

Original Article

**QUANTITATION OF MICROLEAKAGE FROM ROOT CANALS
OBTURATED BY FOUR DIFFERENT TECHNIQUES***

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تمت مقارنة أربع طرق لحشو قنوات الجذور وهي الحشو بالتكثيف العامودي والتكثيف الجانبي وباستخدام الثيرمافيل . تم حساب الانتشار الراجع للنظائر المشعة كطريقة حديثة تمكن ن تقييم كمية التسرب وليس فقط الكيفية وذلك في خلال وقت محدد . فقد وضعت النظائر المشعة بداخل الأقفنية المحشوية وتم حساب كميات التسرب التي حدثت خلال فترات زمنية مختلفة واستمرت عملية التقييم إلى ٤٦٣ يوماً . كان إجمالي العينات المستخدمة ٢٠١٦ عينة .

أظهرت الدراسة أن طريقة الثيرمافيل أقل تسرباً في بداية الدراسة ولكنها أصبحت بعد ذلك أكثر الطرق تسرباً للنظائر . أظهرت طريقة التكثيف الجانبي نتيجة عكسية للثيرمافيل حيث نتج عنها كمية تسرب عالية في البداية أصبحت بعدها أقل الطرق تسرباً مع تقدم الزمن . أظهر التكثيف العامودي أكثر الطرق تسرباً في البداية وكان معدل التسرب الحاد باستخدام الثيرمافيل مشابهاً لهذا الأسلوب في الحشو .

The reference-controlled reverse diffusion method was used to quantitate microleakage from root canals. The canals were obturated using one of four different techniques: Thermafil, Obtura, lateral condensation or vertical condensation. The radioactive material was placed inside the obturated canals and was allowed to leak in the surrounding medium. The quantity of microleakage was determined at various time intervals and up to 463 days relative to microleakage quantities occurring from open canals. Using the quantitative data obtained from 2016 aliquot samples, a linear relationship was established between the predicted quantity of microleakage and time for each obturation method. Each linear relationship yielded a microleakage onset value (K.) and a rate of microleakage (n). While Thermafil resulted in the lowest microleakage onset, it permitted the largest microleakage rate over time. Lateral condensation showed strong microleakage onset, yet the least microleakage rate. Vertical condensation permitted the strongest microleakage onset (2.6 times that of Obtura). yet its microleakage rate was essentially equal to that of Obtura. The quantitative data and its linear analyses explains the diversing literature reports and furnishes an opportunity to objectively quantitate microleakage in endodontics.

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Introduction

The reliability of the reported results of microleakage study methods is questionable. The currently used methods to determine microleakage are destructive, qualitative, of short duration and incapable of reporting microleakage in real time. The literature also reports significant differences between evaluators² using the same qualitative test. Efforts to improve the quality of the testing methods have been reported. These efforts may have eliminated some of the problems of some of the qualitative testing methods, and reduced their subjectivity, yet several other problems remained unresolved. Based on these findings and further review of the microleakage methodology literature, a non-destructive method was re-introduced.⁴ The method objectively quantitates microleakage in real time using reverse diffusion but with reference control. To ascertain the objectivity and reliability of the method, its experimental parameters were tested and standardized.⁵

The literature pertaining to microleakage associated with various endodontic obturation techniques and materials highlights the subjectivity of the current testing methods. While several studies report no significant differences among two or more obturation methods, other studies report significant differences. These conflicting reports suggest the need to use a sensitive, yet well controlled quantitative method to discern the similarities or differences among obturation methods.

The goals of this study were to objectively quantitate microleakage associated with four obturation methods up to 463 days in real time using the reference controlled reverse diffusion method and to mathematically determine a microleakage onset value and a rate constant for each method.

Materials and Methods

Based on a pilot study, the number of specimens that yields meaningful data at the 95% confidence interval was determined to be six.¹⁹ Thirty extracted human single rooted anterior teeth were collected and stored in water. None of the teeth have been previously exposed to endodontic treatment. Crowns of the teeth were cut off at the cervical margins. The working length of all roots was determined with a ruler; the difference did not exceed 2 mm. To maintain a standard patency of the apical foramen, a #15 K-File was set 2 mm longer than the working length. The canals were instrumented using serial filing up to size #50 File and up to 0.5 mm short of the apical foramen. All canals, except the group for Thermafil technique were flared three more sizes using the step back method and circumferentially filed. During canal instrumentation, sodium hypochlorite (2.5%) solution was used for irrigation between file sizes. A spreader was used to check the proper flare-up of the canal. The total surface of the root was covered with two layers of nail varnish leaving the apical 3 mm uncovered to permit leakage. The thirty roots were then divided randomly into five groups of six teeth each. Each of four groups was obturated using one of four techniques using AH-26* as a sealer cement.

