

**“COMPARATIVE EVALUATION OF INTRA
ORIFICE SEALING ABILITY OF NEO MTA PLUS,
CALCIUM ENRICHED MIXTURE AND
CONVENTIONAL GLASS IONOMER CEMENT
UNDER CONFOCAL LASER SCANNING
MICROSCOPE:-AN IN-VITRO STUDY”**

By

Dr. FELBIN TOMY

REG.NO. IE0218004

Dissertation

Submitted to

KLE Academy of Higher Education and Research

In partial fulfillment

of the requirements for the degree of

MASTER OF DENTAL SURGERY

In

**CONSERVATIVE DENTISTRY AND
ENDODONTICS**

(BRANCH – IV)

Under the Guidance of

Dr. SUNITA SHIVANAND M.D.S

**DEPARTMENT OF CONSERVATIVE DENTISTRY AND
ENDODONTICS**

**KAHER VK INSTITUTE OF DENTAL SCIENCES,
BELAGAVI, KARNATAKA**

2018-2021

**KLE ACADEMY OF HIGHER EDUCATION & RESEARCH
BELAGAVI, KARNATAKA**

DECLARATION BY THE CANDIDATE

*I hereby declare that this dissertation/thesis entitled “**COMPARATIVE EVALUATION OF INTRA ORIFICE SEALING ABILITY OF NEO MTA PLUS, CALCIUM ENRICHED MIXTURE AND CONVENTIONAL GLASS IONOMER CEMENT UNDER CONFOCAL LASER SCANNING MICROSCOPE:- AN IN-VITRO STUDY**” is a bonafide and genuine research work carried out by me under the guidance of **Dr. SUNITA SHIVANAND M.D.S** Reader, Department of Conservative Dentistry and Endodontics, **KAHER Vishwanath Katti Institute of Dental Sciences, Belagavi.***

Date:

Dr. FELBIN TOMY

Place: Belagavi

REG.NO. IE0218004

BELGAUM

**KLE ACADEMY OF HIGHER EDUCATION & RESEARCH
BELAGAVI, KARNATAKA**

CERTIFICATE BY THE GUIDE

*This is to certify that the dissertation entitled “COMPARATIVE EVALUATION OF INTRA ORIFICE SEALING ABILITY OF NEO MTA PLUS, CALCIUM ENRICHED MIXTURE AND CONVENTIONAL GLASS IONOMER CEMENT UNDER CONFOCAL LASER SCANNING MICROSCOPE:- AN IN-VITRO STUDY” is a bonafide research work done by **Dr. FELBIN TOMY**, post-graduate student in partial fulfillment of the requirement for the degree of **MASTER OF DENTAL SURGERY** in Conservative Dentistry and Endodontics.*

Date :

Place : Belagavi

Dr. SUNITA SHIVANAND M.D.S

Reader

Department of Conservative Dentistry
and Endodontics,

KAHER V.K. Institute of Dental

Sciences, Belagavi.

BELGAUM

**KLE ACADEMY OF HIGHER EDUCATION & RESEARCH
BELAGAVI, KARNATAKA**

**ENDORSEMENT BY THE HEAD OF DEPARTMENT,
PRINCIPAL OF THE INSTITUTION**

*This is to certify that the dissertation Entitled “COMPARATIVE EVALUATION OF INTRA ORIFICE SEALING ABILITY OF NEO MTA PLUS, CALCIUM ENRICHED MIXTURE AND CONVENTIONAL GLASS IONOMER CEMENT UNDER CONFOCAL LASER SCANNING MICROSCOPE:- AN IN-VITRO STUDY” is a bonafide research work done by **Dr. FELBIN TOMY**, post graduate student under the guidance of **Dr. SUNITA SHIVANAND M.D.S** Reader, Department of Conservative Dentistry and Endodontics, KAHER Vishwanath Katti Institute of Dental Sciences, Belagavi.*

Head of Department

Dr. (Mrs.) Preeti Doddwad M.D.S
Professor & Head,
Department of Conservative Dentistry and
Endodontics,
KAHER V.K. Institute of Dental Sciences,
Belagavi.

Date:
Place: Belagavi

Principal

Dr. (Mrs.) Alka Kale MDS, PhD.
Principal,
KAHER V. K. Institute of Dental
Sciences,
Belagavi

Date:
Place: Belagavi

**KLE ACADEMY OF HIGHER EDUCATION & RESEARCH
BELAGAVI, KARNATAKA**

COPYRIGHT

DECLARATION BY THE CANDIDATE

I hereby declare that KLE Academy of Higher Education & Research, Belagavi, Karnataka shall have the rights to preserve, use and disseminate this dissertation/thesis in print or electronic format for academic/research purpose.

Date:

Dr. FELBIN TOMY

Place: Belagavi

REG.NO. IE0218004

© KAHER, Belagavi, Karnataka

[Established under section 3 of the UGC Act, 1956 vide GOI.

Notification No. F.9-19/2000-U.3(A)]

UNDERTAKING

I, **Dr. FELBIN TOMY**, hereby declare that the information and data mentioned in my thesis entitled “**Comparative evaluation of intra orifice sealing ability of Neo MTA Plus, Calcium Enriched Mixture and Conventional Glass Ionomer Cement under Confocal Laser Scanning Microscope:- An In-Vitro study**” belongs to me and is original.

I am aware of the definition of plagiarism as detailed below:

- An act or instance of using or closely imitating the language and thoughts of another author without authorization and the representation of that author’s work as one’s own, as by not crediting the original author.
- A piece of writing or other work reflecting such unauthorized use or imitation.
- The deliberate or reckless representation of another’s words, thoughts or ideas as one’s own without attribution in connection with submission of academic work, whether graded or otherwise.

I hereby declare that the thesis prepared by me is original and does not involve plagiarism anywhere. In case at later stage it is found that I have indulged in plagiarism, then I am solely responsible for the same and the Institution is at liberty to take any disciplinary action against me including cancellation of dissertation or any other penalties imposed by the University.

Place: Belagavi

Dr.FELBIN TOMY

Date: _____

REG. NO. IE0218004

*This dissertation is dedicated
to*

Almighty God,

My Parents,

&

My Guide

ACKNOWLEDGEMENT

No endeavour can start, continue and complete without the blessings of the Almighty. At the outset I thank the almighty for always being by my side by bestowing strength and patience to complete the task entrusted. With the completion of my dissertation, I am overcome with a sense of satisfaction and even more gratitude towards all my mentors and well-wishers whom I wish to acknowledge.

*I would like to express the deepest appreciation to my honourable teacher and guide, **Dr. Sunita Shivanand** M.D.S., Reader, Department of Conservative Dentistry and Endodontics, KAHER Vishwanath Katti Institute of Dental Sciences, Belagavi. Her patience, encouragement, and meticulous supervision at every step along with his valuable suggestions for improvement enabled me to complete my dissertation which also helped me tremendously during my postgraduate tenure.*

*With deep gratitude, I thank **Dr. Preeti Doddwad** M.D.S., Professor and Head of the Department, Department of Conservative Dentistry and Endodontics, KAHER Vishwanath Katti Institute of Dental Sciences, Belagavi for giving enthusiastic encouragement, consistent support and unequivocal help during my post graduation.*

*I am grateful to **Dr. Alka Kale** M.D.S.,PhD Principal, K.L.E. VK Institute of Dental Sciences, Belagavi for extending her help and co-operation towards the completion of this dissertation.*

*I would like to express my solemn gratitude, respect & special debt of thanks to my beloved teachers **Dr. Sonal Joshi** M.D.S. Professor, **Dr. Anand Patil** M.D.S. Professor **Dr. Avinash Patil** M.D.S. Reader, **Dr. Neha Dhaded** M.D.S. Reader, **Dr. Shavina Patil** M.D.S. Reader, **Dr. Suresh Shenvi** M.D.S. Senior lecturer, **Dr. Sneha***

Patil M.D.S Senior Lecturer and Dr. Aniket Chavan M.D.S Senior lecture who selflessly devoted their time to me with patience whenever needed. Their colossal contribution all through these years will never be history.

My heartfelt thanks to Dr. Niraj Godbole M.D.S Senior lecturer, Dr. Vaidehi Dhopavkar M.D.S Senior lecturer, Dr. Mateen Peerzade M.D.S Senior lecturer and Dr. Olivia Banerjee M.D.S Senior lecturer for their sincere help and support given to me during this dissertation.

I would also like to extend my thanks to Mr. Satveer Jagwani M.PHARM. Research assistant, KLE's Dr. Prabhakar Kore Basic Science Research Center for helping me.

My sincere thanks to Mr. Javali, the statistician, for his services in carrying out the statistical analysis.

I am thankful to Mr. Bagalkoti, store Incharge, Mrs. Geeta, Mr. Mallappa, Materials Incharge and all non-teaching staff of my Department for their immense help during the course of my study.

My sincere thanks to my loving seniors Dr. Suraj, Dr.Neha, Dr. Ankita, Dr. Akash, and Dr. Pratik for having generously shared insight and for their unwavering support.

I extend my heartfelt gratitude to my colleagues Dr. Ashish, Dr. Deepti, Dr. Chaitra and Dr. Shrutika for their constant support and co-operation .

I am thankful to my juniors Dr. Abhijit and Dr. Amrutha for always being helpful and proof reading my dissertation.

*I extend my heartfelt thanks to my junior colleagues **Dr. Mahima, Dr. Priyanka, Dr. Shefali, Dr. Bhavna, Dr. Aishwarya, Dr. Rohit, Dr. Aishika and Dr. Greeta** for their help and co-operation.*

*I would also like to **thank Dr. Maurya M.D.S, Dr. Barath, Dr. Amal, and Dr. Sachin, Dr. Lal, Dr. Meekha, Dr. Liya** for always keeping my morale afloat.*

*I am thankful to **Mr. Anand and Mr. Arun** of Shri Vigneshwara Associates, Belagavi for formatting, printing and binding of my dissertation.*

*I would not have completed this dissertation without the unconditional support of my family, who have always been there for me whenever I needed them, with encouragement and love to empower me all the time. I owe everything of what I am and to my parents, **Mr. Tomy Thomas and Mrs. Rosamma k.p** who gave form to all my dreams and aspirations.*

I owe every success and joy to them and I humbly acknowledge that everything I am today is because of their love and support.

Thank you, one and all.

