CHAPTER 5: DISCUSSION
5.1. Materials and Methods:

An *in-vitro* evaluation is usually the first step for testing any technique or material. Microleakage test is the most common test used to evaluate the efficacy of bonding between two restorative materials or between a restorative material and hard dental tissues. The wide variety of dye materials and different techniques used in *in-vitro* tests make comparing the results of different studies difficult since there are no generally accepted standards for experimental parameters, such as selection of teeth, the way of bringing the materials into operation, the type of dye, type and concentration of the storage solution, time of storage, temperature during storage, type and duration of thermal cycling and/or mechanical cycling and the scoring criteria to examine the microleakage. Although a wide variety of methods had been described with some variations in results, the standardisation of such methods is therefore necessary in order to obtain comparable results from different studies.

In this present study, in order to obtain reliable results, an in vitro standard model was chosen to standardise the quality of the ready made access cavities through PFM crowns and the quantification of leakage to enhance reliability of results. The materials were used according to the various manufacturers’ recommendations and ISO guidelines for testing were followed precisely. All these procedures were done by one evaluator to decrease the number of variables involved in obtaining the results. Also following ISO/TS (6872/1999) and ISO/TS (11405/2003), all procedures were performed at 23 ± 2 ºC in order to limit influences from thermal changes.

A total number of 80 access openings of the PFMs which received 80 restorations were considered sufficient to cover most variables. Due to the fact that the combination of the two types of composites (packable and flowable) was the main focus of this
investigation, all other variables such as the thickness of the PFM samples, the depth of access openings, the roughness of the inner access openings, RMGIC, type of adhesive and all laboratory procedures were standardised to overcome experimental variation as much as possible. All procedural details of this in vitro study were carefully considered and conducted because any minor change may significantly influence the results.

5.1.1. Specimens:
To obtain reliable and reproducible results, a total number of eighty specimens of standard simulated PFMs with their access cavities were prepared for restoration with composite to allow investigation of interfacial coronal leakage along the PFM-restoration interface.

5.1.1.1. The thickness of the specimens of this in-vitro study:
The total thickness of the PFM crowns was 2 ± 0.5 mm (Metal + 2 layers of opaque + 2 layers of body and enamel porcelain) in order to get a standard depth of access cavity through the PFM crowns simulating clinical situations. The total depth of the access cavity was therefore also standardised at 2 mm ± 0.5 thereby obtaining composite resin restorations of similar thickness during the restorative procedures. To simulate clinical access openings, a standard central hole of 5 mm in diameter (Marroquin et al., 1995) and 15.7 mm in circumference was established in each PFM specimen.

The thickness of the metal was 0.4 mm as metal thickness was generally less than 0.5 mm to maximize space for the ceramic (McMullen et al., 1989; Craig et al., 2004). The opaque layer around the access openings was approximately 0.3 ± 0.1 mm in thickness. Barghi and Lorenzana (1982) stated that the recommended thickness of opaque for different brands of porcelain varied from 0.2 to 0.4 mm. Other studies had suggested a minimum dimension of 0.2 mm (Barghi and Lorenzana, 1982) and a maximum
thickness of up to 0.5 mm (O’Brien, 1989) due to the wide variability among opaques and requirements of different shades.

Two body and enamel layers around the access openings added up to approximately 1.5 ± 0.2 mm in thickness in this study. For more accurate shade duplication, estimates of the combined thickness of fired dentine and enamel porcelains had ranged from a minimum of 0.5 to 1.0 mm (Barghi and Lorezana, 1982) to a maximum thickness of 1.5 to 2.0 mm (Naylor, 1992). Porcelain that was built and fired above this 2-mm maximum height was considered unsupported by metal and more prone to fracture as harmful stresses can form in thick, unsupported porcelain sections. For uniformity of shade and maximum strength, it is highly desirable to have an even thickness of porcelain covering the metal substructure (Naylor, 1992).

5.1.1.2. Specimen Fabrications:

The disc shaped specimen was used with modifications by Marroquin et al. (1995) who had gold washers (10 mm in diameter, 1 mm in thickness) to simulate the clinical conditions of cast crowns each with a hole (5 mm in diameter) in the middle to simulate occlusal access cavities for the evaluation of microleakage of gold castings repaired with different materials.

In this present study, Vita VKM® 68 feldspathic porcelain opaque, body and enamel (shade B2, VITA Zahnfabrik, Bad Säckingen, Germany) and Colon Buim Type I® (Ni-Cr-Be free alloy) were used because they were indicated for fabricating PFM restorations.

The use of precious alloys was not considered in this study due to financial considerations and also non-precious Ni-Cr-alloy is most commonly used in dentistry for porcelain fused to metal restoration because of low cost (McCabe and Walls, 1998).
The etching of Ni-Cr-Be alloys removes a Ni-Be phase to create the micro-retention so important to the etched metal-resin-bonded retainer. Also, aluminum is one of the elements that are “etched” from the alloy surface to create micro-retention for resin-bonded retainers. However, Ni-Cr-Be free alloy cannot be etched (Naylor, 1992). Therefore, in this present study, the bonding to Ni-Cr-Be free alloy is enhanced by roughening using diamond bur and 35% phosphoric acid Scotchbond™ Etchant (mechanical) and Rely™ X ceramic primer (chemical).

A limitation of this present study was teeth. However, the specimens were symmetric and standardized, unlike the variation and curvatures found in natural teeth. This was purposely done to control geometric (anatomical variations) variables and allow repairing simulated access cavities.

5.1.2. Restorative Materials:

In this study, the material chosen for restoring access cavities through PFM crowns had been composite resin due to the aesthetics, low cost and easy handling. Specific adhesive bonding systems, including silane primers, with their capacity for improving chemical bonds with organic and inorganic surfaces were used.

The main rationale for using fourth generation Adper™ Scotchbond™ Multi-Purpose Plus (Multiple-step) adhesive system for all the sub-groups is to standardise the bonding agent factor so that the restorative material is the only material-related variable left; this therefore confined the evaluation of the dental composite itself. This also followed the recommendations of the manufacturer of both the Filtek™ P60 packable composite and Filtek™ Z350 flowable composite to be used in combination with an adhesive from the same manufacturer.
Several of the new adhesive systems are considered to be universal dental adhesives because they bond not only to dentine and enamel, but also to metal alloys, porcelain, and composite resin (Swift et al., 1995). Excellent bonding of composite, ceramic and PFM restorations with bond strengths of 20 to 35 MPa are possible (Craig and Powers, 2002).

Ernst et al. (2002) and De Munck et al. (2005) and Tay et al. (2005) and Fabre et al. (2007) reported that the multi-step adhesive demonstrated a better marginal seal than one-bottle adhesive and Ateyah and Elhejazi (2004) concluded that Scotchbond Multi-purpose achieved better sealing ability (less microleakage) and had highest bond strength when compared with other adhesive systems (Frankenberger et al., 2005; De Munck et al., 2005; Tay et al., 2005; Fabre et al., 2007).

