|| Print ISSN: 2589-7837 || Online ISSN: 2581-3935 ||
International Journal of Medical Science and Diagnosis Research (IJMSDR)
Available Online at www.ijmsdr.com
NLM (National Library of Medicine ID: 101738825)
Original Research Article
Volume 4, Issue 6; June: 2020; Page No. 06-11

A COMPARATIVE STEREOMICROSCOPIC EVALUATION OF BIOACTIVITY OF DIFFERENT BIOMIMETIC ROOT END FILLING MATERIALS - AN IN VITRO STUDY

Bhavika Bhavsar[1], Remya Ramchandran[2], Neetu Kharat[3], Shantwana Singh[4], Sapnil Gaidhankar[5]

[1]Professor & HOD, Dept. of Conservative Dentistry and Endodontics, RKDF Dental College, Bhopal
[2]Private Practitioner
[3]Reader, Dept. of Conservative Dentistry and Endodontics, RKDF Dental College, Bhopal
[4]Sr. Lecturer, Rishiraj Dental College, Dept. of Conservative Dentistry and Endodontics, Bhopal
[5]Sr. Lecturer, Dept. of Periodontology, Nanded Rural Dental College and RC, Maharashtra
Conflicts of Interest: Nil
Corresponding author: *Dr. Bhavika Bhavsar*

Abstract:
The interaction between MTA and phosphate-containing fluids results in bioactivity of MTA. Recently, new bio ceramic materials have been introduced as an alternative to MTA. Neo MTA plus and biodentine.

Aim: Hence the aim of the study is to find out and compare the bioactivity of Neo MTA Plus, Pro root MTA white, Biodentine & Glass ionomer cement as root end filling materials using 1% methylene blue as tracer.

Objectives: To comparatively evaluate 1: The sealing ability of different root end materials;2: The change in sealing ability of different root end filling materials with time; 3: To evaluate seal of bioactive materials and glass ionomer cement.

Methodology: Freshly extracted, permanent, single rooted human maxillary central incisor teeth were collected. Samples were, randomly divided into four groups, consisting of 20 samples each, filling Group I with GIC, Group II with pro root MTA, Group III with neo MTA plus and Group IV with biodentine. The evaluation depth dye penetration was done with the help of Stereomicroscope at 2x magnification at 7days and 1 month period intervals.

Result: All the four materials exhibited penetration.

Conclusion: Neo MTA plus and biodentine should be the preferred choice for root end filling.

Key words- Biomimetic Root End Filling, Neo MTA plus, Biodentin, Pro Root MTA

INTRODUCTION

The conducive nature of a material to a given living organism makes it a favourable material, both in medical and dental field. The more conducive the material to the living tissue, the more favourable the material for the application. There are numerous materials that have been identified as being safe for use within the human body. These materials are termed biomimetics. Bioceramic material is one among them which are widely used. They are biocompatible materials used in repair and replacement of the organs in the musculoskeletal system.[1] Emphasizing the practical application, they can be termed as the ceramic materials, applicable for biomedical and dental use.

From the beginning of dental practice to the present time, many materials have been introduced keeping in mind their reactions towards the oral tissues and to be precise considering their bioactivity. The bioactivity is an important property associated with bioceramics. Bioactivity is a particular behaviour of a material, with its reaction with the body fluids, that results in the formation of a surface hydroxyapatite layer which helps the material in the stable chemical bond with the adjacent living tissue.

The mechanism of action of a bioactive material is that the material elicits a specific biological response at the interface of material resulting in the formation of a bond between tissue and the material. The interaction between a biomimetics material and living tissues, results in the formation of an hydroxyapatite layer, bio mineralization, at the material-tissue interface.

Bioactive ceramics are characterized by a dynamic surface. In essence, this is achieved after the formation of a calcium phosphate layer on the surface of the materials. If this surface layer is present at the time of implantation, the success of the tissue regeneration increases. Furthermore, the ceramics' bioactivity will also influence the activity of proteins and cells within the surrounding environment.

MATERIALS AND METHODOLOGY

Freshly extracted, permanent, single rooted human maxillary central incisor teeth were collected. On the basis of following inclusion and exclusion criteria, 80 teeth were selected.

INCLUSION CRITERIA

Freshly extracted, human permanent maxillary central incisor with completely formed root apices and free of defects were selected for this study.

