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Comparative Evaluation of root canal morphology of Human Mandibular permanent second molar using Cone-Beam Computed Tomography and Decalcification & Dye-Penetration Technique -an in vitro study.

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Abstract:
Aim: *This study aims to evaluate the root canal morphology of the human mandibular permanent second molar to determine the root canal morphology according to Vertucci's classification using Cone Beam Computed Tomography (CBCT) and Decalcification & Dye- Penetration technique. A comparative study also has been performed to analyze the accuracy of CBCT taking the Decalcification technique as the reference standard.*
Materials and Methods: *Root canal morphology of 100 (n=100) human permanent mandibular 2nd molars was evaluated by CBCT, then the teeth were subjected to Decalcification (by HNO3) and Die-Penetration (Indian Ink) processes for comparative evaluation of root canal morphology.*
Results: *The predominant root canal morphology for the mesial roots were Vertucci's type II; (45% through CBCT and 42% through Decalcification). For the distal root Vertucci's Type, I (67% through CBCT and 66% through decalcification) was predominant. There was no significant difference between the observed and the expected values of Decalcification & dye penetration technique and CBCT respectively(P>0.05).*
Conclusions: *Human Permanent Mandibular Second molars: usually are two-rooted teeth with three canals; mesial root canals are commonly Vertucci's Type II and distal root canals are commonly Vertucci's Type I. CBCT can be used in-vivo, effectively, and successfully for diagnosis and evaluation of prognosis of endodontic treatment.*
Keywords: *CBCT; decalcification; dye-penetration; root canal morphology; mandibular 2nd molar*

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I. Introduction

Teeth are an integral part of the stomatognathic system. Thus, maintenance of the teeth with sound health is of paramount importance. The basic philosophy in modern dentistry is the prevention and restoration of the functional and aesthetic components of the dental arches. Thus, the Dental Surgeon should know variations and types of the canal configuration of concerned teeth for the success of the endodontic treatment. John Ingle[1] has reported that high rates of failure were attributed to incomplete obturation of various canal spaces. A canal is often left untreated because the dentist fails to recognize its presence.

Among all teeth, Mandibular second molars are one of the most difficult teeth to negotiate because of their intricate root canal morphology due to the presence of an extra canal, curve canal, presence of radix, and C-shaped canal. Mandibular second molars may have one to six canals[2], although the most prevalent configurations are two roots (mesial and distal) and three canals; but four canals or one extra canal at the distal root is also common.

In the quest for root canal anatomy of mandibular permanent second molars, there are some methods had been used in vivo or in vitro, such as tooth sectioning methods[2,3] Decalcification & Die-penetration technique[2,4,8,9], Microscopy (DOM)[2], Intra Oral Peri-Apical X-Ray (IOPA)[2,17.] Stereo-Microscopy[2,25], Digital Radiography[2], Cone-beam computed tomography-(CBCT)[2,14] Micro-CT[2,14,25], peripheral quantitative computed tomography[2] etc. Decalcification & Die-penetration technique[2,4,8,9,11,12,13,15,] described by Barker et al. 1969[2,4] and Vertucci et al .1984[8.9]; was very much popular and still being used by various researchers successfully till date.

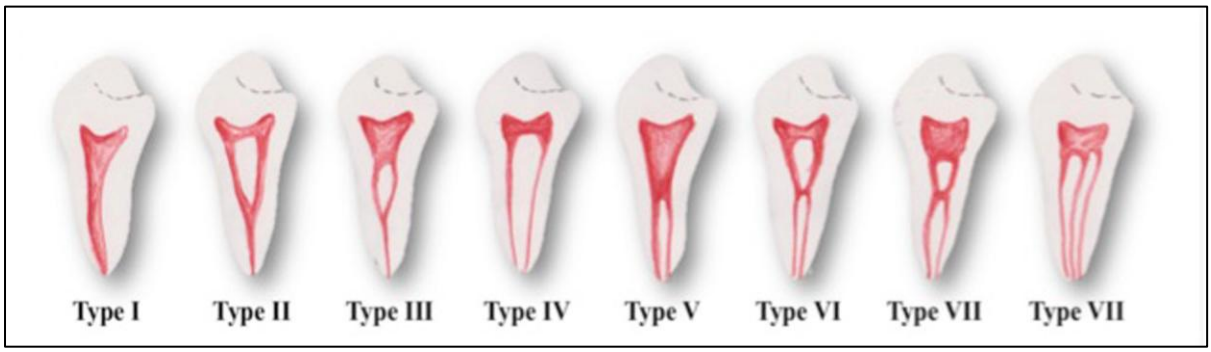
After the advent of digital radiography, CBCT and Micro-CT are considered useful techniques in-vitro as well as in vivo for assessing canal anatomy. CBCT provides three-dimensional information about a tooth in the sagittal, coronal and axial planes. Volume pixels or voxel, a three-dimensional (3D) unit is used like building blocks to form a larger 3D object in this CBCT technique.

