

# Comparison Of The Effect Of Calcium Sodium Phosphosilicate (CSPS) And Fluoride-Containing Desensitizing Agents On Bacterial Biofilm Formation- An In Vitro Study.

Alka Joseph<sup>1</sup>, Gauri SarDessai<sup>2</sup>, James Samuel<sup>3</sup>, Deepthi M<sup>4</sup>

<sup>1</sup>(Periodontics, Goa Dental College and Hospital, India)

<sup>2</sup>(Periodontics, Goa Dental College and Hospital, India)

<sup>3</sup>(Periodontics, Goa Dental College and Hospital, India)

<sup>4</sup>(Oral Medicine and Radiology, Goa Dental College and Hospital, India)

## Abstract:

**Background:** In Dental Practice, Dentine Hypersensitivity (DHS) Is A Prevalent Clinical Concern Characterized By Short, Sharp Pain Arising From Exposed Dentin In Response To Non-Noxious Stimuli. A Desensitizing Agent Used To Alleviate DHS Can Also Influence The Bacterial Biofilm. Our Study Compares The Impact Of CSPS And Fluoride-Containing Desensitizing Toothpastes On The Formation Of Streptococcus Mutans Biofilm Based On The Assumption That Any Changes To The Tooth Surface May Have An Effect On Subsequent Biofilm Formation. The Present Study Might Be The First To Compare The Effects Of CSPS And Fluoride Containing Dentifrices On Biofilm Formation.

**Materials And Methods:** Dentine Discs (15) Obtained From The Coronal Portion Of Extracted Premolars & Molars Were Treated With 17% EDTA And Divided Into 3 Groups: Control, 5% CSPS Toothpaste, And 917ppm Fluoride Toothpaste. The Discs Were Immersed In Toothpaste Slurries For 2 Minutes, Twice Daily For 7 Days Followed By Incubation With Streptococcus Mutans For 24 Hours To Assess The Number Of Colony-Forming Units (CFU).

**Results:** The Control Group Had The Highest Mean CFU Level Of All Groups,  $35.07 \pm 1.39$ . The Lowest Was  $25.33 \pm 2.61$  In The CSPS Group. While In The Fluoride Group, The CFU Count Was  $31.47 \pm 1.92$ . A One-Way ANOVA Test To Compare Groups Was Found To Be Statistically Significant ( $P < 0.001$ )

**Key Words:** Streptococcus Mutans, Bacterial Biofilm, Calcium Sodium Phosphosilicate Toothpaste, Fluoride Toothpaste, Dentin Hypersensitivity

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## I. Introduction

As defined by the American Dental Association, dentin hypersensitivity (DHS) is a short, sharp pain that occurs from exposed dentin in response to non-noxious stimuli, such as thermal, evaporative, tactile, or osmotic stimuli, and cannot be linked to any other type of dental defect<sup>1</sup>. DHS is a common clinical scenario in dental practice and is usually associated with dentin surface exposure.

A desensitizing agent must be incorporated into dentifrices for therapeutic purposes in order to occlude exposed dentinal tubules and alleviate the discomfort caused by dentinal hypersensitivity. Apart from alleviating pain, these compounds can alter dentin surface layers by occluding tubules, influencing bacterial biofilm development. Consequently, the risk of periodontal diseases and dental decay can be reduced. Further, enamel and dentin remineralizations are improved by these agents<sup>2</sup>.

In vitro studies have shown that calcium sodium phosphosilicate (CSPS) deposits a layer between 3 and 5 mm thick over dentin after treatment, altering the entire tooth surface<sup>3</sup>. The layer may be thinner in vivo, but any changes to the tooth surface could affect the formation of salivary pellicles, which might affect bacterial adhesion<sup>4</sup>.

Another desensitizing agent, Fluoride inhibits the bacterial enzyme enolase leading to the suppression of carbohydrate metabolism by the acidogenic microflora present in plaque<sup>5</sup>.