EXCLUSION CRITERIA

1. Presence of developmental defects.
2. Immature apex.
3. Root fracture.
4. Significant apical curvature.
5. Presence of internal resorption.
6. Presence of caries.
7. Presence of morphological defects.
8. Presence of visible cracks.
9. Teeth with previous restoration or endodontic treatment.

Each specimen was sectioned transversely in the cervical area to separate crown from the root, Working length was established at 1mm short of the apex. Biomechanical preparation was done using hand Pro tapers and obturated with gutta-percha using zinc oxide eugenol sealer. The coronal access cavity was filled with GIC. Each sample was resected apically at 90 degree to the long axis of the root. 3mm deep retrograde cavity was prepared with straight fissure bur. .After retrograde cavity preparation, samples were, randomly divided into four groups, consisting of 20 samples each, filling Group I with GIC, Group II with Pro Root MTA, Group III with Neo MTA plus and Group IV with BIODENTINE. Samples were stored in artificial saliva. At the completion of storage period, the respective samples were dried and coated with three coats of nail varnish, except at the apical 1 mm of the resected root. The specimens were suspended in 1% methylene blue for 24hours. Each sample was sectioned longitudinally. Out of the two sections obtained, one with no surface discrepancies was selected and examined for dye penetration. The evaluation depth dye penetration was done with the help of Stereomicroscope at 2x magnification.

Statistical Analysis: The intergroup comparison between different materials at 7 days and one month was done by using ANOVA test and Post Hoc test. The difference between two groups test was done by using independent t Test. Comparison between sealing ability of a root end filling materials between 7 days and one month was done by using paired t test.

RESULTS

Evaluation of the samples at 7 day period

The observed dye penetration was found to be highest with GIC followed by Pro Root MTA, BIODENTINE, and minimum with Neo MTA plus. (fig :1)

Dye penetration at day 7

The mean depth of penetration in samples filled with GIC (fig:2) was displayed to range between maximum: 1.38mm, least value 0.98mm with the mean depth of penetration 1.18 mm, with 95% confidence interval maximum: 1.27mm,least: 1.08mm.

The depth of penetration in samples filled with Pro Root MTA(fig:3) was displayed to range between maximum: 0.73 mm, least value: 0.26mm, with the average depth of penetration 0.43with 95% confidence interval maximum: 0.55mm least: 0.31mm.

The depth of penetration in samples filled with Neo MTA plus (fig: 4) was displayed to range between maximum: 0.19, least value: 0.10mm, with the average depth of penetration0.16mm with 95% confidence interval maximum: 0.17mm, least: 0.12mm.

The depth of penetration in samples filled with BIODENTINE (fig :5) was displayed to range between maximum: 0.28mm, least value: 0.13 mm, with the average depth of penetration 0.18 with 95% confidence interval maximum: 0.22mm, least: 0.14 mm

Dye penetration at 1 month

The depth of penetration in samples filled with GIC was displayed to range between maximum: 1.46 mm, least value: 1.11 mm with the average depth of penetration 1.32mm with 95% confidence interval maximum: 1.40mm, least: 1.23mm

The depth of penetration in samples filled with Pro Root MTA was displayed to range between maximum: 0.75mm, least value: 0.28m, with the average depth of penetration 0.54 with 95% confidence interval maximum: 0.65mm, least: 0.42mm.

The depth of penetration in samples filled with BIODENTINE was displayed to range between maximum: 0.19mm, least value: 0.13 mm, with the average depth of penetration0.17 mm with 95% confidence interval maximum: 0.19 mm, least: 0.14 mm.

The depth of penetration in samples filled with Neo MTA plus was displayed to range between maximum: 0.19 mm, least value : 0.11 mm, with the average depth of penetration 0.15mm with 95% confidence interval maximum:0.19 mm, least: 0.14 mm

INTER GROUP COMPARISION

Table 1: Comparison of Dye penetration among different root end filling material at 7 days by using Post Hoc Test

(I) VAR00001	(J) VAR00001	Mean Difference (I-J)	Std. Error	Sig.
GIC	ProRootMta	.74900*	.04994	.000
	Neo MTA plus	1.03000*	.04994	.000
	BIODENTINE	.99600*	.04994	.000
ProRootMta	Neo MTA plus	.28100*	.04994	.000
	BIODENTINE	.24700*	.04994	.000
Neo MTA plus	BIODENTINE	-.03400	.04994	.904

The inter group comparison at 7 days interval

The observed dye penetration at 7 days period was found to be highest with GIC followed by pro root MTA, biodentine, and minimum with neo MTA plus.