In this study, assessment of the accuracy of the Cone Beam Computed Tomography (CBCT) technique, and determination of the root canal morphology of mandibular permanent second molar teeth, decalcification technique was done as the reference standard, and further, a comparative analytical study in-between those assessments on root canal morphology of mandibular permanent second molar has been performed.

There are various methods[4] to classify the root canal configuration depending upon morphological features. After Weine FS et al.[5,6 were], the first to categorize root canal configurations, Vertucci F.J [7,8], in 1984 introduced the root canal morphology classification into eight types.

Vertucci's classification[7,8,9] is most used and clinically practical to adopt and widely accepted for the classification of root canals in all teeth. So, this classification is used in the present study(Figure No. 1).

Figure No 1: Schematic diagrams showing the root canal configuration according to a Vertucci's classification[7,8]



II. Materials and Methods

100 human permanent mandibular 2nd molar teeth were freshly collected from the Department of Oral and Maxillofacial Surgery of the Dental College and Hospital. The inclusion criteria for the collection of the teeth samples were: having fully formed roots and root apex and with no fracture or loss of tooth structure in the root part of the tooth and exclusion criteria were: tooth with root anatomy having severe alteration, undergone root canal treatment, root resorption, and calcified canals. In this study at first CBCT scans of all the teeth were done sequentially, using the Carestream-CS 9300D CBCT machine, and images were examined with CS- 3D viewer software. The CBCT images were observed in different aspects -coronal, sagittal, and axial sections for evaluation of the root canal configuration based on Vertucci's canal classification.

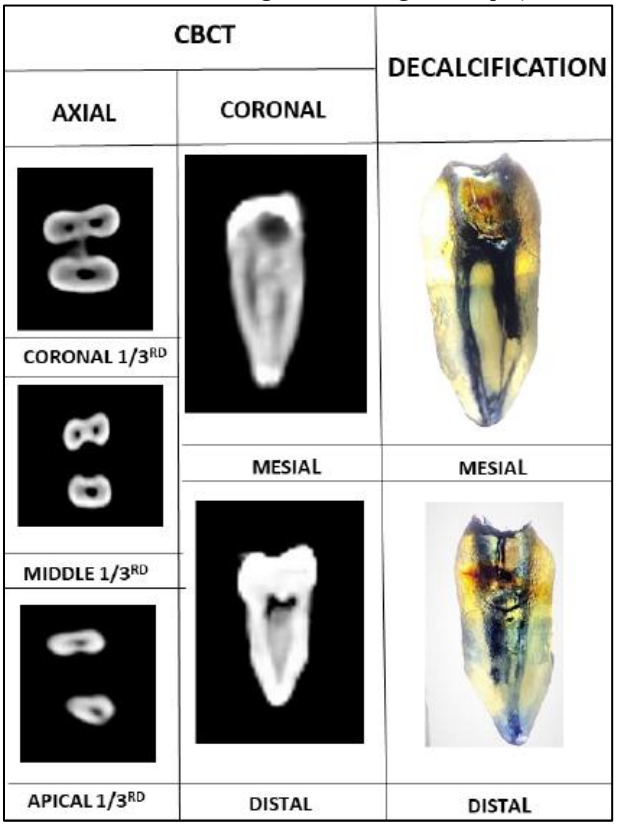
The access cavity was prepared by no 2 round TC burs on each tooth and root canals are negotiated with #10 and #15 K files up to the full length and the samples were placed in 5.25% Sodium hypochlorite(NaOCl) for 24 hours to dissolve organic debris.

After proper cleaning and air drying, Indian Ink was injected into the root canal of all the teeth through the access cavity with the help of a Luer-lock syringe with a 27-gauge needle. During injecting the dye, the root end of the sample was placed on a suction tip for proper distribution of dye under negative pressure.

