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# EFFECT OF CURCUMA LONGA EXTRACT ON BIOFILM FORMATION BY STREPTOCOCCUS MUTANS

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## ABSTRACT

Objective: To find the effect of Curcuma longa extract on biofilm formation by Streptococcus mutans.

Methods: The organism *S. mutans* was isolated from saliva sample using special media (*Mutans* - sanguis agar) and maintained in tryptone soya agar at 4°C in Department of Microbiology, Saveetha Dental College and Hospitals.

**Results:** From the study, we infer that 78.35% of inhibition of the biofilm formation is seen with 100 ug of curcumin extract. From the result, it is evident that curcumin has a very good inhibitory effect on *S. mutans* growth.

Conclusion: The study concludes the inhibitory action of curcumin on S. mutans by preventing biofilm formation.

Keywords: Curcuma, Streptococcus mutans, Biofilm.

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# INTRODUCTION

Streptococcus mutans is one of the main causative agents for dental caries, and thereby, studies are being done inhibiting its growth in the oral cavity by usage of different antibacterial agents. Many members of the genus Streptococcus that cause infections in humans use quorumsensing systems which help to regulate several physiological properties, including incorporating foreign DNA, resisting acids, forming biofilms, and becoming virulent [1]. The dental plaque biofilm grows on all surfaces in the oral cavity, including the teeth, mucosa, and all inserted materials. Dental restoration materials are widely used in the treatment of dental caries [2]. The biofilm can shear off, multiply, disperse, and colonize as planktonic individuals causing relapse and chronic infection, which become a major concern for public health [3]. S. mutans produces glucosyltransferase enzymes that synthesize glucan from the glucose moiety of sucrose that causes the cariogenicity of the dental pathogens [4]. Herbal medicines are more effective and less harmful as they have negligible side effects and show low mammalian toxicity [5,6]. Curcuma longa is a rhizome of ginger family which is native of the South Asia and has a long history of usage from dying to medicinal products. It is also found to have been used in traditional cooking. The beststudied compound is curcumin, which constitutes 3.14% (on average) of powdered turmeric [7]. Both curcumin and the oil fraction suppress the growth of several bacteria such as Streptococcus, Staphylococcus, and Lactobacillus [8], and aqueous extract of turmeric has also shown antibacterial effects [9]. Turmeric oil is also found to have antifungal effects on Aspergillus flavus and Aspergillus parasiticus [10]. This study is done in order to verify the antibacterial effects of Curcuma longa on S. mutans.

### METHODS

The organism *S. mutans* was isolated from saliva sample using special media (*Mutans* – sanguis agar) and maintained in tryptone soya agar at 4°C in Department of Microbiology, Saveetha Dental College and Hospitals.

#### Methodology

Overnight, grown cultures of *S. mutans* from agar plates were inoculated in tryptone soya broth and incubated at 37°C. Individual wells of sterile

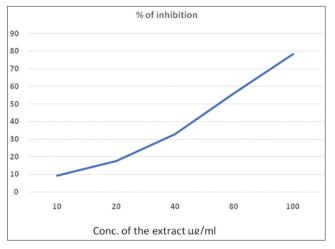
polystyrene 96 well flat bottom microtiter plates were filled with 200  $\mu$ l of culture suspension of the test organism. Uninoculated liters of the curcumin was added from the prepared stock solution of 10, 20, 40, 80, and 100  $\mu$ g/ml, respectively, and incubated at 37°C for 24 hrs. After incubation, content in the wells was removed, washed with 0.2 ml phosphate buffer saline to remove free-floating bacteria. The adherence of the bacteria was fixed with sodium acetate (2%) and stained with crystal violet. The crystal violet was removed, and 250  $\mu$ l of acetone was placed in each well to release the crystal violet. Then, finally, the readings were taken. Optical density (OD) of stained adherent bacteria was determined with an enzyme-linked immunosorbent assay reader (Bio-Rad) at wavelength 570 nm. These OD values were taken as index of bacteria adhering the surface and formed biofilm. The experiment was carried out in triplicate, and their mean was taken for the analysis.

#### RESULTS

On addition of 100 ug/ml curcumin extract into the culture plates of *S. mutans,* it shows 78.35% of inhibition of the biofilm formation. (Graph 1) (Table 1) From the result, it is evident that curcumin has a very good inhibitory effect on *S. mutans* growth (Table 2).

#### DISCUSSION

Since curcumin has good inhibitory effects of *S. mutans*, it is recommended to be an important constituent of toothpastes and oral aids as *S. mutans* is a dangerous causative agent of dental caries and helps prevent growth in the oral cavity. Curcumin [1, 7-bis (4-hydroxy-3-methoxyphenyl)-1, 6-heptadiene-3, 5-dione; diferuloylmethane], a yellow bioactive pigment, is the major component of turmeric which is a dried rhizome and is a rich source of beneficial phenolic compounds [11]. Curcumin is described as prebiotic, which can have beneficial effects on human health [12]. Other research data have showed that the curcuminoids may be effective in controlling dental biofilms and dental cavity formations, suggested that turmeric extracts can be extensively used in the treatment premalignant lesions in the oral cavity [13]. These antibiotics are sometimes associated with adverse effects on the host including hypersensitivity, immunosuppression, and allergic reactions which leads, there is a significant demand to develop



Graph 1: Percentage inhibition versus concentration of extract

Table 1: Biofilm formation and its OD values

Mean OD values	Adherence	<b>Biofilm formation</b>
<0.120-0.240	Non	Non/weak
0.120-0.240	Moderately	Moderate
>0.240	Strong	High

OD: Optical density

Table 2: Inhibition percentage with increase in concentration of curcumin

Concentration of the extract ug/ml	Percentage of inhibition
10	09.15
20	17.37
40	32.90
80	56.05
100	78.35

an alternative antimicrobial drug for the treatment of infectious diseases from medicinal plants [14,15]. It was found that the anti-adhesive effect of curcumin against S. mutans is mediated through collagen and fibronectin. These results support the widespread use of curcumin as a food-based antimicrobial agent [16]. S. mutans could outcompete other species, and occupy additional regions of the mouth, and cause advanced dental plaques, which can be as acidic as pH 4.0 [17]. At 1% concentration, curcumin nanoparticles (curc NPs) have significant antimicrobial activity against cariogenic bacteria with not much adverse effects. However, the insolubility of curc NPs remains a major disadvantage [18]. The antibacterial activity of curcumin is credited to the destruction of peptidoglycan cell wall of bacteria [19]. Turmeric mouthwash can be effectively used as an adjunct to mechanical plaque control methods as 10 mg of curcumin can be dissolved in 100 ml distilled water, and the flavor may be enhanced by using peppermint oil [20]. The killing effect was shown to be dependent on curcumin concentration, radiant exposure, post-irradiation incubation time, bacteria species, and pharmaceutical preparation [21].

#### CONCLUSION

Thus, from the current study, curcumin is found to have very good anti-bacterial effect on *S. mutans* and can be advised to be in prevention of dental caries and also in endodontic procedures.

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