A Comparative Evaluation of Fracture Resistance of Simulated Immature Teeth Using Different Obturating Material: An In-Vitro Study.

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***Abstract*- Aim –**To compare and also evaluate the fracture resistance of immature teeth using different obturating materials like MTA Angelus and Biodentine

Materials and Method: Fifty freshly extracted single rooted human mandibular premolar teeth were used for the study which were decoronated at cementoenamel junction and divided into five groups (*n* = 10per group). Group 1: Samples served as negative control. Group 2 :MTA Angelus apical plug and then backfilling by gutta-percha. Group 3: Filling of root canal system entirely by MTA Angelus. Group 4 : Apical plug of biodentine and backfilling with gutta-percha. Group 5: Root canals completely filled with Biodentine. In four experimental group samples were shaped, cleaned and prepared using ProTaper rotary files. For simulation of immature roots, a #5 Peeso reamer was stepped out from the apex so that apices were enlarged to a diameter of 1.5mm. Group 2 and Group 4 samples were then filled with 5 mm of MTA angelus or Biodentine apical plug and backfilling with gutta-percha using AH Plus sealer. Group 3 and Group 5 rootcanal system samples were completely obturated with MTA Angelus and Biodentine, respectively. All the samples were loaded vertically until root samples fractured with the help of universal testing machine. Statistical Analysis: SPSS 23.0 version software was used for statistical analysis of forces at which fracture of the roots occurred and the results were analyzed with the one-way analysis of variance and *post hoc tukey* test.
Results:In our study, root canal obturation which was done completely with MTA Angelus or Biodentine showed significantly higher fracture resistance ($P<0.05$) when compared to apexification done with MTA or Biodentine. Conclusion: Obturation of the root canals with bioactive materials showed highest fracture resistance when compared to apexification groups.

***Index Terms*-** Apexification, biodentine, mineral trioxide aggregate angelus, universal testing machine, vertical root fracture.

I. Introduction

Thickess of dentin is one of the most vital aspects determining the resistance of teeth to fracture. The tissue loss of tooth reduces the fracture resistance toward occlusal or traumatic forces.[1]The most common site of dental impact injuries in the developing dentition is the maxillary anterior teeth.[2-4] These injuries many times lead to pulpal necrosis, which might cause the termination of root formation in developing teeth.[4-6] It has been stated that the immature teeth which have been endodontically treated have a relatively high incidence (>60%) of cervical root fracture, either spontaneously or even due to minor impacts.[7]

The endodontic treatment of teeth with immature root formation has been a challenge due to wide, open apices and thin dentinal walls.[4,8]Various procedures and materials have been recommended to induce apexification in teeth with immature apexes.[9]Management of openapexes in immature teeth has been accomplished using long-term CH therapy, with success rates ranging from79% to 96%.[10,11]However, these teeth showed a50% reduction in strength vs the controls over 1 year[8] and were compromised by cervical root fractures[7,12] because of changes in the organic matrix of the dentin.

Calcium hydroxide applied to the root canal system to promote the formation of an apical barrieris the conventional treatment in these clinical situations.[8,13,14]However, drawbacks of the long-term calcium hydroxide treatment requires multiple visits, patient adaptation problem, microleakage between the visits and an enhanced risk of root fractures. Hence, other alternatives to Ca $(OH)_2$ have been proposed,of which, the most promising are calcium silicate-based materials, such as mineral trioxide aggregate (MTA) and Biodentine. These root-end repair materials have been claimed to be biocompatible, capable of stimulating biomineralization, andalso offer a superior seal with better bond strength.[15]

MTA has ingredients like tricalcium oxide and other mineral oxides such as tricalcium silicate, silicate oxide, and tricalcium oxide.[16] MTA is biocompatible, less cytotoxic, possess antimicrobial properties, offers low microleakage and can set in presence of blood and moisture.[17,18]Although MTA is a suitable material for clinical use, it shows some disadvantages such as a prolonged time for setting, difficulty in handling, and the probability of discoloration.[19]

Biodentine is a silicate-based biologically active cement that has dentin-like mechanical properties and designed as a "dentin replacement" material. This is formulated using MTA-based cement machinery, also improving its physical and mechanical properties.[20]

Establishment of a proper fracture resistance to the root dentin when obturated with various obturating materials, is an important key point for clinical success. Based on these observations, the aim of this *in vitro* study was to compare and evaluate the influence of MTA angelus and Biodentine as obturating materials on the fracture resistance of immature teeth.

II. Materials and Method

Fifty noncarious, human single-rooted mandibular premolar teeth were selected for the study. Then, each experimental tooth was decoronated from the cementoenamel junction (CEJ) using a flexible diamond disk (Novo Dental Products, Mumbai, India) in a slow-speed handpiece under a copious amount of water. The root of each tooth was standardized. The length of each root was 12 mm as measured from the apex to facial CEJ.