Date:

Place: Belagavi

Dr. FELBIN TOMY

LIST OF ABBREVIATIONS

SR.NO	ABBREVIATIONS	FULL FORM
1	CEM	Calcium Enriched Mixture
2	CH	Calcium hydroxide
3	HA	Hydroxy appetite
4	<i>et al</i>	Additional persons involved in the same study
5	WL	Working Length
6	Ni-Ti	Nickel Titanium
7	CEJ	Cemento-Enamel Junction
8	CFLSM	Confocal Laser Scanning Microscopy
9	° C	Degrees Celsius
10	µm	Micro meter
11	OSHA	Occupational Safety and Health Administration
12	ANOVA	Analysis of Variance
13	SD	Standard Deviation
14	n	Number of specimens
15	p-value	Probability of obtaining a test statistic at least as extreme as the one that was actually observed
16	NaOCl	Sodium Hypochlorite
17	EDTA	Ethylene Diamine Tetra-acetic Acid
18	<	Less than
19	>	Greater than
20	3D	Three dimensional

ABSTRACT

Aim and Objectives: Comparative evaluation of intra orifice sealing ability of Neo MTA Plus, Calcium Enriched Mixture and Conventional Glass Ionomer Cement under Confocal Laser Scanning Microscope:- An In-Vitro study.

Study design:Sixty four single rooted maxillary central incisors were decoronated, cleaned, shaped and obturated. Gutta-percha was removed to a depth of 3.5 mm from the orifice using a # 5 GG drill to facilitate a uniform orifice diameter of 1.3mm at its widest point. The sixty four samples were then divided into four groups (n=64), depending upon the type of intracoronal sealing material used.

Group 1: **Control**

Group 2: **NEO MTA Plus**

Group 3: **Calcium Enriched Mixture**

Group 4: **Conventional Glass Ionomer cement**

The materials were manipulated and placed into the orifice as per the manufacturer's direction. The specimens were submerged in Rhodamine-B dye in vacuum for one week. After which it was sectioned longitudinally and the depth of microleakage was measured using Confocal Laser Scanning Microscope.

Results: Group 3 CEM showed least depth of penetration followed by Neo MTA plus and Glass ionomer cement.

Conclusion: In the current study, Calcium Enriched Mixture was found to be superior over other experimental materials as an intra-orifice barrier.

Key words: Depth of Penetration, Neo MTA Plus, Calcium Enriched Mixture, Glass Ionomer cement, Confocal Laser Scanning Microscope.

TABLE OF CONTENTS

Sl.No.	PARTICULARS	Page No.
1	INTRODUCTION	1-5
2	AIM AND OBJECTIVES	6
3	RESEARCH HYPOTHESIS	7
4	REVIEW OF LITERATURE	8-13
5	MATERIALS AND METHOD	14-26
6	RESULTS	27-35
7	DISCUSSION	36-41
8	CONCLUSION	42
9	SUMMARY	43-44
10	BIBLIOGRAPHY	45-54
11	ANNEXURE	55-57

LIST OF TABLES

Table No.	Particulars	Page No.
1	Depth of penetration in the control group	27
2	Depth of penetration in Group 2 (Neo MTA Plus).	28
3	Depth of penetration in Group 3 (Calcium Enriched Mixture)	29
4	Depth of penetration in Group 4 (Conventional Glass Ionomer)	30
5	Table Showing Normality of depth of penetration (in μm) in four groups by Kolmogorov Smirnov test	33
6	Table showing Summary of depth of penetration (in μm) in four groups	33
7	Table showing Comparison of four groups with mean depth of penetration (in μm) by One way ANOVA.	34
8	Table showing Pair wise comparison of four groups with mean depth of penetration (in μm) by Tukey's multiple Post-hoc procedures.	34

LIST OF GRAPHS

Graph No.	Particulars	Page No.
1	Comparison of four groups with mean depth of penetration.	35

LIST OF FIGURES

Fig No.	Particulars	Page No.
1	Sixty four extracted maxillary central incisor teeth.	21
2	Decoronation with a double diamond disk	21
3	Rotary and Hand Instruments for Biomechanical Preparation along with irrigating solutions	22
4	Biomechanical preparation.	22
5	Irrigation.	23
6	GP removal with GG drills.	23
7	Neo MTA Plus, Calcium Enriched Mixture, Glass Ionomer Cement	24
8	longitudinally sectioned teeth	24
9	Confocal Laser Scanning Microscope	25
10	Representative CLSM image of group 1	26
11	Representative CLSM image of group 2	26
12	Representative CLSM image of group 3	26
13	Representative CLSM image of group 4	26

ANNEXURE

Annexure No.	Particulars	Page No.
I	Ethical Approval Certificate	55
II	Biostatistic Clearance Certificate	56
III	Plagiarism Check Certificate	57

INTRODUCTION

Microbial infection is the root causative factor associated with endodontic failure.¹ Literature has based secondary microleakage as a result of compromised coronal seal to be one of the prominent factor leading to the sequelae of endodontic failure.² Ray and Trope rightly stressed on the fact that the grade of restoration in the coronal preparation might affect the periradicular health than the quality of the obturation.³ The array of factors responsible for coronal microleakage can be listed as the fracture of tooth surface, loss of temporary restorative material, seepage at the restoration margins and recurrent caries.⁴

In absence of a proper coronal seal, there is a posed risk to the breakthrough success in the long run of the case. Furthermore, if this necessary criterion is not followed, it will uncover the obturated root canals to microbes thereby holding back the healing and generating infection in the tissue surrounding the apex of the tooth.⁵

To avoid this undue complication, employing a conducive appropriate material to plug the orifice, in supplementation to restoration temporarily condensed after root canal filling, has been observed as a precautionary layer of protection for the obturated canal.⁶

Orifice barrier technique as popularized is based on the use of a substance medium to secure the orifice and counterbalance any bacterial leakage in circumstances such as when there is a loss or non-functionality of the permanent restoration.^{7,8}

This recent technique comprises of substituting the filling material at the orifice opening with a material that acts as an impediment against any type of leakage from the environment.

The sealing material should furnish with a stable seal and should act as an impervious barrier.⁹ As stated by Wolcott et al. the prerequisites of an intra-orifice barrier is that it must be easy to apply, should bond to tooth structure, should provide a total blockade against microleakage, should be noticeably different from the surrounding tooth tissues, and should not intercede with the post obturation restoration.¹⁰

There has been a commendable study on the material science that can be used for the same and varied materials have been proposed to serve the same.¹¹ The materials widely researched and put to use in this context are Cavit, Glass ionomer cement, Amalgam, Intermediate restorative material (IRM), Super EBA, Composite resins, MTA, Biodentine and Calcium Enriched Mixture.^{12,13} Early in the evolution of the orifice barriers Cavit, Composite resins, IRM, and Super EBA were extensively studied to evaluate the sealing ability which concluded that their ability to seal endodontic access preparation showed lesser durability against the masticatory forces. This paved way for the search of newer materials with satisfactory properties.¹⁴

Neo MTA plus (AVALON BIOMED, USA) is the new advent when concerned with MTA and has come to the market recently. It contains a well ground powder of tricalcium silicate and tantalum oxide which serves as a radiopacifying agent in water-based gel vehicle that provides good handling property. It has clinical implications to be used for vital pulp therapy, apexification of immature root, and

sealing of root canal as per the manufacturer's specifications.¹⁵ Neo MTA Plus has suitable manipulation and doesn't wash off soon which is due to the virtue of its particular gel property and in adjunct, it doesn't exhibit any type of tooth staining.¹⁶ Neo MTA Plus has sulphur and aluminium on a higher value which demonstrates comparatively faster setting than the other MTA products.¹⁶

Calcium Enriched Mixture (BIONIQUE DENT, TEHRAN, IRAN) was pioneered in endodontics by Asgary et al in the year 2006. It is an amalgamation of a variety of calcium compounds dispersed in water which dispenses bioactive calcium and phosphate augmented material.

CEM biomaterial has a higher edge that it can be made use of even in an aqueous environment with the added value of a good handling property. Phosphorous is the prime integrant of CEM and shows competency of setting. It also possesses the capability to set in an water- based environment with a decreased setting time than MTA and offering a seal of orifice analogous to MTA.¹⁷

Glass ionomer cement has been classically put to use for a number of treatment modalities in restorative dentistry and endodontics. It holds its base reaction to be of silicate glass powder in polyacrylic acid.¹⁸ It allows for a generous amount of non-reacted glass which helps fortify the strength of cement after the set occurs in less diluted concentrates in water.

The acid-base reaction reinstates the set after 2-3 min of mixing time. Due to less time at hand, placement and adaptation promptly after mixing is advocated. Glass ionomers impart a substantial adhesion to the tooth surface providing a clinical supremacy.¹⁹ Glass ionomers are prepared from polyacrylic acid, and this compound

in particular has been known to foster adhesion, on the same grounds as done by zinc polycarboxylate cement.^{18,19}

Depth of penetration as evaluated with the aid of dye leakage can be optimally recorded using different microscopy techniques like Stereomicroscopy, Scanning Electron Microscopy, Transmission Electron Microscopy, and Confocal Laser Scanning Microscopy (CLSM). Confocal laser scanning microscopy is a breakthrough approach which has received an upheaved popularity in the recent decenniums in the field of fluorescence imaging and is considered as a prominent apparatus in biological research. Confocal microscopy imparts a precedence of shallow depth of field, blocking of out-of-focus glare, and the capability of assembling optical sectioned assembly from thick specimens.²⁰ *Laser scanning* involves capture of images point by point in the influence of localized laser excitation as against full sample illumination followed in conventional widefield microscopy.²⁰

A multitude of materials was put through investigatory procedures and compared for their effectiveness in sealing off the canal orifices from any type of fluids using divergent approaches.

To enhance the treatment prognosis and render the obturation free of any possibility of reinfection and microleakage a orifice barrier becomes a necessity of the hour. Hence, to provide with an improved and patient friendly treatment and to prevent reinfection in the periradicular and supporting osseous framework around the tooth, a suitable material built up over the coronal gutta-percha as a hitch to coronal microleakage would provide supremacy, increasing the possibilities for success.²¹

To the best of our knowledge there are no studies documented on comparison of intra-orifice sealing ability of Neo MTA Plus, Calcium Enriched Mixture, Conventional Glass Ionomer Cement.

Hence this study aims to evaluate and compare the intra-orifice sealing ability of Neo MTA Plus, Calcium Enriched mixture and Conventional Glass Ionomer cement using Confocal Laser Scanning Microscope.

AIM OF THE STUDY

To evaluate and compare the intra orifice sealing ability of Neo MTA Plus, CEM, and Conventional GIC by dye penetration method after obturation of root canal system under Confocal Laser Scanning Microscope.

