

## **Plaque Ph Changes After Consumption of Sucrose, Milk Containing Sugar and Green Tea Containing Sugar among 8 – 12 Years Old Children**

<sup>1</sup>Dr. Himanshu Gupta, BDS, MDS (Public Health Dentist), Assist. Professor, K. D. Dental College and Hospital, Mathura, Uttar Pradesh

<sup>2</sup>Dr. Navpreet Kaur, BDS, MDS (Public Health Dentist), Head of Department (Department of Public Health Dentistry), K. D. Dental College and Hospital, Mathura, Uttar Pradesh

<sup>3</sup>Dr. Kirti Verma, BDS, MDS (Public Health Dentist), Private Practice Consultation

<sup>4</sup>Dr. Suneel Kumar Gupta, BDS, MDS (Department of Pedodontics and Preventive dentistry), K. D. Dental College and Hospital, Mathura, Uttar Pradesh

<sup>5</sup>Dr. Balkrishn Gaur, BDS, MDS (Department of Oral Medicine and Radiology), Assist. Professor: K. D. Dental College and Hospital, Mathura, Uttar Pradesh

**Corresponding Author:** Dr. Himanshu Gupta, BDS, MDS (Public Health Dentist), Assist. Professor, K. D. Dental College and Hospital, Mathura, Uttar Pradesh

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### **Abstract**

**Aim:** To compare plaque pH changes after consumption of sucrose, milk containing sugar and green tea containing sugar among 8 – 12 years old children.

**Introduction:** Diet is the most important factor to be looked after, as it is not only essential for a growing child or for sustaining good health, but also could be a modifying agent in dental caries.

**Materials and Methods:** A randomized, single exposure study was conducted over a period of 1 week among 45 children (aged 8–12 years) who were selected by simple random sampling. Plaque pH was measured after rinsing

with 10% sucrose (control group;  $n = 15$ ), milk with sugar ( $n = 15$ ), and green tea with sugar ( $n = 15$ ) at an interval of 5, 10, 20, and 30 min.  $p$  value of  $\leq 0.001$  was considered to be statistically significant.

**Results:** Difference in mean plaque pH values between the groups and at different time intervals was statistically analyzed using one way analysis of variance (ANOVA) and repeated measures ANOVA. The final plaque pH results revealed that green tea showed statistically significant difference even after adding sugar when compared to 10% sucrose and milk.

**Conclusion:** The present study proved that local application (oral rinsing) with green tea solution for short time significantly inhibited decrease in plaque pH.

**Keywords:** Children, dental caries, green tea, milk, plaque pH, sucrose.

### Introduction

There are several factors that contribute to an acidogenic environment within the oral cavity. According to the American Dental Association 2008, children consuming sweetened soda and fruit juices were almost twice at risk to develop cavities when compared with milk consumption.<sup>1</sup> Despite the advancements in oral disease science, dental caries continues to be a worldwide health concern, affecting humans of all the ages. It is an infectious bacterial biofilm and pH mediated disease expressed in a predominantly pathologic oral environment. Although acid generating bacteria are the etiologic agents, dental caries has been considered as multifactorial disease since it is influenced by dietary habits as well as host factors.<sup>2</sup> Milk is universally considered as the ideal food for the growing child. It is the most popular form of nutrition from birth to adolescence and beyond, for people both, rich and poor.<sup>3</sup> Dental plaque is a typical biofilm consisting of micro-organisms and their products such as adhesive glucan.<sup>4</sup> Reports suggest that sucrose-containing preparation produces a significantly greater drop in plaque pH than their sucrose-free counterpart.<sup>5</sup> Plaque bacteria produce a large quantity of acids such as lactic acid from fermentable carbohydrates. Acids are entrapped between the tooth surface and plaque biofilm, then the pH of the enamel easily falls below 5.6, and loss of mineral (decalcification) from enamel is induced. This is initiation of dental caries.<sup>6</sup> Disaccharide lactose (4%-5%) is the primary carbohydrate present in the milk. Following fermentation of lactose by biofilm bacteria, the plaque pH lowers to around 6.0 as

