Utjecaj H^+ i Mg^{2+} iona na promjene koncentracije elektrolita u slini mjerene ionsko-selektivnim elektrodama

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Sažetak

Promjene koncentracije elektrolita i pH u slini imaju veliku ulogu u demineralizaciji zubne cakline. Slina je veoma važna u održavanju neutralnoga pH i koncentracije elektrolita potrebne za remineralizaciju cakline. U ovome radu ispitane su promjene koncentracije K^+, Na^+, Cl^- i Ca^{++} nastale padom pH vrijednosti sline te promjene koje su nastale nakon dodatka magnezijevih iona u slinu uz stalnu pH vrijednost. Mjerenja su izvedena potenciometrijski uporabom ionsko-selektivnih elektroda. Rezultati ispitivanja pokazali su da se neutralizacija sline nakon pada pH vrijednosti događa u razdoblju od pet minuta i to samim lučenjem sline. Koncentracija ioniziranoga kalcija u slini povećava se nakon dodatka magnezijevih iona pri pH vrijednosti oko 7.

Ključne riječi: *kalcij, karijes, elektroliti u slini, magnezij.*

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Uvod

Premda se je karijes u djece i adolescenata u nekim razvijenim zemljama smanjio, ta najraširenija bolest zuba još je uvijek uvjetovana lošim prehrambenim i higijenskim navikama. Prema rezultatima ispitivanja provedenih u SAD raširenost karijesa među odraslim osobama obaju spolova jednaka je kao i u djece (1). Zbog svoje raširenosti karijes je predmet brojnih ispitivanja poduzetih s ciljem da se otkrije postupak njegove prevencije. Ta su istraživanja potvrdila povezanost karijesa s mineralnim sastavom sline (2,3). No, unatoč točno poznatim kemijskim procesima koji se događaju tijekom procesa demineralizacije i remineralizacije zuba, još uvijek nema posve djelotvorna odgovora na problem prevencije karijesa. Razlike u jakosti lučenja sline i razgradnji ugljikohidrata iz hrane, te kiseline iz plaka, mogu objasniti velike razlike u osjetljivosti prema karijesu između raznih osoba, ali i između raznih mjesta u usnoj šupljini (4,5). Provedena ispitivanja pokazala su da uporaba fluorida može znatno smanjiti nastanak te bolesti (6,7).

Slina je vrlo važna u održavanju neutralnog pH plaka i usne šupljine. U slučaju naglih promjena pH ili unosa drugih elektrolita hranom, nastaju promjene koje mogu imati štetne posljedice za zubnu caklinu. Slina treba kompenzirati te promjene, a puferski kapacitet sline ovisi o sadržaju bikarbonata, fosfata i drugih iona (8). Koncentracija kalcijeva fosfata u plaku i u slini važna je za sprječavanje karijesa. Ispitivanja provedena dodavanjem kalcija i fosfata u žvakaće gume pokazala su povoljan učinak na smanjenje karijesa (9,10). Primjena kalcijeva fosfata povećala je pH i koncentraciju ukupnog i slobodnog kalcija. Rezultati tih radova pokazuju da se kariozne promjene zuba mogu usporiti akumulacijom kalcija iz gume za žvakanje u plaku i u slini (10).

Svrha ovoga rada bila je ispitati kinetiku promjena koncentracije K^+, Na^+, Cl^- i Ca^{2+} zbog smanjenja pH sline kod ispitanika koji su žvakali žvakaću gumu s dodatkom jabučne i vinske kiseline. U radu je ispitan i utjecaj magnezijevih iona *in vivo* na promjene koncentracije ioniziranoga kalcija u slini, kao potencijalnog iona za sprječavanje demineralizacije zuba. Mjerenje koncentracije ioniziranoga kalcija provedeno je u ispitanika koji su uzeli žvakaću gumu s dodatkom magnezija. U prikazanom ispitivanju rabljene su potenciometrijske metode, jer one pružaju izravnu informaciju o koncentraciji slobodnih iona.