OBJECTIVES

1. To evaluate the intra orifice sealing ability of Neo MTA Plus, CEM, Conventional GIC by dye penetration method using Confocal Laser Scanning Microscope.
2. To compare the intra orifice sealing ability of Neo MTA Plus, CEM and Conventional GIC by dye penetration method using Confocal Laser Scanning Microscope.

HYPOTHESIS

NULL HYPOTHESIS-

There will be no difference in the intra orifice sealing ability of Neo MTA Plus, Calcium Enriched Mixture and Conventional GIC when evaluated under Confocal Laser Scanning Microscope.

ALTERNATIVE HYPOTHESIS-

There will be a difference in the intra orifice sealing ability of Neo MTA Plus, Calcium Enriched Mixture and Conventional GIC when evaluated under Confocal Laser Scanning Microscope.

REVIEW OF LITERATURE

- 1) An ex vivo study was carried out to differentiate the microleakage in the coronal entity of the tooth, of MTA, CEM, and biodentine as intra orifice barriers. The samples were submerged in Rhodamine dye for 1 week and were observed under stereomicroscope under 10x magnification to assess degree of penetration. It was concluded that, although the statistics did not bear significance, CEM cement exhibited least microleakage as intra orifice barrier in endodontically treated teeth.²²
- 2) An in vitro study experiment was directed to evaluate and compare the intra orifice sealing capability of light cure Glass Ionomer Cement, Tetric N Flow and LC GIC + Tetric N Flow. As per the findings of the researchers, LC GIC + Tetric N Flow gave in better seal in comparison to LC GIC. Moreover, the statistical significance showed a lower value difference when leakage was considered.²³
- 3) The sealing ability of GIC, GMTA, WMTA, Composite resin was evaluated meticulously by compacting them over the gutta percha in root canal treated teeth. The results culminated to infer that that groups sealed with GMTA and WMTA showed least dye penetration than other groups and their difference was statistically significant.²⁴
- 4) In a study, endodontic therapy was followed by orifice sealing as a precautionary measure to prevent microleakage and the materials tested were CEM, MTA, Amalgam and composite resin. CEM and MTA groups were devoid of significant difference which was also found with amalgam and composite resin group. The pertinent point noted was that in the context of

average leakage time, MTA and CEM had an upper hand over the amalgam and composite resin group.²⁵

- 5) A study was conducted to test Neo MTA Plus (Avalon Biomed) and MTA plus in regard to physicochemical properties and capability of inducing apatite formation. The study provided a satisfactory base for Neo MTA explaining its properties of valueadded setting time and a up to par radiopacity. Calcium and hydroxyl ion release spurt was predominantly higher and for a longer duration when compared to MTA plus.²⁶
- 6) An in vitro research trial was accomplished to evaluate and compare the intra orifice sealing ability of Fusio, Fuji II, Fuji IX, and MTA “G” when placed in varied thickness. It was concluded that Fusio and MTA “G” were influenced by vertical extent of the cavity when contemplated for their sealing ability. At 2 or 3mm, MTA G showed better combating of microleakage. Fusio followed it in giving persistent conducive results at 2mm depth. Clubbing up the results,2mm of orifice cavity depth provides for better bonding for the adhesive orifice barriers.²⁷
- 7) An in vitro investigation was done to evaluate and compare the intra orifice ability Cavit, ProRoot MTA, or Tetric. It culminated down to justify that there was no significant correlation of the test materials and orifice depths. However, of the entirety of materials tested, tetric set forth a symbolically significant result than Pro Root or Cavit setting aside the parameter of orifice depth.²
- 8) A comparative evaluation of the seal of the orifice was done to differentiate the sealing ability of Mineral Trioxide Aggregate (MTA), Glass Ionomer Cement (GIC) Zinc oxide eugenol (ZOE) sealer and Acroseal sealer. The

study results concluded that MTA group attributed to less leakage than GIC group. Acroseal provided for a better adaptation with the dentinal walls than ZOE sealer. Teeth devoid of intracanal barrier exhibited almost complete leakage.²⁹

- 9 An in vitro study was executed to compare and contrast the orifice plugging efficacy of six varied dental materials: It included the MTA-based cements and standardized filling materials gauged by dye penetration method. The dye penetration order was: ProRoot WMTA<LuxaCore, Angelus White<EndoCem Zr<Fuji II LC<Elite .³⁰
- 10 A study assessed sealing materials Cavit, Vitremer, and Flow-It for the deterrence of microleakage as a post treatment sequelae. The results bore an inference that all groups evidenced dye penetration. Cavit furnished with a better seal than the other groups deterring the leakage in as high as 90% of the investigated specimens. However, the negative toll was taken by Flow-It quantifying to leakage in 65% of specimens. A lack of statistically significant difference was observed in the Vitremer group in which dye penetration calibration dropped down to 55% of specimens.³¹
- 11 A comparison of sealing potential of three restorative materials mineral trioxide aggregate (MTA), composite, glass ionomer cement (GIC) was determined. Results expressed that MTA showed exemplary results. However, 1mm depth GIC and Composite when weighed up showed significant difference. This breach was nullified at the depth of 2mm. By and large, of all the tested categories, MTA and GIC superseded composite at all the vertical dimensions.³²

- 12 CEM and MTA when compared pairwise for their seal of the orifice after intracoronar bleaching, a study summarized that the dye penetration in the specimens of both the groups showed similar penetration rates of the dye.³³

- 13 A study evaluated the ability of mineral trioxide aggregate as a root canal obturating material as against laterally adapted gutta percha and gutta percha. Canals obturated with laterally condensed gutta-percha or softened injectable gutta-percha showed lesser porosities not allowing the detrimental microleakage of dye in the apical region than canals filled with mineral trioxide aggregate. Therefore, the investigation inferred the superiority of gutta-percha obturation as against MTA in context to the apical seal.³⁴

- 14 The intra-orifice tightseal of ProRoot mineral trioxide aggregate (MTA), Biodentine, and Ortho MTA as the root canal obturation materials incorporating the fluid infiltration method. Within the boundaries of this study, microleakage values were similar to MTA ProRoot, Biodentine, and Ortho MTA the fluid infiltration technique.³⁵

- 15 An in vitro study was done to weigh up the coronal microleakage of mineral trioxide aggregate (MTA) plug and gutta-percha in stunted roots which were devised to be receptacle for post integrating the bacterial penetration technique. During the study period, turbidity was observed in all the gutta-percha samples and in only four MTA samples and the results abided by being statistically significant in the two groups. Based on the results, MTA exhibited much better sealing ability compared to gutta-percha as a root canal obturation substance in teeth with short roots.³⁶

- 16 An invitro study was done to compare microleakage in the crown entity of tooth of three compounds, light-cured glass-ionomer (GI), mineral trioxide aggregate (MTA), and composite resin as coronal barriers. MTA owing to its nonporous sturdy set plug made way for less leakage than composite and GI.³⁷
- 17 Bacterial microleakage technique was implemented in a study to evaluate for microleakage in teeth devised for reception of post sub serving either the assimilation or no assimilation of an intracanal glass ionomer barrier. Vitrebond emerged as a promising agent with statistically significant results showing an ability of providing superior coronal seal in teeth requiring post and core.³⁸
- 18 Resilon when weighed for sealing ability against gutta-percha with glass ionomer as an intra-orifice barrier integrating a fluid filtration model. After setting of the sealers, the specimens were put to test for microleakage utilizing a fluid filtration model. An elaborate statistical analysis portrayed significantly reduced seepage for the gutta-percha/glass-ionomer intra-orifice barrier group in contrast to Resilon group.³⁹
- 19 Resected teeth were subjected to obturation either mineral trioxide aggregate (MTA) or calcium enriched mixture (CEM). On a whole CEM proved to be more efficiently productive to cause lesser microleakage as compared to MTA. But, an prominent aspect the difference was meagrely significant.⁴⁰
- 20 Three obturation techniques were time-tested involving the use of mineral trioxide aggregate (MTA) and calcium-enriched mixture (CEM) cement as apical plugs. After cleaning and shaping, an open apex configuration was

obtained. Cement (MTA or CEM) in thickness of 5mm was packed as plugs at the orifice opening. Microleakage was measured by the fluid filtration method and results were analyzed. **It was concluded that** MTA and CEM cement have similar sealing ability as apical plugs and no significant difference was found in microleakage of the three groups.⁴¹

MATERIALS AND METHODOLOGY

MATERIALS

- Human maxillary central incisors
- 5.25% NaOCl (Vishal Dentocare, Ahmedabad)
- 17% EDTA (Ammdent CANALARGE, Punjab)
- Paper points (Diadent, Korea)
- AH-Plus sealer (Dentsply, Germany)
- Gutta-percha points (Dentsply, Germany)
- Neo MTA Plus (Avalon Biomed, USA)
- Calcium Enriched Mixture (Bionique Dent, Tehran, Iran)
- Conventional Glass Ionomer Cement (GC)
- Rhodamine B dye (Sigma Aldrich, Bengaluru)

ARMAMENTARIUM:

- Airotor (NSK Pan Air, Japan)
- Straight fissure bur (Mani, India)
- 10 K- file (Mani, India)
- Pro Taper universal rotary system (Dentsply, Germany)

- Endomotor (X-SMART, Dentsply, Germany)
- 5ml syringes 27gauge needle
- Diamond disc (Double sided)
- Confocal Laser Scanning Microscope (ZEISS LSM T-PMT, Germany)

METHODOLOGY

This study was conducted in Department of Conservative Dentistry and Endodontics, KLE's Vishwanath Katti Institute of Dental Sciences, Belagavi and the laboratory procedures were undertaken in Dr. Prabhakar Kore's Basic Research Laboratory, KLE University, Belagavi.

Specimens were experimentally observed under the Confocal Laser Scanning Microscope at National Center for Biological Sciences, Bengaluru.

Method of collection of data (including sampling procedure)

For this study, sixty four extracted human maxillary central incisors with the following inclusion and exclusion criteria were collected from the Department of Oral and Maxillofacial Surgery and stored in 0.5% NaOCl solution for three days to get rid of any organic debris and disinfect the tooth specimen. All teeth were then rinsed with the flow of tap water to remove NaOCl residues.

INCLUSION CRITERIA

Human permanent maxillary central incisors with single straight canal with closed apex.

EXCLUSION CRITERIA:

Teeth with radicular resorption, cracks/fracture line

- Root canal treated tooth
- Teeth with calcified canal
- Teeth with root caries
- Teeth with multiple canals/anatomic variations

The samples were considered under inclusion and exclusion criteria by using radiographs.

