

**A Comparative Study of the Sealing Ability and Biocompatibility  
of New Modified Glass-Ionomer/Collagen Hybrid versus Mineral  
Trioxide Aggregate and Glass Ionomer as Root-End Filling  
Materials**

A thesis  
submitted to the  
Faculty of Oral and Dental Medicine  
Cairo University

In Partial Fulfillment  
Of the Requirements of  
Master Degree in Endodontics

By  
Hanan Mohammed Nabil Mahmoud  
B.D.S. Cairo University 2004

2011

**Cairo University**  
**Faculty of Oral and Dental Medicine**  
**Department of Endodontics**

## **THESIS APPROVAL FORM**

This thesis titled “A Comparative Study of the Sealing Ability and Biocompatibility of New Modified Glass-Ionomer/Collagen Hybrid versus Mineral Trioxide Aggregate and Glass Ionomer as Root-End Filling Materials” submitted by the student “Hanan Mohamed Nabil Mahmoud” has been approved by the following committee:

Faculty principle supervisor

Dr. Ghada El-Hilaly Eid

Date/

Faculty Examiner

Dr. Maged Negm

Date/

External Examiner

Dr. Naguib Mahmoud Aboul Enein

Date/

# **SUPERVISORS**

## **Dr. Ghada El-Hilaly Eid**

Associate Professor of Endodontics

BDS, MSc, PhD (Cairo University)

## **Dr. Reham Mohamed Said Siam**

Lecturer in Endodontics

BDS, MSc, PhD (Cairo University)

## **Dr. Safaa Khalil Hassan**

Associate Professor of Biophysics, BDS, MSc, PhD

Spectroscopy department, Physics division

National Research Center

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

" إِنَّ الَّذِينَ أُوتُوا الْعِلْمَ مِنْ قَبْلِهِ إِذَا يُتْلَى عَلَيْهِمْ  
يَخِرُّونَ لِلْأَذْقَانِ سُجَّدًا . وَيَقُولُونَ سُبْحَانَ رَبَّنَا  
إِنْ كَانِ وَعْدُ رَبَّنَا لَمَفْعُولًا "

صَدَقَ اللَّهُ الْعَظِيمِ

الاسراء (١٠٧-١٠٨)

## ACKNOWLEDGMENT

*First and foremost, countless thanks to my everlasting support*

**“ALLAH”**

*It is a great honor to express my appreciation to **Dr. Ghada El-Hilaly Eid** Associate Professor of Endodontics, Cairo University, for teaching me how to achieve accurate and valuable work, how to read and understand scientific paper in a right way, how to enjoy the research work and indirectly, how to know my aim and what I am searching for.*

*I wish to express my deep thanks to **Dr. Reham Mohamed Said Siam** lecturer of Endodontics, Cairo University, for believing in my idea through this research and for her flexible, amazing way to guide me into the scientific information.*

*I wish to thank **Dr. Safaa Khalil Hassan** Associate Professor of Biophysics, Spectroscopy department, National Research Center, for being kind, supportive, for always keeping her door open to me, for the wonderful time I spent in the laboratory and finally for believing in the importance of “team work”.*

*I would like to express my deep appreciation to **Dr. Hend Mahmoud Abu El Nasr** lecturer of Endodontics, Cairo University, **Dr. Manal Mohammed Hosni** Professor of Oral Medicine and Periodontology, Cairo University, to my friend **Dr. Ranya Abd Al-Moaty** for guiding me in the “oral pathology world” and to my intimate friend **Dr. Rasha Attia** for always being there whenever I needed and for taking all the photographs in this research.*

## DEDICATION

*To those who shape my mind, educate me, believe in me  
....To my teachers*

*To the man who teaches me how to thank ALLAH, the true  
meaning of satisfaction and honor  
....To my father*

*To the woman who shows me the beauty of life, gives me the  
courage to follow my heart, inspires me, and pushes me  
always to success  
....To my mother*

*To the one who supports me through distance  
....To my brother*

*Finally, to those who share me the road, lightening my life  
with their friendship  
....To Rasha, Ranya, Sarah, Hend, Abeer, Wafaa*