Table 2: Comparison of Dye penetration among different root end filling material at one month by using Post Hoc Test

(I) VAR00001	(J) VAR00001	Mean Difference (I-J)	Std. Error	Sig.
GIC	ProRootMTA	.78650*	.04628	.000
	Neo MTA plus	1.16600*	.04628	.000
	BIODENTINE	1.15400*	.04628	.000
ProRootMta	Neo MTA plus	.37950*	.04628	.000
	BIODENTINE	.36750*	.04628	.000
Neo MTA plus	BIODENTINE	-.01200	.04628	.994

The Inter group comparison at 1 month interval

The observed dye penetration at one month period, was found to be highest with GIC followed by pro root MTA,BIODENTIN, and minimum with Neo MTA plus.

Table 3: Comparison of Dye penetration among different root end filling material at 7 days and 1 Month

Groups	N	Mean		Std. Deviation		F value		P value	
		7 days	1 month	7 days	1 month	7 days	1 month	7 days	1 month
GIC	10	1.18	1.32	0.13	.12	184.11	279.30	0.01	0.01
ProRoot MTA	10	0.43	.54	0.16	.16				
Neo MTA plus	10	0.16	.15	0.03	.03				
BIODENTINE	10	0.18	.17	0.05	.03				
Total	40	0.48	.54	0.43	.48				

The dye penetration was found to be highest with GIC followed by Pro Root MTA, then biodentine and least with Neo MTA plus. Penetration among root end filling material shows statistically highly significant difference in vertical depth of penetration among sealer. NeoMTA plus shows the best sealing property as compared to BIODENTIN, Pro Root MTA and GIC.

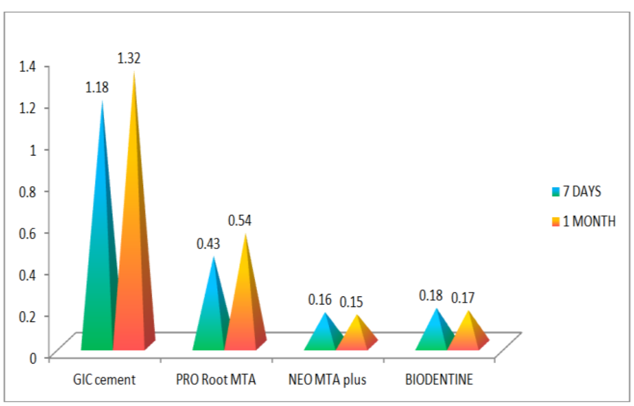

Figure 1: Comparison of dye penetration among different root end filling material at seven days and one month

