



## ORIGINAL ARTICLE

## Laboratory Evaluation of the Effect of Common Snacks Consumption on Oral Saliva pH

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## KEYWORDS

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**ABSTRACT:** The type of diet is one of the important causes of caries and one of the techniques to evaluate the carcinogenicity of foods is to measure the pH of saliva after consuming them. This study aimed to measure the pH of artificial saliva after exposure to a series of common snacks in the community. In this laboratory study, all samples (including 18 common snacks in the community) were completely crushed. Then, 8 g of each sample was dissolved in 50 ml of artificial saliva and continuously mixed with a magnet in a human. The pH of the solutions was then measured by a digital pH meter at intervals of 0, 1, 2, 4, 6, 8, and 10 minutes after the onset of dissolution. Statistical analysis was performed with related tests and SPSS statistical software version 22. The pH of all groups except chocolate and whole wheat biscuits decreased significantly during 10 minutes (P-value <0.05). The obtained results indicated that the groups lowered the pH of artificial saliva below the critical pH (5.5) including vinegar chips, peanut vinegar, cherry industrial dish, home-made dish, industrial swallow, house swallow, and plum. The pH level of the other groups was higher than the limit was critical. The snack with the least pH change and the snack with the most and most severe pH change were whole-grain biscuits and homemade desserts, respectively. All snacks tested except chocolate and whole wheat biscuits significantly reduced the pH of artificial saliva. The pH drop was high for acidic groups.

## INTRODUCTION

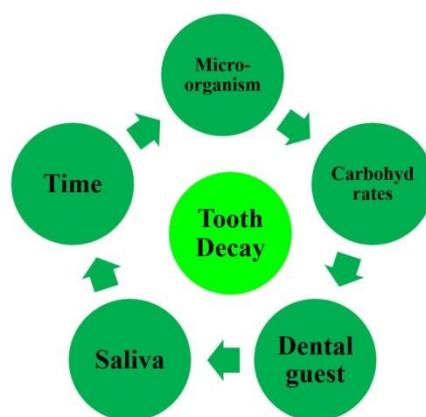
According to the World Health Organization (WHO), oral disease is a major global health problem, with millions of people losing their teeth each year due to caries [1-3]. Caries is a multifactorial disease whose onset and progression are influenced by saliva, genetics, protective factors such as fluoride, and diet. Its consequences include pain and reduced quality of life and the imposition of high costs [3-6]. Over time,

people's lifestyles have changed their tastes and eating habits, so there will be a tendency for people to consume cariogenic products like snacks [5-7]. A snack is a portion of food that is smaller than regular meals and it is usually used as a snack. Frequent consumption of snacks and their persistence on the teeth has been reported in many studies as an important and well-known factor in increasing the process of chemical dissolution (erosion)

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and dental caries [5-9]. Tooth decay begins when carbohydrates are fermented in the mouth by bacteria, which accumulate in a dense matrix called plaque. The time duration of the pH of the oral environment remains low as a critical period for tooth structure. It usually reaches a critical pH when saliva stops being saturated with calcium and phosphorus. Under normal conditions the critical pH of saliva is 5.5, at less the critical pH of the minerals of the tooth may dissolve in the normal resting pH is between 6 and 7. The buffer solution can keep the pH of saliva constant [8-12]. Decreased salivary

buffering power increases the prevalence of erosion and caries in which the saliva secretion changes over time. Adults can produce 1-1.5 liters of saliva per day, which decreases during sleep [11-14]. The rinsing effect of the flushing saliva stream removes free microorganisms from the oral environment by swallowing and this effect increases during chewing. The high volume of saliva can dilute or buffer biofilm acids. In other words, the volume and buffering capacity of saliva play a major role in protecting against decay as shown in Figure 1 [14-17].