Preparation of specimens

Sixty four extracted human permanent maxillary central incisors stored at 100% humidity were selected for the study. The totality of the sample size underwent decoronation with the aid of a double diamond disk under copious amount of water. The root length was standardized upto 11mm for all the samples. Working length was estimated using a # K-10 file sliding it into the canal until it was visible at apical foramen and the file was retracted by 1mm to establish the working length at this level.

Pro Taper Universal system was used for canal preparation upto master apical file size F4. 5.25% NaOCl and 17% EDTA was used after each successive instrument during preparation. Then the canals were rinsed with 2ml of 17% EDTA and a terminating flush was achieved with saline was done. Canals were rendered dry with sterile paper points and obturation was achieved with Pro Taper F4 gutta-percha in

combination with AH-Plus sealer.

The randomization was implemented and specimens were allotted to four groups containing 16 teeth each.

Group 1 =Control (without an orifice barrier)

GROUP 2=Neo MTA Plus

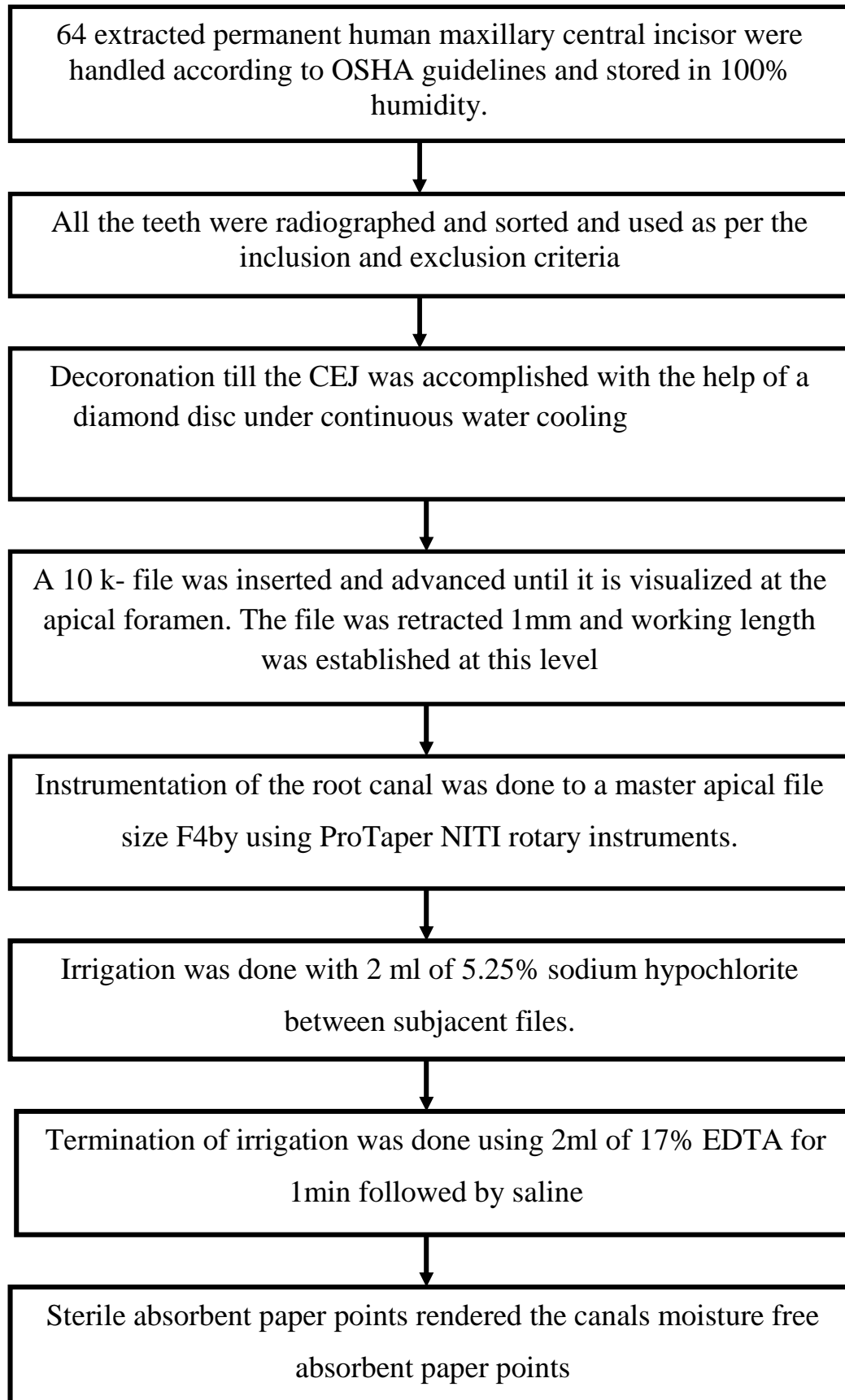
Group 3 =Calcium Enriched Mixture

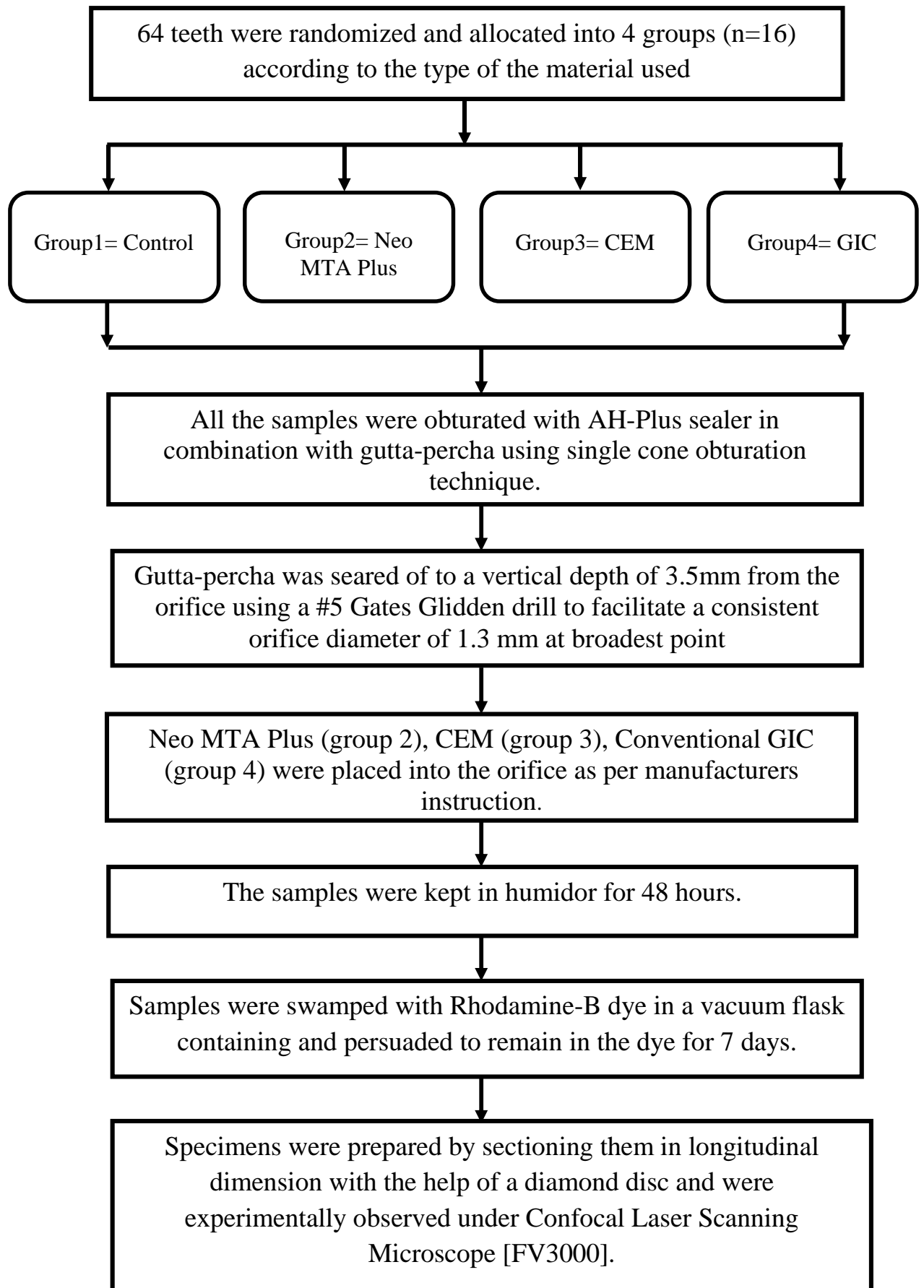
Group 4=Conventional Glass Ionomer Cement

Gutta-percha was seared off in the coronal region to a vertical height of 3.5mm from the orifice with #5 Gates Glidden drill to facilitate a consistent orifice diameter of 1.3 mm at broadest point. The specimens in Group one which was the deemed control group was obturated with gutta-percha and were devoid of any searing of GP and placement of intra-orifice barrier. In Group 2, Group 3, Group 4 the orifices were sealed with different test materials according to the manufacturer's instruction. Each specimen was color coded to represent its liability to a particular group and underwent humidification for 48 hours to provide for the setting of the sealer and test materials. Three layers of nail varnish were spread on all experimental teeth all the way from the root apex to junction where cementum and enamel coincided. Samples were swamped in vacuum flask containing Rhodamine B dye and permitted to hold onto the dye for 7 days. To proceed with the further procedure and assess the microleakage, sectioning was done to acquire longitudinal sections in the bucco-lingual dimension with the aid of a diamond disc and water spray. The splitting was placed given at the level of the restorative material at the intra-orifice and along

the root and the filling material. The samples were then subjected to viewing under Confocal Laser Scanning. The FV10-ASW Viewer provided for gauging the dye penetration in micrometer utilizing the images obtained during confocal microscopic examination. Mean and Standard Deviation of experimental groups were calculated. The data obtained was put through statistical analysis.

