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### Comparative Analysis Of Dental Plaque pH And Streptococcus mutans Count Changes Following Probiotic Curd Consumption: A Prospective Randomized Control Trial

Guru Vishnu C<sup>1</sup>, Deepa Gurunathan<sup>2\*</sup>, Mahesh Ramakrishnan<sup>3</sup>

<sup>1,2,3</sup>Department of Pediatric and Preventive Dentistry, Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences, Chennai – 600 077

\***Corresponding author:** Deepa Gurunathan, Department of Pediatric and Preventive Dentistry, Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences, Chennai – 600 077

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#### ABSTRACT

**Aim:** To evaluate the impact of probiotic curd on the pH and Streptococcus mutans colony count in dental plaque.

**Study Design**: Twenty healthy children between the ages of 3to 6 years participated in this randomized trial. The participants were divided into two groups at: one received probiotic yoghurt for 15 days; the other received no extra supplements. At baseline and 15 days after the intervention, dental plaque's pH and Streptococcus mutans colony count were measured. Pre- and post-values were collated, and paired and unpaired t-tests were used to compare them.

**Results:** Streptococcus mutans levels and plaque pH both drastically decreased in the study group, while there was no discernible difference in the control group.

**Conclusions:** Consuming probiotics, even for a short time, has a negative impact on the organisms that cause tooth decay. With additional benefits for overall health, it can be used as a preventative strategy to manage dental cavities.

Keywords: Early childhood caries, Probiotic, Plaque pH, Streptococcus mutans

#### **INTRODUCTION**

An individual's dental health has a significant impact on their overall health. A dynamic balance of antagonistic and synergistic microbial interactions allows up to 1000 different bacterial species to live happily(1). Disruption of this equilibrium may alter the microbial ecology and lead to common bacterial diseases in humans, such as tooth decay and periodontal disease(2).

The use of fluoride or casein phosphopeptide, amorphous calcium phosphate, personal care

items, antibiotics, antimicrobial treatment using chlorhexidine or povidone iodine, or enamel surface attenuation using an argon laser are just a few of the solutions that have been used to address these issues(3). With the potential for the evolution of resistant strains, antimicrobial therapy results in the indiscriminate killing of both commensal and potentially harmful bacteria(4). Fluoridation stains teeth and necessitates frequent dental checkups. Additionally, dental caries is still a global "silent epidemic" and a financial burden on the health care system.(5)

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Probiotics were used to raise people's general health. Interest in probiotic therapy for preventing and treating dental diseases has increased as a result of the probiotics' beneficial effects on general health. (6)Probiotics have become useful because they can live in an acidic environment, which is present in dental cavities in particular. The bulk of bacteria from the genera Lactobacillus and bifidobacteria that have been discovered as probiotics thus far are grampositive, seldom associated with human diseases, and are. Despite the fact that the precise mechanism by which probiotics stop the growth of cariogenic bacteria(7) is unknown, it is generally accepted that a mix of non-immune defensive mechanisms and local and systemic immune responses is involved. However, direct impacts on the oral cavity are what provide the most plausible explanations. However, the most plausible theories involve direct impacts on the oral microbiota brought on by antagonistic effects against certain kinds of organisms by synthesis of antimicrobial compounds such bacteriocins, or by creating a competition for nutrition and adhesion, or by boosting immunity.

Despite the fact that the precise mechanism by which probiotics stop the growth of cariogenic bacteria is unknown, it is generally accepted that a mix of non-immune defensive mechanisms and local and systemic immune responses is involved(8). The direct effects on the oral microbiome brought on by antagonistic effects against specific groups of organisms by production of antimicrobial substances like bacteriocins, or by posing a competition for nutrients and adhesion1, or by enhancing immunity through immune modulation, are, however, the most likely explanations.(9)

#### MATERIALS AND METHODS

The samples comprised of 20 healthy children who volunteered after receiving verbal and written information. Informed consent was obtained from their parents. The study design was a short-term intervention trial and the approval was obtained by the Ethical Committee of Saveetha Dental College and Hospitals. The study was double blinded and randomized controlled trial.

#### Inclusion criteria for selection of samples

1. Good general health with no significant medical history.

No anti-inflammatory or antibiotic medications taken in the month before the study.
 No chewing gum or mouthwash used in the last week and during the study period.

4. Habit of brushing twice daily with fluoridated toothpaste.

#### Exclusion criteria

1. Children with decayed teeth.

2. Children under antibiotic treatment or topical fluoride or antiseptic mouthwashfor three weeks before and during the course of the study.

3. Children on any other probiotic supplements during the course of the study.

4. Children on use of any xylitol products for 3 weeks before and during the course of the study.5. Children on use of any antiseptic mouthwash.

