

# Comparative Evaluation of Antibacterial Activity of Tooth Pastes with Herbal Formulation, Probiotics and Fluoride Against Oral Pathogens

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## ABSTRACT

**Background:** In recent years, the negative consequences of fluoride toothpaste have come to the attention of consumers, and herbal products have gained popularity as alternatives in the fight against tooth decay. Studies have concentrated on the advantages of herbal essential oils due to their antibacterial properties.

**Aim:** To compare the antibacterial properties of toothpaste with fluoride, probiotics and fluoride against oral pathogens.

**Materials and methods:** Antimicrobial activities of probiotic, fluoridated and herbal toothpastes were assessed at different concentrations. Antimicrobial property was evaluated by determining the zone of inhibition using agar well diffusion method. The plates were incubated for 24 hours at 37°C. After the incubation time the zones of inhibition were measured.

**Results:** It was observed that the sample with probiotics and herbal formulation had higher zones of inhibition than the sample with fluoride.

**Conclusion:** From the results, it can be concluded that usage of toothpaste with probiotics and herbal formulation is more efficient than toothpaste with fluoride

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## INTRODUCTION

One of the most prevalent health issues in the world, dental caries is a chronic condition that damages tooth tissue and can have a negative impact on chewing and aesthetics. The fact that caries do not develop in the absence of plaque or fermentable carbohydrates is the most crucial factor in the development of dental caries. Numerous elements are thought to be potential sources for the emergence of caries, including plaque, cariogenic microbiota, fermentable carbohydrates, and duration(1). These elements work along with host vulnerability to cause dental caries, and the acidogenic bacteria frequently use sucrose as a substrate. Traditional therapy procedures that incorporate alcohol and antibiotics including ampicillin, penicillin, erythromycin, and chlorhexidine have been shown to be effective in preventing dental caries(2). These disorders will advance if the bacteria that make up biofilms become more resistant to drugs. It is being aimed to produce anti-infective drugs that are active against microorganisms as a result of the resistance of bacteria to antibiotics and conventional treatment approaches.(3)

## KEYWORDS:

Toothpaste, antibacterial property, probiotics, herbal formulation, fluoride.

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The most frequent substances utilised to affect bacterial viability in biofilms are antibiotics. Miller claimed that antiseptics may be utilised as an active agent in the prevention of caries while researching the causes of tooth decay. Fluoride-containing dentifrices with great clinical performance are one of the most effective oral hygiene techniques available.(4) Fluoride-containing substances, however, can alter oral and intestinal flora, as well as result in teeth discoloration, vomiting, and mouth cancer.(5) Other antimicrobial agents, such as chlorhexidine, amine fluorides, cetylpyridinium chloride, and products containing such substances, that are used in the treatment and prevention of dental disorders have severe side effects and discolour teeth. Commonly found in mouthwashes, ethanol may increase the risk of developing oral cancer.

In rural areas of underdeveloped nations, herbal remedies are the primary source of healthcare. Many active biological components that can be used to create new compounds for medications are derived from natural products derived from medicinal plants. Herbal medicines' antibacterial, antiviral, and anti-inflammatory properties have made their way into dentistry.(6) Others focused on the prevention of biofilm development, lowering the microbial adhesion that is principally responsible for dental plaque formation. Some studies looked at the impact of plant extracts and plant products on particular oral infections. It has been investigated if plant extracts, essential oils, and phytochemicals can treat or prevent bacterial adhesion. Due to their antibacterial properties against a variety of bacteria, including *S. mutans*, essential oils can be employed.(7)The components of essential oils, such as terpenes and terpenoids, which all have a low molecular weight and aromatic and aliphatic contents, can be used to explain these bactericide or bacteriostatic properties.

## MATERIALS AND METHODS

This study was conducted in Orange Lab at Saveetha Dental College with different toothpastes containing fluoride (sample A), herbal combination(sample B), and probiotics (sample c). Toothpaste with different herbal formulations and probiotics. were chosen for the study and it is compared to toothpaste with fluoride. Muller hinton agar was prepared and sterilised for 15 minutes at 121°C. Media was poured into the sterile petri plate and was left for solidification.

### Isolation and characterization of *Streptococcus mutans*

The samples in the tubes were vortex mixed for a minute to allow the dispersion of bacteria into the medium. Then, 0.1 ml of undiluted inoculum was spread on mitis salivarius bacitracin agar medium. The plates were kept in anaerobic conditions at

37°C for 24-48 h. The colonies on MSB plates were subcultured on a medium containing 5% sheep blood. The colonies having alpha hemolysis were chosen for further identification after a 24-hour incubation period at 37°C.