**Figure 1.** The main factors in causing dental caries.

Buffers are important in reducing the potential for acid production by urea and saline tetrapeptide molecules in saliva, which contains lysine and arginine. Their hydrolysis leads to the production of ammonia and eventually increases leading to a pH value [17-19]. For demineralization of tooth enamel, fermentable carbohydrates and bacterial plaque must be present on the tooth surface for the period to produce acid and cause tooth decay. Carbohydrates provide the necessary substances for the production of acid and extracellular polysaccharides by bacteria and plaque. Therefore, most foods and beverages containing sugar cause a rapid decrease in pH to the point that they cause demineralization of the enamel. The plaque holds the acid for a short time, and it takes about 30 to 60 minutes for it to reach its normal pH=7 [16-19]. The gradual return of the pH to the normal level is due to the release of acid and the presence of buffers in the plaque and the presence of a salivary layer on it. In case of repeated consumption of sugar and elimination of plaque regeneration time to neutralize the pH, acidic plaque remains and causes demineralization of the tooth. Also,

to monitor industrial production and warn factories producing cariogenic products, it is necessary to make scientific and accurate observations about common products in several countries as well as to prevent possible injuries and maintain oral health. The teeth of society, especially children and adolescents, documented awareness of society is essential. With a report of studies on snack consumption and salivary pH reduction, it is discovered that a comprehensive study on the types of snacks and their effect on salivary pH has not been performed in the laboratory. Therefore, to measure salivary pH at different intervals after dissolution for a variety of popular snacks in Iran, author decides to consider their effect using an analytical technique.

## MATERIALS AND METHODS

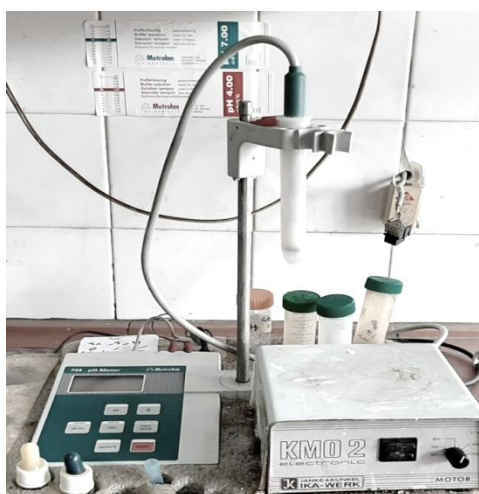
The present study shows an experimental laboratory study was conducted on 18 materials that influence on pH of saliva as shown in Table 1. The statistical population of this study was 18 types of common snacks in the community were listed by their names and brands.

**Table 1.** Average pH of different materials by measurement time

Matter	pH10	pH8	pH6	pH4	pH2	pH1	pH0
Cheetos	6.43	6.44	6.44	6.44	6.45	6.53	7
Simple chips	6.51	6.53	6.54	6.54	6.61	6.79	7
Vinegar	5.43	5.45	5.40	5.42	5.51	5.64	7
--	6.66	6.70	6.76	6.73	6.82	6.84	7
Vinegar peanuts	5.46	5.48	5.48	5.52	5.69	6.14	7
Cherry Lavashka	3.12	3.13	3.16	3.22	3.46	5.51	7
Homemade lavash	4.14	4.29	4.49	5.00	5.62	6.15	7
Industrial Cranberry	3.48	3.49	3.49	3.47	3.45	3.87	7
Homemade crab	3.20	3.19	3.19	3.20	3.24	3.83	7
Plum	3.17	3.25	3.38	3.65	4.21	5.67	7
raw Pistachios	6.67	6.68	6.71	6.72	6.79	6.81	7
Cake	6.83	6.82	6.83	6.81	6.82	6.83	7
Simple Petty Biscuits	6.68	6.66	6.67	6.67	6.67	6.66	7
Wholemeal biscuits	6.95	6.94	6.93	6.93	6.94	6.94	7
Lemon wafer	6.80	6.8	6.78	6.81	6.85	6.90	7
Soft drink pastilles	6.29	6.37	6.45	6.58	6.70	6.81	7
Chocolate	6.79	6.76	6.80	6.81	6.87	6.93	7
Vanilla ice cream	6.82	6.79	6.76	6.78	6.79	6.77	7

Puffs (Chitose) / Simple Chips (Chitose) / Vinegar Chips (Chitose) / Puff (Chitose) / Peanut Vinegar (Chrysanthemum) / Industrial Lavash (Sun) / Homemade Lavash/Industrial Qaraq (Haj Ismail) / Homemade Cranberry / Plum (Shemshak) / Raw Pistachio / Cake (Sweet Honey) / Plain Biscuit (Petibor) / Whole Biscuit (Golden Stem, Mino) / Lemon Wafer (Sweet Honey) / Soft Pastels (Sweet Honey) / Cocoa Chocolate (Baraka) / Vanilla Ice Cream (Homeland). In this study, artificial saliva was made according to the Mondelli method and with Merck raw materials. All samples were completely

crushed separately in a mortar, then 8 g of each material was dissolved in 50 ml of artificial saliva in a human and continuously mixed with a magnetic stirrer. It was measured by a Digital pH meter (METROHM 744) made in Switzerland, at intervals of 0, 1, 2, 4, 6, 8, and 10 minutes after the pH of dissolution of the solutions. This device consisted of an electrode connected to a digital pH meter, which placed electrode in the solution and the sample pH was displayed on the device monitor as can be seen in Figure 2.