Also, the main rationale of using the RelyX™ ceramic primer (silane coupling agent) was for silanisation of ceramic or metal restorations before restoration to enhance bonding to PFM-composite surfaces for improved performance and easy and simple handling. RelyX™ ceramic primer is fully hydrolysed in the bottle and ready for application. This system is designed to be used at room temperature of approximately 21-24 °C and has an increased shelf-life of up to 36 months. Rely X™ ceramic primer proved to be excellent in its ability to prime porcelain surfaces (Diaz-Arnold et al. 1989). This silane is also, however, recommended for treating the surfaces of metals (3M, 2003). Sorensen et al. (1991) and Anagnostopoulos et al. (1993) also suggested that silanes can aid in bonding to Ni-Cr alloys, to improve marginal adaptation and prevent microleakage.

The main contribution to the obtained values was made, not by the mechanical interlocking of the composite resin, but by the formation of siloxane bonds via silane (Söderholm et al., 1984). The ceramic primer activates the porcelain surface, silanating
the porcelain and depositing methyl-methacrylate, which will increase bond strength to the ceramic surface (Kurtzman and Schneider, 2006). Therefore, the use of a Rely X™ ceramic primer single-bottle prehydrolysed silane, as in the present study, makes the procedure easier, reduces error and the roughened surfaces (by super coarse diamond bur and 35% phosphoric acid) prior to silanisation may also have influenced the results in a positive manner (Kurtzman and Schneider, 2006).

The main rationale behind the use of a packable composite was to take advantage of its stiffer consistency with improved handling characteristics that made it easier to condense into the access cavities through the PFM. In this study, the main rationale behind the use of flowable composite (Filtek™ Z350) as a liner under packable composite (Filtek™ P60) was to take advantage of its flow capacity at the base of the access cavities and the formation of an elastic layer that may compensate for polymerization shrinkage stresses. Although several studies demonstrated that the use of flowable composite did not influence the microleakage (Beznos, 2001; Chuang et al., 2001a; Chuang et al., 2001b; Jain and Belcher, 2000; Loguercio et al., 2002; Malmstrom et al., 2002; Neme et al., 2002; Tredwin et al., 2005), other studies showed that the use of flowable composite results in an improved marginal sealing (Estafan et al., 2000; Tung et al., 2000; Leevailoj et al., 2001; Peutzfeldt and Asmussen, 2002; Kubo et al., 2003; Yazici et al., 2003).

5.2. Specimen Preparation:

5.2.1. Access Cavities of PFM Crowns:

In this present study, to ensure that the PFM crowns do not fracture during the making of access openings, ready-made endodontic access cavities in crowns were incorporated. A total number of eighty specimen discs 19 mm in diameter (simulated standard PFM crowns) with 5 mm diameter simulated standard endodontic access cavities in the
center of the discs were obtained by the duplication of standard washer custom made using calibrated green sheet wax 0.4 mm in thickness. The fabrication of all the specimens was accomplished with the same laboratory routine to obtain the standard design for all specimens.

In this present study, a high degree of accuracy for all specific measurements was ensured by using digital caliper, digimatic micrometer and also digimatic indicator for measurement of PFM specimens, cavity-epoxy-mould and depth of the access openings.

5.2.2. The surface treatment of the inner walls of the access cavities in PFM:
Special effort was made to control the treatment of the inner walls of all the access cavities by one evaluator with a standardised super coarse taper diamond bur by using high-speed handpiece turbine of 300,000 rpm with heavy water spray mounted on a dental surveyor. Light pressure in a circular motion in one direction for 10 seconds was used for each specimen. Every five specimens were treated with one bur (Teplitsky and Sutherland, 1985; Stokes and Tidmarsh, 1988; Sutherland et al., 1989). After that, twenty random PFM specimens were selected and measured at 5 areas in various locations along the walls of the inner cavity surfaces using a profilometer. The average of the surface roughness was $Ra = 1.11 \, \mu m$ (refer to Appendix C).

5.2.3. The fixation of the access cavities on to filled cavity-epoxy resin mould:
In this study, the main rationale for using Vitrebond™ liner/base (3M ESPE, Dental Product, St. Paul, MN, USA), a light-cured resin modified glass ionomer cement as a liner under composite resins was to simulate the clinical situation.

Before restoring the standardised access openings, the standardized cavity-epoxy-moulds were made and filled by Vitrebond™ liner/base and then each PFM sample was fixed on to the simulated cavity filled epoxy mould using highly strong glue and epoxy
resin to provide complete sealing from the bottom side. For these reasons, the RMGIC, a light cured glass ionomer Vitrebond™ liner which has high adhesion coupled with good physical properties was used (Mitra, 1991; Pitt Ford et al., 2002; Ruiz and Mitra, 2006).

5.2.4. Technique of Restoration:

The resin composites were inserted into the access cavities by (1) injecting the resin composite with a syringe which reduce incorporation of voids in the composite during insertion and facilitated placement of the material in the areas of retention; and (2) by using a plastic instrument which did not stick to the resin composites during insertion and adaptation (Estafan et al., 2000; Carig and Powers, 2002) to help eliminate bubbles and ensure good contact with the surface of the metal and porcelain (Eames et al., 1977).

According to ISO/TS (11405/2003), syringing high viscosity materials into the cavity reduces the risk of voids along the cavity walls. In this study, no porosities/air bubbles were observed by the evaluator when examining the specimens with the image analyzer system. The application of the flowable composite as a liner (0.5 mm in thickness) at the opaque porcelain-metal junction represented the application of one layer of composite, but this increased the time required for completing the restoration. This method decreased the severity of the marginal leakage as compared to a bulk filling method (Hilton, 2002a; Lopes et al., 2004; Giachetti et al., 2006; Helvatjoglu-Antoniades et al., 2006).

Incremental buildup of resin-based composite materials has been advocated to minimize polymerization shrinkage and to facilitate adequate polymerization of composite material when preparation depths are greater than 2 mm (McCullock and Smith, 1986; Lundin and Noren, 1991; Cobb et al., 2000; Gallo et al., 2000) but it significantly
increased restoration time. In this study, the main reasons for using the layering technique included easier handling, better modeling of the restoration and improved material polymerization. In contrast, the bulk light-curing procedure may result in a low degree of conversion deep inside the restoration (Giachetti et al., 2006). Also, the new packable resin-based composites reportedly have minimal polymerization shrinkage and an increased depth of cure up to 5 mm (Cobb et al., 2000; Helvatjoglu-Antoniades et al., 2006).

In this present study, the adaptation of the composite material was performed by using suitable plastic instruments (Ash No. 6 and 49) (Estafan et al., 2000; Carig and Powers, 2002). Consequently, a Mylar strip and a load of five kg for 5 minutes were used in all sub-groups and carefully applied to the restoration to optimize and standardise the adaptation of the resin composite into the standardised access cavities. Following removal of the static load, the required finishing was minimal; excess materials were removed with a scalpel #11 blade in the direction of restoration to access cavity margin (Neiva et al., 1998); then, the restorations were covered with Mylar strip to counteract the effect of oxygen inhibition and light-cured for 20 seconds by calibrated light-cure unit according to the manufacturer’s recommendations.

