



COMPARATIVE EVALUATION OF EFFECT OF HERBAL INTRACANAL MEDICAMENTS (AZADIRACHTA INDICA and ALOE BARBADENSIS) ON MICROHARDNESS OF ROOT DENTIN.

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Abstract

Aim: The aim of this study was to comparatively evaluate the effect of herbal intracanal medicaments (*Azadirachta Indica* and *Aloe Barbadensis*) on microhardness of root dentin.

Materials and Method: A total of 40 human mandibular single rooted permanent teeth were collected for this study. The selected teeth were cleaned of any debris/deposits using ultrasonic scaler and disinfected using 2% chlorhexidine. The teeth were decoronated at cemento- enamel junction (CEJ) using diamond disc under water coolant. Canals were accessed and their patency was established using #10K file. All the canals were prepared till size #20 K file upto working length followed by preparation using protaper next file (F3) under copious irrigation. The specimens were then divided into 3 groups; Group I (n=20): Control group (N=20) (In which 10 teeth were tested on day 1 and 10 teeth were tested on day 14), Group II (n=10): *Azadirachta Indica* (neem), Group III (n=10): *Aloe Barbadensis* (*Aloe vera*). These medicaments were placed according to assigned groups. After 14 days, medicaments were removed using Endo activator and sodium hypochlorite. For each specimen, transverse root sections of 2 mm thickness was obtained from 2 mm below the CEJ. Vicker's hardness number was calculated after treatment and compared with the untreated control group. Data obtained was evaluated using the Student's *t* test (paired) and the Post Hoc Tukey's test.

Results: The statistical analysis revealed no significant reduction in the microhardness in all treated groups as compared with untreated control group.

Conclusion: Within the limitations of this study, it was concluded that herbal medicaments used in this study i.e *Azadirachta Indica* (*Neem*) and *Aloe Barbadensis* (*Aloe vera*) did not reduce the microhardness of the root dentin and can be effective antimicrobial agents to manage root canal pathogens.

Key Words: Aloe vera, Herbal Intracanal Medicament, Microhardness, Neem, Protaper, Root canal Treatment.

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DOI: - 10.48047/ecb/2023.12.si5a.025

INTRODUCTION

Endodontic therapy focus on eradicating bacteria and their by-products from the root canal system due to the polymicrobial environment of root canal infections, comprising both of aerobic and anaerobic microorganisms.¹ The intricate system of root canal makes it impossible towards decreasing the bacterial count just by cleaning and shaping alone. Therefore, additional methods such as the use of intracanal medicaments are required to effectively disinfect the root canal system and eliminate microorganisms. Hence, the intracanal medicament plays a key role in the success of root canal treatment.^{2,3}

Various chemicals such as calcium hydroxide, triple antibiotic paste, sodium hypochlorite (NaOCl), EDTA (ethylenediaminetetraacetic acid), chlorhexidine, triple antibiotic paste, phenols, Q mix used during root canal procedure may negatively affect the physical and mechanical properties of root dentin.⁴

Chemicals show several drawbacks as sensitivity, cytotoxic reactions and inability to completely eliminate bacteria from root canals. It has been reported that some chemicals used during endodontic procedures are capable of causing surface changes to dentin which in turn cause reduction in microhardness causing dentin to be structurally weak and non-supportive which may affect the final restored tooth.⁵

Nowadays, a wide range of herbal and natural remedies are utilised to prevent the negative effects of chemical irrigants. These products are particularly well-accepted because of their low toxicity, low microbiological resistance, minimal side effects, cost effectiveness, and ease of availability.⁵

World Health Organization (WHO) defines herbal medicine as plant derived material or preparation which contain raw or processed ingredients from one or more plants with therapeutic values. *Aloe barbadensis* (aloe vera) contains aloin and barbadolins as main chemical constituent. It has antimicrobial effect efficacy against caries causing microorganism, i.e. *Streptococcus mutans* and inhibitory effects on *Streptococcus pyogenes* and *E. faecalis* because of anthraquinone.⁶

Azadirachta Indica (Neem) also known as Indian neem/margosa tree is effective against *E. faecalis* and *Candida albicans*. It has strong antioxidant, antiseptic, antihyperglycemic, antiviral, astringent and antimicrobial properties.⁶ Both *Azadirachta Indica* (Neem) and *Aloe barbadensis* (Aloe vera) have inhibitory effect on MMP-2 and MMP-9. Matrix metalloproteinases (MMPs) are believed to

play key role on tissue destruction and are capable of degrading denatured interstitial collagen.²

Thus, the change of calcium/phosphate ratio modifies dentin's microhardness, solubility, permeability and surface roughness. Microhardness test determines the material's resistance to local deformation which is measured on the basis of the permanent surface deformation that remains after removal of a given load.⁷

There are many disadvantages of the currently available intracanal medicaments. As maintaining the microhardness of the tooth and complete or near complete elimination of microbes existing within the root canals is a challenge, there is a need to identify the best herbal intracanal medicament available. Due to the constant increase in antibiotic-resistant strains and side effects caused by synthetic/chemical drugs, it is necessary to look for herbal alternatives in endodontics.⁸ Therefore, the aim of this study was to compare and explore the effect of four different herbal intracanal medicaments namely *curcuma longa* (turmeric), *azadirachta indica* (neem), *aloe barbadensis* (aloe vera) and propolis on microhardness of root dentin.