**Figure 2.** Digital pH meter for determination of pH.

Caries are the destruction of an area of hard dental tissue that is involved in a combination of different factors as shown in Figure 2. These factors include the presence of microorganisms, tooth tissue conditions (as hosts), saliva, and the presence of fermentable carbohydrates [13-15]. This device was calibrated before the test and it should be noted that one molar solution of NaOH was added to the samples to standardize the base pH value. Also, the electrode heads were washed with distilled water after each measurement and then dried. To prevent human and laboratory errors, measurements were performed twice for each sample. Statistical analysis was performed with SPSS version 22 software. Descriptive information was provided by the mean standard deviation ( $\pm$ SD). Tests included one-way ANOVA, Tukey's Multiple comparisons, and the P-value was considered as 0.05. This work was conducted at Medical University of Isfahan with Ethical code number: 398506.

## RESULTS AND DISCUSSION

The average pH of each food group at times 0, 1, 2, 4, 6, 8, and 10 as well as the average of their set at each time is shown in Table 1. The basal pH in all groups is zero after 7 minutes and the range of pH changes was 3.6-12.94 and the groups that brought saliva pH below the critical pH of 5.5 are including the following Vinegar Chips, Vinegar Peanuts, Cherry Industrial Dishes, Homemade Dishes, Industrial Cranberries, Homemade Cranberries, and Plums. In other groups, the pH was above the critical level. Figure 3 shows the trend of pH changes of all snacks after 10 minutes. As it turns out, the snacks with the lowest and highest pH changes were whole-grain biscuits and homemade cranberries,

respectively.

Tukey analysis showed that there was a significant difference between plain chips and vinegar chips (P-value = 0.002). Also, there was a significant difference between home and industrial cherry dishes (P-value = 0.005). There was no significant difference between domestic and industrial cranberries (p-value = 0.983) and the pH differences for each snack at different times significantly using repeated measures analysis of variance for each of the ingredients separately except for chocolate and whole-grain biscuits (P-value <0.05). According to several investigations, the studied snacks in this study can be divided into two groups. The snacks that lower the pH of artificial saliva above the critical level such as whole meal biscuits, cakes, vanilla ice cream, lemon wafers, chocolate, plain biscuits, raw pistachios, plain chips, puff pastry, and soft drink pastilles. However, it is not possible to determine the critical pH value because the critical pH varies between different individuals and parts of the mouth, and the critical pH values have been reported in a wide variety of sources. Comparing the above two groups, we find that the first group contains a variety of acids and is highly acidic which rapidly reduces the pH of saliva. It is known that in the presence of these materials with the teeth and oral mucosa, the risk of damage to the oral structure raises the alteration. Also, the possibility of extensive erosion of teeth in this food group is not unexpected and in people with lesions and caries of the roots and gingival resorption, dentin can be destroyed progressively and quickly like in the group of plates that have high adhesion.