Materijali i metode

Ispitanici

Ispitivanje je provedeno u dvjema različitim skupinama ispitanika. Prva je skupina bila slučajan uzorak od 5 osoba, bez prethodne stomatološke obrade, u dobi od 17 do 45 godina. Druga skupina bila su djeca (N=30) iz dječjeg doma u dobi od 7 do 12 godina, s približno jednakim načinom prehrane. Ta je skupina na temelju stomatološke obrade podijeljena u tri skupine: **skupina A** s intaktnim zubima (N=10), **skupina B** s karioznim zubima (N=10) i **skupina C** sa saniranim zubima (N=10). Promjena koncentracije elektrolita kao posljedica promjena pH u slini proučavana je na prvoj skupini ispitanika, dok je utjecaj magnezija na promjenu koncentracije kalcija ispitivan na drugoj grupi ispitanika. Kontrolnu skupinu za obje studije činili su ispitanici koji su žvakali žvakaću gumu bez šećera i bez drugih dodataka (označena kao Mg - skupina u Tablici 2). Sva ispitivanja provedena su poštujući osnovna načela Helsinške deklaracije (World Health Organization 1975).

Reagensi

Kao sredstvo za sniženje pH vrijednosti sline poslužila je komercijalna žvakaća guma koja je sadržavala jabučnu i vinsku kiselinu. Za ispitivanje utjecaja magnezija uporabljena je žvakača guma bez šećera i s dodatkom magnezija (16 mmol Mg^{2+}/1g žvakaće gume). Magnezij je dodan u obliku magnezijeva klorida analitičke čistoće (Kemika, Zagreb). Ukupna količina magnezija u žvakaćoj gumi iznosila je 48 mmol. Ta je koncentracija magnezija odabrana na temelju rezultata prethodnih mjerenja ukupnoga kalcija u slini (8).

Metoda rada

Uzorci sline uzimani su s dna usne šupljine (7, 11). Prije prvog uzimanja uzorka sline svi su ispitanici isprali usta vodom. Slina je uzimana aspiracijom u polipropilenske šprice. Šprice su potom zatvorene gumenim čepom i pohranjene na hladnome mjestu.

Za ispitivanje utjecaja promjene pH na koncentracije elektrolita u slini ispitanicima je najprije uzet kontrolni uzorak sline, a nakon što su primili žvakaću gumu, uzorak sline uziman je svake minute u razdoblju od pet minuta. Cijelo vrijeme ispitanici su žvakali istu žvakaću gumu.

Za ispitivanje utjecaja magnezija na koncentraciju kalcija u slini svaka od triju skupina ispitanika podijeljena je u dvije podskupine. Jedna podskupina dobila je žvakaću gumu s magnezijem, a druga je bila kontrolna podskupina i primila je *placebo* (žvakaću gumu bez magnezija). Ispitanicima je uzet uzorak sline, a zatim su zamoljeni da žvakaću gumu žvaču 10 minuta. Nakon toga ponovo je uzet uzorak sline.

Mjerenje

Određivanje koncentracije elektrolita izvedeno je na uređaju s protočnim ionsko-selektivnim elektrodama vlastite izvedbe (12). Mjerena je koncentracija ioniziranoga kalcija, natrija, kalija i klorida u slini, te njezina pH vrijednost. Baždarenje instrumenta provedeno je standardnim otopinama Ciba Corning (kat.br. 473385 i kat.br. 483386, Ciba Corning Diagnostic, USA). Kontrola instrumenta komercijalnim kontrolnim otopinama Ciba Corning u sve tri razine koncentracije, provedena prije mjerenja, dala je zadovoljavajuće rezultate.

Određivanje koncentracije ukupnoga kalcija izvedeno je na plamenom fotometru EFOX 5053 (Eppendorf, Njemačka).

