

**Assessment of antimicrobial efficacy of APF gel and the combination of chlorhexidine varnish and xylitol containing chewing gum in children**<sup>1</sup>Dr. Sinjana Jana, Associate Professor, Haldia Institute of Dental Sciences & Research, Haldia.<sup>2</sup>Dr. Amitabha Chakraborty, HOD and Professor, Haldia Institute of Dental Sciences & Research, Haldia.<sup>3</sup>Dr. Bibhas Dey, Professor, Haldia Institute of Dental Sciences & Research, Haldia.**Corresponding Author:** Dr. Sinjana Jana, Associate Professor, Haldia Institute of Dental Sciences & Research, Haldia.**Citation of this Article:** Dr. Sinjana Jana, Dr. Amitabha Chakraborty, Dr. Bibhas Dey, “Assessment of antimicrobial efficacy of APF gel and the combination of chlorhexidine varnish and xylitol containing chewing gum in children”, IJDSIR- June - 2022, Vol. – 5, Issue - 3, P. No. 100 – 106.**Copyright:** © 2022, Dr. Sinjana Jana, et al. This is an open access journal and article distributed under the terms of the creative commons attribution non-commercial License. Which allows others to remix, tweak, and build upon the work non-commercially, as long as appropriate credit is given and the new creations are licensed under the identical terms.**Type of Publication:** Original Research Article**Conflicts of Interest:** Nil**Abstract****Aim:** The aim of the study was to assess the antimicrobial efficacy of APF gel and the combination of Chlorhexidine varnish and Xylitol containing chewing gum on streptococcus mutans levels in 6–10-year-old children.**Materials and methods:** The study was conducted on 30 children studying in a government school in Haldia city aged between 6-10 years who were randomly divided into 3 groups.

Group I (xylitol chewing gum) Group II (xylitol gum and chlorhexidine varnish)

Group III (Fluoride gel)

Group I was given xylitol chewing gum twice daily after breakfast and lunch for 45 days. Group II was given xylitol chewing gum twice daily along with application of chlorhexidine varnish at the beginning of the study and on the 30<sup>th</sup> day. Group III was given Fluoride gel application at the beginning of the study and on the 30<sup>th</sup>day. Microbiological tests were performed to assess the levels of Streptococcus mutans colony forming units (CFU). CFU scores were categorised as follows: 0 = absence of *S. mutans*, 1 = low level (1-10 CFU), 2 = moderate level (11-100 CFU), 3 = high level (101-250 CFU), 4 = very high level (>250 CFU).**Results:** All groups presented a reduction in the Streptococcus mutans levels.

Xylitol and Chlorhexidine varnish together showed the largest reduction throughout the study period, with 58.3% in the first month and 48.2% at the end of the study.

**Conclusion:** The Xylitol chewing gum and Chlorhexidine varnish combination was efficient and superior to single treatments in suppressing Streptococcus mutans.**Keywords:** *S. mutans*, dental caries, APF gel, Varnish, Xylitol

## Introduction

The main pathogen associated with dental caries is *Streptococcus mutans*. It induces mineral loss due to its strong adhesion to the tooth surface and to the acid production resulting from fermentable carbohydrates, which keeps local pH low<sup>1</sup>. The biofilm produced by this pathogen accounts for adhesion of *S. mutans*, and is considered to be cariogenic<sup>2</sup>. Chlorhexidine (CHX)<sup>3,4</sup> and xylitol (XYL)<sup>5</sup> have been used as strategies to prevent and reduce carious lesions<sup>6,7</sup>. Chlorhexidine is a bis-biguanide with antibacterial, anticariogenic and remineralising actions and few toxic effects<sup>4,8,9</sup>. Chlorhexidine acts by damaging the cell membrane of prokaryotes and by disrupting the cytoplasmic constituents. Cell death occurs due to the rapid accumulation of metal ions inside the cells as they become more permeable<sup>10</sup>. Xylitol is also present in human metabolism as a normal metabolic intermediate (in the glucuronate-xylulose cycle). In chemical nomenclature, xylitol is classified similarly to sorbitol and maltitol (i.e., as a sugar alcohol or a polyol). The theoretical calorie value of xylitol is the same as with other dietary carbohydrates (i.e., about 4 kcal/g)<sup>37</sup>. A significant anti-viral effect on streptococci has been seen with Xylitol/Chlorhexidine combination when compared to pure xylitol or chlorhexidine<sup>27</sup>. Fluoride also has antimicrobial action when used in high concentrations, usually higher than those in fluoride-containing products for clinical purposes<sup>19-21</sup>. The present study assessed the antimicrobial effect of APF gel and combining 1% chlorhexidine varnish with xylitol chewing gum on *S. mutans* in 6–10-year-old children.

## Materials and methods

A randomised experimental study was performed at a Government High School, Haldia, West Bengal, India.