*If anything is good about me, it is because of you all*

*Thanks, I owe you a lot*

## LIST OF CONTENTS

No.	Title	Page
	LIST OF TABLES.....	viii
	LIST OF FIGURES.....	ix
	LIST OF ABBREVIATIONS.....	xi
I	INTRODUCTION.....	1
II	REVIEW OF LITERTURE.....	3
	A) Conservative endodontic treatment versus surgical treatment.....	3
	B) Root-end filling materials.....	5
	1) MTA as a root-end filling material.....	6
	2) Glass Ionomer as a root-end filling material.....	7
	3) Enhancing glass ionomer by collagen integration.....	7
	C) Evaluation of the root-end filling materials.....	11
	1) Biocompatibility evaluation.....	11
	a) Biocompatibility of MTA as a retrograde filling material.....	12
	b) Biocompatibility of GI as a retrograde filling material.....	19
	2) Apical leakage evaluation.....	23
	3) Scanning electron microscope examination of retrograde filling Materials.....	33
III	AIM OF STUDY.....	38
IV	MATERIALS AND METHODS.....	39
	A) Preparation of Glass Ionomer /Collagen hybrids in two percentages (0.01% -1%).....	41
	1) Integration of collagen to glass ionomer (GI) liquid.....	41
	2) Mixing of the glass ionomer powder with the GI liquid containing collagen to form GI/collagen hybrid material.....	42
	B) In vivo Biocompatibility Evaluation.....	45
	C) Dye Extraction-Apical leakage Evaluation .....	54
	D) Scanning Electron Microscope Examination.....	62
	E) Statistical Analysis.....	64
V	RESULTS.....	65

<b>No.</b>	<b>Title</b>	<b>Page</b>
	A) In Vivo Biocompatibility Evaluation.....	65
	B) Dye Extraction- Apical leakage Evaluation.....	82
	C) Scanning Electron Microscope Examination.....	83
VI	DISCUSSION.....	92
VII	SUMMARY AND CONCLUSIONS.....	105
VIII	RECOMMENDATIONS.....	108
IX	REFERENCES.....	109
X	ARABIC SUMMARY.....	



## LIST OF TABLES

Table No.	Title	Page
1	Materials and equipments used in the present study.....	39
2	Mean, standard division (SD), median, <i>P</i> -values and score description for MTA in two observation periods.....	67
3	Mean, standard division (SD), median, <i>P</i> -values and score description for GI in two observation periods.....	69
4	Mean, standard division (SD), median, <i>P</i> -values and score description for GI/C 0.01% in two observation periods.....	71
5	Mean, standard division (SD), median, <i>P</i> -values and score description for GI/C 1% in two observation periods.....	73
6	Mean, standard deviation, <i>P</i> -value and scores description for all four test materials at 7-day observation periods.....	77
7	Mean, standard deviation, <i>P</i> -value and scores description for all four test materials at 60-day observation period.....	80
8	Statistical analysis for apical leakage between all groups.....	82

## LIST OF FIGURES

Figure no.	Title	Page
1	Flow chart showing general research outline.....	40
2	Collagen (ETIK™ Collagène) and Glass Ionomer (Ketac™ Cem) .....	43
3	Integration of collagen to glass ionomer liquid.....	44
4	The experimental animals in the cage.....	45
5	Flow chart representing the biocompatibility experimental protocol.....	46
6	Polyethylene tubes loaded with test materials and control tube.....	48
7	Intramuscular anesthesia injection to the rat.....	49
8	Steps of surgical procedure for biocompatibility evaluation of Wistar Albino rats.....	51
9	Retrograde cavity preparation using diamond coated retrograde tip no E32D mounted in the ultrasonic device.....	56
10	Representative samples showing radiographs after retrograde cavity preparation, stereomicroscopic images after root resection and after retrograde cavity preparation.....	56
11	Representative radiographs and stereomicroscopic images after retrograde Filling by MTA, GI, GI/C 0.01%, GI/C 1%.....	59
12	The steps of the dye extraction method.....	61
13	The steps of gold sputtering of the samples.....	63
14	Stereomicroscopic images for four test materials after gold coating.....	63
15	The Scanning Electron Microscope.....	64
16	Microscopic images for the control empty tube of the 7-day observation period in two different samples.....	66
17	Microscopic images for the control empty tube of the 60-day observation period in two different samples.....	66
18	Microscopic images for histological changes of MTA in two observation periods.....	68
19	Microscopic images for histological changes of GI in two observation Periods.....	70

<b>Figure no.</b>	<b>Title</b>	<b>Page</b>
20	Microscopic images for histological changes of GI/C 0.01% in 7-day observation period in two different samples.....	72
21	Microscopic images for histological changes of GI/C 0.01% in 60-day observation period in two different samples.....	72
22	Microscopic images for histological changes of GI/C 1% in 7-day observation period in two different samples.....	74
23	Microscopic images for histological changes of GI/C 1% in 60-day observation period in two different samples.....	74
24	Bar chart representing median inflammatory scores at 7 days and 60 days for all test materials.....	75
25	Bar chart representing mean inflammatory scores at 7-day observation period for all four test materials.....	77
26	Microscopic images for histological changes of the four test materials for 7-day observation period.....	78
27	Bar chart representing mean inflammatory scores at 60 days for all four test materials.....	80
28	Microscopic images for histological changes of the four test materials for 60-day observation periods.....	81
29	Bar chart representing mean leakage of all groups.....	82
30	Scanning micrographs for MTA at three different magnifications.....	84
31	Scanning micrographs for glass ionomer at two different magnifications.....	85
32	Scanning micrographs for GI/C 0.01% at three different magnifications.....	87
33	Environmental scanning micrographs for GI/C 0.01% at four high different magnifications.....	88
34	Scanning micrographs for GI/C 1% at three different magnifications.....	90
35	Environmental scanning micrographs of GI/C 1% at four high different magnifications.....	91