Group 1 was filled with the lateral condensation technique. A size 50 Master cone that fits snugly at 0.5 mm short of the apical foramen was used. Accessory cones, size fine-fine, were condensed laterally.

Group 2 was filled with the vertical condensation method using warm gutta-percha and an endo plugger.

Groups 3 was filled with the high temperature thermoplasticized molded

*De Trey Division, Dentsply Ltd., Weybridge Surrey, England

gutta-percha technique . Group 4 was filled using the correct size of plastic core Therafil device in accordance with the manufacturer's instructions*. A radiograph was taken to confirm the length and total canal obturation for the four groups.

The pulp chamber area of each root was cleaned and the gutta-percha at the orifice of the canal was removed from groups 1-4 such that the apical 6 mm remained obturated.

Group 5 was instrumented in the same manner but was not obturated. Each root was split along the root canal to two halves. The standardized reference controlled reverse diffusion method of quantitating microleakage in real time was used.⁴

Two microliters of ³H-Alanine radiotracer⁵ were deposited in each canal of the experimental groups or half canal of the control group and dried under vacuum. The access opening of the experimental group were then filled with IRM paste then painted with two layers of nail varnish.

Each root (half root for control group) was placed in a double orifice 50 mL conical flask* containing 20 mL artificial saliva^{**} and was shaken continuously at 80 times/minutes in a water bath⁵⁵ at 37°C to simulate the oral condition. The radioactive material diffused from the root canal into the artificial saliva (in a reverse sense when compared to qualitative methods).

Samples of 50 µL of saliva containing the leaked radiotracer were drawn at 0.33, 0.66, one, two, three and four hours, one day, then every day up to one week, every two days up to thirty days, every one week up to six months, and every month up to 16 months. During the 16 months period,

47 readings were taken from the aliquot surrounding each root. Saliva was replenished at each time so that its volume remained at 20 mL throughout the experiment. Each aliquot sample was added to 5 mL of scintillation cocktail*. The amount of radioactivity in the sample was determined as counts per minute (CPM's) using a scintillation counter⁵.

The CPM's in each sample drawn at a time (X) was used to calculate the amount of microleakage from each obturated canal (L_x) at that time. The mean leakage of the 6 obturated canals at time X, (L_x) together with the standard error were then computed. The mean leakage for each experimental group at time X, (L_x) was then divided by the mean leakage of the 12 specimens (unobturated canals) serving as control i.e. L_c. Therefore, the quantity of microleakage that occurred at time X from the obturated canals relative to the quantity of possible leakage from the open canals, i.e., the true or relative microleakage (M_r) was calculated, in terms of percent, as:

$$M_r = (L_x/L_c)100$$

Using this method, microleakage of the obturated canals was determined in real time (while it is occurring) and was computed relative to the maximum leakage that can occur from unobturated canals (true).

To delineate influence of the canal dentin in chemisorping and retaining the radioactivity, the mean quantity of microleakage that occurred at time X from the restored cavity (L_w) as CPM's of radioactivity, relative to the number of radioactive units placed originally in the canal (N), i.e. the apparent or absolute microleakage (M_a) was also calculated in terms of percent as :

$$M_a = (L_w/N)100$$

* Obtura
Tulsa Dental Products, Tulsa, OK
Radiochemical Centre, Amersham, England
**Quickfit, Germany
**VA-Oralube formula
^GFL 1083-Germany

*Opti Phase MP, LKB Scintillation Products, England, made for LKB
Wallace, Finland
⁵1211 Rackbeta and 1215 Rackbeta, LKB Wallace, Finland

The number of aliquot samples taken during this study was 2016 (36 Flasks x 56 samples).

Analyses to determine whether the change in microleakage by time (the rate) could be presented as a linear relationship and whether a microleakage constant could be computed for each material were made. The method of analysis consisted of inputting the relative (true) microleakage data for each of the four obturation methods and the corresponding times up to 463 days in a computer. Relative microleakage as leak %, log leak %, radioactivity remaining % in the canal, and log remaining % radioactivity were computed to represent the microleakage data set. The duration as days, log days, and square days, were computed to represent the time data set. Using correlation analysis, the Pearson correlation coefficients between members of the microleakage data set and those of the time data set were calculated. The coefficient values indicated the best correlation between one member from each of the two data sets.

Once the best correlation was determined, the General Linear Model (GLM) for the relationship between the two members was computed in accordance with the relationship:

$$M_t = K t^n$$

where M_t is the relative microleakage %, K is the rate constant, n is the slope and t is time in days. The equation is an adaptation of that presented by Korsmeyer et al. (1983) in analyzing data of water soluble drug diffusion through polymeric structures for comparative purposes.

An actual plot relating the two members with best correlation is then made. Once the linear plots were obtained the rate constants for each obturation method up to 463 days was determined.