The study restorative materials have the same shade B2 to reduce variation in the extent of polymerization shrinkage caused by differences in colour of resin composites and cured at the same duration without exceeding layer thickness of 2 ± 0.5 mm to ensure uniform and maximum polymerization. Also, in both the light-cure materials to obtain as high energy as possible, the light guide tip was placed close to the surface of the restoration during curing for 20 seconds with a light intensity of 400 mW/cm² (Phillips, 1991; Craig and Ward, 1997; Yap, 2000; Lui et al., 2006). All these factors were kept constant using one light-cure unit in this study.
All restored access cavities were first finished on # 600 silicon carbide papers and polished by one operator using new Sof-Lex discs for ten seconds. The discs were kept in uniform light pressure and constant motion during the polishing procedures (Lui and Low, 1982; Lopes et al., 2002). The Sof-Lex polishing system was designed by 3M to polish the surface of its composite resins. It is a series of four aluminum oxide coated abrasive discs that decrease in particle size and abrasiveness from coarse (150 grit), through medium (360 grit), fine (600 grit) and super-fine (1200 grit). Previous research indicated that the Sof-Lex disc system was highly effective and the average surface roughness appeared to be sufficiently low to produce a highly polished surface that was light reflective when visually inspected (Lui and Low, 1982; Lopes et al., 2002).

In this present study, all the restored access cavities were finished and polished after 24 hours of distilled water storage, the rationale behind the delayed finishing/polishing was to allow more complete cure of the composite resin. It had been demonstrated that visible light activated resin-based composite will continue to polymerize after light curing is complete, usually having achieved a maximum by 24 hours (Watts et al., 1986; Rueggeberg and Craig, 1988; Pilo and Cardash, 1992). Studies had also demonstrated that immediate finishing of resin-based composite restorations can result in significantly increased wear (Glasspoole and Erickson, 1989) and microleakage (Fusayama and Kohno, 1989).

5.3. Specimen Evaluation:

To evaluate the influence of two important parameters of water storage and thermocycling on leakage at the PFM-composite interface, a sample size of ten for each sub-group was determined to be necessary (Mixson et al., 1991).
5.3.1. Water Storage and Thermocycling:

In this present study, thermocycling was delayed for one day so as to reduce the leakage as Crim and Gracia-Godoy (1987) had observed that restored teeth with composite that were cycled immediately exhibited a slightly higher extent of dye penetration than those stored for 24 hours in water before testing. Storage of specimens for 24 hours would allow some water sorption by the resin and subsequent expansion of the restoration. This expansion would not establish a perfect marginal seal but could contribute to less dye penetration. Conversely, immediate cycling would not permit the time necessary for this phenomenon to occur (Crim and Gracia-Godoy, 1987).

Storage factors such as time, temperature and medium may also influence microleakage patterns (Jodaikin, 1981). It was reported that polymerization of the commercially available composite resins produced shrinkage and that subsequent water sorption led to the expansion of the restorations. The length of time involved in achieving this compensation was approximately seven days (Hirasawa et al., 1983). It was reported that the water sorption of visible light and chemically activated composite resins ranged from 12.9 to 36.9 µg/mm³ and that it was characterized by a rapid increase within the first seven days which attained equilibrium after one or two months (Øysæd and Ruyter, 1986).

Water storage and thermocycling are common in-vitro methods of testing dental materials to establish their suitability for in vivo use. Exposing the specimens to thermocycling speeds up the diffusion of water in between the composite resin and the metal or ceramic. Changing the temperature creates stress at the interface of the two materials because of different coefficients of thermal expansion (Özcan, 2003). Crim and Mattingly (1981) indicated that in vitro testing to evaluate microleakage of restorations should include thermocycling. Crim et al. (1985) had shown that if
thermocycling was used, there would be no difference among techniques regardless of whether a dye or isotope was used as tracer. Research had indicated that thermocycling would hasten the process of microleakage and caused marginal deterioration of all types of restorative materials as it washed out debris, so it was more potent in demonstrating leakage than the non-cycled method (Crim et al., 1985; Hakimeh et al., 2000). Dye penetration appeared to be independent of the dwell time in thermal bath but directly related to the temperature difference (Crim et al., 1985). Short-term cycling appeared as effective in demonstrating marginal leakage as protracted cycling regimens (Crim and Garcia-Godoy, 1987).

The storage time recommended by the ISO/TS (11405/2003) was 24 hours which is normally sufficient to discriminate between those materials that cannot and those that can withstand a wet environment or 6 months when the longer periods of water storage may be necessary to show the durability of the bond in water at 37 °C. A six months storage time was a constraint as time is limited in this study. One week storage was adopted in this study was done by Hirasawa et al. (1983); Øysæd and Ruyter (1986); Sorensen et al. (1991); and Momoi and McCabe, (1994). The specimens were placed in distilled water at 37 °C for 7 days. This allowed adequate water sorption by the composite restorations (Hirasawa et al., 1983). Söderholm et al. (1984) also found that accelerated water diffusion in between the bonded materials may weaken the adhesive interface (Söderholm et al., 1984).

In the present study, the specimens of sub-group 1 (A1 and B1) and sub-group 2 (A2 and B2) were aged for 1 day in water at 37 °C as described in the ISO/TS (11405/2003). The specimens of sub-group 3 (A3 and B3) and sub-group 4 (A4 and B4) were aged for 7 days in water storage at 37 °C (Hirasawa et al., 1983; Øysæd and Ruyter, 1986; Sorensen et al., 1991; Momoi and McCabe, 1994). The temperatures of 5 °C ± 2 and 55 °C ± 2 were used in the thermocycling machine baths as an accelerated aging test. The
range of temperatures used in thermal cycling tests have an upper limit of 45-68 °C for the hot bath and a lower limit of 0-15 °C for the cold bath (Bauer and Henson, 1984; Noguera and McDonald, 1990; Gale and Darvell; 1999) ISO/TS (11405/2003) recommendations were followed in this study. However the present study had a limitation to simulate the oral cavity temperature completely, after hot or cold application because the specimens should return to normal oral temperature (37 °C) clinically by the circulation of blood and/or saliva (Kwon et al., 2002). It was not possible to immerse the specimens in another bath at 37 °C due to the limitations of the thermocycling equipment used. This therefore exposed the specimens to a more rigorous thermocycling regime.

The time of immersion of specimens in hot and cold baths had varied from a few seconds to several hours, with most investigators using 30 or 60 second exposure times. In the present study, 30 second was used in each bath as recommended by Gale and Darvell (1999). However, when testing clinically relevant dwell times (10 sec or lower), there was no significant differences in microleakage in thermocycled composite specimens compared to non-thermocycled specimens (Rossomando and Wendt, 1995). Increases in the dwell times exceeding clinical conditions may hide the thermal isolation characteristics of the resin composites (Rossomando and Wendt, 1995), resulting in fatigue to this material. The number of cycles used in many studies also varied and seemed to be selected by convenience (Chan and Glyn Jones, 1994). In addition, microleakage may be dependent on the size of the restoration; that is; the thermal conductance in relation to the total volume of the material (Rossomando and Wendt, 1995).

In the present study, the specimens of sub-group 2 (A2 and B2) were thermocycled for 500 cycles after one day water storage. The 500 cycles were chosen as recommended by
ISO/TS 11405/2003. Gale and Darvell (1999) in their review also suggested 500 thermal cycles. So far, there is no evidence of the number of cycles likely to simulate \textit{in vivo} condition and this requires further investigation. Gale and Darvell (1999) suggested an estimate of approximately 10,000 cycles per year. However, the specimens of subgroup 4 (A4 and B4) were thermocycled for 7 days (during water storage) to a total of 504 cycles (72 cycles per day) (Crim et al., 1985; Noguera and McDonald, 1990).

In this study load cycling was not used; so this is also a limitation to simulate the masticatory load process. Load cycling effects had not been examined in such great detail, but there had been a study by Hakimeh et al. (2000) who concluded that the load cycling did not have a significant effect on microleakage which confirmed the observation of Darbyshire et al. (1988).

