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EVALUATION AND COMPARISON OF ANTIMICROBIAL EFFICACY OF DIFFERENT ROOT CANAL IRRIGANTS ON ENTEROCOCCUS FECALIS: AN INVITRO COMPARATIVE STUDY

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ABSTRACT

Background: Endodontic treatment failure may occur due to number of factors which includes bacterial persistence, improperly cleaned and obturated root canals, faulty coronal seal, and missed canals. Enterococcus faecalis is the most important bacterial species which most often cause endodontic failure. **Aim:** To Evaluate the Antimicrobial Efficacy of Normal Saline, Sodium Hypochlorite, Octenidine dihydrochloride, Simvastatin as irrigant solution against Enterococcus fecalis. **Methodology:** A total of 9 agar plates were taken for the study. Samples were placed on each agar plate as Group A: Saline, Group B: 5.2% Sodium hypochlorite, Group C: Octenidine Dihydrochloride (OCT), Group D: Simvastatin solution 32µg/ml concentration. Linear measurement was done by measuring the zones of inhibition around the endodontic irrigants in millimeters. **Results:** The results of the study have shown that the zone of inhibition of Group B is 19.3 mm, which confirmed the best outcome. While Group C has 11.4 mm, followed by Group D which is 9 mm. **Conclusion:** 5.2 % sodium hypochlorite has better antimicrobial property than OCT, Simvastatin and Normal saline.

KEYWORDS : antimicrobial, irrigant, simvastatin, chlorohexidine, octenidine dihydrochloride

INTRODUCTION:

During endodontic therapy, it is imperative to eradicate dentinal debris, live microorganisms all pulpal tissues from the root canal system. Because pulpal and peri-radicular inflammation are caused by bacteria and their metabolites, getting rid of them is essential for effective endodontic therapy [1]. The architecture of root canals is intricate, with multiple anatomical abnormalities. In addition to invading the anatomic abnormalities of the root canal system, the bacteria found in root canals are also found in the dentinal tubules [2]. Endodontic infections are multi-microbial however obligatory anaerobic bacteria predominates [3]. Because it is such a tenacious organism, Enterococcus plays a considerable role in the formation of periradicular lesions following root canal therapy [4]. An optimal root canal irrigant must have strong antibacterial action, dissolve any remaining pulp tissues without posing a systemic risk, reduce instrument friction during mechanical preparation, and be readily available [5].

Since Walker's 1936 initial report of using sodium hypochlorite as a endodontic irrigant, the substance has gained widespread acceptance. It is an effective organic solvent for vital, necrotic and fixed tissue and acts as a potent antimicrobial agent[6].It has been found that the disadvantages of using the highest concentration of sodium hypochlorite as an root canal irrigant are related to its cytotoxicity.Its extrusion may cause sudden edema & profuse bleeding [7]

Simvastatin is a structural analogue of HMG-CoA (3-hydroxy-3-methylglutaryl-coenzyme). It shows antibacterial properties as one of its pleiotropic effects. Numerous studies have revealed its antimicrobial efficacy against periodontal pathogens [8,9]. OCT is a bispyridine antimicrobial compound. It exhibit broad-spectrum antimicrobial effects, covering both gram-positive and gram-negative bacteria, fungi, and several viral species [10] .

OCT (Octenidine dihydrochloride) is currently widely used in mouthwash formulations and other dental applications, as well as in the medical field for skin burns, decontaminating mucous membranes, and open wounds [11]. Its component include phenoxyethanol, an ethanol derivative that acts as a preservative, and 0.1% OCT[12].OCT has been shown in numerous investigations to have a strong antimicrobial action against E. faecalis[13,14].

MATERIALS AND METHODS

Preparation Of SMV Solution

Pure powdered simvastatin was obtained using SMV. To achieve a concentration of 32µg/ml of SMV under aseptic conditions, SMV powder was mixed with 10 mL of distilled water using a vortex device. A single-use Eppendorf tube was filled with 0.5 mL of the prepared SMV, which was then stored at 4 °C [18].

