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## Saliva Changes in Pediatric Patients with Eating Disorders

### Promjene u slini pedijatrijskih bolesnika s poremećajima u jedenju

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#### Abstract

**Objectives:** To determine the differences between pediatric patients with eating disorders (ED) and the control group in the amount of saliva and the concentration of total amylase and electrolytes in saliva, and to evaluate the correlation between the saliva changes and nutritional status. **Material and methods:** The study included 101 participants (14.34 ± 1.99 years), out of which 50 participants with ED subgroups and 51 participants in the control group. Data were statistically analyzed (Mann-Whitney, Kruskal-Wallis, chi-square, Spearman rank correlation test,  $\alpha=0.05$ ). **Results:** No significant differences in salivary volume between the groups were found. A significant difference in the volume of saliva secreted in the 5th and 15th minute was found between the anorexia nervosa and bulimia nervosa subgroups. The examined anthropometric parameters were marginally or significantly positively associated with saliva volume at 5 and 15 minutes, noting a more significant correlation of the same at 15 than at 5 minutes. The patients with ED had a significantly higher concentration of inorganic phosphates in saliva while the concentrations of other electrolytes and total amylase in saliva did not differ significantly. **Conclusions:** Nutritional status affects salivation. There is a difference in saliva volume in pediatric patients with different ED disorders. Variations in saliva electrolytes in pediatric patients with ED are possible.

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#### Introduction

Eating disorders (ED) are mental health conditions which are, according to the Diagnostic and Statistical Manual of Mental Disorders (DSM V), classified as Anorexia nervosa (AN) restrictive type (AN-RT) or binge/purge type (AN-BP), Bulimia nervosa (BN), Binge eating (BE), Other Specified Feeding or Eating Disorder (OSFED) and Unspecified Feeding or Eating Disorder (UFED) (1). The last two disorders, that is, OSFED and UFED, came from what used to be called eating disorders not otherwise specified (EDNOS) in DSM IV (2). ED is a relatively rare disorder occurring relatively rarely in the general population. Lifetime prevalence estimates for anorexia nervosa, bulimia nervosa, and binge-

#### Uvod

Poremećaji u jedenju (PUJ) psihijatrijske su bolesti koje su klasificirane prema Dijagnostičkom i statističkom priručniku za mentalne poremećaje (DSM V) kao anoreksija nervoza (AN) koja može biti restriktivna (RT) ili bulimično-purgativna (BP), zatim kao bulimija nervoza (BN), poremećaj prejedanja (BE), drugi specificirani poremećaji u hranjenju ili prehrani (OSFED) i nespecificirani poremećaj u hranjenju ili prehrani (UFED) (1). Posljednja dva – OSFED i UFED – potječu od onoga što se nekada nazivalo poremećajima u jedenju koji nisu drukčije specificirani (EDNOS) u DSM-u IV (2). PUJ-i su razmjerno rijetki u općoj populaciji. Procjena životne prevalencije AN-a, BN-a i BE-a u SAD-u iznosi 0,3

eating disorder in USA were 0.3%, 0.9%, and 1.6%, respectively (3). Among European women, the prevalence of AN is <1-4%, BN <1-2%, overeating disorders 1-4%, other eating disorders 3% with significant differences depending on the country/region, age groups and ethnicity. Among men, 0.3% of them met the criteria for one of the eating disorders (4). There is no accurate data on the prevalence of eating disorders in Croatia. However, there are a few studies showing that a significant number of adolescents exhibit abnormal eating habits. And even though ED usually occurs in early puberty, we are witnessing a shift towards younger age groups (5–8).

Numerous changes involving all organ systems are developing as a result of dysfunctional eating behavior in patients with eating disorders (ED). The changes also affect the oral cavity. The most commonly described oral findings are dental plaque, marginal gingivitis, calculus, caries, exfoliative cheilitis, angular cheilitis and burning tongue (9–13). Some changes in the volume and composition of saliva are also described (14). Serum amylase levels are often elevated in patients with BN and AN-BP, most often due to an increase in salivary amylase due to frequent and intense vomiting (15). Patients with ED often complain of a feeling of reduced salivation and consequently dry mouth (16).

Although numerous studies about composition and volume of saliva, electrolytes and total amylase in saliva in patients with eating disorders (10, 14, 17–25) were performed, the existing data about these parameters in pediatric patients with ED are scarce.

The aim of this study was to determine differences between the group of pediatric patients with ED and the control group in the amount of saliva and the concentration of total amylase and electrolytes in saliva and to evaluate the correlation between saliva changes and nutritional status.

## Material and methods

This cross-sectional study was carried out from September 2016 until November 2017 at the Department of Pediatric Gastroenterology in University Hospital Center Sestre milosrdnice Zagreb and School of Dental Medicine, Zagreb, Croatia in accordance with the Declaration of Helsinki and approved by the Hospital Ethics Committee and by the Ethics Committee of the School of Dental Medicine, University of Zagreb (05-PA-26-1/2016). The study included 101 female participants with a mean age of  $14.34 \pm 1.99$  years, of which 50 participants had eating disorders and 51 participants were included in the control group. The patients included in the study as well as their parents received both written and oral information about the study and a written informed consent was signed by the parents. The study group consisted of 50 patients with ED who were hospitalized in the Department of Gastroenterology, Hepatology and Eating Disorders of the Pediatric Clinic (University Hospital Center Sestre milosrdnice, Zagreb, Croatia). The diagnosis of ED was made according to DSM V criteria. Participants were classified into subgroups of ED: AN-RT, AN-BP, BN, BE, UFED and OSFED. Participants with AN-RT and AN-BP, due to the small number of participants, were merged into one group (AN). Also, for the

