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Correlation between Apical Microleakage and Penetration of Various Root Canal Sealers: An In Vitro Study

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ABSTRACT

Introduction: This study aimed comparatively evaluate the relationship between penetration in simulated lateral canals and apical microleakage of AH Plus Jet, TotalFill BC and GuttaFlow Bioseal root canal sealers.

Materials and methods: Thirty single-rooted extracted human mandibular premolars were decoronated and the root canal length was standardized at15mm, simulated lateral canals were made. After chemo-mechanical preparation, the specimens were randomized into three experimental groups according to the root canal sealers: TotalFill BC, AH Plus Jet, and GuttaFlow Bioseal root canal sealers. In all groups, root canal filling was performed with a single cone technique. The specimens were immersed in 2% methylene blue dye solution and then, subjected to the clearing technique. A stereomicroscope was used to observe the specimens. The penetration depth of root canal sealers in simulated lateral canals and apical microleakage were assessed by a four-grade scoring system (0.3). Data were statistically analyzed by Spearman correlation test at 5% significance.

Results: Statistically, there was no significant correlation between penetration depth and apical microleakage of AH Plus Jet sealer (P>0.05). Instead, there was a significant correlation between penetration depth and apical microleakage of TotalFill BC sealer at apical simulated lateral canals whereas GuttaFlow Bioseal sealer showed a significant correlation at coronal simulated lateral canals (P<0.05).

Conclusions: GuttaFlow Bioseal and TotalFill BC sealers showed an association between their penetration depth and apical microleakage whereas AH Plus Jet sealer showed no association between its penetration depth and apical microleakage.

Key words: Root canal sealers, Clearing technique, Apical microleakage technique, Penetration depth

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INTRODUCTION

Root canal filling is clinically a challenge to be succeeded since it is the final operative phase of root canal treatment which completes the clinical procedures of accurate diagnosis, and chemo-mechanical preparation [1. Moreover, Gutmann et al. [2] stated that along with the removal of debris and microorganism, good adaptation of filling material to the dentinal wall is essential to achieve hermetic sealing of root canal filling through the penetration of the filling material into the dentinal tubules thus, complete cleaning and sealing of the anatomical ramifications aid in achieving tridimensional sealing of the entire root canal cavity, and consequently root canal filling success [3]. Also, it had been suggested by Ingle et al. [4] in the so-called "Washington study" that apically infected periapical exudate into the incompletely filled root canal space represented about 60% of root canal treatment failure therefore, tridimensional sealing of root canal system is of utmost important factor influencing root canal treatment success.

Root canal sealers should have good sealing ability in static and dynamic conditions since; sealing ability reduces any space that permits penetration of oral fluid between the gutta-percha, the dentin wall, and resisting filling dislodgment following treatment [5]. The sealing ability of the root canal sealer is frequently assessed by microleakage, and bond strength tests [6]. Hence, the good sealing ability of root canal sealer is related to improved adhesion and reduced microleakage values [7].

Theoretically, root canal sealer penetration into dentinal tubules could improve the sealing ability of a root canal filling due to micro-mechanical interlocking [8]. Subsequently, the concept of the root canal sealer penetrating dentinal tubules as a requirement to improve the sealing ability of root canal fillings has been created [9]. Hence, this in vitro study aimed to estimate the association between penetration depth and apical microleakage among experimental root canal sealers in a single experimental group.

MATERIALS AND METHODS

A total of 30 human single-rooted mandibular premolars were used as study specimens, the teeth were kept in a 0.1% thymol solution at room temperature till the time of the experiment [10].

A preoperative periapical radiograph (Carestream, USA) was taken for teeth in bucco-lingual directions and mesio-distal direction to ensure the presence of a single straight root canal and exclude teeth with prior root canal treatment, calcification, internal or external resorption [11].

The teeth were horizontally decoronated at/below the cemento–enamel junction (CEJ) by a high speed fissure bur under continuous water cooling [12] to obtain a standardized root canal length of 15 mm [13].

Simulated lateral canals preparation

The simulated lateral canals were made by parallel milling machine (bio-art 1000MAX, Brazil), measured and prepared at 3 mm (apical third), 6 mm (middle third), and 10 mm (coronal third) from the root apex by a 0.3 mm active end cylinder-shaped bur [14]. The connection between the outer root surface and the main root canal by simulated lateral canals was identified by a 21mm, size 08 K-file (Dentsply Maillefer, Switzerland) and a periapical radiograph was taken if the K-file did not enter the main root canal, and the specimen was excluded from the experimental procedure [15].

