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Impact of Turpentine-Containing Chewing Gum on Saliva Characteristics and *Streptococcus mutans* in Young Adults

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ABSTRACT

Background and Aim: Tooth decay is one of the most common oral diseases. The pH and amount of saliva affect the growth rate of caries-causing bacteria. Chewing gum can help reduce caries by affecting saliva characteristics. The aim of this study was to compare the effect of several types of chewing gums available in the Iranian market on pH, volume, buffering capacity of saliva and the number of salivary *Streptococcus (S.) mutans*.

Materials and Methods: Forty dentistry students were divided into four groups. Each group was given a brand of gum (Van-xylitol/Van-glucose/Chic/Triadent). The participants were asked to chew gum for 20 minutes after each meal. Non-stimulated saliva was collected by spitting method at the beginning of the study and three weeks after gum consumption. The pH, volume, and salivary buffering capacity were determined. In addition, the amount of *S. mutans* in the saliva was evaluated.

Results: The amount of salivary *S. mutans* in all groups of consumers dropped (P<0.05) and saliva volume increased after three weeks of gum chewing. Saliva pH, volume, buffering capacity, and the number of salivary Streptococci all changed significantly after chewing Van-xylitol gum.

Conclusion: Van-xylitol and Triedent gums were most effective in improving salivary volume, and reducing the number of *S. mutans* in saliva, but Van-xylitol was the most effective among the groups in improving salivary pH and increase in the buffering capacity of saliva. It seems that turpentine can significantly enhance the properties of xylitol-containing gums.

Keywords: Chewing Gum, pH, Saliva, *Streptococcus mutans*, Turpentine

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1. Introduction

Dental caries is a chronic and common infectious disease characterized by demineralization of the inorganic component and loss of organic materials in the tooth **(1)**. In developing countries, it is still the most common chronic disease in children and adults **(2)**. Dental caries

is multifactorial tooth decay that the volume of saliva, oral microbial flora, dry mouth, time, consumption of carbohydrates, socio-economic status, educational level of people, and lifestyle are all factors that contribute **(3)**. *Streptococcus (S.) mutans* is the primary cause of tooth decay in humans **(4)**. The typical oral flora consists of microorganisms that are naturally present in the mouth and are completely safe. These colonies adhere to the tooth outer surface and produce plaque, which causes tooth decay **(5)**.

Saliva has various defense mechanisms such as rinsing and cleansing microorganisms, reducing the acidity of the mouth, lubricating the tooth surface, and protecting the teeth through the buffering system **(6)**. Due to mechanical and digestive stimulation, saliva flow rate increases during chewing gum, especially in the first minutes. After a meal, chewing sugar-free gum decreases plaque acidity and, as saliva output increases, it neutralizes the process of saliva pH dropping that happens after each meal. Bicarbonate-containing sugar-free chewing gums increase saliva pH **(7, 8)**. As it is impossible to eliminate sugar and other fermentable carbohydrates from people's diets, and because caries is so common in Iran, finding ways to increase the pH, volume, and buffering capacity of saliva, as well as reduce the number of salivary *S. mutans*, is critical. Studies have shown that natural products can be effective in reducing the number of *S. mutans* bacteria and improving oral health **(9-11)**.

Pistacia terebinthus, scientifically known as *Pistacia atlantica*, with the local name of Baneh, produces a type of resin (turpentine), which is called van. This substance is naturally antifungal and antibacterial, and is used in production of medicinal and health products. It is a great importance in traditional Iranian medicine and is prescribed in the treatment of some digestive diseases **(12-14)**.

Research suggests that chewing turpentine gum after surgery, until the first bowel movement occurs, may help accelerate the recovery of bowel function following a cesarean section **(15)**. Given that turpentine or *Pistacia atlantica* resin is used to strengthen and tighten the gums due to its firm consistency, it is used in chewing gum industry **(16)**. It has been shown that chewing gum is a good alternative to brushing teeth, and that chewing gum is especially helpful for use in situations where brushing is not possible or convenient, such as after lunch, while traveling, or while working. During chewing gum, saliva flow increases, especially during the first few minutes, due to mechanical and digestive stimulation **(17)**. Therefore, in recent years, efforts have been made to optimize chewing gums and their use in promoting oral health.

Amorphous calcium phosphate casein phosphopeptide (CPP-ACP) is an agent that saturates saliva and biofilms and contributes to the tooth remineralization process by providing Ca2+ and -3PO4 ions, which become available after its application and reduce the risk of dental hard tissue destruction **(18)**. Another ingredient used in chewing gum today is xylitol. Xylitol is a five-carbon sugar alcohol obtained from the Birch tree and naturally there is in some fruits, vegetables (cauliflower), strawberries, and mushrooms, which *S. mutans* and other microorganisms cannot ferment. Xylitol has bacteriostatic properties and inhibits the production of acid from sucrose and glucose in dental plaque. When xylitol is consumed repeatedly and for a long time, it causes changes in the dental flora and reduces acid production by bacteria **(19, 20)**.

In the herbal medicine supply industry, a type of gum containing turpentine has been widely available and commonly purchased by customers for a long time. According to the contents on the packaging, this gum contains glucose, which is a type of cariogenic sugar. On the other hand, according to oral and untested reports from consumers, the consistency of this gum is tighter than other gums on the market, which stimulates more saliva secretion in mouth. The two seemingly contradictory features of this gum include cariogenic sugar (disadvantage) and stimulation of more saliva secretion (advantage), which may cause a decrease in pH and subsequently an increase in caries, and the aforementioned condition can lead to an increase in pH and even a decrease in dental and salivary bacterial plaque and a decrease in caries.

