

A Study to Compare Antibacterial Efficacy of Turmeric Extract, Neem Extract and Sodium Hypochlorite as Root Canal Irrigants

Namita Agrawal^{1*} Jayesh Billore² Ankita Dixit³ Gagandeep Singh Randhawa⁴ Amrita Soni⁵ Kunal Gupta⁶

¹Senior Lecturer, Department of Conservative Dentistry and Endodontics, Modern Dental College & Research Centre, Madhya Pradesh, India.

²Reader, Department of Endodontics & Conservative Dentistry College of Dental Sciences and Hospital, Indore, Madhya Pradesh, India.

³Assistant Professor, Department of Pedodontics and Preventive Dentistry, Ahmedabad Dental College and Hospital, Ahmedabad, Gujarat, India.

⁴Reader, Department of Oral and Maxillofacial Surgery, Ahmedabad Dental College and Hospital, Ahmedabad, Gujarat, India.

⁵Consulting Prosthodontist, Ujjain, Madhya Pradesh, India.

⁶Senior Lecturer, Department of Oral and Maxillofacial Surgery, Modern Dental College and Research Centre, Madhya Pradesh, India.

ABSTRACT

Background: Thorough debridement and complete elimination of micro-organisms are objectives of an effective endodontic treatment. *E. faecalis* is the predominant micro-organism recovered from root canal of the teeth where previous endodontic treatment has failed. For many years, intracanal irrigants have been used as an adjunct to enhance antimicrobial effect of cleaning and shaping in endodontics. Aim: The aim of the study is to compare the antibacterial effectiveness between Turmeric extract as irrigant with neem extract irrigant and sodium hypochlorite against *E. faecalis*.

Materials and Methods: The present study is a randomized controlled *in vitro* study conducted on 90 extracted permanent single rooted teeth. The teeth were divided into three groups- Group 1 was irrigated with Turmeric extract, Group 2 was irrigated with neem extract irrigant and Group 3 with 3% sodium hypochlorite. The colonies in the reduction of *E. faecalis* was noted as CFU values per ml. Wilcoxon signed ranked test was used to compare in between the three groups. Mann-Whitney's test was used for inter group comparison between the three groups.

Results: Our results showed that the Turmeric extract had the maximum reduction in the CFU colonies followed by sodium hypochlorite. Neem extract irrigant had the least reduction in the colonies of *E. faecalis*. ($P < 0.05$).

Conclusion: Turmeric extract irrigant show better reduction in *E. faecalis* as compared to 3% sodium hypochlorite and neem extract irrigant.

Keywords: Turmeric extract, endodontic irrigant, Enterococcus faecalis, neem extract, sodium hypochlorite.

INTRODUCTION

The microorganisms in the oral cavity are most often the opportunistic pathogens. They have the ability to invade and establish an infectious process. Longer the root canal remains infected, more the number of facultative anaerobes. The elimination of microorganisms from the infected root canal defines the success of endodontic treatment.^{1,2}

Sodium hypochlorite fulfils most of the requirements of the ideal endodontic irrigant and is widely used in dentistry. It has been the choice of irrigant due to its pulp dissolving capacity, antimicrobial action and ability to remove smear layer.¹⁻⁶ However, cases have been reported of allergic reaction due to the use of sodium

Received: Jan. 3, 2021; Accepted: Mar. 22, 2021

*Correspondence Dr. Namita Agrawal.

Department of Conservative Dentistry and Endodontics, Modern Dental College & Research Centre, Madhya Pradesh, India.

Email: namita0903@gmail.com

hypochlorite.⁷⁻¹⁰ There have been cases of tissue toxicity and accidental mishaps associated with sodium hypochlorite. A bad smell and taste is also associated with irrigation using sodium hypochlorite. To overcome this disadvantage, other herbal alternatives have been considered over time. Neem also known as *Azadirachta indica* is widely used in clinical dentistry due to its antibacterial and antiviral properties. Studies have been done to evaluate the antibacterial effectiveness of neem against wide variety of micro-organism.¹¹⁻¹⁵ The advantage of using neem as an endodontic irrigant is due to its antibacterial and antioxidant properties. It shows a wide spectrum of action against gram positive and gram negative organism. Turmeric also known as *Curcuma longa* also has gained popularity in recent time due its antibacterial activity.¹⁶⁻²⁰ It has shown effectiveness against *S. mutans* and *E. fecalis* organisms. It is also known to show a wide spectrum of action on gram positive and gram negative organism. Therefore, the aim of the present study is to compare neem extract, Turmeric extract and sodium hypochlorite as irrigant. The comparison is based on the effectiveness of these irrigants against *E. fecalis* in extracted permanent teeth.