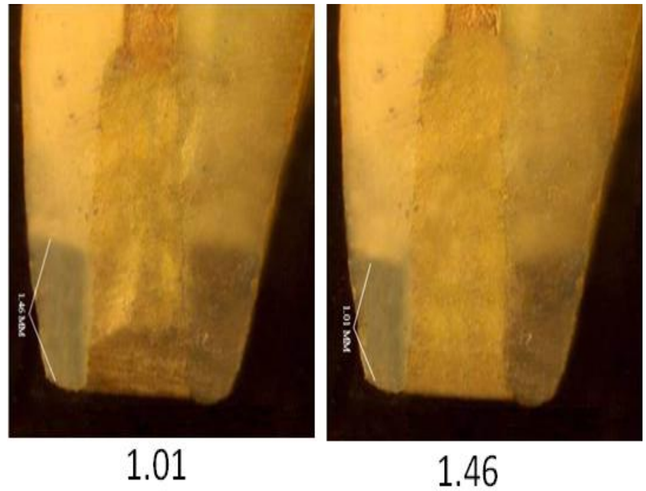

Figure 2: Leakage analysis in GIC group in 7 days and 1 month

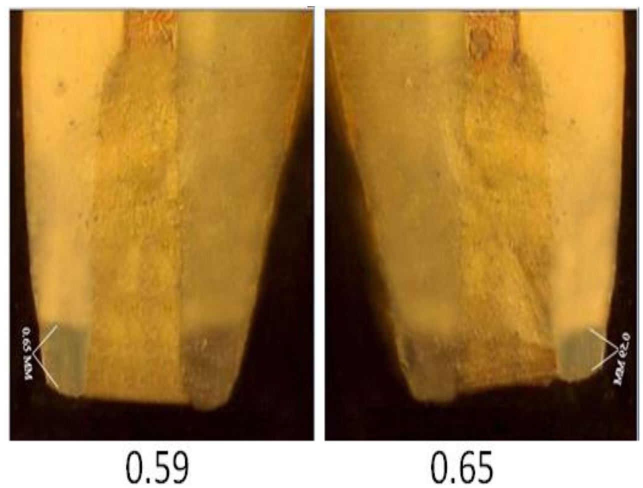

Figure 3: Leakage analysis in Pro-Root- MTA group in 7 days and 1 month period

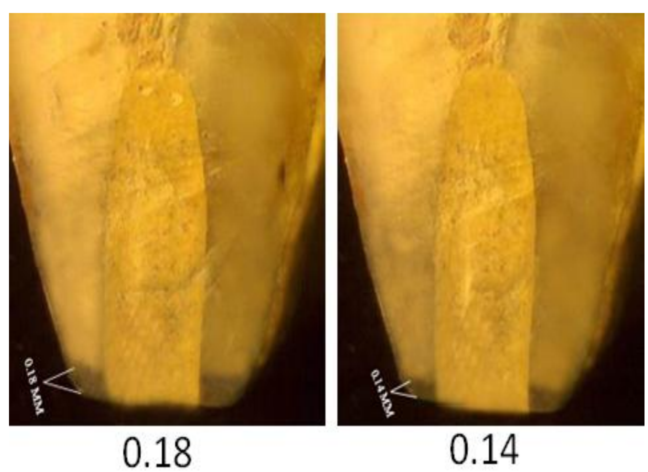

Figure 4: Leakage analysis in Neo MTA plus group in 7 days and 1 month period

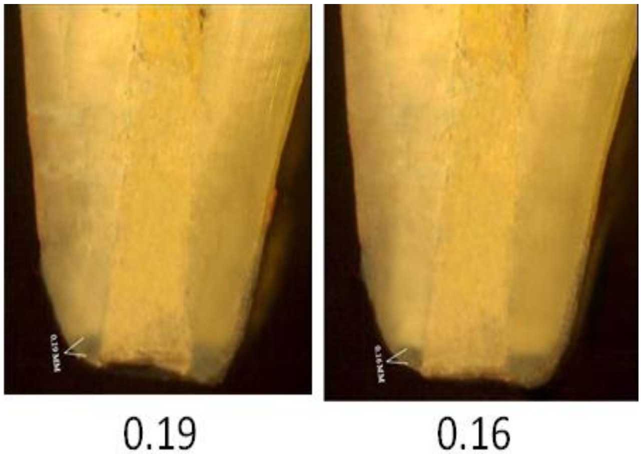

Figure 5: Leakage analysis in BIODENTIN group in 7 days and 1 month period

Discussion:

An ideal root-end filling material should meet the following characteristics- non-resorbable, non-toxic, non-carcinogenic, biocompatible, and dimensionally stable, and provide a hermetic seal. Added to the sealing ability and biocompatible nature, an ideal root end filling material should not be affected by blood or other fluid, should induce bone formation, healing and mineralization.[2] The past two decades have seen the emergence of biomimetics materials as a promising alternative.

It was from the year 1970 that Glass ionomer cement (GIC) was widely used in dentistry. The good mechanical properties and transparency of GIC make them superior material, compared to other types of water based cements. In addition, fluoride release over a prolonged period and good biocompatibility make GIC useful as a dental material. However, GIC has some disadvantages such as relatively high brittleness and moisture sensitivity during the early stage of setting.[3] Another disadvantage of GIC is that the bonding between the GIC and tooth is weak.[4] However according to Kokub, a stronger chemical bond between GIC and teeth could be achieved by rending a GIC with true bioactive properties, i.e. formation of an hydroxyapatite layer.[5]

In the present study, each specimen was sectioned transversely in the cervical area to separate crown from the root, keeping a standard length of the remaining root at 12mm, so as to maintain standardization among samples. Working length was established at 1mm, short of the anatomical apex using 15 K file after ensuring that the file was within the canal.