As a result, we decided to investigate the effects of four types of chewing gum available in the Iranian market that contain various ingredients including xylitol, CPP-ACP, turpentine, etc. on pH, volume, buffering capacity, and number of *S. mutans* in saliva.

2. Materials and Methods

This research was approved by the ethics code IR.MUQ.REC.1399.286 in Qom University of Medical Sciences and was conducted on 40 female dental students aged 18 to 26 who were randomly divided into 4 groups of 10 people (individuals were selected using a block randomization method). The sample size was calculated based on Khoramian et al **(21)** study, with mean bacterial reduction in the case and control groups ($\mu_1 = 44072.3 \pm 8793$, $\mu_2 = 32696.6 \pm 6158$) and significance level (α) of 0.05, and a power ($1-\beta$) of 90%. The first group used Van-xylitol gum, second group used Van-glucose gum, third group used Chic gum, and fourth group used Trident gum for three

weeks (Table 1). These students had at least 20 functional teeth in the mouth and healthy gums, no significant systemic disease or quickly advancing caries. Also, they had not used xylitol gum, probiotic items, systemic antibiotics, or topical fluoride for 4 weeks prior to test initial. (Exclusion criteria included: students with decayed teeth, chronic dry mouth, systemic, infectious or inflammatory disease, undergoing radiotherapy, take certain medications such as antihypertensive or contraceptives, abnormal diet and eating habits, active periodontitis, consumption of any antiseptic (except toothpaste) in the last month, oral breathing problems, and pregnant and smokers' women **(22)**.

Table 1. Types of chewing gum studied.

Gum brand name	Main ingredients	Manufacturer
Van-xylitol	Pistacia atlantica resin (turpentine) and xylitol	Van company, Kurdistan, Iran
Van-glucose	Pistacia atlantica resin (turpentine), sucrose, glucose, and α-penine	Van company, Kurdistan, Iran
Chic	Glucose and base, glucose sweetener, carnauba wax, antioxidant, and titanium dioxide (as a colorant)	Minoo Company, Karaj, Iran
Trident	Base, thickeners and stabilizers, acesulfame, antioxidants, glycerol, essential oil, color, carnauba wax powder (as a polisher), CPP-ACP and xylitol	Mondelez global LLC. East Hanover, USA

Groups were as follow:

Group 1: Consumer sugar-free turpentine gum with the commercial name Van-xylitol (group Vx)

Group 2: Consumer turpentine gum containing sucrose and glucose with the commercial name Van-glucose (group Vg)

Group 3: Consumer chewing gum with the commercial name Chic (group C)

Group 4: Consumer chewing gum containing CPP-ACP and xylitol with the commercial name Trident (group T)

At first Decay, Missing, Filling Teeth (DMFT) participants were determined and those with DMFT, between 8-12, were selected. The students were asked to chew gum for 20 minutes three times daily after each meal, use a specified type of toothpaste (Nasim) and having regular and the same number of brushing times during the study for three weeks **(21)**. In addition, the participants were instructed not to use any gum other than the mentioned gum during the study period.

Non-stimulated saliva was collected in two times; before consuming chewing gum, and 3 weeks after for specific culture and assessment of the number of salivary *S. mutans* colonies, and evaluation of salivary buffering capacity, the volume and pH of saliva. Samples were labeled 1 to 4 and sent to the laboratory for analysis and statistical evaluation.

Each sample was prepared at 9:00 a.m before washing the mouth, brushing the teeth, and eating breakfast. Saliva was collected by spitting method and putting non-stimulated saliva into a specific sterilized container with a cover **(23)**. Saliva pH and volume were determined immediately after collection, and the samples were sent to microbiology lab on dry ice to assess the buffering capacity and the number of *S. mutans*.

2.1 Determination of Saliva volume

The volume of saliva was determined by the amount of saliva collected in the Falcon calibrated tube for 4 minutes **(24)**.

2.2 Determination of the Saliva pH

The pH of saliva samples was determined immediately after sampling using a calibrated pH meter. First, the electrode of the instrument was cleaned with distilled water, and then pH of saliva was measured. To obtain a perfectly accurate value, this process was performed twice **(25)**.

2.3 Determination of Saliva Buffering Capacity

To determine the buffering capacity of saliva 0.05 N hydrochloric acid (1 ml) was added to the saliva sample and the pH was read and recorded using a pH meter. This was repeated until the saliva pH decreased dramatically (a few more times to be sure), until the point the pH variations were reduced. Then, the diagram of each sample was plotted and the buffering capacity was calculated **(6)**. According to this approach, a solution with a higher buffering capacity has more acid resistance, whereas a solution with a lower buffering capacity has less acid resistance. For example, if the pH of saliva declines dramatically in the

range of 6.79 and reaches 6.52 the sample buffering capacity is 6.79.

2.4 Determining *S. mutans* number

As the quantity of bacteria in 1 mL saliva can exceed one million, and it is preferable to count 100-150 bacteria for each colony, consequently, 0.5 ml of saliva was 10-fold diluted in sterile phosphate buffered saline (PBS) (0.05M; pH 7.3) and mixed on shaker. Then, using the Streak technique, 20 microliters of this mixture were cultivated on the surface of a particular culture medium MSA with bacitracin and 10% sucrose. The MSA plate was then incubated for 48 hours at 37°C. The colonies that were suspected of being *S. mutans* were then assessed, and *S. mutans* was identified using Gram staining, catalase, mannitol fermentation, and Voges-Proskauer (VP) tests. In the final stage, confirmed colonies of *S. mutans* were counted and scored as following Kamate et al **(26)**, criteria (CFU/mL):

0 colonies: no growth/mL saliva

1 colony: 1-10^3 bacteria/mL saliva

2 colonies: 10^3-10^5 bacteria/mL saliva

3 colonies: more than 10^5 bacteria/mL saliva **(26)**.

