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# Effects of Xylitol in Chewing Gum on Dental Plaque and *Streptococcus mutans*

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## ABSTRACT

Effects of "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint" on plaque accumulation and *Streptococcus mutans,* two indices of potential dental caries and periodontal diseases, were examined in healthy adult volunteers. Forty healthy volunteers aged between 20 and 39 were randomly assigned to either the experimental or control group. Subjects in the experimental group chewed 5 sticks of "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint" daily for two weeks; those in the control group did not chew any gum. In the beginning and the end of study, plaque on all teeth of the subjects was collected and weighed. Results indicated that the group that chewed xylitol sugar free chewing gum for 2 weeks showed significant decrease in both plaque accumulation (21.58%, $p < 0.01$) and *S. mutans* counts (23.14%, $p < 0.05$) while those in the control group showed no significant changes. The results thus indicated that chewing xylitol chewing gum could decrease the likelihood of dental caries.

Key words: plaque accumulation, *Streptococcus mutans*, xylitol, chewing gum, dental caries

## INTRODUCTION

Dental caries and periodontal diseases are two of the most common and serious dental problems that can result in the loss of teeth[1,2]. The history of caries dates back to as early as 22,000 years ago (Cro-Magnon Period) when caries were depicted on wall paintings[2]. In the early days, dental caries occurred mostly in the cemento-enamel junction in the mouth while today they are mostly found in the pits and fissures of the teeth[2,3]. Before the scientific evidences were available, caries were believed to occur as a result of dental bugs "eating out" the teeth. Around the turn of the 20th century, a series of animal experiments and clinical trials indicated that multiple factors contribute to the occurrence of caries[2-5]. One of the contributing factors is carbohydrates in the diet. For example, sucrose and glucose would be fermented by micro-organisms, mainly *S. mutans*, to yield lactic and other organic acids in the mouth. As the result, erosion and decalcification create cavities and cause the teeth to decay and develop caries. Therefore, the amount of dental plaque and densities of *S. mutans* are the two of the most reliable indices which directly reflect the state of dental health.

In 1890, Fischer and Stahel separated a new compound from Beech and named it "xylitol"[6]. In 1969, Scheinin and Makinen first proposed that replacing sucrose with xylitol could reduce caries formation[7]. They found that the dental plaque accumulation on the subjects who consumed snacks and soft drinks containing xylitol was 45~50% less than those who consumed snacks and soft drinks which contained sucrose after 4 days of consumption[8]. Further researches indicated that sugar free gum containing xylitol could decrease the dental plaque accumulation and plaque levels of *S. mutans* for both long-term and short-term uses[9-12]. Decreases in plaque accumulation and *S. mutans* counts appeared to be related to the xylitol content and frequency of usage. During the past 30 years, several studies have indicated that high-content (> 50%) xylitol confections, including candies and chewing gum, are non-cariogenic and possibly inhibit dental caries. Furthermore, the higher manufacturing costs of xylitol as compared with sorbitol necessitated a study to investigate lower content of these polyols; partial substitution of sorbitol with xylitol may keep manufacturer's cost down while still provide dental benefit. Therefore, a simultaneous comparison of the cariologic significance between chewing gums containing as low as 21.82% of xylitol, and without gum was undertaken.

The present study was conducted to test the effectiveness of "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint" manufactured by Wrigley's Taiwan Ltd. on decreasing the dental plaque accumulation and plaque levels of *S. mutans*. The result were used to test against the criteria of "Estimated Health Food Effect for Dental Health" promulgated by Department of Health (DOH), Taiwan, stating that of 50% or more of the subjects had *S. mutans*

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counts lower than the baseline level after using the test food, then the test food can be regarded as having an effect of decreasing the *S. mutans* counts in the plaque.

## MATERIALS AND METHODS

### I. *Content Analyses of "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint"*

The composition of "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint" was analyzed by Hwayo Tech & Lab Co., Ltd. According to the analysis, the chewing gum weighed about 2.7 g and contained 15.32% xylitol, 37.90% sorbitol, 11.97% mannitol, 4.78% maltitol syrup and 0.25% aspartame. Each gum contained about 0.41 g of xylitol. On the other hand, the total sweetener in the gum was 70.22%, and the content of total sweetener in xylitol was 21.82%. Therefore, the intake of xylitol by the volunteer in this experiment was 2.07 g per day.