To Evaluate The Anti-microbial Efficacy The Groups Were Divided As-

- Group A (n=9)-Normal Saline
- Group B (n=9)- 5.2%Sodium Hypochlorite
- Group C (n=9)- 0.1%Octenidine hydrochloride
- Group D (n=9)- 32µg/ml Simvastatin

Agar Diffusion Test

Mueller-Hinton agar was used in this experiment. Mueller-Hinton agar was used to produce a pure culture of E. faecalis.MTCC 2729 was the strain of E. faecalis utilized in this investigation. It was inoculated into Mueller-Hinton Broth and incubated for the entire night at 37 °C. After that, streaks were made on the agar plates.

In a Petri dish, small wells (6 mm diameter) were dug in the agar and filled with the study irrigants. Four cavities were poured with irrigants - the quantity used for each cavity was 20µL. For 48 hours, the Petri dishes were incubated at 37 °C. The purpose of the study was to use the agar diffusion test to evaluate the zone of inhibition. The zone of inhibition's linear measurement in millimeters was used to compute the results[19,20].

RESULTS:

- Antimicrobial efficacy total of four groups of irrigating solutions were checked for E. faecalis.
- The diameter of zones of inhibition is tabularized for all the groups with their mean and standard deviation. Results were recorded based on the diameter of the zones.

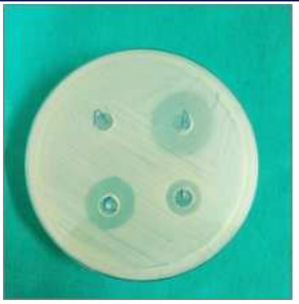


Figure 1: Inhibitory zone (mm) of different irrigants against Enterococcus Faecalis.

The results by using the agar diffusion method have shown that the Group B has maximum zone of inhibition followed by Group C, Group D, Group A. This Shows that Group B has better antimicrobial effect as compared to other group.

Statistical Anaylsis:

Table 1 - Comparison Of Zone Of Inhibition (In Mm) Among Four Groups

Group	N	Mean	SD	Minimum	Maximum	F-value	p-value
Group A	9	0.00	0.00	0	0	138.135	<0.001*
Group B	9	19.33	3.74	10	23		
Group C	9	11.56	1.33	10	14		
Group D	9	8.22	0.97	7	10		

One-way ANOVA test; * indicates a significant difference at $p \leq 0.05$

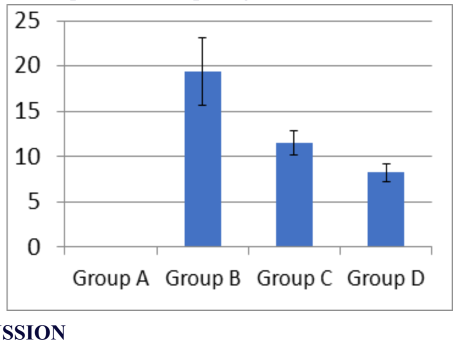
This table compares zone of inhibition (in mm) among four groups. No zone of inhibition was seen in Group A. Zone of inhibition in Group B, Group C and Group D were 19.33mm, 11.56mm and 8.22mm respectively. This difference in the zone of inhibition (in mm) among four groups was statistically significant.

Table 2 - Intergroup Comparison Of Zone Of Inhibition

Group	Mean difference	P-value
Group A vs Group B	-19.33	<0.001*
Group A vs Group C	-11.56	<0.001*
Group A vs Group D	-8.22	<0.001*
Group B vs Group C	7.77	<0.001*
Group B vs Group D	11.11	<0.001*
Group C vs Group D	3.34	<0.001*

- Post hoc Tukey test; * indicates a significant difference at $p \leq 0.05$
- This table presents the pairwise comparison of zone of inhibition (in mm) among four groups. Group B showed a significantly greater zone of inhibition as compared to the other three groups. Group C showed a significantly greater zone of inhibition as compared to Group A and Group D. Group D showed a significantly greater zone of inhibition as compared to Group A.

