ORIGINAL RESEARCH

Evaluation of Nanomagnesium Oxide in Combination with Garlic Extract as an Endodontic Irrigant: An *In Vitro* Study

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Abstract

Aim: The aim of this *in vitro* study was to evaluate the effectiveness of the combination of garlic extract in combination with magnesium oxide (MgO) for use as an endodontic irrigant at various contact times.

Materials and methods: All 48 teeth were divided into 6 groups according to irrigation used after inoculation with *Enterococcus faecalis* and incubation. The control groups consisted of saline and sodium hypochlorite (NaOCl) used as irrigants and the test groups employed garlic extract combined with nano-magnesium oxide (nano-MgO) used as irrigant with two contact times, namely, 2 and 5 minutes, and garlic extract and nano-MgO used solely for 5 minutes each. Colony-forming units (CFUs) were counted after plating and incubation.

Results: In NaOCl, and in both combination groups, there was a significant reduction in CFU counts. The saline group showed no decrease. Statistical analysis showed no difference in efficacy between NaOCl and the two combination groups. There was a statistical difference between the combination group and garlic/nano-MgO alone at both 2 and 5 minutes.

Conclusions: Under the conditions of this study, a novel irrigant, a combination of nanoparticles of MgO and garlic extract was as effective as NaOCl against *E. faecalis* in an *in vitro* model at two tested contact times.

Clinical significance: Combination of MgO nanoparticles and garlic extract achieves disinfection comparable to gold standard NaOCl without harmful caustic effects of hypochlorite.

Keywords: *Enterococcus feacalis*, Nanoparticles, Natural irrigant, Root canal irrigant.

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Highlights

- Success of endodontic treatment relies on irrigation protocol.
- Sodium hydrochlorite (NaOCl) tissues when extruded beyond the canal.
- Garlic extract combined with magnesium oxide (MgO) nanoparticles used as an irrigant for prepared root canals infected with *Enterococcus faecalis*.
- Efficacy of this irrigant, at 2 and 5 minutes, equals to NaOCl.

Introduction

The success of endodontic treatment relies largely on the effectiveness of the instrumentation protocol, which then enables thorough cleaning of the root canal space. The effectiveness of the irrigating solution to disinfect the root canal thus estimates the success, longevity, and reliability of any endodontic procedure.[1] The outcome seems more favorable if the microbial remnants and necrotic tissue are eradicated completely before obturation.

Sundqvist and Fidgor famously published that "root canal infection is not a random event."[2] Microorganisms that invade the complex root canal space have shared characteristics that include the capacity to penetrate and invade dentin, a growth pattern of chains or cohesive filaments, resistance to antimicrobials used in endodontic treatment, also an ability to grow in monoinfections, to survive periods of starvation and to successfully evade the host response.

Enterococcus faecalis has proved to be a potentially important microorganism to colonize in endodontic infections and has been seen as the dominant microorganism in post-treatment apical periodontitis. In mixed infections, *E. faecalis* is typically the dominant isolate. Enterococci survive very harsh environments including extreme alkaline pH of 9.6. Also, *E. faecalis* can easily overcome the challenges of survival within the root canal system in many ways. It has been shown to exhibit widespread genetic polymorphisms. It possesses serine protease, gelatinase, and collagen-binding protein, which helps it adhere to dentin. Furthermore, *E. faecalis* is small enough to proficiently invade and live within dentinal tubules. It has the capacity to endure prolonged periods of starvation until adequate nutrition is available. It also has a proton pump to maintain pH homeostasis and thus can survive high pH.

However, studies regarding root canal irrigants are very few, which may be due to the fact that NaOCl has been the gold standard and has proven to be very effective in dissolving any remanent pulp tissue as well as disinfection of the complex root canal anatomy

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and effective destruction of all forms of microorganisms.[3–5] Also, NaOCl is so highly toxic, it causes necrosis when it comes in contact with normal tissues. This has led to a number of NaOCl accidents due to the extrusion of this caustic substance into normal tissue space.[6,7]

Consequently, in a recent search for alternative, safer products such as natural phytochemicals isolated from plants would be reasonable.[8] More so, due to their low toxicity, and better biocompatibility due to no added chemical products.

Natural ingredients tested recently include garlic, Aloe vera, and black mulberry as these have active phytochemicals that work as antimicrobials. *Allium sativum*, or garlic, has been extensively used in traditional medicine due to its anti-bacterial, anti-fungal, and anti-viral properties.[9] Its extract has been shown to have a wide antibacterial spectrum and can inhibit the growth of both Gram-positive as well as Gram-negative bacteria.