%, 0,9 %, odnosno 1,6 % (3). Među europskim ženama prevalencija AN-a je < 1 do 4 %, BN-a < 1 do 2 %, BE-a 1 do 4 %, a ostalih PUJ-a 3 % sa znatnim razlikama ovisno o zemlji/regiji, dobnim skupinama i etničkoj pripadnosti. Među muškarcima njih 0,3 % zadovoljava kriterije za jedan od PUJ-a (4). Ne postoje točni podatci o učestalosti PUJ-a u Hrvatskoj. No u nekoliko studija autori su istaknuli da znatan broj adolescenata ima nenormalne prehrambene navike. Iako se PUJ obično pojavljuje u ranom pubertetu, svjedoci smo pomaka prema mlađim dobnim skupinama (5 – 8).

Mnogobrojne promjene koje zahvaćaju sve organske sustave nastaju kao posljedica disfunkcionalnog ponašanja u prehrani bolesnika s PUJ-om. Promjene zahvaćaju i usnu šupljinu. Najčešće opisani oralni nalazi su zubni plak, marginalni gingivitis, kamenac, karijes, eksfolijativni heilitis, angularni heilitis i pečenje jezika (9 – 13). Također su opisane promjene u volumenu i sastavu sline (14). Razine serumske amilaze često su povišene kod pacijenata s BN-om i AN-BP-om, a najčešće su prouzročene zbog čestog i intenzivnog povraćanja (15). Pacijenti s PUJ-om često se žale na osjećaj smanjenoga izlučivanja sline i posljedično na suha usta (16).

Unatoč studijama o sastavu i volumenu sline, elektrolita i ukupne amilaze u slini pacijenata s PUJ-om (10, 14, 17 – 25), oskudni su dosadašnji podatci o tim parametrima kod pedijatrijskih bolesnika s tim poremećajima.

Cilj ovog istraživanja bio je ustanoviti razlike između pedijatrijskih bolesnika s PUJ-om i kontrolne skupine u količini sline i koncentraciji ukupne amilaze i elektrolita u slini te procijeniti korelaciju između promjena u slini i nutritivnog statusa.

## Materijal i metode

Ovo presječno istraživanje provedeno je u skladu s Helsinškom deklaracijom od rujna 2016. do studenoga 2017. u Zavodu za pedijatrijsku gastroenterologiju KBC-a Sestre milosrdnice u Zagrebu i na Stomatološkom fakultetu u Zagrebu. Odobrilo ga je Etičko povjerenstvo KBC-a i Etičko povjerenstvo Stomatološkog fakulteta Sveučilišta (05-PA-26-1/2016). Istraživanjem je obuhvaćena 101 sudionica prosječne dobi od  $14,34 \pm 1,99$  godina, od čega 50 s PUJ-om i 51 u kontrolnoj skupini. Pacijentice uključene u studiju i njihovi roditelji dobili su pismene i usmene informacije o istraživanju, a roditelji su potpisali informirani pristanak. Studijsku skupinu činilo je 50 pacijentica s PUJ-om koje su bile hospitalizirane na Odjelu za gastroenterologiju, hepatologiju i poremećaje u prehrani Klinike za pedijatriju (KBC Sestre milosrdnice, Zagreb, Hrvatska). Dijagnoza PUJ-a postavljena je prema kriterijima DSM-a V. Bile su razvrstane u podskupine AN-RT, AN-BP, BN, BE, UFED i OSFED. Sudionice s AN-RT-om i AN-BP-om zbog malobrojnosti su bile svrstane u jednu grupu (AN). Iz istog razloga su i poremećaji BE, UFED i OSFED spojeni u jednu skupinu pod nazivom EDNOS (naziv je preuzet iz kriterija DSM-a IV). Kontrolnu skupinu činila je 51 sudionica iste dobi i one su tako-

same reason, BE, UFED and OSFED disorders were merged into one group named EDNOS (the name is taken from the DSM IV criteria). The control group consisted of 51 participants of the same age who were also hospitalized in the Pediatric Clinic where they were admitted for treatment due to symptoms of diseases outside the gastrointestinal tract, i.e. due to conditions and diseases in which no changes in the oral cavity were expected. Each participant underwent anthropometric measurements, determination of body height in centimeters and body weight in kg. The measurements were performed with a scale with an altimeter and measuring scale, in the morning on an empty stomach after waking up, wearing only underwear. The Body Mass Index (BMI) was calculated by dividing weight and height squared ( $\text{kg}/\text{m}^2$ ). The nutritional status was expressed in z-score for BMI, since the values of BMI change with age in children. Whole unstimulated saliva samples were collected between 10 and 12 o'clock in the morning, at least 2 hours after the last meal or fluid intake. Participants sat with their head tilted forward and, after initially swallowing saliva, they expelled unstimulated saliva every 60 seconds into calibrated tubes (0.1 ml) for 15 minutes. Salivary flow, expressed in milliliters in the fifth and fifteenth minutes, was determined for each participant. Samples of collected unstimulated saliva were submitted for processing to the Clinical Department of Chemistry of the University Hospital Centre Sisters of Mercy Zagreb within one hour of sampling, frozen at  $-60^\circ\text{C}$  and stored until used for measuring electrolyte concentration and total amylase. The determination of electrolytes in saliva was performed using the method of indirect potentiometric on the analytical platform Architect c8000 (Abbott Laboratories, Chicago, IL, USA). The determination of a total salivary amylase was performed using the spectrophotometric enzymatic method on the same analyzer.