Thereafter the teeth were placed in test tubes containing 10% Nitric Acid(HNO_3) for consecutive 2 days. Thereupon the samples were placed in 5% HNO_3 for the next consecutive 3 days for smoother Decalcification. Then samples, placed in test tubes containing HNO_3, were checked with a paper pin for their completion of decalcification. After achieving desired decalcification, the samples were thoroughly washed under tap water for 2-3 hours to remove traces of acid, then the samples were placed in the successive range of ethyl alcohol for dehydration. Subsequently, the samples were placed in test tubes containing methyl salicylate for 3-4 hours for transparency. When all the teeth became transparent and the total root-canal system became visible, due to the presence of the dye [Indian Ink] within it, all the teeth were then taken out consecutively from methyl salicylate and placed in a glass Petri-dish over a LED light, for evaluation of Root Canal Morphology under Dental Operative Microscope [DOM]; according to the Vertucci's classification. Photographs were taken of the cleared tooth on all sides. A null hypothesis was assumed as no difference to be existent between CBCT and the decalcification technique in determining root canal configuration in human permanent mandibular second molars. All data were collected and recorded for further comparative evaluation with statistical analysis. Subsequently, statistical analysis was done using IBM SPSS Statistics for Windows, Version 26.0. (Armonk,

NY: IBM Corp).between two sets of data collected through CBCT and the Decalcification technique for the determination of the root canal morphology of human mandibular permanent second molar teeth (Figure No 2 .

Figure No 2: Canal visualization by CBCT(axial and coronal views) and by Decalcification technique (using India Ink staining and clearing technique)



III. Results

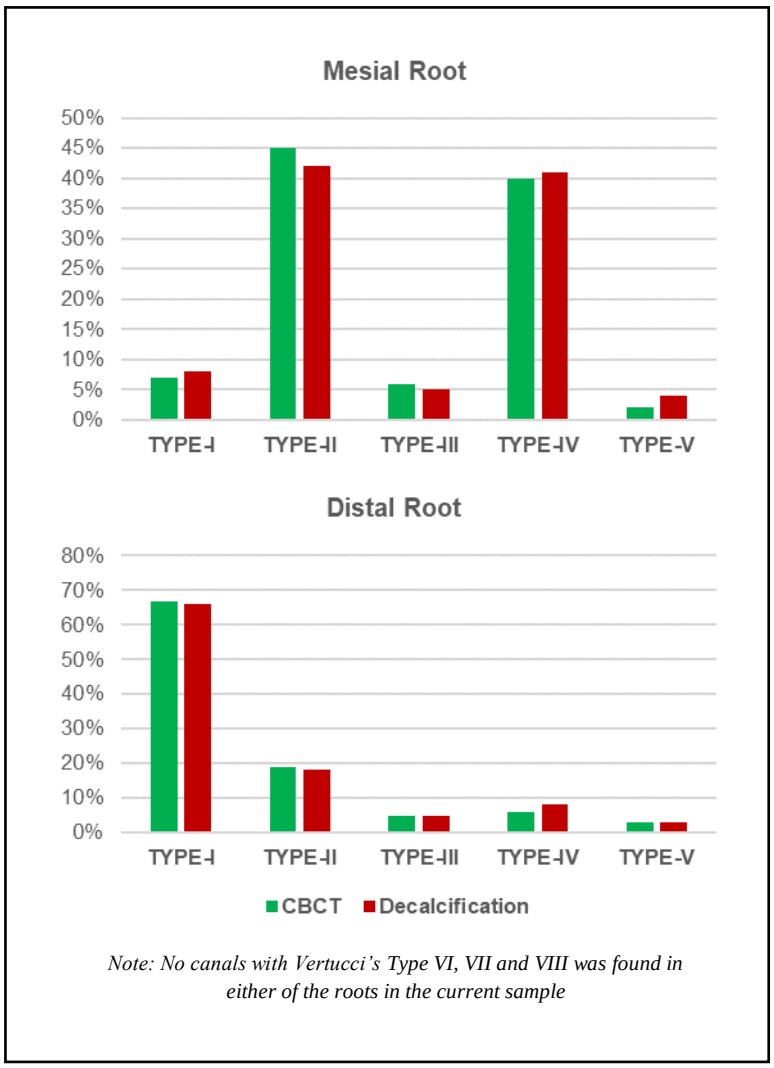
In this present study, the number of roots, number of root canals, root canal anatomical configurations along with its variations, and the correlations between no of root canals and root apex were analyzed. The results are evaluated in bar diagrams and pie charts for quantitative percentage analysis. Analysis of the 100 specimens through CBCT and decalcification technique shows the following result: -

The most common root canal configuration was found in Human Mandibular Permanent Second Molar: two-root and three-root canals (66% through CBCT, and 67% through Decalcification). The predominant root canal morphology in the **mesial roots** was **Vertucci's type II for both techniques:** (45% through CBCT and 42% through Decalcification) and followed by Type IV (through CBCT 40% and Decalcification technique 41%); Type I (by CBCT 7% and by decalcification 8%); Type III (6% by CBCT and 5% through decalcification) and Type V (2% by CBCT and 4% by decalcification) (Figure No.3).