METHODOLOGY WITH FLOWCHART-





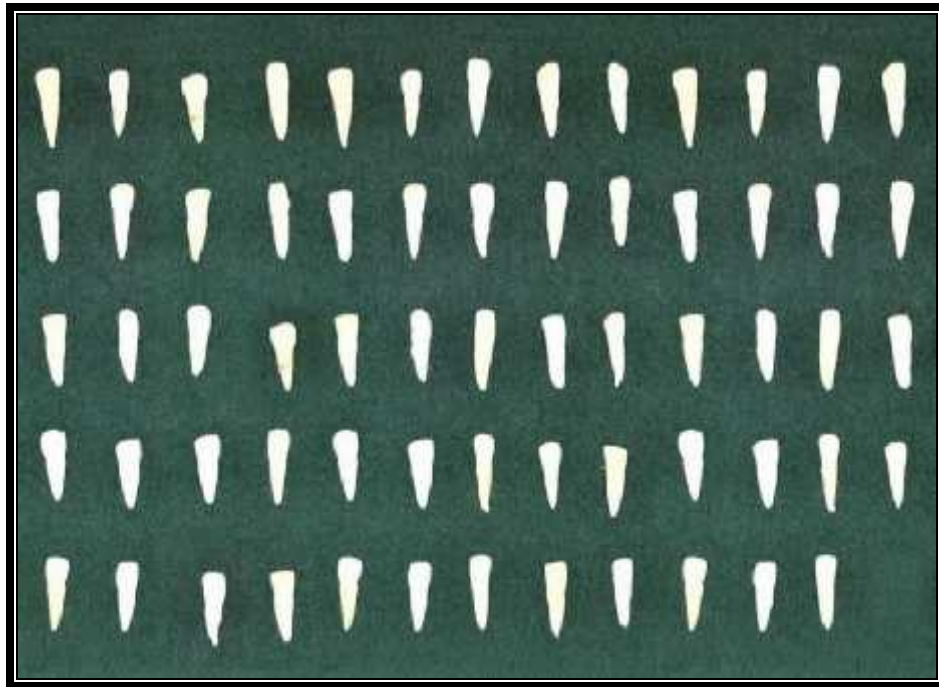


Fig 1: Teeth specimens (n = 64)



Fig 2: Decoronation with a double diamond disk



Fig 3: Rotary and Hand Instruments for Biomechanical Preparation along with irrigating solutions



Fig 4: Biomechanical Preparation



Fig 5: Irrigation done with 5.25% NaOCl and RC prep and final rinse was done with 2ml of 17% EDTA and saline



Fig 6: Gutta-percha was removed to a depth of 3.5mm from the orifice with #5 Gates Glidden drill



Fig 7: Specimens divided into different groups according to the intra-orifice barrier material used Neo MTA Plus, Calcium Enriched Mixture, Glass Ionomer Cement



Fig 8: Teeth longitudinally sectioned and dye penetration observed



Fig 9: Confocal Laser Scanning Microscope

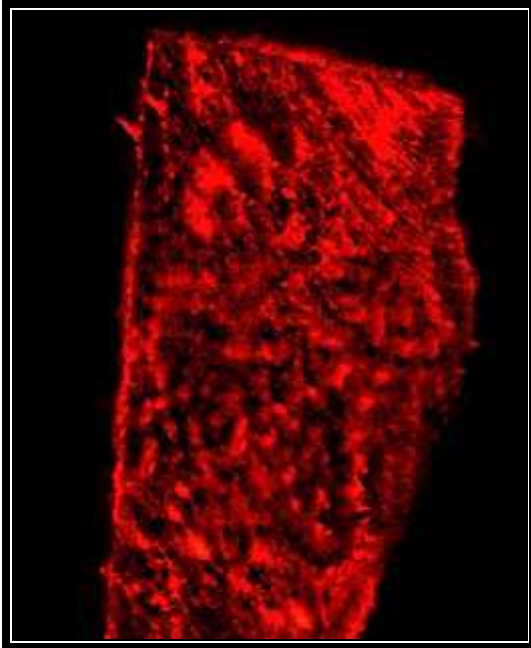


Fig 10: Confocal image representing dye penetration in the specimens of group1 [Control]



Fig 11: Confocal image representing dye penetration in the specimens of group 2 [Neo MTA Plus]



Fig 12: Confocal image representing dye penetration in the specimens of group 3 [Calcium Enriched mixture]



Fig 13: Confocal image representing dye penetration in the specimens of group 4 [Glass Ionomer Cement]

RESULTS

Table 1: GROUP 1 (Control Group)

Sample No.	Depth of penetration (in μm)
1	6.08
2	6.07
3	6.09
4	6.05
5	6.13
6	6.1
7	6.06
8	6.08
9	6.09
10	6.07
11	6.13
12	6.06
13	6.08
14	6.09
15	6.1
16	6.09

Table 2 : GROUP 2 (Neo MTA Plus)

Sample No.	Depth of penetration (in μm)
1	2.44
2	2.47
3	2.41
4	2.45
5	2.43
6	2.39
7	2.49
8	2.47
9	2.41
10	2.45
11	2.43
12	2.44
13	2.45
14	2.39
15	2.41
16	2.42

Table 3 : GROUP 3 (Calcium Enriched Mixture)

Sample No.	Depth of penetration (in μm)
1	2.31
2	2.33
3	2.29
4	2.35
5	2.27
6	2.34
7	2.28
8	2.34
9	2.27
10	2.35
11	2.29
12	2.33
13	2.31
14	2.34
15	2.35
16	2.31

Table 4 : GROUP 4 (Glass Ionomer Cement)

Sample No.	Depth of penetration (in μm)
1	3.9
2	4.1
3	3.7
4	4
5	3.8
6	3.6
7	4.2
8	3.6
9	4.1
10	4.2
11	3.5
12	3.7
13	4
14	3.9
15	3.8
16	3.9

The data on depth of penetration was obtained for all the samples in each group. Descriptive statistics of parameters like mean, standard deviation and 95% Confidence Interval were obtained for each group. One-way analysis of variance was performed to determine the statistical significance of difference in the means of parameters. Further, Tukey's post-hoc test was performed to determine the pair wise significance of difference between the means.

The description of methods used in the study are as below

TABLE 5: Normality of depth of penetration (in μm) in four groups by Kolmogorov Smirnov test.

TABLE 6: Shows the number of samples per group, their mean, standard deviation and 95% confidence interval for mean in all 4 groups.

The depth of penetration (in μm) in four groups follow a normal distribution, therefore, the parametric tests i.e. one way ANOVA followed by Tukey's multiple Post-hoc procedures were applied.

TABLE 7: Comparison of four groups with mean depth of penetration (in μm) by one way ANOVA.

Table 8: Pair wise comparison of four groups with mean depth of penetration (in μm) by Tukey's multiple Post-hoc procedures.

According to this it was found that

Group 1 → Control Group showed maximum depth of penetration and was statistically significant when compared to the other test groups.

Group 2→Neo MTA Plus Group showed significant difference with Group 1, group 3, and Group 4

Group 3→Calcium Enriched Mixture Group showed least depth of penetration and was statistically significant when compared to Group1, Group 2 and Group 4.

Group 4→showed statistical difference with all the groups.

Table 5: Normality of depth of penetration (in μm) in four groups by Kolmogorov Smirnov test

Groups	Mean	SD
Control	0.6920	0.7250
Neo MTA Plus	0.4570	0.9850
Calcium enriched mixture	0.7300	0.6600
Glass ionomer cement	0.4330	0.9920

Table 6: Summary of depth of penetration (in μm) in four groups

Groups	N	Min	Max	Mean	SD	SE	95% CI for mean	
							Lower bound	Upper bound
Control	16	6.05	6.13	6.09	0.02	0.01	6.07	6.10
Neo MTA Plus	16	2.39	2.49	2.43	0.03	0.01	2.42	2.45
Calcium enriched mixture	16	2.27	2.35	2.32	0.03	0.01	2.30	2.33
Glass ionomer cement	16	3.50	4.20	3.88	0.22	0.05	3.76	3.99

Table 7: Comparison of four groups with mean depth of penetration (in μm) by one way ANOVA.

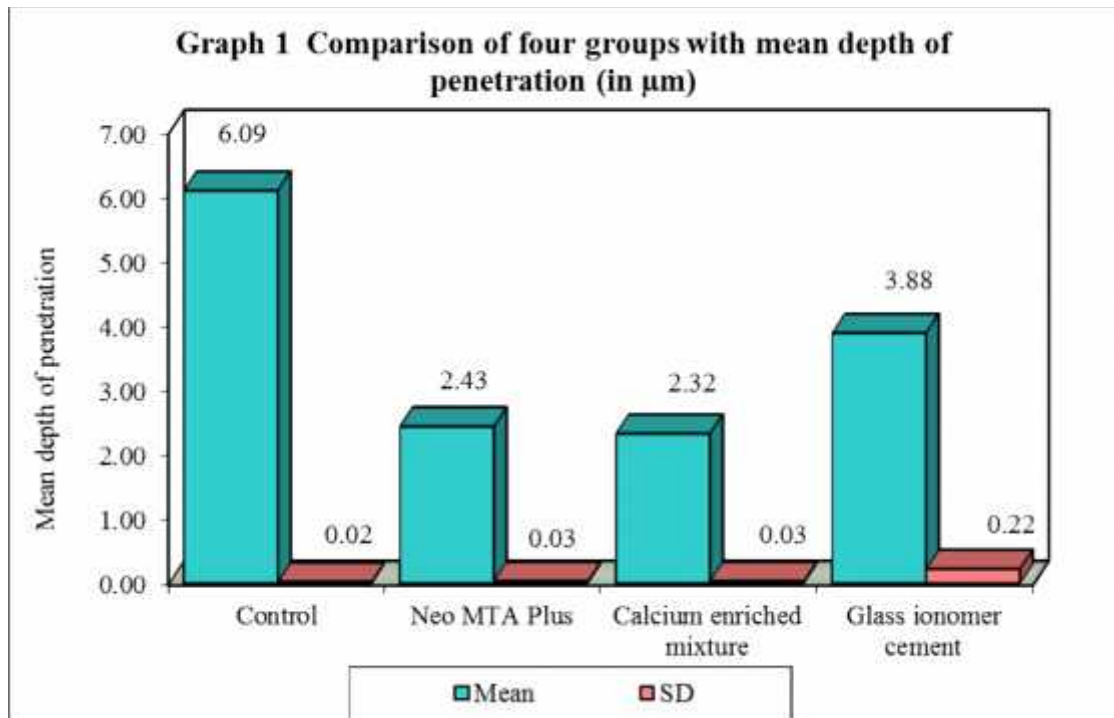
Sources of variation	Degrees of freedom	Sum of squares	Mean sum of squares	F-value	p-value
Between groups	3	147.78	49.26	3980.3500	0.0001*
Within groups	60	0.74	0.01		
Total	63	148.53			

* $p < 0.05$

Table 8: Pair wise comparison of four groups with mean depth of penetration (in μm) by Tukey's multiple Post-hoc procedures.