### II. *Screening of Participants*

Volunteers consisted of students and staff in the Dental School of the National Defense Medical Center, in addition to a few test subjects recruited via the internet. Eighteen males and 22 females ranging from 20 to 39 years old with an average age of 25.8 years participated in this study. Volunteers were healthy adults, non-smokers, non-alcoholics, free of periodontal diseases and without habit of using mouthwashes. The initial screening of saliva from the 55 volunteers indicated that 40 of them had > $10^5$ CFU/mL *S. mutans* in the saliva. The mean DMFS index of the volunteers was 9.95 (range between 0 and 27) with 7.43 (range between 0 and 27) mean FS index and 1.28 (range from 0 to 19) mean DS index. Participants were randomly assigned to either the experimental or control group containing 20 volunteers each.

### III. *Determination of S. mutans Counts in Human Saliva*

Volunteers were asked to chew parafilm® (American National Can Co., Greenwich, CT, USA) for 3 min. Two to four milliliter of saliva was then collected. One hundred microliter aliquots of the saliva were then diluted with 4.90 mL of 0.05 M phosphate buffer (pH 7.3) and further diluted 10 fold for several times. Six aliquots of 50 μL of diluted saliva were plated on Bacitracin-mitis-salivarius (MSB) agar (Difco, Detroit) and incubated for 3 days in an anaerobic chamber (37°C, 5% $CO_2$ plus 95% $N_2$) following bacterial colony counts[13].

### IV. *Experimental Design*

The study followed the guidelines promulgated in the "Estimate the Health Food Effect for Dental Health" published by DOH, Taiwan. Changes in dental plaque accumulation and densities of *S. mutans* were used for the assessment of their effects on dental health. The experimental period was approximately 4 weeks. Subjects maintained normal dietary and oral hygiene habits throughout the study but were restricted to using "Darlie" toothpaste (a popular toothpaste brand in Taiwan). Use of mouthwash was forbidden. During the experimental period, volunteers were required to refrain from eating any foods containing xylitol and any gum not provided as part of the study. For 48 hr before the second and third examinations, subjects were asked to refrain from cleaning their teeth by brushing or using dental floss. Volunteers could return to their normal dietary and oral hygiene habits after each examination. The experimental protocol is illustrated in Figure 1.

Volunteers were informed of the purpose, protocols, methods and any potential risks following the first examination. Written consent was then obtained from each volunteer agreeing to take part in the trial. DMFS index of each volunteer's oral cavity was then recorded. Teeth were then cleaned by ultrasonic full mouth scaling.

At the secod examination following the washing-out period, the supra-gingival plaque was scraped from the subjects' teeth and collected on 2 cm × 2 cm aluminum foil and weighed using a digital electronic balance. The plaques were then transferred to sample vials containing 10 mL of 0.05 M phosphate buffer (pH 7.3) and sonicated for 5 min. Aliquots of 0.5 mL of the plaque solution were diluted with 4.50 mL of 0.05 M of phosphate buffer. Following serial 10× dilutions, aliquots of the solution were plated on MSB agar for *S. mutans* counts and incubated for 3 days in an anaerobic chamber (37°C, 5% $CO_2$ plus 95% $N_2$). The di-

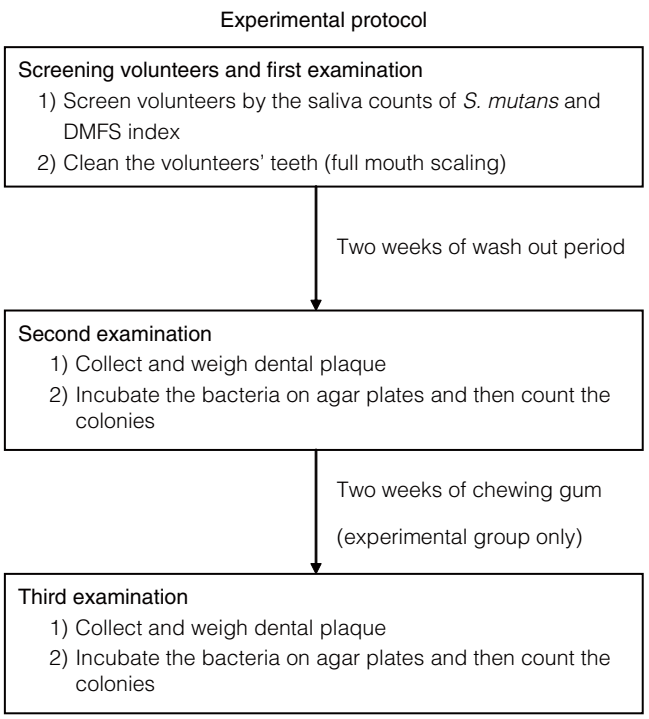


**Figure 1.** The experimental procedure and investigation schedule.

luted solution was also placed separately and simultaneously on Brain Heart Infusion (BHI) agar (Difco, Detroit) for microorganism counts and further incubated in an anaerobic chamber (37°C, 5% $CO_2$ plus 95% $N_2$) for 5 days. The *S. mutans* counts on MSB agar were divided by their total number of bacteria counts on BHI agar, which resulted in the percentage of *S. mutans* in plaque.

