

## Can coffee combat caries? An *in vitro* study

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### Abstract

**Background:** Dental caries is a multifactorial disease caused by oral microflora, diet and oral environment. Oral microflora is crucial in initiation and progression of caries. An important strategy in the prevention of dental caries is to inhibit *Streptococcus mutans*, a caries causing bacteria. Use of natural products in the disease prevention has been increasing recently. One among such products is coffee which has anticariogenic properties against *S. mutans*. The aim of the present study is to evaluate the anticariogenic efficacy of coffee against *S. mutans*.

**Materials and Methods:** The study is carried out using saliva samples collected from 120 healthy individuals of 18–25 years' age group divided into two groups (cariou and noncariou). *S. mutans* is isolated from the saliva samples, cultured on mitis salivarius–bacitracin agar plates and used for checking the inhibitory activity of coffee in different formulations on these bacteria.

**Results:** Our study results showed that coffee, when used alone, has highest antibacterial activity against *S. mutans* and its anticaries activity reduces when it is mixed with milk and sugar ( $P < 0.01$ ).

**Conclusion:** Our study proved that coffee has anticariogenic activity against *S. mutans*.

**Keywords:** Anticariogenic, caries, coffee, *Streptococcus mutans*

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### INTRODUCTION

Dental caries is one of the most common diseases with high prevalence among human population. It is a multifactorial infectious disease in which diet, nutrition, microorganisms and host response plays a major role.<sup>[1]</sup>

In Indian children, the prevalence of dental caries is as high as 60%–80%.<sup>[2]</sup> It has been observed that during 1940, the prevalence of dental caries in India was 55.5%, and during 1960, it was reported to be 68%. A comprehensive national health survey conducted throughout India in 2004 has shown dental caries prevalence as follows: 51.9% in

5-year-old children, 53.8% in 12-year-old children and 63.1% in 15-year-old teenagers.<sup>[3]</sup> There is an overall general impression that the prevalence and severity of dental caries has increased in urban and cosmopolitan population over the last couple of decades.<sup>[4]</sup> Pain from untreated caries in children can affect school attendance, eating and speaking and, subsequently, growth and development. A practical solution to this “silent epidemic” is a preventive approach in a public health scenario. There is a tremendous need for the prevention of dental caries by population-oriented oral disease prevention programs and health promotion. In addition to these, not only preventive restorations and

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fluoride applications but also certain everyday elements of our diet can also help prevent caries.<sup>[2]</sup>

Dental caries is the consequence of the interaction among the oral microflora, the diet, the dentition and the oral environment. Bacteria are crucial to the initiation and progression of carious lesions. *Streptococcus mutans* has been implicated as a primary causative agent of dental caries in humans.<sup>[5]</sup>

Studies on anticariogenic effects of dietary components and natural products have increased in number only in the recent years, especially when there was a realization that foods such as milk and cheese can reduce the effects of acids and can help restore the enamel that may have been lost during eating. The caries inhibitory activity of coffee has recently come into focus when many researchers have started to investigate its bioactive components.<sup>[6]</sup>

A cup of coffee contains 100–150 mg of caffeine. This compound was effective against Gram-positive and Gram-negative reference strains, such as *S. mutans*, the organism causing dental caries. The roasted coffee also has antiadhesive properties.<sup>[7]</sup>

Hence, the present study is aimed to evaluate the *in vitro* antimicrobial activity of coffee-based solutions, obtained by three distinct methods, on *S. mutans*.

## MATERIALS AND METHODS

The study sample included 120 individuals of age group of 18–25 years with no significant past or present medical history. The study sample consisted of two groups. Group 1 included 60 healthy individuals of 18–25 years' age group with caries and Group 2 included 60 healthy individuals of 18–25 years' age group without caries. The caries experience of the individuals was considered based on the WHO criteria. One milliliter of unstimulated saliva was obtained in sterile containers from all the individuals. Written consent for obtaining saliva sample from the individuals was obtained after giving the necessary instructions. Ethical approval was obtained from the institutional review board.

Individuals below 18 and above 25 years were excluded from the study and individuals with any significant past or present medical history were also excluded from the study.

Coffee solution was made in different formulations by boiling for 2 min, either alone or in combination with sugar and milk. One milliliter of saliva of the individuals was

diluted in 100 ml of distilled water and 1 ml of this 100 ml, which contained saliva, and distilled water was added to brain–heart infusion broth and incubated for 48 h at 37°C. One microliter of this turbid broth was plated on a culture plate containing mitis salivarius–bacitracin (MSB) agar along with sucrose, bacitracin and potassium tellurite and again was incubated anaerobically at 37°C for 48 h. Raised, convex, undulate, opaque, granular pale blue colonies were identified and counted using colony counter [Figure 1]. Confirmation for *S. mutans* was done using Gram's stain and catalase test.

Filter paper was cut into circular disks of size 5 mm in diameter and soaked in the following solutions – (1) boiled coffee with sugar and milk, (2) boiled coffee with milk and no sugar and (3) boiled coffee without sugar and milk. The soaked disks were placed on blood agar plate streaked with *S. mutans* colonies obtained from the MSB agar plate. Dilution factor of coffee was 2.2 g of coffee (BRU instant) in 5 ml of milk. The different combinations of milk and sugar with coffee were taken as this is the method by which the coffee is consumed in the world. Zone of inhibition around the filter paper disks was measured in millimeters [Figure 2]. The results were analyzed using unpaired *t*-test and ANOVA single factor.