Statistical analysis was performed by Statistical software package version 13.5 (TIBCO Inc., Palo Alto, CA, USA). The normality of data distribution was tested by the Shapiro-Wilk's W test and homogeneity of variance by Leven test. The results of descriptive analyses were expressed as median (min-max) for continuous data and percentages for categorical data. The differences in distributions of continuous data were evaluated by nonparametric tests (Mann-Whitney, Kruskal-Wallis) since the assumption of homogeneity of variance for tested variables had not been proven. The multiple comparison tests were used for post-hoc analysis. The differences in distributions of categorical data were assessed by the Chi-square test. The Spearman rank correlation test was used to evaluate the direction and the strength of the associations between the tested variables;  $p < 0.05$  was considered statistically significant.

## Results

The study included 101 participants with a mean age of  $14.34 \pm 1.99$  years, of which 50 participants were patients with ED and 51 participants were included in the control group. Participants from both groups did not significantly differ by age ( $p=0.261$ ). Patients with ED were significantly malnourished. They had significantly lower BMI z-value ( $t=9.822$ ,  $p<0.001$ ) than the participants in the control group. Table 1 shows subjective feeling of saliva volume and

der bile hospitalizirane u Klinici za pedijatriju, a primljene su na liječenje zbog simptoma bolesti izvan gastrointestinalnog trakta, odnosno zbog stanja i bolesti kod kojih se ne očekuju promjene u usnoj šupljini. Svaka sudionica bila je podvrgnuta antropometrijskim mjerenjima, određivanju tjelesne visine u centimetrima i tjelesne težine u kilogramima. Mjerenja su obavljena vagom s visinomjerom i mjernom ljestvicom, ujutro natašte nakon buđenja, samo u donjem rublju. Indeks tjelesne mase (ITM) izračunat je dijeljenjem težine i visine na kvadrat ( $\text{kg}/\text{m}^2$ ). Prehrambeni status izražen je kao z-vrijednost za ITM, zato što se vrijednosti ITM-a kod djece mijenjaju s dobi. Ukupna nestimulirana slina prikupljena je između 10 i 12 sati prije podne, najmanje dva sata nakon posljednjega obroka ili pijenja tekućine. Sudionice su sjedile nagnute glave prema naprijed i nakon što su najprije progutale slinu, izbacivale su 15 minuta nestimuliranu slinu svakih 60 sekunda u kalibrirane epruvete (0,1 mL). Za svaku sudionicu određen je protok sline izražen u mililitrima u petoj i petnaestoj minuti. Uzorci nestimulirane sline proslijeđeni su na obradu Kliničkome odjelu za kemiju KBC-a Sestre milosrdnice u roku od jednog sata od uzorkovanja, zamrznuti su na  $-60^\circ\text{C}$  i pohranjeni do mjerenja koncentracije elektrolita i ukupne amilaze. Određivanje elektrolita u slini provedeno je metodom neizravne potencijometrije na analitičkoj platformi Architect c8000 (Abbott Laboratories, Chicago, IL, SAD). Određivanje ukupne amilaze u slini obavljeno je spektrofotometrijskom enzimskom metodom na istom analizatoru.

Statistička analiza provedena je u softverskom paketu Statistica verzije 13.5 (TIBCO Inc., Palo Alto, CA, SAD). Normalnost distribucije podataka testirana je Shapiro-Wilkovim W testom, a homogenost varijance Levenovim testom. Rezultati deskriptivne analize izraženi su kao medijan (min. – maks.) za kontinuirane podatke i postotci za kategoričke podatke. Razlike u distribucijama kontinuiranih podataka evaluirane su neparametrijskim testovima (Mann-Whitneyjev, Kruskal-Wallisov) zato što pretpostavka o homogenosti varijance za testirane varijable nije bila ispunjena. Za post-hoc analizu korišten je test višestruke usporedbe. Razlike u distribucijama kategoričkih podataka procijenjene su Hi-kvadrat testom. Spearmanov koeficijent korelacije ranga korišten je za procjenu smjera i jačine povezanosti između testiranih varijabli. Statistički značajnim smatran je  $p < 0,05$ .

## Rezultati

U istraživanje je bila uključena 101 sudionica, a prosjek godina iznosio je od  $14,34 \pm 1,99$ . Njih 50 patilo je od poremećaja u jedenju, a 51 je bila u kontrolnoj skupini. Sudionice iz obje skupine nisu se statistički značajno razlikovale prema dobi ( $p = 0,261$ ). Pacijentice s PUJ-om bile su znatno pothranjene. Imale su znatno nižu ITM z-vrijednost ( $t = 9,822$ ,  $p < 0,001$ ) u usporedbi sa sudionicama u kontrolnoj skupini. Tablica 1. prikazuje subjektivni osjećaj količine sline i količinu

volume of secreted saliva in examined groups. Table 2 shows subjective feeling of saliva volume and volume of saliva secretion in patients with ED divided into subgroups according to the type of eating disorder. A post hoc multiple comparison test showed a significant difference in the volume of saliva secreted in the 5th ( $p=0.007$ ) and 15th minute ( $p=0.028$ ) only between the AN and BN subgroups.