Statističke metode

Za statističku obradu rezultata uporabljen je program Microsoft Excel®. Rezultati mjerenja prikazani su kao medijani, a prikazan je i raspon vrijednosti mjerenja. Razina znatnosti ispitivana je Studentovim t-testom uz p = 0,05.

Rezultati

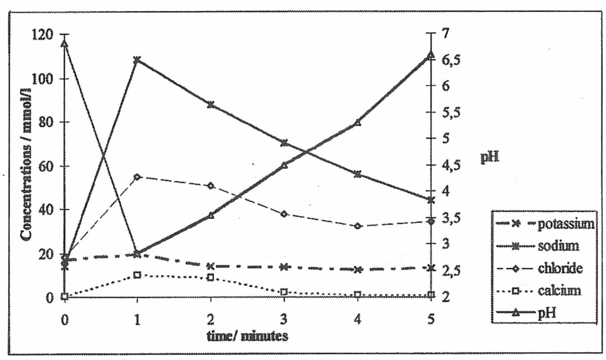
Ispitivanje utjecaja pH

Rezultati ispitivanja utjecaja pH prikazani su u Tablici 1. Zbog velikoga raspona izmjerenih vrijednosti, kao mjera srednje vrijednosti odabran je medijan, a ne aritmetička sredina. Na Slici 1 ti su podaci prikazani grafički. Početne vrijednosti pH i kon-

Tablica 1. *Rezultati mjerenja koncentracije elektrolita tijekom žvakanja gume sa sadržajem jabučne kiseline (N = 5). Podatci za kalij, natrij, kloride i kalcij su u mmol/L. Prikazani su medijan i raspon izmjerenih vrijednosti*

Table 1. *Results of electrolyte concentration determination during mastication of chewing gum containing malic acid (N = 5). Data for potassium, sodium, chlorides and calcium are expressed in mmol/L. The median and range of the measured values are presented*

Vrijeme/Time (min)	pH	K^{+}	Na^{+}	Cl^{-}	Ca^{2+}
0	6.83 6.79 - 6.95	16.9 15.2 - 18.2	14 9.9 - 17.7	17.9 13.6 - 18.8	0.60 0.5 0- 0.73
1	2.83 2.57 - 3.15	19.3 16.6 - 19.6	108.3 77.3 - 121.7	54.6 47.0 - 61.8	9.95 0.48 - 12.45
2	3.55 2.69 - 5.83	13.9 13.3 - 19.2	87.5 69.9 - 102.2	50.6 38.5 - 69.3	8.87 0.38 - 12.52
3	4.50 3.36 - 6.96	13.5 10.2 - 15.6	69.7 46.2 - 80.6	37.5 37.0 - 62.1	2.30 0.56 - 8.27
4	5.29 3.40 - 7.50	12.2 9.2 - 15.4	55.4 34.7 - 61.0	32.0 31.7 - 50.0	0.96 0.71 - 4.99
5	6.60 3.58 - 7.63	13.1 9.2 - 14.4	43.9 32.0 - 51.6	34.0 24.0 - 43.8	1.03 0.82 - 4.49



Slika 1. *Promjene koncentracije elektrolita tijekom žvakanja žvakaće gume s dodatkom jabučne kiseline (podatci iz Tablice 1).*

Figure 1. *Changes in electrolyte concentration during the mastication of chewing gum containing malic acid (data from Table 1).*

centracije ioniziranoga kalcija, kalija, natrija i korida u slini predstavljaju vrijednosti navedene u prvome redu u Tablici 1, a odgovaraju vrijednostima izmjerenim u trenutku t=0. Početna vrijednost pH u slini kreće se u području od 6,79 do 6,95; natrija između 9,9 i 17,7 mM; kalija između 15,2 i 18,2 mM; klorida između 13,6 i 18,8 mM, a ioniziranoga kalcija u području od 0,5 do 0,73 mM.

