steier *et al.* (2010), who studied the interface between sealer-dentine using SEM. They found that the AH Plus demonstrated significantly more gap than RealSeal.

In general, the gaps that created between the core filling material and sealer, and/or between sealer and dentine, would serve as pathways that allow leakage of bacteria or

their by-products into the dentinal tubules and periapical tissues (Kubo *et al.*, 2005, Paque and Sirtes, 2007) which, in turn, may adversely affect the RCT outcome (Siqueira and Rôças, 2008). A gap-free interface would be preferable.

Within the limitation of SEM observations, the gap was present in all GP techniques, with the gap being larger in WLC than CLC or SC technique. However, SC presented with a large amount of sealer, compared to others, filling (some of) the gap and making it small. It is against general belief to find the gap in WLC being larger than the gap in CLC, this might be due to that only 1 specimen was used in SEM observation.

In ER group, resin tags are identified in all techniques regardless to the gap free and gap containing areas (Tay *et al.*, 2005a). The finding of this study was disagree with the manufacturer of the ER materials in that the SC is the recommended technique for use with this material, since CLC and WLC yield better in maximizing the amount of core filling and minimizing the amount of sealer inside the root canal space.

5.3. Limitation of this study

1. The variation of human root canal anatomy was one of the difficult parameters to control in this study. (More than 200 extracted single-rooted mandibular premolars were examined to obtain the study sample).
2. This *in-vitro* study attempted to represent root canal obturation quality that can be applied to clinical situations; however, results may vary for *in-vivo* conditions.
3. Two different sealers (AH Plus® and ER) were used. It was rather difficult to standardize the amount of sealer applied in the canal even through a standardized method of application that was employed.
4. Only one specimen was used for SEM observation due to the financial restriction, and thus measurements made could not be analyzed statistically.

5.4. The significance of the study

In spite of the high cost of ER sealer and ER point, they are able to produce a good obturation quality in the root canal. In fact, this conclusion was obviously noticed in SC technique where the amount of core filling material was higher and inversely the amount of sealer + voids was lower in ER group than GP group. Therefore, if the dental practitioners want to use the simple and fastest obturation technique (SC), they can select ER point/ER sealer to obturate root canal space.

CHAPTER SIX

CONCLUSIONS AND SUGGESTIONS

6.1. Conclusions

ER point/ER sealer was compared with GP/AH Plus® using three different obturation techniques (CLC, WLC and SC). Within the limitation of this study, it may be concluded that:

1. Root canal obturation with ER and GP using CLC, WLC and SC techniques showed no significant differences in the occurrence of apical extrusion of material.
2. ER point/ER sealer is superior to GP/AH Plus® in the percentage of core filling material when different obturation techniques are used:
    i. CLC technique: ER group was better than GP group for maximizing the amount of filling core material at all levels with statistically significant difference at 1 and 3 mm levels.
    ii. WLC technique: ER group was better than GP group for maximizing the amount of core filling material at all levels with statistically significant difference at 1, 3 and 9 mm levels.
    iii. SC technique: ER group was better than GP group for maximizing the amount of core filling material at all levels with statistically significant difference at all levels.
3. SEM observation (based on 1 specimen only) at a different magnification showed that ER points/ER sealer seemed to suggest a better adaptation to dentine as compared to GP/AH Plus®.

6.2. Suggestions

Further studies are recommended for evaluating the following:

- The response of periradicular tissue to extruded EndoREZ® obturation materials.
- The mechanical and thermal properties of EndoREZ® points and EndoREZ® sealer.
- The impact of different temperature setting on EndoREZ® points and EndoREZ® sealer relative to their constituents and performance.
- The measurement of the depth of spreader penetration in EndoREZ® points and its effect on the apical seal by using different taper EndoREZ® points.
- Shear bond strength between EndoREZ® points and other resin-based sealer.
- The measurement of the resin tags penetration into dentinal tubules by using confocal laser scanning microscope and its influence on sealing ability.