Preparation of specimens

The specimens were accessed then, a size 10 K-file was introduced into root canal until it was just visible at the apical foramen, and the length of K-file was measured, working length was established by subtracting 1 mm from this length [16]. The instrumentation procedure had been done by the crown down technique by Protaper Next rotary system nickel-titanium files (Dentsply Maillefer ,Switzerland) in sequential order from (X1-X2) [11]. The irrigation procedure had been done using 2 ml of 5% sodium hypochlorite (NaOCl) (CHLORAXID, ul. Kwiatkowskiego) was used as an irrigant, and 17% Ethylene diamine tetra acetic acid gel (EDTA) (Dentsply Maillefer, Switzerland) was used as a lubricant during the instrumentation procedure [17].

After end of the instrumentation, all specimens were washed with 5ml of 5% NaOCl solution for 1 min subsequently 5ml of distilled water, 5ml of 17% EDTA solution for 1 min, and finally with 5ml of distilled water [18].

Specimens grouping and root canal filling

The specimens were randomly allocated into three experimental groups (N=10) according to the type of the root canal sealers: AH Plus Jet, TotalFill BC, and Gutta Flow Bioseal sealers. All experimental root canal sealers were handled along with manufacturer's instructions and root canal filling was completed by a single cone filling technique.

Apical dye leakage technique

A coat of nail polish (FloDerm, P.R.C) was carried out on the external surface of specimens except the apical part about 1mm free from nail paint then, after one hour another coat was applied upon the coat of nail paint had completely dried, the specimens were immersed in a 2% methylene blue dye solution for 48 hours [19].

Clearing technique

This technique was accomplished by following the phases: First phase was decalcification process by immersing the specimens into 5% nitric acid for 4 days that had been changed every day, shaken three times in a day and on the 4th day the specimens were tested by trying to thrust a thin needle through the coronal third. If the needle went easily through, the specimens were soft and ready for the next phase [17]. The next phase was dehydration process by immersing the specimens in 70% ethyl alcohol solution for 12 hours followed by 80% ethyl alcohol solution for 12 hours, 90% ethyl alcohol solution for 6 hours and finally in 100% ethyl alcohol solution. The final phase was transparency by immersing the specimens in 100% methyl salicylate for 2 hours till the specimens made transparent [20].

Stereomicroscopic evaluation

All the specimens were examined under a stereomicroscope (OPTIKA, Italy) at 10X magnification, linear apical leakage from the apex of the root to the most coronal extent of dye penetration was evaluated [21] and measured in millimetres on digital images of specimens that had been captured by an attached camera (OptikamB5, Italy) on a stereomicroscope [22].

Apical microleakage values were scored as: Score 0: If no leakage. Score 1: If leakage less than or equal 0.5 mm. Score 2: If leakage from 0.51mm to less than 1 mm .Score 3: If leakage more than 1 mm [17].

The penetration of root canal sealer in simulated lateral canals was examined under a stereomicroscope (OPTIKA, Italy) at 10X magnification and evaluated on digital images of specimens that had been captured by an attached camera (OptikamB5, Italy) on a stereomicroscope [23].

The depth of penetration into the simulated lateral canals thirds (apical, middle and coronal) was scored as: Score 0: No sealer penetration. Score1: If sealer was present in less than half of the lateral canal. Score 2: If sealer covered more than half of the lateral canal. Score 3: Lateral canal was completely filled with sealer [13].

Statistical analysis

Data were analyzed by the Statistical Package for Social Sciences (SPSS, version 25). Spearman correlation coefficient was estimated to measure the strength of correlation between the penetration depth scores and the apical microleakage scores at a 5% significance level ($p < .05$).

RESULTS

All experimental root canal sealers revealed the ability to penetrate simulated lateral canals, the measured mean of depth of penetration scores of root canal sealers in the apical, middle and coronal thirds simulated lateral canal, number of specimens(N), and percentage(%) were assessed (Table 1 and Figure 1).

The highest mean apical microleakage was observed on AH Plus Jet sealer (1.290) mm whereas the least mean apical micro leakage was observed on GuttaFlow Bioseal sealer (0.670)mm as shown in Figure 2.

All experimental root canal sealers showed no apical microleakage in various distributions. The highest distribution of score 0 in which no leakage was observed on GuttaFlow Bioseal sealer which was 30.0% whereas TotalFill BC sealer, and AH Plus Jet showed 20.0% as shown in Table 2.

