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„Ponižavati me može ko god hoće,
ali da me ponizi može samo jedan,
a taj jedan sam ja“
Patrijarh Pavle

Prošla je godina dana od pokušaja da „paralizom života“ pobedimo virus, a uspeli smo jedino da u toj agoniji ogolimo našu stvarnost i tu borbu učinimo besmislenijom i sumanutijom. Sopstvenu svakodnevicu oslikali smo jezom, gde svaki dan započinje i završava se strahom.

Neukus kultura kiča i prostakluka izviru iz svakog segmenta opipljive pseudostvarnosti, a javni „nadriveštaci“ se jako trude da izvuku esenciju iz sopstvene neograničene praznine. Rafali pretećih rečenica bez ikakvog smisla i „šminkanje besmisla“ anatemišu svaki razuman glas koji odudara od sopstvene tvrde dogme i velikodušno nude nesavladivo beznađe kao još jedan dokaz naše apokalipse. Vlasnici zaumnih govora i grupovođe glasne monoumnosti su vidno obnevideli od besa i mržnje prema svima koji ne misle i ne vole ono što i oni. Poganim lažima i patetičnim transferom svoje nemoći unižavaju svaku slobodnu misao, a „pseudovestima“ zamajavaju i skreću pažnju sa važnih stvari i ponosno nas usmeravaju u začarani krug koloseka besmisla.

Svemogući poganog jezika i još poganijih postupaka uzrok su opšte starletizacije i estradizacije svega pa i kompletne duhovne intelektualne destrukcije. U društvenoj plimi banalnosti i atmosferi laži koja se „rukom može uhvatiti“ put u nestajanje je jedino izvestan.

Tamo gde je laž sinonim za izgovorenu reč, a istina slučajni incident, kada se u laži pogreši neophodna je ozbiljna „dekontaminacija i deratizacija“ društva.

U društvenoj hipokriziji koju živimo, borba za istinu mora biti proces koji traje i najsvetiji cilj i zadatak slobodnih, čestitih i hrabrih. Neko je jednom rekao da je najveći prijatelj istine vreme, njen najveći neprijatelj su predrasude, a njen stalni pratilac poniznost. U takvoj atmosferi je uloga univerziteta, kao centralne institucije svakog društva, upravo borba za istinu i izlazak iz peska besmisla aktuelne svakodnevice.

Akademska čestitost, znanje, odgovornost i poštovanje osnovnih zakonskih, etičkih i moralnih načela je jedini lek protiv nerealne i izvitoperene realnosti i jedina sigurna vakcina protiv dominirajuće „pandemije neistina“.

Neodustajanje od borbe za istinu, istrajnost i upornost je privilegija obrazovanih, slobodnih i hrabrih koji realnost i budućnost vide u saglasju sa sopstvenim vrednosnim okvirima baziranim na visokim moralnim, naučnim i nadasve profesionalnim kriterijumima.

Čestitost i moralni integritet učenih mora biti iskorak iz kruga drugorazrednih i neukih čiji moralni kodeksi dopiru jedino do poniznosti, poltronstva i slepe poslušnosti. Ovaj iskorak je istovremeno i beg iz intelektualne „kaljuge“ koja nedri jedino kupljene diplome i plagirane doktorate.

U maglovitom društvenom trenutku gde ne postoji ništa što nije „isprljano“ i gde se mnogo toga bazira na snazi maslačka (čitaj brzo se raspada) jedini izlaz je „deestradizacija“ sistema i eliminacija nekulture kao nametnutog modela ponašanja.

Sopstveni moralni kodeks i elementarna savest hrabrih, učenih i slobodnih u borbi za istinske vrednosti je jedina „putanja“ za spas sopstvenog života iz kandži bezizlaza koje prete našem postojanju.

Ovaj urednički komentar ću i završiti onako kako sam i počeo – citatima našeg velikog patrijarha Pavla: „Proći će sve. Samo duša, obraz i ono što je dobro ostaju večno“, jer je to paradigma vremena u kojem živimo, ali istovremeno i najbolji savet i odgovor na izazove i strahove našeg aktuelnog trenutka.

Prof. dr Slavoljub Živković

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Oral health status of young adults in Serbia – Clinical and non-clinical assessment of undergraduate students in Belgrade

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SUMMARY

Introduction Since 1987, there has been little information on oral health (OH) status of young adults aged 19 to 26 in Serbia. This study aimed to investigate the trends in OH status and OH-related quality of life (OHRQoL) of undergraduate student population in Belgrade in 2012.

Materials and methods The study included a total of 699 students of different study fields who underwent a dental examination, with 530 of them agreeing to an assisted survey and 275 forgoing a periodontal assessment. The survey consisted of six sections – basic information, socio-demographic data, oral healthcare use and habits, risk factors, OH self-assessment and OHRQoL. Clinical assessment was performed using the Decayed, Missing, Filled teeth (DMFT) index; Gingival Index; Clinical Attachment Level; Community Periodontal Index, and Plaque index. OHRQoL was evaluated through a modified OH Impact Profile (OHIP-14).

Results The mean DMFT of the examined population was 10.24 (standard deviation [SD] 5.33). Students from East and South Serbia (DMFT=8.69 [SD=4.93]), technical study field (DMFT=8.84 [SD=4.94]), with mother's having a university degree (DMFT=9.33 [SD=5.15]) and with satisfactory OH (DMFT=8.94 [SD=4.76]) were all significantly associated with lower DMFT. A significantly lower OHRQoL score was observed in students with satisfactory self-reported OH (Score=9.48 [SD=1.44]). The periodontal assessment showed no significant differences across all values of observed indexes.

Conclusion The mean DMFT of Belgrade's young adults' population has decreased from 1987 to 2012 from 12.5 to 10.4. Self-reported OH is significantly associated with both the DMFT values and OHRQoL.

Keywords: oral health status; oral health related quality of life; students; Belgrade

INTRODUCTION

Oral health represents an integral component of general health necessary for the overall physical and mental well-being of an individual [1]. Recently, The Global Burden of Disease (GBD) Study determined that as many as 2.5 billion people have been affected with untreated dental caries, while recent reports suggest the total costs due to dental disease amounted to $544.41 billion in 2015 [2, 3]. In 2017, the OECD study also indicated that spending on dental treatments in high-income countries accounted for 20% of out-of-pocket health expenditure [4]. In the wake of these data, most dental epidemiology studies have been focusing on gathering information on children and adolescents' oral health status. These form a basis for preventive policy recommendations to lower the socio-economic burden of dental disease in the nearby future, especially in terms of state financed dental care plans. That said, the oral health of some age groups, such as the one of young adults aged 19 to 26 has been unjustifiably disregarded. Certainly, this is one of the most important transition periods of a person's life, particularly seen in university students who are faced with rapid physical and social development [5]. New environments, often away from home, come with dynamic changes to their lifestyle and can have major influence on their oral and overall health [6]. There are only a handful of reports presenting data on these age groups' dental status and oral health-related quality of life (OHRQoL). In Serbia, the oral health status of student population was evaluated in 1987 as part of a more extensive oral health assessment of general population in Belgrade, when a total of 4943 of students participated in the study [7].

Since then, two different aspects could have had significant implications. Firstly, the economic collapse and wars of the 90' in former Yugoslavia were never explored in terms of the impact they had on this population's oral health status. Secondly, during the country's transition years at the beginning of the 21st century, a new Health Care Law from 2004 stopped recognizing the oral health care of students aged 19 to 26 as part of a mandatory state-financed health care plan. For the next seven years, around 250,000 students spent out-of-pocket money for their dental treatment.

This study aimed to provide new data on young adults (19 to 26 years of age) oral health status in the Republic of Serbia in 2012 and give a comprehensive overview of their OHRQoL.

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MATERIAL AND METHODS

The study comprised an assisted oral health interview, a dental status evaluation and a periodontal assessment of the participating population.

The participants in this cross-sectional study were recruited onsite between October 2011 and January 2012 at the University of Belgrade, Republic of Serbia. A total of 699 undergraduate students aged 19 to 26 gave a verbal and written informed consent and agreed to schedule a dental examination at the Department of Restorative Dentistry, School of Dental Medicine, University of Belgrade. The study population included students from the fields of medical (n=173), social (n=290), life (n=16) and technical (n=116) sciences as well as other public and private higher education institutions located in Belgrade (n=104). All students of the School of Dental Medicine, University of Belgrade were excluded from the study to avoid selection bias [8].

Of the total number of participating students, 530 agreed to an assisted oral health interview. The questionnaire used in this study comprised six sections and represented a modified version of the European Global Oral Health Indicators Development Project's (EGOHID II) "Full Standard Oral Health Interview Questionnaire for Adults" [9]. The survey was conducted at the Department of Conservative Dentistry, School of Dental Medicine, University of Belgrade before the dental status evaluation procedure. Section 1 of the questionnaire collected basic screening information (age, sex, childhood residence and faculty attended). Section 2 focused on socio-demographic variables customized for the student population – year(s) of study; parents' education status (without a University degree/ with a University degree); parents' employment status (employed/ unemployed/ inactive); means of financing the studies (by parents/ scholarship/ student loan/ self-financed/ other). Section 3 assessed the oral health care use and oral hygiene habits – last dental visit (within the past 12 months/ more than 12 months ago/ not sure); reasons of the last visit (check-up/ routine treatment/ emergency treatment/ prosthodontic reasons/ orthodontic reasons); dental care provider (private/ public/ not sure); daily tooth brushing frequency (once a day or less/ at least twice a day); toothbrush replacement frequency (monthly/ quarterly/ bi-annual/ annually); usage of fluoride-based toothpaste (yes/ no/ not sure); the use of interdental brush or floss (yes/ no). Section 4 provided data on the risk factor habits – number of food and drink meals per day (up to three times a day/ more than three times a day); sweetened beverages consumption (yes, daily or occasionally/ never/ not sure); cigarette smoking and alcohol intake frequency (yes, regularly or occasionally/ never). Section 5 gave inputs on the oral health self-assessment status (satisfactory/ unsatisfactory). Section 6 gathered data on dental anxiety presence and the OHRQoL, using a modified OHIP-14 instrument which encompassed eight items – difficulties with eating and drinking due to teeth issues; toothache and painful gums experience; bad breath experience; embarrassment because of the appearance of teeth; avoiding smiling/laughing because of the appearance of teeth; avoiding conversation with other individuals because of the appearance of teeth; difficulties with studying and other daily activities due to teeth issues; avoiding any form of social activity due to teeth issues. The frequency of each item was measured using a 4-point Likert scale ("never" = 1, "sometimes" = 2, "often" = 3, and "daily" = 4). Based on the OHRQoL students were divided into two groups – one with a low OHRQoL (at least one answer on any of the eight items was either "sometimes", "often" or "daily") and one without a low OHRQoL [10].

The maximum possible OHRQoL score in this study was 32 for the DMFT (DT – decayed teeth; MT – missing teeth; FT – filled teeth) index correlation and 28 for the GI (Gingival Index) and PI (Plaque Index) indexes. Reliability analysis based on Cronbach's alpha was initiated to determine the validity and internal consistency of the given OHRQoL survey [11]. In two instances (DMFT/ GI, PI), Cronbach's alpha for standardized items was 0.78, and 0.73, respectively. The average inter-item correlation was 0.31 (DMFT) and 0.28 (GI, PI; "bad breath" item was excluded after item correction), with no negative correlations and following the recommended threshold of 0.2 [10, 12].

Dental status evaluation

Fourth and fifth-year dental students performed the clinical dental status evaluation with a senior clinician's verification at the Department of Restorative Dentistry. A total of 699 participants were examined. The assessment followed the standard World Health Organization (WHO) Guidelines and the Oral Health Clinical Surveys Guidelines of the EGOHID II project [13, 14]. The DMFT index was used to measure dental caries experience and a "Classification of Lesions of the Exposed Tooth Surfaces" system proposed by Mount et al. to record the tooth sites affected by caries (Site 1 – pits, fissures and minor defects on exposed enamel surfaces of all teeth; Site 2 – approximal enamel surfaces immediately cervical to the contact area between any pair of adjacent teeth; Site 3 – the cervical one-third of the crown around the full circumference of any tooth or, following a gingival recession, the exposed root surface) [15]. All the assessments were completed visually and using a standard dental explorer and mirror.

Periodontal assessment

Fifth-year dental students performed the periodontal assessment with a senior periodontist's verification at the Department of Periodontology and Oral Medicine, School of Dental Medicine, University of Belgrade. A total of 275 participants were examined. The assessment followed the standard WHO Guidelines and the Oral Health Clinical Surveys Guidelines of the (EGOHID II) project [13, 14]. The evaluation included the identification of changes to soft gingival tissues – gingivitis (yes, type of gingivitis – catarrhal, acute ulcero-necrotic, hyperplastic, gingival fibromatosis, desquamative/ no) and periodontal disease presence (yes, type of periodontitis – chronic periodontitis, acute periodontitis, necrotizing periodontitis, periodontitis as a manifestation of systemic diseases/ no). Three indices were used to determine the state of periodontal

tissue – the Loe & Silness Gingival Index – GI; the clinical attachment level – CAL; a community periodontal index – CPI, modified for assessing periodontal pocket depth if present. All measurements were taken on six teeth (code numbers 16, 12, 24, 36, 32, 44), on four surfaces (mesial, distal, lingual and buccal) and with a WHO Community Periodontal Index probe. Additionally, the Silness & Loe Plaque Index - PI was used to record the position and the amount of dental plaque present in gingival and subgingival areas of examined teeth.

Statistical Analysis

The DMFT, GI, CAL, CPI and PI indexes are shown in their mean and standard deviation values accordingly. The Mann-Whitney U test was used to compare data between two and the Kruskal-Wallis test to compare more than two groups of students and in compliance with the interview survey results. *P-values* <0.05 were considered statistically significant. The chi-square and Fisher's exact tests were applied to compare the proportion of decayed, filled, extracted and healthy teeth across two or more groups of students and in compliance with the interview survey results. *P-values*<0.05 were considered statistically significant. All the statistical analyses were carried out with the XLSTAT statistical software Trial Version 2021.1.1.1082 (Addinsoft, Paris, France, EU).

RESULTS

As shown in Table 1, the mean DMFT index of the total number of participating students (n=699) was 10.24 (standard deviation [SD] 5.33; range = 0-31). There were no significant differences in DMFT values between female and male students in terms of socio-demographic aspects covered in this study, while obvious ones could be seen across other observed variables (Table 2). The lowest mean DMFT was seen in students originally from the region of East and South Serbia. Mean DMFT values did not differ significantly between Belgrade, West and Central Serbia and other regions and countries. Students studying technical sciences had a significantly lower mean DMFT index, followed closely by medical field students. Life science students and ones from other higher education institutions were impacted by the highest values of the DMFT index. Mother's education level was strongly associated with the DMFT score, with lower scores found among students whose mother possessed a university degree.

All the variables used to determine students' oral health behavior showed no significant differences between their mean DMFT values, except for the time of the last dental visit. Students who visited a dental professional less than a year ago had a higher mean DMFT of 10.61 (SD 5.11) compared to the ones who did it more than a year ago – 8.64 (SD 4.83) The results presented in Table 3 also showed no significant differences in DMFT values of students receiving dental care from private dental professionals opposite to the ones receiving it from dental professionals working in public clinics.