Each sample was resected apically at 90 degree to the long axis of the root, using cross cut fissure bur, removing 3mm of the apex.

During preparations of root end cavity, good visualization and easy access are main criteria for choosing 0', 30' or 45' resection angles. However, angled root-end resection also opens dentin tubules which can increase the risk of bacterial contamination and microleakage resulting in failure of endodontic surgery. Gilheanyet al in their study stated that the microleakage increased significantly with increased angulations of the resected root-end.[6]

The root ends of the teeth could have variations in the root anatomy, becoming the source of treatment failure. All these considerations make it mandatory to eliminate last apical three millimetres for maximum security.[7] Studies advocate that the depth of penetration should be ideally 3mm.[8] The plane of sectioning is equally important consideration in technique of root resection 90^o angulations have been proved to be most acceptable by research. Hence this angle was chosen in this study.

Dye penetration method the most commonly used technique for microleakage assessment of root-end filling materials was used in our study. Because dyes are relatively easy to be stored, applied and have their penetration assessed quantitatively, no reactive chemicals are used along with no radiation hence the method is safe, the technique is highly feasible and easily reproducible.[9] However, the dye penetration method is said to have certain drawbacks including the smaller molecular size of the dye molecules when compared to the bacteria.

The various advantages of methylene blue compared to other dyes have influenced the researchers to use it as medium. Literature proves that a large number of studies have used methylene blue as a dye[10] and state that it is inexpensive, easy to handle, has a high degree of staining. This dye has some disadvantages such as dissolution during the demineralization and clearing processes; in addition, it is difficult to observe its maximum penetration point in some cases. On the other hand, Barthel *et al.* propounded that the particle size of the dye might not be an important factor in leakage studies.[11]

In the present study, stereomicroscopic evaluation has been used for observation. According to Keerthi et al it has proved to be more accurate and convenient. Hence many of the researchers rely on stereomicroscopic evaluation.[12]

In our study, least dye penetration was observed with Neo MTA plus followed by BIODENTINE Pro Root MTA, and GIC. It denotes that the biocermic materials have better sealing ability compared to GIC. Bioceramic materials possess this better sealing ability because of their biocompatible and bioactive nature. In general, bioceramic sealers use the water inherent in the dentinal tubules for the setting reaction thus beginning the hydration reaction of the material and thereby reducing the setting time. Dentin is believed to contain about 20percent water (by volume)[13] this water is responsible for the setting of the material.

The results of the present study show that both Neo MTA plus and BIODENTINE have almost similar figures in the dye penetration after 7 days and one month with a statistically insignificant difference for Neo MTA. It could be attributed to similarities in their components, their setting reactions and bioactive properties.

Neo MTA plus is tricalcium silicate-based materials with tantalum as opacifier. Dammaschke et al found that Neo MTA plus has higher amount (0.2 ± 0.02) of sulfur and the presence of gypsum.[14] Li et al concluded that during setting reaction, gypsum plays an important role to harden the material as yielding ettringite.[15]

BIODENTINE is a two components material. The powder is mainly composed of tricalcium silicates. It also contains dicalcium silicate as a second core material and calcium carbonate and oxide as filler. The powder contains zirconium oxide as a radio-opacifier. The liquid contains calcium chloride as a setting accelerator and a water reducing agent. The presence of a setting accelerator allows the material setting in 12 min and the presence of a water reducing agent avoids the formation of cracks within the material.

Khan and Koubi et al in their studies found that the microleakage of BIODENTINE is comparatively less. The reasons they assigned for this finding were shorter setting time (12 min), hydrophilic nature and mild expansion on setting.

Chng H et al in their study regarding the setting time of Pro Root MTA found it to be 40 minutes and the final setting time to be 140-170 minutes.[16]

In the present study, the microleakage in Pro Root MTA, although not increased significantly from one week to one month i.e. from 0.43 mm to 0.54 mm, it remains at higher value of leakage on both.