Spearman correlation test indicated no significant correlation between the penetration depth scores and the apical microleakage scores of AH Plus Jet sealer. Furthermore, there was a significant positive Spearman correlation in TotalFill BC sealer between penetration depth scores and the apical microleakage scores in the apical third. Regarding the coronal third, there was a significant positive Spearman correlation in GuttaFlow Bioseal sealer between penetration depth scores and the apical microleakage scores (Table 3).

Table 1: The penetration depth scoring distribution of experimental root canal sealers.

	Bio		AH		GF	
Penetration score	**N**	**(%)**	**N**	**(%)**	**N**	**(%)**
Apical						
0	5	(50.0)	5	(50.0)	4	(40.0)
1	0	0	1	(10.0)	0	0
2	0	0	3	(30.0)	3	(30.0)
3	5	(50.0)	1	(10.0)	3	(30.0)
Middle						
0	4	(40.0)	3	(30.0)	4	(40.0)
1	2	(20.0)	0	0	1	(10.0)
2	2	(20.0)	4	(30.0)	2	(20.0)
3	2	(20.0)	3	(30.0)	3	(30.0)
Coronal						
0	3	(30.0)	2	(20.0)	2	(20.0)
1	1	(10.0)	3	(30.0)	2	(20.0)
2	3	(30.0)	2	(20.0)	4	(40.0)
3	3	(30.0)	3	(30.0)	2	(20.0)

Table 2: Apical microleakage scoring distribution of experimental root canal sealers.

	Bio		AH		GF	
Leakage score	**N**	**(%)**	**N**	**(%)**	**N**	**(%)**
0	2	(20.0)	2	(20.0)	3	(30.0)
1	1	(10.0)	1	(10.0)	2	(20.0)
2	3	(30.0)	0	0	2	(20.0)
3	4	(40.0)	7	(70)	3	(30.0)
Total	10	(100.0)	10	(100.0)	10	(100.0)

Table 3: Analysis of the Spearman correlation among experimental root canal sealers.

	Leakage Score					
	Bio		AH		GF	
Penetration Score	Spearmen correlation coefficient (rho)	p-value	Spearmen correlation coefficient (rho)	p-value	Spearmen correlation coefficient (rho)	p-value
Apical	0.84	*0.002	-0.162	0.655	-0.238	0.507
Middle	0.06	0.87	-0.222	0.537	-0.338	0.34
Coronal	0.37	0.293	0.386	0.27	0.808	*0.005

* Spearman correlation is significant at the 0.05 level.

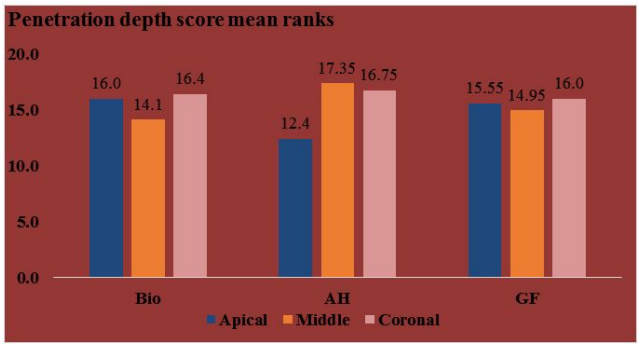

Figure 1: Comparison of the depth of root canal sealers penetration in simulated lateral canal thirds.

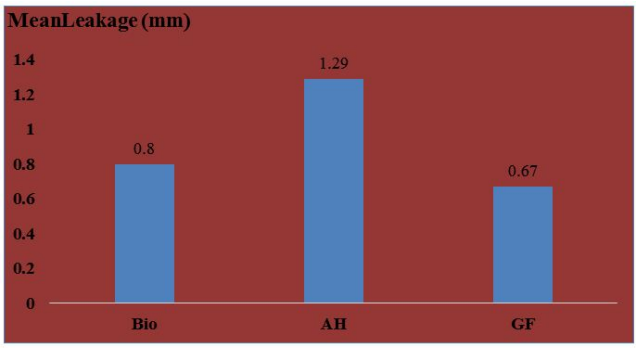

Figure 2: Comparison of mean apical microleakage of root canal sealers.