Table 1. Mean DMFT value
Tabela 1. Srednje vrednosti KIP indeksa

Index	N broj	Mean (SD) Srednja vrednost (SD)
DMFT KIP indeks	699	10.24 (5.33)

Table 2. Student's socio-demographic context of the mean DMFT index values
Tabela 2. Sociodemografske karakterisitke studentske populacije u funkciji KIP-a

Variable Promenljiva	n (%) broj	DMFT mean (SD) KIP (SD)	p
Sex/Pol			
Male/muški	185 (34.9)	10.07 (5.18)	0.994
Female/ženski	344 (65.1)	9.98 (5.07)	
Region/Regija			
Belgrade/Beograd	223 (42.1)	10.26 (4.95)	< 0.05 (0.046)
West and Central Serbia Zapadna i Centralna Srbija	152 (28.7)	10.29 (5.39)	
East and South Serbia Istočna i Južna Srbija	89 (16.8)	8.69 (4.93)	
Other Ostalo	66 (12.4)	10.29 (4.91)	
Study field / Oblast studiranja			
Medical / Medicinske nauke	151 (28.5)	9.67 (5.01)	< 0.05 (0.027)
Social / Društvene nauke	231 (43.6)	10.29 (5.09)	
Life / Prirodne nauke	11 (2.1)	11.00 (4.27)	
Technical / Tehničke nauke	82 (15.5)	8.84 (4.94)	
Other/Ostalo	55 (10.3)	11.33 (5.46)	
Mother's education / Obrazovanje majke			
<University degree <Fakultetsko	333 (62.8)	10.41 (5.03)	< 0.05 (0.013)
University degree Fakultetsko	197 (37.2)	9.33 (5.15)	

Table 3. The impact of student's oral health behaviour on their mean DMFT index values
Tabela 3. Uticaj odnosa studenata prema oralnom zdravlju na vrednosti KIP-a

Variable Promenljiva	n (%) broj	DMFT mean (SD) KIP (SD)	p
Last dental visit / Poslednja poseta stomatologu			
< 1 year / < 1 godine	353 (70.5)	10.61 (5.11)	< 0.001
> 1 year / >1 godine	148 (29.5)	8.64 (4.83)	
Daily tooth brushing / Svakodnevno pranje zuba			
Once a day or less Jednom dnevno i manje	49 (9.4)	10.06 (5.12)	0.981
More than once a day Više od jednom dnevno	475 (90.6)	9.98 (5.09)	
Interdental brushing / Interdentalno pranje zuba			
Regularly/Occasionally Redovno/Ponekad	286 (54.8)	10.25 (5.28)	0.161
Never/Nikada	236 (45.2)	9.7 (4.87)	
Use of fluoride tooth paste / Upotreba fluoridne paste za zube			
Yes/Da	392 (75.7)	10.04 (5.13)	0.841
No / Not sure / Ne / Nisam siguran(a)	126 (24.3)	10.15 (5.06)	
Dental care / Stomatološka nega			
Private/ Privatni stomatolog	205 (59.9)	11.03 (5.2)	0.067
Public/ Državni stomatolog	137 (40.1)	10.07 (5.01)	

When looking at various risk factor habits presented in Table 4, only the alcohol intake significantly influenced the mean DMFT of students. The students who consumed alcohol on a daily/occasional level had a significantly lower DMFT index than those who stated never to drink alcohol (9.71 & 10.69).

Students who deemed their oral health satisfactory had a lower DMFT of 8.94 (SD 4.76) than those unsatisfied with it – DMFT of 12.13 (SD 5.18). The same trend could be seen when comparing the average values of the OHRQoL score – satisfied students with a score of 9.48 (SD 1.44) and unsatisfied ones with a mean score of 11.11 (SD 2.85). Students without low OHRQoL showed no clear advantages to the ones with reported low OHRQoL. The same pattern was visible in students who confirmed they suffered from dental anxiety compared to those who did not (Table 5).

Table 4. Risk habit determinants of the student's mean DMFT
Tabela 4. Faktori rizika i vrednost KIP-a kod studenata u Beogradu

Variable Promenljiva	n (%) broj	DMFT mean (SD) KIP (SD)	p
Daily intake of food and drinks / Dnevni unos hrane i pića			
Up to three times / Do tri puta	146 (28.6)	9.31 (4.76)	0.063
> 3 times / > 3 puta	365 (71.4)	10.32 (5.18)	
Sweetened beverages intake / Unos zaslađenih pića			
Daily/Svakodnevno	183 (34.9)	9.99 (4.88)	0.672
Occasionally/Ponekad	248 (47.3)	10.10 (5.02)	
Never/Nikada	93 (17.8)	9.67 (5.70)	
Smoking / Pušenje			
Daily/Occasionally Svakodnevno/Ponekad	96 (19.0)	9.54 (4.82)	0.382
Never/Nikada	410 (81.0)	10.19 (5.12)	
Alcohol / Alkohol			
Daily/Occasionally Svakodnevno/Ponekad	345 (67.5)	9.71 (5.09)	< 0.05 (0.028)
Never/Nikada	166 (32.5)	10.69 (5.04)	

Table 5. DMFT index values in terms of self-assessed oral health, dental anxiety and OHRQoL
Tabela 5. Vrednosti KIP, GI and PI indeksa u odnosu na samoprocenu oralnog zdravlja, strah od odlaska stomatologu i vrednost KŽPsOZ-a

Variable Promenljiva	n (%) broj	DMFT mean (SD) KIP (SD)	p
Self-assessed oral health / Samoprocena oralnog zdravlja			
Satisfactory Zadovoljavajuće	338 (66.3)	8.94 (4.76)	< 0.001
Unsatisfactory Nezadovoljavajuće	172 (33.7)	12.13 (5.18)	
Dental anxiety / Strah od odlaska stomatologu			
Yes/Da	158 (31.0)	10.24 (5.12)	0.608
No/Ne	351 (69.0)	9.92 (5.08)	
OHRQoL / KŽPsOZ			
With Low / Sa niskom	131 (24.9)	9.39 (5.41)	0.088
Without Low / Bez niskog	396 (75.1)	10.20 (4.99)	
OHRQoL mean (SD) / KŽPsOZ srednja vrednost (SD)			
Self-assessed oral health / Samoprocena oralnog zdravlja			
Satisfactory Zadovoljavajuće	338 (66.3)	9.48 (1.44)	< 0.001
Unsatisfactory Nezadovoljavajuće	172 (33.7)	11.11 (2.85)	

Table 6 depicts data on the status (decayed, filled, extracted, healthy) of as many as 14,840 teeth (530 × 28) evaluated in context of several questionnaire aspects – sex (529 × 28); region (530 × 28); study field (530 × 28); mother's education (530 × 28); self-assessed oral health (510 × 28); last dental visit (501× 28) and dental anxiety (509 × 28). Significant differences are seen in almost every percentage comparison of decayed, filled, extracted and healthy teeth. Female and male students had approximately the same proportion of healthy and extracted teeth (≈68%; ≈3%), with the female ones having significantly more filled and significantly less decayed teeth. Medical students reported the lowest number of decayed teeth, while the same could be said of students originally from East and South Serbia. Greatest differences were seen in numbers of decayed and healthy teeth in groups of students who perceived their oral health as satisfactory opposite to those perceiving it as unsatisfactory. There were also no significant differences between the proportions of filled teeth in students whose mothers had a university degree and those whose mothers did not.

Among the teeth most affected, the first and second molars in both the upper and lower jaw had the highest frequency (percentage) of dental caries (Figure 1a). The least affected teeth were the ones in the lower jaw's inter-canine region (Figure 1b). On the other hand, the teeth surfaces having dental caries were more evenly distributed across different tooth groups (Figure 2). The prevalence of site one lesions was the highest in molars, while the site two lesions dominated in premolars and the upper jaw's inter-canine region. Site three lesions were featured mostly in lower jaw premolars.

Based on the periodontal assessment of 275 students, gingivitis was identified in 99 (36%) and periodontal disease in 20 (7.3%) of them. The most prominent type of gingivitis was the catarrhal one (Figure 3a). Localized chronic periodontitis was the most prevalent one in terms of periodontal disease presence (Figure 3b). As displayed in Table 7, the mean values of all analyzed periodontal indexes were under 1.0 (n = 275).

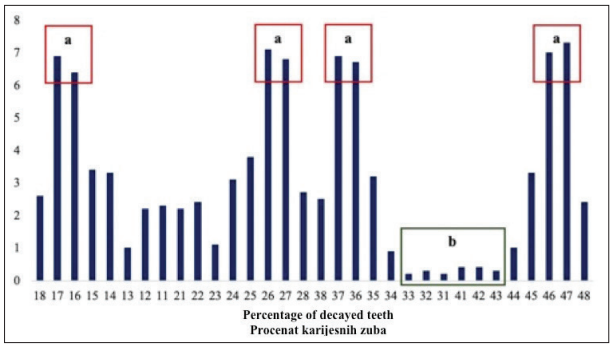

Figure 1. Percentages of decayed teeth per tooth group: a) highest percentages of decayed teeth, b) lowest percentages of decayed teeth.
Numbers on the x-axis are a representation of the standard human dentition code system
Slika 1. Procenti karijesnih zuba: a) najviši procenat karijesnih zuba, b) najniži procenat karijesnih zuba.
Brojevi na x-osi predstavljaju šifre 32 zuba prisutna u humanoj denticiji.

Table 6. Proportion of decayed, filled, extracted and healthy teeth across different questionnaire aspects
Tabela 6. Zastupljenost karijesnih, plombiranih, ekstrahovanih i zdravih zuba prema različitim aspektima u okviru predloženog upitnika

Variable Promenljiva	n (%) DT n (%) K	n (%) FT n (%) P	n (%) ET n (%) E	n (%) HT n (%) Z
Sex/Pol (X^2 p < 0.01)				
Male/Muški	686 (13.24)	866 (16.72)	150 (2.9)	3478 (67.14)
Female/Ženski	971 (10.08)	1784 (18.52)	301 (3.13)	6576 (68.27)
p	< 0.01	< 0.01	0.452	0.161
Region/Regija (X^2 p < 0.01)				
Belgrade/Beograd	690 (11.05)	870 (13.93)	140 (2.24)	4544 (72.77)
West and Central Serbia / Zapadna i Centralna Srbija	442 (10.39)	816 (19.17)	107 (2.51)	2891 (67.93)
East and South Serbia / Istočna i Južna Srbija	248 (9.95)	362 (14.53)	40 (1.61)	1842 (73.92)
Other/Ostalo	246 (13.31)	277 (14.99)	29 (1.57)	1296 (70.13)
p	< 0.05	< 0.01	< 0.05	< 0.05
Study field / Oblast studiranja (X^2 p < 0.01)				
Medical / Medicinske nauke	446 (10.55)	779 (18.42)	107 (2.53)	2896 (68.5)
Social / Društvene nauke	770 (11.9)	1215 (18.78)	245 (3.79)	4238 (65.52)
Life / Prirodne nauke	38 (12.34)	70 (22.73)	9 (2.92)	191 (62.01)
Technical / Tehničke nauke	253 (11.02)	393 (17.12)	48 (2.09)	1602 (69.77)
Other/Ostalo	192 (12.47)	247 (16.04)	54 (3.51)	1047 (67.99)
p	< 0.05	< 0.05	< 0.01	< 0.05
Mother's education / Obrazovanje majke (X^2 p < 0.01)				
<University degree/ <Fakultetsko	1117 (11.98)	1662 (17.82)	311 (3.34)	6234 (66.86)
University degree/ Fakultetsko/	523 (9.48)	960 (17.4)	135 (2.45)	3898 (70.67)
p	< 0.01	0.519	< 0.05	< 0.01
Self-assessed oral health / Samoprocena oralnog zdravlja (X^2 p < 0.01)				
Satisfactory/Zadovoljavajuće	821 (8.67)	1631 (17.23)	187 (1.98)	6825 (72.12)
Unsatisfactory/Nezadovoljavajuće	779 (16.18)	927 (19.25)	154 (3.2)	2956 (61.38)
p	<0.01	<0.01	<0.01	<0.01
Last dental visit / Poslednja poseta stomatologu (X^2 p < 0.01)				
< 1 year / < 1 godine	1062 (10.47)	1948 (19.71)	282 (2.85)	6592 (66.69)
> 1 year / > 1 godine	508 (12.26)	583 (14.07)	113 (2.73)	2940 (70.95)
p	< 0.05	< 0.01	0.696	< 0.01
Dental anxiety/ Strah od odlaska stomatologu (X^2 p < 0.01)				
Yes/Da	562 (12.7)	716 (16.18)	151 (3.41)	2995 (67.7)
No/Ne	1018 (10.36)	1794 (18.25)	246 (2.5)	6770 (68.88)
p	< 0.01	< 0.01	< 0.01	0.161

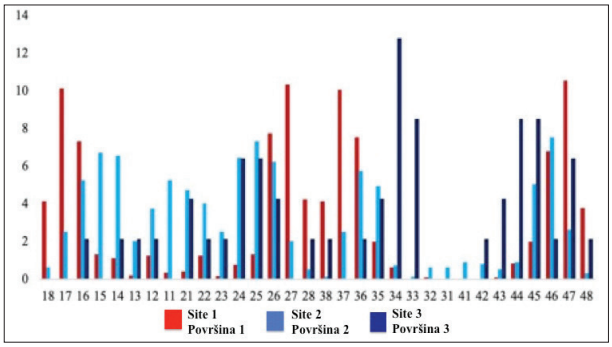

Figure 2. Distribution of caries lesion sites across the whole dentition (%).
Site 1 – pits, fissures and minor defects on exposed enamel surfaces of all teeth; Site 2 – approximal enamel surfaces immediately cervical to the contact area between any pair of adjacent teeth; Site 3 – the cervical one-third of the crown around the full circumference of any tooth or, following gingival recession, the exposed root surface. Numbers on the x-axis are a representation of the standard human dentition code system
Slika 2. Distribucija površina zuba zahvaćenih karijesom (%).
Površina 1 – jamice, fisure i okluzalne površine bočnih zuba; površina 2 – aproksimalne površine zuba; površina 3 – gingivalna trećina zuba. Brojevi na x-osi predstavljaju šifre 32 zuba prisutna u humanoj denticiji.

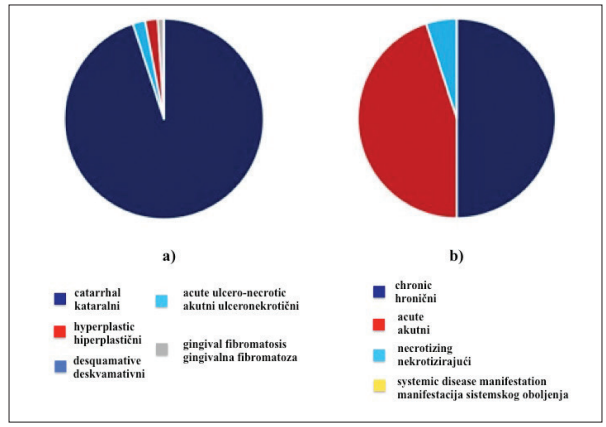

Figure 3. Results of periodontal assessment: a) distribution of gingivitis per type, b) periodontal disease distribution per type
Slika 3. Rezultati procene stanja parodoncijuma: a) distribucija različitih tipova gingivitisa, b) distribucija različitih tipova parodontopatija

Table 7. Mean of the GI, CAL, CPI and PI indexes for the total population of students
Tabela 7. Srednje vrednosti GI, NPE, CPI i PI indeksa za ukupnu populaciju učesnika

Index Indeks	n	Mean (SD) Srednja vrednost (SD)
GI GI	275	0.51 (0.5)
CAL NPE	275	0.14 (0.38)
CPI* CPI*	275	0.26 (0.6)
PI PI	275	0.55 (0.47)

Table 8. GI and PI index values in terms of self-assessed oral health, dental anxiety and OHRQoL
Tabela 8. Vrednosti KIP, GI and PI indeksa u odnosu na samoprocenu oralnog zdravlja, strah od odlaska stomatologu i vrednost KŽPsOZ-a

Variable Promenljiva	n (%) broj	GI mean (SD) GI (SD)	p
Self-assessed oral health/ Samoprocena oralnog zdravlja			
Satisfactory/Zadovoljavajuće	93 (62.4)	0.47 (0.56)	0.055
Unsatisfactory/Nezadovoljavajuće	56 (37.6)	0.61 (0.53)	
Dental anxiety / Strah od odlaska stomatologu			
Yes/Da	37 (25)	0.6 (0.59)	0.264
No/Ne	111 (75)	0.49 (0.53)	
OHRQoL / KŽPsOZ			
With Low / Sa niskim	101 (65.2)	0.54 (0.54)	0.257
Without Low / Bez niskog	54 (34.8)	0.46 (0.54)	
PI mean (SD) / PI (SD)			
Self-assessed oral health/ Samoprocena oralnog zdravlja			
Satisfactory/Zadovoljavajuće	93 (62.4)	0.56 (0.51)	0.300
Unsatisfactory/Nezadovoljavajuće	56 (37.6)	0.60 (0.43)	
Dental anxiety / Strah od odlaska stomatologu			
Yes/Da	37 (25)	0.6 (0.51)	0.818
No/Ne	111 (75)	0.56 (0.47)	
OHRQoL / KŽPsOZ			
With Low / Sa niskim	101 (65.2)	0.59 (0.47)	0.308
Without Low / Bez niskog	54 (34.8)	0.53 (0.49)	

No significant differences were found between the effect of self-assessed oral health, dental anxiety, and the OHRQoL on periodontal health of participating students (Table 8).