Groups	Control	Neo MTA Plus	Calcium enriched mixture	Glass ionomer cement
Mean	6.09	2.43	2.32	3.88
SD	0.02	0.03	0.03	0.22
Control	-			
Neo MTA plus	P=0.0002*	-		
Calcium enriched mixture	P=0.0002*	P=0.0199*	-	
Glass ionomer cement	P=0.0002*	P=0.0002*	P=0.0002*	-

* $p < 0.05$



Experimental samples of the Group 1 leaked 6.09 μm into the gutta-percha. Group 2 showed leakage at 2.43 μm mean penetration while Group 3 showed the least at 2.32 μm mean penetration. Group 4 showed 3.88 μm leakage. There was statistically significant difference between all the groups.

DISCUSSION

The purpose of root canal therapy is to eradicate bacteria from the infected 3D root canal system to favour the interception of recurrence of infection and facilitate the healing of the periapical pathology.⁴²

This virtue has been accomplished by establishing a compact and three-dimensional obturation seal in the entirety of root length from the coronal aperture to the apical constriction. A sustainable amount of evidence suggests that coronal invasion by microbes can persuade the penetration of the entire root canal system.⁴³ Therefore, a proper barrier on the canal orifice is equally crucial as that of apical seal as the salivary fluid can dissolve the root canal filling material, culminating into contamination of the root canal space. Moreover, it also spreads the infection to the apex and the periradicular region.⁴³

In augmentation, coronal restoration is a prime requirement for long-standing success of the therapy.

The null hypothesis of this study was rejected as there was difference among all the four experimental groups in terms of dye penetration with coronal barrier materials.

Extracted human teeth with only one canal were chosen in the present study because their preparation is relatively less cumbersome when the disclosure of the orifices and restoration of the same was concerned.⁴⁴

The teeth included were decoronated to acquire a stipulated root length of 11 mm by dissecting them at the level where the cementum and enamel coalesce forming

a boundary with the aid of a diamond disk under copious water cooling. Chemomechanical preparation was done with the Pro Taper Universal system upto a Master apical file size F4, 5.25% NaOCl and 15% EDTA were the irrigating solutions dispensed in the breach of use of successive instrument. Once instrumentation was completed, the canals received a rinse of 2 ml of 17% ethylene diamine tetra acetic acid solution followed by 2.5% sodium hypochlorite, and a final flush was executed with 5 ml of saline. All the canals were obturated with single cone gutta percha point and AH Plus sealer. After obturation Gutta-percha was extirpated to a vertical dimension of 3.5 mm from the orifice using a # 5 Gates Glidden drill. This facilitated an uniform orifice diameter of 1.3 mm at its broadest point.⁴⁵

This investigatory trial has utilized a barrier thickness of 3.5mm to establish the seal at the coronal aperture which is in accordance to the evidence in the literature that sets forth that a 3.5 mm of material is the minimal obligatory thickness paramount in coronal restoration to curb any type of leakage.^{45,46,47,48,49}

An array of *ex vivo* investigatory procedures were adopted to appraise microleakage through obturated roots such as dye penetration, bacterial leakage, electrochemical method, fluid filtration, radioisotope labelling, and scanning electron microscope analysis.⁵⁰ In the aforementioned study, dye penetration method was chosen to check for coronal microleakage. Dye penetration method was used due to its known leverage of properties of being inexpensive, easy manipulation, and in particular its superior degree of staining.⁵¹ Dyes possess a molecular weight subordinate than that of bacterial toxins and therefore they have a diffusability similar to butyric acid, an observed microbial byproduct of bacterial metabolism.⁵² In this study we have used Rhodamine-B dye owing to its considerable properties of having

small particle size, enhanced penetration, dissolution capacity in water, diffusability, and compatibility with the hard tissues. The dye was integrated under vacuum penetration method as it succors to extract entrapped air which can counter entire dye penetration.^{53,45}

Depth of tubule penetration of dye has been determined using several microscopy techniques, including Stereomicroscopy, Scanning Electron Microscopy, Transmission Electron Microscopy, And Confocal Laser Scanning Microscopy (CLSM). Amidst these, CLSM extends an upper hand of the use of intact hard tissue specimens which do not stand in the need of a special sectioning technique (sputter coating), normalizing the conditions of observation. It further benefits in giving an inward eye up to a depth of 10 μm subjunct to the exterior expanse of the specimen.^{54,55,56}

In the present study, Neo Mta Plus, Calcium Enriched Mixture, and Glass Ionomer Cements were evaluated and compared for coronal microleakage. With respect to the results, all the experimental groups exhibited leakage within the test materials. Calcium Enriched Mixture showed the least coronal leakage, followed by Neo MTA Plus and then Glass Ionomer Cement.

The Control group i.e Group 1 which had no coronal barrier exhibited the maximum leakage.

Group2 Neo MTA plugs exhibited significantly lower leakage than groups with GIC plugs. This is in accordance with various other studies that reported MTA to have better sealing ability than GIC. Torabinejad *et al* reported a levelled up marginal adaptation of MTA which accounted for its capability of resisting leakage.⁵⁷

The enhanced sealing ability has been credited to its hydrophilic nature by virtue of which it expands during setting in the moist environment.⁵⁸ GIC has a leaning to entrap air bubbles while setting forming a relatively porous set barricade as against MTA which is condensable and builds a less porous barrier leading to less microleakage.⁵⁹ Owing to the material shrinkage on setting, GIC potentiates gap formation when in association with dentin ensuing a poor seal .^{60,61}

The probability for air bubble generation emanating in voids can be the guessed extrapolation for hireling findings of GIC. ⁶³

Group 3 Calcium Enriched Mixture showed least coronal microleakage when compared with all other expiremental groups.

Studies on CEM have suggested that it can liberate phosphorus and calcium ions which boost the alkalinity of environment and persuade the mineralization, which testified its hard tissue inductivity.⁶³ CEM has already been investigated for numerous treatment options like management of furcal perforation, vital pulp therapies in permanent and primary teeth , root end filling , management of root resorption and pathologic/iatrogenic perforations, periradicular surgery and revascularization of necrotic immature permanent molars .^{64,65,66,67} CEM has presented some well documented benefits in comparison to MTA of a decreased setting time and also symbolically high-calibre results in context of film thickness and flow, ease of handling, and intensified antibacterial effects. ⁶⁸ It also has a established proof as an inducer of hydroxyapatite formation in the presence of normal saline⁶⁹

The prominent components of CEM are alkaline earth metal oxides and hydroxides (for example, calcium oxide and calcium hydroxide [CH]), calcium phosphate, and calcium silicate.⁷⁰ The distinguishing factor between CEM and MTA is that considering the chemical composition phosphorous is the major fraction of CEM whereas it is in close approximation to the detection limit in MTA.⁷¹ In disparity with MTA, CEM shows surface composition similar to the enclosing dentin. With the similarity between CEM and dentin in the presence of hydroxyapatite, there is an increased feasibility of cementogenesis over dentin.⁷¹

In compliance with the International Standard Organization (ISO) 6876 standard for dental root canal sealing materials, CEM when as a mixture in water as a solvent releases bioactive calcium and phosphate fortified molecules. In the process of mixing it undergoes hydration reaction to produce CH. This generation is largely because of the reactions involving calcium silicates, calcium phosphate, and calcium oxide in adjunct to the presence of CH. CH splits up to give out calcium and hydroxyl ions, taking the pH to the higher side and elevates the calcium concentration.⁷⁰ Furthermore, this futuristic cement liberates calcium and phosphorus ions from aboriginal sources that culminate into abundance of OH⁻, Ca²⁺, and PO₄⁻ ions which serve to form the building blocks of the Hydroxyapatite crystals.⁷² Backing up the formation of these crystals are numerous studies which certify HA formation in normal saline solution apart from simulated body tissue fluid.⁷¹

Foregone studies have revealed that CEM exhibited reasonable film thickness 25 µm and flow 1 mm, variably different in the statistical mannerisms from MTA (63 µm and 0.79 mm), respectively. The sustainable expansion and rational flow and film

thickness of CEM could warrant a constructive closure post the setting, and lessen the ensuing seepage.⁷³

By and large, it appears that all three materials tested in the current study are suitable for use as intra-orifice barriers in endodontically ventured teeth

In conclusion, immediate placement of a suitable intra-orifice barriers such as CEM cement, Neo MTA, Glass Ionomer Cement prior to final restoration of tooth can effectively decrease the coronal microleakage and recontamination of root canal contents.

CONCLUSION

Thus, the present study concludes that double seal is required to prevent coronal microleakage which could be achieved by using an intra-orifice barrier. In the current study, Calcium Enriched Mixture was found to be superior over other experimental materials as an intra-orifice barrier.

SUMMARY

The study was executed following all the norms, in the Department of Conservative Dentistry and Endodontics, KLE Academy Of Higher Education And Research, Vishwanath Katti Institute of Dental Sciences, Belagavi aiming to evaluate and compare the intra orifice sealing ability of Neo MTA Plus, Calcium Enriched Mixture, and Conventional Glass Ionomer Cement by dye penetration method after obturation of root canal system under Confocal Laser Scanning Microscope.

To accomplish this study, sixty four extracted human maxillary central incisors were assembled. All the samples underwent the process of decoronation, shaping and cleaning and obturation in order. Pro Taper Universal system was used for canal preparation upto master apical file size F4. 5.25% NaOCl and RC prep was used after each successive instrument during preparation. The canals acquired the final rinse with 2ml of 17% EDTA followed by saline. Canals were dried with sterile paper points and obturation was done with Pro Taper F4 gutta-percha and AH-Plus sealer.

The samples were randomly allocated to four groups containing sixteen teeth each.

Group 1 =Control

GROUP 2=Neo MTA Plus

Group 3 =Calcium Enriched Mixture

Group 4=Conventional Glass Ionomer Cement

Gutta-percha in the orifice region was extirpated to a depth of 3.5mm with #5 Gates Glidden. The Group one control group was obturated with gutta-percha at the level of the orifice and received no intra orifice barrier. In Group 2, Group 3, Group 4 the orifices were sealed with different test materials meticulously following the instructions by manufacturers. After sequential coding of each specimen separately, they were subjected to a humidifier for 48 hours to provide for setting of the sealer and the test materials. Three layers of nail varnish were applied on all experimental teeth all the way from the root apex to the level of the cemento-enamel junction. Samples were swamped in vacuum flask containing Rhodamine B dye and permitted to hold onto the dye for 7 days. To proceed with the further procedure and assess the microleakage, sectioning was done to acquire longitudinal sections in the bucco-lingual dimension with the aid of a diamond disc and water spray. The cut was given at the level of the restorative material at the intra-orifice and along the root and the filling material. The samples were then subjected to viewing under Confocal Laser Scanning Microscope. The FV10-ASW Viewer provided for gauging the dye penetration in micrometer utilizing the images obtained during confocal microscopic examination. Mean and Standard Deviation of experimental groups were obtained. The data obtained was made liable to statistical analysis.