Magnesium oxide is an important inorganic material with a wide band gap. Also, MgO nanoparticles have shown promise for application in tumor treatment. They are a promising antibacterial agent due to their high resistance to harsh processing conditions. Three main antibacterial mechanisms have been proposed, which include the formation of reactive oxygen species (ROS), the interaction of nanoparticles with bacteria, subsequently damaging the bacterial cell, and an alkaline effect.[10]

Various studies have individually reported the efficacy of garlic extract as well as nano-MgO. Rao et al. in 2017 demonstrated the low pulp-dissolving ability of garlic extract,[11] whereas Birring et al. in 2015 demonstrated that garlic extract was able to disrupt biofilm as well as prevent colony formation of *E. faecalis*.[12] Whereas studies have been performed showing the action of nano-MgO against both Gram-negative and Gram-positive colonies. Kishen et al. compared 5.25% NaOCl, MgO nanoparticles at 5 mg/L and chitosan to compare their long-lasting efficacy in the eradication of *E. faecalis*.[13] He concluded that both chitosan and MgO showed superior results to the gold standard NaOCl irrigant.

Recent studies have shown the use of phytochemicals from plants, such as *Swertia chirayaita*, can be used to produce nanoparticles of MgO and show antibacterial activity against various pathogenic strains.[14] Garlic or *A. sativum* also has a similar reducing agent action to the *S. chirayaita* genus. Hence, in a search for newer non-toxic irrigants, a combination of garlic extract with MgO was proposed and to test its efficacy against *E. faecalis* present in root canals.

Materials and Methods

The approval for the research was obtained from the committee for the student's proposal, a constituent of the Institutional Ethical Committee of Sri Ramachandra Institute of Higher Education and Research (Deemed to be University), Chennai, Tamil Nadu, India (CSP/22/MAY/110/330). This study was conducted in accordance with the Declaration of Helsinki. This study was conducted from 1 August 2022 to 31 December 2022.

Study Design and Sample Size Calculation

An *in vitro* study design was selected. Based on a previous study, the sample size was calculated using G*Power 3.1.9.2, which indicated that 6 samples per group (total = 36 samples) would provide 80% power in determining statistically significant differences among the efficacy of the four irritants.[15] The α-value was set at 5% and the effect size was 0.6.

Thus six groups were formed, namely: group I—Saline irrigation, group II—NaOCl irrigation (3%; 2 minutes), group III—garlic extract with nano-MgO (2 minutes), group IV—garlic extract with nano-MgO (5 minutes), group V—garlic extract (5 minutes), and group VI—Nano-MgO (5 minutes). Each group consisted of eight samples that were randomly allocated.

Extracted Teeth Selection

A total of 48 human-extracted single-rooted single canal mandibular incisors with completely formed roots were collected and stored in 0.5% thymol. Teeth with fracture lines, decay, abnormal root canal morphology, and cracks or previous endodontic or restorative treatment were excluded. The teeth were measured and decoronated with a diamond disk to a specific length of 15 mm and stored till the instrumentation was performed.

Instrumentation of Teeth

The root apex was sealed with resin. The teeth were immobilized in wax, after which an endo access bur was used to gain access to the root canal space. Following this, instrumentation was performed after the glide path was established. Apical enlargement was performed till three sizes above the initial binding file after which circumferential filing was done.

Following this, irrigation was performed first with 1 mL 3% NaOCl and then 17% ethylenediaminetetraacetic acid (EDTA). The canals were finally flushed with saline solution.

Tooth samples were then sterilized using an autoclave for 30 minutes at 121°C. Also, 10 µL (10 microliter) of 1.5×10^8 colony-forming unit (CFU)/mL suspension of ATCC 29212 *E. faecalis* was used to incubate the root canals using sterile 1 mL syringes. The contaminated tooth samples were incubated for 48 hours and divided into 6 groups.[15]

- Group I—Saline irrigation (2 minutes)
- Group II—NaOCl irrigation (3%; 2 minutes)
- Group III—(Combination) Garlic extract with nano-MgO (2 minutes)
- Group IV—(Combination) Garlic extract with nano-MgO (5 minutes)
- Group V—Garlic extract (5 minutes)
- Group VI—Nano-MgO (5 minutes)

Irrigation was performed according to the group assigned.