MATERIALS AND METHOD-

Collection of specimen

Forty freshly extracted human mandibular single rooted permanent teeth extracted for orthodontic/periodontal reasons were collected for this study from the Department of Oral and Maxillofacial Surgery, Darshan Dental College and Hospital.

Teeth with dental caries, any fracture lines, resorption, anatomical defects, immature apex, curved canals, multiple canals, calcified or obliterated canals were excluded from the study.

Preparation of specimens

A total 40 human mandibular single rooted permanent teeth were collected. All the teeth were cleaned of any debris and calculus using ultrasonic scaler and disinfected with 2 % chlorhexidine.

The selected teeth were decoronated at the level of cemento-enamel junction using diamond disc in a slow speed handpiece under copious water coolant. The access to the canal was established and apical patency was achieved with a #10 K stainless steel file. Working length was determined by #15 K stainless steel file and the specimens were instrumented till # 20 K files, followed by sequence of rotary Pro-Taper upto size F3 under copious irrigation with 2mL of 3% sodium hypochlorite (NaOCl) and normal saline. The root canals received a final irrigation of 5 mL 5 % NaOCl solution and 5mL of 17% ethylene diamine tetra acetic acid (EDTA) after which the canals

were flushed with normal saline to avoid the prolonged effect of NaOCl and EDTA.

The prepared specimen were then randomly divided into one control group [without medicament (n=20)] and two experimental groups (n=10) with herbal medicaments.

- Group I (n=20): Control group
- Group IA- 10 teeth were evaluated for microhardness on day 1 and
- Group IB - 10 teeth were evaluated on day 14
- Group II (n=10): *Azadirachta Indica* (Neem)
- Group III(n=10): *Aloe Barbadensis* (Aloe vera)

Medicament gel was prepared using methylcellulose as a vehicle. Prepared gel for each medicament was introduced into the root canal using lentulo-spiral mounted on a slow speed contra angle handpiece in a clockwise direction to ensure that medicament reaches apical foramen and no medication was applied for control group. After medicament placement, the root canal access was sealed with cotton pellet and cavitated followed by incubation at 37°C temperature and at 100% humidity for 14 days.

After 14 days, the medicament was removed with the help of EndoActivator and 5% NaOCl. Any leftover NaOCl was washed with 2 mL distilled water as a part of final irrigation. Paper points were used for drying the root canal. From each root, a section was obtained of 2 mm thickness by transverse resection with diamond disc under water coolant, 2 mm apical to the CEJ. The sections were then mounted in cold cure acrylic resin to be evaluated for microhardness of dentin.

To evaluate microhardness three indentations were made on the dentinal surface, 1mm from the root canal, towards the root surface, using Vicker's microhardness tester. All the indentations were made with 50gm load with dwell time of 10 seconds.

RESULTS

The data obtained was tabulated and statistically analyzed using SPSS software V.21.0. The results of microhardness were analyzed using 't' test followed by Post hoc Tukey test.

Table 1 shows descriptive statistics of microhardness of dentin for control, *Azadirachta indica* and *Aloe barbadensis*. *Aloe barbadensis* (58.37±3.39) group exhibited highest mean microhardness value followed *Azadirachta indica* (55.03± 6.26).

Paired 't' test showed (Table 2) significant difference in the microhardness of dentin between control 1 and 2. Control 2 exhibited reduced

microhardness of dentin after incubation for 14 days compared to control 1.

Intergroup comparison of microhardness between herbal medicament groups using Post Hoc Tukey's test, indicates no significant difference at $p \leq 0.05$ among the tested medicaments. (Table 3)

DISCUSSION

A full eradication/disinfection of the bacteria and their by-products from the affected root canal is required for an effective endodontic therapy. Root canal therapy is a chemo-mechanical process that makes use of chemical irrigants and medications to ensure that root canals are properly disinfected.⁹ Debridement and cleaning of the canal is essential for sustained endodontic success. Intracanal medications are "medications temporarily inserted into root canals with strong biocompatibility for the objective of preventing micro-organisms penetration."¹

An intracanal medication is used to remove any bacteria that may still be present in the canal after instrumentation; to lessen inflammation in the periapical tissues and pulp remnants; to make the contents of the canal inert and neutralise tissue debris; to dry chronically damp or "weeping" canals and act as a barrier against leaking from an interappointment dressing in symptomatic instance.¹⁰