The **distal root** in contrast showed **Vertuci'sType I** (67% through CBCT and 66% through decalcification) as the predominant canal configuration, followed by Type II (through CBCT 19% and Decalcification 18%); Type IV (by CBCT 6% and through decalcification 8%), Type III (6% through both CBCT and decalcification) and Type V (2% through both CBCT and decalcification).

The data obtained from the present study were statistically analyzed using the chi-square test. The result of the study shows that there is no significant difference between the observed and the expected values of Decalcification & dye penetration technique and CBCT, as $P > 0.05$**.**There is no need to reject the null hypothesis and there is likely significance between the canal classifications based on decalcification and CBCT.

Figure No 3: Bar Graph showing Root canal configuration of Mesial & Distal roots through vertucci's classification type



Note: No canals with Vertucci's Type VI, VII and VIII was found in either of the roots in the current sample

IV. Discussion

The present study delineates the evaluation of morphological variations, number of root canals in a root, and number of apical foramina of the human mandibular permanent second molars through decalcification & dye penetration technique[2] and CBCT. Subsequently, a comparative statistical analysis has been performed between those techniques owing to the advancement of diagnosis and prognosis of Endodontic treatment.

After the advent of digital radiography, CBCT became a popular and useful technique both in-vitro as well as in vivo for assessing canal anatomy. In this study, CBCT and decalcification & tooth-cleaning techniques were done to the determination of the root canal morphology of the human mandibular permanent second molar and an assessment of the accuracy of the Cone Beam Computed Tomography (CBCT) technique. For the assessment of the accuracy of CBCT, Decalcification & tooth-clearing technique was taken as a reference standard, and a comparative analytical study in-between that assessment on root canal morphology of mandibular permanent second molar has been performed.

The result of the study shows that there is no significant difference between the observed and the expected values of the Decalcification & Dye penetration technique and CBCT respectively. Upon the result and statistical analysis of this study, we can state that: CBCT can be used in-vivo, effectively, and successfully in identifying intricate Root canal Morphology along with C- shaped canals, accessory canals, and canal communications, etc, for a successful endodontic treatment.

The most commonly observed root morphology was Vertucci's Type II predominating in the mesial root canal and Vertucci's Type-I for the distal root of the mandibular second molar. We also observed that a double root canal is common in the mesial root and a single root canal is common in the distal root; a single root

apex is predominant (58% by CBCT & 55% by Decalcification technique), but findings of double root apex are also prominent (42% by CBCT & 45% by Decalcification technique) for the Mesial root and in the Distal roots single root apex is undoubtedly predominant (Table No.1).

Various studies uphold our present study such as the study of Sert et al 2008[10] where Vertucci's type II canal was predominant (31.5%); the study of Gulabivala et al. 2001[11] where Type II was found (35.9%) predominant; study of Weine et al 1988[6] where they found that type-II canal was predominant (52%) and study of Frank Vertucci 1984[8] where Type II was found (38%) predominant in the mesial canal of mandibular 2nd molar. For the distal root, no root canal, and no apical foramina per root, almost all the studies uphold our present study.

According to the present study Human Permanent Mandibular Second molars usually are two-rooted teeth with three canals[2,6,8,15,20,] and the observation was supported by almost all researchers[2,7,8,10,11,12,27].

Table No 1: Root canal configuration (number of root canals and number of root canal foramen at root apex)of Mesial & Distal roots through Vertucci's classification type

Configurations	CBCT	Decalcification	CBCT	Decalcification
Single Root Canal	7	8	67	66
Double Root Canal	93	92	33	34
Single Foramen at Root Apex	58	55	91	89
Two Foramens at Root Apex	42	45	9	11

V. Conclusion

The study reveals that the Human mandibular permanent second molars are two-rooted teeth with three canals according to both the technique, Decalcification & Dye penetration, and CBCT. Human Permanent Mandibular Second Molars teeth are commonly seen to have a double root canal in the mesial root and a single root canal in the distal root predominantly. CBCT can be used as a promising tool to accurately determine the root canal morphology of mandibular second molar teeth, thus aiding in the instrumentation and disinfection of the root canals and ultimately achieving an improved treatment outcome.

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