DISCUSSION

Mean DMFT score of the student population examined in our study was above 10 (10.24). The presented findings indicate that the lower average DMFT index value correlates to students from East and South Serbia, the technical study field, whose mothers possess a university degree, students who drink alcohol regularly and regard their oral health as satisfactory. Significantly higher DMFT index was seen in students who made a dental visit during the last 12 months. The nature of the utilized dental care, private or public, seems to play a minor role. Opposite to the proportion of decayed or filled teeth, sex and dental anxiety did not have a significant impact. Tooth brushing, interdental cleaning, use of fluoride toothpaste, sweetened beverages consumption, smoking and OHRQoL appear to have little effect on the students' DMFT index values. As a parameter, a lower OHRQoL score that leans toward the absence of OHRQoL issues corresponds tightly with a satisfactory view students have of their oral health. On the other hand, the proportion of decayed, filled, extracted and healthy teeth in the examined population and its relation to the questionnaire offers a somewhat different perspective. Lower proportion of decayed teeth is strongly related to female students, students originally from East and South Serbia and the ones studying medical sciences. The same could be said of those whose mothers have higher level of education (university degree), who have visited dentist's office in the past 12 months, do not experience dental anxiety and students who perceive their oral health as satisfactory. It is also apparent that female students, together with those who visited dentist more regularly and did not suffer from dental anxiety, had a significantly higher percentage of filled teeth. When it comes to teeth and tooth surfaces mostly affected by caries, this study's results follow conventional trends [16]. Highest caries prevalence is seen on occlusal surfaces of molars and approximal surfaces of incisors, canines and premolars, especially in the upper jaw. The cervical one-third of canines and premolars of the lower jaw is also the surface significantly affected. In terms of periodontal disease, only a handful of students have been diagnosed with some level of periodontitis. At the same time, the average values of GI, CAL, CPI and PI indices well below one did not indicate severe periodontal issues in the examined population.

The average DMFT index value of the total number of participants in this study was higher than the same score in similar studies. The average DMFT score of 10.24 (SD 5.33) was closest to the DMFT value of undergraduate students of dentistry and medicine in Russia – 7.46 (SD 4.43) followed by a score reported in Korean students – mean DMFT of 6.1 (SD 4.0) [10, 17]. When only the DMFT values of medical students from this study were analyzed, the differences became less apparent (DMFT=9.67 [SD 5.01]). Compared to the population of 30-year-olds in Adelaide, Australia (DMFT=2.1), 18-year-olds in Hong Kong, China (DMFT=1.4 [SD 1.8]) and first-year students in Okayama, Japan (DMFT=2.01 [SD 2.88]) the values were significantly higher [18, 19, 20]. It is also worth mentioning that the number of filled teeth in these studies accounts for most of the DMFT score (80-90%) while in the current study that is not the case – around 60% [10, 19]. Such finding indicates that the undergraduate students in Belgrade have more active caries lesions and extractions than their fellow students elsewhere.

Additionally, Russian students differ significantly between groups of low and without low OHRQoL, which is not typical in Belgrade students (Table 6) [10]. In that sense, undergraduate students in Belgrade were more similar to the 19-year-old Swedes [21]. Average GI index of Belgrade students was almost twice the value reported for Northwest Russian area (0.51 and 0.27) [10]. However, neither value surpassed the score of one, indicating low gum inflammation in both cases.

Compared to the 1987 oral health assessment study, the mean DMFT had decreased from 12.5 in 1987 to 10.2 in

2012. In terms of the percentage of teeth affected by caries, the number had increased from 31% in 1987 to 38% in 2012 [7]. It is not yet clear if this is due to economic reasons, ineffective childhood prevention programs, regulatory issues or the individual-level factors. Further, the current study results advocate that the higher the students' mother education, the more healthy teeth are present. The result also falls in line with suggestions that higher level of mother's education and mother's oral health positively influences oral health during childhood with a prolonged effect [22]. The proportion of filled teeth has also increased from 46.4% to 54.5%, while the percentage of extracted teeth has seen a large decrease from 22.4% to 7.4% [7]. That potentially indicates a move from surgical to more conservative approach in dental treatment of patients from this population during the period between the two studies.

To our knowledge, this is the first study since 1987 in Belgrade and Serbia that gives a comprehensive and detailed overview of the oral health status of students aged 19 to 26. In contrast to most of the other studies, it excluded dental students to bring more generalization to the findings and included various groups of students with different backgrounds and divergent study fields. It also provided a sound basis for further investigation on what effect OHRQoL possibly has on clinical determinants of oral health.

The OHRQoL section of the questionnaire contained only eight questions that related to the original OHIP-14 instrument. Although the internal consistency of the applied OHRQoL survey is acceptable (Cronbach's alpha>0.7 [0.778;0.703]) this is still well below the levels that were higher than 0.85 in other studies that applied the original OHIP-14 concept [10, 19]. The other limitation is the fact that dental students performed dental status and periodontal assessments. Besides the senior clinicians' immediate supervision, it remains unclear if the examination quality was uniformly desirable.

CONCLUSION

This study's findings suggest a decrease in the average DMFT of young adults during the last 30 years and a rise in the percentage of decayed teeth on a population level. It is also clear that students in Belgrade had worse overall oral health than their peers from Russia, China, Japan or Sweden. Furthermore, it is indicative that the student's self-awareness on the topic of oral health could have a significant influence on some of the clinical variables examined in this study. Further research is necessary on specific socio-economic and loco-regional determinants that can potentially explain oral health discrepancies between the students from different regions observed in this study. Future studies should also focus on multi-year prospective research into changes that potentially occur over time to the same participants. That is why more active inclusion of private and public dental professionals backed by governing bodies and effective regulation is an essential aspect of a reliable and successful community approach to the nation's wellbeing in terms of oral health.

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Conflict of interest: No conflict of interest.

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Oralno zdravlje mladih u Srbiji – kliničke i nekliničke determinante kod studenata osnovnih studija u Beogradu

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KRATAK SADRŽAJ

Uvod U Srbiji je malo informacija o statusu oralnog zdravlja (OZ) mladih od 19 do 26 godina. Ova studija imala je za cilj da istraži trendove u statusu OZ-a i kvalitetu života povezanom sa oralnim zdravljem (KŽPsOZ) u populaciji studenata osnovnih studija u 2012. godini u Beogradu.

Materijal i metode Studija je obuhvatila ukupno 699 studenata sa različitih fakulteta kod kojih je analiziran status zuba. Njih 530 je uzelo učešće u anketi o oralnom zdravlju, a kod 275 je evaluiran parodontalni status. Anketni upitnik sastojao se iz šest odeljaka – osnovnih ličnih informacija, sociodemografskih podataka, oralnozdravstvenih navika i korišćenja stomatološke službe, faktora rizika, samoprocene OZ-a i KŽPsOZ-a. Klinička procena izvršena je korišćenjem prosečnog karijes indeksa (KIP); gingivalnog indeksa; nivoa pripojnog epitela; parodontalnog i plak indeksa. Procena KŽPsOZ-a sprovedena je kroz primenu modifikovanog OHIP-14 instrumenta.

Rezultati Vrednost KIP-a ispitivane populacije iznosila je 10,24. Kod studenata iz Istočne i Južne Srbije ove vrednosti su bile nešto manje (KIP = 8,69), kao i kod grupacije fakulteta tehničkih nauka (KIP = 8,84), kod onih čija majka poseduje fakultetsku diplomu (KIP = 9,33), odnosno onih koji svoje OZ-e smatraju zadovoljavajućim (KIP = 8,94). Značajno niža vrednost KŽPsOZ-a uočena je kod studenata sa zadovoljavajućom procenom OZ-a (KIP = 9,48). Procena zdravlja potpornog aparata zuba nije pokazala značajne razlike u vrednostima analiziranih indeksa.

Zaključak Vrednost KIP-a u populaciji mladih u Beogradu bila je manja u periodu između 1987. i 2012. godine (sa 12,5 na 10,4). Samoprocena OZ-a značajno korelira sa vrednostima KIP-a i KŽPsOZ-om.

Ključne reči: status oralnog zdravlja; kvalitet života povezan sa oralnim zdravljem; studenti

UVOD

Oralno zdravlje predstavlja sastavni deo opšteg zdravlja neophodnog za celokupno fizičko i mentalno blagostanje pojedinca [1]. Studija globalnog tereta bolesti (GBD) iz 2015. godine utvrdila je da je čak 2,5 milijarde ljudi pogođeno nelečenim zubnim karijesom, a nedavni izveštaji sugerišu da ukupni troškovi zbog bolesti zuba za 2015. godinu iznose 544,41 milijarde dolara [2, 3]. Studija OECD-a iz 2017. godine je takođe pokazala da su troškovi stomatološkog lečenja u zemljama sa visokim prihodima iznosili 20% od ukupnih zdravstvenih troškova finansiranim od samih građana [4]. Na osnovu postojećih podataka, uočeno je da je većina epidemioloških studija fokusirana na prikupljanje informacija o oralnom zdravstvenom stanju dece i adolescenata. Ovi podaci su ujedno i osnova za preporuke preventivne politike za smanjenje socijalno-ekonomskog tereta dentalnih oboljenja u bliskoj budućnosti, a posebno sa stanovišta javnozdravstvenih izdataka. Međutim, neke starosne grupe, poput mladih od 19 do 26 godina, u tom procesu ostale su manje ili više zapostavljene. Svakako, ovo je jedan od najvažnijih prelaznih perioda u čovekovom životu, posebno izražen kod studenata univerziteta suočenih sa dinamičnim fizičkim i socijalnim promenama [5]. Nova okruženja, često daleko od kuće, imaju za posledicu novi način života i mogu imati velikog uticaja na oralno i celokupno zdravlje [6]. U stručnoj literaturi samo je nekoliko epidemioloških studija u ovoj oblasti koje prikazuju podatke o zubnom statusu navedene populacije i kvalitetu života povezanih sa njihovim oralnim zdravljem (skr. KŽPsOZ). U Republici Srbiji poslednji put je oralno zdravstveno stanje navedene starosne grupe procenjivano 1987. godine, u okviru opsežnije studije oralnog zdravlja stanovništva u Beogradu, a analiza je obuhvatila ukupno 4943 ispitanika navedene starosne dobi [7].

U periodu od poslednje studije dva aspekta su od potencijalno velikog značaja. Stručnoj javnosti nisu dostupni podaci o uticaju ekonomskog kolapsa i ratova 90-ih u bivšoj Jugoslaviji na oralno zdravlje obuhvaćene ciljne grupe. Tokom tranzicionih godina u Srbiji početkom 21. veka, uredbama Zakona o zdravstvenoj zaštiti iz 2004. godine, studentima uzrasta od 19 do 26 godina nije priznato pravo na stomatološku zdravstvenu zaštitu kao deo obaveznog zdravstvenog plana finansiranog iz državnog budžeta. Sledećih sedam godina oko 250000 studenta troškove stomatoloških usluga finansiralo je sopstvenim prihodima.

Cilj ovog istraživanja bio je da se utvrdi status oralnog zdravlja studenata Beogradskog univerziteta od 19 do 26 godina u Republici Srbiji za 2012. godinu i analizira sveobuhvatan pregled kvaliteta života povezanog sa oralnim zdravljem.

MATERIJAL I METODE

Studija je uključila anketu o oralnom zdravlju, procenu statusa zuba i stanje potpornog aparata zuba.

Učesnici u ovoj studiji bili su studenti na fakultetima Univerziteta u Beogradu između oktobra 2011. i januara 2012. godine. Ukupno 699 studenata osnovnih studija uzrasta od 19 do 26 godina dalo je usmenu i pismenu saglasnost i pristalo na stomatološki pregled na Klinici za bolesti zuba Stomatološkog fakulteta Univerziteta u Beogradu. Populacija ispitanika obuhvatila je studente iz oblasti medicinskih (n = 173), društvenih (n = 290), prirodnih (n = 16) i tehničkih (n = 116) nauka, kao i drugih javnih i privatnih visokoškolskih ustanova smeštenih u Beogradu, Srbija (n = 104). Svi studenti Stomatološkog fakulteta Univerziteta u Beogradu bili su isključeni iz studije kako bi se izbegla pristrasnost u izboru [8].

Od ukupnog broja studenta koji su učestvovali, 530 je pristalo na anketu o oralnom zdravlju. Upitnik korišćen u ovoj studiji sastojao se od šest odeljaka i predstavljao je izmenjenu verziju „Standardnog upitnika za oralno zdravlje odraslih“ evropskog

projekta razvoja globalnih indikatora oralnog zdravlja – EGOHID II [9]. Anketa je sprovedena na Klinici za Bolesti zuba Stomatološkog fakulteta Univerziteta u Beogradu pre sprovođenja stomatološkog pregleda. U Odeljku 1 upitnika prikupljene su osnovne informacije o učesnicima studije – starost, pol, mesto rođenja i pohađani fakulteti (medicinske nauke / društvene nauke / prirodne nauke / tehničke nauke / ostalo [druge visokoškolske ustanove u Beogradu]). Odeljak 2 se fokusirao na sociodemografske karakteristike prilagođene studentskoj populaciji – godine studija; nivo obrazovanja roditelja (bez fakultetske diplome – visoka i viša škola / sa fakultetskom diplomom); radni status roditelja (zaposleni / nezaposleni / neaktivni); sredstva za finansiranje studija (roditelji / stipendije / studentski kredit / samofinansiranje / drugo). U Odeljku 3 fokus je postavljen na procenu korišćenja stomatološke zdravstvene zaštite kao i oralnohigijenskih navika studenata – poslednja poseta stomatologu (u poslednjih 12 meseci / pre više od 12 meseci / nisam siguran); razlozi poslednje posete (kontrola / rutinsko lečenje / hitno lečenje / protetski / ortodontski razlozi); pružalac stomatološke zaštite (privatni sttomatolog / državna služba / nisam siguran); učestalost pranja zuba (jednom dnevno ili manje / najmanje dva puta dnevno); učestalost zamene četkica za zube (mesečno / tromesečno / dvogodišnje / godišnje); upotreba paste za zube na bazi fluorida (da / ne / nije siguran/a); upotreba interdentalne četkice ili konca (da / ne). Odeljak 4 je uključivao podatke o prisustvu loših navika a u vezi sa potvrđenim faktorima rizika – broj obroka (hrana i piće) dnevno (do tri puta dnevno / više od tri puta dnevno); konzumacija zaslađenih pića (da, svakodnevno ili povremeno / nikad / nisam siguran/a); pušenje cigareta i konzumacija alkohola (da, redovno ili povremeno / nikada). Odeljak 5 pružio je podatke o samoprocenjivanju oralnog zdravlja (zadovoljavajuće / nezadovoljavajuće). U Odeljku 6 su bili podaci o prisustvu straha od odlaska stomatologu i KŽPsOZ-a, korišćenjem modifikovanog OHIP-14 instrumenta koji je obuhvatio osam aspekata – poteškoće sa hranom i pićem zbog problema sa zubima; zubobolja i bolne desni; iskustvo neprijatnog zadaha; osećaj sramote zbog izgleda zuba; izbegavanje smejanja / smeha zbog izgleda zuba; izbegavanje razgovora sa drugim osobama zbog izgleda zuba; poteškoće sa učenjem i drugim svakodnevnim aktivnostima zbog problema sa zubima; izbegavanje bilo kog oblika društvene aktivnosti zbog problema sa zubima. Učestalost svake stavke izmerena je pomoću bodovanja u okviru Likertove skale („nikada" = 1, „ponekad" = 2, „često" = 3 i „svakodnevno" = 4). Studenti koji su koristili mogućnost odgovora nisam siguran(a) / ne znam bili su isključeni iz daljih analiza. Na osnovu odgovora u okviru procene KŽPsOZ-a studenti su podeljeni u dve grupe – prvu sa niskim KŽPsOZ-om (najmanje jedan odgovor na bilo koju od osam stavki bio je „ponekad", „često" ili „svakodnevno") i drugu bez niskog KŽPsOZ-a [10].