MATERIALS AND METHODS

The present study followed an *in vitro* study design. The study design was approved by the institutional review board of the dental institution. The present study uses ethanolic extract of neem and Turmeric to prepare the irrigation solution. The ethanolic extract of neem was obtained from neem leaf ground into coarse and medium coarse particles. These particles were mixed with 50% ethanol and this suspension was placed on the magnetic stirrer for 4 hours. Whitman's filter paper was used to obtain the filtrate which was then dried to obtain the extract. The extract was dried to evaporate the solvent to prevent the influence of extracting agent (ethanol) on the final extract. Turmeric extract was obtained using Turmeric powder which was pulverized into medium coarse granules. The extracting agent and the procedure to obtain the final extract was similar to that for neem extract. These extracts were stored in dark coloured bottles at 4 degree Celsius until use. The irrigating solution was prepared by mixing the crude extract in DMSO (dimethyl sulfoxide) solvent. Previous studies have noted MIC for neem extract to be 25% and for

Turmeric extract to be 20%. Neem extract was prepared by dissolving 25 grams of crude extract in 100 ml DMSO solution. Similarly, Turmeric extract was prepared by dissolving 20 grams of crude extract in 100 ml of DMSO. The final solution were refrigerated at 4 degree Celsius. The present study includes a total of 90 single rooted extracted teeth for periodontal or orthodontic purpose. Access opening was done with No. 4 round carbide bur (DENTSPLY Maillefer, OK, USA). The working length was determined using radiograph and was kept 0.5 mm short of the radiographic apex. After initial instrumentation up to no 30 stainless steel hand files, rotary instrumentation was done using ProTaper (DENTSPLY Maillefer, OK, USA) up to F3. After complete instrumentation the tooth were scheduled for the inoculation of *E. fecalis*. A pure culture of test strain of *E. fecalis* ATCC 29212 was prepared in sterile nutrient broth. The extracted teeth placed in the nutrient broth containing *E. fecalis* and incubated for 21 days at 37 degree Celsius. The level of the nutrient broth was checked daily, and new broth inoculated with *E. fecalis* was added to maintain all the extracted teeth completely submerged in the inoculated broth for 21 days. The inoculated teeth were divided into three groups for the irrigation protocol. 30 teeth were randomly assigned to each group. Each tooth was assigned a serial number. The allocation of each sample into individual groups was done using block randomization using lots. Group 1 was irrigated using 3% sodium hypochlorite, group 2 was irrigated using neem extract irrigant and group 3 was irrigated using Turmeric extract irrigant. The solution was irrigated using flexible tip attached to a 5 ml irrigating syringe. Each tooth was irrigated with 5 ml of irrigating solution. The presence of *E. fecalis* was verified using an eosinophilic and gentian violet staining to verify gram positive organism. The data obtained were entered in the spreadsheet and statistical analysis was done using SPSS software version 22. (IBM Corp, Armonk, NY, USA). Kruskal-Wallis test was used to compare the CFU values per ml between the three groups. Mann-Whitney's test was used was used in multiple pairwise comparison between the groups.

RESULTS

A total of 90 permanent single rooted teeth are included in the study. The mean CFU values obtained using neem as irrigant was 0.45 ×

10^3 having a standard deviation of 0.35. The mean CFU values obtained using Turmeric extract as irrigant was 0.28×10^3 having a standard deviation of 0.08. The mean CFU values obtained using sodium hypochlorite as irrigant is 0.40×10^3 having a standard deviation of 0.30 [Table 1].