All examined anthropometric parameters were marginally or significantly positively associated with saliva volume at 5th and 15th minute (Table 3, Figures 1 and 2), noting a more significant correlation of the same at 15th than at 5th minute.

Total amylase in saliva did not differ significantly between patients with ED and the control group (med. 82802 vs. 77891 U/L;  $p=0.666$ ). Table 4 shows concentration of total salivary amylase in patients with ED divided into subgroups according to the type of eating disorder. A marginally higher concentration of total serum amylase was found in patients with ED than in the control group (med. 56.5 vs. 50 U/L,  $p=0.052$ ) but no association between serum amylase and salivary amylase concentrations has been demonstrated ( $p>0.05$ ).

Patients with ED had a significantly higher concentration of inorganic phosphates in saliva than participants in the control group ( $p=0.025$ ), while the concentrations of other electrolytes in saliva did not differ significantly (Table 5). There was no significant difference in salivary electrolyte concentration among patients with different types of ED ( $p>0.05$ ) (data not shown). BMI z-score and salivary volume at 5 and 15 minutes were marginally or significantly negatively associat-

izlučene sline u ispitivanim skupinama.

Tablica 2. prikazuje subjektivni osjećaj količine sline i količinu izlučene sline kod pacijentica s PUJ-om, podijeljenih u podskupine prema tipu poremećaja u jedenju. Post-hoc testom višestruke usporedbe utvrđena je značajna razlika u količini sline izlučene u 5. minuti ( $p = 0,007$ ) i 15. minuti ( $p = 0,028$ ) samo između podskupina AN i BN.

Svi proučavani antropometrijski parametri bili su marginalno ili značajno pozitivno povezani s količinom sline u 5. i 15. minuti (tablica 3., slika 1. i 2.), s time da je uočena značajnija korelacija u 15. minuti nego u 5. minuti.

Ukupna amilaza u slini nije se značajno razlikovala između pacijentica s PUJ-om i kontrolne skupine (med. 82802 prema 77891 U/L;  $p = 0,666$ ). U tablici 4. zabilježena je koncentracija ukupne amilaze u slini pacijentica s PUJ-om podijeljenih u podskupine prema tipu poremećaja u jedenju. Marginalno viša koncentracija ukupne amilaze u serumu ustanovljena je kod sudionica s PUJ-om u usporedbi s kontrolnom skupinom (med. 56,5 prema 50 U/L,  $p = 0,052$ ), ali nije pronađena veza između koncentracije amilaze u serumu i amilaze u slini ( $p > 0,05$ ).

Pacijentice s PUJ-om imale su znatno višu koncentraciju anorganskih fosfata u slini u usporedbi s onima u kontrolnoj skupini ( $p = 0,025$ ), a koncentracije ostalih elektrolita nisu se značajno razlikovale (tablica 5.). Nije utvrđena značajna razlika u koncentraciji elektrolita u slini između pacijentica s različitim tipom PUJ-a ( $p > 0,05$ ) (podatci nisu prikazani). ITM z-vrijednost i količina sline u 5. i 15. minuti bili su mar-

**Table 1** Subjective sensation of saliva volume and volume of saliva secreted in participants divided into two groups according to the presence/absence of ED.

**Tablica 1.** Subjektivni osjećaj količine sline i izlučena količina sline kod sudionica podijeljenih u dvije skupine prema prisutnosti/odsutnosti PUJ-a

	PUJ pacijenti • ED patients (n = 50)	Kontrolna skupina • Control group (n = 51)	Chi <sup>a</sup> Z <sup>b</sup>	p
Subjective sensation of saliva volume* • Subjektivni osjećaj količine sline*	20/72/8	12/86/2	3.590 <sup>a</sup>	0.166 <sup>a</sup>
Saliva volume in the 5th minute (mL)** • Količina sline u 5. minuti (mL)**	1, 0 - 3.3	1.4, 0.1 - 8.5	1.609 <sup>b</sup>	0.107 <sup>b</sup>
Saliva volume in the 15th minute (mL)** • Količina sline u 15. minuti (mL)**	2.5, 0.5 - 8	3.5, 0.6 - 15	1.434 <sup>b</sup>	0.143 <sup>b</sup>

\* percentages • postotak; \*\* med, min-max • med., min. – maks.; a Chi-square test  $df=2$  • Hi kvadrat test  $df = 2$ ; b Mann-Whitney test • Mann-Whitneyjev test; Subjective sensation of saliva volume: decreased/normal/increased • Subjektivni osjećaj količine sline: smanjen/normalan/povećan; ED - eating disorders • PUJ – poremećaji u jedenju

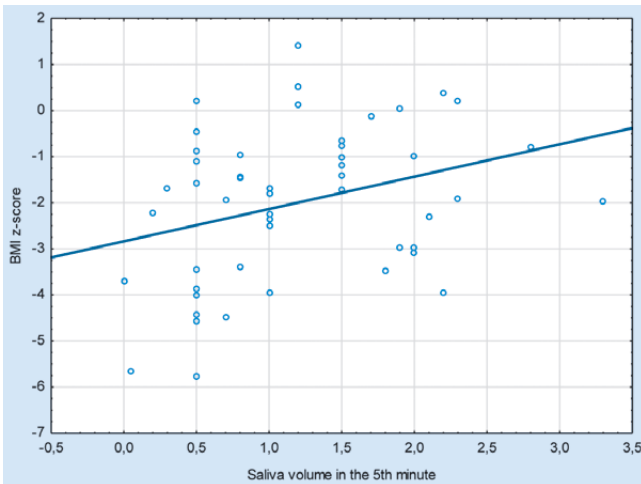
**Table 2** Subjective sensation of saliva volume and volume of saliva secretion in patients with ED divided into subgroups according to the type of eating disorder.