Institutional ethical clearance was obtained. All the children of the school were examined out of which only 30 were selected who were within the age group of 6-8 years with early mixed dentition and without any active carious lesions.

The children should not have taken any anti-microbial medication for at least three months before the study and a signed informed consent was obtained from the parents.

Dental examination was performed in the classroom using a mouth mirror and a straight probe.

The children were randomly divided into three groups. In Group I (XYL) (n = 10), xylitol chewing gum was given to the children twice a day, after breakfast and after lunch for a period of 45 days by the in charge care taker. In Group II (CHX + XYL) (n = 10), chlorhexidine varnish at 1% (CERVITEC – Ivoclar) was applied on each child's dentition using an applicator at the beginning of the study and on the 30<sup>th</sup> day, totalling two applications throughout the study.

These children also used xylitol chewing gum during the study period as except on those days of chlorhexidine varnish application.

In Group III (n = 10), acidulated phosphate fluoride gel, was applied at the beginning of the study and on the 30<sup>th</sup> day, totalling two applications.

A sterile tongue blade (180 mm × 18 mm) was inserted into the child's oral cavity and then moved around up to ten times, with both sides being then pressed on a plate containing 12 ml of Mitis salivarius agar base containing 0.2 g/ml sorbitol, 0.01 mg/ml potassium tellurite, 1.66 µg/ml bacitracin and 1.275 µg/ml kanamycin sulphate.

The plates were then incubated at 37°C for 72 hours in an anaerobiosis jar with an atmosphere of 80% N<sub>2</sub>, 10% H<sub>2</sub> and 10% CO<sub>2</sub>.

The period of time lapsed between inoculation and anaerobic incubation did not exceed 4 hours.

Colony forming unit scores (CFU) are counted in the spatula impression area by the same operator using a stereoscopic microscope.

The CFU scores for *S. mutans* are expressed according to the criteria described by Weber as follows:

0 = absence of *S. mutans*

1 = low level (1-10 CFU)

2 = moderate level (11-100 CFU)

3 = high level (101-250 CFU)

4 = very high level (>250 CFU)

Data were analysed by using the statistical program SPSS 11.0. Kruskal-Wallis, Mann-Whitney and ANOVA statistical tests were performed with a 5% significance level.

## Results

Thirty children were selected for this study. The mean age was 7.27 years old, and the 8-year-old children were mostly (75%) seen in groups G2 (CHX + XYL) and G3 (APF).

All groups had statistically significant reductions in *S. mutans* levels ( $p < 0.001$ ). Groups G1 and G2 were found to have the largest reductions, on average 1.97. The mean scores (0 to 4) for *S. mutans* ranged from 3.40 and 3.50 at the baseline (Fig2) to 2.85 and 2.60 after 45 days (Fig 4) for groups G1 (XYL) and G2 (CHX+XYL) respectively. At the baseline, 70% of the children were evaluated as having moderate *S. mutans* levels (11-100 CFU), whereas 17% had high levels and 12% the highest. After 45 days (last saliva collection), however, the levels of *S. mutans* had gradually decreased and were found to be lower than those at the baseline: 70% of the children had low levels, 22% had moderate levels of *S. mutans*.

None of the children had high or very high levels of this micro-organism at the end of the study. *S. mutans* was gradually suppressed in all groups during the study and all the comparisons were statistically significant ( $p < 0.05$ ). The group G2 (CHX + XYL) showed the highest reduction in *S. mutans* level of 58.3% in the first month and 48.2% at the end of the study.

## Discussion

The first micro-organisms described as being involved in the formation of a pellicle adhering to dental surfaces were *S. mitis*, *S. oralis*, and *S. sanguinis*. *S. mutans* is able to demineralise hydroxyapatite, thus demonstrating its relevance in the development of caries<sup>1,8,24</sup>. This key role played by *S. mutans* in the formation of cariogenic biofilm is a hypothesis already accepted in dentistry, since it is the best explanation for caries aetiology<sup>25</sup>. Therefore, reducing the levels of this micro-organism in the oral cavity seems to be crucial for controlling caries in general.

Chlorhexidine (CHX) is known to be an excellent antimicrobial agent, particularly in reducing the levels of *S. mutans*, which is very sensitive to this substance<sup>4,6,8,11,26</sup>. At low concentrations, chlorhexidine can be considered a bacteriostatic agent<sup>3</sup> because it affects the metabolic activity of microorganisms by acting on cell membranes, causing intracellular disruption<sup>27</sup>. However, at high concentrations, this product seems to affect the cellular content, thus being bactericidal<sup>8,28</sup>.

With regard to *S. mutans* levels, the largest reduction was observed in group G2 (CHX + XYL) (91.3%) than G1 (XYL) (87%). These results support other previous studies<sup>3,26,28</sup>, which showed that individuals using chlorhexidine had the best results regarding biofilm reduction compared to those using either fluoride or only

xylitol (G3 and G1), respectively. Chlorhexidine has the ability to remain inside the oral cavity for a long period of time, mainly as varnish, a finding already proved by other authors<sup>3,4,26,29</sup>.

