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The effect of sugar-sweetened carbonated soda and carbonated mineral water on the salivary pH value

Nataša Puškar[1], Milica Puškar[1], Milica Jeremić Knežević[1], Daniela Đurović Koprivica[1], Ljiljana Andrijević[2]

[1]University of Novi Sad, Faculty of Medicine, Department of Dental Medicine, Novi Sad, Serbia;
[2]University of Novi Sad, Faculty of Medicine, Department of Biochemistry, Novi Sad, Serbia

SUMMARY

Introduction Saliva plays an important role in maintaining oral health. Its buffer capacity helps neutralise acidic products from food, drinks and dental plaque. Saliva composition, quantitative and qualitative properties, such as acidity, are associated with the occurrence of dental caries, non-carious lesions and periodontitis. The aim of the study was to examine the change in salivary acidity after taking carbonated mineral water and sugar-sweetened carbonated soda in subjects aged 18 to 25 years.

Materials and method The study research was conducted at the Dentistry Clinic of Vojvodina on 30 adult subjects. Salivary acidity was measured with a digital pH meter before, and 5, 10 and 20 minutes after taking 200 ml of carbonated mineral water and 200 ml of sugar-sweetened carbonated soda. The obtained results were statistically processed by mixed, combined analysis of variance.

Results Consumption of carbonated mineral water and sugar-sweetened carbonated soda led to a change in salivary acidity in the examined time intervals. Carbonated mineral water reduced the salivary acidity, and after 20 minutes the acidity recovers reaching levels close to the initial pH value. After taking sugar-sweetened carbonated soda, the salivary pH value dropped. It reached the lowest mean value 5 and 10 minutes after taking the beverage, and then increased slightly without reaching the initial value. In some subjects, the minimum value of the salivary pH obtained was below the value considered critical for the occurrence of enamel and dentin demineralisation.

Conclusion Carbonated mineral water and sugar-sweetened carbonated soda affected the salivary pH value within a period of 20 minutes after consumption. Consuming sugar-sweetened carbonated soda can have adverse effects on oral health.

Keywords: saliva; pH; beverage

INTRODUCTION

Oral cavity, as the beginning of digestive tract, is in direct contact with the external environment. Saliva, which is secreted by the exocrine salivary glands, is of great importance for oral biochemistry. It is an aqueous solution secreted by large salivary glands (parotid, submandibular and sublingual), small salivary glands located in the mucous membrane of the lips, cheeks, tongue, palate and pharynx and gingival cervical fluids [1]. There is 700–800 ml of saliva secreted daily on average, which is swallowed and absorbed in the digestive tract under physiological conditions [1]. Saliva is composed of 99.5% water and 0.5% salts and proteins [2]. The most important proteins are the alkaline glycoproteins that protect tooth enamel and are responsible for the viscosity of saliva and lubrication [2]. Saliva also contains immunoglobulins and lysosomes that lyse bacteria, the enzymes alpha amylase and hyaluronidase [2]. In part of population, antigens of blood groups A and/or B are also found in saliva [2].

Saliva plays an important role in maintaining oral health. It protects and wets the oral mucosa, plays an important role in the processes of mineralisation and demineralisation of both enamel and dentin [3]. With its immunological and non-immunological components, it exhibits antimicrobial activity, enhances the taste of food and drink and initiates the process of food digestion [3]. Its buffering capacity helps neutralise acidic products from food, drinks and dental plaque [2, 3]. Disruption of homeostasis, which is maintained by saliva, leads to poor oral health [4].

The acidity of saliva, i.e., salivary pH depends on bicarbonate, phosphate and protein. Calcium and phosphate are supersaturated at physiological ("normal") pH, and the buffering effect of saliva depends mainly on bicarbonate and phosphate [3]. With sufficient salivary secretion rates (more than 1 ml per minute), the concentration of bicarbonate is 30-60 mmol/l, and the pH is 7.5-7.8 [3].

Saliva prevents damage to the mucous membranes of the mouth and teeth as well as the occurrence of dental erosions by washing away pathogenic microorganisms, breaking down food and destroying the bacteria as such, including cariogenic ones [4]. After eating and drinking, there is a drop in the salivary pH value, which can have harmful consequences, the appearance of dental caries and dental erosions in particular [5]. It is believed that any factor that can affect the change of oral microflora and local environmental conditions, such as a changed

Address for correspondence: Nataša PUŠKAR, 5 Pavleka Miškine St, 21000 Novi Sad, Serbia; *na_puskar@yahoo.com*

salivary pH value, leads to an imbalance in the biofilm and multiplication of cariogenic bacteria, which leads to the occurrence of dental caries [5]. Thus, the buffer system of saliva plays a key role in the prevention of both dental caries and dental erosions, which are dental lesions on the tooth surface caused by a multitude of factors, but without bacterial action [5].

There are numerous authors who have examined the influence of various beverages on the salivary pH. The studies have mostly been done *in vitro*, focusing on examining the buffering capacity of saliva or the ability of acidic beverages to damage the teeth enamel and dentin [6–12]. *In vivo* studies have been carried out on children and young people aged 18-25 by measuring the salivary pH value at different intervals after taking fruit juices, teas, wine or consuming certain types of sweets or foods [1, 4, 5,13–20]. *In vivo* findings on the salivary pH values in humans after taking carbonated mineral water and sugar-sweetened soda at different time intervals in the scientific literature are few.

This study was aimed at examining the influence of sugar-sweetened carbonated soda and carbonated mineral water on the salivary pH value 5, 10 and 20 minutes after consumption in subjects aged 18 to 25.