The phenomenon of leakage showed up in all the groups with all the test materials. Moreover, the ascending order in the performance ability in context to microleakage was Calcium Enriched Mixture, followed by Neo MTA Plus and Glass Ionomer Cement.

BIBLIOGRAPHY

1. Friedman S, Abitbol S, Lawrence HP. Treatment outcome in endodontics: The Toronto study. Phase 1: Initial treatment. *J Endod* 2003;29:787-93.
2. Saunders WP, Saunders EM. Coronal leakage as a cause of failure in root-canal therapy: a review. *Endod Dent Traumatol* 1994;10(3):105-8.
3. Ray HA, Trope M. Periapical status of endodontically treated teeth in relation to the technical quality of the root filling and the coronal restoration. *Int Endod J*. 1995;28(1):12-8.
4. Tselnik M, Baumgartner JC, Marshall JG. Bacterial leakage with mineral trioxide aggregate or a resin-modified glass ionomer used as a coronal barrier. *J Endod*. 2004;30(11):782-4.
5. Pisano DM, DiFiore PM, McClanahan SB, Lautenschlager EP, Duncan JL. Intraorifice sealing of gutta-percha obturated root canals to prevent coronal microleakage. *J Endod* 1998;24:659-62.
6. Jenkins S, Kulild J, Williams K, Lyons W, Lee C. Sealing ability of three materials in the orifice of root canal systems obturated with gutta-percha. *Journal of Endodontics*. 2006 Mar 1;32(3):225-7.
7. Chailertvanitkul P, Saunders WP, MacKenzie D. Coronal leakage in teeth root-filled with gutta-percha and two different sealers after long-term storage. *Dental Traumatology*. 1997 Apr;13(2):82-7.
8. Wolcott JF, Hicks ML, Himel VT. Evaluation of pigmented intraorifice barriers in endodontically treated teeth. *Journal of endodontics*. 1999 Sep 1;25(9):589-92.

9. Parolia A, Kundabala M, Acharya SR, Saraswathi V, Ballal V, Mohan M. Sealing ability of four materials in the orifice of root canal systems obturated with gutta-percha. *Endodontology*. 2008;20(2):65-70.
10. Verma MR, Desai VM, Shahani SN. Importance of coronal seal following routine endodontic therapy. *Endodontology*. 1992;1(2):17-9.
11. Roghanizad N, Jones JJ. Evaluation of coronal microleakage after endodontic treatment. *J Endod*. 1996;22(9):471-3.
12. Chailertvanitkul P, Saunders WP, Saunders EM, MacKenzie D. An evaluation of microbial coronal leakage in the restored pulp chamber of root-canal treated multirooted teeth. *International Endodontic Journal*. 1997 Sep;30(5):318-22.
13. Beckham BM, Anderson RW, Morris CF. An evaluation of three materials as barriers to coronal microleakage in endodontically treated teeth. *J Endod*. 1993;19(8):388-91.
14. Lee YC, Yang SF, Hwang YF, Chueh LH, Chung KH. Microleakage of endodontic temporary restorative materials. *Journal of endodontics*. 1993 Oct 1;19(10):516-20.
15. Siboni F, Taddei P, Prati C, Gandolfi MG. Properties of NeoMTA Plus and MTA Plus cements for endodontics. *International Endodontic Journal*, 50, e83–e94, 2017.
16. J. Camilleri , staining potential of neo MTA plus, MTA plus and biodentine used for pulpotomy procedure, *journal of endodontia* , vol .41 , no . 7 , pp. 1139-1145,2015.
17. Asgary S, Shahabi S, Jafarzadeh T, Amini S, Kheirieh S. The properties of a new endodontic material. *J Endod*. 2008;34(8):990-3.

18. McCabe, John F.; Walls, Angus W.G. (2008). *Applied Dental Materials* (9 ed.). Oxford, United Kingdom: Wiley-Blackwell (an imprint of John Wiley & Sons Ltd)
19. Sidhu SK, Nicholson JW. A review of glass-ionomer cements for clinical dentistry. *Journal of functional biomaterials*. 2016 Sep;7(3):16.
20. Sungur DD, Aksel H, Purali N. Effect of a Low Surface Tension Vehicle on the Dentinal Tubule Penetration of Calcium Hydroxide and Triple Antibiotic Paste. *Journal of endodontics*. 2017 Mar 31;43(3):452-5
21. Roghanizad N, Jones JJ. Evaluation of coronal microleakage after endodontic treatment. *J Endod*. 1996;22(9):471-3.
22. Fatemeh Ramezanali, Sasan Aryanezhad, Fatemeh Mohammadian , Fatemeh Dibaji , Mohammad javad kharazifard , *Doi: 10.22037/iej.2017.41*
23. Bandish Parekh, Rukshin S. Irani¹, Sucheta Sathe¹, Vivek Hegde¹ IP: 223.237.163.211]
24. K T Divya¹, G Satish², T S Srinivasa³, Veera Reddy⁴, K Umashankar⁵, B Mohan Rao⁶ *Journal of International Oral Health 2014; 6(4):12-17*
25. Hamid Reza Yavari , Mohammed Samiei , Shahriar Shahi, Mohammed Aghazadeh , Farnaz Jafari , *IEJ Iranian Endodontic Journal 2012;7(1):25-30*
26. F. Siboni¹ , P. Taddei², C. Prati³ & M. G. Gandolfi¹ *International Endodontic Journal*, 50, e83–e94, 2017 doi:10.1111/iej.12787
27. Yavari H, Samiei M, Eskandarinezhad M, Shahi S, Aghazadeh M, Pasvey Y. An in vitro comparison of coronal microleakage of three orifice barriers filling materials. *Iranian endodontic journal*. 2012;7(3):156.

28. Jenkins S, Kulild J, Williams K, Lyons W, Lee C. Sealing ability of three materials in the orifice of root canal systems obturated with gutta-percha. *Journal of Endodontics*. 2006 Mar 1;32(3):225-7.
29. Malik G, Bogra P, Singh S, Samra RK. Comparative evaluation of intracanal sealing ability of mineral trioxide aggregate and glass ionomer cement: An in vitro study. *Journal of conservative dentistry: JCD*. 2013 Nov;16(6):540.
30. Lee KS, Kim JS, Lee DY, Kim RJ, Shin JH. In vitro microleakage of six different dental materials as intraorifice barriers in endodontically treated teeth. *Dental materials journal*. 2015 May 28:2014-42.
31. Sauáia TS, Gomes BP, Pinheiro ET, Zaia AA, Ferraz CC, Souza-Filho FJ. Microleakage evaluation of intraorifice sealing materials in endodontically treated teeth. *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*. 2006 Aug 1;102(2):242-6.
32. Salim B, Hassan N. Effect of different intraorifice barriers in endodontically treated teeth obturated with gutta-percha. *International Arab Journal of Dentistry*. 2015 Nov 15;6(3).
33. Moghadam N, Abdollahi AA, Fakhim HA, Borna Z. In vitro sealing properties of calcium-enriched mixture and mineral trioxide aggregate orifice barriers during intra-coronal bleaching. *Iranian endodontic journal*. 2017;12(2):231.
34. Vizgirda PJ, Liewehr FR, Patton WR, McPherson JC, Buxton TB. A comparison of laterally condensed gutta-percha, thermoplasticized gutta-percha, and mineral trioxide aggregate as root canal filling materials. *Journal of endodontics*. 2004 Feb 1;30(2):103-6.

35. Mousavi SA, Khademi A, Soltani P, Shahnasari S, Poorghorban M. Comparison of sealing ability of ProRoot mineral trioxide aggregate, biodentine, and ortho mineral trioxide aggregate for canal obturation by the fluid infiltration technique. *Dental Research Journal*. 2018 Sep;15(5):307
36. Khademi AA, Shekarchizade N. Evaluation of coronal microleakage of mineral trioxide aggregate plug-in teeth with short roots prepared for post placement using bacterial penetration technique. *Indian Journal of Dental Research*. 2016 May 1;27(3):295.
37. Yavari H, Samiei M, Eskandarinezhad M, Shahi S, Aghazadeh M, Pasvey Y. An in vitro comparison of coronal microleakage of three orifice barriers filling materials. *Iranian endodontic journal*. 2012;7(3):156.
38. Vijay R, Indira R. Effect of glass-ionomer cement as an intra-canal barrier in post space prepared teeth: An in vitro study. *Journal of conservative dentistry: JCD*. 2009 Apr;12(2):65.
39. Jack RM, Goodell GG. In vitro comparison of coronal microleakage between Resilon alone and gutta-percha with a glass-ionomer intraorifice barrier using a fluid filtration model. *Journal of endodontics*. 2008 Jun 1;34(6):718-20.
40. Milani AS, Shakouie S, Borna Z, Deljavan AS, Jafarabadi MA, Azar FP. Evaluating the Effect of Resection on the Sealing Ability of MTA and CEM Cement. *Iranian endodontic journal*. 2012;7(3):134.
41. Tabrizizade M, Asadi Y, Sooratgar A, Moradi S, Sooratgar H, Ayatollahi F. Sealing ability of mineral trioxide aggregate and calcium-enriched mixture cement as apical barriers with different obturation techniques. *Iranian endodontic journal*. 2014;9(4):261.

42. Siqueira JF Jr, Rocas IN (2008) Clinical implications and microbiology of bacterial persistence after treatment procedures. *J Endod* 34(11):1291–1301 e1293
43. Salim B, Hassan N. Effect of different intraorifice barriers in endodontically treated teeth obturated with gutta-percha. *International Arab Journal of Dentistry*. 2015 Nov 15;6(3).
44. Tselnik M, Baumgartner JC, Marshall JG. Bacterial leakage with mineral trioxide aggregate or a resin-modified glass ionomer used as a coronal barrier. *Journal of Endodontics*. 2004 Nov 1;30(11):782-4.
45. Parekh B, Irani RS, Sathe S, Hegde V. Intraorifice sealing ability of different materials in endodontically treated teeth: An in vitro study. *Journal of conservative dentistry: JCD*. 2014 May;17(3):234.
46. Webber RT, Carlos E, Brady JM, Segall RO. Sealing quality of a temporary filling material. *Oral Surgery, Oral Medicine, Oral Pathology and Oral Radiology*. 1978 Jul 1;46(1):123-30.
47. Pisano DM, DiFiore PM, McClanahan SB, Lautenschlager EP, Duncan JL. Intraorifice sealing of gutta-percha obturated root canals to prevent coronal microleakage. *Journal of endodontics*. 1998 Oct 1;24(10):659-62.
48. Wolcott JF, Hicks ML, Himel VT. Evaluation of pigmented intraorifice barriers in endodontically treated teeth. *Journal of endodontics*. 1999 Sep 1;25(9):589-92.
49. Sauáia TS, Gomes BP, Pinheiro ET, Zaia AA, Ferraz CC, Souza-Filho FJ. Microleakage evaluation of intraorifice sealing materials in endodontically treated teeth. *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*. 2006 Aug 1;102(2):242-6.