against sucrose, following fermentation of which the plaque pH lowers to 5.<sup>7</sup> The anticariogenicity of milk lies within its high buffering capacity and its components i.e, calcium, phosphate, proteins and phosphoproteins. Masih et al. and Danchaivijitr et al. showed that milk can be cariostatic and its consumption with cariogenic food dominates this property.<sup>8</sup> In studies conducted, these polyphenols found in the content of green tea reported bactericidal activity against *S. mutans*, *S. sobrinus* and *Lactobacillus*. However, there are limited number of studies showing that regular green tea consumption reduces tooth decay.<sup>9</sup> Very few studies have reported on in vivo dental plaque pH changes in relation to caries prevalence or activity.<sup>10</sup> Thus, the present study intended at evaluating the anticariogenic effects of green tea and milk with sugar in the oral cavity by measuring the plaque pH changes in children aged 8–12 years and comparing them with sucrose.

### Materials and methods

A randomized, single exposure study was conducted over a period of 1 week among 45 children (aged 8–12 years) who were selected by simple random sampling from Saraswati Vidya Mandir, Mathura. The sample size was determined based on the number of participants present on the day of study. The participants were selected from the same school after obtaining the required Institutional Ethical Committee approval (SVM/Acad/1686).

Oral cavity examination was carried out, and participants were selected as per the following inclusion and exclusion criteria:

#### Inclusion criteria

- Healthy cooperative participants.

#### Exclusion criteria

- History of oral prophylaxis in the past 3 months.
- Participants who ate or drank in the past 2 hours.
- History of antibiotic therapy in the past 3 months.

- History of topical fluoride or mouthwash usage in the past 48 hours.
- Participants with orthodontic appliances.

Carious lesions if present were taken into consideration for appropriate treatment after the study.

### **Methodology**

Eligible participants and their parents were given an informational summary to read, and the study was explained verbally. A signed informed consent form was obtained from the parents. All the demographic data and medical history (especially diabetes and allergy) were taken. Emphasis was given on history of diabetes as sugar is being used in the study to make the green tea palatable for the children, and also, allergic reactions to any of the components of green tea were taken into consideration while selecting the participants.

A prerinsing baseline pH was measured for all the 45 selected children.

They were then randomly divided into three groups of 15 each. Group 1: 10% sucrose already prepared (Department of Biochemistry, K. D. Dental College and Hospital, Mathura), Group 2: milk with sugar, and Group 3: 2% green tea with sugar.

### **Preparation of solutions**

Two percent green tea was prepared by adding 2 g in 100 ml of deionized water and brewed for a constant period of 5 minutes. An equal quantity of sugar, i.e., 1 teaspoon (approximately 5 g) was added to both the test samples, green tea and milk (bovine packed milk, pH - 6.5–6.7 and temperature - 7°C).

Students were then made to rinse their mouth with the respective solution as per the allotted intervention for 2 minutes, and plaque samples were collected after 5, 10, 20, and 30 minutes.

One gram per sample was collected from lingual surface of permanent maxillary right first molar and palatal

surface of permanent mandibular left first molar, permanent mandibular left first premolar, permanent mandibular right first molar, and permanent mandibular right first premolar using four occlusally directed strokes with the help of sterile curettes or probes and was then stored in sterile polypropylene tubes. They were then transferred to laboratory for further biochemical analysis. [Figure 1]

After the collection of plaque samples, i.e., after 30 minutes, students were made to rinse vigorously with distilled water to prevent pooling of the test samples in their mouth. [Figure 2]

### **Plaque pH testing**

The procedure was carried out in Department of Biochemistry, K. D. Dental College and Hospital, Mathura. Calibration of pH meter was done using buffer solutions with pH 4 and 7, prepared using the buffer capsules, every day before the commencement of experiment. pH was separately evaluated for each time interval, and 1 g of plaque was dissolved in 1–2 ml of distilled water. [Figure 3]

### **Data analysis**

Data were analyzed using the SPSS Statistics for Windows, Version 21.0. Difference in mean plaque pH values between the groups and at different time intervals was statistically analysed using one way analysis of variance (ANOVA), repeated measures ANOVA and Least significant difference multiple comparison test (post hoc test). p value of  $\leq 0.001$  was considered to be statistically significant.



Figure 1: Armamentarium



Figure 2: Collection of plaque samples



Figure 3: Digital pH meter

## Results

The present study was conducted for comparison of plaque pH changes after consumption of sucrose, milk containing sugar and green tea containing sugar. A randomized, single exposure study was conducted over a period of 1 week among 45 children (aged 8–12 years)

who were selected by simple random sampling from Saraswati Vidya Mandir, Mathura.