\textbf{5.3.2. Dye Application:}

Microleakage is usually evaluated with \textit{in-vitro} models. A number of techniques including bacterial, chemical or radioactive tracer molecule infiltration are available. Colour dye penetration studies are the most commonly employed techniques. Since new materials are constantly being introduced on to the market, short-term laboratory assessments are required because clinical evaluations are expensive, time consuming and require ethical approval. In contrast, in vitro studies such as microleakage tests can provide important information on possible clinical performance of new restorative materials (Mota et al., 2003; Fabianelli et al., 2007). These are methods of screening dental materials and determining the presence of microleakage with the theoretical ability to transfer the findings to the clinical environment (Roulet, 1994; Fabianelli et al., 2007). Microleakage tests are very common research methods (Raskin et al., 2001), even if these studies had often proven contradictory and were performed with different
procedures and without standardization. Nonetheless, it was reported that microleakage tests might be reliable parameters to predict in vivo performance (Fabianelli et al., 2007).

Currently, no outstanding method is available to determine microleakage (Alani and Toh, 1997; Hilton, 2002b; Fabianelli et al., 2007). One of the oldest and most frequently used methods for the study of microleakage around restorations was the use of organic dyes (Going, 1972; Kidd, 1976a). Despite the limitations, dye leakage methodology remains a popular tool to investigate the sealing ability of restorative materials due to its low cost and very simple technique (Baure and Henson, 1984; Raskin et al., 2001). The dye penetration technique, a quantitative method, is adequate to show leakage along the restoration margin (Crim, 1987; Fayyad and Shortall, 1987, 1989; Darbyshire et al., 1988). It has been reported that the use of either dye or radioisotopes is equally effective to show microleakage (Crim and Garcia-Godoy, 1987; Fabianelli et al., 2007).

Examination of the literature reveals that whilst there are wide variations in the choice of dyes. The most dyes frequently used in dental research are provided as either solutions or particle suspensions of differing particle size depending the manufacture and the individual behaviour of the dye (Taylor and Lunch, 1992). Many dyes can be used with different particle sizes and affinity to substrates but this does not seem to influence the test results significantly (Hilton, 2002b; De Munck et al., 2005; Verissimo and do Vale, 2006).

A commonly used method for marginal leakage detection is therefore the dye penetration technique; a quantitative method showing the leakage along the restoration margin (Kopjar et al., 2006). The dye particles are small enough to penetrate and colour the surface where the adhesion of restorative material and cavity wall is insufficient
(Alani and Toh, 1997; Kopjar et al., 2006) thereby making them appropriate for this research. Some researchers believe that in vitro microleakage studies overestimate the amount of leakage that will occur clinically. This is because of the considerably smaller molecular size and weight of the tracer molecules versus larger entities such as bacteria or endotoxins that would be responsible for pathosis and secondary caries (Hilton, 2002b).

The dye method was also chosen for this study because of its extensive use and ease of use (Alani and Toh, 1997; Verissimo and do Vale, 2006; Fabianelli et al., 2007). With regard to dyes, particle molecule size, pH and chemical reactivity are expected to affect the degree of penetration (Ahlberg et al., 1995). On the other hand, Barthel et al. (1999) suggested that the molecular size of the dye may not be a relevant parameter in leakage tests.

A concern with microleakage studies is that the results might be influenced by the particular dye used. Microleakage assessment was accomplished using different methods. Some used ordinal scales with different ranges while the others used a continuous scale measurement. Of those that used the continuous scale, most reported the directly-measured microleakage in µm or mm; the while one (Sano et al., 1995) related the amount of leakage as a percentage of the total cavity-restoration interface length as reported by Hilton (2002b). Schafer and Olthoff (2002) stated that although greater linear dye penetration did not furnish data about area, it provided sufficient data about apical leakage.

In the pilot of this study, Parker Quink black ink was used. Martin-Gil et al. (2006) stated that the composition of the Parker Quink black ink was as follows: 48% carbon in the elementary state; 23% bituminous or resinous organic compounds with high carbon
content; 16% sodium sulfate; 7% calcium sulfate; 4% potassium sulfate; 1% iron(II) sulfate and 1% mineral species (not characterized) containing Ni, Cr, Cu and Zn. The presence of carbon black and indigo had the advantage of imparting a preservative effect to the ink (Martin-Gil et al., 2006). However, it was found that the black ink could not be distinguished from the colour of metal in the digital image. Thus, Parker Quink blue ink was chosen instead due to its good contrast between composite resin restorations and PFM specimens in digital images. Parker Quink blue ink manufactured using dyes (Lee et al., 2004) is a soluble dye-based ink. Parker Quink blue ink contains a dye which can stain and is generally pretty stable (Table 3 Appendix A).

Historically, the common dyes which are used in the production of blue ink are Prussian blue, Indigo and Aniline blue (Martin-Gil; Personal communication, 2007). However, the Parker Quink blue ink does not contain aniline nor Prussian blue dyes. Ink formulae are pretty closely guarded secrets of the manufacturers (Kok, personal communication, 2008), so it is difficult to tell exactly which dye is used in the ink. The activation energy of Quink blue ink is 15.6 kJ mol⁻¹ and its density is 1.193 g cm⁻³. The viscosity coefficient of Quink blue ink is 0.98 cp and is inversely proportional to the mobility. Therefore, the ink with a higher viscosity coefficient is more difficult to diffuse than the ink with a lower viscosity coefficient. On the other hand, the viscosity coefficient of Quink blue ink is 0.98 cp, which indicates that the ink with a smaller viscosity coefficient would result in an expected lower energy barrier for ink diffusion in water (Lee et al., 2004).

The pigment which is principally used in the production of blue ink is *Prussian* blue. It is first digested for two or three days with either strong hydrochloric acid and sulfuric acid or nitric acid. The digested mass is largely diluted with water after setting the
supernatant liquid. This sediment is repeatedly washed till all traces of iron and free acid disappear from the used water. Then, it is dried and mixed with oxalic acid in the proportion of 8 parts of Prussian blue to 1 part of the oxalic acid. In this condition the material must be ready to be dissolved in water to the degree of colour intensity necessary (Martin-Gil, personal communication, 2007).

Dye-based ink, such as Parker Quink blue ink, can produce much more colour of a given density per unit of mass. An additional advantage of dye-based ink system is that the dye molecules interact chemically with other ink ingredients. This means that it can benefit from optical brighteners and colour-enhancing agents designed to increase the intensity and appearance of the dye. As the dye gets its colour from interaction of electrons in their molecules, the way in which the electrons can move is determined by the charge and extent of electron delocalization in the other ink ingredients. The colour emerges as a function of the light energy that falls on the dye. Nevertheless, in spite of all its advantages, dye-based ink can be more susceptible to fading especially when exposed to ultraviolet radiation as in sunlight (Lee et al., 2004).