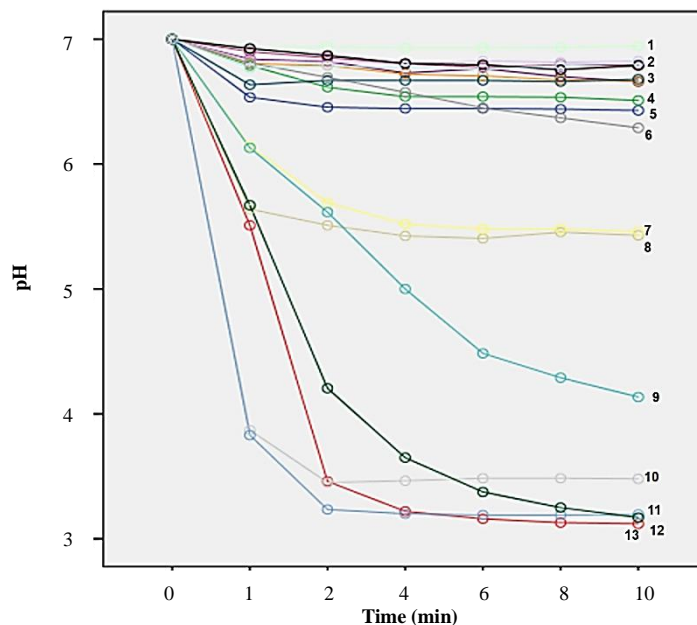


Figure 3. Changes in pH by the material overtime after 10 minutes

The second category contains more carbohydrates, which is not able to lower the pH of saliva as much as snacks containing acid. However, due to their high carbohydrate content and high adhesion to the teeth and placement in the peat and fissure of the teeth, and high potential. In addition, in the oral environment, cariogenic bacteria in

the biofilm metabolize small carbohydrates for energy and by-products of organic acid, which double the pH. Figure 4 demonstrate the comparison of pH changes of plain, vinegar chips/home, and industrial cranberries/home.

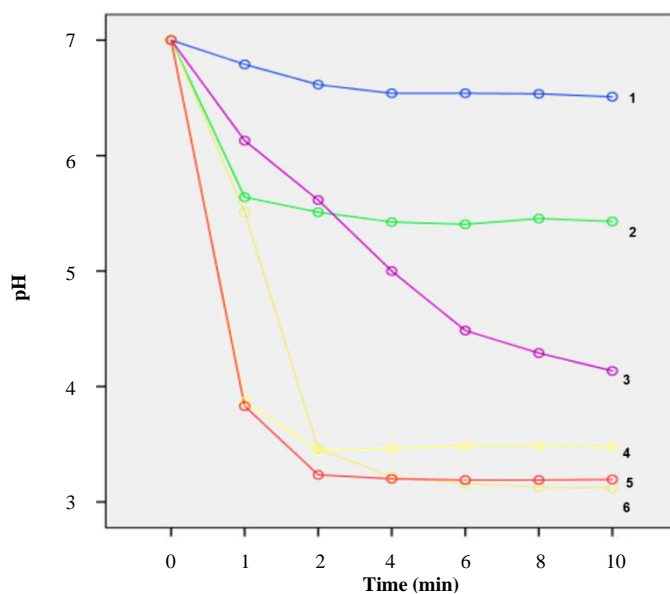


Figure 4. Comparison of pH changes trend of plain and vinegar chips/home and industrial cranberries/home and industrial.

Davies et al. [10] consider the range of initial pH changes of sour chocolate in the range of 2.30-3.14. However, the lowest pH was for cherry industrial dessert at 3.12 in this study, which is more acidic than sour

chocolate. Two reasons can be mentioned for this difference, the difference in the concentration of the substances, and another reason is the use of deionized water as a solution. Therefore, it can be said that artificial

saliva due to the presence of sodium ions, calcium, and urea is more resistant than deionized water to lower the pH. Comparisons indicate that it can be predicted that saliva plays an important role in its proteins and buffering power. Brand et al. [11], found that candies have a highly acidic pH (1.3-2.3) and reported that after consuming candies, the pH of saliva reaches the range of 4.5-4.4 and among 7 types of candies, 5 of them reduce the pH below the critical level. They stated that the pH and saliva flow returned to normal after 2-3 minutes. In the present study, the pH of all groups (except for housewives and plums) decreased significantly after 3 minutes and the graph was almost stable, but did not return to normal and basic pH.

The difference between the two studies can be seen in clinical factors. In the first study, due to the circulating flow of saliva in the mouth and swallowing, as well as proteins and enzymes in saliva, the pH of saliva returned to normal. In our study, the pH remained approximately constant after 3 minutes. Researchers concluded that despite the artificial saliva, sour candies had a lower pH and a higher acidic titer than flavorless candies (pH 7-67 / 27). Also, the pH of tasteless candies in water was lower than their pH in artificial saliva, which the author attributes to the presence of sodium bicarbonate ions in artificial saliva. With these descriptions, the power and capabilities of saliva can be understood, even if it is synthetic [9]. Classifying food into caries and non-caries groups allows for a proper diet to reduce the risk of caries. Sugars and other fermentable carbohydrates are the basis for the onset of decay. On the other hand, sugars are other foods that have a protective role and act