Prikazana je dinamika promjene koncentracije elektrolita u slini zbog promjene pH. Iz slike se mogu vidjeti velike promjene u koncentraciji natrija, klorida i kalcija, te male promjene koncentracije kalija. Vidljivo je da se pH vrijednost i koncentracija kalcija vraćaju na početne vrijednosti nakon 5 minuta.

Utjecaj magnezijevih iona

Promjene koncentracije elektrolita nastale žvakanjem žvakaće gume s magnezijem prikazane su u Tablici 2. Vrijednosti koncentracije slobodnoga kalcija bile su veće u skupinama A i B, a medijan za kalcij u skupini C bio je veći u kontrolnoj podskupini koja nije primila magnezij, ali te razlike nisu statistički znatne ($p<0,05$). Razlike pH vrijednosti između spomenutih skupna također nisu bile statistički znatne ($p<0,05$), premda je skupina s karioznim zubima (skupina B) imala niži pH od preostalih dviju skupina. Koncentracija klorida viša je u podskupinama koje su primile magnezij (osim u skupini C), što je posljedica egzogenog unosa klorida (s pomoću magnezijeva klorida), ali ni tu razlika nije statistički znatna ($p<0,05$). Promjene koncentracije kalija i natrija također se nisu znatno statistički razlikovale ($p<0,05$), jednako kao ni koncentracija ukupnoga kalcija.

Diskusija

Demineralizacija zubne cakline uzrokuje nastanak karijesa. Zubna caklina građena je od kalcijeva hidroksiapatita. Topljivost toga spoja izrazito ovisi o pH vrijednosti medija: u području niskoga pH za održavanje zasićenosti potrebna je veća koncentracija ioniziranoga kalcija nego kod višega pH (8). Slina kao tekućina usne šupljine ima zadaću ispirati i čistiti plak od ostataka hrane (ugljikohidrata), dok

Tablica 2. *Promjene koncentracije elektrolita u slini u nazočnosti iona magnezija (c (Mg^{2+}) = 16 mmol/g žvakaće gume). Vrijednosti su prikazane kao medijan i raspon (oznake u tablici su Mg^+ = žvakaća guma s magnezijem; Mg^- = žvakaća guma bez magnezija). iCa^{2+} = ionizirani kalcij, tCa = ukupni kalcij (vezan u kompleks + ioniziran)*

Koncentracija elektrolita izražena je u mmol/L, oznake skupina jesu: A = skupina s intaktnim zubima; B = skupina s karijesom; C = skupina sa saniranim zubima. Broj ispitanika u svakoj skupini N = 5

Table 2. *Changes in salivary electrolyte concentrations in the presence of magnesium ions (c (Mg^{2+}) = 16 mmol/g of chewing gum). The values are presented as median and ranges (designations in the table are: Mg^+ = magnesium-containing chewing gum, Mg^- = chewing gum without magnesium). iCa^{2+} = ionized calcium, tCa = total calcium (bounded + ionized)*

Electrolyte concentrations are expressed in mmol/L, group designations are: Group A with intact teeth; Group B with carious teeth; Group C with treated teeth. Number of subjects in each group: N = 5

			pH	K^+	Cl^-	Na^+	iCa^{2+}	tCa
A	Mg^+	median	7.00	19.7	24.0	7.3	0.77	1.60
		range	6.61-7.27	18.6-31.7	10.5-38.2	5.6-26.2	0.41-0.98	1.48-1.68
	Mg^-	median	7.10	22.6	19.7	11.9	0.57	1.76
		range	6.84-7.27	17.6-25.1	18.8-30.0	5.1-23.5	0.57-0.78	1.43-1.92
B	Mg^+	median	6.78	22.6	24.4	14.1	0.8	1.56
		range	6.67-7.14	19.7-22.7	19.4-29.9	8.8-23.4	0.56-0.91	1.43-1.75
	Mg^-	median	7.30	22.7	16.3	5.7	0.72	1.54
		range	6.74-7.43	17.8-22.9	16.3-20.2	3.8-10.1	0.55-0.80	1.43-1.59
C	Mg^+	median	6.83	19.0	20.9	6.4	0.61	1.49
		range	6.70-7.56	16.1-22.3	17.5-27.4	4.8-11.3	0.54-0.97	1.31-1.76
	Mg^-	median	7.34	18.3	21.6	11.4	0.71	1.40
		range	7.15-7.57	14.5-20.1	14.2-22.5	3.2-21.0	0.51-0.73	1.08-1.75