MATERIALS AND METHOD

The research was conducted at the Clinic for Dentistry of Vojvodina and was approved by the Ethics Committee of the Clinic. The sample consisted of 30 subjects, Dentistry students at the Faculty of Medicine in Novi Sad, aged 18 to 25, of both sexes. Before participating in the study, the subjects were informed about the method and objectives of the research and signed their voluntary consent. The selection was made by the method of random choice. The first 30 students who registered for the offered sessions and met the criteria for inclusion in the research were selected as subjects.

The research exclusion criteria were:

- candidates undergoing orthodontic or dental restorative treatment;
- pregnant women;
- candidates suffering from systemic diseases;
- candidates suffering from salivary gland diseases;
- candidates on medications that can affect the salivary gland function (antihistamines, anticancer drugs...) in a period of at least two months before the start of the experiment and those who had been taking antibiotics during the same period.

Saliva sampling was carried out at 9:00 a.m., and the subjects were instructed not to brush their teeth in the morning on the day samples were to be taken and not to eat or drink until the beginning of the experiment. Before the start of the experiment, the pH value of the tested beverages and the pH value of saliva of all subjects were determined. Saliva was collected by spitting into a sterile test tube through a sterile funnel. The pH value was measured with a digital pH meter (InoLab, Xylem analytics, Germany) in the biochemistry laboratory of the Faculty of Medicine in Novi Sad, which is located at the Dental Clinic of Vojvodina. Calibration of the pH meter was performed before measuring. The glass electrode was immersed in the test tube, and after each measurement, it was carefully cleaned with distilled water and dried with filter paper. The salivary pH value was determined by 3 consecutive immersions of the pH meter glass electrode in the test tube with the sample, and the mean value was taken as the obtained value. When immersing the glass electrode, the measurement was performed after 10 seconds, in order to allow the sampled liquid to stabilise.

The examination was performed on two consecutive days, on the first day for carbonated mineral water, and on the second day for sugar-sweetened carbonated soda. After the initial salivary pH determination, all subjects were given 200 ml of carbonated mineral water (bottled natural mineral water Vrnjci, Voda Vrnjci a.d., Vrnjačka Banja, Republic of Serbia) at room temperature on the first day, and 200 ml of sugar-sweetened carbonated soda (Coca Cola®, HBC-Srbija d.o.o. Beograd) at room temperature on the second day. They were instructed to drink the beverage within one minute. After that, saliva samples were taken again and its pH value was measured.

Saliva was collected 5, 10 and 20 minutes after the start of drinking the beverage, and the sample was measured 5 minutes after the sample collection. The obtained results were statistically analysed. A mixed, combined, analysis of variance: "mixed design ANOVA" was used.

RESULTS

The study included 30 subjects, of whom 70% were female, and 30% male. A total of 720 pH measurements of saliva and 18 pH measurements of beverages were made. The average pH value of carbonated mineral water was 6.32, while the pH of the sugar-sweetened soda was 2.62. The salivary pH values were influenced by two independent variables: time – when the saliva pH measurements were performed (0, 5, 10, 20 minutes after the beverage consumption) and the type of liquid that was taken – carbonated mineral water (Water Group) or sugar-sweetened carbonated soda (Soda Group).

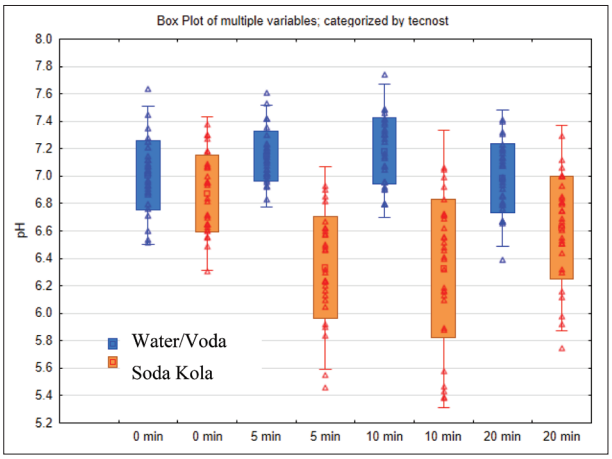
The salivary pH values for measurements before and after taking carbonated mineral water and sugar-sweetened carbonated soda are shown in Table 1. Different average pH values are observed for the two groups, as well as for different measured intervals.

The pH values for two groups (Water Group and Soda Group) for different measuring intervals are presented by a box plot in Graph 1, and the average values of this variable by a line chart in Graph 3.

There was statistically significant difference between the salivary pH value in the group that consumed carbonated mineral water and the group that consumed sugar-sweetened carbonated soda, $F(1,58)=60.02$, $p=0.00$, regardless of the measurement time intervals. For the main effect of the group, the effect size measure, the partial eta squared $\eta^2=0.51$, which is a very significant variance between the tested beverages.

Table 1. The salivary pH values in the measured intervals for Water and Soda Groups
Tabela 1. PH vrednosti pljuvačke u merenim vremenskim intervalima za grupe voda i kola