At the second examination, we distributed "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint" to participants in the experimental group. Volunteers in the experimental group chewed gums five times/day, one stick at a time and each for 10 min. The chewing times were after breakfast, at 10:00 a.m., after lunch, after dinner, and at 9:00 p.m. Volunteers in the control group could not chew any gum.

The experimental process of the third examination was the same as the second examination. Both second and the third examination were executed between 10:00 and 12:00 a.m. Before 6 hr of each examination, volunteers couldn't eat but could only drink water. Volunteers couldn't eat or drink water 2 hr before each examination. All plaque collections from the volunteers were executed by the same dentist.

V. *Statistical Analyses*

We chose SigmaStat (version 2.0) statistics software for statistical analysis and compared the result of the third examination with the baseline level derived from the second examination. If the plaque accumulation change of the experimental group was 20% lower than the control group ($p < 0.05$), gum is believed to reduce the dental plaque accumulation on the teeth. If more than 50% of the volunteers showed a decrease in *S. mutans*, gum is believed to reduce counts of *S. mutans* in the plaque.

## RESULTS

Table 1 shows the dental plaque accumulation before (baseline) and after (2 weeks after) chewing "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint". The average accumulation of dental plaque before chewing was 74.40 mg while that after chewing was 56.30 mg. Based on the Student's paired *t*-test analysis ($p < 0.01$), the after chewing showed a significant decrease of 18.10 mg. The average reduction in dental plaque (after/before – 100%) was 21.58%. Trend-wise, plaque accumulation decreased in 15 volunteers (75%) but increased in the remaining 5 volunteers (25%).

Table 2 shows the effects of two week chewing of "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint" on plaque *S. mutans* counts. The average before chewing count of *S. mutans* was 0.836% of the total bacteria count in the plaque while the average after chewing count was 0.751%, yielding a mean before and after chewing difference of -0.085%, which was significantly different, based on the Wilcoxon Signed Rank Test ($p < 0.05$). Similarly, the mean (after/before – 100%) difference was -23.14% with 15 subjects (75%) showing decrease and 5 subjects (25%) showing increase. The subjects increased in *S. mutans* counts were totally different from the subjects increased in dental plaque accumulations.

In the control group, results from the second and third examinations yielded mean dental plaque accumulations of 53.60 mg and 55.72 mg respectively (Table 1), which were not significantly different from each other ($p > 0.4$). Similarly, the percentage of *S. mutans* in the second examination (before) was 0.567% and that of the third examination (after) was 0.546% (Table 2), which again was not significantly different from each other ($p > 0.6$).

Statistical analyses of before and after two weeks of chewing as well as no chewing on dental plaque accumulation and *S. mutans* counts are presented in Table 3. Statistical analysis indicated significant changes at $p < 0.01$ and $p < 0.05$ for plaque accumulation and *Streptococcus mutans* counts, respectively.

## DISSCUSSION

Saliva is a very important factor for prevention of caries[2]. The most attractive saliva stimulation method is chewing sugarless gum. Anti-caries agents such as fluoride, green tea extract and xylitol have been added to chewing

**Table 1.** Effects of chewing "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint" on dental plaques

| Groups / Statistics | Amount of dental plaque (mg) | | | | | |
|---|---|---|---|---|---|---|
| | Experimental group (n = 20) | | | Control group (n = 20) | | |
| | Before | After | Change (%)[a] | Before | After | Change (%) |
| Mean | 74.40 | 56.30 | -21.58 | 53.60 | 55.72 | 7.28 |
| S.D.[b] | 51.16 | 47.79 | 29.28 | 17.01 | 17.86 | 29.55 |
| S.E.[c] | 11.44 | 10.73 | 6.55 | 3.80 | 3.99 | 6.61 |
| Range | 22.1 to 196.8 | 14.6 to 213.8 | -59.22 to +38.40 | 26.7 to 90.7 | 30.6 to 91.7 | -25.23 to +70.45 |

[a](After / Before) – 100%.
[b]Standard deviation.
[c]Standard error.
Student's paired *t*-test: experimental group p = 0.0040; control group p = 0.4958.