The fifty root samples were randomly divided into five groups:

Group 1 (*n* = 10): Sound roots with no root canal treatment (negative control)

Group 2 (*n* = 10): MTA Angelus apical plug and backfilling by gutta-percha

Group 3 (*n* = 10): Filling of root canal system entirely by MTA Angelus

Group 4 (*n* = 10): Biodentine apical plug, and backfilling by gutta-percha

Group 5 (*n* = 10): Root canals completely filled with Biodentine

For all the test root samples, coronal access was prepared using #245 bur in a high-speed handpiece and the canal patency was checked by probing with endodontic explorer DG-16 and #10 K-file (DentsplyMaillefer, Switzerland). The working length was determined followed by cleaning and shaping of the root canals which was completed with ProTaper rotary Ni-Ti files (Dentsply Maillefer, Switzerland) upto F3(#30/09) at a speed of 300 rpm using an X smart plus endomotor (DentsplyMaillefer, Switzerland). The canals were irrigated with 2 ml of 3% NaOCl after every instrumentation using 27-gauge side vented needle and syringe. #1–5 Peeso reamers were used for simulation of immature roots with open apexes. Then, a #5 Peeso reamer waspassed beyond the apex so that apices were enlarged to a diameter of 1.5 mm. Following cleaning and shaping, all the root canals were irrigated with 5 ml of normal saline. Before the obturation, the rootcanals in all the root samples were irrigated with 17% ethylenediamine tetraacetic acid (EDTA)(Canalarge, Ammdent, Mohali, India) for 1 min to remove the smear layer. Final flush was done with5 ml of normal saline and excess moisture from the canals was removed with sterile absorbent points.

In Group 2 root samples, MTA angelus (Angelus, Londrina,Brazil) mix was placed into the canal with MTA messing gun and advanced apically with endodontic pluggers of different sizes 9/11, 5/7, 1/3(Dentsply Maillefer, Germany) and 5 mm thickness of the apical plug was maintained. After 24 hour, remaining part of the root canals was obturated with F3 master gutta-percha point and AH Plus sealer.

In samples of Group 3, 5 mm of MTA apical barrier was placed and then the remaining canal was filled with MTA up to 1 mm short of CEJ. In root samples of Group 4, an apical plug of 5 mm with Biodentine and backfilling with gutta-percha and AH Plus sealer was done. In Group 5, Biodentine apical plug was prepared, and then immediately, remainder of coronal part of the canal was filled with Biodentine. For all the experimental root samples, post obturation radiographs were taken in both labiolingual and mesiodistal directions to ensure homogeneous and adequate root filling without voids. The filled roots were stored in an incubator for 1 month at 37°C and 100% relative humidity for allowing obturating materials to set completely. Acrylic resin blocks with 10 mm height and 20 mm width were prepared. All the roots were embedded in a vertical direction in these blocks with a distance of 2 mm between the top of the acrylic and the cement – enamel junction.

III. Fracture Resistance Evaluation

Fracture resistance was measured by universal testing machine. A cylindrical ball indenter of 2.2mm diameter with a sharpened conical tip was attached to the upper part of universal testing machine to apply force to the root causing vertical root fracture. The root was placed under the plunger on the lower platen, and the plunger was driven downward exactly along the long axis of the root. A vertical load was applied at a crosshead speed of 0.5 mm/min until the root fractured. The maximum load at the time of fracture recorded in Newtons was then converted into Megapascals

(MPA) using the formula:

$$\text{MPa} = \frac{\text{Maximum load in Newtons (N)}}{\pi/4\,(\text{Area of cross-section of ball indendator})^2}$$

Where $\pi = 3.14$ (constant value)

IV. Statistical Analysis

All the collected data about forces at which fracture of teeth occurred in MPa were tested to statistical analysis using SPSS 23.0version IBM (USA). A one-way analysis of variance was used to compare the values of forces at which the fracture of roots obturated with different materials occurred. Post Hoc Tukey's HSD test was carried out to assess whether the mean difference between a pair of group is significant or not. A p value of <0.05 was considered as statistically significant where as a p value <0.001 was considered as highly significant.