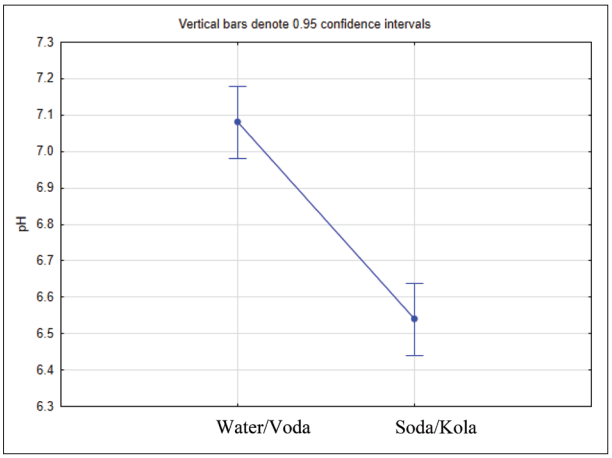
Time Vreme	Group Grupa	pH value PH vrednost						
		Number Broj	Average Srednja vrednost	95% confidence interval 95% Int. poverenja		Min	Max	Std Dev
0 min	Water Voda	30	7.01	6.91	7.10	6.52	7.64	0.25
	Soda Kola	30	6.87	6.77	6.98	6.31	7.38	0.28
5 min	Water Voda	30	7.15	7.08	7.21	6.83	7.61	0.18
	Soda Kola	30	6.33	6.19	6.47	5.46	6.93	0.37
10 min	Water Voda	30	7.18	7.09	7.28	6.80	7.74	0.24
	Soda Kola	30	6.33	6.14	6.52	5.38	7.06	0.51
20 min	Water Voda	30	6.99	6.89	7.08	6.39	7.41	0.25
	Soda Kola	30	6.62	6.48	6.76	5.75	7.29	0.37



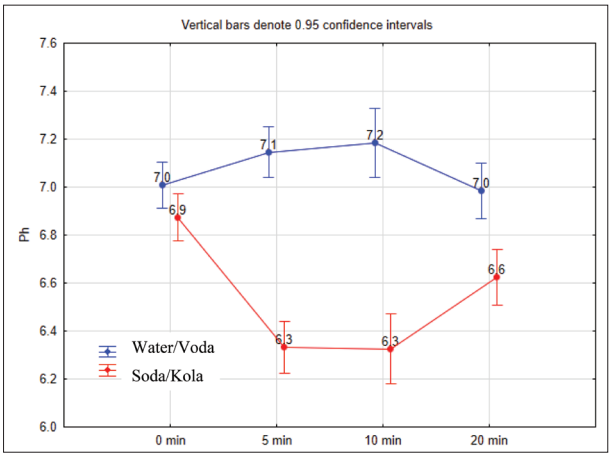
Graph 1. Box plot for salivary pH values during different time intervals for Water Group and Soda Group
Grafikon 1. Boks dijagram za vrednosti pH pljuvačke tokom različitih vremenskih intervala za grupe voda i grupe kola

The change in the pH value over time was significantly different in both subjects who took water and those who took soda (Graph 3). For the interaction of group and time, the effect size measure partial eta squared is η^2=0.443, which is a medium effect size.

When the differences between the groups were observed, by *post hoc* analysis, Tukey's test can be used to determine which average values are statistically significantly different. It is found that in the intervals of 5 min., 10 min. and 20 min., the average salivary pH values in the Water Group were significantly higher than the corresponding average salivary pH values in the Soda Group. The first measurement of the salivary pH value, before consumption of beverages, the average showed no difference in values. The results are presented in Table 2.



Graph 2. Linear chart for average salivary pH values for Water Group and Soda Group, regardless of the time elapsed since the consumption of beverages, with corresponding 95% confidence intervals
Grafikon 2. Linijski dijagram za prosečne vrednosti pH pljuvačke za grupe voda i grupe kola, bez obzira na vreme proteklo od momenta konzumiranja napitaka, sa odgovarajućim 95% intervalima poverenja



Graph 3. Linear chart for average pH values over time for Water Group and Soda Group, with corresponding 95% confidence intervals
Grafikon 3. Linijski dijagram za prosečne vrednosti pH tokom vremena za grupu koja je konzumirala vodu i grupu koja je konzumirala kola napitak, sa odgovarajućim 95% intervalima poverenja

Table 2. Results of Tukey's *post hoc* test for comparison of average salivary pH values between the Water Group and Soda Group
Tabela 2. Rezultati Takijevog post hoc testa za poređenje prosečnih pH vrednosti pljuvačke između grupe voda i kola

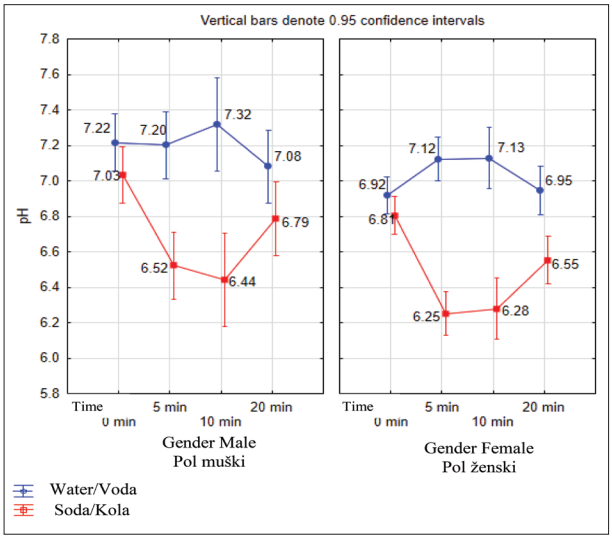
	Water average Voda – srednja vrednost	Soda average Kola – srednja vrednost	p-value p-vrednost
0 min	7.01	6.87	0.74224
5 min	7.15	6.33	0.00012
10 min	7.18	6.33	0.00012
20 min	6.99	6.62	0.00088

Table 3 shows which average values in Water Group and Soda Group differ in which measurement interval.