Parker Quink blue ink, however, is not the same as carbon particles of known diameter, and therefore leakage is likely to be greater. Parker Quink blue ink was chosen as a suitable tracer to determine the microleakage at the PFM-restoration interface. It was an efficient tracer as it could be detected immediately. The detection of the Parker Quink blue ink was easy because its colour differed from that of PFM-restorative materials. Parker Quink blue ink is also a chemically non-reactive agent (Kok, personal communication, 2008), unlike some other dyes like methylene blue dye, for example, which in contact with reducing agents that exist in restorative materials, may be reduced to a colourless substance (Wu et al., 1998). Furthermore, Parker Quink blue ink is a pH-
neutral solution of dye-based ink and therefore does not require buffering. Conversely, other dyes like methylene blue, silver nitrate and basic fuchsin need to be neutralized before being used (Youngson et al., 1998). It is also a simple and cheap and has a well-defined, excellent, visible colour contrast in digital images; hence providing the evaluators with a clear reference point from which to score and measure quantitatively when using a computerized image analyzer system (Fayyad and Shortall, 1987, 1989; Almeidia et al., 2003).

Fayyad and Shortall (1987) assessed dye penetration by using an image analysis apparatus linked to a stereomicroscope. Digital imaging microscopy was used to record the actual length of the dye penetration along the interface. The area of dye penetration was selected, since this would indicate the amount of dye penetration along the PFM-composite restoration interfaces. The specimens were photographed and colour transparencies were made. The transparencies were then evaluated using an image analyzer. Nevertheless, image analysers are prone to some subjective problems in that the threshold for distinguishing between the presence and absence of stain usually needs to be set prior to analysis by the operator (Glyn Jones et al., 1988; Taylor and Lynch, 1992).

Two coats of fingernail varnish were painted 1 mm from the interfaces between the PFM and the restored access openings to prevent dye penetration from anywhere else other than at the interfaces. Special attention was also given to ensure maximum sealing capability in other areas of the specimens. The possible route of dye penetration was therefore along the PFM-composite interface.

All eight sectioned sides of each specimen were determined and their depths of dye penetration were measured in millimeters using an image analyzer. This measurement of the interfacial microleakage was considered more acceptable than score assessment because it permitted quantitative and parametric statistical analyses of the data. Although
the dye penetration test is not a quantitative test such as a three-dimensional leakage test, stereomicroscope examination of dye penetration has been used as a qualitative test (Yap et al., 1997). However, computerized image analysis allowed quantitative measurement of the length of dye penetration which has occurred (Fayyad and Shortall, 1987, 1989) and the percentage area of crown dentine exhibiting staining (Glyn Jones et al., 1988). These measurements can therefore be used subsequently as an assessment of the degree of restoration leakage (Youngson et al., 1990).

5.3.3. Sectioning and Assessment:

Before sectioning, the restored PFM specimens were embedded completely in a clear epoxy resin in order to accurately identify the midline plane. Because the way (direction) that the specimens are cut may influence leakage measurements (Raskin et al., 2001), in this present study, the length of the PFM-composite interface was equally cut on all 8 sectioned sides of each specimen, so that the extent of dye penetration in each sub-group might be compared.

The specimens were measured by one evaluator (Darbyshire et al., 1988) with a single blind technique (Darbyshire et al., 1988). Three readings for the eight sides of each specimen were taken at three different times (Tjan et al., 1992; Baldissara et al., 1998; Howdle et al., 2002) with a three day interval between each evaluation (Annuar and Abdullah, 2003). Dehydration procedures could cause shrinkage of the composite resins but this did not compromise evaluation of leakage patterns. Each evaluation was carried out independently without reference to the previous measurement to eliminate evaluator bias.

For maximum penetration of dye leakage, for the eight sides the greatest measurement was taken as representative of each specimen (Déjou et al., 1996; Trautmann et al., 2001b; Bijella et al., 2001; Howdle et al., 2002; Raskin et al., 2003) and the results were
analyzed by parametric and nonparametric tests. In the mean measurement, the depth of dye leakage was taken from all eight sides for each specimen (PFM-filling materials interface) as representative data (Darbyshire et al., 1988; Déjou et al., 1996; Trautmann et al., 2001b; Bijella et al., 2001) and parametric tests were used to analyse them. This study was based on earlier studies (Faayad and Shortall, 1987, 1989; Darbyshire et al., 1988, Déjou et al., 1996; Trautmann et al., 2001b; Bijella et al., 2001) where two different microleakage assessment techniques were combined with the purpose of clarifying the data on the outcomes of the statistical analysis in a dye penetration study. Each measurement may be considered as a statistical criterion (dependent data) (Déjou et al., 1996).

Gale et al. (1994) clearly demonstrated that microleakage is a 3-D phenomenon and that different locations and angles of sectioning might result in completely different penetration scores. Therefore, methods for three-dimensional evaluation of microleakage have been proposed (Gwinnett et al., 1995) because in a single section, the amount of microleakage is underestimated. Therefore, in this study, in order to take this into account, three sectionings giving eight interfaces of each specimen were examined.

Lucena-Martin et al. (2002) affirmed that it was clear that three-dimensional evaluation techniques revealed more accurate and more extensive dye penetration than sectioning techniques that provided a two-dimensional view only. However, the three-dimensional techniques are more time consuming and expensive and some do not allow good visualization of dentin tubule leakage (Hilton and Ferracane, 1998). When a sectioning technique is to be utilized for dye assessment, multiple sections, should be utilized (Hilton, 2002b).
Commonly, microleakage had been assessed by ascribing a score to each surface exhibiting dye penetration, the scores being weighted where it was felt that a particular surface was of greater importance (Glyn Jones et al., 1979). These scoring systems, although widely used, were felt to be arbitrary and insensitive and therefore more objective methods of assessment have been sought. Fayyad and Shortall (1987) endeavoured to overcome the deficiencies of a scoring system by evaluating the length of dye penetration along the cavity-restoration interface using an image analysis apparatus linked via a viewing tube to a stereomicroscope. The image analysis apparatus was considered to offer the degree of sensitivity required, the extent of dye penetration was measured along the interface in a given time in the present study. This method allowed a more objective assessment to be made of the extent of marginal leakage in comparison to traditional methods where qualitative judgments had been made for the depth and path of tracer penetration (Fayyad and Shortall, 1987). Linear dye penetration expressed in millimeters may have different significance, depending on the variations in specimen’s size and shape. The importance of 2 mm microleakage in a restored access cavity with a 2 mm high clinical crown is likely to be greater than in a crown 8 mm high. Thus, linear measurement allowed a better evaluation of coronal microleakage in the restored access cavities of PFM crowns.

5.4. Results:

The wide variety of dye materials and different techniques used in in-vitro tests make comparing results of different studies difficult since there is no generally accepted standard for experimental parameters, such as selection of teeth, the way of bringing the materials into operation, the type of dye, type and concentration of the storage solution, time of storage, temperature during storage, type and duration of thermal cycling and/or mechanical cycling and the criteria to examine the microleakage (Helvatjoglu-
Antoniades et al., 2000). In addition, there is lack of correlation between in-vitro and in-vivo studies (Roulet, 1994), since in-vivo studies present some conditions that could hardly be reproduced in-vitro. Nevertheless, results of in-vivo studies are often less negative than in vitro studies. But, in-vitro testing is essential for development purposes. Therefore, it follows that in-vitro results should be viewed as a theoretical level of maximum leakage which may be expected in-vivo (Pashley, 1990). Although a wide variety of methods had been described with some variations in results, the standardization of such methods is therefore necessary in order to obtain comparable results from different studies (Fabianelli et al., 2007).