je njezin puferski učinak manje izražen. Ispitivanja drugih autora pokazala su da su reakcije plaka na pad pH vrijednosti sline u osoba otpornih na karijes jednake reakcijama plaka u osoba osjetljivih na karijes. No, vrijednosti pH kod kojih se te reakcije događaju bitno su različite: u osoba otpornih na karijes početna vrijednost pH je viša, a pad pH vrijednosti manji je nego u osoba osjetljivih na karijes. Istraživanja su pokazala da je puferski kapacitet plaka veći u osoba otpornih na karijes u usporedbi s osobama osjetljivima na karijes (7,13). U ovom je radu provedeno istraživanje s ciljem da se utvrde promjene u koncentraciji elektrolita u slini, posebice kalcija, nastalih naglim padom pH vrijednosti sline.

Prigodom pada pH vrijednosti sline nastaje vrlo brz porast koncentracije natrija: već u prvoj minuti ta vrijednost poraste za oko 8 puta. Natrij se izlučuje u acinarnim stanicama aktivno, a kloridi slijede natrij pasivnim prijenosom pa je njihov porast nešto manji. Najvažnija posljedica pada pH vrijednosti sline jest porast koncentracije kalcija zbog otapanja zubne cakline. Kao što se vidi iz Tablice 1, koncentracija kalcija porasla je za više od 10 puta. Istodobno, velika koncentracija natrija uzrokuje porast pH vrijednosti sline, pa se već u petoj minuti pH vrijednosti sline vraćaju na početnu vrijednost. Porast pH vrijednosti prati i pad koncentracije slobodnog (ioniziranog) kalcija, što pokazuje da proces remineralizacije zubne cakline nastupa vrlo brzo nakon procesa njezine demineralizacije. Velik raspon izmjerenih vrijednosti upozorava na veliku interindividualnu varijabilnost u odgovoru na promjene pH vrijednosti sline, što vjerojatno može imati za posljedicu i različitu individualnu podložnost karijesu. Procesi demineralizacije i remineralizacije isključivo su kemijske reakcije ovisne o pH vrijednosti medija; u neutralnom pH području ravnoteža je gotovo posve pomaknuta u smjeru taloženja kalcijeva apatita. S druge strane, pri tim pH vrijednostima koncentracije kalcija i fosfata u području su supersaturacije. Zbog toga će svaki porast koncentracije kalcija uz istodobno održavanje pH vrijednosti sline oko 7 uzrokovati njegovo taloženje u obliku kalcijeva apatita, odnosno remineralizaciju zubne cakline.

Prijašnja istraživanja provedena *in vitro* pokazala su da se dodatkom magnezijevih iona u slinu može istisnuti kalcij koji je vezan na proteine u slini, te na taj način odrediti ukupni kalcij u slini (11). Istraživanje opisano u ovome radu, provedeno *in vivo* na istoj populaciji kao i prije spomenuto, pokazalo je da u nazočnosti magnezijevih iona nastaje sličan učinak. Uz vrijeme žvakanja od 10 minuta, izmjerene koncentracije ioniziranog kalcija u podskupinama u skupinama A (osobe sa zdravim zubima) i B (osobe s razvijenim karijesom), koje su primile magnezij u žvakaćoj gumi, bile su više nego kod podskupina koje nisu primile magnezij, ali te razlike nisu bile statistički znatne. No, vrijednosti ukupnoga kalcija nisu se povećale, što znači da se dodatkom magnezija oslobađa više kalcija iz kompleksa dajući tako slobodni kalcij, a istodobno vrijednost pH ostaje u neutralnom području.