### Antibacterial Activity

The toothpastes were diluted in sterile water to concentrations of 100 mg/mL, 50 mg/mL and 25 mg/mL. Antimicrobial activity of toothpaste with different formulations was assessed against *S. mutans* using modified agar well diffusion technique. A standardised inoculum matched with McFarland 0.5 turbidity standard of the isolate was inoculated onto a Mueller Hinton agar plate with a sterile cotton swab stick. The plates were dried for 15 minutes. Using a sterile cork borer of 10 mm in diameter, wells were bored into the inoculated agar plates and the different concentrations of the toothpastes were added to the wells, in a volume of 0.2 mL. Sterile distilled water was added into a separate well as a control. The plates were left for 40 minutes for pre-diffusion and then incubated at 37 °C for 24 hours. Zones of inhibition were measured in millimetres

## RESULTS

The antibacterial activity of toothpastes were tested against *S. mutans* and the results are given in the table below. The Fig 1 shows 3 different dentifrices, demonstrating various levels of antimicrobial activity against tested isolated *S. mutans*. Among all the tested toothpastes, Sample B (Herbal toothpaste) and SampleC ( probiotic toothpaste) had a zone of inhibition of 18mm compared to other toothpastes with fluoride. Sample A ( fluoridated toothpaste)was found to be the less effective dentifrice out of all those tested, however there weren't many variances.



Fig 1: Antibacterial activity of toothpaste with different formulations against *S. mutans*

**Table 1:** Zone of inhibition ( in millimetre diameter) of toothpaste with different formulations against S.mutans

Sample	Zone of inhibition
Sample with probiotics	18mm
Sample with fluoride	17mm
Sample with herbal formulation	18mm

## DISCUSSION

High levels of cariogenic bacteria are seen in dental caries, a multifactorial illness. In the current investigation, *S. mutans*, the most often isolated bacterium from human dental plaque, was employed since it is thought to be the primary cariogenic microbe for dental caries. The acids of *S. mutans* and the by-products of carbohydrate metabolism are responsible for the loss of superficial tooth structures.(8) Due to its virulence characteristics, primarily its adhesion and acidogenicity, *S. mutans* has the ability to cause caries. *S. mutans* has the potential to cause caries, hence oral hygiene practises are utilised to lessen its buildup on oral biofilm. Periodontal disease, gum inflammation, and cavities can all be reduced with professional dental care and teeth brushing techniques. It is clear from the rise in these oral illnesses' prevalence around the globe that brushing alone is insufficient to stop tooth decay. Because of this, chemotherapeutic supplements may be added to the regular brushing procedure. Fluoride and triclosan are common ingredients in toothpastes that are suggested by the WHO (World Health Organization), ADA (American Dental Association), and FDI (World Dental Federation). It has been demonstrated that the use of triclosan in toothpastes lowers gingiva and plaque index scores as well as in vivo bacterial viability.(9)

Several plants that have carvacrol as a key structural element are referred to as oregano. *Origanum* species are a rich source of chemicals with insecticidal, antibacterial, antifungal, antioxidant, and anti-carcinogenic effects, according to phytochemical and biological research(10). Numerous investigations have been done on the antibacterial properties of the *Origanum* species. *O. dubium* was tested in earlier research for its antibacterial effectiveness against a variety of bacterial strains, including *E. coli*, *S. aureus*, *C. albicans*, and *S. mutans*. Depending on the sorts of phenolic chemicals they contain, *Origanum* species are classified. Carvacrol and thymol are two of oregano oil's primary ingredients. They are monoterpenic phenols that are produced by osmosynthesizing p-cymene with -terpinene.(11)

To ensure that the pure essential oils utilised in this investigation were effective, they were put to agar plates, before adding them to the examined toothpastes, they had not been contaminated. A measurement of the diameter of the pure oils' inhibitory zones were particularly sizable. This is explained by the fact that only pure oxidative stress is a result of essential oils. (12)

A rising trend is the use of natural remedies to treat various illnesses. Modern medicine has benefited greatly from the use of herbal remedies. There are a lot of herbal toothpastes on the market, and they all tout their efficacy. Instead than just assuming that a new product is effective based on manufacturer claims, it is crucial to confirm its effectiveness through clinical or in vitro testing.(13) Therefore, the objective of the current study was to assess the efficiency of herbal toothpastes from specific brands against *S. mutans*. The majority of research have contrasted the effectiveness of toothpaste with a herbal base with toothpaste without. Six toothpastes made from herbs were tested for their anticariogenic properties in an earlier study. Studies have shown that there is no discernible difference in the anticariogenic properties of herbal and conventional toothpastes(14). Choosing herbal toothpaste over regular toothpaste with harsh chemicals is a sensible choice. In the previous study, it was found that *S. mutans*, a cariogenic bacteria, was inhibited by all of the toothpastes examined(15-24)

From the above results, it is observed that the sample with probiotics and herbal formulation had more zone of inhibition than the sample with fluoride sample. So it is preferred to use toothpastes which contain natural herbal ingredients like clone, ginger etc, and even probiotics. They enhance the capability of tolerance of the oral pathogens within our oral cavity and give us protection against various oral diseases.

## CONCLUSION

*S. Mutans* was shown to be the most common type of bacteria in people with dental plaque. Significantly, the toothpaste with herbal combination and toothpaste with probiotics had high zone of inhibition than the toothpaste with fluoride combination.(25) The toothpaste with probiotic and herbal formulation was found to be effective among all the tested dentrifices. Numerous dentrifices have antibacterial activity, but more study is needed to understand how it works. Comprehensive pharmacological investigations are also needed to assess the toxicity of these products as well as their clinical usefulness for use in the prevention and treatment of dental caries. Although several plant extracts and phytomedicines have been shown to be effective in dentistry thus far, future research should take the products' safety and potential adverse effects into account.

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### Authorship contribution

SB compiled the manuscript RVG conducted the study LT designed the study

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