In this study, to be acceptable the measuring was done by a single operator in a single blind design and to evaluate the effect of potential measurement errors, each independently coded section was measured three times. An intra-class correlation coefficient in excess of 0.98 was obtained in the analysis of intra-rater reliability. The results of this study demonstrated the high reproducibility of results of the measuring system. The use of a similar measurement system had been reported by Crim (1987), Darbyshire et al. (1988), Hakimeh et al. (2000) and Trautmann et al. (2001b).

In this study, the experimental design included two restorative procedures, two periods of water storage and with/without thermocycling. This experimental design lends itself most naturally to a 3-way ANOVA, which not only takes into account each of the three independent variables but also allows an analysis of interaction among the variables. It requires that the data be normally distributed; if the data are not normally distributed, then transformation of the data (such as logarithmic or square root) is undertaken. Only if non-normality persists is it necessary to use a non-parametric test.
The results of this study, statistically, showed that the measuring validity test (intra-rater correlation) to be excellent and the reproducibility of results is high reliable. However, at times the variances were found to be different that made the parametric test invalid and so the Mann-Whitney U test was used for confirming the parametric analysis due to a lack of normal data distribution. Therefore, both the parametric and nonparametric procedures were used to test the objectives and when the assumption for the parametric test was not valid, the non-parametric procedure was used.

5.4.1. General Results

Microleakage tests are the most frequently used laboratory tests to study the mechanisms that may minimize, or eliminate, the leakage around dental restorations. A microleakage test is a useful method in the investigation of resin composite restorations.

Nonparametric scoring methods (Kidd, 1976a; Glyn Jones et al., 1979; Swift, 1991; Mixson et al., 1991; Tjan and Chiu, 1989; Tjan et al., 1992; Baldissara et al., 1998; Raskin et al., 2003), widely used in microleakage studies, are limited if compared with a continuous variable evaluation scale (Crim, 1987; Faayad and Shortall, 1987, 1989; Darbyshire et al., 1988; Lacy et al., 1992; Dejou et al., 1996; Howdle et al., 2002). However, linear dye penetration expressed in millimeters may have different significance, depending on the standardization in specimens’ size and shape or on the variations in specimen’s size and shape. From this point of view, linear measurement allows a better evaluation of coronal microleakage in restored access cavity of PFM crowns (Camps and Pashley, 2003; Bodrumlu and Tunga, 2007). The image analysis apparatus was considered to also offer the degree of sensitivity required. Fayyad and Shortall (1987) had endeavoured to overcome the deficiencies of a scoring system by evaluating the length of dye penetration along the cavity-restoration interface using an
image analysis apparatus linked via a viewing tube to a stereomicroscope. In this study, the results were obtained by linear measurement. However, it represented a bi-dimensional perspective of a three-dimensional phenomena (Gwinnett et al., 1995).

As seen in Tables 4.1 and Table 4.6, an examination of the means and standard deviations (mm) for restorative technique, water storage and thermocycling in both criteria indicated that, for the maximum dye penetration, leakage (mm) was higher than the mean dye criteria. Also leakage at the restored access cavities of PFM-composite resin interfaces was found to be significantly greater thermocycling compared to other factors in both criteria. From this, it appeared that the interfaces between the composite resins restored access cavities in PFM were affected significantly by thermocycling in both evaluation criteria (mean and maximum dye penetration) than by water storage.

Considering the microleakage mean values (Table 4.1), the results of the 3-way ANOVA revealed that the restorative techniques (packable composite without and packable with flowable composites) had no significant effect on the coronal microleakage ($P=0.135$). Whereas the effect of water storage and thermocycling had significant effect on the mean coronal microleakage ($P=0.000$). However, when 2-way interaction effect between each study factor was checked, it was found that there was a significant interaction effect between thermocycling and water storage on the mean microleakage ($P=0.009$) as shown in Table 4.2. It indicated that thermocycling effect on coronal microleakage might differ between one-day and seven-day water storage. Therefore, a separate analysis was done to locate the effect of thermocycling and water storage on coronal microleakage.

Based on T-test, there was a significant interaction between the specimens, showing that thermocycling influenced the tested materials in different ways. Also, based on Mann-
Whitney U test, there was a significant interaction between the specimens, showing that water storage influenced the tested materials and seven-day water storage specimens had lesser coronal microleakage than one-day water storage specimens.

Considering the maximum microleakage values (Table 4.7), the result of Mann-Whitney U test, there was no significant difference in maximum dye penetration between the two restorative techniques on coronal microleakage ($P=0.176$). Thus, it indicated that restorative technique had no significant effect on the coronal microleakage. However, water storage ($P=0.031$) and thermocycling ($P=0.000$) had significant effect on coronal microleakage.

The results of this study revealed that the maximum dye penetration seemed to reduce the mean criteria as it lowered the results. Ranking the restorative systems as a function of maximum dye penetration seemed to comply with the role as defined by Pashley (1990), Dejou et al. (1996), Bijella et al. (2001), Trautmann et al. (2001b) and Howdle et al. (2002). Thus, the maximum dye penetration was able to explain the results in this study which were in agreement with those obtained by Dejou et al. (1996) who stated that the maximum penetration parameter is the most suited comparative measure for clinical conditions, because even if it was small, but when the significance level of 5% was applied, it was possible to see the interaction between materials as also confirmed by the observations of Trautmann et al. (2001b) and Bijella et al. (2001).

Wenner et al. (1988), in a composite study, had two independent raters score six surface from three sections per restoration and assigned the highest of the six site scores to each restoration. This worst-case-analysis scoring method could overestimate microleakage, since it would have the disadvantage of being unable to distinguish between massive and isolated microleakage around restorations. Nevertheless, whichever microleakage
evaluation criterion is selected, it is important for the investigator to justify its use. Some investigators had argued that it was necessary to score multiple-sectioned surfaces of a composite restoration to describe microleakage accurately (Wenner et al., 1988).

The maximum degree of leakage in a clinical situation might be considered to be far more important than the average leakage (Trautmann et al., 2001b). The maximum dye penetration is the most important measurement, because it represents the greatest potential for bacterial penetration and hence the potential for failure of the root fillings (Howdle et al., 2002). Also, it had been reported that bacterial recontamination along the entire length of a coronally unsealed root restoration could occur within 19 days (Torabinejad et al., 1990), therefore, leakage to the root filling would indicate the potential for periradicular pathosis (Howdle et al., 2002).

5.4.2. Comparison of Results:

Various studies have reported on the microleakage of resin composite restorations. Conflicting results are probably due to experimental inconsistency and material properties. The effects of restorative placement technique, cavity design, water storage time, thermocycling and occlusal loading procedure all seem to affect the amount of microleakage (Crim and Mattingly 1981; Crim and Garcia-Godoy 1987; Chan and Glyn Jones, 1994; Hakimeh et al., 2000; Yap, 2000; Trautmann et al., 2001b; Hilton, 2002a, 2002b; Braga et al., 2003; De Munck et al., 2005; Fabianelli et al., 2007).

5.4.2.1. The Effect of Restorative Technique on Coronal Microleakage:

The results of this study agree with the literature, when all specimens leaked; this could be explained by the initial shrinkage of composite resins due to polymerization shrinkage stress which resulted in a pull back of the restoration toward the light source.
and pulling with it the adhesive bonding system with subsequent gap formation and increased microleakage (Craig and Ward, 1997). Since polymerization shrinkage is dependent on the amount of resin matrix in non-polymerized form and the greater the concentration of inorganic filler, the smaller the matrix concentration in that form, it can be stated that a high concentration of inorganic filler reduces the final polymerization shrinkage. The percentage of volumetric polymerization shrinkage for Filtek P60 is 2.1% (Leinfelder et al., 1999; Aguiar et al., 2003; da Silva et al., 2007), while Filtek Z350 is 4% (Bayne et al., 1998; Labella et al., 1999; Stavridakis et al., 2005; Tredwin et al., 2005).