Maksimalni mogući rezultat KŽPsOZ-a u ovoj studiji bio je 32 za korelaciju sa KIP indeksom i 28 za gingivalni i indeks plaka. Analiza pouzdanosti zasnovana na Kronbahovoj alfi pokrenuta je da bi se utvrdila validnost i unutrašnja konzistentnost navedene KŽPsOZ ankete [11]. U dva slučaja (KIP / GI, PI), Kronbahova alfa za standardizovane stavke iznosila je 0,78, odnosno 0,73. Prosečna korelacija među stavkama iznosila je 0,31 (KIP) i 0,28 (GI, PI; stavka „neprijatan zadah" je isključena iz analize nakon neophodne korekcije). Negativnih korelacija nije bilo i obezbeđena je minimalno prihvatljiva vrednost korelacije ponuđenih stavki od 0,2 [10, 12].

Kliničku procenu zubnog statusa sproveli su studenti četvrte i pete godine stomatologije uz verifikaciju iskusnijeg kliničkog lekara na Klinici za bolesti zuba Stomatološkog fakulteta Univerziteta u Beogradu. Pregledano je 699 ispitanika. Procena je sledila standardne smernice Svetske zdravstvene organizacije (SZO) i smernice za kliničko ispitivanje oralnog zdravlja projekta EGOHID II [13, 14]. KIP indeks (prosečna vrednost karijesnih, plombiranih i ekstahovanih zuba kod pregledanih osoba) korišćen je za merenje iskustva zubnog karijesa populacije, a „Klasifikacija lezija izloženih površina zuba" koju su predložili Mount i saradnici za identifikovanje površina zuba zahvaćenih karijesom (Površina 1 – jamice, fisure i okluzalne površine bočnih zuba; Površina 2 – aproksimalne površine zuba; Površina 3 – gingivalna trećina zuba) [15]. Sve procene su sprovedene vizuelnom inspekcijom i sondiranjem (korišćenjem ogledalca i standardnih stomatoloških sondi).

Procenu stanja potpornog aparata zuba sproveli su studenti pete godine stomatologije uz verifikaciju kliničkog parodontologa na Klinici za parodontologiju i oralnu medicinu Stomatološkog fakulteta Univerziteta u Beogradu. Parodontološku procenu prošlo je 275 ispitanika. Ispitivanje je sledilo standardne smernice SZO i smernice za kliničko ispitivanje oralnog zdravlja projekta EGOHID II [13, 14]. Evaluacija je obuhvatala identifikaciju promena na mekim gingivalnim tkivima – gingivitis (da, vrsta gingivitisa – kataralni, akutni ulcero-nekrotični, hiperplastični, gingivalna fibromatoza, deskvamativni / ne) i prisustva oboljenja parodoncijuma (da, vrsta parodontopatije – hronična parodontopatija, akutna parodontopatija, akutni nekrotizirajući parodontitis, parodontopatija kao manifestacija sistemskih bolesti / ne). Tri indeksa korišćena su za određivanje stanja parodontalnog tkiva – gingivalni indeks po Lou i Silnesu – GI; nivo pripojnog epitela – NPE; parodontalni indeks – CPI, modifikovan za procenu dubine parodontalnog džepa ako je isti prisutan. Sva merenja sprovedena su na šest zuba (brojevi zuba – 16, 12, 24, 36, 32, 44), na četiri površine (mezijalna, distalna, jezična i vestibularna) i kalibrisanom parodontalnom sondom. Uz navedene indekse, primenjen je i plak indeks po Lou i Silnesu – PI, korišćen za registrovanje položaja i količine zubnih naslaga prisutnih u gingivalnom i subgingivalnom pojasu pregledanih zuba.

Indeksi KIP, GI, NPE, CPI i PI prikazani su u svojim srednjim vrednostima sa pripadajućom standardnom devijacijom. Mann-Whitney U test korišćen je za poređenje podataka između dve, a Kruskal-Vallis test za poređenje vrednosti navedenih indeksa za više od dve grupe učesnika i u skladu sa rezultatima sprovedene ankete. Vrednosti $p < 0,05$ smatrane su statistički značajnim. Hi-kvadrat i Fišerovi testovi primenjeni su za poređenje udela karijesnih, plombiranih, ekstrahovanih i zdravih zuba kod dve ili više grupa studenta i u skladu sa rezultatima sprovedene ankete. Vrednosti $p < 0,05$ smatrane su statistički značajnim. Sve statističke analize izvršene su pomoću probne verzije 2021.1.1.1082 statističkog softvera XLSTAT (Addinsoft, Pariz, Francuska, EU).

REZULTATI

U Tabeli 1 prikazan je KIP indeks ukupnog broja studenata podvrgnutih pregledu (n = 699), koji je iznosio 10,24 (standardna devijacija [SD] 5,33; opseg = 0–31).

Kao što je prikazano u Tabeli 2, nije bilo značajnih razlika u vrednostima KIP-a između devojaka i mladića u pogledu

sociodemografskih aspekata, ali je bilo kod ostalih parametera. Najniža vrednost KIP-a zabeležena je kod studenata poreklom iz regiona Istočne i Južne Srbije. Vrednosti KIP-a nisu se značajno razlikovale između Beograda, Zapadne i Centralne Srbije i ostalih regiona i država. Ispitanici koji studiraju tehničke nauke imali su znatno niži KIP, a potom studenti medicinskih nauka. Najviše vrednosti KIP-a uočene su kod studenata prirodnih nauka i studenata drugih visokoškolskih ustanova. Nivo majčinog obrazovanja imao je značajnu ulogu u vrednosti KIP-a studenata, pri čemu su oni čija je majka imala fakultetsku diplomu imali značajno niže vrednosti.

Ni kod jednog parametra nisu uočene značajne razlike u vrednostima KIP-a, osim u slučaju vremena poslednje posete stomatologu. Studenti koji su stomatologa posetili u poslednjih godinu dana imali su viši KIP, i to u vrednosti od 10,61, u poređenju sa onima koji su posetu obavili pre više od godinu dana – 8,64. Rezultati prikazani u Tabeli 3 takođe pokazuju da nema statistički značajne razlike u vrednostima KIP-a u odnosu na stomatološku negu dobijenu kod privatnog ili državnog stomatologa.

Kada se posmatraju navike studenata prema faktorima rizika prikazanih u Tabeli 4, samo je konzumacija alkohola značajno uticala na vrednost KIP-a kod studenata. Studenti koji su svakodnevno/povremeno konzumirali alkohol imali su znatno niži KIP od onih koji su izjavili da nikada ne piju alkoholna pića (9,71 i 10,69).

Studenti koji su ocenili da je njihovo oralno zdravlje zadovoljavajuće imali su niži KIP, vrednosti 8,94, od onih koji su oralnim zdravljem nezadovoljni – KIP vrednosti 12,13. Isti trend primetan je i kada se uporede prosečne vrednosti KŽPsOZ-a – zadovoljni studenti sa ocenom 9,48 i nezadovoljni 11,11. Studenti bez niskog KŽPsOZ-a nisu pokazali prednosti u odnosu na one sa registrovanim niskim KŽPsOZ-om. Isti obrazac moguće je uočiti i kod studenata koji su potvrdili da imaju strah od odlaska stomatologu u poređenju sa onima koji takav problem nemaju (Tabela 5).

Tabela 6 prikazuje status (karijesni, plombirani, ekstrahovani, zdravi) zuba analiziranih u odnosu na region; fakultet i obrazovanje majke. Nešto manji broj zuba analiziran je u kontekstu pola, samoprocene oralnog zdravlja, poslednje posete stomatologu i straha od odlaska stomatologu (509 × 28). Značajne razlike su uočene u gotovo svakom poređenju procentualne zastupljenosti karijesnih, plombiranih, ekstrahovanih i zdravih zuba. Studenti ženskog i muškog pola imaju približno jednak udeo zdravih i ekstahovanih zuba, dok se kod devojaka uočava značajno veći procenat plombiranih i značajno manji udeo karijesnih zuba. Studenti medicine imaju najniži procenat karijesnih zuba, dok bi se isto moglo reći i za studente poreklom iz Istočne i Južne Srbije. Najznačajnije razlike se vide u grupama studenata koji svoje oralno zdravlje doživljavaju kao zadovoljavajuće u odnosu na one koji ga doživljavaju kao nezadovoljavajuće. Takođe nisu uočene značajne razlike u udelu plombiranih zuba kod učesnika čija je majka fakultetski obrazovana i onih čija majka to nije.

Među zubima najčešće pogođenim karijesom preovlađuju prvi i drugi molari i u gornjoj i u donjoj vilici (Slika 1a). Najmanje zahvaćeni bili su zubi u interkaninoj regiji donje vilice (Slika 1b).

Sa druge strane, površine zuba pod zubnim karijesom ravnomernije su distribuirane po različitim tipovima zuba (Slika 2). Prevalencija lezija površine 1 najviša je kod molara, dok lezije površine 2 dominiraju kod premolara i u interkaninom regionu gornje vilice. Lezije na površini 3 uglavnom su zastupljene kod premolara u donjoj vilici.

Procena stanja potpornog aparata zuba kod 275 studenta rezultirala je identifikovanjem gingivitisa kod 99 (36%) studenata i nekog od oblika parodontopatije kod njih 20 (7,3%). Najčešći tip gingivitisa bio je kataralni (Slika 3a). Lokalizovana hronična parodontopatija bila je najzastupljenija u pogledu parodontalnih oboljenja (Slika 3b).

Srednje vrednosti svih analiziranih parodontalnih indeksa prikazanih u Tabeli 7 bile su ispod 1,0 (n = 275).

U ispitivanju ukupnog uticaja samoprocene oralnog zdravlja, straha od odlaska stomatologu i KŽPsOZ-a na zdravlje parodoncijuma, a naročito po pitanju prosečne vrednosti GI i PI indeksa, nisu uočene statistički značajne razlike (Tabela 8).

DISKUSIJA

Vrednost KIP-a studentske populacije u ovoj studiji je iznad 10,24. Niže vrednosti KIP-a su povezane sa rodnim mestom studenata (Istočna i Južna Srbija), sa fakultetima tehničkih nauka, odnosno studentima čije majke poseduju fakultetsku diplomu, studentima koji redovno konzumiraju alkohol i onima koji smatraju svoje oralno zdravlje zadovoljavajućim. Znatno viši KIP primećen je kod ispitanika koji su posetili stomatologa u poslednjih godinu dana, a priroda stomatološke službe (privatne ili javne) ne igra bitniju ulogu, kao ni pol i strah od odlaska stomatologu. Učestalost pranja zuba, interdentalno čišćenje, upotreba fluoridne paste za zube, konzumiranje zaslađenih pića, pušenje i KŽPsOZ takođe nemaju značajnijeg uticaja na vrednosti KIP-a studentske populacije. Niže vrednosti prosečnog KŽPsOZ-a, koje podrazumevaju odsustvo tegoba i problema, potpuno odgovara pogledu studenata na svoje oralno zdravlje. S druge strane, zastupljenost karijesnih, plombiranih, ekstrahovanih i zdravih zuba donosi nešto drugačiju perspektivu oralnog zdravlja učesnika ovog istraživanja. Niža zastupljenost karijesnih zuba u velikoj meri povezana je sa studentima ženskog pola, studentima poreklom iz Istočne i Južne Srbije i studentima medicinskih nauka. Slično je i sa onima čije majke imaju viši stepen obrazovanja (fakultetsku diplomu), odnosno studentima koji su posetili stomatološku ordinaciju u proteklih 12 meseci i nemaju strah od odlaska stomatologu. Takođe je evidentno da studenti ženskog pola, zajedno sa onima koje redovno posećuju stomatologa i nemaju strah od odlaska stomatologu, poseduju značajno veći broj plombiranih zuba. Kada je reč o zubima i zubnim površinama koje su uglavnom zahvaćene karijesom, rezultati ove studije prate uobičajene trendove [16]. Najveća prevalencija karijesa primećuje se na okluzalnim površinama molara i aproksimalnim površinama sekutića, očnjaka i premolara, a naročito u gornjoj vilici. Gingivalna trećina očnjaka i premolara donje vilice takođe je bila značajnije zahvaćena karijesom. Samo nekolicini ispitanika je dijagnostikovan neki vid parodontopatije. Istovremeno, prosečne vrednosti GI, NPE, CPI i PI indeksa znatno ispod jedan ne ukazuju na ozbiljnija oboljenja parodoncijuma u ispitivanoj populaciji.

Prosečna vrednost KIP-a ukupnog broja učesnika ovog istraživanja veća je od vrednosti istog parametra u studijama sličnog sadržaja. Prosečni KIP od 10,24 najbliži je vrednostima studenata stomatologije i medicine u Rusiji – 7,46, a potom kod korejskih studenata – 6,1 [10, 17]. Kada se analizira samo KIP vrednost studenata medicine koji su učestvovali u ovoj studiji, razlike su manje izražene (KIP = 9,67). U poređenju sa populacijom

30-godišnjaka u Adelejdu, Australija (KIP = 2,1), 18-godišnjaka u Hong Kongu, Kina (KIP = 1,4) i studenata prve godine studija u Okajami, Japan (KIP = 2,01), vrednosti su znatno više [18, 19, 20]. Vredno je napomenuti da broj plombiranih zuba u ovim studijama zauzima veći udeo KIP-a (80-90%), dok u našoj studiji to nije slučaj – oko 60% [10, 19]. Takav nalaz ukazuje na to da studenti osnovnih studija u Beogradu imaju veći broj aktivnih karijesnih lezija, kao i veći broj ekstrakcija od kolega sa određenih univerziteta u inostranstvu.

Uz sve navedeno, ruski studenti se značajno razlikuju po grupama sa niskim i bez niskog KŽPsOZ-a, što nije tipično za beogradske studente [10]. U tom smislu, studenti osnovnih studija u Beogradu sličniji su 19-godišnjim Šveđanima [21]. Prosečni GI indeks beogradskih studenta gotovo je dvostruko veći od vrednosti istog parametra za studente iz severozapadne Rusije (0,51 i 0,27) [10]. Međutim, obe vrednosti ne prelaze graničnu vrednost od jedan, što ukazuje na odsustvo inflamacije gingive.

U poređenju sa studijom procene oralnog zdravlja iz 1987. godine, KIP je opao sa 12,5 u 1987. na 10,2 u 2012. U pogledu procenta zuba pogođenih karijesom, broj je porastao sa 31% u 1987. na 38% u 2012. [7]. Ostaje nejasno da li su razlozi navedenog skoka ekonomske prirode, neefikasnosti preventivnih programa u dečjem dobu, regulatornih pitanja ili individualnih faktora. Takođe, rezultati ove studije pokazuju da što je veći nivo obrazovanja majki studenata, to isti poseduju veći broj zdravih zuba. Navedeni rezultat podudara se sa konstatacijama da visok nivo majčine edukacije i adekvatno oralno zdravlje majke pozitivno utiču na oralno zdravlje tokom detinjstva, a kasnije i tokom odrastanja [22]. Udeo plombiranih zuba takođe se uvećao sa 46,4% na 54,5%, dok je procenat ekstrahovanih zuba zabeležio značajan pad sa 22,4% na 7,4% [7]. To potencijalno ukazuje na prelazak sa hirurškog na konzervativniji pristup u stomatološkom lečenju pacijenata studentske populacije u periodu između dve studije.

Prema saznanjima autora ovog istraživanja, ovo je prva studija od 1987. godine u Beogradu i Srbiji koja daje sveobuhvatan i detaljan pregled stanja oralnog zdravlja studenata uzrasta od 19 do 26 godina. Za razliku od većine ostalih studija, ona isključuje studente stomatologije, čime doprinosi diversifikaciji nalaza i obuhvata brojne druge grupe studenata različitog geografskog porekla i sa različitih fakulteta. Takođe obezbeđuje zdravu osnovu za dalja ispitivanja uticaja KŽPsOZ-a na kliničke determinante oralnog zdravlja.