Results

Mean quantities of relative (true) microleakage expressed as % at various times is shown in Fig. 1. While the 95% confidence interval for all sample means was computed, it was not reported in the figure for clarity. The line representing leakage for the control group is also omitted from the figure but one mean value at 463 days is shown. At 463 days, the control (unobtured) canals leaked 94% of the radioactivity placed. None of the obturated canals leaked that much up to 463 days.

While there was no statistically significant differences between the vertical, lateral and obtura techniques with mean values of 26, 25 & 31% at 463 days, Thermafil showed a significantly higher value of 41% (student t-test, $P < 0.05$).

When the microleakage values were considered progressively in terms of time, the figure shows that at one day, there was no difference among the four methods. Beyond one day, microleakage of the vertically condensed canals was higher than that for the three other methods which showed no significant differences among them up to 50 days. From 50 days and up to 99 days, microleakage with both vertical and lateral condensation were essentially equal and significantly higher than those for the Thermafil and Obtura techniques which were essentially equal.

Between 99 and 127 days, Thermafil continued to show less microleakage even than Obtura. While Obtura and Thermafil showed lower microleakage means than the vertical and lateral condensation methods between 127 and 162 days, the difference was not statistically significant. From 162 days and up to 463 days, microleakage with Thermafil was significantly higher than the other three obturation techniques.

The absolute microleakage values expressed as per cent of the CPM's of radioactivity placed originally in the canals reflected the same trends shown in Fig. 1 except that all the mean microleakage values were lower.

The results of computing correlation coefficients between members of the microleakage data set and those of the time data set up to 463 days are shown in Table 1. Positive correlation coefficient values for microleakage associated with the four obturation techniques ranged from 0.728 to 0.990.

Among all correlations, the % leakage vs days were selected since their values as a group were among the highest. Using equation (1) and these correlation values, the predicted (best fit) leak % was plotted against time up to 463 days for each of the four obturation techniques as shown in Fig. 2.

The computed intercepts (K) and microleakage rate constants (n) for the four obturation methods up to 463 days were also computed and are shown in Table 2.

The intercept (K) of the best fit curve up to 463 days was lowest (0.219) for Thermafil and highest (4.862) for vertical condensation.

The rate (n) of microleakage up to 463 days was highest (0.101) for Thermafil and lowest (0.055) for lateral condensa-

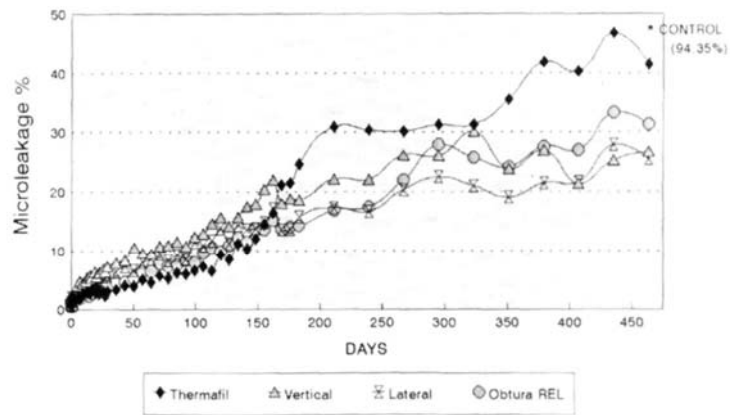


Figure 1: Relative microleakage as a function of time.

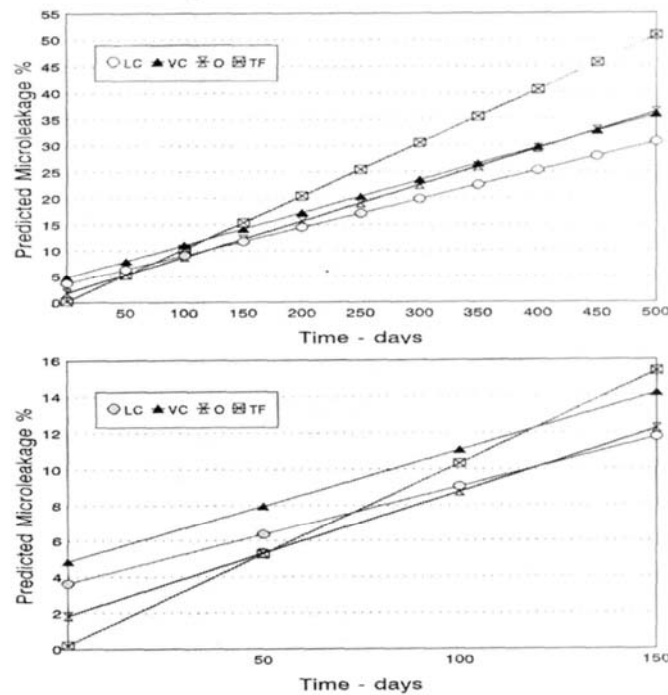


Figure 2: Rate of change of microleakage as a function of time. Intercept is microleakage onset (TF: Thermafil; VC: Vertical Condensation; O: Obtura; LC : Lateral Condensation). Bottom figure is expanded scale up to 150 days.

tion.