Several chemicals used in root canal procedures are calcium hydroxide, triple antibiotic paste, phenols, formaldehyde, halogens, steroids preparation, polyantibiotic paste, eugenol and chlorhexidine.³

Chemicals used during root canal treatment may adversely influence the mechanical and physical characteristics of root dentin. These substances dramatically decreased the flexural strength, microhardness, solubility, permeability, and surface roughness of dentin, according to research.³

There has been a significant increase in interest in the development of alternative types of antimicrobials for the control of illness as a result of the rise in bacterial resistance to antibiotics. Researchers are currently looking for herbal alternatives in endodontics as a result of the ongoing rise in strains that are resistant to antibiotics and the adverse effects brought on by synthetic medications. Herbal plant extracts can be used to offset the drawbacks of the chemical medications that are now available.⁶

Herbs contains phytochemicals, including terpenoid, lignans, flavonoids, polyphenolic, carotenoid sulphide, saponin, coumarins and

curcumins, that impart them anti-inflammatory, antioxidant and antimicrobial properties.¹¹

Aloe vera is made of the Arabic term "Alloeh," which implies a bright, bitter substance, and the Latin word "vera," which means "truth". Aloe Vera known as *Aloe Barbadensis* Miller, is a member of the Liliaceae family. All the three extract of Aloe vera, i.e., aqueous, ethanol, and acetone has antimicrobial effect against Gram-positive bacteria and Gram-negative bacteria. Aloe vera has very strong antioxidant property and anti-inflammatory action.^{12,13}

Azadirachta indica commonly known as neem, has analgesic, anthelmintic, antibacterial, antiyeast, antiulcer, antifertility, antifilarial, antifungal, antihyperglycemic, anti-inflammatory, antiviral, antimalarial, diuretic, antinematodal, antipyretic, antispasmodic, insecticidal, antispermato-genic, antitumor, hypercholesteremic, hypoglycaemic, immunomodulatory properties.^{14,15}

The microhardness of dentin depends upon the degree of mineral content, amount of hydroxyapatite in intertubular substance, and tubular density and tubule diameter. The change of Ca/PO₄ ratio modifies dentin's microhardness, solubility, permeability and surface roughness.¹⁶

In this study Aloe vera improved the microhardness, which may be due to their MMP inhibiting activity and neutral pH. Aloe vera contains an active component alloin; which shows MMP inhibiting property i.e. it reduces collagen degradation and loss of Ca/PO₄ ions from dentin.² Whereas, neem shows minimal changes in the microhardness of the root dentin similar to study by Gondi et al., in which *Azadirachta indica* and green tea groups did not show any significant changes in root dentin microhardness.¹⁷ It may be attributed to the pH of these medicaments, as many researchers have demonstrated that more acidic and alkaline pH causes the calcium and phosphate to dissolve from the hydroxyapatite crystals, which has a negative impact on the microhardness of the dentin. The pH of Neem is 8.4, which reduces the chances of Ca/PO₄ loss due to pH changes i.e. no changes in the microhardness of the root dentin is observed.¹⁸

CONCLUSION

Within the limitations of this study, it was concluded that herbal medicaments used in this study i.e. *Azadirachta Indica* (*Neem*) and *Aloe Barbadensis* (*Aloe vera*) exhibits positive results in terms of minimal effects on the microhardness of the root dentin and can be used as an intracanal medicament in the endodontic treatment. Further

studies are required to evaluate the antimicrobial efficacy of these medicaments.

CONFLICTS OF INTEREST

Authors declare that there are no conflicts of interest.

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Table 1: Descriptive statistics of microhardness of dentin for control, *Curcuma longa*, *Azadirachta indica*, *Aloe barbadensis*, Propolis.

GROUPS	N	MEAN	SD
Group I (Control)			
Group Ia [Before]	10	57.67	3.28
Group Ib [After 14days]	10	52.27	5.34
Group II(<i>Azadirachta indica</i>)	10	55.03	6.26
Group III(<i>Aloe barbadensis</i>)	10	58.37	3.39

Table 2: Paired 't' test to evaluate significant difference in microhardness after medication application for the various test groups.

Groups	Mean	N	Std. Deviation	Std. Error Mean	Mean Difference	't'	p value
Group IA	57.666	10	3.284	1.039	2.632	1.133	0.286
Group II (<i>Azadirachta indica</i>)	55.034	10	6.258	1.979			
Group IA	57.666	10	3.284	1.039	0.701	0.416	0.687
Group III (<i>Aloe barbadensis</i>)	58.367	10	3.390	1.072			

Table 3: Intergroup comparison of microhardness between herbal medicament groups using Post Hoc Tukey's test.

Groups		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
Group II (<i>Azadirachta Indica</i>)	Group III (<i>Aloe Barbadensis</i>)	-3.333	2.075	0.388	-8.921	2.255
Group III (<i>Aloe Barbadensis</i>)	Group II (<i>Azadirachta Indica</i>)	3.333	2.075	0.388	-2.255	8.921