Material used in the study were basic examination kit consisting of disposable mouth mask and gloves, mouth mirror, explorer, tweezers and sterile cotton, Amul probiotic yogurt – Flavvyo, Digital ph meter, Mitissalivarius bacitracin (MSB) agar (himedia), Calibrated inoculating loop and Bunsen burner, Incubator and Autoclave.

Using simple random sampling (lottery method) the sample was allocated to either test or control groups. The subjects were encouraged to maintain their normal oral hygiene habits and to continue to brush their teeth.

Group 1: The subjects were provided by the probiotic yogurt, 200 ml of which they were supposed to consume daily at dinnertime. No tooth brushing was allowed for at least 1 hour after yogurt consumption.

Group 2: Consisted of patients who received no probiotic treatment (control group). They were encouraged to maintain oral hygiene and follow the routine dietary pattern.

All subjects will be asked to brush twice daily with their regular fluoridated toothpaste. The patients were instructed to avoid chewing gums, mouthwashes, and antibiotics during the study. Those subjects who fell sick and required antibiotic therapy were dropped from the study and replaced by other children. Samples were collected twice: before the study began and 15 days.

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#### Estimation of plaque pH

Plaque specimens were collected from the labial surfaces of the maxillary teeth with a sterilized scaler. A digital ph meter was used to analyse the pH of collected plaque.

#### Estimation of Streptococcus mutans count

The rest of the plaque was dispersed in sterile containers containing transport media and carried to the microbiologic laboratory for analysis of Streptococcus mutanscount. Plaque Streptococcus mutans were enumerated immediately before (baseline) and after (followup) period.

#### Statistical analysis

The readings obtained were tabulated and compared. The results were derived using SPSS and Excel software for data entry and statistical analysis

#### RESULTS

#### Change in the pH

In the study group as well as the control group, a drop in the pH was recorded. However the drop was significant in the study group, while that in the control group was statistically non-significant (Table 1). When the change in the pH of the two groups was compared, the mean pH of study group was lower in study group as compared to control, but the difference was statistically non-significant. (p-value = 0.09) (Table 2).

## **TABLE 1:** Comparison of the pH between test group and control group at baseline and at 15 days interval

	Mean pH at baseline	Mean pH after 15 days	p value
Probiotic group	$5.1 \pm 0.6$	$5.9 \pm 0.5$	0.04
Control group	$6.2 \pm 0.35$	$5.9 \pm 0.26$	0.3

**TABLE 2:** Comparison of pH between test group and control group at 15 days interval

	Mean	Std dev	p value
Probiotic group	$5.9 \pm 0.5$	0.5	0.9
Control group	$5.9 \pm 0.26$	0.26	

*Change in Streptococcus mutans colony counts* The drastic decline in the colony of Streptococcus mutans in the plaque was recorded after the consumption of probiotic yogurt. On other hand the control group showed no significant difference (Table 3) . On comparing the colony count of the two groups a highly significant decrease was seen in the experimental group as compared to control (Table 4) .

**TABLE 3:** Comparison of the number of colonies between test group and control group at baseline and at 15 days interval

	Mean of baseline	count at	Mean of count after 15 days	p value
Probiotic group	$25.4 \pm 6.33$		$15.8 \pm 5.40$	0.00
Control group	$28.9\pm9.12$		$27.2\pm8.55$	0.3

**TABLE 4:** Comparison of the number of colonies between test group and control group at 15 days interval

	Mean	Std dev	p value
Probiotic group	15.8	5.40	0.03
Control group	27.2	8.55	

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#### DISCUSSION

Dental caries is a microbiological condition that is brought on by a variety of circumstances, thus treating it requires a multifaceted approach. Although it is difficult to sterilise the mouth, probiotic replacement treatment can be used to eradicate harmful organisms while preserving the rest of the oral ecology by balancing the microbial community(6). Studies have shown that probiotics, which were initially identified by Elen Metchnikoff, are good for dental health. Given that dental caries affects children most severely between the ages of 3 and 6, the current study focused on this age range(10). Children who lived in the same location and drank water from the same source were chosen in order to eliminate any potential biases(1). Yoghurt containing the probiotic strains Lactobacillus acidophilus La-5 and Bifidobacterium bifidum was chosen as the probiotic vehicle because it is widely available, has a good buffering capacity, has a low risk of causing caries, and is popular in the Indian market. Despite having acidproducing bacteria, yoghurt is safe to eat due to its ability to buffer and low acid level. Since saliva samples are not specific to dental surfaces, plaque samples were used in the study to evaluate the change in pH and the number of Streptococcus mutans(11), the most aggressive caries-producing bacteria. After 15 days of yoghurt consumption, the study discovered a significant drop in the number of Streptococcus mutans in the study group, showing that the probiotic bacteria can limit the survival of Streptococcus mutans(12). These results were consistent with earlier research that shown a decrease in Streptococcus mutans(13) in the oral cavity as a result of Lactobacillus ingestion.

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