Odeljak 6 upitnika, a na temu KŽPsOZ-a, sadrži svega osam pitanja koja se odnose na originalni OHIP-14 obrazac. Iako se unutrašnja konzistentnost primenjene ankete na temu KŽPsOZ može smatrati prihvatljivom (Kronbahova alfa > 0,7 [0,778; 0,703]), ona je i dalje značajno ispod nivoa vrednosti iznad 0,85 dostignutog u studijama koje primenjuju originalni koncept OHIP-14 obrasca [8, 16]. Drugi evidentan nedostatak ove studije jeste i činjenica da su procenu stomatološkog statusa i stanja parodoncijuma sprovodili studenti stomatologije. I pored neposrednog nadzora kliničkih lekara, ostaje nejasno da li je kvalitet obavljenog pregleda u svim slučajevima bio na zadovoljavajućem nivou.

ZAKLJUČAK

Nalazi ove studije ukazuju na smanjenje KIP-a kod mladih starosti od 19 do 26 godina tokom poslednjih 30 godina i porast procenta karijesom zahvaćenih zuba na nivou populacije.

Na osnovu komparativnih analiza uočava se da populaciona grupa studenata u Beogradu sveukupno ima lošije oralno zdravlje od svojih kolega iz Rusije, Kine, Japana ili Švedske.

Samoprocena studenata o kvalitetu svog oralnog zdravlja ima značajan uticaj na neke od kliničkih determinanti analiziranih u okviru ovog istraživanja.

Dalja ispitivanja neophodna su u kontekstu određenih socio-ekonomskih i lokalno-regionalnih specifičnosti koje mogu objasniti razlike u oralnom zdravlju između studenta poreklom iz različitih regiona. Buduća istraživanja takođe je neophodno usredsrediti na prospektivna istpitivanja promena koje se potencijalno dešavaju u funkciji vremena kod istih ispitanika. Iz tih razloga akcenat je potrebno staviti na aktivnije uključivanje privatnih i državnih stomatologa i na obezbeđivanje podrške regulatornih tela radi pouzdanijeg i uspešnijeg pristupa zajednice u pogledu oralnog zdravlja.

Zahvalnica: Ova studija izvedena je u okviru projekta „Zagrizi Znanje Zdravim Zubima“ finansiranog sredstvima Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije (Projekat br. 451-02-266 / 2011-05). Želimo da se zahvalimo osoblju Klinike za bolesti zuba i Klinike za parodontologiju i oralnu medicinu Stomatološkog fakulteta Univerziteta u Beogradu na njihovom predanom radu i doprinosu ovoj studiji. Takođe bismo želeli da se zahvalimo gđi Sonji Nektarijević na vremenu i radu koje je posvetila pripremi aplikacione dokumentacije i celokupnom dizajnu projekta.

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Evaluation of surface characteristics of new rotary nickel-titanium instruments – SEM-EDS analysis

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SUMMARY

Introduction Modern endodontic procedure implies the use of rotary Ni-Ti instruments during chemomechanical treatment of root canals. The aim of this study is to analyze the surfaces of new (unused) rotary endodontic instruments using the SEM-EDS method and determine how frequently manufacture defects or impurities appear on their working surfaces.

Material and method Five new different sets of rotary endodontic Ni-Ti instruments were included in this study: K3, Mtwo, ProTaper Universal, HyFlex and BioRaCe. The working part of endodontic instrument was analyzed using SEM-EDS method (magnifications ×150 to ×2000), which determined the morphological characteristics of the instrument surface and chemical composition of the found impurities. Statistical analysis was performed using the Fisher's test ($p < 0.05$).

Results The results of SEM-EDS analysis showed that there is no new instrument without defects on its surface. The most common defects were observed in K3 (27.43%) and ProTaper Universal group (27.21%) and the least were in BioRaCe instruments (7.67%). The most common type of defect in tested instruments was fretting. In addition, the presence of debris and metal strips was found on all instruments, while corrosion of the working part was observed only in K3, ProTaper Universal and Mtwo systems in a small percentage.

Conclusion Based on the results of this research, it can be concluded that manufacturing defects were noticed in all examined instruments. The most common defect is pitting. Impurities such as debris and metal strips have also been registered. No organic debris was observed on electropolished surface of BioRaCe instruments, but a small percentage of other types of defects were registered.

Keywords: Ni-Ti instruments; manufacturing defects; SEM-EDS

INTRODUCTION

The application of rotary nickel-titanium (Ni-Ti) instruments with shape memory properties, biocompatibility and corrosion resistance, has introduced a new era into the endodontic procedure. Rotary Ni-Ti instruments have enabled faster and more efficient preparation i.e. reduced the possibility of procedural errors during cleaning and shaping of root canals of different morphology [1]. Numerous innovations in instrument design, in recent years related to the surface and heat treatment of Ni-Ti alloys, have affected the efficiency and required safety during endodontic treatment [2–4].

In the production of endodontic instruments, Ni-Ti alloys are used in the ratio 56: 44 = Ti: Ni, which achieves their equiatomic relationship. Although only one manufacturer (Dentsply, Maillefer Instruments SA, Ballaigues, Switzerland) published the absolute composition and detailed technological manufacturing process, it is assumed that this is the best ratio that gives the alloy super-elastic properties [5].

The production of Ni-Ti rotary endodontic instruments is much more complicated compared to the process of making steel instruments by cold twisting of pre-profiled wire cones [2]. Ni-Ti instruments are created by specific grinding, i.e. by carving a certain profile into the central stem of the Ni-Ti wire [5]. Newer production techniques include a combination of heat treatment of alloy and simultaneous twisting, for greater flexibility and better resistance to torsion and cyclic fatigue [3]. Although modern computer technology is used in the process of making a complicated design of Ni-Ti instruments, surface defects often occur in the form of fretting, pitting, cracks and impurities that can increase vulnerability to fracture [6]. It has been observed that surface defects act as points of stress concentration, leading to the initiation and spread of cracks i.e. frequent fractures during instrument activation [7].

Various metal residues and impurities of organic and inorganic origin can be found on the surface of new endodontic instruments. During instrumentation, these metal shavings can be incorporated into dentinal wall or pushed into periapical tissue and cause an allergic reaction [8]. The use of instruments with organic impurities also carries the risk of potential cross-infection [9].

The aim of this study is to analyze the surfaces of new rotary endodontic instruments using Scanning Electron Microscopy with Energy-Dispersive Spectrometry

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Table 1. Characteristics of tested sets of Ni-Ti rotary instruments
Tabela 1. Osnovne karakteristike ispitivanih setova Ni-Ti rotirajućih instrumenata

Instrument Manufacturer Instrument Proizvođač	Activation Aktivacija	Cross-section Special Features Poprečni presek, specifičnosti dizajna	Diameter Dijametar	Taper Koničnost	Manufacturing Proces proizvodnje
K3, SybronEndo	Rotary centric Puna rotacija	Triple-fluted, Positive rake angle with asymmetric radial lands Trostruka sečiva sa pozitivnim uglom i asimetričnim radijalnim površinama	25	0.12-0.02	Micromilling, conventional Ni-Ti alloy Mikroglodanje, konvencionalna Ni-Ti legura
Mtwo, VDW	Rotary centric Puna rotacija	S-shaped with two active cutting edges S-oblika sa dva aktivna sečivna ugla	10-35	0.04-0.05, 0.06	Micromilling, Conventional Ni-Ti alloy Mikroglodanje, konvencionalna Ni-Ti legura
ProTaperUniversal Dentsplay-Sirona	Rotary centric Puna rotacija	Convex triangular, Variable and progressive tapers along the instrument Konveksni trougao, varijabilna progresivna koničnost duž instrumenta	17-30	Regressive Tapers Regresivna koničnost	Micromilling, Conventional Ni-Ti alloy Mikroglodanje, konvencionalna Ni-Ti legura
HyFlex CM Coltene	Rotary centric Puna rotacija	Double fluted Hedstroöm design with positive rake angle Dvostruki Hedstrom dizajn sa pozitivnim upadnim uglom	20-40	0.04, 0.06, 0.08	Micromilling, Post- manufacture heat treatment: CM-wire Mikroglodanje CM-žica
BioRaCe, FKG	Rotary centric puna rotacija	Triangular with alternating cutting edges along the instrument Trougaoni sa alteracijama sečivnih ivica duž instrumenta	15-40	0.04, 0.05, 0.06, 0.08	Micromilling, Electropolishing Conventional Ni-Ti alloy Mikroglodanje, konvencionalna Ni-Ti legura, elekropolirana površina

(SEM-EDS) to determine how frequently manufacture defects or impurities appear on their working surfaces.

MATERIAL AND METHOD

The study used three basic sets (each set of six instruments) of five different systems of rotary endodontic Ni-Ti instruments: K3 (SybronEndo Co, USA), Mtwo (VDW, Munich, Germany), ProTaper Universal (Dentsply Maillefer, Switzerland), HyFlex (Coltene Whaledent group, Switzerland) and BioRaCe (FKG DENTAIRE Swiss Dental Products, Switzerland) (Table 1).

Scanning Electron Microscopy with Energy-Dispersive Spectrometry (SEM-EDS) was performed in the Laboratory for SEM, Faculty of Mining and Geology, University of Belgrade, using the type of SEM - JEOL JSM-6610LV, Japan. The instruments were analyzed without any preparation (directly from the factory packaging). Images were made using Secondary Electron Detector (SE images – second electron) at magnifications ranging from 150x to 2000x. Chemical analysis was performed on unpolished samples using the EDS detector (type X-Max Large Area Analytical Silicon Drifted spectrometer, Oxford Instruments) using internal standards. Obtained chemical composition is presented as the content of chemical elements in weight percent (wt%), normalized to 100%. Detection limit for most elements was about 0.1 wt%. This type of chemical analysis is considered semi-quantitative, because it was performed on unpolished surfaces.

A total of 540 recordings of apical and middle thirds of instruments were made from two different directions. Three SE images were taken for each surface of the instrument. Two researchers reviewed the images and their results were reconciled by Cohen Kappa analysis.

A qualitative analysis of various irregularities and errors present on the working surface of Ni-Ti instruments was applied in accordance with recommendations of Kristina Egert et al. [10]. The instruments registered the presence of: pitting, fretting, microfractures, complete fractures, metal flash, metal strips, blunt cutting edge, disruption of cutting edge, corrosion and presence of debris.

Statistical analysis of obtained results was performed using the Fisher's test ($p < 0.05$).

RESULTS

The results of SEM-EDS analysis of the new Ni-Ti sets are presented in Tables 2-4 and Figures 1-9.

The analysis of SE images showed the existence of surface contamination on the working part of tested instruments, and the subsequent EDS analysis determined its chemical composition. This way, a division was made into instruments contaminated with debris and instruments contaminated with metal strips. Examples of SEM-EDS analysis with the appearance of performed spectrum are presented in Figures 1 and 2.

SEM-EDS analysis in point 1 (spectrum 1, Figure 1, Table 2) shows that dominant element in the examined sample was carbon (88.1 wt%) with low oxygen content (1.5 wt%), and impurity on this ProTaper Universal instrument is characterized as a debris of organic origin. Nickel and titanium contents (spectrum 1) reflect the composition of instrument.

Based on SEM-EDS nalysis, the impurities on K3 instrument are characterized as a combination of debris of organic origin and contamination with metal strips. The quantity of nickel and titanium in analysis 1 (spectrum 1, Figure 2) and slightly more quantity of these two elements in analysis 2 (spectrum 2, Figure 2) represents the distribution of these elements in the structure of instruments, just like in the previous case.

The results of SEM analysis indicate the most frequent occurrence of defects and impurities in systems

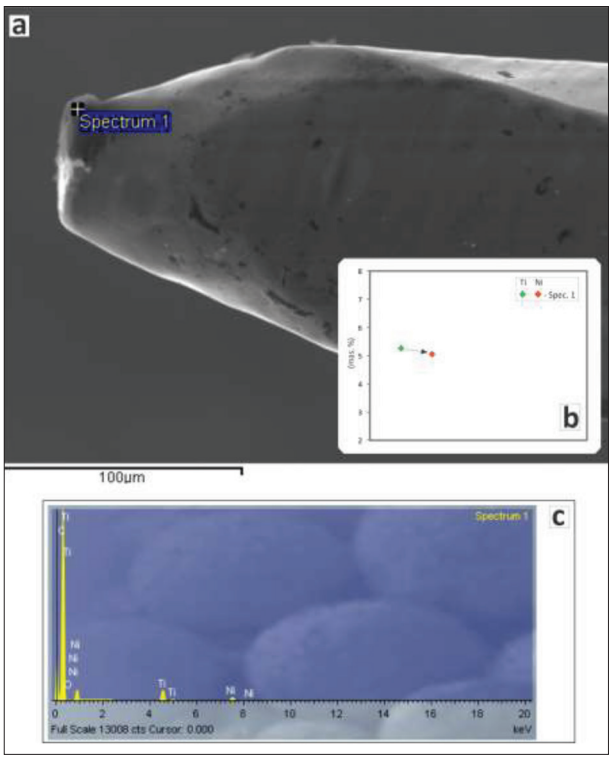

Figure 1. SEM-EDS analysis of the new ProTaper (Sx) instrument
a) SE image of the tip of ProTaper (Sx) instrument with the marked point where EDS analysis was performed, parts of its surface, b) nickel and titanium contents (wt%), c) spectrum of the analysed point
Slika 1. SEM-EDS analiza površine novog ProTaper (Sx) instrumenta
a) SE snimak vrha ProTaper (Sx) instrumenta sa označenom tačkom u kojoj je izvršena EDS analiza, dela njegove površine,
b) sadržaji nikla i titanijuma (mas %),
c) izgled spektra 1 za analizu u tački 1 (a)

Table 2. EDS analysis of the sample of ProTaper Universal (Sx) instrument in point 1 (Figure 1)
Tabela 2. SEM-EDS analiza uzorka vrha ProTaper Universal (Sx) instrumenta u tački 1 sa Slike 1

Spectrum	C	O	Ti	Ni	Total Ukupno
Spectrum 1	88.1	1.5	5.3	5.1	100.0

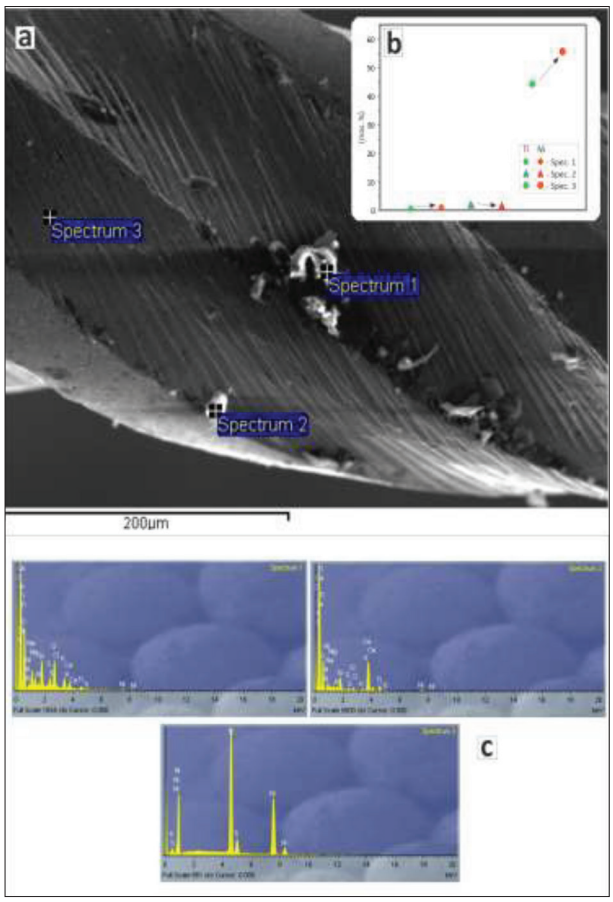

Figure 2. SEM-EDS analysis of a new K3 (25-0.10) instrument
a) SE image of K3 instrument surface with marked points where EDS analyzes were performed, b) nickel and titanium contents (wt%) at the analysed points (spectra 1–3),
c) spectrums of the analysed points 1–3
Slika 2. SEM-EDS analiza površine novog K3 (25-0.10) instrumenta
a) SE snimak površine K3 instrumenta sa označenim tačkama u kojima su vršene EDS analize, b) sadržaji nikla i titanijuma (mas %) u tačkama analiziranja (spektri 1–3),
c) izgled spektara 1–3

Table 3. EDS analysis of sample K3 (25-0.10) in points 1–3 (Figure 2)
Tabela 3. SEM-EDS analiza površine uzorka K3 (25-0.10) instrumenta u tačkama 1–3 (Slika 2)

Spectrum	C	O	Na	Mg	Al	Si	S	Cl	K	Ca	Ti	Ni	Total Ukupno
Spectrum 1	47.6	37.4	2.3	1.0	0.2	2.5	1.0	3.6	1.6	1.1	0.6	1.1	100.0
Spectrum 2	49.8	34.0	0.8	0.6	0.4	1.8	0.3	0.5	0.4	7.7	2.0	1.7	100.0
Spectrum 3	0.0	0.0	0.0	0.0	0.0	0.0	0,0	0.0	0.0	0.0	44.3	55.7	100.0

of K3 (27.43%) and ProTaper Universal group (27.21%). Occurrence of defects and impurities on new Ni-Ti instruments was recorded in Mtwo group in 20.43% and in HyFlex group in 17.2%. The lowest percentage of defects on the apical and middle surface was shown by BioRaCe sets (7.67%). Analyzing the results, a higher occurrence of defects on the apical surfaces of the working part of tested instruments can be noticed (53.49% on the apical and 46.51% on the middle third) (Table 4).