Catalase was tested after Gram staining by transferring bacteria from the center of a colony to the surface of a glass slide using a wooden applicator. The production of bubbles on the slide was examined when a drop of H_2O_2 was added to the colony. The test was considered positive when boiling occurred quickly and persistently (foam).

2.5 Fermentation of Mannitol

Before examining, the target colonies were inoculated onto mannitol salt agar medium and incubated at 35°C in CO_2-free condition for 24 to 48 hours. The plates were kept for 48 hours because of slow fermentation process of some staphylococcal species. Staphylococcus colonies were appeared in yellow with a yellow halo around them **(27)**.

2.6 Vogues-Proskauer (VP) Test

A screw tube holding 25 ml of VP / MR Broth was infected with a pure bacterial culture and remained for 24-48 hours at 35°C. , Two drops of 40% KOH and one drop of 5% α-naphthol were added. The tube was gently shaken for 5 to 15 minutes. The presence of dystyl and a positive VP test was indicated by formation of red color for 2 minutes or longer. A negative VP test was indicated by yellow color or no response. This test should not be read more than one hour for the possibility of false positive **(28)**.

Finally, Gram-positive and catalase-negative cocci were cultured for 48 hours at 37°C under biochemical assays, and colonies positive for mannitol and Vogues-Proskauer (VP) were chosen as target colonies (*S. mutans*) **(21)**.

2.8 Data Analysis

Quantitative variables were summarized using means and standard deviations. The normality of the data was evaluated using the Kolmogorov-Smirnov test. Since all variables exhibited a non-normal distribution, comparisons among four groups were conducted using the Kruskal-Wallis and Mann-Whitney tests, while intra-group comparisons were made using the Wilcoxon signed-rank test. The results were analyzed using SPSS version 24.0 statistical analysis software. Data were considered significant at $P<0.05$.

3. Results

The pH, volume, and quantity of *S. mutans* in saliva of all four groups were measured before and after chewing gum (Table 2).

3.1 Saliva pH

Before chewing gum, pH means of non-stimulated saliva in groups Vx, Vg, C, and T were 6.86, 6.49, 7, and 6.75, respectively, and after chewing gum were 7.05, 6.72, 6.71, and 6.93, respectively (Table 3). In the comparison between groups (Figure 1), consumption of chewing gum in groups Vx and T significantly increased pH of saliva compared to group C (*P*=0.029 and *P*=0.043, respectively). These changes were not significant among other groups.

3.2 The Volume of Non-Stimulated Saliva

Before chewing gum intake, the average amounts of non-stimulated saliva in groups Vx, Vg, C, and T were 5.7, 4.5, 4.35, 3.35 mL, respectively and after chewing gum consumption the results were 6.59, 5.5, 4.55, and 4.5 mL, respectively (Table 4).

An increase in saliva volume was reported in all groups after consuming chewing gum, but this change in saliva volume was only significant in groups Vx and T. The highest and the lowest rates of increase in saliva volume after chewing gum belonged to Trident Chic gums, respectively. As shown in Figure 2, gum consumption in groups Vx and T significantly increased saliva volume compared to group C (P=0.029 and P=0.043, respectively). These changes were not significant among other groups.

3.3 Salivary Buffering Capacity

Before chewing gum, the averages of salivary buffering capacity in groups Vx, Vg, C, and T were 6.65, 7.08, 7.06, and 7.08, respectively, whereas after chewing gum consumption were 6.77, 5.66, 5.85, and 4.87, respectively (Table 5). The averages of salivary buffering capacities among 40 participants, before and after chewing gum were 6.96± 0.28 and 5.78 ± 0.76, respectively. The buffering capacity of saliva decreased significantly (P=0.005) in all groups after chewing gum, except for group Vx, which reported an increase in buffering capacity of saliva, however, it was not statistically significant.

In the comparison between groups as shown in Figure 3, these changes in group Vx were significant compared to all groups (P=0.000). The changes in group T were also significant compared to groups Vg (P=0.002) and C (P=0.001). It means that group T decreased buffering capacity of saliva compared to groups Vg and C. This difference was not significant in groups Vg and C.

3.4 *S. mutans* Numbers

Before and after chewing gum, the average numbers of *S. mutans* in four groups of Vx, Vg, C and T were measured 65.06, 40.80, 53.70, 53.50 CFU/mL, and 9.3, 30.9, 48.6 and 18 CFU/mL, respectively (Table 6). According to Table 6, consumption of all 4 types of chewing gums reduced significantly the number of *S. mutans* bacteria in the saliva of consumers. Comparing these changes between groups (Figure 4), groups Vx and T showed significant decrements in the number of bacteria compared to groups Vg and C (1&2: *P*=0.03/ 1&3: *P*=0.029/ 4&2: *P*=0.03/ 4&3: *P*=0.02) but this difference was not significant in other groups. In other words, gum types Vx and T performed the same in terms of effectiveness on reducing the number of bacteria and more effective than the other two groups.

Table 2. Variables examined in 40 participants before and after chewing gum.

Saliva variable	Mean ± standard deviation	Saliva variable
pH	6.77 ± 0.46	6.85 ± 0.58
volume (mL)	4.47 ± 1.58	5.28 ± 1.73
buffering	6.96 ± 0.29	5.78 ± 0.77
S. mutans (CFU/mL)	53.04 ± 27.22	27.15 ± 21.21

Table 3. Comparison of saliva pH values among four groups.