for remineralization and are called anti-cariogenic or cariostatic materials such as milk, plant foods, and sugar substitutes [33-37]. Tayab et al. [12], reported that the pH of saliva decreased in the first 5 minutes of chocolate consumption (from 6.94 to 6.74) and returned to normal after 60 minutes. In our study, the pH of artificial saliva after exposure to chocolate decreased from 7 to 6.79, which is similar to the above study. The author further states that the pH of saliva has increased significantly after consuming cheese, which is due to its high protein and phosphate content. This content suggests chewing some cheese after consuming carbohydrate snacks to keep the saliva pH high. The author also suggests that pH test strips are an effective educational tool in that children can easily see the acidic effects of snacks on saliva due to the color codes on the strip [26-34]. Bhat et al., [13] stated that the pH reached 7.99 in the first 10 minutes after consuming plain milk, and after 30 minutes the pH reached more than its baseline value. The pH after consuming plain milk rose slightly above the baseline of 6.53 and reached this level again after 10 minutes [14].

Figure 5 shows the evolution of the pH of the home-grown cranberry over time which has a downward trend after 1 and 2 minutes. Figure 6 indicates the evolution of the pH of the home-grown cranberry over time. In the present study, the pH of artificial saliva during dissolving vanilla ice cream decreased from 7 to 6.81. From these studies, it can be concluded that milk and its products either do not change the pH of saliva or cause a slight decrease in its amount, which may be due to minerals, casein, and their protein components.

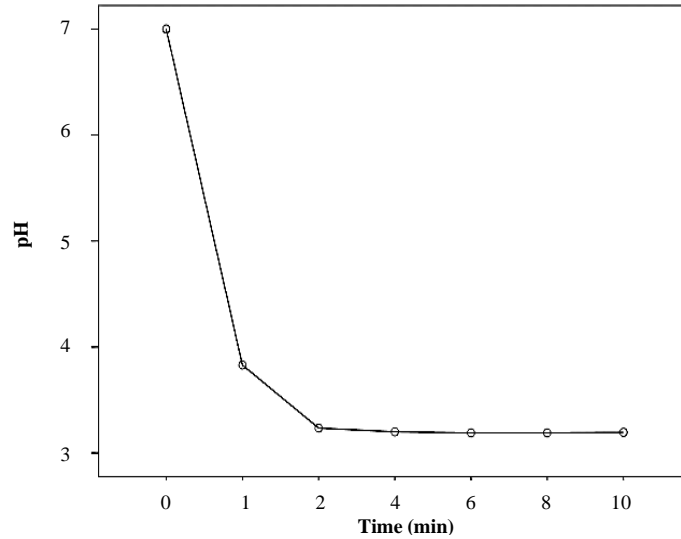


Figure 5. The evolution of the pH of the home-grown cranberry over time.

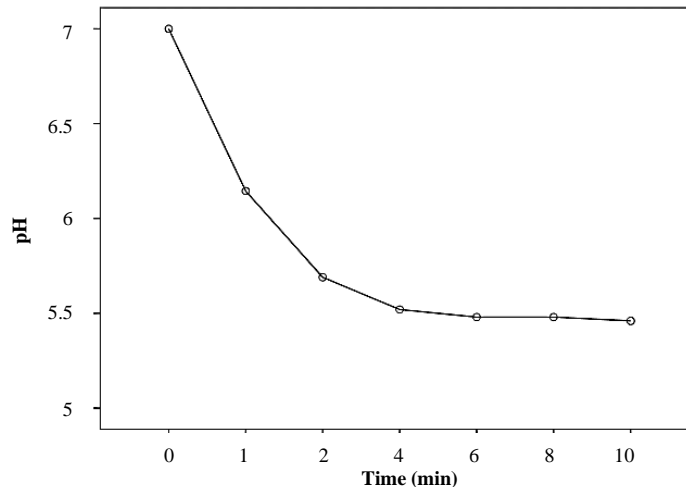


Figure 6. The evolution of the pH of the home-grown cranberry over time.