**Tablica 2.** Subjektivni osjećaj količine sline i izlučena količina sline kod pacijentica s PUJ-om podijeljenih u skupine prema tipu poremećaja u jedenju

	AN (n = 7)	BN (n = 6)	EDNOS (n = 17)	Chi <sup>a</sup> H <sup>b</sup>	p
Subjective sensation of saliva volume* • Subjektivni osjećaj količine sline *	22/70/8	33/50/17	12/82/6	2.513 <sup>a</sup>	0.642 <sup>b</sup>
Saliva volume in the 5th minute (ml)** • Količina sline u 5. minuti (ml)**	0.8, 0-2.3	1.9, 1.2-2.8	1.2, 0.5-3.3	10.449 <sup>b</sup>	<b>0.005<sup>b</sup></b>
Saliva volume in the 15th minute (ml)** • Količina sline u 15. minuti (ml)**	2.2, 0.5-7	4.7, 2.5-8	3.6, 0.7-7	9.274 <sup>b</sup>	<b>0.009<sup>b</sup></b>

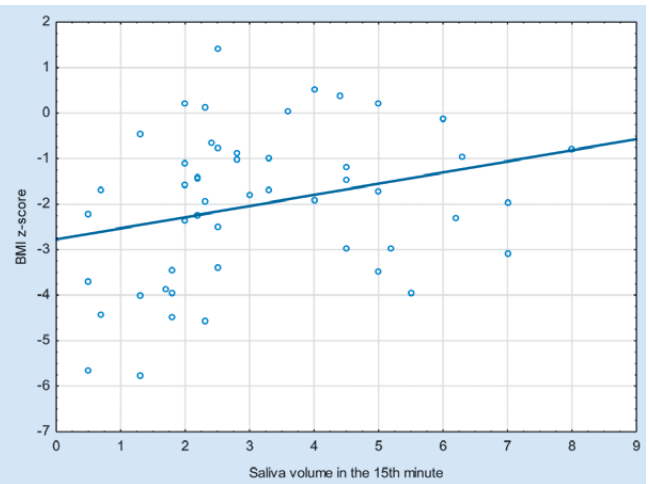
\* percentages • postotak; \*\* med, min-max • med., min. – maks.; <sup>a</sup> Chi-square test  $df=4$  • Hi kvadrat test  $df = 4$ ; <sup>b</sup> Kruskal-Wallis test • Kruskal-Wallisov test; Subjective sensation of saliva volume: decreased/normal/increased • Subjektivni osjećaj količine sline: smanjen/normalan/povećan; AN – anorexia nervosa • anoreksija nervoza; BN – Bulimia nervosa • bulimija nervoza; EDNOS – eating disorders not otherwise specified • poremećaj u jedenju, neodređen





**Figure 1** Relationship of BMI z-score with saliva volume at 5 minutes (all participants).

**Slika 1.** Povezanost ITM z-vrijednosti s količinom sline u 5. minuti (sve sudionice); ITM – indeks tjelesne mase.



**Figure 2** Relationship of BMI z-score with saliva volume at 15 minutes (all participants).

**Slika 2.** Povezanost ITM z-vrijednosti s količinom sline u 15. minuti (sve sudionice); ITM – indeks tjelesne mase.

**Table 3** Correlations of anthropometric parameters with saliva volume at 5 and 15 minutes (all participants).

**Tablica 3.** Korelacija antropometrijskih parametara s izlučenom količinom sline u 5. i 15. minuti (sve sudionice)

		BMI • ITM	BMI z-score • ITM z-vrijednost	BMI percentile • ITM percentil
Saliva volume in the 5th minute • Količina sline u 5. minuti	Spearman R	0.225	0.197	0.191
	t(N-2)	2.295	1.999	1.933
	p	<b>0.024</b>	<b>0.048</b>	0.056
Saliva volume in the 15th minute • Količina sline u 15. minuti	Spearman R	0.238	0.213	0.204
	t(N-2)	2.435	2.168	2.070
	p	<b>0.017</b>	<b>0.033</b>	<b>0.041</b>

BMI - body mass index • ITM – indeks tjelesne mase

**Table 4** Concentration of total salivary amylase in patients with ED divided into subgroups according to the type of eating disorder.

**Tablica 4.** Koncentracija ukupne salivarne amilaze kod pacijentica s PUJ-om podijeljenih u podskupine prema tipu poremećaja u jedenju

	AN (n = 27)	BN (n = 6)	EDNOS (n = 17)	H	P
Total amylase in saliva (U/L)* • Ukupna amilaza u slini (U/L)*	71030, 11007-357796	86212, 60658-383484	91907, 2998-309260	1.554	0.459

\* med, min-max • med., min-maks.; Kruskal-Wallis test • Kruskal-Wallisov test; AN – anorexia nervosa • anoreksija nervozna; BN – Bulimia nervosa • bulimija nervozna; EDNOS – eating disorders not otherwise specified • poremećaj u jedenju, neodređen

**Table 5** Salivary electrolyte concentration in participants divided into two groups according to the presence/absence of ED.