Discussion

Gathering data could have theoretically continued until X time = ∞. However, since the radioactivity in the open canals was essentially completely released by the end of 16 months, data gathering was stopped at that time.

Table 1. Correlation analysis "Pearson correlation coefficients" up to 463 days.

Obturation Technique	Coefficient					
	% Leak vs Days	% Leak vs Log Days	% Leak vs Square Days	Log % Leak vs Days	Log % Leak vs Log Days	Log % Leak vs Square Days
Thermafil	0.976	0.648	0.916	0.903	0.874	0.973
Vertical Condensation	0.922	0.838	0.977	0.736	0.965	0.890
Lateral Condensation	0.958	0.815	0.986	0.771	0.933	0.919
Obtura	0.990	0.728	0.963	0.854	0.920	0.961

Table 2: Intercepts (K) and rate constants (n) for the four obturation methods.

Obturation Technique	K	n
Thermafil	0.219	0.101
Vertical Condensation	4.862	0.062
Lateral Condensation	3.658	0.055
Obtura	1.860	0.069

The controls (unobtured canals) leaked 94% of the radioactivity only after 463 days of leakage. This suggests that the radioactivity was adsorbed on the canal dentin and was released slowly. Hence, the radioactivity placed originally in the root canal was not used as a base for calculating microleakage.⁴ Rather, the mean radioactivity released from the open canals at each given time was used as a base to compute microleakage from the obturated canals at that same time.

After 463 days, none of the four obturation methods allowed microleakage greater than 41% suggesting that the four obturation methods are of value in decreasing microleakage.

While there was no statistically significant difference between vertical, lateral and Obtura techniques, Thermafil showed significantly higher (41%) microleakage at 463 days and in agreement with prior studies suggesting that the technique is not better than vertical condensation in barring microleakage.

Vertical condensation showed the highest microleakage up to 50 days which is in agreement with the general convention that the technique is least effective in preventing microleakage. At that time, the three other techniques showed lower and essentially equal microleakage values in agreement with prior reports.

By 99 days both vertical and lateral condensation leaked equally and as reported previously.^{8,9} At that same time Thermafil and Obtura were superior to vertical and lateral condensation in preventing microleakage and in agreement with prior reports.¹⁶ Apparently, the microleakage that occurs with any of the obturation methods is a function of the time at which the values were determined. Between 127 and 163 days the microleakage associated with any of the four methods was essentially the same. Again this will explain the literature

reporting no difference in microleakage among the various techniques. It is only beyond 163 days and up to 463 days that Thermafil showed the greatest microleakage among the four methods.

Up to this point, this study agrees, at various time durations, with all reported microleakage data comparing various obturation methods^{6,18} despite the disparity of such data.

Nonetheless, reporting data in a narrative manner as above could be confusing and does not lead to concrete conclusions about the long time performance of the obturation. Hence, since the data is quantitative, plotting it in a linear form was attempted. The success of such an attempt is depicted in Figure 2 and Table 2.

Should one assume that the microleakage that occurred at one day (log time = zero) is the microleakage onset value (K) then, it is quite clear from Table 2 and Figure 2 that Thermafil was associated with the least onset of microleakage in the early stages after obturation (up to 50 days). The microleakage onset (K) values for the other three obturation methods in an increasing order were Obtura, lateral condensation and vertical condensation. This efficacy of obturation of Thermafil is true only in the early days following obturation. The change in the amount of microleakage by time (rate = n) is the slope of the best-fit line shown in Figure 2 for each of the methods which value is reported in Table 2. The rate of microleakage with Thermafil was highest and that for lateral condensation was lowest while Obtura and vertical condensation showed moderate values. Yet, the slopes (n) of Obtura and vertical condensation are much closer in value to that of lateral condensation than they are to Thermafil.

The above linear analysis suggests that an objective and quantitative method for

computing microleakage for long time durations and in real time is capable of resolving the disagreements reported in the microleakage literature. Using the reference controlled reverse diffusion method to determine microleakage associated with four obturation methods in real time showed that while Thermafil resulted in the lowest microleakage onset in the early days, it permitted the largest microleakage over extended time periods. The lateral condensation obturation method showed stronger microleakage onset in the early days than Thermafil, yet permitted the least microleakage over extended periods of time.¹ While the vertical condensation method showed the strongest microleakage onset, it should not be discounted since it maintained a lower rate of microleakage than Thermafil and one nearly equals to that of Obtura over a much longer period of time.

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