Table 1: Mean Values Of Zone Of Inhibition Between Group A, Group B, Group C And Group D Against E.faecalis.



DISCUSSION

The present study compared the antimicrobial effectiveness of four irrigants against E. faecalis. E. faecalis was selected because in root canal infection it is persistent. Irrigation is essential for a root canal procedure's success. It reduces the amount of friction between the dentine and the instrument, improving the files' ability to cut, dissolving tissue, lowering the temperature of the file and the tooth [5]. With the exception of saline, every experimental irrigant had antibacterial qualities. When compared to simvastatin and octenidine hydrochloride, NaOCl exhibits the strongest antibacterial action against Enterococcus fecalis.

E. faecalis is the most often isolated bacteria from root canals in individuals who have failed endodontic procedures. In addition to being able to survive as a single organism or as a significant part of the oral flora, it possesses a variety of virulence characteristics that enable it to persist as a pathogen in root canals. Because of its characteristics, which include the capacity to proliferate and endure in situations where treatment is unsuccessful and to cause chronic infections, this organism is significant and an essential choice for numerous antimicrobial studies. Since it enables a direct comparison of the materials against the organisms, the agar diffusion method is considered the most accepted method to test the antimicrobial activity of different dental materials and irrigants, indicating the potential of the test materials to eradicate microorganisms in the local microenvironment of the root canal system. For this reason, it was used in the study for antimicrobial activity assessment [21].

The most often used "root canal irrigant" at the moment is NaOCl. It functions as a lubricant for machinery, provides an efficient tissue solvent, has a broad range of antibacterial activity, and may eliminate loose material through the root canals. It is used in concentrations, ranges from 0.5 percent to 5.25 percent. The production of hypochlorous acid (HOCl) and its oxidative effect on the sulfhydryl groups of bacterial enzymes provide NaOCl its antibacterial properties. This action will disturb the microorganism's metabolism. Despite being a powerful antibacterial agent, NaOCl is unpleasant to taste, smell, and toxic when inject into the periradicular tissues [22].

A limited number of in vitro studies have found that OCT, when administered as a root canal irrigant, exhibits substantial antibacterial effect [15, 16, 17].

Octenidine's cation-active structure, which tends to adhere easily to the negatively charged bacterial cell envelope and instantly interfere with the vital processes of the cell membrane, is most likely the cause of its enhanced antibacterial action.

The high antimicrobial efficacy without negatively impacting human epithelia or wound tissue is explained by preliminary findings that indicate a strong adherence, particularly to lipid components (such as cardiolipin) prominent in bacterial cell membranes[23]. Because of its greater antibacterial action, low cytotoxicity, and excellent biocompatibility, OCT is being explored as a possible CHX substitute [18].

For the management of hyperlipidemia, statins are the first-choice medication class. SMV has shown to have a number of advantageous properties, such as antibacterial, anti-inflammatory, immunomodul atory, antioxidant, and bone-forming actions [9].

By inhibiting the activity of 3 hydroxy 3 methylglutaryl coenzyme A (HMG CoA) reductase, statins lower the amount of endogenous cholesterol produced [9].

In the present study, Group B (5.2%Sodium hypochlorite) shows highest average inhibition zone, indicating it has the strongest antibacterial effect among other groups.

CONCLUSIONS

Within the limitations of this study, it is concluded that 5.2 % sodium hypochlorite has better antibacterial properties compared to OCT, Simvastatin and Normal saline. The following is a ranking of the most effective irrigants' in vitro antimicrobial effects, from strongest to weakest: 5.2% sodium hypochlorite, OCT, simvastatin, and normal saline. Further clinical research is necessary to assess the efficacy of simvastatin at different dosage levels because the trial was experimental in nature and results may vary in real-world situations.

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