## RESULTS

When the colony count was done, the mean colony count in carious individuals was 2744 colonies/ml and that of noncarious individuals was 2595 colonies/ml. The difference in the colony count between the two groups was statistically nonsignificant ( $P > 0.05$ ) [Table 1]. The mean values of zones of inhibition (in millimeters) observed on blood agar plate due to each solution in Group 1 were  $1.49 \pm 1.41$  (mean  $\pm$  standard deviation) with Solution 1,



**Figure 1:** Photograph showing *Streptococcus mutans* colonies on mitis salivarius–bacitracin agar plate

2.37 ± 2.32 with Solution 2 and 2.99 ± 2.95 with Solution 3. The mean values of zones of inhibition (in millimeters) observed due to each solution in Group 2 were 1.5 ± 1.46 with Solution 1, 2.16 ± 1.87 with Solution 2 and 3.37 ± 3.09 with Solution 3. When the mean values of inhibition zones were compared between the two groups for each solution, there was no statistically significant difference. The difference was statistically highly significant when compared between the three different solutions individually within each group ( $P < 0.01$ ) [Table 2].

## DISCUSSION

The coffee tree belongs to the Rubiaceae family, genus *Coffea*. More than 80 coffee species were identified worldwide, of which only two are economically important. *Coffea arabica*, also known as Arabica coffee, is responsible for approximately 70% of the global coffee market, and *Coffea canephora* or Robusta coffee (commercial name of one of the main *C. canephora* cultivars) accounts for the rest.<sup>[8]</sup> Coffee is the most commonly consumed beverage everywhere. According to the online survey by Cintin 2018, on beverages consumed regularly in India, 53.51% of respondents of India stated they drink coffee regularly.<sup>[9]</sup> Coffee majorly contains higher amounts of antioxidant compounds and caffeine. The nonvolatile fraction of coffee is composed primarily of water, carbohydrates and fiber, proteins and free amino acids, lipids, minerals, organic acids, chlorogenic acids and trigonelline.<sup>[8]</sup> Few studies are found in the literature that showed the antimicrobial activity of coffee-based solutions. Toda et al. related the effects of coffee on microbial species such as *Staphylococcus aureus*, *Salmonella typhi*, *Shigella dysenteriae*, *Vibrio cholerae*, *Vibrio parahaemolyticus* and *Yersinia enterocolitica* and proposed that bactericidal effect of coffee might be due to tannic acid.<sup>[10]</sup> Studies showed that caffeine and trigonelline exert a strong inhibitory performance against several microorganisms such as *S. mutans*. Few studies showed that, among all substances

studied, trigonelline was the most effective against the growth of the oral pathogens. These compounds exhibit their antimicrobial action by inactivation of cellular enzymes, which depends on the rate of penetration of the substance into the cell or caused by membrane permeability changes. Increased membrane permeability is a major factor in the mechanism of antimicrobial action, where compounds may disrupt membranes and cause a loss of cellular integrity.<sup>[11]</sup> Coffee anticaries potential is related to its capacity of altering the biosynthesis of extracellular polysaccharides (mainly mutans).<sup>[12]</sup> The commercial coffee powder (BRU Instant) used in the present study is a combination of 70% coffee and 30% chicory. Few studies demonstrated that chicory also exert antibacterial activity against *S. mutans*.<sup>[11]</sup> Some studies done to assess the dietary habits (which included coffee) in relation to caries experience stated that there was less caries experience in individuals who consumed coffee in breakfast meal regularly.<sup>[13]</sup> All these findings showed the anticariogenic effect of coffee. This study proved the antibacterial efficacy of coffee on *S. mutans* by the large significant zone of inhibition. There was a difference observed in the colony counts between Group 1 and Group 2 individuals, but it was not statistically significant. Solution 3, i.e., coffee without milk and sugar, showed greater mean values of zone of inhibition followed by Solution 2 (coffee with milk but no sugar) and Solution 1 (coffee with milk and sugar). This might be because of the

**Table 1: Colony count difference between the groups**

Group	Colony count (colonies/ml)	t-statistic	P	Inference
I (cariou)	2744	0.897	0.371	NS
II (noncariou)	2595			

NS: Not significant

**Table 2: The difference in mean values of zones of inhibition between the groups**

Group	Inhibition (mm), mean±SD			ANOVA (F-statistic)	P	Inference
	Solution-I	Solution-II	Solution-III			
Cariou	1.49±1.41	2.37±2.32	2.99±2.95	6.33	0.0022	HS
Noncariou	1.5±1.46	2.16±1.87	3.37±3.09	10.57	0.001	HS
Cariou (vs.) noncariou, P	0.94 (NS)	0.58 (NS)	0.48 (NS)			

SD: Standard deviation, NS: Not significant, HS: Highly significant



**Figure 2:** Photograph showing zones of inhibition on blood agar plate with different formulations of coffee

sugar in Solution 1 which favors the growth of *S. mutans*. Our study results show that black coffee shows highest antibacterial activity against *S. mutans* and its anticaries activity reduces when it is mixed with milk and sugar.

## CONCLUSION

Dental caries is still considered one of the widespread problems of oral health and there is a need for new strategies to prevent the occurrence of this process. Although caries is a multifactorial disease, mutans streptococci are the main etiologic agents of caries. Our study proved that coffee has antibacterial effect against *S. mutans*. Although there are adverse effects with high amount of coffee consumption, it has more useful effects when consumed in limits. Hence, with further research, anticariogenic applications can be developed by incorporating coffee in tooth pastes, mouth washes and chewing gums. As coffee is one of the most consumed beverages in India, its taste and odor are largely appreciated. Coffee can be used at reasonable cost and help in developing new avenues of caries preventive and protective research as oral health is essential to general health and quality of life.

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## Conflicts of interest

There are no conflicts of interest.

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