The presence of pitting and fritting (apical and middle third 100%), the presence of defects in the form of metal strips (apical third 83.33%, middle third 38.89%) and corrosion on only one instrument (apical and middle third) (Figure 3) were noticed on the surface of working part of new K3 instruments. Debris was observed in apical third in 77.8% of instruments and in middle third in 44.44% of K3 instruments.

SEM analysis of Mtwo instruments indicated the presence of metal strips in apical (50%) and middle third (22.2%). The corrosion was observed only on one instrument (the thinnest one) (10 / 0.4) in its middle third (Figure 4).

Blunt cutting edge of Mtwo instruments was observed on apical surface of one instrument (25 / 0.6) and defect in the form of disruption of cutting edge in middle third of

Table 4. Presence of defects and impurities on the working part of new Ni-Ti instruments
Tabela 4. Prisustvo defekata i nečistoća na radnom delu novih Ni-Ti instrumenata

		K3		*Mtwo*		*ProTaper Universal*		*HyFlex*		*BioRaCe*	
		Apical third % apikalna trećina %	**Middle third % srednja trećina %**	**Apical third % apikalna trećina %**	**Middle third % srednja trećina %**	**Apical third % apikalna trećina %**	**Middle third % srednja trećina %**	**Apical third % apikalna trećina %**	**Middle third % srednja trećina %**	**Apical third % apikalna trećina %**	**Middle third % srednja trećina %**
1	No Bez vidljivih defekata	0	0	0	0	0	0	0	0	0	0
2	Pitting Jamičasta udubljenja	100	100	0	0	83.3	88.8	0	0	0	0
3	Fretting Žljebovi	100	100	100	100	100	100	100	100	33.3	27.7
4	Microfractures Mikrofrakture	0	0	0	0	0	0	0	5.5%	0	0
5	Complete fractures Kompletne frakture	0	0	0	0	0	0	0	0	0	0
6	Metal flash Metalna uglačanost	0	0	0	0	0	0	0	0	0	0
7	Metal strips Metalni opiljci	83.3	38.8	50.0	22.2	50.0	38.8	5.5	0	50.0	38.8
8	Blunt cutting edge Zatupljene sečivne ivice	0	0	5.5	0	0	0	0	0	0	0
9	Disruption of cutting edges Prekid sečivne ivice	0	0	0	5.5	5.5	5.5	0	0	0	0
10	Corrosion Korozija	5.5	5.5	5.5	0	11.1	11.1	0	0	0	0
11	Debris Debris	77.7	44.4	100	100	100	55.5	100	100	11.1	22.2
	∑	15.34	12.09	10.93	9.53	14.65	12.56	8.60	8.60	3.95	3.72
	∑	27.43		20.46		27.21		17.2		7.67	

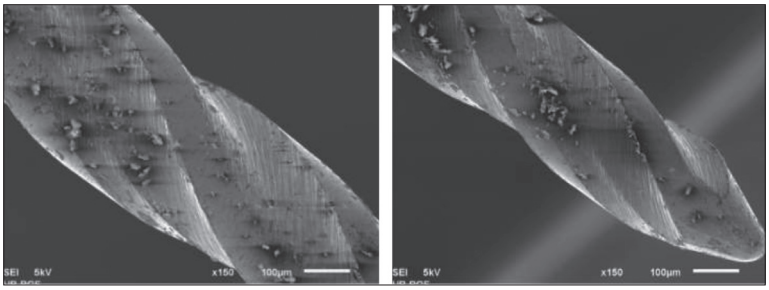

Figure 3. SE image of new K3 instruments: a) surface of middle third (25/0.10) with defects in the form of metal strips, pitting and fretting (×150) b) surface of apical third (25/0.08) where defects in the form of metal strips, pitting and fretting can be observed (×150).
Slika 3. SE snimak površine novih K3 instrumenata: a) površina srednje trećine (25/0,10) sa defektima u vidu metalnih opiljaka, žljebova i jamičastih udubljenja (×150); b) površina apikalne trećine (25/0,08), gde se uočava prisustvo defekta u vidu metalnih opiljaka, žljebova i jamičastih udubljenja (×150)

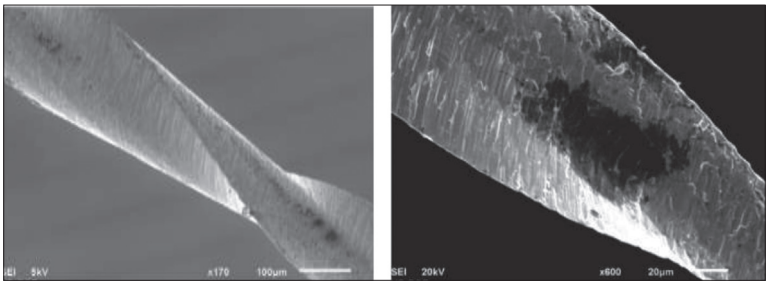

Figure 4. SE image of new Mtwo instrument: a) surface of middle third (10/0.4) with the presence of corrosion (×170); b) detail from the previous image at higher magnification (×600)
Slika 4. SE snimak novog Mtwo instrumenta: a) površina srednje trećine (10/0,4) sa prisustvom korozije (×170); b) detalj sa prethodnog snimka na većem uvećanju (×600)

the instrument (20 / 0.6) (Figure 5). Debris (confirmed by EDS analysis) was observed on all instruments, both in the apical and middle third (100%).

The most common defects on the working surface of new ProTaper Universal instruments were changes in the form of fretting (apical and middle third, 100%) and pitting (apical third 83.3% and middle third 88.8%) (Table 4, Figure 6). Metal strips were detected on the apical (50%) and middle third (38.8%). A defect on the cutting edge (disruption of its continuity) was observed on one, the most conical instrument (Sx) (apical and middle third), and corrosion on the apical (11.1%) and middle part (11.1%) (Figure 7). Contamination in the form of debris was noticed in apical third (100%) and in middle third of slightly more than a half (55.5%) of ProTaper Universal sets. The most common defects on new HyFlex instruments were the appearance of fretting in the form of debris on apical and middle segments of all instruments (100%) (Figure 8). A defect in the form of a microfracture was observed on the apical part of the instrument (25-0.08) as well as the appearance of metal strips (25-0.04).

The results of SEM analysis of BioRaCe sets show the most frequent occurrence of metal strips (apical 50% and middle third

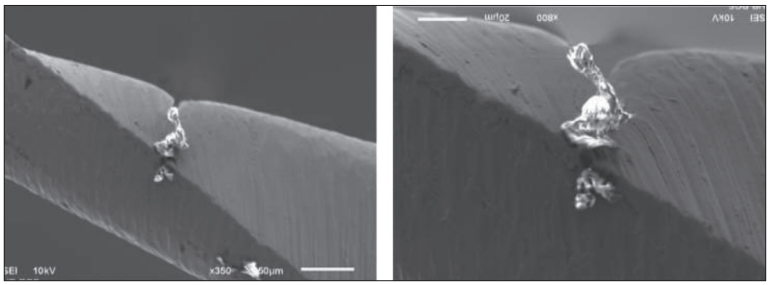

Figure 5. SE image of new Mtwo instrument (20/0.6): a) area of middle third with a disruption of cutting edge defect (×350); b) detail from the previous image at higher magnification (×800)
Slika 5. SE snimak novog Mtwo instrumenta (20/0,6): a) površina srednje trećine na kojoj se uočava prisusutvo defekta u vidu prekida sečivne ivice (×350); b) detalj sa prethodnog snimka na većem uvećanju (×800)

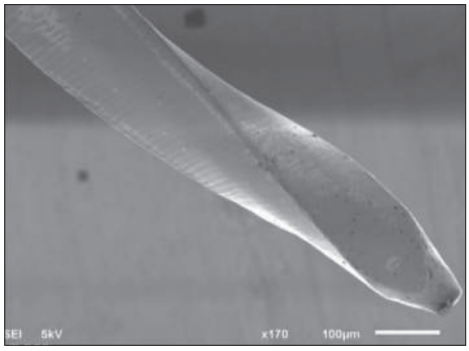

Figure 7. SE image of apical third of new ProTaper instrument (Sx) with corrosion and discontinuity of cutting edge (apart from the presence of pitting and fretting) (×170)
Slika 7. SE snimak apikalne trećine novog ProTaper instrumenta (Sx) sa korozijom i prekidom kontinuiteta (pored prisustva jamičastih udubljenja i žljebova) (×170)

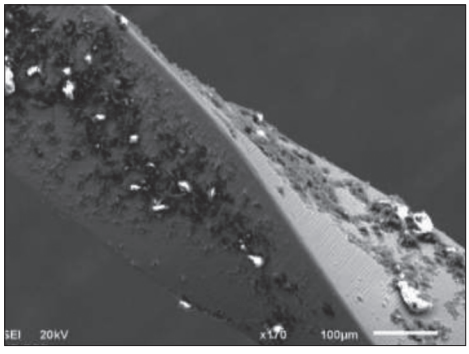

Figure 9. SE image of the surface of new BioRaCe instrument (No. 3) i.e. its middle third with the presence of metal strips and fretting (×170)
Slika 9. SE snimak površine srednje trećine novog BioRaCe instrumenta (br. 3) sa prisusutvom metalnih opiljaka i žljebova (×170)

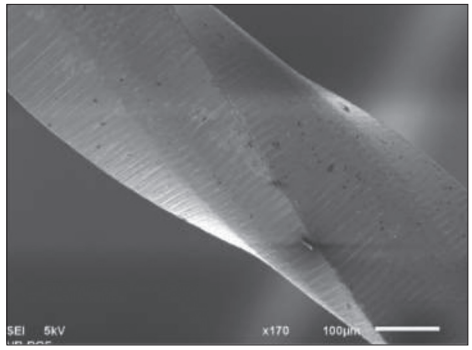

Figure 6. SE image of middle part of ProTaper Universal instrument (F2) with pitting and fretting (×170)
Slika 6. SE snimak srednjeg dela ProTaper Universal instrumenta (F2) sa jamičastim udubljenjima i žljebovima (×170)

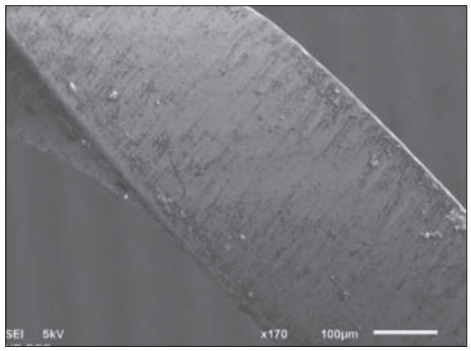

Figure 8. SE image of the middle third surface new HyFlex instrument (30-0.04) with the presence of fretting and debris (×170)
Slika 8. SE snimak površine srednje trećine novog HyFlex instrumenta (30-0.04) sa prisusutvom žljebova i debrisa (×170)

38.8%) and fretting (apical 33.3% and middle third 27.7%) (Table 4, Figure 9). Debris contamination detected by EDS analysis was observed on the apical (11.1%) and middle surface (22.2%) of the instruments.

A statistically significant difference in the presence of fretting was observed between K3, MTwo, ProTaper and Hyflex instruments between the apical (by $p < 0.05$) and middle third (by $p < 0.05$).

Significant differences related to the presence of metal strips were observed between K3 and HyFlex instruments (by $p < 0.05$), between ProTaper Universal and HyFlex (by $p < 0.05$), MTwo and HyFlex (by $p < 0.05$) and BioRaCe and HyFlex instruments (for $p < 0.05$)). The difference was also significant in the occurrence of metal strips in apical third between K3 and BioRaCe group of instruments (by $p < 0.05$) between K3 and MTwo (by $p < 0.05$) and K3 and ProTaper group, respectively (by $p < 0.05$).

In the middle third, a statistically significant difference in the occurrence of metal strips was observed between the HyFlex group and K3, BioRaCe, ProTaperUniversal and MTwo instruments (by $p < 0.05$). The difference was also significant between the apical and middle third of K3 instruments (by $p < 0.05$).

The difference was also significant in the values of debris in the apical segment between K3 and ProTaper Universal group (for $p < 0.05$), K3 and MTwo group (for $p < 0.05$), and between K3 and HyFlex group of instruments (for $p < 0.05$). A statistically significant difference was also observed between K3 and BioRaCe group (for $p < 0.05$), ProTaperUniversal and BioRaCe group (for $p < 0.05$), MTwo and BioRaCe group (for $p < 0.05$), respectively, HyFlex and BioRaCe groups (for $p < 0.05$). In the middle third, the difference was significant between K3 and MTwo (for $p < 0.05$) and K3 and HyFlex instruments (for $p < 0.05$). In the ProTaper Universal group, a statistically significant difference in the occurrence of debris was observed between the apical and middle third (by $p < 0.05$).

DISCUSSION

SEM analysis of various surface irregularities, manufacturing defects and contamination of new Ni-Ti rotating instruments showed that there is not a single tested instrument without defects or impurities on the working surface. In this study, the presence of various defects and impurities was found in all new Ni-Ti instruments (five different commercial sets), with a slightly higher prevalence observed in their apical third. Although there is confirmation of their sterility on the factory packaging, the presence of defects and dirt on the active part of new Ni-Ti instruments is a proven reality, which is documented by the results of various studies [8, 10–19]. The complicated process of machining the initial Ni-Ti wire often causes the occurrence of surface deformations and cracks due to the traces of milling and machining, but also the appearance of polished surfaces on the cutting edges of instruments [5, 10].

Changes on the surface can compromise blade efficiency of instruments and become sites for potential corrosion. Also, these points represent the sites of initiation of defects, contributing to degradation of mechanical properties and occurrence of micro or complete fractures during their clinical use [5, 7, 13]. Arens et al. presented an interesting study on the incidence of fractures after the first use of new Ni-Ti instruments (0.9%), while Shen cites inadequate manipulation and existence of manufacturing defects as the cause of this complication [12, 13].

Due to the higher forces and speed that are necessary for the processing of Ni-Ti alloys, it is possible to cause burning sawdust and formation of hardened places. These are the hardened parts that are more difficult to process, and they represent the zones with higher probability of deformation and fractures [16].

The manner in which defects are formed during the formation of Anusavica and Phillips alloys has been attributed to the specific phase transformation and recrystallization of Ni-Ti alloy [20]. Recrystallization represents the change in the type of lattice depending on the temperature (e.g. titanium at 882 ° C changes from a hexagonal to a monoclinic structure), where the rate of crystallization affects the regularity of crystal structure [20].

The most common type of surface irregularities on working surface of new instruments in this study was the appearance of fretting. Clinical significance of fretting is potentially increased possibility of its screwing (due to the friction that is caused by uneven surface) and increased incidence of fracture [13].