An inorganic filler content, which also interferes during the polymerization shrinkage process, may be found in Filtek P60 as round zircon and silicon particles that are not treated with silane, in proportions of 61% by volume or 83% by weight. Inorganic fillers in dental composites are typically coated with silanes in order to improve the bond to the resin matrix and increase the service life of the composite (Chen and Brauer, 1982; Bayne et al., 1994; Wahab et al., 2003; Blalock et al., 2006).

The results of this present study, revealed that none of the studied materials was able to totally prevent microleakage at the PFM-composite resin interface (though packable composite restorations had 2 specimens and packable/flowable composite restorations had 3 specimens which did not show dye leakage in any location), although there was no statistically significant difference between them. P60/Z350 presented the smallest microleakage and the P60 alone presented the highest leakage although the polymerization state of all specimens was relatively identical at the time of finishing and thermocycling and simulated cavities were standardized and their depths were equal.
In this study, in both mean and maximum dye penetration, the results demonstrated that the restorative technique did not significantly affect the mean microleakage at PFM-resin composite interface although it was slightly decreased in group B compared to group A (subjected to the same conditions). This can be due to the effect of the following factors:

- Standardization of the model of this study and/or thickness of flowable composite (0.5 mm) (Malmström et al., 2002; Braga et al., 2006). Consequently, the C-Factor was the same for all specimens. Feilzer et al. (1987) described the ideal C-Factor as being lower than 1, and that when the C-Factor is greater than 1, the results are unexpected (Aguiar et al., 2003; Braga et al., 2006; da Silva et al., 2007).

- Variables, such as cavity size and shape, techniques of placement, polymerization of the composite, silane coupling agent and adhesive system, were eliminated. Also, only one packable and one flowable composite were used and the resin increments were not larger than 2 mm in order to achieve optimal polymerization.

- Similarity of their chemical compositions; the matrix of P60 (61% vol, 83% wt) is almost based on Bis-GMA monomers with Bis-EMA and UDMA resins (3M Technical Profile, 1998), the matrix of Z350 (55% vol., 65% wt) are also based on Bis-GMA, TEGDMA and Bis-EMA resins (3M Technical Profile, 2005).

- The fillers of packable resin-based composite are added in the form of irregular rounded zirconia/ silica particles (Filtek P60- 61% filled by volume, particle size range 0.01-3.5 μm) (Gallo et al., 2000; Leevailoj et al., 2001; Bala et al., 2003). The filler loading and particle size can affect the leakage (Craig et al., 1996; Wahab et al., 2003; Braga et al., 2006; Papadogiannis et al., 2007).
- The viscosity controllers might be responsible for the different behaviour of these materials with more viscosity controllers in P60 than Z350, though the difference is slight (Braga et al., 2003; Tredwin et al., 2005; Pongprueksa et al., 2007).

The results of this study revealed that there was no statistically significant difference between two restorative techniques (packable composite without and packable with flowable composites) in both mean and maximum dye penetration, which is in agreement with several studies (Beznos, 2001; Chuang et al., 2001a; Chuang et al., 2001b; Jain and Belcher, 2000; Loguercio et al., 2002; Malmstrom et al., 2002; Neme et al., 2002; Tredwin et al., 2005) who demonstrated that the use of flowable composite did not influence the microleakage, and, disagreement from other studies showed that the use of flowable composite results in an improved marginal sealing (Estafan et al., 2000; Tung et al., 2000; Leevailoj et al., 2001; Peutzfeldt and Asmussen, 2002; Kubo et al., 2003; Yazici et al., 2003).

Although the literature is equivocal on the benefit attributed to flowable materials in reducing microleakage or gaps at the bonded interface, the benefit associated with the reduction of voids or gaps during placement of traditional composite is intuitive. Marginal adaptation under packable resin composites has shown to be improved by the use of a flowable, as reported by Fabianelli et al. (2003) and Helvatjoglu-Antoniades et al. (2006). It had been suggested that the use of a low modulus flowable as a liner may absorb some of the polymerization stress of a conventional composite. Although Braga et al. (2003) measured some stress relief when a flowable was modeled for use as a liner, its magnitude was found to be low and variable depending upon the flowable examined. The authors also note, however, that the lower modulii of the flowable composites could be beneficial in relieving stress at the bonded interface created by
mechanical and thermal stimuli. Thus far, this benefit as measured by lower microleakage or reduced gap formation had been found in some laboratory studies (Ferdianakis, 1998; Tung et al., 2000; Leevailoj et al., 2001; Yazici et al., 2003), but not in others (Jain and Belcher, 2000; Wibowo and Stockton, 2001; Tredwin et al., 2005).

5.4.2.2. The Effect of Water Storage on Coronal Microleakage:

In this study, in both mean and maximum dye penetration, the results demonstrated that water storage was significantly affect the mean coronal microleakage at the PFM-resin composite interface; there was a slight decrease in mean coronal microleakage in seven-day water storage specimens than the one-day water storage specimens. These slight differences could be explained by slightly more water uptake (water sorption) at 7 days than 1 day (hygroscopic expansion). Previous studies examining the degree of hygroscopic expansion in resin-based composites had shown an inverse relationship between filler loading and water sorption (Øysæd and Ruyter, 1986; Momoi and McCabb, 1994). As the volume of filler increases, the amount of water absorbed into the matrix is reduced. This was in accordance to the findings of this study.

The phenomenon of water sorption of resin composites and their resulting hygroscopic expansion can compensate for resin composite shrinkage. Although hygroscopic expansion may lead to a substantial relaxation of polymerization contraction stress, bonded surfaces are kept from direct contact with water and are restricted in their expansion. As a result, hygroscopic expansion will contribute to the relaxation of shear stress parallel to the adhesive interface. In contrast to the rather rapid polymerization contraction stress development, hygroscopic relief proceeds slowly and might require days. Neither the original contraction stress nor the hygroscopic expansion will be uniform throughout the restoration (Giachetti et al., 2006). Water exerts a plasticizing
effect on the polymer, which degrades its mechanical properties. As a result, the quality of the bonding interface might be compromised (Fabre et al., 2007). Thus, both water sorption and solubility would lead to a variety of chemical and physical processes that may result in deleterious effects on the structure and function of dental polymers, including their retentive capacity in adhesive dentistry (Malacarne et al., 2006).

5.4.2.3. The Effect of Thermocycling on Coronal Microleakage:

In this study, in both mean and maximum dye penetration, the results demonstrated that thermocycling significantly increased the mean coronal microleakage at the PFM-resin composite interface.

Exposing the specimens to thermocycling speeds up the diffusion of water in between the composite resin and the metal or ceramic. Changing the temperature creates stress at the interface of the two materials because of different coefficients of thermal expansion (Özcan, 2003). This was due to the high differences between the linear coefficient of thermal expansion of porcelain (14 x 10^-6 pp/°C), nickel-chromium beryllium free alloy (15.38 x 10^-6 pp/°C) and composite resins (packable composite: 28-35 x 10^-6 pp/°C; and flowable composite: 35-50 x 10^-6 pp/°C) (Craig and Ward, 1997; Roberson et al., 2002) and confirmed by Bullard et al. (1988), Momoi et al. (1990), Sorensen et al. (1991), Marroquin et al. (1995), Bala et al. (2003) and Wahab et al. (2003).