**Tablica 5.** Koncentracije elektrolita u slini sudionica podijeljenih u dvije skupine prema prisutnosti/odsutnosti PUJ-a

	ED patients • PUJ (n = 50)	Control group • Kontrolna skupina (n = 51)	Z	p
Total calcium (mmol/L)* • Ukupni kalcij (mmol/L)*	1.21, 0.92-2.94	1.11, 0.66-1.85	-1.446	0.147
Inorganic phosphates (mmol/L)* • Anorganski fosfati (mmol/L)*	6.08, 3.41-14.69	5.11, 2.89-19.2	-2.234	<b>0.025</b>
Magnesium (mmol/L)* • Magnezij (mmol/L)*	0.30, 0.25-0.92	0.27, 0.25-0.58	-1.653	0.098
Potassium (mmol/L)* • Kalij (mmol/L)*	24.4, 15.9-47.8	23.2, 15.5-44.6	-1.120	0.262
Sodium (mmol/L)* • Natrij (mmol/L)*	20, 20-28	20, 20-24	-0.173	0.862
Chlorides (mmol/L)* • Kloridi (mmol/L)*	22, 20-53	21, 20-34	-1.300	0.193

\* med, min-max • med., min. – maks.; Mann-Whitney test • Mann-Whitneyjev test; ED - eating disorders • PUJ – poremećaji u jedenju

ed with the concentrations of most of the tested electrolytes in saliva (Table 6), except for the concentrations of inorganic phosphates and a total salivary amylase with which they were not bounded, hence these results are not shown in any tables.

ginalno ili značajno negativno povezani s koncentracijom većine testiranih elektrolita (tablica 6.), osim s koncentracijom anorganskih fosfata i ukupnom amilazom u slini s kojom nisu bili povezani, te zato ti rezultati nisu u tablici.

**Table 6** Correlations of BMI z-score and saliva volume at 5 and 15 minutes with salivary electrolyte concentration.  
**Tablica 6.** Korelacija ITM z-vrijednosti i količine sline u 5. i 15. minuti s koncentracijom elektrolita u slini

Electrolytes in saliva • Elektroliti u slini		BMI z-score • ITM z-vrijednost	Saliva volume in the 5th minute • Količina sline u 5. minuti	Saliva volume in the 15th minute • Količina sline u 15. minuti
Total calcium • Ukupni kalcij	Spearman R	-0.098	-0.333	-0.280
	t(N-2)	-0.983	-1.767	-1.459
	p	0.328	0.074	0.157
Magnesium • Magnezij	Spearman R	-0.167	-0.279	-0.324
	t(N-2)	-1.688	-1.450	-1.817
	p	0.094	0.159	0.061
Potassium • Kalij	Spearman R	-0.070	-0.375	-0.336
	t(N-2)	-0.701	-2.022	-1.763
	p	0.485	0.054	0.066
Sodium • Natrij	Spearman R	-0.025	-0.329	-0.303
	t(N-2)	-0.252	-1.729	-1.589
	p	0.801	0.079	0.125
Chlorides • Kloridi	Spearman R	-0.165	-0.392	-0.354
	t(N-2)	-1.664	-2.131	-1.890
	p	0.099	<b>0.043</b>	0.059

BMI - body mass index • ITM – indeks tjelesne mase

## Discussion

Eating disorders are often diagnosed late. They are most commonly developed during adolescent age (15–19 years) (26–28). Complications of ED develop in all organ systems, and numerous changes in the oral cavity have been described (9–11). So far, changes in saliva have been described in patients with eating disorders in adulthood, with the exception of research conducted by Paszynska et al (29), which likewise in the present study included younger girls.

The results of a series of studies showed that patients with ED are more likely to report a subjective feeling of decreased saliva and have a smaller volume of unstimulated saliva at 5 and 15 minutes than participants in the control group (10,15–21), which could be considered similar to the results of this study (Table 1). However, the differences found between the groups did not reach statistical significance (Table 1), which coincides with the results published by Johansson et al (10). Decreased salivary flow (in the ED group in this study) was not due to antidepressant therapy and/or the use of appetite-suppressing drugs, which have reduced salivation as a side effect, because none of the participants used them. The results of a study by Paszynska et al (29), which included 23 girls with AN RT with an average age of 15.5 ± 2.1 years, showed a statistically significant difference in the flow of unstimulated saliva between the group with AN RT and the control group. The girls in Paszynska's study (29), as well as the patients with AN RT in this study, were significantly malnourished, which, along with dehydration, is a known cause of decreased saliva, which could explain the lower saliva flow in AN patients compared to BN patients (Table 2). This conclusion (29) was confirmed by a determined borderline or statistically significant positive correlation between the degree of nutrition and the volume of non-stimulating saliva, which was more pronounced in the 15th minute compared to the 5th minute of the study (Table 3, Figure 1 and 2).

The literature review has revealed different results for total amylase in saliva. Riad et al (17) found elevated amylase values in unstimulated saliva in bulimic patients. Elevated amylases in 25–60 % of patients with BN were also observed

## Rasprava

PUJ se često kasno dijagnosticira. Najčešće se pojavljuje u adolescenciji (15 – 19 godina) (26 – 28). Pritom komplikacije nastaju u svim organskim sustavima pa su opisane mnogobrojne promjene u usnoj šupljini (9 – 11). Zasad su promjene u slini opisane kod pacijenata s poremećajima u jedenju u odrasloj dobi, osim u istraživanju Paszynske i suradnika (29) koji su uključili mlade djevojke, kao i u ovom istraživanju.