The mean and standard deviation (SD) of plaque pH in experimental groups at different time intervals in between the three groups showed that there was no significant difference between the groups for prerinsing values. At 5, 10, 20, and 30 minutes, one-way ANOVA showed a significant difference in plaque pH between the three experimental groups ( $p < 0.001$ ). When least significant difference (LSD) post hoc test was applied, the lowest pH was observed in 10% sucrose group and the highest in green tea group at all time intervals (except prerinsing). [Table 1]

Results showed a significant difference in plaque pH in 10% sucrose group at different time intervals when repeated measures one way ANOVA was used ( $p < 0.001$ ). When LSD post hoc test was applied for pairwise comparison, the highest pH was observed in 30 minutes at all time intervals. [Table 2]

When the comparison of plaque pH in milk group at different time intervals using repeated measures one way ANOVA was done, a significant difference in plaque pH was found ( $p < 0.001$ ). When LSD post hoc test was applied for pairwise comparison, it showed that 30 minutes plaque pH was significantly higher than other time intervals. [Table 3]

When the comparison of plaque pH in green tea group at different time intervals using repeated measures one way ANOVA was done, results showed a significant difference in plaque pH ( $p < 0.001$ ). When LSD post hoc test was applied for pairwise comparison, the highest pH was significantly in 30 minutes at all-time intervals. [Table 4]

## Discussion

Dental caries is slowly becoming a health problem which can be attributed to use of various therapeutic products, food and beverages which are acidic in nature. The



etiology of dental caries is multifactorial, but sucrose normally plays a major role because of the ability of the micro-organisms in dental plaque to convert this important dietary constituent into various organic acids. For sugar to be metabolized by dental plaque, it must first dissolve in the oral fluids and then diffuse into the plaque.<sup>4</sup>

Modern diet is more refined with increased preference to more natural and healthier products that can be conveniently prepared at home. Dairy products are important for one's overall and dental health and health drinks are the one form through which parents make their children consume milk.<sup>7</sup>

Acid production is an important trait of cariogenic bacteria and its assessment by measuring plaque pH can be a useful indicator of caries risk assessment. Addition of sugar to both the proven anticariogenic agents, i.e., milk and green tea, was essential to make the drinks palatable for children.<sup>11</sup>

Thus, the present study intended to compare plaque pH change after consumption of green tea and milk with sugar with an objective to evaluate the effect of sweetened milk and green tea on plaque pH knowing the fact that it can be easily ingested by children with added benefits of anticariogenicity of the two agents.<sup>1</sup>

The present study included school going children of 8–12 years as developing permanent dentition and changing dietary patterns make them more susceptible to develop caries.<sup>12</sup>

In the present study, the mean and standard deviation (SD) of plaque pH in experimental groups at different time intervals in between the three groups showed that there was no significant difference between the groups for prerinsing values. At 5, 10, 20, and 30 minutes, one-way ANOVA showed a significant difference in plaque pH between the three experimental groups. In previous study conducted by Nidhi Telreja et al.,<sup>1</sup> similar results were

found where there was no significant difference between the groups for prerinsing values. At 5, 10, 20, and 30 min, one-way ANOVA showed a significant difference in plaque pH between the three experimental groups.

The mean value of plaque pH in 10 % Sucrose group at different time intervals between the plaque pH was higher in 30 minutes than at prerinse, 20, 5, and 10 minutes. In previous study conducted by Nidhi Telreja et al.,<sup>1</sup> at 30 minutes, plaque pH was significantly higher than at 20, 10, 5 minutes and prerinse values. Sezin Ozer et al.,<sup>13</sup> found that the plaque pH after rinsing with 10% sucrose solution was significantly lower than baseline for all times tested except 60 min. The minimum pH value was observed at 5 min.

Results showed a significant difference in plaque pH in 10% sucrose group at different time intervals when repeated measures one way ANOVA was used ( $p < 0.001$ ). Also, in previous study conducted by Nidhi Telreja et al.,<sup>1</sup> the significant difference in plaque pH in 10% sucrose group at different time intervals was seen ( $p = 0.030$ ).