Analysis of Variance shows that there is no statistically significant correlation of the beverage consumed (soda, water), gender (male, female) and time (0, 5, 10, 20 min) in which the salivary pH is measured. The change in the salivary pH value over time is not statistically different for

Table 3. Results of Tukey's post hoc test for comparison of average salivary pH values in different measurement intervals within one group
Tabela 3. Rezultati Takijevog post hoc testa za poređenje prosečnih vrednosti pH pljuvačke u različitim intervalima merenja u okviru grupe

		Average Srednja vrednost		p-values P-vrednost
Water Voda	0 min. vs. 5 min.	7.01	7.15	0.1327
	5 min. vs. 10 min.	7.15	7.18	0.9955
	10 min. vs. 20 min.	7.18	6.99	0.0031
Soda Kola	0 min. vs. 5 min.	6.87	6.33	0.00032
	5 min. vs. 10 min.	6.33	6.33	1.000
	10 min. vs. 20 min.	6.33	6.62	0.00032



Graph 4. Linear chart for average pH values over time for soda and water, for men and women, with corresponding 95% confidence intervals
Grafik 4. Linijski dijagram za prosečne vrednosti pH tokom vremena za kolu i vodu, za muškarce i žene sa odgovarajućim 95% intervalima poverenja

men and women, if the beverage consumed is taken into account (Graph 4). For the interaction of gender, beverage and time, the effect size measure, the partial eta squared is η^2=0.019, which is a small effect size. Thus, the pH values for men and women change similarly over time, if the type of beverage is taken into account.

DISCUSSION

The pH value is the negative decimal logarithm of the hydrogen ion concentration in the solution. The pH values of solutions can be measured using several methods: electrochemical method, optical method (indicator colour and optical fibre pH sensor), indicator strips and pH meters [21]. A digital pH meter is an electronic device used to measure the pH value of a liquid and consists of an electrode and an electronic part of the device that measures and displays the pH value [21]. Due to its user-friendliness, availability and accuracy, it is used in recent scientific research where the salivary pH value is determined [12–15]. The use of pH indicator strips requires additional costs for test strips and is considered a convenient but less accurate method compared to a digital pH meter [21]. A digital pH meter with a glass electrode was chosen for measuring the salivary pH value in the presented research due to the accuracy, precision and simplicity of the method.

The pH of saliva can be influenced by individual factors, such as the salivary secretion rate and buffering capacity [4]. The influence of individual factors was not examined in the presented research. The salivary pH is also affected by external factors such as time of day when the experiment is performed, food and drink intake, the quantity of the beverage taken, its temperature, and taking medications in a 6-month period before the start of the experiment [4]. In order to minimise the influence of external factors, all subjects were instructed not to eat, drink or brush their teeth in the morning on the day of the experiment until it is completed, the experiment started at the same time of day and the same quantity and temperature of the experimental beverages were defined. By observing the experimental protocol and the study exclusion criteria, the influence of external factors that were not the subject of the study was minimised to the greatest extent possible.

The findings about salivary pH value after taking carbonated mineral water in the scientific literature are very few. The results of the present research can only be compared with the findings of Uma et al. from 2018 [4]. They recorded an increase, but no statistically significant difference was found in the pH value of saliva 5, 10 and 15 minutes after consuming mineral water, in contrast to the presented findings, where the pH value increased significantly after 5 and 10 minutes (Table 3). After 20 minutes, the salivary pH dropped to the level of the initial values measured before the drink had been taken, which matches the findings of Uma et al. [4]. The presented findings can be explained by a low buffering capacity of mineral water and the gustatory stimulation of salivary secretion. However, it must be taken into account that the pH value of the mineral water used in the research by Uma et al. had a pH of 7.02, while the pH of the mineral water in the present research was 6.32, thus slightly more acidic, which could have had an impact on differences in findings after 5 and 10 minutes.

In contrast to the findings of the salivary pH obtained after consuming carbonated mineral water, when taking sugar-sweetened carbonated soda with the average pH value of 2.62, the salivary pH decreased after 5 and 10 minutes, and after 20 minutes it increased, but predominantly failed to reach the initial pH value before the beverage had been taken. The lowest average pH value was determined 5 and 10 minutes after taking the sugar-sweetened soda and was pH= 6.33, which is 0.6 lower than the average initial pH value (Table 1). The findings of the authors who examined the change in the salivary pH after consuming fruit, chocolate and sweetened beverages also indicate a drop in the pH value in the time intervals examined [4, 12, 19]. In the findings of Almenara et al. from 2016, a drop in the salivary pH value was determined 5, 10 and 15 minutes after taking soda, but the research did not include a time interval of 20 minutes [19].

In vitro studies have proven the correlation between taking acidic beverages and the occurrence of dental erosions [13, 14]. The critical pH value for the appearance of enamel demineralisation is considered to be pH≤5.5, and pH≤6.5 for dentin [19]. Although in the present research the lowest mean pH value was 6.33, in some subjects the pH value below the critical was measured 5 and 10 minutes after taking soda. The minimum value 5 minutes after taking soda was pH 5.46, and after 10 minutes it was pH 5.38. The results of the present research indicate that in some subjects the salivary pH value dropped below the critical value and that taking sugar-sweetened soda can affect the occurrence of enamel demineralisation, while the mean values are indicative of the possibility of dentin demineralisation.

The question arises as to whether the lowered salivary pH value was due to the acidity of the soda as such (pH=2.62) or the decomposition of sucrose and the creation of acidic products by the microorganisms of the oral cavity had already occurred, to which it is not possible to answer based on the present research. In any case, the pH value decrease to 6.33 on average with minimum values of 5.38 and 5.46 in some subjects is a very unfavourable environment for the preservation of oral health and increases the risk of the occurrence of hard dental tissue demineralisation, the occurrence of dental caries, soft tissue diseases and the appearance of periodontitis. The presented research results indicate that the consumption of a sugar-sweetened carbonated soda can lead to a drop in the salivary pH value below the critical value for the occurrence of enamel and dentin demineralisation, which persists even 10 minutes after its consumption.