Group	Number	pH Conditions	Mean±SD	*P. Value*
Vx	10	Basic	6.86 ± 0.30	0.010
		After chewing gum	7.05 ± 0.38	
Vg	10	Basic	6.49 ± 0.58	0.332
		After chewing gum	6.72 ± 0.66	
C	10	Basic	7 ± 0.41	0.096
		After chewing gum	6.71 ± 0.61	
T	10	Basic	6.75 ± 0.44	0.149
		After chewing gum	6.93 ± 0.62	

group 1: Vx, group 2: Vg, group 3: C, group 4: T According to this table, the changes in pH before and after chewing gum were significant only in group Vx (*P*=0.01). This means that pH of saliva increases significantly with consumption of Van-xylitol gum. There was an increase in saliva pH in all groups, except for group C.

Table 4. Comparison of mean salivary volume values among four groups (mL).

Group	Number	pH Conditions	Mean±SD	*P. Value*
Vx	10	Basic	6.86 ± 0.30	0.010
		After chewing gum	7.05 ± 0.38	
Vg	10	Basic	6.49 ± 0.58	0.332
		After chewing gum	6.72 ± 0.66	
C	10	Basic	7 ± 0.41	0.096
		After chewing gum	6.71 ± 0.61	
T	10	Basic	6.75 ± 0.44	0.149
		After chewing gum	6.93 ± 0.62	

Table 5. Comparison of salivary buffering capacity values in four groups.

Group	Number	pH Conditions	Mean±SD	*P. Value*
Vx	10	Basic	6.65 ± 0.25	0.189
		After chewing gum	6.77 ± 0.39	
Vg	10	Basic	7.08 ± 0.20	0.005
		After chewing gum	5.66 ± 0.44	
C	10	Basic	7.06 ± 0.23	0.005
		After chewing gum	5.85± 0.37	
T	10	Basic	7.08 ± 0.25	0.005
		After chewing gum	4.87 ± 0.33	

Table 6. Comparison of mean values of salivary *S. mutans* in four groups (CFU/mL).

Group	Number	pH Conditions	Mean±SD	*P. Value*
Vx	10	Basic	65.06 ± 43.76	0.011
		After chewing gum	9.33 ± 4.12	
Vg	10	Basic	40.80 ± 31.47	0.008
		After chewing gum	30.90 ± 24.32	
C	10	Basic	53.70 ± 12.02	0.051
		After chewing gum	48.60± 11.67	
T	10	Basic	53.50 ± 7.80	0.007
		After chewing gum	18 ± 17.07	

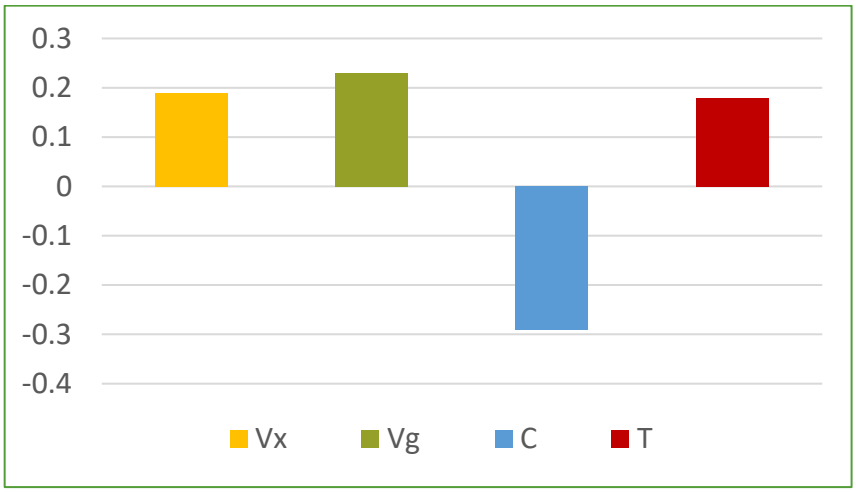


Figure 1. Salivary pH changes in the studied groups. Vx: Van-xylitol, Vg: Van-glucose, C: Chic, T: Trident **(Prepared by Authors, 2025)**.

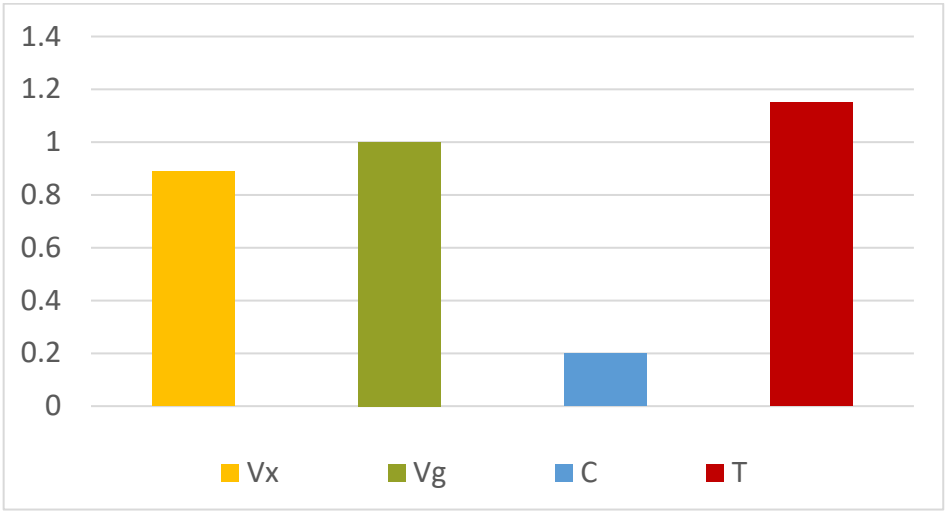


Figure 2. Salivary volume changes in the studied groups. Vx: Van-xylitol, Vg: Van-glucose, C: Chic, T: Trident **(Prepared by Authors, 2025)**.