V. Results

The mean fracture resistance value and standard deviation were obtained. Significant variations ($p<0.05$) between the groups were observed in ANOVA test and also Post hoc Tukey analysis which were used to determine significant differences among experimental and control groups. The mean fracture resistance value was higher in Group 1 (control group, without instrumentation)when compared to experimental groups with statistically significant difference ($P < 0.05$) [Table 1]. When experimental Group 2 compared to Group 3, Group 4 and Group 5 show significant difference while Group 3 when compared to Group 5 show not significant results.(Table 2)

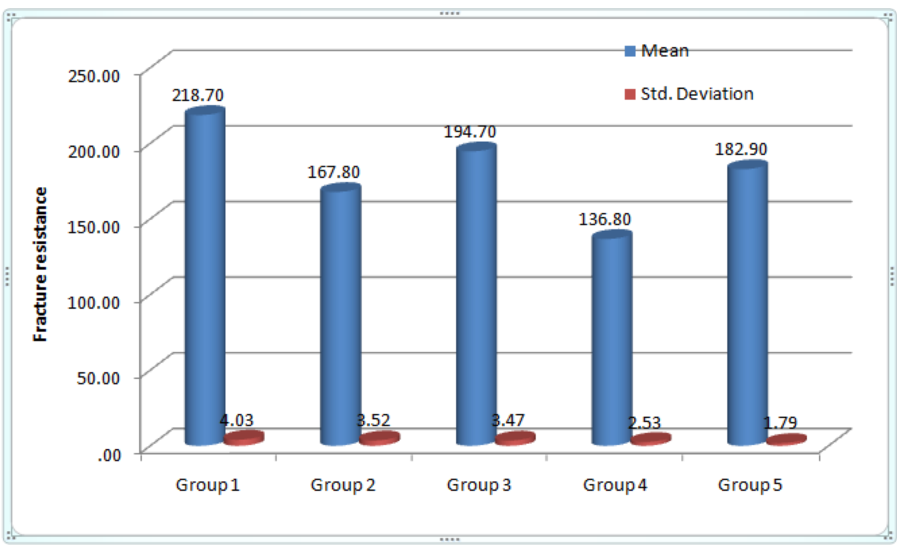
TABLE 1
Comparison of -Mean, Standard Deviation and P-value of fracture resistance values in MPa for experimental and control groups

	N	Mean	Std. Deviation	F	Df	P	Inference
Group 1	10	218.70	4.03	929.52	4	0.0001 (<0.001)	Highly significant
Group 2	10	167.80	3.52				
Group 3	10	194.70	3.47				
Group 4	10	136.80	2.53				
Group 5	10	182.90	1.79				
Total	50	180.18	27.78				

TABLE 2
Pair wise comparisons of five groups (1, 2, 3, 4, 5) with respect to fracture resistance (Mpa) by Post Hoc Tukey's HSD to see whether the mean difference between individual group is significant or not

	Group 2	Group 3	Group 4	Group 5
Group 1	50.9*	24.0*	81.9*	35.8*
Group 2		-26.9*	31.0*	-15.1*
Group 3			-57.9*	11.8
Group 4				-46.1*

*Indicates that the difference in the mean is significant at 0.05 level.



GRAPH 1 - Comparison of Mean, Standard Deviation and P-value of fracture resistance values in MPa for experimental and control groups

VI. DISCUSSION

The occurrence of trauma in the permanent dentition has been reported to range from 2.6% to 35% with the greater incidence occurring between the ages of 7 and 15 when most permanent teeth are in an incomplete root development stage. Unfortunately, approximately 50% of the traumatized teeth are diagnosed with pulpal necrosis and incomplete root formation.[21]Immature teeth have thin dentin walls and, thus, are more fragile than the mature teeth, posing difficult for the clinicians.[22] In spite of current feasibility of treatment of open apex, immature teeth remain very sensitive to fracture, especially in the cervical area.[23]Therefore, a material that reinforces strength must be selected in such cases, which should be easy to manipulate, prevent microleakage, removed easily when necessary and can adhere consistently to the dentin walls.[24] For such open apices, apexification involving induction of a calcific barrier at the apex with calcium hydroxide has been tried. The root-end closure procedure using calcium hydroxide is standardized but time-consuming and require on an average of 7–8 months for apical bridge formation. It is also associated with long treatment time and thus increasing the risk of failure of treatment[25]and decreases the fracture resistance of root dentin.[26]

Various materials including composite resins, resin-reinforced glass ionomers, resin-based root canalfillings (Resilon), different post systems and different root-end filling materials like MTA and BA have been used previously to reinforce the immature permanent teeth.[23,26,27]In this study, the efficiency of MTA angelus and Biodentine on root fracture resistance was evaluated and also compared in human simulated immature premolar teeth with predetermined and same diameter and length. When restoring immature teeth, single-visit apexification by using a MTA barrier offers several advantages over traditional apexification. These include lesser appointments for the patient and development of an immediate apical seal and less potential to weaken the tooth structure compared with long-term Ca $(OH)_2$. MTA offers a biologically active substrate for bone cells which stimulates interleukin production owing to its alkaline pH and calcium ion release. It also initiates the calcific bridge formation within first postoperative week.[16] But the procedure of apexification with MTA poses various disadvantages such as difficult handling characteristics, prolonged setting time, potential coronal discoloration, and less compressive strength as compared to dentine and Biodentine.[28]The roots were placed into acrylic block for homogeneous distribution of the force. The influence of the periodontium was not considered in this study. The root length was standardized to 12 mm, and the apex was enlarged using peeso reamers (No. 1–5).Stuart *et al*.,[29] Tanalp *et al.*,[14] and Seto *et al.*[30]had used asimilar method for preparation of root canals.