In our study, we observed that in those groups where chlorhexidine was one of the agents (G2), the levels of *S. mutans* continued to decrease even during the one-month interval between the first and second evaluations. This finding confirms its substantivity.

With regard to the amount of chlorhexidine in the varnish, different concentrations can be used, although 1% chlorhexidine yields better results according to the literature<sup>3,26,29-31</sup>. The varnish vehicle was also justified due to the recommendation of not using gels or mouthwashes for young children.

Xylitol also reduces both biofilm and *S. mutans* levels when added to a patients' diet as a sweetener, thus preventing early development of caries. The use of chewing gum as a means of administering xylitol was important for achieving co-operation among the children, in addition to remaining for a longer time in the oral cavity, thus prolonging its effect<sup>12,14,17</sup>. Nevertheless, the frequent use of xylitol chewing gum should be controlled to prevent the occurrence of microbial resistance<sup>15-17</sup>. This finding may be related to factors such as formation of acids resulting from glucose and/or polysaccharides resulting from sucrose<sup>33</sup>.

The optimum length of time for using xylitol should be further evaluated in order to avoid or minimise the emergence of microbial resistance.

Although the results in group G3 (fluoride) were found to be inferior compared to other groups, this agent has antimicrobial action when used in high concentrations, usually higher than that in fluoride-containing products for clinical purposes.

## Conclusion

Based on the results found, we can conclude that combining 1% chlorhexidine with xylitol chewing gum was efficient and superior to single treatments in controlling biofilm and suppressing *S. mutans*. Further investigation should be carried out in order to confirm the results and develop strategies for using such products to prevent dental caries.

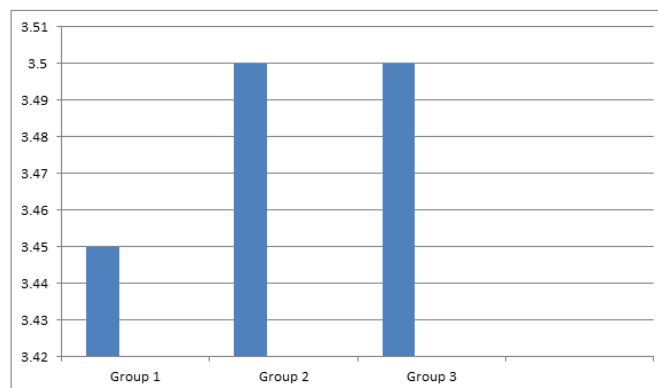


Fig 1: Number of Colony forming units (CFU) of streptococcus mutans in the saliva samples at baseline.

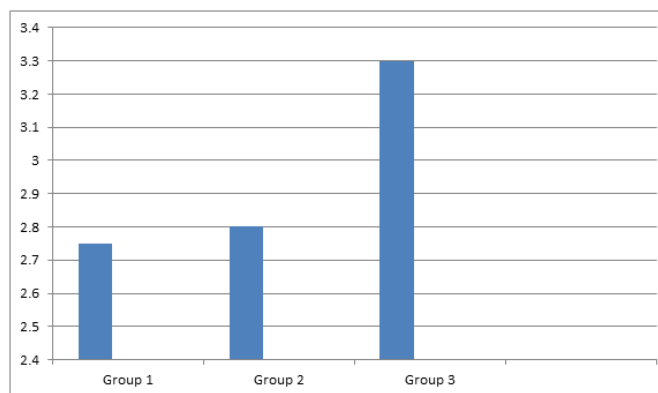


Fig 2: Number of Colony forming units (CFU) of streptococcus mutans in the saliva samples after 30 days.

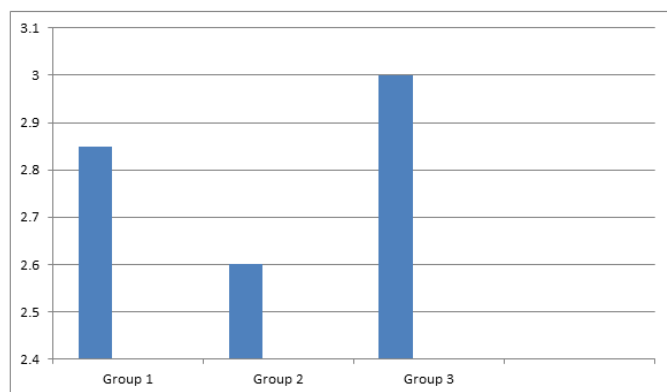


Fig 3: Number of Colony forming units (CFU) of streptococcus mutans in the saliva samples after 45 days

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Fig 4: Procedures followed in the study