When thermocycled and non-thermocycled specimens were compared, in both mean and maximum dye penetration, there was a statistically significant difference between their means. The thermocycled specimens had significantly higher microleakage than non-thermocycled specimens. This could be due to thermal fatigue (stress) and mismatch of coefficient of thermal expansion and the thermal conductivity of metal.
However, in non-thermocycled specimens, since the restorations were not thermally challenged, the extent of leakage of the composite restorations could be attributed to the polymerization shrinkage of the resin system or to the mechanical destruction of the bond during application. A possible explanation for these results was that the polymerization shrinkage was responsible for the microleakage values. Even though the bond between PFM-composite resin interfaces may be disturbed initially during curing because of shrinkage, water sorption can cause gap reduction by hygroscopic expansion over time (Thonemann et al., 1997; Fabianelli et al., 2007). This factor may help these restorations achieve a better seal over time and, thus, add to their success and longevity (Gallo et al., 2000), though it may impair the mechanical properties of the resins (Hansen and Asmussen, 1989; Momoi and McCabe, 1994; Giachetti et al., 2006). The importance of water uptake in the long-term durability of restorations is still under question, but it may be related to water tree formation as described by Tay and Pashley (2003). Water diffusion into the bonding interface causes the resinous components to swell and become plasticized (Fabre et al., 2007). The extent of nanoleakage increases with time in relation to water absorption (Tay et al., 2003; Fabianelli et al., 2007). During the thermocycling test, hot water might accelerate the hydrolysis of the resin and extract poorly polymerised resin oligomers (Asaka et al., 2006). This process can possibly lead to marginal gap formation and resulting leakage (Bala et al., 2003). Furthermore, the differences in the coefficients of thermal expansion of the resin-based composites and PFM can lead to different volumetric changes which could directly affect leakage (Bullard et al., 1988; Momoi et al., 1990; Sorensen et al., 1991; Marroquin et al., 1995; Schuckar and Geurtsen, 1997).

For the thermocycling test, the specimens were subjected to thermal cycles that simulated the intraoral temperature. However, the literature showed that there was a
wide range in temperature extremes, transfer times between baths and dwell times (Shortall, 1982; Crim et al., 1985; Gale and Darvell, 1999). Thus, there was no standard for thermocycling methodology in microleakage studies and this permitted contradictory discussions and results in various laboratory studies. In some studies, the variables chosen were only restricted to the thermocycling method and were not intended to understand the meaning of these effects (Gale and Darvell, 1999). Because of this, in the present study the temperature was standardized at 5ºC-55ºC ± 2ºC and the dwell time was 30 seconds. These variables seem able to be tolerated by the oral tissues and should be suitable for clinical conditions. Also, in this study, a constant number of cycles (500 cycles after one day water storage and 504 cycles within seven days) were selected for evaluation to determine if there was a direct relation with the increase of microleakage in the restored access cavities.

For this study, in both mean and maximum dye penetration, the results showed that there were significant differences in the coronal microleakage between one-day and seven-day water storage with thermocycling ($P>0.05$). Thus, it indicated that thermocycling for 500 cycles after one-day water storage was more pronounced than thermocycling for 504 cycles within seven-day water storage. The increase in coronal microleakage of the thermocycled specimens following 500 cycles after one day water storage could be due to thermal fatigue (stress) and mismatch of coefficient of thermal expansion and the thermal conductivity of metal. However, the thermocycled specimens for 504 cycles within seven day water storage had less coronal microleakage; this was because they could not reach threshold fatigue (72 cycles per day so, less thermal stress). A possible explanation for these results was that the polymerization shrinkage was responsible for the coronal microleakage values. Thus, 72 cycles per day did not cause enough fatigue as 500 cycles; this meant composite resins in the oral cavity should be able to withstand coronal microleakage over time.
The low thermal conductivity of resin composite suggests that a 15 s dwell time is not sufficient to transfer the temperature through resin composite restoration to fatigue the adhesive interface and rupture it (Shortall, 1982). Although, Crim et al. (1985) reported that microleakage seemed to be independent of the dwell time used, studies by Darbyshire et al. (1988) and Schuckar and Geurtsen (1997) showed that thermocycling with a 30s dwell time promoted an increase in microleakage. This 30s dwell time was observed in the present study in the thermocycled groups.

Another study showed a gradual increase in microleakage during the 7 weeks of the thermocycling test (Momoi et al., 1990). However, the dwell times used were 2 minutes and, possibly, the dye penetration could have occurred due to degradation of the adhesive interface in this period of time and not due exclusively to the increase of thermal cycles. In addition, microleakage may be dependent on the size of the restoration, that is, thermal conductance in relation to the total volume of the material (Rossomando and Wendt, 1995).

Various microleakage studies that compared thermocycled and non-thermocycled groups (Wendt et al., 1992; Chan and Glyn Jones, 1994; Rossomando and Wendt, 1995; Wibowo and Stockton, 2001; Bijella et al., 2001; Aguiar et al., 2003) and also the different numbers of cycles (Crim and Garcia-Godoy, 1987) observed no statistically significant difference. However, in some studies (Crim and Mattingly, 1981; Crim et al., 1985; Litkowski et al., 1989; Schuckar and Geurtsen, 1997; Hakimeh et al., 2000; Wahab et al., 2003) there were significant differences in marginal microleakage of resin composite restorations between thermocycled and non-thermocycled groups. These results suggest that the thermocycling method was a suitable test to simulate the real significance of temperature changes in clinical conditions.
The relevance of the studies in which thermocycling was applied for a shorter period of time should be questioned. There seems to be a lack of agreement that water storage and thermocycling have decreasing effects on the resin–ceramic bond. The main reason for this could be attributed to various thermocycling times in the experiments (Gale and Darvell, 1999; Özcan, 2003; Fabianelli et al., 2007).

An in vitro evaluation is the first step for testing any technique or material. Although the specimens and testing conditions of the present study were designed to simulate the clinical use of restoring access cavities of PFM crowns, in-vivo tests are needed to confirm the results. Moreover, this study tested interfacial microleakage in restored access cavities of PFM crowns in relation to the resin material only. Other aspects of the PFM-resin interface such as, bond strength and the effect of different retention systems should also be investigated.

Regarding thermocycling, in vitro studies on microleakage demonstrated that, in spite of influencing the materials in different ways, it resulted in statistical differences. As to water storage, in vitro studies on microleakage demonstrated that, in spite of influencing the materials in different ways, it did lead to statistical differences. As to the methods for the evaluation of microleakage, maximum penetration seemed to have greater clinical value since it detected more precisely the extent of microleakage.

In order to reduce coronal leakage along all margins of PFM-restoration interfaces, surface treatment (sandblasting, etching, bonding agent), need to be developed that bond strength to both metal and porcelain is strong enough to resist polymerization shrinkage forces, water and thermocycling. Additionally, composite resin materials with reduced shrinkage, thermal expansion and resistance to water sorption are needed. Additionally, an understanding of choice appropriate restorative materials is needed when restoring these access cavities.