## LIST OF ABBREVIATIONS

---

<b>Abbreviation</b>	
AFM	Atomic Force Microscopy.
BHI	Brain Heart Infusion.
BII	Blue India Ink.
CER	Cimento Endodôntico Rápido (fast endodontic cement).
CFP	Capillary Flow Porometry.
CHX	Chlorhexidine.
EBA	Ethoxy Benzoic Acid.
ECMs	Extracellular matrices.
ESEM	Environmental Scanning Electron Microscope.
FTM	Fluid Transport Model.
GI	Glass Ionomer.
GI/C 0.01%	Glass Ionomer/Collagen hybrid 0.01%.
GI/C 1%	Glass Ionomer/Collagen hybrid 1%.
H&E	Hematoxylin & Eosin stain.
HEMA	Hydroxyethyle methacrylate.
IRM	Intermediate Restorative Material.
MTA	Mineral Trioxide Aggregate.
Na <sub>2</sub> HPO <sub>4</sub>	Disodium Hydrogen Phosphate.
OP	Optical Density.
PC	Portland Cement.
PDL	Periodontal Ligament.
SEM	Scanning Electron Microscope.
WMTA	White Mineral Trioxide Aggregate.
ZOE	Zinc Oxide Eugenol.

---

## **I) INTRODUCTION**

Endodontic surgery is an important adjunct to conservative root canal treatment and is sometimes the only option for treating some endodontic conditions. Resection of the root end during periradicular surgery results in an exposed apical dentine surface bounded by cementum with central root canal. Following retrograde cavity preparation, a retrograde filling material is used to seal the root-end cavity. Various root-end filling materials include amalgam, resin reinforced zinc oxide-eugenol cements and glass ionomer cement (GI). Nowadays, Mineral Trioxide Aggregate (MTA) is considered a standard for root end-filling materials.

Glass ionomer is a bioactive tooth adhesive material. Despite the strong chemical bond between the GI and tooth structure, the material is brittle and sensitive to moisture during setting. Several modifications have been made to GI in order to improve its properties. Silver particles were mixed with GI powder in order to improve strength and wear resistance of the material. Collagen is the most abundant extracellular matrix component in the human body and periodontal tissue. It serves as the matrix component which may enhance the mechanical properties, allow cell binding and proliferation which starts the repair process and promote tissue interface biocompatibility. The integration of collagen to GI might enhance compatibility, promote wound healing and raise the possibility of using it as a retrograde filling material.

As the materials used in surgical endodontics are placed in intimate contact with the hard and soft tissues of periodontium, root-end filling materials must be tested for biocompatibility. The capacity of sealing the

apical region is an important aspect when choosing a root-end filling material, aiming to prevent apical microleakage, i.e., the passage of bacteria, fluid, molecules or ions between the cavity wall and the filling material. The quality of apical seal obtained by root-end filling materials can be assessed by the dye penetration, radioisotopes, bacteria leakage or electro-chemical means as well as fluid filtration technique and dye extraction method. The efficiency of marginal adaptation and the interface between tooth structure and restorative materials should also be evaluated.

To accept the possibility of using glass ionomer/collagen hybrids as a retrograde filling material, the biocompatibility, sealing ability and marginal adaptation, still need to be investigated.

## II) REVIEW OF LITERATURE

### A) Conservative endodontic treatment versus surgical treatment

For many years the term conservative treatment has been used as a synonym for nonsurgical treatment. The root canal system has the capacity to harbor several species of microorganism with their toxins and byproducts. Egress of such irritants from the root canal system into the periapical tissue results in the formation of the periapical lesions. Removal of these irritants and three dimensional obturation of root canal system are the major goal of conservative root canal system. <sup>(1)</sup> *Johnson (1999)* discussed clinical evidences of success as follows: (1) absence of symptoms; (2) absence of swelling, sinus tract, and other signs of infection; (3) radiographic evidence of healing; and (4) continued normal functioning of the tooth. <sup>(2)</sup>

Although conservative endodontic treatment is successful in most cases, sometimes shifting to surgical approach is necessary. <sup>(2)</sup> The decision to shift to endodontic surgery may be indicated in the following situations: if there is a strong possibility of failure from nonsurgical treatment, if failure has resulted from nonsurgical endodontic treatment, and retreatment is impossible or would not achieve a better result and if a biopsy is necessary at or near the tooth apex. <sup>(3)</sup>

Steps of endodontic surgery consist of exposure of the involved area, root-end resection (apicoectomy), the preparation of retrograde cavity and finally, the insertion of root-end filling material.