Culture of *E. faecalis*

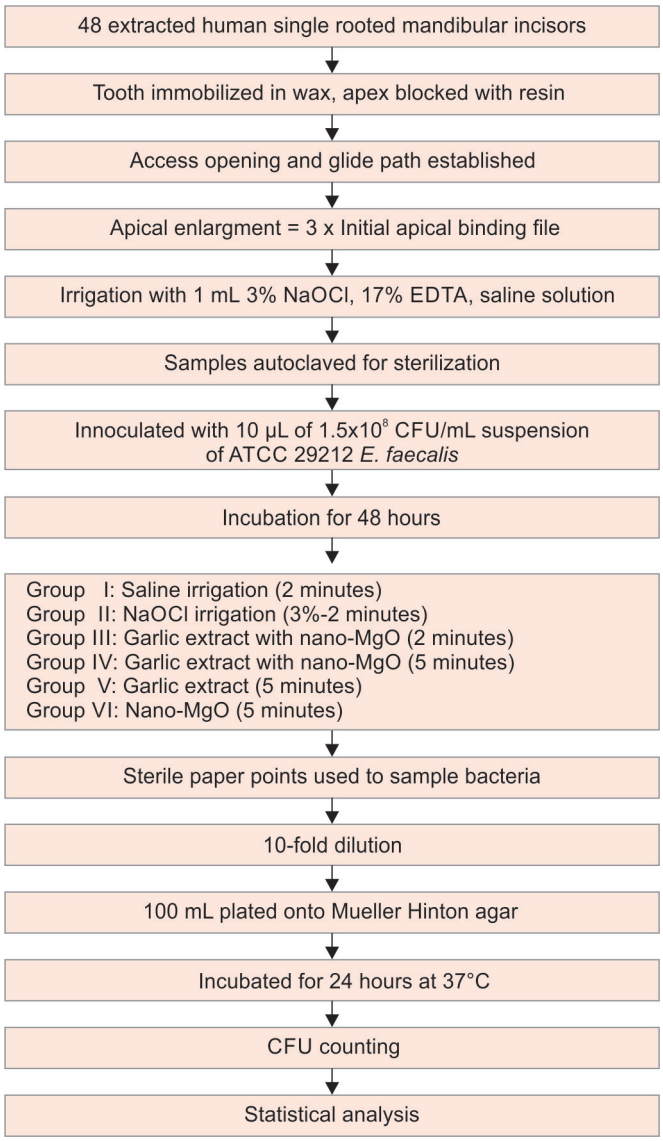
ATCC 29212 *E. faecalis* strain was obtained from pure culture from the Department of Microbiology (Sri Ramachandra University) and subcultured on a blood agar plate. Then 10 µL (10 microliter) of 1.5×10^8 CFU/mL suspension was created by dilution in BHI broth and CFU count accuracy was checked using a McFarland densitometer.

Nanomagnesium Oxide Aqueous Solution Preparation

Magnesium oxide nanoparticles, purity of 99.9%, particle size 30–50 nm were procured (Batch No. NRL01068574/252, Nano Research Lab, Jamshedpur, Jharkhand, India). It was manufactured using the chemical precipitation method.

A stock solution of 1 gm/L was prepared by adding 1 gm nanoparticle powder to 1 L ultrapure water. The nanoparticle stock solution was diluted with ultrapure water to prepare a concentration of 10 mg/mL Nano-MgO solution.[15]

Flowchart 1: Flowchart explaining methodology



Garlic Extract Preparation

Freshly peeled cloves of organically grown garlic were shade dried and powdered. A total of 100 gm of garlic bulbs was cleaned and crushed; 5 gm of garlic powder was macerated with 100 mL of distilled water. The homogenate was filtered using Whatman's filter paper No. 1. The supernatant was centrifuged at 10000 rpm for 20 minutes. This was stored at –20°C until use.[16]

Preparation of Combined Solution

As 1-mg extract from the garlic that had been filtered was mixed in 1 mL of aqueous nano-MgO vehicle and centrifuged at 5000 rpm for 10 minutes. The upper layer, which was clear, was used as the irrigant.

Culture Method

The sterile paper point method was selected to sample the bacteria from the root canals. The paper points were then transferred to tubes containing 2 mL of normal saline and serial 10-fold dilutions were made. Furthermore, 100 mL of the sample from each tube was pipetted and spread onto Mueller Hinton agar plates and incubated for 24 hours at 37°C. The colonies were then counted manually to obtain CFU/mL.[15]

Table 1: Cross-tabulations of groups

	CFU colonies			*Chi-square*		
Group	*0 Cells*	*1–10 cells*	*10–1000 cells*	*statistic*	*df*	*p-value*
I	0	0	8	43.81	6	0.0001
II	6	2	0			
III	4	4	0			
IV	6	2	0			
V	7	6	0			
VI	9	5	0			

Chi-square test. The test shows that there is a significant difference in colonies between the groups

Data Analysis

The CFU were counted on the agar plates manually and statistically analyzed using the statistical package for the social sciences (SPSS) software, version 28.0 (IBM, India). Non-parametric methods were applied to analyze data. To compare the mean values between groups, the intergroup analysis, Chi-square test, was used. The significance level was set at 5% ($\alpha = 0.05$). A flowchart explaining the methodology is provided in Flowchart 1).