Previous researches had a primary focus on assessing the dentinal tubule patency in order to investigate the desensitizing property of various kinds of toothpaste. Assuming that any changes to the tooth surface may have an impact on the microbial biofilm that forms and may also have the additional benefit of reducing the risk of developing caries, our study aims to compare the effects of CSPS and fluoride-containing desensitizing

toothpaste on the formation of Streptococcus mutans biofilm. This might be the first study to compare the effect of two distinct dentifrices on biofilm.

## II. Material And Methods

The study was conducted in the Department of Periodontics, Goa Dental College & Hospital, and was approved by the Goa Dental College & Hospital Institutional Ethics Committee. Microbiological analysis was done at Maratha Mandal's Central Research Laboratory, Belgaum, Karnataka.

### Sample preparation

A total of 15 extracted molars and premolars were collected from the Oral Surgery Department. Diamond discs were used to prepare dentin discs measuring 4mm X 4mm and 1mm in thickness from the collected extracted teeth. To simulate a sensitive dentin, the discs were immersed in EDTA solution (17%) at a pH of 7.4 [Figure 1]. The samples were divided into 3 groups: [Figure 2].

Group 1 (5 discs): No treatment

Group 2 (5 discs): Treatment with desensitizing toothpaste containing CSPS 5% (SHY-NM)

Group 3 (5 discs): Treatment with desensitizing toothpaste containing fluoride 917 ppm (SHY-XT)

The dentin discs were immersed in toothpaste slurries created by using 1 part of distilled water and 3 parts of the tested toothpaste, for 2 minutes, twice daily for 7 days. The discs were rinsed with and stored in distilled water [Figure 3].

### Biofilm preparation and Streptococcus Mutans analysis

The discs were sterilized using an autoclave and later incubated with Streptococcus mutans (pure form) for 24 hours in brain heart infusion broth (BHI). The number of colony-forming units (CFU) was counted after the inoculum was cultured on BHI agar plates [Figure 4-6].

### Statistical analysis

The software used for statistical analysis was STATA. The distribution of the sample was checked using the Shapiro-Wilk Normality test. One-way ANOVA was used for inter-group comparison followed by pair-wise comparison of groups. p-value <0.05 was considered statistically significant.

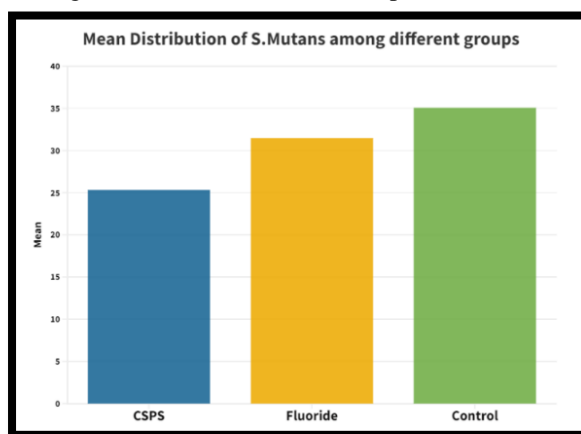
## III. Result

The control group had the highest mean CFU level of all groups,  $35.07 \pm 1.39$ . The lowest was  $25.33 \pm 2.61$  in the CSPS group. While in the fluoride group, the CFU count was  $31.47 \pm 1.92$ . A one-way ANOVA test to compare groups was found to be statistically significant ( $p < 0.001$ ) [Table 1, Graph 1].

**Table no 1:** Descriptive table showing the mean, standard deviation, normality and One-way ANOVA test for different groups.