Presence of metal strips as a consequence of the production process was observed on the work surface of all tested instruments. The correlation between the high prevalence of metal strips and the higher conicity of K3 instruments in this study (conicity greater than .06) is in accordance with the results of Marending et al. who indicated that metal strips are formed as a result of the production process of Ni-Ti instruments [11]. Using SEM analysis, Van Eldik et al. noticed the presence of a large amount of metal strips on the surface of new Ni-Ti instruments, immediately after opening the original packaging [14]. This type of contamination leads to a decrease in cutting efficiency, and metal strips can be retained in the dentinal walls of the canal or in the periapical tissue during instrumentation. Van Eldik proved that possible contamination of periapical tissue with these metal strips could reduce the course of tissue repair and compromise the success of endodontic therapy [14]. According to the results of Stefanescu et al. metal particles can be transported during instrumentation and active irrigation through the apical foramen and cause an allergic reaction of the periapical tissue [8]. It has been shown that metal ions as potential hapten allergens can cause type 1 reaction, with a possible immediate or delayed dermal or mucosal reaction. Allergic reactions in endodontics are extremely rare, but the consequences of allergic reactions such as symptoms of delayed apical healing, persistent discomfort after canal obstruction, can increase their number significantly [8].

The presence of debris was also observed on the working surfaces of all types of tested Ni-Ti instruments. Titanium alloys are difficult to machine due to their elasticity and require higher cutting forces compared to steel. Ni-Ti alloys are intensively glued to the tool with which they are processed, so the protection of materials is achieved by oxidizing the surface or metal coating, which are removed chemically after processing, but may still remain on their surface [16]. Electropolishing the surface of BioRaCe instruments increases cutting efficiency, while reducing defects in the production process and possible debris contamination [21]. The significant frequency of debris in the apical third of K3 and ProTaper Universal instruments in relation to their middle third confirms higher contamination of the apical segment due to the more complex production of thinner apical part. This finding is consistent with studies by Eggert and Alapati, which indicated a higher incidence of debris in the apical segment of new Ni-Ti instruments [10, 22].

Working surface defects in the form of pitting were observed only in two groups of new instruments, but in a high percentage (K3 and ProTaper Universal). The appearance of pitting occurs during the production process, as during melting of elemental nickel and titanium, the rates of their mutual diffusion during heating differ, which leads to the formation of void spaces [23]. Nickel atoms diffuse faster into titanium than titanium atoms in the opposite direction. Thus, the mass transport is not balanced which can lead to the formation of void spaces in the nickel after alloying. These cavities are known as the Kirkendal porosity or Kirkendal effect [23].

Nagumo presented the evidence on the significance that these defects have on mechanical characteristics of Ni-Ti instruments, as well as the exact mechanism of their influence [24]. He observed that alloy could absorb hydrogen from saliva and form hydride bonds with Ni-Ti lattice atoms that are stable at room temperature. This change in the molecular structure leads to the change in physical properties of the alloy, causing hydrogen porosity. Asaoka also pointed out that diffusion of hydrogen through a Ni-Ti alloy forms hydride phases on the surface of a material that has a more brittle structure [25]. This newly formed hydride layer on the active surface of Ni-Ti instrument is of different thickness thus causing microcracks during

clinical work. By providing an absolutely dry working field, this mechanism is not important, but it can have an impact during the process of cleaning and sterilization of instruments, when the instruments are exposed to a longer action of ionizing liquids [25].

Corrosion of the working part of Ni-Ti instruments was not observed on HyFlex and BioRaCe instruments, and in other groups it was observed in a small percentage. The low degree of corrosion on Ni-Ti instruments confirms the resistance of this alloy to corrosion, but also the non-exposure of new instruments to corrosive factors [26].

Findings of defects on the surface of new Ni-Ti instruments in the form of blunting of the cutting edge, disruption of the blade edge and microfracture, only confirm the problems of their production. Microfractures on new instruments are, according to research by Marending and Barbakow, the result of manufacturing process of larger and more conical but less flexible instruments [11]. According to the most researchers, cracks or microfractures are the most dangerous defects that a file can have [24, 25]. If instruments with this defect are activated in the canal, during rotation and screwing, they break immediately. Microfracture affects high sensitivity of the instrument to the accumulation of cyclic fatigue and inevitable fractures [11, 24, 25].

Subsequent heat treatment of finished Ni-Ti instruments (HyFlex) potentially offers the most promising method of manufacturing rotating instruments [27]. These instruments do not have the shape memory that traditional Ni-Ti instruments have, and a special thermomechanical procedure significantly increases their flexibility [28]. The research results in this study show the lowest contamination of Hyflex system with metal strips. The low prevalence of this contamination can be explained by their specific heat treatment that reduces irregularities on their surface. Heat treatment, apart from the change in microstructure (increased flexibility), also leads to the appearance of a cleaner and more regular surface of these instruments [28, 29].

Following the results of our study, a significantly lower prevalence of defects in BioRaCe group is observed. This finding is in accordance with the results of a research on a significant reduction of surface irregularities of electropolished instruments [3]. Electropolishing creates a homogeneous oxide layer during the production process, which reduces the appearance of surface defects and increases resistance to corrosion and fracture [27]. Electropolished surface of instruments is visibly brighter than the untreated surface [26]. By introducing current through the solution, a thin passive layer is formed and the surface dissolves into the electrolyte, which also leads to the selective removal of surface defects [4].

In order to improve microstructure of the working surface of Ni-Ti instruments and improve mechanical properties, flexibility, fatigue resistance, i.e. cutting efficiency, manufacturers have used various techniques in recent years (ionic application, plasma immersion, titanium oxide coating, thermal nitriding, thermal treatments and cryogenic treatments, electropolishing) [3, 26, 28, 30].

CONCLUSION

Based on the results of this study, it can be concluded that production defects or impurities (one or more) were observed on all new tested instruments. The most common type of irregularity was the existence of fretting, debris and metal strips on the working part of instruments. No organic debris was observed on electropolished surface of BioRaCe instruments. The results of our study indicate that cleaning and sterilization of instruments before the first use is mandatory. However, further research is needed in order to start manufacturing instruments without defects and impurities.

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Karakteristike površine novih rotirajućih nikl-titanijumskih instrumenata – SEM-EDS analiza

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KRATAK SADRŽAJ

Uvod Moderna endodontska procedura podrazumeva upotrebu mašinskih rotirajućih Ni-Ti instrumenata tokom hemomehaničke obrade kanala korena.

Cilj ovog rada je bio da se primenom SEM-EDS metode analiziraju površine novih (neupotrebljenih) rotirajućih endodontskih instrumenata i utvrdi učestalost pojave proizvodnih defekata ili nečistoća na njihovim radnim površinama.

Materijal i metoda U istraživanje je uključeno pet različitih novih setova mašinskih endodontskih Ni-Ti instrumenata: K3, Mtwo, ProTaper Universal, HyFlex i BioRaCe. Radni deo svakog instrumenta je podvrgnut SEM-EDS analizi (uvećanja ×150 do ×2000), čime su utvrđene morfološke karakteristike površine instrumenata i hemijski sastav zatečenih nečistoća. Statistička analiza je urađena primenom Fišer testa ($p < 0,05$).

Rezultati Rezultati SEM-EDS analize su pokazali da ne postoji nijedan novi instrument bez defekta na svojoj površini. Najučestaliji defekti su uočeni kod K3 (27,43%) i ProTaper Universal grupe (27,21%), a najmanje ih je bilo na BioRaCe instrumentima (7,67%). Najzastupljeniji tip defekata kod testiranih instrumenata je bilo prisustvo žljebova. Osim toga, utvrđeno je i prisustvo debrisa i metalnih opiljaka na svim instrumentima, dok je korozija radnog dela primećena samo kod K3, ProTaper Universal i Mtwo sistema u malom procentu.

Zaključak Na osnovu rezultata ovog istraživanja može se zaključiti da su na svim ispitivanim instrumentima uočeni proizvodni defekti, od kojih su najzastupljeniji žljebovi. Takođe su registrovane i nečistoće kao što su debrisa i metalni opiljci. Na elektropoliranoj površini BioRaCe instrumenata nije uočeno prisustvo organskog debrisa, ali je registrovan mali procenat ostalih tipova defekata.

Ključne reči: Ni-Ti instrumenti; proizvodni defekti; SEM-EDS

UVOD

Primena mašinskih nikl-titanijumskih (Ni-Ti) instrumenata sa svojstvima pamćenja oblika, izrazite biokompatibilnosti i otpornosti na koroziju, uvela je endodontsku proceduru u novu eru. Rotirajući Ni-Ti instrumenti su omogućili bržu i efikasnu preparaciju, odnosno smanjenu mogućnost pojave proceduralnih grešaka tokom čišćenja i oblikovanja kanala različite kanalske morfologije [1]. Brojne inovacije u dizajnu instrumenata, tokom poslednjih godina vezane za površinsku i termičku obradu Ni-Ti legure, uticale su na efikasnost i neophodnu sigurnost tokom endodontskog tretmana [2, 3, 4].

U izradi endodontskih instrumenata koristi se Ni-Ti legura u odnosu 56 : 44 = Ti : Ni, čime se postiže njihov ekviatomski odnos. Iako je samo jedan proizvođač (Dentsply, Maillefer Instruments SA, Ballaigues, Švajcarska) objavio apsolutni sastav i detaljan tehnološki proces njihove izrade, pretpostavlja se da je to najbolji odnos koji leguri daje superelastična svojstva [5].

Proizvodnja Ni-Ti mašinskih endodontskih instrumenata je mnogo komplikovanija u poređenju sa procesom izrade čeličnih instrumenata koji se izrađuju hladnim uvijanjem prethodno profilisanih žičanih konusa [2]. Ni-Ti instrumenti nastaju specifičnim brušenjem, odnosno urezivanjem određenog profila u centralno stablo Ni-Ti žice [5]. Novije tehnike proizvodnje uključuju kombinaciju termičke obrade legure i istovremenog uvijanja, radi veće fleksibilnosti i bolje otpornosti na torziju i ciklični zamor [3]. Iako se u procesu izrade komplikovanog dizajna Ni-Ti instrumenata koristi savremena kompjuterska tehnologija, često se javljaju površinski nedostaci u vidu žljebova, jamičastih udubljenja, pukotina i nečistoća koji mogu povećati vulnerabilnost na frakturu [6]. Uočeno je da defekti na površini deluju kao tačke koncentracije napona, dovode do inicijacije i širenja pukotina, odnosno čestih pojava frakture tokom aktivacije instrumenta [7].

Na površini novih endodontskih instrumenata se mogu naći i različiti metalni ostaci i nečistoće organskog i neorganskog porekla. Tokom instrumentacije ovi metalni opiljci se mogu inkorporirati u dentinski zid ili potisnuti u periapeksno tkivo i izazvati alergijsku reakciju [8]. Upotreba instrumenata na kojima su se zatekle organske nečistoće nosi rizik i od moguće unakrsne infekcije [9].

Cilj ovog rada je bio da se primenom skenirajuće elektronske mikroskopije sa energetsko-disperzivnom spektrometrijom (SEM-EDS) analiziraju površine novih rotirajućih endodontskih instrumenata i utvrdi učestalost pojave proizvodnih defekata ili nečistoća na njihovim radnim površinama.

MATERIJAL I METOD

U istraživanju su korišćena po tri osnovna seta (svaki set po šest instrumenata) pet različitih sistema mašinskih endodontskih Ni-Ti instrumenata: K3 (SybronEndo Co, USA), Mtwo (VDW, Munich, Germany), ProTaper Universal (Dentsply Maillefer, Switzerland), HyFlex (Coltene Whaledent gruppe, Switzerland) i BioRaCe (FKG DENTAIRE Swiss Dental Produkts, Switzerland) (Tabela 1).

Skenirajuća elektronska mikroskopija sa energetsko-disperzivnom spektrometrijom (SEM-EDS) realizovana je u Laboratoriji za SEM Rudarsko-geološkog fakulteta Univerziteta u Beogradu, na SEM-u tipa JEOL JSM-6610LV, Japan. Instrumenti su analizirani bez pripreme (direktno iz fabričkog pakovanja). Izrađeni su snimci pomoću detektora za sekundarne elektrone (SE snimci – *second electron*) na uvećanjima u rasponu od 150 do 2000 puta. Hemijska analiza je urađena na nepoliranim uzorcima pomoću EDS detektora (tip X-Max Large Area Analytical Silicon Drifted spectrometer, Oxford Instruments) uz upotrebu

unutrašnjih standarda. Dobijeni hemijski sastav je predstavljen kao sadržaj hemijskih elemenata u masenim procentima (wt%), normalizovan na 100%. Granica detekcije je za većinu elemenata iznosila oko 0,1 wt%. Ovako urađena hemijska analiza se smatra semikvantitatvnom, jer je urađena na nepoliranim površinama.

Ukupno je napravljeno 540 snimaka apeksne i srednje trećine instrumenata iz dva različita pravca. Za svaku površinu instrumenta napravljena su po tri SE snimka. Snimke su pregledala dva istraživača, a usaglašavanje njihovih rezultata izvršeno je analizom Cohen Kappa.

Primenjena je kvalitativna analiza prisustva različitih nepravilnosti i grešaka na radnoj površini Ni-Ti instrumenata u skladu sa preporukama Kristine Egert i sar. [10]. Na instrumentima je registrovano prisustvo: jamičastih udubljenja, žljebova, mikrofraktura, fraktura, metalne uglačanosti, metalnih opiljaka, tupih sečivnih ivica, korozije, debrija i uočen je prekid sečivnih ivica.

Statistička analiza dobijenih rezultata je urađena primenom Fišerovog testa ($p < 0,05$).

REZULTATI

Rezultati SEM-EDS analize novih Ni-Ti setova prikazani su u tabelama 2, 3 i 4 i na slikama 1–9.

Analizom SE snimaka utvrđeno je postojanje kontaminacije na površini radnog dela ispitivanih instrumenata, a naknadnom EDS analizom je utvrđen njen hemijski sastav. Na taj način je urađena podela na instrumente kontaminirane debrisom i kontaminirane metalnim opiljcima. Primeri SEM-EDS analize sa izgledom izvršenog spektra su dati na slikama 1 i 2.

EDS analiza u tački 1 (spektar 1, Slika 1, Tabela 2) pokazuje da je dominantni element u uzorku ugljenik (88,1 mas%) sa malim sadržajem kiseonika (1,5 mas%), te je nečistoća na ovom ProTaper Universal instrumentu okarakterisana kao debris organskog porekla. Sadržaji nikla i titanijuma (spektar 1) odražavaju sastav instrumenta.

Na osnovu EDS analize nečistoća na ovom K3 instrumentu je okarakterisana kao kombinacija debrisa organskog porekla i kontaminacija metalnim opiljcima.

Pojava nikla i titanijuma u analizi 1 (spektar 1, Slika 2) i nešto više u analizi 2 (spektar 2, Slika 2), kao i u prethodnom slučaju, predstavlja distribuciju ovih elemenata u strukturi samih instrumenata.

Rezultati SEM-EDS analize ukazuju na najučestaliju pojavu defekata i nečistoća u sistemima K3 (27,43%) i ProTaper Universal grupe (27,21%). Pojava defekata i nečistoća na novim Ni-Ti instrumentima je u Mtwo grupi zabeležena u 20,43%, a u HyFlex grupi u 17,2%. Najmanji procenat zastupljenosti defekata na apikalnoj i srednjoj površini pokazali su BioRaCe setovi (7,67%). Analizirajući rezultate, uočava se veća pojava defekata na apikalnim površinama radnog dela testiranih instrumenata (53,49% na apikalnoj i 46,51% na srednjoj trećini) (Tabela 4).

Na površini radnog dela novih K3 instrumenata uočeno je prisustvo jamičastih udubljenja i žljebova (apikalna i srednja trećina 100%), prisustvo defekta u vidu metalnih opiljaka (apikalna trećina 83,33%, srednja trećina 38,89%) i pojava korozije samo na jednom instrumentu (apikalna i srednja trećina) (Slika 3). Debris je uočen na apikalnoj trećini kod 77,8% instrumenata i na srednjoj trećini kod 44,44% K3 instrumenata.

SEM-EDS analiza Mtwo instrumenata je ukazala na prisustvo metalnih opiljaka na apikalnoj (50%) i na srednjoj trećini (22,2%). Defekt u vidu korozije je primećen samo na jednom (najtanjem) instrumentu (10/0,4), u njegovoj srednjoj trećini (Slika 4).