Table 1: Comparison of colony forming units values between Turmeric extract irrigant, neem extract irrigant and sodium hypochlorite

Group	Number of Samples	Mean	SD	P
Sodium Hypochlorite	30	0.45×10^3	0.35	0.008*
Neem Extract	30	0.28×10^3	0.29	
Turmeric Extract	30	0.80×10^3	0.79	

*Significant: $P < 0.05$, Kruskal – Wallis test . SD = Standard deviation

Kruskal-Wallis test was used to compare CFU values between the three groups. Kruskal Wallis test showed a significant difference between the three groups with Turmeric extract irrigant having the least CFU values ($P = 0.008$). Mann-Whitney's test was used for multiple pairwise comparison between the three groups. Turmeric extract irrigant showed significantly less CFU values as compared to neem extract irrigant ($P = 0.004$). However, Turmeric extract irrigant as compared to sodium hypochlorite showed lower CFU values but the results was not statistically significant ($P = 0.189$). On the other hand sodium hypochlorite irrigant showed statistically significant lower CFU values as compared to neem extract irrigant ($P = 0.035$). Thus, Turmeric extract showed statistically significant difference to reduce the *E. fecalis* colonies as compared to other two groups.

DISCUSSION

The final goal of root canal treatment is to clean the root canal system free of debris and microbiota thereby leading to well sealed microbial tight filling. Chemical irrigants have been in practice over a long time in the disinfection of infected root canal. Due to its antibacterial action and pulp dissolving ability, sodium hypochlorite is being used as an ideal irrigant for root canal treatment. Other

irrigants with increased antibacterial effectiveness such as chlorhexidine has also been advocated in necrotic teeth. However, there has been an increased allergic reaction and adverse effects associated with the use of chemical irrigants. To overcome this, the use of herbal alternatives having comparable antibacterial effectiveness should be considered. Neem extract is the most common herbal extract used as an irrigant due to its antioxidant properties which aid in antibacterial action against *E. fecalis*. Dutta and Kundabala²⁰ in 2014 showed similar antibacterial efficacy of neem extract as irrigant with sodium hypochlorite and chlorhexidine irrigant against *E. fecalis*. Turmeric extract has recently gained a limelight as an irrigant in endodontics due its antibacterial properties. Gupta-Wadhwa *et al.*²² have demonstrated comparable antibacterial effectiveness of Turmeric than other herbal irrigants. Gupta *et al.*²¹ also shows higher antibacterial effectiveness of Turmeric extract as compared to other herbal agents. The present study shows Turmeric extract irrigant to have better antibacterial effectiveness followed by sodium hypochlorite. The neem showed to have least antibacterial effectiveness. These results are contradictory to previous study by Gupta-Wadhwa *et al.*,²² where sodium hypochlorite showed higher effectiveness as compared to Turmeric extract. This can be attributed to a lower concentration of sodium hypochlorite used in the present study as compared to other study.¹⁵⁻²¹ The present study showed similar results in relation to comparison of neem and sodium hypochlorite. The present study compares the antibacterial effectiveness of only two herbal agents. Further improvement in the study can be done by including more herbal agents in the comparison. The present study evaluates the colony forming units per ml to determine the antibacterial efficacy. However, further improvement in the study can done by evaluation of the antibacterial efficacy by more standardized methods like PCR. This can aid in achieving more standardized results. This study thus acts as a preliminary study to compare Turmeric extract, neem extract irrigant and sodium hypochlorite irrigant. This study recommends further studies at a molecular level to understand the exact mechanism of the action of pharmacological agents.

CONCLUSION

The present study shows Turmeric extract to have better effectiveness followed by sodium hypochlorite and neem extract irrigant. This study thus acts as a preliminary study to compare the antibacterial effectiveness of Turmeric extract irrigant and neem extract irrigant with sodium hypochlorite.

CONFLICTS OF INTEREST

The authors declare they have no potential conflict of interests regarding this article.