Group	N	Mean	Standard Deviation	Shapiro Wilk W	Shapiro Wilk p	One way ANOVA	
						F	p
CSPS	15	25.33	2.61	0.94	0.392	82.34	<0.001*
Fluoride	15	31.47	1.92	0.86	0.023		
Control	15	35.07	1.39	0.91	0.140		

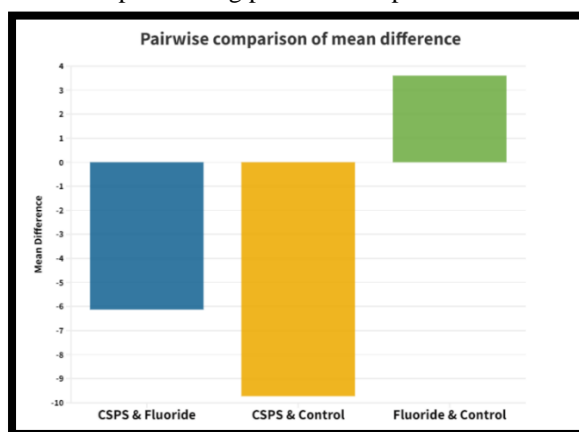
**Graph no 1:** Graph showing the mean distribution of streptococcus mutans among different groups



**Table no 2:** Tukey's post hoc pairwise comparison of groups according to the S. mutans count

Dependent Group (I)	Dependent Group (J)	Mean Difference	P
CSPS	Fluoride	-6.13	<0.001*
CSPS	Control	-9.73	<0.001*
Fluoride	Control	-3.60	<0.001*

**Graph no 2:** Graph showing pairwise comparison of mean difference



### III. Discussion

A species-rich heterogeneous ecological system colonizes the oral cavity, encompassing numerous small microbial habitats. The main inhabitants of this ecosystem are bacteria. Streptococcus mutans is the main component of the oral microbiota and dental plaque. It is also the primary pathogen implicated in the pathogenesis of dental caries. Hence, Streptococcus mutans biofilm was introduced to the discs in our current study<sup>6</sup>.

The salivary pellicle that forms on dentin as a result of the selective adherence of salivary biopolymers such as glycoproteins may be thinner in vivo, but any modification to the tooth surface may have an effect on this layer. When CSPS comes into contact with an aqueous environment, an outer layer of carbonated hydroxyapatite crystals forms on dentin [4]. This might interfere with bacterial adhesion to the pellicle and subsequent co-adhesion of other bacteria to the pioneer microorganisms, forming polymicrobial biofilms<sup>7</sup>.

Fluoride directly inhibits the glycolytic enzyme enolase in an irreversible manner, which in turn has an impact on bacterial metabolism and results in a decrease in microbial count<sup>8</sup>.

The present study demonstrates a reduction in the growth of Streptococcus mutans on dentin discs in the CSPS as well as in the fluoride group as compared to the control group ( $p < 0.001$ ) with CSPS showing greater inhibitory effect than fluoride ( $p < 0.001$ ). This is in contrast to a previous study conducted by Coulter J et al which showed no significant effect of CSPS on bacterial biofilms<sup>9</sup>.

Samuel V et al. however reported a maximal reduction in Streptococcus mutans CFU/ml in saliva in the first week after initiating brushing with a remineralizing product containing CSPS which is consistent with the findings of our study<sup>10</sup>.

Concerning fluoride, the result is in accordance with a previous in-vitro study by Randall J et al which has demonstrated clear zones of inhibition against Streptococcus mutans with a fluoride concentration of 1000ppm<sup>11</sup>.

Although our study certainly has room for improvement, we believe that it has demonstrated the anti-bacterial properties of CSPS and of fluoride-containing desensitizing toothpastes. By refining this study, we would be able to investigate the anti-microbial properties of various kinds of desensitizing materials.

### IV. Conclusion

In the current investigation, CSPS-containing desensitizing toothpaste outperformed fluoride-containing desensitizing toothpaste in terms of its antibacterial activity against Streptococcus mutans. The two may have an effect on the microbial biofilm that grows on and around a tooth, as well as the tooth's caries risk. This study had a small sample size and did not replicate the oral environment. As a result, large samples mounted in an oral environment and uniform samples need to be tested in the future.

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