Zaključak

Provedena ispitivanja pokazala su da se nagle promjene koncentracije elektrolita u slini nastale zbog pada pH vrijednosti sline mogu djelotvorno i brzo popraviti samim lučenjem sline. Također se je pokazalo da se koncentracija slobodnoga kalcija u slini povećava dodatkom magnezija pri pH vrijednostima sline oko 7. Potvrđeno je da potenciometrijska mjerenja omogućuju pouzdana, brza i simultana mjerenja koncentracije elektrolita i pH vrijednosti sline. No, opažene interindividualne razlike u koncentraciji elektrolita i pH vrijednosti sline, neovisne o dobi i spolu te prehrambenim i higijenskim navikama ispitanika, premda nisu bile predmet ovog istraživanja, otežavaju tumačenje rezultata i potvrđuju prijašnja opažanja o slini kao kompleksnom dijagnostičkom uzorku.

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Electrolyte Concentration Change in Saliva after the Addition of Hydrogen and Magnesium Ions

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Summary

Changes in electrolyte concentration and pH in saliva have an important role in the enamel demineralization of teeth. Saliva is very important in maintaining neutral pH and concentration of electrolytes necessary to promote enamel remineralization. In this study we have investigated the changes of K^+, Na^+, Cl^- *and* Ca^{++} *concentrations in saliva due to pH decrease and after the addition of magnesium ions. All measurements were obtained by potentiometry using ion selective electrode. The results of this study showed that significant pH decrease could be effectively improved in five minutes by saliva secretion only. Concentration of calcium ion increased in all subgroups supplemented by magnesium ion in the chewing gum, while the pH values remained about 7.*

Key words: *caries, calcium, electrolyte in saliva, magnesium, pH*

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Introduction

Despite the data on diminished caries frequency in children and adolescents in some developed countries, it still remains the most widespread dental disease conditioned by nutritional and hygiene habits. According to a USA study, the incidence of caries among adults of both sexes equals that in children (1). Due to its high incidence, this disease has been the subject of numerous investigations performed with the aim of detecting the preventive procedures. These investigations have confirmed an association between caries and salivary mineral content (2,3). Despite precise knowledge of the chemical processes of demineralization and remineralization of teeth, a completely effective answer to this problem has not yet been given. Differences in the intensity of saliva secretion and degradation of carbohydrates from food, and of acids from plaque, may explain considerable differences in sensitivity towards caries in different individuals, but also between various sites in the dental cavity (4,5). Conducted studies have shown that fluoride use could significantly decrease the prevalence of this disease (6, 7).

Saliva is very important in maintaining the neutral pH of plaque and the dental cavity. Changes with possible harmful effects on enamel occur in the case of sudden pH changes or intake of other electrolytes through food. Saliva should compensate for these changes, while saliva buffer capacity depends on the content of bicarbonates, phosphates and other ions (8). Calcium phosphate concentration in enamel and saliva plays a very important role in tooth decay prevention. Studies including calcium and phosphate additives to chewing gums have shown favorable effects on tooth decay decrease (9, 10). The use of tricalcium phosphate has lead to an increase in pH and total and free calcium concentration. Results of these studies suggest that calcium accumulation in plaque and saliva, resulting from chewing gum use, may decelerate carious changes in teeth (10).

The primary aim of this study was to investigate the changes in electrolyte concentration caused by a pH decrease in subjects to whom chewing gums with added malic and tartaric acids were given. Besides monitoring saliva neutralization, the effect of pH on changes in the concentration of ionized calcium, potassium, sodium and chloride in saliva, was also observed during 5 minutes. The influence of magnesium ions in vivo on changes in ionized calcium concentration in saliva, as a potential ion for prevention of tooth demineralization, was also investigated. Determination of ionized calcium concentration was performed in subjects who were given magnesium-containing chewing gums.

Potentiometric methods were used in the study because they provide information on free ions concentration.