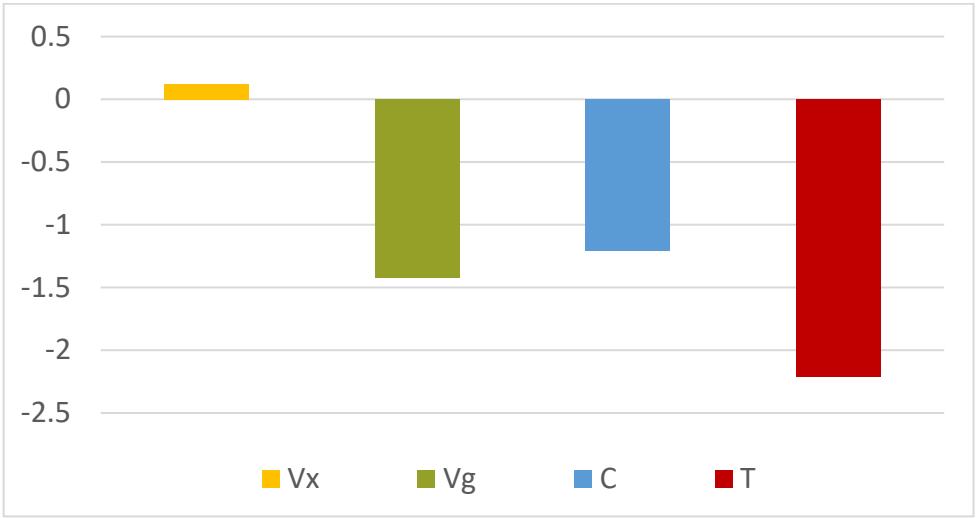


Figure 3. Salivary buffering changes in the studied groups. Vx: Vx: Van-xylitol, Vg: Van-glucose, C: Chic, T: Trident **(Prepared by Authors, 2025)**.

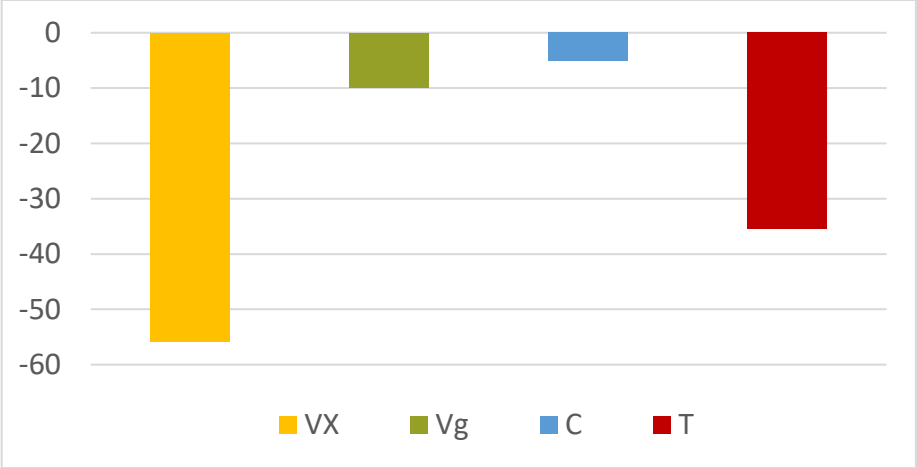


Figure 4. Salivary *S. mutans* changes in the studied groups. Vx: Van-xylitol, Vg: Van-glucose, C: Chic, T: Trident **(Prepared by Authors, 2025)**.

4. Discussion

Tooth decay is one of the most frequent and serious oral health issues worldwide. Dental decay affects everyone at same point in their lives, and roughly 36% of individuals worldwide **(29, 30)**. As non-stimulated saliva is present in the mouth for long time during the day and is more significant in maintaining oral health, it better represents the physiological state of the oral and the entire body **(31)**. Saliva helps the demineralization-remineralization process and the elimination of bacterial substrates by neutralizing pH, although chewing gum influences the saliva flow rate and pH **(21)**. Xylitol has an inhibitory effect on acid production from sucrose and glucose in dental plaque. When xylitol is consumed repeatedly for a long time, it changes the dental flora and reduces acid production by bacteria **(20)**. In this study, pH of saliva increased significantly with consumption of Van-xylitol gum (P=0.010). Comparison between pH changes in four different types of chewing gums showed that Van-xylitol and Trident significantly increased pH of saliva compared to Chic gum (P=0.029 and P=0.043, respectively). Although Van-xylitol, Trident, and Van-glucose gums increased saliva pH, Chic gum containing glucose and sugar decreased saliva pH. Furthermore, despite the fact that group Vg chewing gum include glucose, the salivary pH of this group increased, possibly due to the presence of van (turpentine) in the ingredients of this type of chewing gum. In a study that investigated the impact of chitosan-based chewing gum on *S. mutans* counts and salivary pH, the results showed that salivary pH had no significant change **(32)**. Another trial, which looked at the effect of xylitol and sorbitol chewing gum on the volume and pH of saliva in children aged 13 to 18, found that chewing gum increases saliva flow but reduces saliva pH **(33)**, but in our study, Van-xylitol increased both volume and pH of saliva.