The null hypothesis was set as "NaOCl was more effective as an antimicrobial agent than garlic–nano-MgO irrigant for root canals infected with *E. faecalis*."

Results

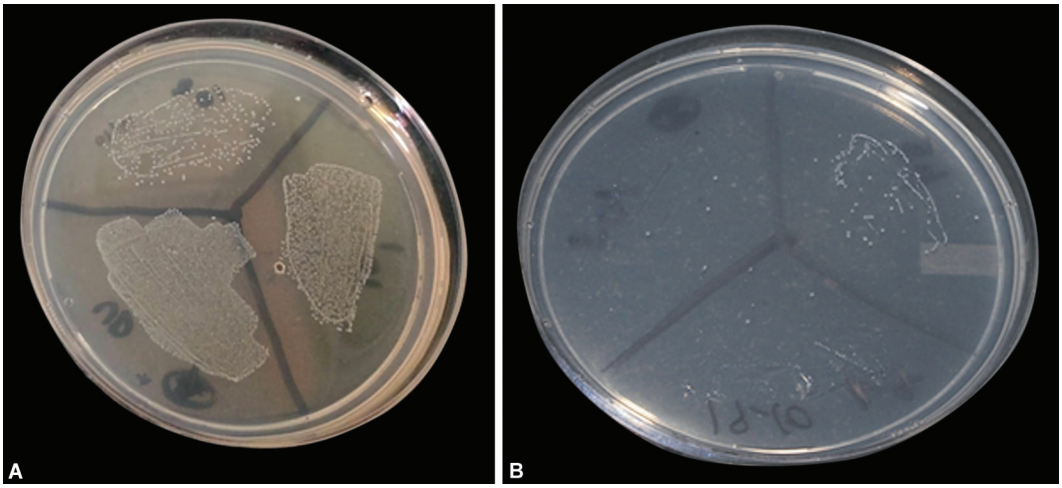
The efficacy of the irrigants was assessed by counting the CFUs on Mueller Hinton agar plates after final irrigation and an incubation period of 24 hours. The values obtained were calculated for mean and standard deviation and an intergroup comparison of the number of colonies between the groups was done.

Crosstabulation of the six groups was done (Table 1), and Chi-square test was applied for comparison. Statistical analysis showed that there was a significant difference in colonies between the groups with $p = 0.0001$.

Group I (Fig. 1A), where saline was used as an irrigant the plate showed an average of 110×10^3 CFU/mL. The CFU ranged from 110–208 CFU/mL, whereas when NaOCl (Fig. 1B) was used, the positive control showed an average of 0–1000 CFU/mL only as shown in Table 1.

Groups III and IV (Figs 2 and 3) in which garlic extract combined with nanoparticles of MgO were used as irrigant showed a mean value of 1–1000 CFU/mL and had no significant differences from the NaOCl group as shown in Tables 2 and 3, respectively.

Group V using garlic extract (Fig. 4A) showed an average CFU range of 53–75 CFU/mL. Group VI (Fig. 4B) using MgO nanoparticles showed CFU range of 34–63 CFU/mL. Thus, even though they were efficacious as sole irrigant. A combination of the two showed better efficacy. A pictorial comparison of CFU has been represented in Figure 5 after the elimination of negative control – group I (saline) (Fig. 5). The bacterial colony was confirmed under a light microscope (Fig. 6).



Figs 1A and B: Colonies formed on (A) MHA plate – Group I (Saline – 2 minutes) and (B) Group II (3% NaOCl – 2 minutes)

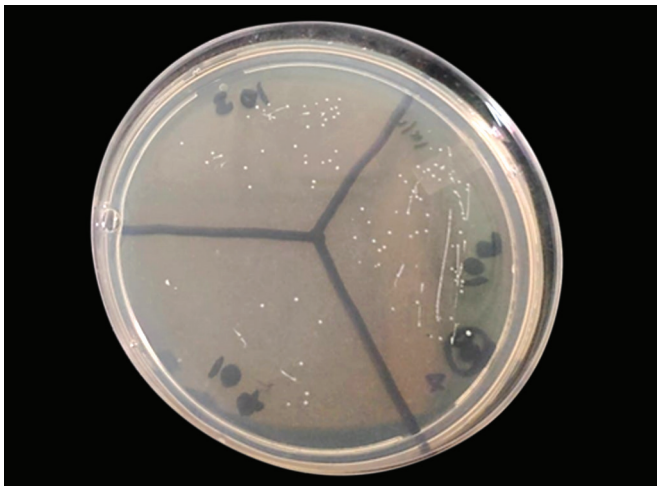


Fig. 2: Colonies formed on MHA plate – Group III (Garlic Extract + MgO NP – 2 minutes)