REFERENCES

1. Fabricius L, Dahlén G, Holm SE, Möller AJ. Influence of combinations of oral bacteria on periapical tissues of monkeys. *Scand J Dent Res* 1982;90:200-6.
2. Gomes BP, Pinheiro ET, Gadê-Neto CR, Sousa EL, Ferraz CC, Zaia AA, et al. Microbiological examination of infected dental root canals. *Oral Microbiol Immunol* 2004;19:71-6.
3. Ruoff KL, de la Maza L, Murtagh MJ, Spargo JD, Ferraro MJ. Species identities of enterococci isolated from clinical specimens. *J Clin Microbiol* 1990;28:435-7.
4. Love RM. *Enterococcus faecalis* – A mechanism for its role in endodontic failure. *Int Endod J* 2001;34:399-405.
5. Chugal NM, Clive JM, Spångberg LS. A prognostic model for assessment of the outcome of endodontic treatment: Effect of biologic and diagnostic variables. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2001;91:342-52.
6. Roudsari RV, Jawad S, Taylor C, Darcey J, Qualtrough A. Modern endodontic principles. Part 8: The future of endodontics. *Dent Update* 2016;43:430-2, 435-8, 441.
7. Young GR, Parashos P, Messer HH. The principles of techniques for cleaning root canals. *Aust Dent J* 2007;52:S52-63.
8. Zehnder M. Root canal irrigants. *J Endod* 2006;32:389-98.
9. Ayhan H, Sultan N, Cirak M, Ruhi MZ, Bodur H. Antimicrobial effects of various endodontic irrigants on selected microorganisms. *Int Endod J* 1999;32:99-102.
10. Byström A, Sundqvist G. Bacteriologic evaluation of the effect of 0.5 percent sodium hypochlorite in endodontic therapy. *Oral Surg Oral Med Oral Pathol* 1983;55:307-12.
11. Guivarc'h M, Ordioni U, Ahmed HM, Cohen S, Catherine JH, Bukiet F. Sodium hypochlorite accident: A systematic review. *J Endod* 2017;43:16-24.
12. Faras F, Abo-Alhassan F, Sadeq A, Burezq H. Complication of improper management of sodium hypochlorite accident during root canal treatment. *J Int Soc Prev Community Dent* 2016;6:493-6.
13. Mehdipour O, Kleier DJ, Averbach RE. Anatomy of sodium hypochlorite accidents. *Compend Contin Educ Dent Jamesburg NJ* 1995 2007;28:544-6, 548, 550.
14. Chaugule VB, Panse AM, Gawali PN. Adverse reaction of sodium hypochlorite during endodontic treatment of primary teeth. *Int J Clin Pediatr Dent* 2015;8:153-6.
15. Mustafa M. Antibacterial efficacy of neem (*Azadirachta indica*) extract against *Enterococcus faecalis*: An in vitro study. *J Contemp Dent Pract* 2016;17:791-4.
16. Nabavi SF, Di Lorenzo A, Izadi M, Sobarzo-Sánchez E, Daglia M, Nabavi SM. Antibacterial effects of Turmeric: From farm to food, cosmetic and pharmaceutical industries. *Nutrients* 2015;7:7729-48.
17. Gupta A, Duhan J, Tewari S, Sangwan P, Yadav A, Singh G, et al. Comparative evaluation of antimicrobial efficacy of *Syzygium aromaticum*, *Ocimum sanctum* and *cinnamomum zeylanicum* plant extracts against *Enterococcus faecalis*: A preliminary study. *Int Endod J* 2013;46:775-83.
18. Ghonmode WN, Balsaraf OD, Tambe VH, Saujanya KP, Patil AK, Kakde DD. Comparison of the antibacterial efficiency of neem leaf extracts, grape seed extracts and 3% sodium hypochlorite against *E. faecalis* – An in vitro study. *J Int Oral Health* 2013;5:61-6.
19. Venkatadri B, Arunagirinathan N, Rameshkumar MR, Ramesh L, Dhanasezhian A, Agastian P, et al. In vitro antibacterial activity of aqueous and ethanol extracts of *Aristolochia indica* and *Toddalia asiatica* against multidrug-resistant bacteria. *Indian J Pharm Sci* 2015;77:788-91.

20. Dutta A, Kundabala M. Comparative anti-microbial efficacy of Azadirachta indica irrigant with standard endodontic irrigants: A preliminary study. J Conserv Dent 2014;17:133-7.
21. Gupta-Wadhwa A, Wadhwa J, Duhan J. Comparative evaluation of antimicrobial efficacy of three herbal irrigants in reducing intracanal E. faecalis populations: An in vitro study. J Clin Exp Dent 2016;8:e230-5.