During apexification procedure, the most common disadvantage with gutta-percha usageas a backfilling material is microleakage.[31]Previous research has indicated that no known method with various techniques of cold or warm compaction of gutta-percha can predictably produce a coronal bacterial tight seal when the material is exposed to microorganisms and their by-products.[32]This has led to the current focus in endodontic research over exploring various alternatives to gutta-percha as obturating material.

Studies have proved that MTA apexification and backfilling with gutta-percha have less fracture resistance than MTA apexification and backfilling with different types of prefabricated intracanal metalposts or fiber posts.[14,33]One study has also stated that there is no statistical difference between MTAapexification with gutta-percha backfilling and complete root canal obturation with MTA.[22]

Considering these conflicting results, the present study compared the complete root canal obturation using Biodentine or MTA Angelus. Mandibular premolars were used which have a circular cross-section in the mid toapical region that would result in uniform distribution of load to fracture. They also simulate clinical situation better, where chewing forces are maximum. Some other studies have stated that smear layer might act as a coupling agent enhancing MTA bonding to root dentin.[34]However, recent studies have showed that the removal of smear layer must be done with 17% EDTA toimprove the sealing ability of MTA.[35] Hence, in the present study, 17% EDTA was used to removethe smear layer before obturation of canals.A 90° angle was applied for placement of the teeth into the testing machine as previously demonstrated by Tuna *et al*.[26]Although the force applied in ex vivo studies cannot completely simulate the clinical situations,standardizing the force in all of the study groups makes it possible to compare the strengthening effect of materials tested.

In present study, the mean bond strength values were higher for control group when compared to the experimental groups with a difference that was statistically significant. Contrary to these findings, a few studies concluded that control group exhibits less fracture resistance than the canals completely obturated with MTA.[16,36] Also, it appears that long-term (tested for 1 year period) placement of MTA in the canal system not just provides increased resistance to fracture but also increases the strength of the tooth with time. Researchers postulated that MTA might prevent the destruction of collagen by inducing the expression of a tissue inhibition of metalloproteinase-2 (TIMP-2) in the dentin matrix. According to histological analysis, expression of collagen type I, matrix metalloproteinase (MMP)-2, -14, and TIMP-2 on the dentin were noticed in MTA-treated teeth. TIMP-2 prevented the organic matrix from degradation caused by MMP-2,-14.[34] Hence, Hatibovic-Kofman *et al*.[37]stated that the reason for high fracture resistance of dentin at long-term MTA placement might lie in the inhibitor activities of TIMP-2 and reduced expression of MMP-2, -14 in the dentine matrix.

The mean fracture resistance values were higher for root canals obturated completely with MTA Angelus when compared to Biodentine. However, the difference was not statistically significant. This finding was in line with results of a study conducted by Elnaghy and Elsaka.[38] Biodentine has shown high compressive strength, reduced setting time (9–12 min), and less solubility and better handling characteristics as compared to MTA. A special feature of Biodentine is its capacity to continue improving the compressive strength with time over several days. It reaches up to 300 MPa after 1month, which is actually more than compressive strength of natural dentin (297 MPa).[39]

The mean fracture resistance values were higher for root samples of MTA angelus apexification group as compared to for root samples of Biodentine apexification group, and the difference between them was statistically significant. This can be due to the superior sealing ability of MTA angelus as compared to Biodentine.

The present mechanical testing was performed with greater effort to simulate the exact clinical conditions and to achieve standardization. However, *in vitro* conditions do not completely simulate *invivo* conditions. The teeth used in the present study may simulate the shape of immature teeth but do not simulate the tissue composition and physical properties. The ability of these materials to reinforce the strength of the immature teeth should be evaluated clinically.

VII. CONCLUSION

Complete obturation of root canals with bioactive materials (MTA Angelus/Biodentine) displayed improved fracture resistance of immature teeth when compared to apexification groups. Further *in vivo* and *invitro* studies with larger sample sizes are necessary to support or refute the efficacy of MTA Angelus and Biodentine as a root canal filling materials.

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