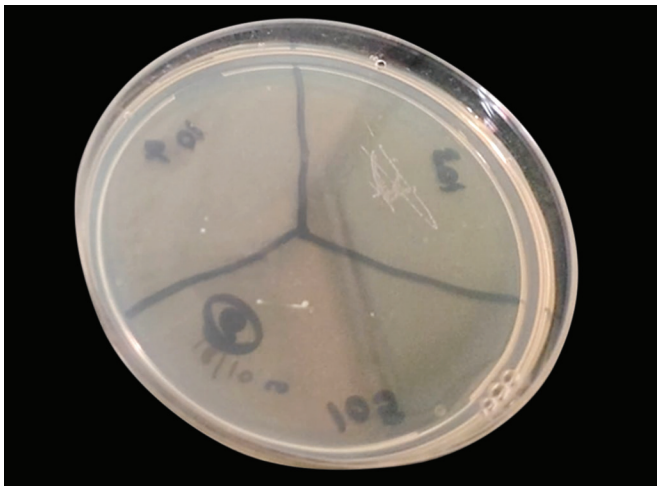


Fig. 3: Colonies formed on MHA plate – Group IV (Garlic Extract + MgO NP – 5 minutes)

From the intragroup analysis, it was derived that there were no differences between the eight samples in each group.

Table 2: Group II vs group III

Number of colonies	*Group II*		*Group III*		*Chi-square statistic*	*df*	*p-value*
	N	*%*	*N*	*%*			
0 cells	6	80.0	4	50.0	5.2	2	0.068
1–10 cells	2	20.0	4	50.0			
10–1000 cells	0	0	0	0			

There is no difference in efficacy noted between groups 2 and 3; both groups are similarly efficacious

Table 3: Group II vs group IV

Number of colonies	*Group II*		*Group IV*		*Chi-square statistic*	*df*	*p-value*
	N	*%*	*N*	*%*			
0 cells	6	80.0	6	80.0	0	2	1.0
1–10 cells	2	20.0	2	20.0			
10–1000 cells	0	0	0	0			

Groups II and IV activities are exactly similar and both groups are similarly efficacious

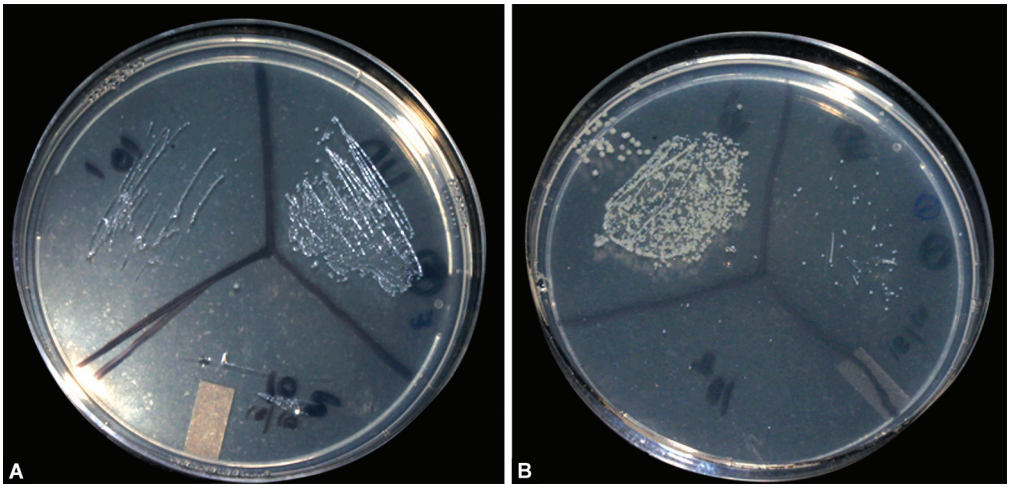
Thus, it was observed that garlic extract in combination with nano-MgO used as an irrigant was as efficacious as NaOCl against *E. faecalis*.

Hence, the null hypothesis was rejected.

Discussion

The basic goal of clinical endodontics is to clean and debride the root canal space, to remove any pathogenic microorganisms, necrotic tissue, and any remnant organic content. Creating completely sterile conditions is difficult. Even when done meticulously, mechanical preparation cannot cover substantial regions (>35%) of the canal walls, especially in the apical third of the root. As a result, chemical irrigation is critical for root canal disinfection.