50. Veríssimo DM, do Vale MS. Methodologies for assessment of apical and coronal leakage of endodontic filling materials: a critical review. *Journal of oral science*. 2006;48(3):93-8.
51. Mayer T, Eickholz P. Microleakage of temporary restorations after thermocycling and mechanical loading. *Journal of Endodontics*. 1997 May 1;23(5):320-2.
52. Yavari H, Samiei M, Eskandarinezhad M, Shahi S, Aghazadeh M, Pasvey Y. An in vitro comparison of coronal microleakage of three orifice barriers filling materials. *Iranian endodontic journal*. 2012;7(3):156.
53. Wimonchit S, Timpawat S, Vongsavan N. A comparison of techniques for assessment of coronal dye leakage. *Journal of Endodontics*. 2002 Jan 1;28(1):1-4.
54. Chandra SS, Shankar P, Indira R. Depth of penetration of four resin sealers into radicular dentinal tubules: a confocal microscopic study. *Journal of endodontics*. 2012 Oct 1;38(10):1412-6.
55. Ma J, Wang Z, Shen Y, Haapasalo M. A new noninvasive model to study the effectiveness of dentin disinfection by using confocal laser scanning microscopy. *Journal of endodontics*. 2011 Oct 1;37(10):1380-5.
56. Vadhana S, Latha J, Velmurugan N. Evaluation of penetration depth of 2% chlorhexidine digluconate into root dentinal tubules using confocal laser scanning microscope. *Restorative dentistry & endodontics*. 2015 May 1;40(2):149-54.





57. Torabinejad M, Smith PW, Kettering JD, Ford TR. Comparative investigation of marginal adaptation of mineral trioxide aggregate and other commonly used root-end filling materials. *Journal of Endodontics*. 1995 Jun 1;21(6):295-9.
58. Torabinejad M, Chivian N. Clinical applications of mineral trioxide aggregate. *J Endod* 1999;25:197-205.
59. Zakizadeh P, Marshall SJ, Hoover CI, Peters OA, Noblett WC, Gansky SA, Goodis HE. A novel approach in assessment of coronal leakage of intraorifice barriers: a saliva leakage and micro-computed tomographic evaluation. *Journal of Endodontics*. 2008 Jul 1;34(7):871-5.
60. Beckham BM, Anderson RW, Morris CF. An evaluation of three materials as barriers to coronal microleakage in endodontically treated teeth. *Journal of endodontics*. 1993 Aug 1;19(8):388-91.
61. Zaia AA, Nakagawa R, De IQ, Gomes BP, Ferraz CC, Teixeira FB, Souza-Filho FJ. An in vitro evaluation of four materials as barriers to coronal microleakage in root-filled teeth. *International Endodontic Journal*. 2002 Sep;35(9):729-34.
62. Malik G, Bogra P, Singh S, Samra RK. Comparative evaluation of intracanal sealing ability of mineral trioxide aggregate and glass ionomer cement: An in vitro study. *Journal of conservative dentistry: JCD*. 2013 Nov;16(6):540.
63. Asgary S, Ahmadyar M. Vital pulp therapy using calcium-enriched mixture: An evidence-based review. *Journal of conservative dentistry: JCD*. 2013 Mar;16(2):92.
64. Moradi S, Disfani R, Ghazvini K, Lomee M. Sealing ability of orthograde MTA and CEM cement in apically resected roots using bacterial leakage method. *Iranian endodontic journal*. 2013;8(3):109.

65. Haghgoo R, Arfa S, Asgary S. Microleakage of CEM cement and ProRoot MTA as furcal perforation repair materials in primary teeth. *Iranian endodontic journal*. 2013;8(4):187.
66. Asgary S, Nosrat A, Seifi A. Management of inflammatory external root resorption by using calcium-enriched mixture cement: a case report. *Journal of endodontics*. 2011 Mar 1;37(3):411-3.
67. Nosrat A, Seifi A, Asgary S. Regenerative endodontic treatment (revascularization) for necrotic immature permanent molars: a review and report of two cases with a new biomaterial. *Journal of endodontics*. 2011 Apr 1;37(4):562-7.
68. Asgary S, Eghbal MJ, Parirokh M, Ghoddusi J, Kheirieh S, Brink F. Comparison of mineral trioxide aggregate's composition with Portland cements and a new endodontic cement. *Journal of endodontics*. 2009 Feb 1;35(2):243-50.
69. Samiee M, Eghbal MJ, Parirokh M, Abbas FM, Asgary S. Repair of furcal perforation using a new endodontic cement. *Clinical oral investigations*. 2010 Dec 1;14(6):653-8.
70. Asgary S, Shahabi S, Jafarzadeh T, Amini S, Kheirieh S. The properties of a new endodontic material. *Journal of endodontics*. 2008 Aug 1;34(8):990-3.
71. Utneja S, Nawal RR, Talwar S, Verma M. Current perspectives of bio-ceramic technology in endodontics: calcium enriched mixture cement-review of its composition, properties and applications. *Restorative dentistry & endodontics*. 2015 Feb 1;40(1):1-3.

72. Ghazvini SA, Tabrizi MA, Kobarfard F, Baghban AA, Asgary S. Ion release and pH of a new endodontic cement, MTA and Portland cement. *Iranian endodontic journal*. 2009;4(2):74.
73. Bhatia C, Chandak M, Adwani D, Chandak R, Rahul A. Calcium enriched mixture cement. *Dent. Health Sci*. 2015;2(4):905-10.

ANNEXURE – I

ETHICAL CLEARANCE CERTIFICATE

 KLE UNIVERSITY www.kleu.ac.in	Research and Ethics Committee KLE V K INSTITUTE OF DENTAL SCIENCES KLE University Accredited 'A' Grade by NAAC Placed in Category 'A' by MHRD (Govt) Nehru Nagar, Belagavi - 590 010, Karnataka State ☎: 0831-2470362 Web: http://www.kledental-bgm.edu.in FAX: 0831-2470640 E-mail: principal@kledental-bgm.edu.in	 KLE UNIVERSITY www.kleu.ac.in
SI. No. : 1390		
CERTIFICATE		
<p><i>This is to Certify that the synopsis titled</i></p> <p><u>Comparative evaluation of intra orifice sealing ability of Normtra Plus, Calcium enriched mixture and Conventional glass ionomer cement under Confocal Laser scanning microscope - An In vitro study. Submitted by</u></p> <p><u>Dr. Felbin Tomy</u> P. G. Student /</p> <p><u>Staff, Guided by Dr. Sunita Shivnanand</u> from Department of</p> <p><u>Conservative dentistry & Endodontics</u> has been critically evaluated by</p> <p>committee members and granted ethical clearance to conduct the above</p> <p>mentioned study</p>		
<p>Date :</p>		
 Member Secretary Research and Ethical Committee KLEVK Institute of Dental Sciences Belagavi	 Chairman Research and Ethical Committee KLEVK Institute of Dental Sciences Belagavi	

ANNEXURE – II

BIostatistic CLEARANCE CERTIFICATE



KLE V.K. Institute of Dental Sciences

(A Constituent unit of KLE Academy of Higher Education & Research
Deemed-to-be-University u/s 3 of the UGC Act, 1956)
Nehru Nagar, Belagavi-590 010 INDIA

Re-Accredited 'A' grade by NAAC (2nd Cycle) & Placed in Category 'A' by MHRD (GoI)

☎: 0831-2470362
FAX: 0831-2470640

Web: <http://www.kledental-bgm.edu.in>
E-mail: principal@kledental-bgm.edu.in




Biostatistics Clearance Certificate

This is to certify that the Biostatistics aspect of the Dissertation / Research work of....*Dr. Felbin Tomy*.....




entitled...*Comparative evaluation of intra-orifice sealing ability of Neo-MTA plus, Calcium Enriched Mixture and conventional Glass Ionomer cement under confocal laser scanning microscope - An In vitro study*
has been done under my guidance and considered satisfactory.

Place : Belagavi
Date : 22/09/2020 .


Name & Signature of Biostatistician
(Dr. S. B. Javali)

ANNEXURE – III

PLAGIARISM CHECK CERTIFICATE

Scientific Correspondence and Review Committee	
KLE VK Institute of Dental Sciences	
	A Constituent Unit of KLE Academy of Higher Education and Research (Deemed-to-be-University u/s 3 of the UGC Act, 1956) Nehru Nagar, Belagavi - 590 010, Karnataka State Accredited 'A' Grade by NAAC (2nd Cycle) Placed in Category 'A' by MHRD (GoI)
☎: 0831-2470362 FAX: 0831-2470640	Web: http://www.kledental-bgm.edu.in E-mail: principal@kledental-bgm.edu.in
Date : 28-9-2020	Serial No. : 045
PLAGIARISM CHECK REPORT	
Name of the Applicant : <i>Dr. Felbin Jomy</i> UG / PG / Ph.D / Staff : <i>Post Graduate</i> Batch & Year : <i>2018-2021</i> Department : <i>Conservative dentistry and Endodontics</i>	
The soft copy of Research Work / Manuscript by <i>Dr. Felbin Jomy</i> entitled <i>Comparative evaluation of extra-orifice sealing ability of</i> <i>"res-MTA plus; CEM and conventional GIC after obturation of</i> <i>root canal system under confocal laser scanning microscope</i> <i>- An in vitro study.</i> under the guidance of has been submitted for Anti-Plagiarism check to the Scientific Correspondence & Review Committee of KLE VK Institute of Dental Sciences using "Turn-it-in" software.	
The scan has been carried out and the scanned output reveals a Similarity Index of <i>10</i> %, which is within / not within the acceptable limits of 10% as per the UGC guidelines.	
 Member Secretary Scientific Correspondence and Review Committee KLEVK Institute of Dental Sciences KAHER-Belagavi	 Chairman Scientific Correspondence and Review Committee KLEVK Institute of Dental Sciences KAHER - Belagavi



Introduction



Aim and Objectives



Hypothesis



Review of Literature



Methodology



Results



Discussion



Conclusion



Summary



Bibliography



Annexures