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**Table 2.** Effects of chewing "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint" on the levels of *S. mutans*

| Groups / Statistics | Percentage of *S. mutans* (%) | | | | | |
|---|---|---|---|---|---|---|
| | Experimental group (n = 20) | | | Control group (n = 20) | | |
| | Before | After | Change (%)[a] | Before | After | Change (%) |
| Mean | 0.836 | 0.751 | -23.136% | 0.567 | 0.546 | 1.449% |
| S.D.[b] | 1.270 | 1.521 | 35.320% | 0.371 | 0.320 | 24.771% |
| S.E.[c] | 0.284 | 0.340 | 7.898% | 0.083 | 0.072 | 5.539% |
| Range | 0.14 to 4.55 | 0.06 to 6.87 | -80.952% to 50.989% | 0.142 to 1.765 | 0.144 to 1.350 | -59.597% to 45.318% |

[a](After / Before) – 100%.
[b]Standard deviation.
[c]Standard error.
Wilcoxon Signed Rank Test: experimental group p = 0.0266; control group p = 0.674.

**Table 3.** Comparison of dental plaque accumulation and *S. mutans* counts between experimental and control group

| Groups | After/before change (%) | | | |
|---|---|---|---|---|
| | Dental plaque accumulation[a] | | *S. mutans* count[b] | |
| | Experimental group (n = 20) | Control group (n = 20) | Experimental group (n = 20) | Control group (n = 20) |
| Mean | 78.42 | 107.28 | 76.864 | 101.449 |
| S.E.[c] | 6.55 | 6.61 | 7.898 | 5.539 |

[a]Mann-Whitney Rank Sum Test, p = 0.0093.
[b]Student's paired *t*-test, p = 0.015.
[c]Standard error.

gums to prevent caries[1,14,15]. Xylitol containing gums are recently introduced and commercially available in the Taiwan market.

Results from several studies indicated that chewing sugar free gums containing xylitol for a short-term period decreased the amount of dental plaque accumulation and *S. mutans* counts in saliva and the plaque[16-19]. In addition, chewing sugarfree gum containing xylitol for a prolonged period could decrease the incidence of dental caries[20-25]. On the other hand, short-term (4 to 28 days) effect of chewing sugarfree gum appeared to be related to the frequency of chewing and the xylitol content of the chewing gum. Chewing 10 sticks of gum containing 100% or 75% xylitol daily for 4 days decreased dental plaque accumulation[16,17] while chewing 2 sticks of gum containing 75% xylitol daily for 28 days resulted in the decrease of levels of *S. mutans*[18]. Chewing 10 sticks of gum containing 75% xylitol every day for 14 days resulted in the decrease of both the dental plaque accumulation and the *S. mutans* count[9].

In the long-term, chewing gums containing xylitol can decrease the possibility of caries development. It has been reported that daily chewing of gum that contains 6.7 g of xylitol resulted in the decrease of caries by 82% within a year[21] while in the case of chewing gum containing 1 to 3.9 g xylitol for 1 to 2 years, caries development would decrease by 52%[22].

The present study shows that chewing "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint" at 5 sticks/day for 2 weeks resulted in significant reduction of dental plaque accumulation (21.58%). Trend-wise, plaque accumulation decreased in 15 volunteers (75%) but increased in the remaining 5 volunteers (25%). The mean counts of *S. mutans* in plaque also significantly decreased by 23.14% with again 15 subjects (75%) showing decrease and 5 subjects (25%) showing increase. This result is consistent with the claim that "Wrigley's Extra® Sugarfree Chewing Gum • Peppermint" containing xylitol in place of sucrose had a beneficial effect on dental health. Furthermore, this result satisfies the DOH criteria.

Test subjects of this study chewed gum containing 2.07 g of xylitol. Based on the previous studies, it can be inferred that 1 to 2 years of chewing the gum might lead to reduction in caries development at certain levels. More studies on the long-term effect will be required to support such projections.

## CONCLUSIONS

Dental plaque accumulation and intra-plaque densities of *S. mutans* are two major indices of dental diseases. The present study found that both plaque accumulation and plaque densities of *S. mutans* were significantly decreased by chewing "Wrigley's Extra® Chewing Gum • Peppermint" daily (5 sticks/day) for two weeks, which contained xylitol in place of sucrose, appeared to reduce the likelihood of caries development.

## ACKNOWLEDGEMENTS

This study was accomplished on behalf of Wrigley Taiwan Ltd. which applied for Health Food Registration

license for its Extra Sugarfree Gum. This product was approved by the Department of Health, Taiwan on Feb. 26, 2005 (Licensure No. A00058).

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