Even though Pro Root MTA appears to be the preferred material with many positive features, the cement does have several drawbacks. Karabucak B et al stated that compared to other root end filling materials, the handling characteristics of Pro Root MTA is difficult that can eventually cause discrepancies in the procedure.[17]

In our study, the results show that the filling with BIODENTINE does not show significant difference in mean microleakage at one week and one month. The microleakage in biodentine remains almost similar (decreased slightly) from one week to one month. Soundappan et al found that BIODENTINE has the added advantage of better handling and mechanical properties and biocompatibility and ability to induce odontoblast differentiation mineralization.[18] Ravichandra et al in their in vitro study compared the marginal adaptation of BIODENTINE and other commonly used root end filling materials and found that BIODENTINE had a better marginal adaptation when compared with the commonly used root-end filling materials.[19]

Han et al compared BIODENTINE and white Pro Root MTA in terms of Ca and Si uptake by adjacent root canal dentine and observed that both materials formed tag-like structures. They observed that dentine element uptake was more prominent for BIODENTINE than Pro Root MTA.[20]

Although BIODENTINE has the advantages of smaller particle size, faster setting time and bioactive properties that make them better root end filling material, some of the studies have shown their demerits too. Shetty et al in their comparative study of various root end filling materials concluded that BIODENTINE showed significant microleakage.[21]

The result of the present study shows that the filling with Neo MTA plus does not produce significant difference in mean microleakage at one week and one month. Kubo et al in their study found that hydration of MTA powder by moisture can result in an increase in the compressive strength and decrease leakage.[22] According to Sarkar et al MTA has the ability to precipitate hydroxyapatite crystals in the presence of fluid which may be relevant in minimizing leakage.[23]

As regard the similarity of leakage in MTA and BIODENTINE, they noted that BIODENTINE is actually formulated using the MTA-based cement technology and the improvement of some properties of these types of cements, such as physical qualities and handling. Since the basic components of BIODENTINE are similar to MTA, these materials are expected to have similar properties and effects. In their study, Ozbayet al confirmed this reason for the similarity and concluded that MTA has least leakage compared to biodentine.[24]

Various studies shows that the minor variation in the lower microleakage values of MTA may be attributed to its superior marginal sealing ability resulting from its hydrophilic properties and formations of an interfacial layer between the material and dentin. The interfacial layer reduces the risk of marginal percolation and gives promising long-term clinical success.[25]

The result of the present study show that dye penetration was least in Neo MTA plus followed by BIODENTINE, Pro Root MTA and GIC respectively. As mentioned in the discussion above, this study is in confirmation with the result of majority of the studies showing better results for both Neo MTA plus and BIODENTINE. The similarity in the results can be attributed to the similarities in the chemical and component combination of both the materials.

Pro Root MTA, although possesses some of the characteristics of a bioceramic material, falls very short of the other two materials with regard to the dye penetration. According to the various studies, discussed above, although similar or smaller in the particle size, the material's longest setting time and its difficult handling properties could have caused this demerit.

In this regard, the shortest setting time of both Neo MTA plus and BIODENTINE make them ideal root end filling materials. An interesting observation in the result of the present study is that there is a slight decrease in the dye penetration of samples filled with Neo MTA plus and BIODENTINE after one month, showing the clinical advantage of these materials in a long term. It could be attributed to the bioactive nature of these materials that evoke a biological response within the surface.

Conclusion:

Neo MTA plus has higher sealing followed by BIODENTINE, Pro Root MTA and glass ionomer cement. All the three bioactive materials, used in this study, Pro Root MTA, BIODENTINE and Neo MTA plus have better sealing ability than GIC. Neo MTA plus, BIODENTINE displayed enhanced sealing with passage of time, whereas Pro Root MTA and glass ionomer display decreased sealing ability. The result of this study suggest that Neo MTA plus and BIODENTINE should be the preferred choice for root end filling.