Milk and dairy consumption have also been shown to be inversely related to the number of decayed teeth, which is due to the absorption of milk protein by tooth enamel. Gupta et al. [15] stated that although milk reduced the pH of saliva, it did not bring it to the critical pH value. Contrary to the existing reports and the results of the present study of changes in pH during the consumption of chocolate, biscuits, and chips, Pachori et al. [16] study shows a decrease in pH in the range of 0.2 to 0.5. This difference may be due to the different combinations of these products in Iran and India [16]. In the study of Gupta et al., Saliva pH was measured for 12 minutes after consuming 5 types of snacks and 3 types of drinks. Meanwhile, all products lowered the pH except chewing gum. In the above study, a decrease in pH was observed

during the consumption of biscuits and chocolate, which was not as critical as the pH of 5.5. The decrease in pH was related to snacks and drinks for up to 4 minutes [15]. Caries is a multifactorial disease whose onset and progression are influenced by saliva, genetics, protective factors such as fluoride, and diet whose consequences include pain and reduced quality of life, and the imposition of high costs [5-7, 35-37]. Over time, people's lifestyles have changed their tastes and eating habits, so there is a tendency for people to consume cariogenic products like snacks [3, 29-34]. A snack is a portion of food that is smaller than regular meals and usually used as a snack. Frequent consumption of snacks and their persistence on the teeth has been reported in many studies as an important and well-known factor in

increasing the process of chemical dissolution (erosion) and dental caries. Tooth decay is an infectious and nutritionally related disease. Nutritional patterns such as the frequency of consumption of carbohydrate-containing products and their presence in the mouth have been identified as important factors in caries. Diet counseling plays a key role in preventive dentistry, and oral health care providers should provide their patients with the necessary diet-based advice based on scientific evidence and persuade them to improve their eating behaviors. Kumar et al. [17], introduced the saliva pH measurement after consuming 5 types of biscuits (sugar biscuits, salt biscuits, bran biscuits, chocolate biscuits, and cream biscuits). Sugar and chocolate biscuits had the lowest pH, but none of them reached the critical pH

range, even the critical pH of ivory (6.2). In the present study, products such as biscuits, wafers, and cakes had pH values similar to those mentioned in the study. In the above study, whole wheat biscuits have a curve with small changes in saliva pH, which is similar to whole wheat biscuits with very limited changes in our study [17]. Saliva is very important for maintaining oral health, because increased saliva flow increases pH, and promotes remineralization and buffering capacity [24-27]. Salivary buffering capacity is one of the factors that affect a person's risk of caries. The ability of saliva to buffer acids is essential to maintain a pH above the critical pH to protect teeth from demineralization as shown in Table 2.

**Table 2.** Saliva compounds and their functions

Functions	Components
Lubrication	Mucin, proline-rich glycoproteins, water
Antimicrobial action	Lysozyme, lactoferrin, lactoperoxidase, mucins, cysts, statins, immunoglobulins, proline-rich glycoproteins, IgA
Maintaining mucosa integrity	Mucins, electrolytes, water
Cleansing	Water
Buffer capacity and remineralization	Bicarbonate, phosphate, calcium, statin, proline-rich anionic proteins, fluoride
Preparing food for swallowing	Water, mucins
Digestion	Amylase, lipase, ribonucleases, proteases, water, mucins
Taste	Water, Justin
Phonation	Water, mucin

Shetty et al. [18] conducted a study on the acidity of 4 common crispy snacks in India. These products are made from frying and roasting potatoes, which is similar to chips in Iran. They stated that the plaque pH decreased in the first minutes but returned to normal after 30 minutes. Although the range of pH changes was often above 7, the range of changes similar to the pH changes of artificial saliva after dissolving chips is simple.

## CONCLUSIONS

The results of this study showed that all snacks lowered the pH of saliva. These pH changes were greater for groups that contained acids or an acidic flavor added to

them. Most of the pH drop was up to 3 minutes, in which the pH of the snacks became more stable. The results of this study cannot provide a definitive prediction of changes in saliva pH during snack consumption. Some effective factors including salivary proteins, salivary flows in the mouth, chewing and swallowing as well as food adhesion were available. Extensive clinical studies on snacks and their effect on oral conditions including pH changes should be evaluated. One of the limitations of this study is the laboratory nature of the design, which causes other shortcomings and limitations. These include the use of artificial saliva, adjacent to snacks and saliva as desired. The lack of a continuous salivation system and the



effect of flushing and salivary clearance, the lack of salivary proteins and enzymes that certainly have a double buffering effect. One of the strengths of this study is the extensive study of common snacks in Iranian society. It can be mentioned that there is no similar study in previous texts and also the use of artificial saliva, which in some studies used deionized water.

#### ACKNOWLEDGEMENTS

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#### Conflict of interest

The authors declare that they have no conflict of interest.

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