CONCLUSION

The type of beverage taken affects the salivary pH value 5, 10 and 20 minutes after consumption. After taking carbonated mineral water, the pH value of saliva rises, and after 20 minutes it drops to a value close to the value before measuring. After taking sugar-sweetened carbonated soda, there is a drop in salivary pH value. It reaches the lowest mean value 5 and 10 minutes after taking the beverage, and then increases slightly without reaching the initial value. In some subjects, a minimum pH value below the value considered critical for the occurrence of enamel demineralisation was measured, while the mean values of the pH drop indicate the possibility of dentin demineralisation. Consuming sugar-sweetened carbonated soda can have adverse effects on oral health.

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Uticaj zaslađenog napitka kola i mineralne gazirane vode na pH vrednost pljuvačke

Nataša Puškar[1], Milica Puškar[1], Milica Jeremić Knežević[1], Daniela Đurović Koprivica[1], Ljiljana Andrijević[2]

[1]Univerzitet u Novom Sadu, Medicinski fakultet, Klinika za stomatologiju, Novi Sad, Srbija;
[2]Univerzitet u Novom Sadu, Medicinski fakultet, Klinika za biohemiju, Novi Sad, Srbija

KRATAK SADRŽAJ

Uvod Pljuvačka ima značajnu ulogu u održavanju oralnog zdravlja. Njen puferski kapacitet pomaže u neutralizaciji kiselih produkata iz hrane, pića i dentalnog plaka. Sastav pljuvačke, kvantitativne i kvalitativne osobine, kao što je aciditet, povezani su sa pojavom karijesa zuba, nekarijesnih lezija i parodontopatije.

Cilj rada je bio da se ispita promena aciditeta pljuvačke posle uzimanja mineralne gazirane vode i zaslađenog gaziranog napitka kola kod ispitanika starosti 18 do 25 godina.

Materijal i metod Istraživanje je sprovedeno na Klinici za stomatologiju Vojvodine na 30 odraslih ispitanika. Aciditet pljuvačke je meren digitalnim pH metrom pre, 5, 10 i 20 minuta posle uzimanja 200 ml gazirane mineralne vode i 200 ml zaslađenog gaziranog napitka kola. Dobijeni rezultati su statistički obrađeni mešovitom, kombinovanom analizom varijanse.

Rezultati Konzumiranje mineralne gazirane vode i zaslađenog napitka kola dovelo je do promene aciditeta pljučke u ispitivanim vremenskim intervalima. Mineralna gazirana voda smanjuje aciditet pljuvačke, da bi se posle 20 minuta vratila na vrednost blisku onoj pre početka merenja. Nakon uzimanja gaziranog zaslađenog napitka kola vrednost pH pljuvačke opada. Najnižu srednju vrednost dostiže 5 i 10 minuta posle uzimanja napitka, a zatim blago raste ne dostižući početnu vrednost. Kod pojedinih ispitanika izmerena je minimalna vrednost pH pljuvačke ispod vrednosti koja se smatra kritičnom za nastanak demineralizacije gleđi i dentina.

Zaključak Konzumiranje gazirane mineralne vode i zaslađenog napitka kola utiče na promenu vrednosti pH pljuvačke u vremenskom periodu od 20 minuta posle konzumiranja. Konzumiranje zaslađenog gaziranog napitka kola može imati štetne posledice po oralno zdravlje.

Ključne reči: pljuvačka; pH; napitak

UVOD

Usna duplja, kao početak digestivnog trakta, u neposrednom je kontaktu sa spoljašnjom sredinom. Za oralnu biohemiju pljuvačka, koju luče egzokrine pljuvačne žlezde, ima veliki značaj. Ona je vodeni rastvor koga sekretuju velike pljuvačne žlezde (parotidne, submandibularne i sublingvalne), male pljuvačne žlezde koje se nalaze u sluzokoži usana, obraza, jezika, nepca i ždrela i gingivalnih cervikalnih fluida [1]. Dnevno se u proseku izluči 700–800 ml pljuvačke, koja se u fiziološkim uslovima proguta i apsorbuje u digestivnom traktu [1]. U sastvav pljuvačke ulazi voda, koja čini 99,5%, a 0,5% čine soli i proteini [2]. Od proteina najznačajni su bazni glikoproteini, koji štite gleđ zuba i odgovorni su za viskozitet pljuvačke i lubrikaciju [2]. U pljuvački se nalaze još i imunoglobulini i lizozomi, koji liziraju bakterije, enzimi alfa-amilaza i hijaluronidaza [2]. U jednom delu populacije u pljuvački se nalaze i antigeni krvnih grupa A i/ili B [2].

Pljuvačka ima značajnu ulogu u održavanju oralnog zdravlja. Ona štiti i vlaži oralnu sluzokožu, igra važnu ulogu u procesima mineralizacije i demineralizacije i gleđi i dentina [3]. Svojim imunološkim i neimunološkim komponentama ispoljava antimikrobnu aktivnost, pojačava ukus hrane i pića i započinje proces varenja hrane [3]. Njen puferski kapacitet pomaže u neutralizaciji kiselih produkata iz hrane, pića i dentalnog plaka [2, 3]. Poremećaj homeostaze, koja se održava pomoću pljuvačke, dovodi do lošeg oralnog zdravlja [4].

Aciditet, tj. pH pljuvačke, zavisi od bikarbonata, fosfata i proteina. Kalcijum i fosfat su prezasićeni pri fiziološkom („normalnom“) pH, a pufersko dejstvo pljuvačke zavisi uglavnom od bikarbonata i fosfata [3]. Pri dovoljno brzom lučenju pljuvačke (više od 1 ml po minutu) koncentracija bikarbonata je 30–60 mmol/l, a pH je 7,5–7,8 [3].