References

1. Wei G, Ma P X. Nanostructured biomaterials for regeneration. Adv Funct Mater 2008; 18 (22): 3566–3582.
2. Torabinejad M, Pitt Ford T R. Root end filling materials: A Review. Endod Dent Traumatol 1996; 12 (4): 161-178.
3. Moshaverinia A, Roohpour N, Rehman I U. Synthesis and characterization of a novel fast-set proline-derivative- containing glass ionomer cement with enhanced mechanical properties.ActaBiomaterialia 2008; 5 (1): 498-507.
4. Kamitakahara M, Kawashita M, Kokubo T, Nakamura T. Effect of polyacrylic acid on the apatite formation of a bioactive ceramic in a simulated body fluid: Fundamental examination of the possibility for obtaining bioactive glass ionomer cements for orthopaedic use. Biomaterials 2001; 22 (23): 3191-3196.
5. Kokub T. Bioactive glass ceramics properties and applications.Biomaterials 1990; 12:155-163.
6. Gilheany P A, Figdor D, Tyas M J. Apical dentin permeability and microleakage associated with root end resection and retrograde filling. J Endod 2012; 20 (1): 22-26.
7. Gagliani M, Taschieri S, Molinari R. Ultrasonic root end preparation: Influence of cutting angle on apical seal. J Endod 1998; 24 (11): 726-730.
8. Min M M, Brown C E, Legan J J, Kafrawy A H. Invitro evaluation of effects of ultrasonic root end preparation on resected root surfaces. J Endod 1997; 23(10): 624-628.
9. Subramaniam P, Pandey A. Assessment of microleakage of a composite resin restoration in primary teeth following class III cavity preparation using Er, Cr:YSGG laser: An invitro study. J Lasers Med Sci 2016; 7 (3): 172-176.
10. Yavari H, Samiei M, Eskandarienezhad M, Shahi S, Aghazadeh M, Pasvey Y. An in vitro comparison of coronal microleakage of three orifice barriers filling materials.IranEndodJ2012;7 (3): 156–160.
11. Barthel C R, Moshonov J, Shuping G, Orstavik D. Bacterial leakage versus dye leakage in obturated root canals.IntEndod J 1999; 32 (5): 370–375.
12. Narayan V K, Varsha V K, Girish H C, Murgod S. Stereomicroscopic study on unsectioned extracted teeth. JFDS 2017; 9 (3): 157-164.
13. Koch K A, Brave D G, Nasseh A A.Bioceramic technology: Closing the endo-restorative circle, Part I, Dent. Today 2010; 29 (2): 100–105.
14. Dammaschke T, Gerth H U, Zuchner H, Schafer E. Chemical and physical surface and bulk material characterization of white pro root MTA and two portland cements. Dent Mater 2005; 21 (8): 731–738.
15. Li Q, Coleman N J.The hydration chemistry of pro root MTA. Dent Mater J 2015; 34 (4): 458–465.
16. Chng H K, Islam I, Yap A U, Tong Y W, Koh E T. Properties of a new root-end filling material. J Endod 2005;31(9): 665-668.
17. Karabucak B, Li D, Lim J, Iqbal M. Vital pulp therapy with mineral trioxide aggregate. Dent Traumatol 2005; 21 (4): 240–243.
18. Soundappan S, Sundaramurthy J L, Raghu S, Natanasabapathy V. Biodentine versus mineral trioxide aggregate versus intermediate restorative material for retrograde root end filling: An in vitro study. J Dent Tehran Univ Med Sciences 2014;11(2):143–149.
19. Ravichandra P V, Vemisetty H, Deepthi K, Reddy J, Ramkiran D, Negendrakrishna M J, Malathi G. Comparative evaluation of marginal adaptation of biodentineand other commonly used root end filling materials - An in vitro study. J ClinDiagn Res 2014; 8 (3): 243-245.
20. Han L, Okiji T. Uptake of calcium and silicon released from calcium silicate-based endodontic materials into root canal dentine.IntEndod J 2011; 44 (12): 1081–1087.
21. Shetty S, Hiremath G, Yeli M. A comparative evaluation of sealing ability of four root end filling materials using fluid filtration method: An in vitro study. J Conserv Dent 2017; 20 (5): 307-310.
22. Kubo C H, Gomes A P, Mancini M N.Invitroevaluation of apical sealing in root apex treated with demineralization agents and retrofilled with mineral trioxide aggregate through marginal dye leakage. Braz Dent J 2005; 16 (3): 187-191.
23. Sarkar N K, Caicedo R, Ritwik P, Moiseyeya R, Kawashima I. Physicochemical basis of the biologic properties of mineral trioxide aggregate.J Endod 2005; 31 (2): 97-100.
24. Ozbay G, Kitiki B, Peker S, Kargul B. Apical sealing ability of a novel material: Analysis by fluid filtration technique. ActaStomatol Croat 2014;48 (2): 132-139.
25. Yatsushiro J D, Baumgartner J C, Tinkle J S. Longitudinal study of the microleakage of two root-end filling materials using a fluid conductive system. J Endod 1998; 24 (11): 716-719.