DISCUSSION

In this study, there is no significant correlation between the penetration depth scores and the leakage scores of AH plus jet sealer indicated that there was no significant monotonically increased association. This could be caused by AH Plus Jet sealer contains epoxide component [24] that has to soften the effect on gutta-percha being as a partial solvent [25], bond with root dentin by adamantine, and good flow ability [26]. In addition to low solubility on complete setting altogether improve the penetration ability into root canal complexities through micro-mechanical interlocking hence, AH Plus Jet sealer only bonds mechanically with the root dentin [27]. On the other hand, fast polymerization reaction and subsequently sealer shrinkage was occurred during an early stage of polymerization reaction resulted in micro gap formation, also this sealer is naturally acidic and hydrophobic while the root dentin is hydrophilic in nature and the dentin-sealer interface is relatively hydrophilic [28]. Hence, adhere poorly to humid root dentin and decrease the ability of complete adaptation and filling of the hydrophilic root canal [29]. Moreover, the sealing ability of AH Plus Jet sealer could be affected by silicone oils resulted in poor wettability and high surface tension [30]. Also, this sealer consists of large-sized particles (1.5-8) µm which could not easily penetrate small dentinal tubules, particularly at the apical area hence, high risk of apical microleakage that enhances dye penetration [31].

There is no significant association in TotalFill BC sealer between the penetration depth scores and the leakage scores except in the apical third of the simulated lateral canal ($p<0.05$) which was $p = 0.002$ whereas the Spearman correlation coefficient was positive (rho = 0.84) indicated that there was a significant monotonically increased association at the apical third of the simulated lateral canal. This could be caused by the ability of the nanoparticles sized less than 2 µm in diameter of this sealer to penetrate apical dentinal tubules to form micro–mechanical interlocking then, hydroxyapatite formation along the mineral infiltration zone hence, this sealer bonds mechanically and chemically [32].

There was no significant correlation in GuttaFlow Bioseal sealer except between the penetration depth scores and the leakage scores in the coronal third of the specimens ($p<0.05$) which was $p = 0.005$ whereas the Spearman correlation coefficient was positive (rho= 0.808) indicated that there was a significant monotonically increased association between the penetration scores and the leakage scores at the coronal third of the simulated lateral canal. This could be caused by the ability of nanoparticles sized less than 30µm of gutta-percha, thixotropic property, and physically, it is comparable to the gutta-percha core material. In addition to the ability of this sealer to expand as much as 0.2% after setting hence, filling the irregular spaces of the root canal wall and into dentinal tubules hence, GuttaFlow Bioseal bonds mechanically, physically and chemically with the root dentin [32].

TotalFill BC and GuttaFlow Bioseal sealers showed a significant monotonically increased association between the penetration depth scores and the leakage scores at the apical, coronal thirds of the simulated lateral canal respectively. Hence, if sealer penetration into simulated

lateral canal had been increased, apical microleakage had not been decreased which could be due to the advantageous biological properties of calcium silicate-based TotalFill BC and GuttaFlow Bioseal sealers primarily resulted from their bioactivity ability [33] which considered a significant property for chemical bonding resulted from water sorption and solubility even after complete setting resulted in sealer mass loss, decrease dimensional stability, and high risk of apical microleakage [28]. Hence, a negative impact on the sealing ability of root canal sealer [34]. Moreover, the bio mineralization ability has not been absolutely confirmed that the mineral infiltration zone influences the outcome of root canal filling treatment [35], either positively, calcium ions will react with the carbon dioxide forming calcite crystals [36]. Hence, decrease micro gaps and porosity as well as increase the retention of the root canal sealer [37] or negatively, hydroxyapatite formation did not decrease microleakage values due to its porous shape [38].

In this study a single experimental group had been standardized, designed and aimed to evaluate the potential correlation between root canal sealer penetration and apical microleakage in which the same specimens had been made two different experiments to examine two independent variables; hence, it could be possible to evaluate the cause and result in correlation thus, increase the power of statistical analysis since typical correlation analysis is assumed that any random variable factor affects only one subject and this requirement could not possible when two or more different experiments [39].

The results of this study are in conformance with De-Deus et al. [39] showed that no significant correlation between tubular penetration and leakage of AH Plus sealer. In contrast, Attur et al. [40] revealed that there was a positive correlation between microleakage and tubular penetration of AH26 sealer.

CONCLUSIONS

The penetration ability of AH Plus jet sealer was not a dependent factor influencing its apical microleakage values as no significant association was accepted between penetration depth scores and microleakage scores of AH Plus jet sealer. The penetration ability of TotalFill BC sealer in apical root thirds was a dependent factor influencing its apical microleakage values as a positive association was accepted between penetration depth scores and microleakage scores. The penetration ability of GuttaFlow Bioseal sealer in coronal root thirds was a dependent factor influencing its apical microleakage values as a positive association was accepted between penetration depth scores and microleakage scores.

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