Pljuvačka sprečava proces oštećenja sluzokože usta, zuba, kao i nastanak dentalnih erozija spirajući patogene mikroorganizme, razgrađujući hranu i uništavajući same bakterije, uključujući i one kariogene [4]. Nakon uzimanja hrane i pića dolazi do pada vrednosti pH pljuvačke koji može imati štetne posledice, naročito na nastanak karijesa zuba i dentalnih erozija [5]. Smatra se da bilo koji faktor koji može da utiče na promenu oralne mikroflore i lokalnih uslova sredine, kao što je promena pH pljuvačke, dovodi do disbalansa u biofilmu i porasta broja kariogenih bakterija, što dovodi do nastanka dentalnog karijesa [5]. Time puferski sistem pljuvačke igra ključnu ulogu u prevenciji i dentalnog karijesa i dentalnih erozija, koje predstavljaju dentalne lezije na površini zuba uzrokovane mnoštvom faktora, ali bez učešća bakterija [5].

Brojni autori su ispitivali uticaj uzimanja različitih napitaka na pH pljuvačke. Istraživanja su mahom rađena *in vitro*, gde je ispitivan puferski kapacitet pljuvačke ili sposobnost kiselih napitaka da oštete gleđ i dentin zuba [6–12]. *In vivo* istraživanja su vršena na ispitanicima dečjeg uzrasta i na mladima 18–25 godina starosti, merenjem vrednosti pH pljuvačke u različitim intervalima posle uzimanja voćnih sokova, čajeva, vina ili konzumiranja određene vrste poslastica ili hrane [1, 4–5, 13–20]. *In vivo* nalazi o vrednostima pH pljuvačke kod čoveka posle uzimanja gazirane mineralne vode i zaslađenog napitka kola u različitim vremenskim intervalima u naučnoj literaturi su veoma oskudni.

Cilj rada je bio da se ispita uticaj zaslađenog gaziranog napitka kola i mineralne gazirane vode na vrednost pH pljuvačke 5, 10 i 20 minuta nakon konzumiranja kod ispitanika starosti 18 do 25 godina života.

MATERIJAL I METOD RADA

Istraživanje je sprovedeno u Klinici za stomatologiju Vojvodine i odobreno je od strane Etičkog odbora Klinike. Uzorak je činilo 30 ispitanika, studenata stomatologije Medicinskog fakulteta u Novom Sadu starosti 18 do 25 godina oba pola. Ispitanici su pre uključivanja u studiju bili informisani o načinu i ciljevima istraživanja i potpisali su svoj dobrovoljni pristanak. Odabir je vršen metodom slučajnog izbora. Prvih 30 studenata koji su se javili u ponuđene termine a zadovoljavali su kriterijume za uključivanje u studiju bili su izabrani kao ispitanici.

Kriterijumi za isključivanje iz studije su bili:

– ispitanici kojima je u toku ortodontski tretman i izrada zubnih nadoknada;
– trudnice;
– ispitanici koji boluju od sistemskih bolesti;
– ispitanici koji imaju oboljenja pljuvačnih žlezda;
– ispitanici koji uzimaju lekove koji mogu imati uticaja na funkciju pljuvačnih žlezda (antihistaminici, antikancerski lekovi...) u periodu od najmanje dva meseca pre početka eksperimenta i oni koji su u ovom periodu uzimali antibiotike.

Uzimanje uzoraka pljuvačke vršeno je u 9 sati ujutru, a ispitanicima je naloženo da tog jutra ne peru zube i ne uzimaju hranu ni piće do početka eksperimenta. Pre početka eksperimenta određena je vrednost pH ispitivanih napitaka i vrednost pH pljuvačke svih ispitanika. Sakupljanje pljuvačke vršeno je ispljuvavanjem u sterilnu epruvetu kroz sterilni levak. Merenje vrednosti pH je vršeno digitalnim pH metrom (InoLab, Xylem analytics, Germany) u laboratoriji za biohemiju Medicinskog fakulteta u Novom Sadu, koja se nalazi na Klinici za stomatologiju Vojvodine. Kalibracija pH metra je vršena pre početka merenja. Staklena elektroda je uranjana u test epruvetu, a nakon svakog merenja pažljivo je očišćena destilovanom vodom i osušena filter-papirom. Vrednost pH pljuvačke određivana je sa tri uzastopna uranjanja staklene elektrode pH metra u epruvetu sa uzorkom, a srednja vrednost se uzimala kao dobijena vrednost. Prilikom uranjanja staklene elektrode merenje je vršeno nakon 10 sekundi, da bi se stabilizovala uzorkovana tečnost.

Ispitivanje je vršeno u dva uzastopna dana, prvog za gaziranu mineralnu vodu, a drugog za gazirani zaslađeni napitak kola. Posle početnog određivanja pH pljuvačke svim ispitanicima je prvog dana dato 200 ml mineralne gazirane vode (flaširana gazirana prirodna mineralna voda Vrnjci, Voda Vrnjci a.d., Vrnjačka Banja, Republika Srbija), a drugog dana 200 ml zaslađenog gaziranog napitka kola (Coca Cola®, HBC-Srbija d.o.o. Beograd) na sobnoj temperaturi. Naloženo im je da napitak popiju u roku od jednog minuta. Nakon toga ponovo su uzimani uzorci pljuvačke i merena je njena vrednost pH.

Pljuvačka je sakupljana 5, 10 i 20 minuta od početka konzumiranja napitka, a merenje je izvršeno u roku od 5 minuta od uzimanja uzorka.