The results of current study showed that the volume of saliva after chewing gum increased in all four groups, but this change was significant in groups Vx and T (groups containing xylitol). In comparison between groups, gums with xylitol (groups Vx and T) increased saliva volume significantly compared to Chic gum (P=0.029 and P=0.043, respectively). This finding was similar to the results of previous studies **(34-36)**. A considerable rise in the mean saliva pH and flow rate was seen in both groups' chewing gum consumers (xylitol and CPP-ACP gums) from the beginning until after spitting gum (similar to our study results for group Vx). Consuming these two gums was suggested in this study because it improved salivary features and avoided additional problems **(34)**. A systematic review study has shown that chewing gum can enhance the unstimulated salivary flow rate in elderly individuals and those with medical conditions who experience dry mouth (xerostomia). The longer the gum is chewed over several days, the greater the improvement in saliva production **(37)**. Therefore, using these gums can also be useful in treating dry mouth.

In our study, buffering capacity of saliva decreased significantly (P=0.005) in groups Vg, C and T compared to before consuming gum, but in group Vx an increase in buffering capacity of saliva was reported after consuming gum, that was not statistically significant. Changes in group Vx were significant compared to other three groups (P=0.000), and group T showed a greater decrease in buffering capacity of saliva compared to groups Vg and C (P=0.002 and P=0.001). These results show that van together with xylitol will have a great effect on improving the buffering

capacity of saliva and as a result, it is beneficial in the field of oral hygiene. In a study to investigate the effects of xylitol and CPP-ACP chewing gums on salivary properties of children with molar incisor hypomineralization, buffering capacity improved in both groups from the start until shortly after spitting gum **(34)**. Also, another study on the effects of CPP-ACP and xylitol-containing chewing gums on salivary flow rate, pH, and buffering capacity in children, showed statistically significant increases in salivary flow rate, pH, and buffering capacity in both groups from the start to immediately after spitting gum (38). Current evidence indicates that adding CPP-ACP to milk, chewing gum, or candy may help remineralization tooth enamel and exhibit some antibacterial effects against dental biofilm. However, further clinical studies are necessary **(39)**.

The effects of four types of chewing gums were studied on the number of salivary *S. mutans*. It was discovered that chewing all four brands of chewing gums (Van-xylitol, Van-glucose, Chic, and Trident) significantly reduces the quantity of *S. mutans* saliva. Trident and Van-xylitol chewing gums had the greatest effect on reducing the bacterial population of *S. mutans*. Also, these two gums were the same in terms of effectiveness on reducing the number of bacteria that were more effective than the other two groups. Chewing gum, particularly gum containing xylitol, has shown to reduce the bacterial population of salivary *S. mutans* in the current and other investigations. The effect of short-term xylitol chewing might be related to decrease in the quantity of *S. mutans*, which used qPCR technique for the first time **(40)**. It was reported in 2020 that xylitol gum decreased bacterial strains linked to caries and periodontal disease from subgingival plaque *in vitro*. The use of xylitol to prevent periodontal disease was recommended in this study **(41)**.

Xylitol chewing gum is recommended as a supplement to tooth brushing for reducing the levels of *S. mutans* and plaque accumulation, which helps in controlling and preventing tooth decay in both children and adults. Additionally, adults with other plaque-related issues, such as periodontal disease, may also benefit from xylitol chewing gum **(42)**. Also, CPP-ACP and xylitol gums alone have a considerable potential to suppress *S. mutans* levels **(5, 43)**.

Limitations

Limitations of this study include the small sample size, short-term duration of intervention, population (all female dental students), and generalizability. It is recommended that clinical trial studies be conducted with larger sample sizes, different populations, and longer durations.

5. Conclusion

Van-xylitol and Triedent gums were the most effective in improving salivary volume and reducing the number of *S. mutans* in saliva, but Van-xylitol was the most effective among the groups in improving salivary pH and increase in buffering capacity of saliva. It seems that turpentine can significantly enhance the properties of xylitol-containing gums. As a result, among those with poor dental hygiene or mental or physical limitations, the use of these gums might be advised as a viable alternative to mechanical treatments. Furthermore, the inclusion of van (turpentine) in chewing gum containing xylitol improves its effectiveness. Turpentine gums have advantages such as adjunct preventive therapy, cost-effectiveness, and user acceptability.

6. Declarations

6.1 Acknowledgment

The authors appreciate the College of Dentistry, Qom University of Medical Sciences, for supporting this work.

6.2 Ethical Considerations

This research was approved by ethics code IR.MUQ.REC.1399.286 in Qom University of Medical Sciences.

6.3 Authors' Contributions

Conceptualization: SH/ Data curation: SH, EK, ZP/ Formal analysis: RF, MS/ Investigation: SH, EK, ZP/ Methodology: SH, ZP, RF/ Project administration: SH/ Supervision: SH/ Validation: SH, MS, EK/ Writing original draft: ZP and EK/ Writing, review and editing: SH, ZP, RF, MS, EK.

6.4 Conflict of Interests

The authors declare no conflict of interest.

6.5 Financial Support and Sponsorship

No financial support or sponsorship was obtained for this research.

6.6 Using Artificial Intelligence Tools (AI Tools)

All authors declare that there is no use of AI Tools in this study, including the writing of this manuscript.