U nizu studija istaknuto je da će pacijenti s PUJ-om vjerojatnije prijaviti subjektivni osjećaj smanjene sline i imat će smanjenu količinu nestimulirane sline u 5. i 15. minuti u usporedbi s ispitanicima u kontrolnoj skupini (10, 15 – 21), što se može smatrati rezultatima sličnima u ovom istraživanju (tablica 1.). Ipak, razlike između skupina nisu dosegnule statističku značajnost (tablica 1.), što je u skladu s rezultatima Johanssona i suradnika (10). Smanjeno izlučivanje sline (PUJ skupina) nije nastalo zbog terapije antidepresivima i/ili upotrebe lijekova za suzbijanje apetita koji mogu prouzročiti smanjenu salivaciju kao nuspojavu jer ih nijedna sudionica nije koristila. Rezultati istraživanja Paszynske i suradnika (29) u koje su bile uključene 23 djevojke s AN-RT-om s prosjekom godina od 15,5 ± 2,1, pokazali su statistički značajnu razliku u izlučivanju nestimulirane sline između kontrolne skupine i skupine s AN-RT-om. Ispitanice u toj studiji (29), kao i pacijentice s AN-RT-om u ovom istraživanju, bile su znatno pothranjene, što je zajedno s dehidracijom poznat uzrok smanjene sline i može objasniti smanjeno izlučivanje u slučaju AN-a u odnosu prema BN skupini (tablica 2.). Taj zaključak (29) potvrđen je s graničnom ili statistički značajnom pozitivnom korelacijom između stupnja uhranjenosti i količine nestimulirane sline, što je u ovom istraživanju bilo izraženije u 15. minuti u usporedbi s 5. minutom (tablica 3., slike 1. i 2.).

U literaturi mogu se pronaći različiti rezultati za ukupnu amilazu u slini. Riad i suradnici (17) pronašli su povišene vrijednosti amilaze u nestimuliranoj slini kod bulimičnih pacijenata. Povišene vrijednosti amilaze kod 25 do 60 % kod pacijenata s BN-om utvrđene su u raznim studijama, s pretpostavkom da je riječ o salivarnoj amilazi (16, 23, 30 – 32).

by a number of other researchers with the assumption that it was salivary amylase (16, 23, 30-32). Contrary to these studies (16,23,30-32), Paszynska et al (29) found no differences in amylase concentration between the group with AN RT and the control group, nor did Johansson et al (10), which is close to the present study results. Paszynska et al (29) explained these findings by the shorter duration of the disease with respect to the age of the patients. They have concluded that further research is needed to determine the effects of nutritional status on amylase excretion.

There are very few studies describing salivary electrolyte concentrations in ED patients. In the first studies examining salivary electrolyte levels, no significant differences were found between patients and control groups (17, 18). In the study by Johansson et al (14), significantly higher concentrations of phosphate in unstimulated saliva were determined which is similar to the results of the present study (Table 5). A higher concentration of phosphate which was obtained in unstimulated saliva could be linked with a decreased salivary flow in patients with ED (15-21) (Table 1). Yet, other electrolyte concentration did not differ between ED patients and the control group despite lower salivary flow in ED patients (Table 1). Johansson et al (14) also found significantly higher concentrations of chloride and magnesium (14). Blazer et al (24) found significantly higher concentrations of magnesium, and there was no significant difference in the concentrations of sodium, calcium, phosphate, potassium, and chloride. Statistically significantly higher calcium concentrations in unstimulated saliva in adult BN patients (23.8 ± 4 years) were noted by Dynesen et al (19), which is contrary to the results obtained in the present study (Table 5). However, some limitations in the present study should be pointed to such as the use of combined subgroups with distinct clinical characteristics (AN, BN and EDNOS within ED group). Based on previous investigations (14, 17-19, 24), and on the results of the present study (Table 5), the effect of ED on the state of electrolytes in saliva remains unclear. Nevertheless, some studies showed common electrolyte (serum) disturbances in children with malnutrition (33, 34). With this in mind, along with determined different concentrations of saliva electrolytes (Table 5) (14, 24, 30), variations in saliva electrolytes in ED can be considered possible. To confirm or dispute these findings, further studies are needed, especially with phosphate in saliva in pediatric patients with different types of ED.

## Conclusions

Nutritional status affects salivation. There is a difference in saliva volume in pediatric patients with different ED disorders. Variations in saliva electrolytes in pediatric patients with ED are possible.

## Conflict of interest

The authors declare that they do not have conflict of interest related to the present study.

**Author's Contribution:** D.V.J., A.C.R., T.L. – conceptualization; T.L., S.K.Š., A.V., N.N.G. – methodology; T.L., M.T. – analysis; T.L., D.V.J., A.V., N.N.G. – investigation; S.K.Š., A.V., A.C.R. – resources; T.L., S.Č., M.T. – writing, original draft preparation; S.Č., A.C.R. – writing, review and editing; S.K.Š., D.V.J. – supervision.