Materials and Methods

Subjects

The study included two different groups of subjects. The first group comprised a randomly formed group of 5 individuals aged from 17 to 45 years without prior stomatologic treatment. The second group consisted of children (N=30) from the Institution of Child Accommodation aged from 7 to 12 years, with a similar diet. This group was, on the basis of prior stomatological treatment, further divided into three groups: Group A with intact teeth (N=10), Group B with carious teeth (N=10), and Group C with filled teeth (N=10).

The effect of pH on salivary electrolyte concentration was monitored in the first group of patients, while the effect of magnesium was examined in the second group. The control group for both studies were subjects who took commercial sugarfree chewing gum without additives (marked as Mg-Group in Table 2). All examinations were performed in accordance with basic principals of the Helsinki Declaration (World Health Organization 1975).

Reagents

Commercial chewing gum containing malic and tartaric acids was used as a mean for decreasing pH, while sugarfree magnesium-containing chewing gum (16 mmol Mg^{2+}/ 1g of the chewing gum) was used to examine the effects of magnesium.

Magnesium of analytical purity was added in the form of magnesium chloride (Kemika, Zagreb). Total magnesium concentration in the chewing gum amounted to 48 mmol. This magnesium concentration was chosen on the basis of results already reported (8).

Method

Saliva specimens were taken from the bottom of the oral cavity (7,11). Before the first sampling of saliva all subjects washed out their mouths. Saliva was aspirated into polypropylene syringes which were subsequently closed by a rubber stopper and stored in a cool place.

Saliva samples were taken from the subjects in order to investigate the effect of pH change, while other samples were taken each minute during five minutes after they had been given chewing gum.

The subjects had the same chewing gum during the whole period of the observation.

For examination of magnesium influence on the calcium concentration in saliva, all three groups were divided into subgroups. The first Group-ewas given the magnesium-containing chewing gum, while a placebo was administered to the other group (i.e. chewing gum without the addition of magnesium). Subjects were on both occasions asked to rinse out their mouths. Saliva samples were taken from subjects who were then asked to masticate for 10 minutes. After which saliva samples were taken again.

Measurement

Determination of electrolyte concentration was performed on a laboratory-made instrument with flow-through design ion-selective electrodes (12). Concentrations of ionized calcium, sodium, potassium, chlorides as well as pH, were determined. Instrument calibration was performed using standard solutions by Ciba Corning (Cat. Nos.: 473385 and 483386, Ciba Corning Diagnostics, USA). Prior to the measurement, the instrument was controlled using commercial aqueous control solutions by Ciba Corning at all three levels of concentrations, yielding satisfactory results.

Total calcium concentration was determined on a flame photometer EFOX 5053 (Eppendorf, Germany).

Statistical methods

Windows Excel application was used for statistical processing of results. Results are presented as medians with determination ranges. The level of significance was examined using Student's t-test ($p = 0.05$).

Results

Study of pH effects

Results of pH effect studies are presented in Table 1. Due to the high range of determined values, median was taken as the mean value, rather than mean arithmetic value. These data are presented schematically in Fig. 1. Initial values of pH and concentration of ionized calcium, potassium, sodium and chloride in saliva are shown as values at zero time. Initial values for pH in saliva were in the range 6.79-6.95, sodium 9.9-17.7 mM, potassium 15.2 - 18.2 mM, chloride 13.6 - 18.8 mM and for ionized calcium 0.5 - 0.73 mM.

Dynamics of change in electrolyte concentration in saliva occurs due to the change in pH. Significant changes in sodium, chloride and calcium concentrations and slight changes in potassium concentration are evident in Figure 1. Figure 1 also shows that pH and calcium values are restored to their initial values after five minutes.