References

1. Signori C, Laske M, Bronkhorst EM, Huysmans MCD, Cenci MS, Opdam NJ. Impact of individual-risk factors on caries treatment performed by general dental practitioners. J Dent. 2019;81:85-90. [DOI:10.1016/j.jdent.2018.12.016] [PMID]
2. Pitts NB, Wright JP. Reminova and EAER: keeping enamel whole through caries remineralization. Adv Dent Res. 2018;29(1):48-54. [DOI:10.1177/0022034517737026] [PMID]
3. Gao X, Jiang S, Koh D, Hsu CYS. Salivary biomarkers for dental caries. Periodontol 2000. 2016;70(1):128-41. [DOI:10.1111/prd.12100] [PMID]
4. Wei SH, Crall JJ, Wefel JS. Dental caries: resistance factors-enamel chemistry and saliva. Systemized Prevent Oral Dis. 2019:43-59. [DOI:10.1201/9780429283758-4]
5. Padminee K, Poorni S, Diana D, Duraivel D, Srinivasan MR. Effectiveness of casein phosphopeptide-amorphous calcium phosphate and xylitol chewing gums on salivary pH, buffer capacity, and Streptococcus mutans levels: An interventional study. Indian J Dent Res. 2018;29(5):616-21. [DOI:10.4103/ijdr.IJDR_166_17] [PMID]
6. Bakhshi M, Kavei D, Asayesh H, Mehdipour A. Evaluating the effect of pregnancy on streptococcus mutans, ph and buffering capacity of saliva. Stud Med Sci. 2015;26(6): 467-74.
7. Afshar H, Nahvi A. Investigating the sweeteners of current gums present in Iranian market. Iran J Pediatr Dent. 2014;9(2):19-24. [DOI:10.29252/ijpd.9.2.19]
8. Mazhari F, Izadi EF. Comparison of the Effect of Natural Turpentine and Synthetic Sugar Free Gums on Dental Plaque pH Recovery. J Dent Mater Tech. 2018;7(3):111-6.
9. de Siqueira Fraga EG, Neto EMR, Campos FMT, da Silva Cavalcante MP, Amorim FA, Martins LFB, et al. Evaluation of chewing gums containing natural products on salivary concentrations of streptococcus mutans in Children. J Young Pharm. 2022;15(1):182-8. [DOI:10.5530/097515050561]
10. Ahmadi-Motamayel F, Akbari E, Mahjoub R, Alikhani MY, Poorolajal J. Effect of chewing gum containing glycyrrhiza glabra, honey, and vitamin E on oral health. J Herb Med 2024;43: 100831. [DOI:10.1016/j.hermed.2023.100831]
11. Weber J, Scholz KJ, Schenke IM, Pfab F, Cieplik F, Hiller KA, et al. Randomized controlled clinical trial on the efficacy of a novel antimicrobial chewing gum in reducing plaque and gingivitis in adolescent orthodontic patients. Clin Oral Investig. 2024;28(5):272. [PMID] [PMCID] [DOI:10.1007/s00784-024-05669-4]
12. Daryaei M, Hoseiny S, Taheri K, Mirzaei J, Mzbani A. Effect of morphological variables of Pistacia atlantica on gum and seed production. Iran J Biol. 2012;25(2):303-15.
13. Niculescu A-G, Grumezescu AM. Natural compounds for preventing ear, nose, and throat-related oral infections. Plants. 2021; 10(9):1847. [DOI:10.3390/plants10091847] [PMID] [PMCID]
14. Esentürk-Güzel İ, Abdo L, Topuzoğlu S, YILDIZ C, YILMAZ F, DÖŞLER S. Natural ingredients included antimicrobial lozenge formulations for oral care. J Res Pharm. 2024;28(1):248-57. [DOI:10.29228/jrp.692]
15. Mobaraki N, Amani F, Najafi E, Gamisi J. Comparison of the Effect of Coffee and Chewing Gum on the Return of Gastrointestinal Motility in Patients Undergoing Cesarean Section. Complement Med Res. 2024;14(3):21-6.
16. Tahmourespour A, Aminzadeh A, Salehifard I. Anti-adherence and anti-bacterial activities of Pistacia atlantica resin extract against strongly adherent Streptococcus mutans strains. Dent Res J. 2022;19(1):36. [PMID] [DOI:10.4103/1735-3327.344159]
17. Ly KA, Milgrom P, Rothen M. The potential of dental-protective chewing gum in oral health interventions. J Am Dent Assoc. 2008;139(5): 553-63. [PMID] [DOI:10.14219/jada.archive.2008.0215]
18. Madrid-Troconis CC, Perez-Puello SdC. Casein phosphopeptide-amorphous calcium phosphate nanocomplex (CPP-ACP) in dentistry: State of the art. Rev Fac Odontol Univ Antioq. 2019;30(2):248-62. [DOI:10.17533/udea.rfo.v30n2a10]
19. Agarwal B, Singh LK. Sugar and Sugar Alcohols: Xylitol. High Value Fermentation Products: Human Health. 2019;1:285-307. [DOI:10.1002/9781119460053.ch12]
20. Kumar K, Singh E, Shrivastava S. Microbial xylitol production. Appl Microbiol Biotechnol. 2022; 106(3):971-9. [PMID] [DOI:10.1007/s00253-022-11793-6]

21. Tusi SK, Jafari A, Marashi SM, Niknam SF, Farid M. Effect of teucrium polium-containing chewing gum on reducing salivary streptococcus mutans counts. J Mashhad Dent Sch. 2018;42(2):141-50.

22. Rafeek R, Carrington CV, Gomez A, Harkins D, Torralba M, Kuelbs C, et al. Xylitol and sorbitol effects on the microbiome of saliva and plaque. J Oral Microbiol. 2019;11(1):1536181. [PMCID] [DOI:10.1080/20002297.2018.1536181][PMID]

23. Navazesh M. Methods for collecting saliva. Ann N Y Acad Sci. 1993;694(1):72-7. [PMID] [DOI:10.1111/j.1749-6632.1993.tb18343.x]

24. Mahjoub S, Ghasempour M, Gharage A, Bijani A, Masrourroudsari J. Comparison of total antioxidant capacity in saliva of children with severe early childhood caries and caries-free children. Caries Res. 2014;48(4):271-5. [DOI:10.1159/000355581] [PMID]

25. Yaghini J, Shirani F, Mogharehabed A, Karimipour H, Sarsangi M. Evaluation of DMFT, salivary pH and salivary buffering capacity in patients with and without calculus referring to Isfahan Faculty of Dentistry. J Isfahan Dent Sch. 2011;6(5):820-7.