Dobijeni rezultati su statistički obrađeni. Korišćena je mešovita, kombinovana, analiza varijanse: „mixed design ANOVA" (engl. prim. autora).

REZULTATI

U ispitivanje je bilo uključeno 30 ispitanika. Ženski ispitanici su činili 70% uzorka, dok su muški ispitanici činili 30% uzorka.

Ukupno je načinjeno 720 merenja pH pljuvačke i 18 merenja pH napitaka.

Prosečna vrednost pH gazirane mineralne vode iznosila je 6,32, dok je pH zaslađenog napitka kola bila 2,62.

Na vrednosti pH pljuvačke uticale su dve nezavisne promenljive: vreme – momenti u kojima su izvršena merenja pH pljuvačke (0, 5, 10, 20 minuta posle uzimanja napitka) i vrsta tečnosti koja je uzimana – mineralna gazirana voda (grupa voda) ili zaslađeni gazirani napitak kola (grupa kola).

Vrednosti pH pljuvačke za merenja pre i posle uzimanja gazirane mineralne vode i zaslađenog gaziranog napitka kola prikazani su u Tabeli 1. Uočavaju se različite prosečne vrednosti pH za dve grupe, kao i za različite momente merenja.

Vrednosti pH za dve grupe (grupa voda i grupa kola) za različite momente merenja predstavljene su boks dijagramom na Grafikonu 1, a prosečne vrednosti ove promenljive linijskim dijagramom na Grafikonu 3.

Analizom varijanse utvrđeno je da postoji statistički značajan glavni efekat grupe, odnosno da postoji statistički značajna razlika između vrednosti pH pljuvačke u grupi koja je konzumirala gaziranu mineralnu vodu i grupe koja je konzumirala zaslađeni gazirani napitak kola, $F(1,58) = 60,02$, $p = 0,00$, bez obzira na vremenske intervale merenja. Za glavni efekat grupe, mera veličine efekta, parcijalni eta kvadrat η^2 iznosi 0,51, što predstavlja veoma značajnu razliku između ispitivanih napitaka.

Promena vrednosti pH tokom vremena statistički se značajno razlikuje i kod ispitanika koji su uzimali vodu i kod onih koji su uzimali kolu (Grafikon 3). Za interakciju grupe i vremena, mera veličine efekta, parcijalni eta kvadrat je $\eta^2 = 0,443$, što predstavlja srednju veličinu efekta.

Ako se gledaju razlike između grupa, *post hoc* analizom, Takijevim testom, može se utvrditi koje prosečne vrednosti se statistički značajno razlikuju. Dobija se da su u intervalima 5 min., 10 min. i 20 min. prosečne vrednosti pH pljuvačke u grupi voda statistički značajno veće od odgovarajućih prosečnih vrednosti pH pljuvačke u grupi kola. Prilikom prvog merenja vrednosti pH pljuvačke, pre uzimanja napitaka, prosečne vrednosti se nisu razlikovale. Rezultati su prikazani u Tabeli 2.

U Tabeli 3 prikazano je koje se prosečne vrednosti u grupi voda i grupi kola razlikuju u kom intervalu merenja.

Analiza varijanse pokazuje da ne postoji statistički značajna interakcija tečnosti koja se pije (kola, voda), pola (muško, žensko) i vremena (0, 5, 10, 20 min.) u kojima se meri pH pljuvačke. Promena vrednosti pH pljuvačke tokom vremena statistički se ne razlikuje za muškarce i žene, ako se uzima u obzir tečnost koja se pije (Grafikon 4). Za interakciju pola, tečnosti i vremena, mera veličine efekta, parcijalni eta kvadrat, jeste $\eta^2 = 0,019$, što predstavlja malu veličinu efekta. Dakle, vrednosti pH se i za muškarce i žene slično menjaju tokom vremena, ako se uzima u obzir vrsta napitka.

DISKUSIJA

Vrednost pH predstavlja negativan dekadni logaritam koncentracije jona vodonika u rastvoru. Vrednosti pH rastvora mogu se meriti na više načina: elektrohemijskom metodom, optičkom metodom (indikatorske boje i pH sonde od optičkih vlakana), indikatorskim trakama i pomoću pH metara [21]. Digitalni pH metar je elektronski uređaj koji se koristi za merenje vrednosti

pH tečnosti i sastoji se od elektrode i elektronskog dela uređaja koji meri i prikazuje vrednost pH [21]. Zbog svoje jednostavnosti pri radu, dostupnosti i tačnosti koristi se u savremenim naučnim istraživanjima u kojima se određuje vrednost pH pljuvačke [12–15]. Upotreba pH indikatorskih traka iziskuje dodatne troškove za merne trake i smatra se pogodnom ali manje tačnom metodom u odnosu na digitalni pH metar [21]. Za merenje vrednosti pH pljuvačke u prezentovanom istraživanju izabran je digitalni pH metar sa staklenom elektrodom zbog tačnosti, preciznosti i jednostavnosti metode.

Na pH pljuvačke mogu uticati individualni faktori, kao što su brzina lučenja pljuvačke i njen puferski kapacitet [4]. Uticaj individualnih faktora nije bio predmet ispitivanja u prezentovanom istraživanju. Na pH pljuvačke utiču i spoljašnji faktori, kao što su uzimanje hrane i pića, doba dana u kom se vrši eksperiment, količina uzetog napitka, njegova temperatura i uzimanje lekova u periodu od šest meseci pre započinjanja eksperimenta [4]. Da bi se isključio uticaj spoljašnjih faktora, svim ispitanicima je naloženo da ne uzimaju hranu, piće, niti da peru zube ujutru na dan izvođenja eksperimenta do njegovog završetka, eksperiment je započinjao u isto vreme i definisana je jednaka količina i temperatura eksperimentalnih napitaka. Poštovanjem eksperimentalnog protokola i kriterijuma za isključivanje iz studije u najvećoj mogućoj meri je isključen uticaj spoljašnjih faktora koji nisu bili predmet istraživanja.