The biological function of any irrigant is highly dependent on its antimicrobial action.[17] An ideal irrigant should have high efficacy against anaerobic and facultative microorganisms both in their planktonic state and in their biofilms, and against inactive toxins. The irrigant should also be nontoxic when they come in contact with vital tissues and should not cause anaphylactic reactions.[18]



Figs 4A and B: Colonies formed on (A) MHA plate – Group V (Garlic extract – 5 minutes) and (B) Group VI (nano-MgO – 5 minutes)

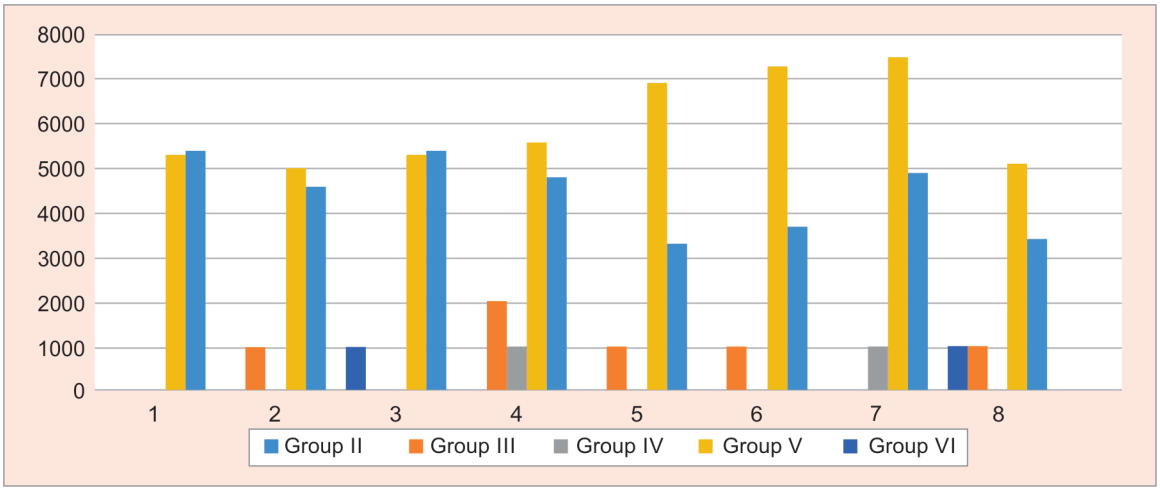


Fig. 5: Graph showing intergroup CFU counts after irrigation (after removing negative control – group I)

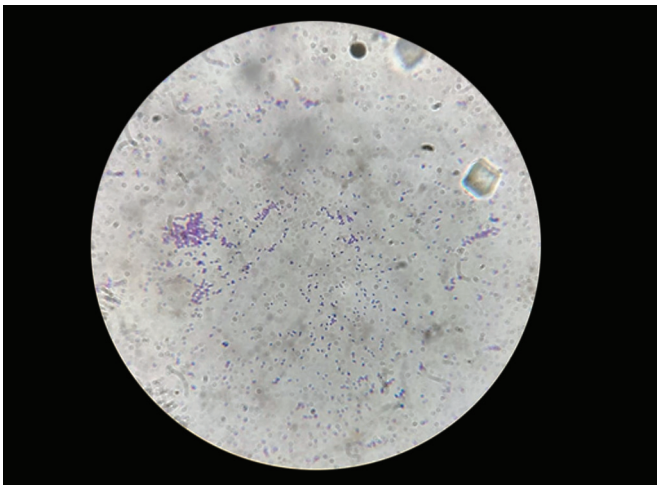


Fig. 6: Confirmation of bacterial colony under light microscope – Gram-positive cocci

A variety of irrigants have been used in combination to form an ideal irrigation protocol during root canal therapy, as presently no one solution can be regarded as optimal.[17]

Sodium hypochlorite is the most commonly used solution during endodontics.[19] This is due to its ability to dissolve necrotic tissue, vital pulp tissue, and organic part of biofilms in a matter of seconds.[4] Concentrations ranging from 0.5 to 6% have been used. After extensive research into the ideal concentration, it was established that lower concentrations of 2–3.5% may be used if the irrigant is agitated and activated to improve efficacy.[20] Time of contact has also been controversial, but a generally accepted contact time is 2 minutes with continuous replenishment.

The concern with NaOCl arose when it was seen that it has a highly caustic action on vital tissue. Other undesirable characteristics include unpleasant taste, allergic reactions, and hypochlorite accidents. If advertently extruded through the apex, severe reactions may occur. An episode can involve severe pain, edema, ecchymosis, profuse bleeding from the canal, paresthesia, and swelling of the face.[17]

Other irrigants available to endodontists include chlorhexidine, EDTA, hydroxyethylidene-1, 1-bisphosphonate (HBPT-1); however, none of these were as efficacious as hypochlorite to eradicate all necrotic organic tissue and microorganisms.