The mean values of plaque pH in Milk group at different time intervals between the plaque pH was higher at 30 minutes than at 20, 10, 5 and prerinse values. Also, in previous study conducted by Nidhi Telreja et al.,<sup>1</sup> at 30 minutes, plaque pH was significantly higher than at 20, 10, 5 minutes and prerinse values. Sezin Ozer et al.,<sup>13</sup> found that the plaque pH after rinsing with milk was significantly lower than baseline for all times tested. The minimum pH value was observed at 10 min.

When the comparison of plaque pH in milk group at different time intervals using repeated measures one way ANOVA was done, a significant difference in plaque pH was found ( $p < 0.001$ ). In previous study conducted by Nidhi Telreja et al.,<sup>1</sup> the significant difference in plaque pH in milk group at different time intervals was found ( $p = 0.005$ ).

The mean values of plaque pH in green tea group at different time intervals between the plaque pH was higher in 30 minutes than in 20, 10, 5 and prerinse values. In previous study conducted by Nidhi Telreja et al.,<sup>1</sup> at 30 minutes, plaque pH was significantly higher than other time intervals. The lowest pH was observed at 10 minutes. Demir T. et al.,<sup>9</sup> found that the highest pH value in the 30 minute groups was reached in the green tea group.

When the comparison of plaque pH in green tea group at different time intervals using repeated measures one way ANOVA was done, results showed a significant difference in plaque pH ( $p < 0.001$ ). In previous study conducted by Nidhi Telreja et al.,<sup>1</sup> the significant difference in plaque pH in 2% green tea group at different time intervals was seen ( $p = 0.0001$ ).

Since the study was conducted in a single geographical area, generalization can not be done. It is recommended that further studies can be undertaken with larger sample size and over a longer period of time.

### Conclusion

Oral rinsing with green tea solution for short time significantly inhibited decrease in plaque pH in the study. Oral application of green tea solution in the form of mouthwashes or when added to dentifrices might be a cost-effective caries preventive measure especially for people who can not afford more costlier preventive measures.

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**Legend Figure**

Experimental groups	n	Pre-rinsing (Mean±SD)	5 min (Mean±SD)	10 min (Mean±SD)	20 min (mean±SD)	30 min (mean±SD)
10% Sucrose group	15	6.20±0.08	6.03±0.04	6.01±0.09	6.13±0.10	6.25±0.07
Milk group	15	6.27±0.19	6.44±0.11	6.49±0.06	6.69±0.05	6.78±0.04
Green Tea group	15	6.29±0.04	6.67±0.05	6.71±0.04	6.80±0.03	6.90±0.03
One way ANOVA		2.113*p=0.1 34 (<0.001)	271.044*p=0. 0001 (<0.001)	378.085*p=0. 0001 (<0.001)	355.164*p=0.0 001 (<0.001)	651.678*p=0.000 1 (<0.001)
LSD post hoc test (significant results)		Not applicable	2>1 3>1,2	2>1 3>1,2	2>1 3>1,2	2>1 3>1,2

For LSD Post Hoc, 1=Group 1, 2=Group 2, 3=Group 3

Table 2: Comparison of plaque pH in 10% Sucrose group at different time intervals using Repeated Measures one way ANOVA

F	P	LSD post hoc test (significant results)
23.866	0.0001 (<0.001)	1>2,3,4 5>1

\*For LSD post hoc 1=Pre-rinsing, 2=5 Min, 3=10 Min, 4=20 min, 5=30 min

Table 3: Comparison of plaque pH in Milk group at different time intervals using Repeated Measures one way ANOVA

F	P	LSD post hoc test (significant results)
48.472	0.0001 (<0.001)	2>1 3>1,2 4>1,2,3 5>1,2,3,4

\*For LSD post hoc 1=Pre-rinsing, 2=5 Min, 3=10 Min, 4=20 min, 5=30 min

Table 4: Comparison of plaque pH in green tea group at different time intervals using Repeated measures one way ANOVA

<i>F</i>	<i>P</i>	LSD post hoc test (significant results)
434.175	0.0001 (<0.001)	2>1 3>1,2 4>1,2,3 5>1,2,3,4

\*For LSD post hoc 1=Pre-rinsing, 2=5 Min, 3=10 Min, 4=20 min, 5=30 min