26. Kamate WI, Vibhute NA, Baad RK. Estimation of DMFT, salivary streptococcus mutans count, flow rate, pH, and salivary total calcium content in pregnant and non-pregnant women: a prospective study. J Clin Diagn Res. 2017;11(4): ZC147. [DOI:10.7860/JCDR/2017/24965.9516] [PMID] [PMCID]

27. Higuchi M. The effect of oxygen on the growth and mannitol fermentation of Streptococcus mutans. Microbiology. 1984;130(7):1819-26. [DOI:10.1099/00221287-130-7-1819] [PMID]

28. Ma J, Hunjan M, Smith R, Kelly C, Lehner T. An investigation into the mechanism of protection by local passive immunization with monoclonal antibodies against Streptococcus mutans. Infect Immun. 1990;58(10):3407-14. [DOI:10.1128/iai.58.10.3407-3414.1990] [PMID] [PMCID]

29. Karpiński TM, Szkaradkiewicz AK. Microbiology of dental caries. J Biol Earth Sci. 2013;3(1):M21-4.

30. Vos T, Flaxman AD, Naghavi M, Lozano R, Michaud C, Ezzati M, et al. Years lived with disability (YLDs) for 1160 sequelae of 289 diseases and injuries 1990-2010: a systematic analysis for the Global Burden of Disease Study 2010. Lancet. 2012;380(9859):2163-96. [DOI:10.1016/S0140-6736(12)61729-2] [PMID]

31. Mohammadzadeh M, Babaeifar F, Babaei F. Combination thrapy of hydroalcoholic extract of Pistacia atlantica kurdica and fluvoxamine on spatial memory of immobilization rat. J Gorgan Univ Med Sci. 2017;19(3):24-31.

32. Khamverdi Z, Farhadian F, Khazaei S, Adabi M. Efficacy of chitosan-based chewing gum on reducing salivary S. mutans counts and salivary pH: a randomised clinical trial. Acta Odontol Scand. 2021;79(4):268-74. [PMID] [DOI:10.1080/00016357.2020.1836392]

33. Shinde MR, Winnier J. Comparative evaluation of Stevia and Xylitol chewing gum on salivary Streptococcus mutans count-A pilot study. J Clin Exp Dent. 2020;12(6):e568. [DOI:10.4317/jced.55720] [PMID] [PMCID]

34. Prathima GS, Narmatha M, Selvabalaji A, Adimoulame S, Ezhumalai G. Effects of Xylitol and CPP-ACP Chewing Gum on Salivary Properties of Children with Molar Incisor Hypomineralization. Int J Clin Pediatr Dent. 2021;14(3):412. [PMID] [PMCID] [DOI:10.5005/jp-journals-10005-1779]

35. Shinde MR, Winnier J. Effects of stevia and xylitol chewing gums on salivary flow rate, pH, and taste acceptance. J Dent Res Rev. 2020; 7(2):50-5.

36. Nazri WA, Daud W, Rumiati F. Characteristics of saliva in FK Ukrida students after chewing paraffin, xylitol and sucrose gum. Indones J Biotechnol Biodivers. 2021;5(1):24-30. [DOI:10.47007/ijobb.v5i1.73]

37. Dodds MW, Haddou MB, Day JE. The effect of gum chewing on xerostomia and salivary flow rate in elderly and medically compromised subjects: a systematic review and meta-analysis. BMC Oral Health. 2023;23(1):406. [DOI:10.1186/s12903-023-03084-x] [PMID] [PMCID]

38. Hegde RJ, Thakkar JB. Comparative evaluation of the effects of casein phosphopeptide-amorphous calcium phosphate (CPP-ACP) and xylitol-containing chewing gum on salivary flow rate, pH and buffering capacity in children: An in vivo study. J Indian Soc Pedod Prev Dent. 2017;35(4):332-7. [DOI:10.4103/JISPPD.JISPPD_147_17] [PMID]

39. Giacaman RA, Maturana CA, Molina J, Volgenant C, Fernández CE. Effect of casein phosphopeptide-amorphous calcium phosphate added to milk, chewing gum, and candy on dental caries: a systematic review.

Caries Res. 2023;57(2):106-18. [DOI:10.1159/000530638] [PMID]

40. Akgül Ö, Topaloğlu Ak A, Zorlu S, Öner Özdaş D, Uslu M, Çayirgan D. Effects of short-term xylitol chewing gum on pro-inflammatory cytokines and Streptococcus mutans: A randomised, placebo-controlled trial. Int J Clin Pract. 2020; 74(9):e13623. [DOI:10.1111/ijcp.13623] [PMID]
41. Cocco F, Cagetti MG, Majdub O, Campus G. Concentration in Saliva and Antibacterial Effect of Xylitol Chewing Gum: In Vivo and In Vitro Study. Appl Sci. 2020;10(8):2900. [DOI:10.3390/app10082900]
42. Söderling E, Pienihäkkinen K. Specific effects of xylitol chewing gum on mutans streptococci levels, plaque accumulation and caries occurrence: a systematic review. BMC Oral Health. 2025;25(1):1275. [PMID] [PMCID] [DOI:10.1186/s12903-025-06602-1]
43. Arora N, Hande A. The Effect of Chewing Gum Containing Casein Phosphopeptide-Amorphous Calcium Phosphate on Salivary Streptococcus mutans. J Datta Meghe Inst Med Sci Univ. 2019; 14(3):171-4. [DOI:10.4103/jdmimsu.jdmimsu_47_19]