Nalazi o stanju vrednosti pH pljuvačke posle uzimanja gazirane mineralne vode u naučnoj literaturi su veoma oskudni. Rezultati prezentovanog istraživanja mogu se uporediti samo sa nalazom Uma i saradnika iz 2018 [4]. Zabeležili su porast, ali nije pronađena statistički značajna razlika u vrednosti pH pljuvačke 5, 10 i 15 minuta posle konzumiranja mineralne vode, za razliku od prezentovanih nalaza gde je vrednost pH značajno porasla posle 5 i 10 minuta (Tabela 3). Nakon 20 minuta vrednost pH pljuvačke je opala na nivo početne vrednosti pre uzimanja napitka, što se slaže sa nalazom Uma i saradnika [4]. Prezentovani nalazi se mogu objasniti niskim puferskim kapacitetom mineralne vode i gustativnim stimulisanjem lučenja pljuvačke. Mora se uzeti u obzir i činjenica da je vrednost pH mineralne vode koja je korišćena u istraživanju Uma i saradnika bila 7,02, dok je pH mineralne vode u prezentovanom istraživanju bila 6,32, dakle nešto kiselija, što je moglo imati uticaja na razliku u nalazima posle 5 i 10 minuta.

Za razliku od nalaza pH pljuvačke dobijenog posle uzimanja mineralne gazirane vode, pri uzimanju zaslađenog napitka kola, čija je prosečna vrednost pH bila 2,62, pH pljuvačke opada posle 5 i 10 minuta, da bi se posle 20 minuta povećala, ali u najvećem broju slučajeva ne dostiže početnu vrednost pH od pre uzimanja napitka. Najniža prosečna vrednost pH utvrđena je 5 i 10 minuta posle uzimanja zaslađenog kola napitka i iznosila je 6,33, što je za 0,6 manje od srednje početne vrednosti pH (Tabela 1). Nalazi autora koji su ispitivali promenu pH pljuvačke posle uzimanja voćnih, čokoladnih i zaslađenih napitaka takođe ukazuju na pad pH u ispitivanim vremenskim intervalima [4, 12, 19]. U nalazima Almenare i saradnika iz 2016. utvrđen je pad vrednosti pH pljuvačke 5, 10 i 15 minuta posle uzimanja napitka kola, ali istraživanje nije obuhvatilo vremenski interval od 20 minuta [19].

In vitro studije su dokazale vezu između uzimanja kiselih napitaka i pojave dentalnih erozija [13, 14]. Kao kritična vrednost za pojavu demineralizacije gleđi navodi se vrednost od pH ≤ 5,5, dok je za dentin pH ≤ 6,5 [19]. Iako je u prezentovanom istraživanju najniža srednja vrednost pH bila 6,33, kod pojedinih ispitanika izmerena je vrednost pH ispod kritične vrednosti 5 i 10 minuta posle uzimanja napitka kola. Minimalna vrednost 5 minuta posle uzimanja kola napitka je bila pH 5,46, a posle 10 minuta pH 5,38. Rezultati prezentovanog istraživanja ukazuju na to da se kod pojedinih ispitanika vrednost pH pljuvačke spustila ispod kritične vrednosti i da uzimanje zaslađenog napitka kola može uticati na pojavu demineralizacije gleđi, dok srednje vrednosti čak ukazuju na mogućnost demineralizacije dentina.

Postavlja se pitanje da li snižena vrednost pH pljuvačke potiče od kiselosti samog napitka kola (pH = 2,62) ili je već došlo do razgradnje saharoze i stvaranja kiselih produkata od strane mikroorganizama usne duplje, na koje na osnovu prezentovanog istraživanja nije moguće dati odgovor. U svakom slučaju sniženje pH na 6,33 u proseku sa minimalnim vrednostima od 5,38 i 5,46 kod pojedinih ispitanika predstavlja vrlo nepovoljnu sredinu za očuvanje oralnog zdravlja i povećava rizik za pojavu demineralizacije tvrdih zubnih tkiva, pojavu karijesa zuba, za oboljenja mekih tkiva i pojavu parodontopatije. Rezultati prezentovanog istraživanja ukazuju na to da konzumiranje zaslađenog gaziranog napitka kola može dovesti do pada vrednosti pH pljuvačke ispod kritične vrednosti za pojavu demineralizacije gleđi i dentina, koja perzistira i 10 minuta posle njegovog konzumiranja.

ZAKLJUČAK

Vrsta uzetog napitka utiče na vrednost pH pljuvačke 5, 10 i 20 minuta posle konzumiranja. Nakon uzimanja gazirane mineralne vode vrednost pH pljuvačke raste, da bi posle 20 minuta opala na vrednost blisku vrednosti pre početka merenja. Posle uzimanja gaziranog zaslađenog napitka kola vrednost pH pljuvačke opada. Najnižu srednju vrednost dostiže 5 i 10 minuta posle uzimanja napitka, a zatim blago raste ne dostižući početnu vrednost. Kod pojedinih ispitanika izmerena je minimalna vrednost pH ispod vrednosti koja se smatra kritičnom za nastanak demineralizacije gleđi, dok srednje vrednosti pada pH ukazuju na mogućnost pojave demineralizacije dentina. Konzumiranje zaslađenog gaziranog kola napitka može imati štetne posledice po oralno zdravlje.