Suprotno tim istraživanjima (16, 23, 30 – 32), Paszynska i suradnici (29) nisu pronašli razlike u koncentraciji amilaze između AN-RT-a i kontrolne skupine ispitanika, kao ni Johansson i suradnici (10), što je slično rezultatima u ovom istraživanju. Paszynska i suradnici (29) ove nalaze objašnjavaju kraćim trajanjem bolesti s obzirom na godine pacijenata te zaključuju da su potrebna dodatna istraživanja kako bi se ustanovio utjecaj nutrijcijskoga statusa na izlučivanje amilaze.

U samo nekoliko studija opisana je koncentracija elektrolita u slini pacijenata s PUJ-om. U prvim istraživanjima čiji su autori proučavali elektrolite u slini nisu pronađene značajne razlike između pacijenata i kontrolnih skupina (17, 18). Johansson i suradnici ustanovili su u svojem istraživanju (14) znatno višu koncentraciju fosfata u nestimuliranoj slini, što je u skladu s ovim istraživanjem (tablica 5.). Utvrđena viša koncentracija fosfata u nestimuliranoj slini može se povezati sa smanjenim izlučivanjem sline kod pacijenata s PUJ-om (15 – 21) (tablica 1.). Ipak, koncentracije drugih elektrolita nisu se znatno razlikovale između PUJ-a i kontrolne skupine sudionika, unatoč smanjenom izlučivanju sline kod pacijenata s PUJ-om (tablica 1.). Johansson i suradnici (14) pronašli su i znatno veće koncentracije klorida i magnezija (14). Blazer i suradnici (24) zabilježili su znatno veće koncentracije magnezija, a nije bilo značajnih razlika u koncentraciji natrija, kalcija, fosfata, kalija i klorida. Statistički znatno veću koncentraciju kalcija u nestimuliranoj slini kod odraslih BN pacijenata (23, 8 ± 4 godine) ustanovili su Dynesen i suradnici (19), što je u suprotnosti s rezultatima dobivenima u ovoj studiji (tablica 5.). Potrebno je napomenuti ograničenje ovog istraživanja, a to je kombinacija podskupina s različitim kliničkim karakteristikama (AN, BN i EDNOS unutar PUJ skupine). S obzirom na rezultate prijašnjih istraživanja (14, 17 – 19, 24) i ovoga (tablica 5.), ostaje nejasan utjecaj PUJ-a na stanje elektrolita u slini. Ipak, u istraživanjima su istaknuti uobičajeni poremećaji elektrolita (serum) kod djece s malnutricijom (33, 34). S obzirom na te poremećaje, a uz utvrđene različite koncentracije elektrolita u slini (tablica 5.) (14, 24, 30), varijacije elektrolita u slini kod pacijenata s PUJ-om smatraju se mogućima. Da bi se potvrdili ili odbacili ovi nalazi, potrebna su daljnja istraživanja, posebice s fosfatom u slini pedijatrijskih bolesnika s različitim tipovima PUJ-a.

## Zaključak

Nutrijcijski status utječe na salivaciju. Postoji razlika u količini sline kod pedijatrijskih pacijenata s različitim poremećajima PUJ-a. Moguće su varijacije elektrolita u slini pedijatrijskih pacijenata s PUJ-om.

## Sukob interesa

Autori nisu bili u sukobu interesa.

**Doprinos autora:** D. V. J., A. C. R., T. L. – konceptualizacija; T. L., S. K. Š., A. V., N. N. G. – metodologija; T. L., M. T. – analiza; T. L., D. V. J., A. V., N. N. G. – istraživanje; S. K. Š., A. V., A. C. R. – resursi; T. L., S. Č., M. T. – pisanje, priprema rukopisa; S. Č., A. C. R. – pisanje, pregled i ureditanje; S. K. Š., D. V. J. – nadzor

**Sažetak**

**Ciljevi:** Ustanoviti razlike između pedijatrijskih bolesnika s poremećajima u jedenju (PUJ) i kontrolne skupine u količini sline i u koncentraciji ukupne amilaze i elektrolita u slini te procijeniti korelaciju između promjena u slini i nutritivnog statusa. **Materijal i metode:** Istraživanjem je obuhvaćena 101 sudionica (14,34 ± 1,99 godina), od čega 50 s podskupinama PUJ-a i 51 u kontrolnoj skupini. Podatci su statistički obrađeni (Mann-Whitneyev, Kruskal-Wallisov test, Hi-kvadrat test, Spearmanov koeficijent korelacije ranga,  $\alpha = 0,05$ ). **Rezultati:** Nisu pronađene značajne razlike u volumenu sline između skupina. Ustanovljena je značajna razlika u volumenu izlučene sline u 5. i 15. minuti između podskupina anoreksije nervoze i bulimije nervoze. Ispitani antropometrijski parametri bili su marginalno ili značajno pozitivno povezani s volumenom sline u 5. i 15. minuti, uz njihovu značajniju korelaciju u 15. minuti nego u 5. Bolesnici s PUJ-om imali su značajno veću koncentraciju anorganskih fosfata u slini, a koncentracije ostalih elektrolita i ukupne amilaze nisu se značajno razlikovale. **Zaključak:** Status uhranjenosti utječe na izlučivanje sline. Postoji razlika u volumenu sline pedijatrijskih bolesnika s različitim poremećajima PUJ-a. Moguće su varijacije elektrolita u slini pedijatrijskih bolesnika s PUJ-om.

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**Autorske ključne riječi:** stanje uhranjenosti; amilaze sline; elektroliti sline

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