Researchers have been searching for treatments with natural and herbal products, and research has indicated that natural substitutes to be used for endodontic practice are quite

encouraging, given the unfavorable and inadequate specifications of existing irrigant solutions, the continuous enhancement in the number of strains resistant to solutions, and the side effects of synthetic medicines.[21]

Thus, in this study, the objective was to explore the antibacterial efficacy of a combination of garlic extract with nano-MgO.

Garlic (*A. sativum*) has been used since ancient times for its anti-inflammatory, antibacterial, and antioxidant characteristics that effectively limit infections while causing little damage to tissues. They not only help heal but they also protect by boosting host immunity.[22]

The antimicrobial constituent of garlic has been identified as the oxygenated sulfur compound, thio-2-propene-1-sulfunic acid S-allyl ester. This compound is also known as allicin.[23] Allicin is not present in raw garlic, it is formed when garlic is crushed by the enzyme alliinase lyase. The main antimicrobial action comes from the interaction with thiol-containing enzymes. These enzymes are essential for bacterial metabolism and thus, it has been shown that the development of resistance to allicin in bacteria arises 1000-fold less easily than to synthetic antibiotics.[24]

Allicin has been isolated in the pure state as a colorless liquid. Its pH in aqueous form is approximately 6.5 and the acidity slowly increases. The antibacterial action is not affected by the presence of p-aminobenzoic acid, thus again making it more efficacious than traditional antibiotics such as sulfonamides.[23]

Allicin shows a wide spectrum of action including action on *Escherichia*, *Salmonella*, *Staphylococcus*, *Streptococcus*, *Klebsiella*, *Proteus*, *Clostridium*, *Mycobacterium*, and *Helicobacter* species.[25] Garlic extract has shown high activity against Gram-negative bacteria and lower activity against Gram-positive organisms. Also, *E. faecalis* has a specific minimum inhibitory concentration (MIC) for allicin of 27.5 µg/mL.[26] Studies done by Nair;[27] Bokaeian and Bameri[28] among others, have demonstrated good antibacterial activity even against antibiotic-resistant strains and multi drug resistant (MDR) enterococci. Since Gram-negative bacteria are shown to be abundant in the apical one-third of root canal systems, this makes garlic extract ideal to be used for disinfection.

Allicin has been tried and tested for disinfection of the root canal space for the last decade. Birring et al. in 2015, assessed the penetration of the agent into the root canal dentin when used as an irrigant.[12] He demonstrated that 70% garlic extract shows as much dentin penetration as 5.25% NaOCl, making it quite efficacious.

Most importantly, it is imperative to preserve the potency of allicin in the garlic extract through an effective preparation process. Chavan et al. in 2010, have shown that heat deactivates allicin.[29] Thus, in this study, throughout the preparation, care was taken that the garlic was handled without subjecting it to any high temperatures.

Garlic extract as an irrigant has also been evaluated in clinical trials. Ghoddusi et al. used *A. sativum* as an irrigant in necrotic teeth.[30] The results highlighted that the efficacy of aqueous garlic extract was almost comparable to 2.5% NaOCl since the CFU count was low in both groups. Siddique et al. conducted a double-blinded clinical trial with garlic in combination with lemon extract, and the results showed bacterial reduction rates higher than even NaOCl.[31]

Hence, in this study, fresh garlic was crushed with a sterile mortar, then filtered to obtain an extract, which was then centrifuged at 10000 rpm for 20 minutes to precipitate the garlic particles, with reference to a previous study.[16] Sterilization was ensured through ultrafiltration. Due to apical complexities that exist in the root canal systems, it is preferable to have a more aqueous vehicle to carry a bioactive substance to the apex. Included a vehicle that has some bioactive properties may allow for supplemented antimicrobial activity of the irrigant. Thus, in this study, a slurry of MgO nanoparticles was combined with the garlic extract. Furthermore, a 10-mg/mL concentration of nano-MgO powder in distilled water has inherent antimicrobial properties, which have been tested *in vitro* in several studies.[15,32]

Metal oxides when manufactured as nanoparticles have high surface area due to the numerous edges and corners. This allows for the destructive adsorption of halogens, which are well-known bactericides. Halogens cannot be solely used due to their high toxicity and vapor pressure, but when doped on metal nanoparticles they become biocompatible.[33] Various metal oxides have been tested by antibacterial efficacy including zinc oxide, MgO, and calcium oxide, however, MgO has shown higher activity than its counterparts.[34] Koper et al., in 2002, used nano-MgO against *Escherichia coli*, *Bacillus megaterium*, and *Bacillus subtilis* and observed its activity under various microscopic techniques, which showed immediate coagulation of bacteria upon contact.[33]