Effect of magnesium ions

Changes in electrolyte concentration caused by the use of the magnesium-containing chewing gum are presented in Table 2. Free calcium concentrations were higher in Groups A and B, whereas the median for calcium in Group C was increased in the subgroup without magnesium supplementation, although these differences were not statistically significant ($p< 0.05$). Differences in pH values between the above groups were also not statistically significant ($p< 0.05$). However, the group with decayed teeth (B) had lower pH than the other two groups. Chloride concentration was higher in the group with magnesium supplementation, which could be ascribed to the exogenous chloride intake (magnesium chloride), although the difference was also not statistically significant ($p<0.05$). Changes in potassium and sodium concentration also failed to differ statistically significantly ($p< 0.05$). Total calcium concentration also did not differ significantly.

Discussion

Demineralization of dental enamel is the cause of tooth decay. Dental enamel consists of calcium hydroxy-apatite. Solubility of this mineral depends to a large extent on the pH of the medium: a higher concentration of calcium ions is necessary to maintain saturation at the low pH (8). As the oral cavity fluid, saliva has the task of rinsing and cleansing the plaque from leftover food (carbohydrates), while its buffer effect is less pronounced. Studies by other authors have shown equal plaque reactions from caries-resistant and caries-prone individuals to pH decrease. However, the level at which these events occur is basically different: initial pH value is higher in caries-resistant individuals and pH decline is less pronounced.

This study showed that buffer capacity was higher in caries-resistant individuals (7,13). It was undertaken with the aim of investigating changes in electrolyte concentration, particularly calcium, caused by sudden pH decline in saliva.

During the pH decrease, sodium concentration increases very rapidly: there is an 8-fold increase after only one minute. Sodium is excreted in acinar cells actively, followed by passively transported chlorides whose increase is less pronounced. Calcium concentration due to dissolving of dental enamel is the most significant consequence of the pH decrease: apparently, there is a more than 10-fold calcium concentration increase. Sodium concentration at the same time leads to a salivary increase of pH which regains its initial value after only five minutes. The increase in pH value is followed by a decrease in free (ionized) calcium concentration, which points to very rapid interchange of the processes of remineralization and demineralization of dental enamel. The broad range of determined values indicates considerable interindividual variability as a response to pH changes, which may probably result in different individual susceptibility to caries. Highly variable saliva composition, as well as a probable different ability for sodium excretion, require different response to the decline in pH value. The processes of remineralization and demineralization are exclusively chemical reactions depending on pH; the pH balance around 7 is almost completely shifted in the direction of calcium appatite precipitation. The calcium and phosphate concentrations in this pH region are at the level of supersaturation. Therefore, each calcium concentration increase with the same pH value of 7 will, according to the law of mass action, contribute to the precipitation, i.e. remineralization of dental enamel.

Previously in vitro studies have shown that complex-bound calcium may be released by the addition of magnesium ions to saliva and thus determine the total calcium (tCa) in saliva (11). This study, conducted in vivo on the same population as those mentioned above, have shown that similar effect occurs in the presence of magnesium ions. With a mastication period of 10 minutes, the ionized calcium (iCa^{2+}) concentrations determined in subgroups of Group A (individuals with healthy teeth) and Group B (individuals with developed caries) who were administered magnesium-containing chewing gums, were higher than in subgroups without this supplementation, although the differences were not statistically significant. Total calcium values were, however, not increased, which means that the addition of magnesium helps the release of more complex-bound calcium and thus the concentration of free calcium increases, while the pH level remains in the neutral region.

Conclusion

The studies performed showed that significant pH changes of saliva could be very effectively and rapidly a treated through saliva secretion only. Also, free calcium concentration increases in saliva as a result of released magnesium after mastication of chewing gum containing magnesium, while pH remains close to 7. However, the observed considerable interindividual differences in salivary content independent of age, sex and nutritional habits, were not the subjects of the studies, but make the result interpretation more complex and confirm earlier observations regarding the use of saliva as a diagnostic sample. The results obtained by potentiometric measurements proved that this method is rapid and reliable and could be proposed for simultaneous determination of electrolyte concentrations and pH, allowing current assessment of events.

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