Nano-MgO has good bactericidal performance in an aqueous environment due to the formation of superoxide anions on its surface. Thus, it shows high efficacy against bacteria, spores, and even viruses.[33] Also, MgO is readily hydrated and forms a layer of $Mg(OH)_2$ on its surface, oxygen then dissolves in the solution and generates superoxide anions by single electron reduction, which is very stable in a basic environment.[35] As the surface area increases and the MgO because smaller the amount of hydroxide (OH) on the surface increases and activity increases. Recently, the use of these superactive nano-MgO has found its way into endodontics.

In vitro models have shown that nano-MgO represents more efficient and long-term antibacterial activity comparable to gold standard NaOCl 5.25%.[32] When toxicity was assessed against HeLa cells 24-hour results showed a minimal effect.[36–38] The Minimum inhibitory concentration of MgO was calculated as 10mg/mL against *E. faecalis*. It has been shown to disrupt biofilm growth even seen in SEM analysis.

Since both MgO and garlic extract have proven antibacterial properties, previously tested in several studies, it was decided to test its combination. Recently, studies have shown that plants with phytochemicals and reducing agents, such as *S. chirayaita* and *Rhizophora lamarckii*, are able to photosynthesize and enhance the effect of inorganic nanoparticles.[14,39] Since *A. sativum* has similar reducing properties like genera of *Swertia* (Gentianaceae) and *Rhizophora* (Rhizophoraceae), it was thought that this combination will also act synergistically and enhance the antibacterial properties of MgO nanoparticles.

In the current study, the efficacy of the irrigant was evaluated in an *in vitro* model. The root canal spaces of prepared teeth were inoculated with 10^1 concentrations of 1.5×10^8 CFU/mL suspension of *E. faecalis*, to obtain single colony biofilms for antibacterial efficacy evaluation.

Sodium hypochlorite and saline were chosen as positive and negative controls respectively due to their role as a gold standard. Colony-forming units were counted after plating samples that were collected using paper points from the inoculated root canals. Paper points are the easiest method to collect bacterial samples from a prepared tooth. Furthermore, CFU counting was chosen as the method of analysis because it gives a quantitative and definitive number of colonies present, in comparison to using more qualitative methods such as PCR.

Two contacts times—2 minutes and 5 minutes—of this new irrigant were tested. Results showed a significant reduction in CFUs with contact times of both 2 minutes[27] and 5 minutes. Saline showed an average of 132.8×10^3 colonies/mL, whereas hypochlorite showed around 1–3 colonies/mL. The tested irrigant was as efficacious as hypochlorite both at 2 and 5 minutes contact times, showing colonies around 1–2 colonies/mL.

The already proved MIC of 10 mg/mL was combined with a fresh garlic extract and used for irrigation. This was comparable to our gold standard NaOCl. There was no statistical difference between the hypochlorite group and nano-MgO combined with garlic extract groups.

Due to the already proven biocompatibility, ease in procurement and extreme economic factors, and lack of fear of toxic accidents on extrusion, this novel irrigant may have a promising role to play in the irrigation of infected root canals.

Hence, this *in vitro* study clearly shows garlic extract combined with MgO nanoparticles have a comparable if not superior activity to NaOCl even at a short contact time of 2 minutes. This activity is seen without the fear of untoward effects if the irrigant is extruded from the apex, unlike hypochlorite. Thus, this irrigant may have a promising place in the endodontic armamentarium.

However, since this study was performed *in vitro*, long-term biological and toxicological effects, proper dosage, and surface area to particle ratio are required to be determined in order to come to proper conclusions about the dosage and long-term application of metal oxide nanoparticles in clinical practice. In the future, these long-term effects and toxicities should be assessed for translating the use of this irrigant into clinical practice.

This novel irrigant was thus an attempt to combine easily available, natural ingredients with a low propensity to cause toxicity and maybe be the answer to replace NaOCl, in turn reducing the fear of extrusion accidents. Thus, clinicians may appreciate an irrigant with lesser negative effects but equal efficacy of NaOCl.

Conclusion

This *in vitro* study assessed the use of a novel irrigant in root canals inoculated with *E. faecalis*. The combination of garlic extract combined with MgO nanoparticles was efficacious against *E. faecalis* and significantly reduced colony count when used in extracted human teeth. The use of a bioactive vehicle such as MgO enables superior efficacy of an irrigant. This novel irrigant was as efficacious as the gold standard, NaOCl at even a 2-minute contact time.

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