Zatupljenost sečivne ivice Mtwo instrumenata je uočena na apikalnoj površini jednog instrumenta (25/0,6) a defekt u vidu prekida sečivne ivice na srednjoj trećini instrumenta (20/0,6) (Slika 5). Debris (koji je potvrđen EDS analizom) uočen je na svim instrumentima, i u apikalnoj i srednjoj trećini (100%).

Najzastupljeniji defekti na površini radnog dela novih ProTaper Universal instrumenata bile su promene u vidu žljebova (apikalna i srednja trećina, 100%) i jamičastih udubljenja (apikalna trećina 83,3% i srednja trećina 88,8%) (Tabela 4, Slika 6). Metalni opiljci su detektovani na apikalnoj (50%) i srednjoj trećini (38,8%). Defekt na sečivnoj ivici (prekid njenog kontinuiteta) uočen je na jednom, najkoničnijem instrumentu (Sx) (apikalna i sredna trećina), a korozija na apikalnom (11,1%) i srednjem delu (11,1%) (Slika 7). Kontaminacija u vidu debrisa je bila zastupljena na apikalnoj trećini (100%) i u nešto više od polovine (55,5%) u srednjoj trećini ProTaper Universal setova.

Najzastupljeniji defekt na novim HyFlex instrumentima je bila pojava žljebova kao prisustvo nečistoće u vidu debrisa na apikalnim i srednjim segmentima svih instrumenata (100%) (Slika 8).

Defekt u vidu mikrofrakture je uočen na apikalnom delu instrumenta (25-0,08), kao i pojava metalnih opiljaka (25-0,04).

Rezultati SEM analize BioRaCe setova pokazuju najučestaliju pojavu metalnih opiljaka (apikalna 50% i srednja trećina 38,8%) i žljebova (apikalna 33,3% i srednja trećina 27,7%) (Tabela 4, Slika 9). Kontaminacija debrijem detektovana EDS analizom je uočena na apikalnoj (11,1%) i srednjoj površini (22,2%) instrumenata.

Statistički značajna razlika u prisustvu žljebova je uočena između intrumenata K3, MTwo, ProTaper i Hyflex između apikalne (za $p < 0,05$) i srednje trećine (za $p < 0,05$).

Značajne razlike vezane za prisusutvo metalnih opiljaka uočene su između K3 i HyFlex instrumenata (za $p < 0,05$), između ProTaper Universal i HyFlex (za $p < 0,05$), MTwo i HyFlex (za $p < 0,05$) i BioRaCe i HyFlex instrumenata (za $p < 0,05$). Razlika je bila značajna i u pojavi metalnih opiljaka u apikalnoj trećini između K3 i BioRaCe grupe instrumenata (za $p < 0,05$), između K3 i MTwo (za $p < 0,05$), odnosno K3 i ProTaper grupe (za $p < 0,05$).

U srednjoj trećini statistički značajna razlika u pojavi metalnih opiljaka uočena je između HyFlex grupe i K3, BioRaCe, ProTaperUniversal i MTwo intrumenata (za $p < 0,05$). Razlika je bila značajna i između apikalne i srednje trećine instrumenata K3 (za $p < 0,05$).

Razlika je bila značajna i u vrednostima pojave debrisa u apikalnom segmentu između K3 i ProTaper Universal grupe (za $p < 0,05$), K3 i MTwo grupe (za $p < 0,05$), odnosno između K3 i HyFlex grupe instrumenata (za $p < 0,05$). Statistički značajna razlika je uočena i između K3 i BioRaCe grupe (za $p < 0,05$), ProTaperUniversal i BioRaCe grupe (za $p < 0,05$), MTwo i BioRaCe grupe (za $p < 0,05$), odnosno HyFlex i BioRaCe grupe (za $p < 0,05$). U srednjoj trećini razlika je bila značajna između K3 i MTwo (za $p < 0,05$) i K3 i HyFlex instrumenata (za $p < 0,05$). U ProTaper Universal grupi statistički značajna razlika u pojavi debrisa uočena je između apikalne i srednje trećine (za $p < 0,05$).

DISKUSIJA

SEM analiza prisustva različitih nepravilnosti površine, proizvodnih grešaka i kontaminacija novih Ni-Ti rotirajućih instrumenata pokazala je da ne postoji nijedan ispitivani instrument bez defekta ili nečistoća na radnoj površini. U ovoj studiji je utvrđeno prisustvo različitih defekata i nečistoća kod svih novih Ni-Ti instrumenata (pet različitih komercijalnih setova), pri čemu je nešto veća zastupljenost uočena u njihovoj apikalnoj trećini. Iako na fabričkom pakovanju postoji potvrda o njihovoj sterilnosti, prisustvo defekata i prljavštine na aktivnom delu novih Ni-Ti instrumentima je dokazana realnost, što je dokumentovano rezultatima različitih studija [8, 10–19].

Komplikovani proces mašinske obrade početne Ni-Ti žice često uzrokuje nastanak površinskih deformacija i pukotina usled tragova glodanja i obrade, ali i pojavu uglačanih površina na sečivnim ivicama instrumenata [5, 10].

Površinske promene mogu kompromitovati sečivnu efikasnost instrumenata i postati mesta za mogući nastanak korozije. Takođe, ove tačke predstavljaju mesta inicijacije defekata, doprinoseći degradaciji mehaničkih svojstava, i pojavi mikro ili komletne frakture tokom njihove kliničke upotrebe [5, 7, 13]. Arens i saradnici su prezentovali zanimljivu studiju o učestalosti frakture posle prve upotrebe novih Ni-Ti instrumenata (0,9%), dok Shen kao uzrok ove komplikacije navodi neadekvatnu manipulaciju i postojanje proizvodnih defekata [12, 13].

Usled većih sila i veće brzine koje su neophodne za obradu Ni-Ti legure, postoji mogućnost paljenja strugotine i formiranja otvrdnutih mesta. To su delovi veće tvrdoće koji se teže obrađuju, te predstavljaju zone sa većom verovatnoćom za pojavu deformacija i fraktura [16].

Način na koji dolazi do formiranja defekata tokom nastanka legure Anusavice i Phillips su pripisali karakterističnoj faznoj transformaciji i prekristalizaciji Ni-Ti legure [20]. Prekristalizacija je pojava promene tipa rešetke u zavisnosti od temperature (npr. titan pri 882°C prelazi iz heksagonalne u monokliničnu strukturu), pri čemu brzina kristalizacije utiče na pravilnost strukture kristala [20].

Najučestaliji tip površinskih iregularnosti na radnoj površini novih instrumenata u ovoj studiji je bila **pojava žljebova**. Klinički značaj pojave žljebova je u povećanju mogućnosti njegovog ušrafljivanja (usled trenja koje postoji zbog neravne površine) i povećane incidence loma [13].

Prisustvo metalnih opiljaka kao posledica proizvodnog procesa zapaženo je na radnoj površini svih ispitivanih instrumenata. Korelacija između velike zastupljenosti metalnih opiljaka i veće koničnosti instrumenata K3 u ovoj studiji (koničnost veća od 0,06) u saglasnosti je sa rezultatima Marendinga i saradnika, koji su ukazali da metalni opiljci nastaju kao rezultat proizvodnog procesa i to češće kod debljih i koničnijih instrumenata [11].

Primenom SEM analize Van Eldik je sa saradnicima uočio prisusutvo velike količine metalnih opiljaka na površini novih Ni-Ti instrumenata, neposredno po otvaranju iz originalnog pakovanja [14]. Ovaj tip kontaminacije dovodi do smanjenja sečivne efikasnosti, a metalni opiljci se tokom instrumentacije mogu zadržati u dentinskim zidovima kanala ili u periapeksnom tkivu. Van Eldik je dokazao da moguća kontaminacija periapeksnog tkiva ovim metalnim opiljcima može redukovati tok reparacije tkiva i kompromitovati uspeh endodontske terapije [14]. Stefanesku i saradnici u svojim rezultatima dokazuju da čestice metala mogu biti transportovane tokom instrumentacije i aktivne irigacije kroz apeksni foramen i uzrokovati alergijsku reakciju periapeksnog tkiva [8]. Dokazano je da joni metala kao potencijalni haptenski alergeni mogu uzrokovati reakcije tipa 1, uz moguću trenutnu ili odloženu dermalnu ili sluzokožnu reakciju. Alergijske reakcije u endodonciji izuzetno su retke, ali posledice alergijskih reakcija, kao što su simptomi odloženog apikalnog zarastanja, uporne nelagodnosti nakon opturacije kanala, mogu značajno povećati njihov broj [8].

Prisustvo debrisa je, takođe, uočeno na radnim površinama svih tipova ispitivanih Ni-Ti instrumenata. Titanijumske legure se teško obrađuju zbog svoje elastičnosti, te zahtevaju veće sile rezanja u odnosu na čelik. Ni-Ti legura se intenzivno lepi za alat kojim se obrađuje, pa se zaštita materijala postiže oksidiranjem površine ili metalnim premazima, koji se nakon obrade uklanjaju hemijskim putem, koji može zaostati na njihovoj površini [16]. Elektropoliranjem površine instrumenata BioRaCe povećava se sečivna efikasnost, a istovremeno smanjuju defekti nastali u proizvodnom procesu i smanjuje mogućnost kontaminacije debrisom [21]. Značajnija učestalost pojave debrisa u apikalnoj trećini K3 i ProTaper Universal instrumenata u odnosu na njihovu srednju trećinu potvrđuje veću kontaminiranost apikalnog segmenta zbog kompleksnije izrade gracilnijeg apikalnog dela. Ovaj nalaz je u saglasnosti sa studijama koje su sproveli Eggert i Alapati, a koje su ukazale na veću pojavu debrisa u apikalnom segmentu novih Ni-Ti instrumenata [10, 22].

Defekti radne površine u vidu jamičastih udubljenja uočeni su samo kod dve grupe novih instrumenata ali u visokom procentu (K3 i ProTaper Universal). Pojava jamičastih šupljina nastaje tokom proizvodnog procesa, jer se tokom topljenja elementarnog nikla i titanijuma razlikuju brzine njihove međusobne difuzije pri zagrevanju, što dovodi do formiranja praznina [23]. Atomi nikla difunduju brže u titan nego atomi titanijuma u obrnutom smeru. Dakle, maseni transport nije izbalansiran, što može dovesti do stvaranja praznina u niklu nakon legiranja. Ove šupljine su poznate pod nazivom Kirkendalova poroznost ili Kirkendalov efekat [23].

Nagumo je izneo dokaze o značaju ovih defekata na mehaničke karakteristike Ni-Ti instrumenata, kao i tačnom mehanizmu njihovog uticaja [24]. On je zapazio da legura može da apsorbuje vodonik iz pljuvačke i formira hidridne veze sa atomima Ni-Ti rešetke koje su stabilne na sobnoj temperaturi. Ovakva promena molekularne strukture dovodi do promene fizičkih svojstava legure izazivajući hidrogensku poroznost. Asaoka je, takođe, ukazao da difuzija vodonika kroz Ni-Ti leguru formira faze hidrida na površini materijala koji ima krtiju strukturu [25]. Ovaj novonastali hidridni sloj na aktivnoj površini Ni-Ti instrumenta je različite debljine i tokom kliničkog rada dolazi do formiranja mikropukotina. Obezbeđenjem apsolutno suvog radnog polja, ovaj mehanizam nema značaja, ali može imati uticaja tokom procesa čišćenja i sterilizacije instrumenata, kada su instrumenti izloženi dužem dejstvu jonizujućih tečnosti [25].

Korozija radnog dela Ni-Ti instrumenata nije primećena na HyFlex i BioRaCe instrumentima, a i u ostalim grupama je zapažena u malom procentu. Mali stepen zastupljenosti korozije na Ni-Ti instrumentima potvrđuje otpornost ove legure na koroziju, ali i neizlaganje novih instumenata korozivnim faktorima [26].

Nalazi defekata na površini novih Ni-Ti instrumenata u vidu **zatupljenosti sečivne ivice**, **prekida sečivne ivice i mikrofrakture** samo potvrđuju problematiku njihove proizvodnje. Mikrofrakture na novim instrumentima su, prema istraživanjima Marendinga i Barbakowa, rezultat proizvodnog procesa većih i koničnijih, manje fleksibilnih instrumenata [11]. Pukotine ili mikrofrakture su po većini istraživača najopasniji defekti koji turpija može imati [24, 25]. Ukoliko se instrumenti sa ovim defektom aktiviraju u kanalu, prilikom rotacije i ušrafljivanja, oni se lome odmah. Mikropukotina utiče na veliku osetljivost instrumenta na akumulaciju cikličnog zamora i neminovne frakture [11, 24, 25].

Naknadna termička obrada gotovih Ni-Ti instrumenata (HyFlex) potencijalno nudi najperspektivniju metodu proizvodnje rotirajućih instrumenata [27]. Ovi instrumenti nemaju memoriju oblika koju imaju tradicionalni Ni-Ti instrumenti, a poseban termomehanički postupak značajno povećava njihovu fleksibilnost [28]. Rezultati istraživanja u ovoj studiji pokazuju najmanju kontaminiranost Hyflex sistema metalnim opiljcima. Mala zastupljenost ove kontaminacije se može objasniti njihovom specifičnom termičkom obradom koja smanjuje nepravilnosti na njihovoj površini. Toplotni tretman osim promene mikrostrukture (povećana fleksibilnost), dovodi do pojave čistije i pravilnije površine ovih instrumenata [28, 29, 29].

Prateći rezultate ove studije, zapaža se i značajno manja zastupljenost defekata u grupi BioRaCe. Ovaj nalaz je u saglasnosti sa rezultatima istraživanja o značajnom smanjenjenju površinskih nepravilnosti elektropoliranih instrumenata [3]. Elektropoliranjem se tokom procesa proizvodnje stvara homogeni oksidni sloj koji smanjuje pojavu površinskih defekata i povećava otpornost na koroziju i lom [27]. Elekropolirana površina instrumenata je vidljivo sjajnija u odnosu na netretiranu površinu [26]. Uvođenjem struje kroz rastvor dolazi do formiranja tankog pasivnog sloja i otapanja površine u elektrolit, što takođe dovodi do selektivnog uklanjanja površinskih defekata [4].

U svrhu poboljšanja mikrostrukture radne površine Ni-Ti instrumenata i poboljšavanja mehaničkih osobina, fleksibilnosti, otpornosti na zamor, odnosno sečivne efikasnosti, proizvođači poslednjih godina primenjuju različite tehnike (jonska implementacija, plazma imerzija, formiranje premaza titanijum-oksida, termalna nitridacija, termalni tretmani i kriogeni tretmani, elektropoliranje) [3, 26, 28, 30].

ZAKLJUČAK

Na osnovu rezultata ovog istraživanja može se zaključiti da su na svim ispitivanim novim instrumentima uočeni proizvodni defekti ili nečistoće (po jedan ili više). Najučestaliji tip nepravilnosti je bilo postojanje žljebova, debrisa i metalnih opiljaka na radnom delu instrumenata. Na elektropoliranoj površini BioRaCe instrumenata nije uočeno prisustvo organskog debrisa. Rezultati ove studije ukazuju na potrebu obaveznog čišćenja i sterilizacije instrumenata pre prve upotrebe ali su neophodna i dalja istraživanja u cilju dobijanja instrumenata bez defekata i nečistoća.

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Viruses as potential nanomachines

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SUMMARY

In this paper viruses are considered as very efficient nano-machines that produce numerous copies of them. Observing these nano-architectures, the question arises: which molecular forces and processes make up the set of such structures, given that they are extremely inspiring for development of new technologies at the nano level. There is a need for deep understanding of individual molecular building blocks and their structures, properties of their assemblies and dynamic behavior.

Keywords: viruses; bacteriophages; DNA nanomachines

INTRODUCTION

The high efficiency of bacteriophage-based therapy in suppressing colonies of many resistant bacteria, as in the case of cystic lung fibrosis causing their very serious infection, has made bacteriophages one of the most important new advances in molecular biology, biophysics and bio-nanotechnology. Filamentous viruses, such as bacteriophage M13, have a virion architecture that allows them to precisely build ordered two-dimensional and three-dimensional structures without damaging at the level of nanometer dimensions. This would not be possible without a detailed knowledge of the protein coat's structure and dynamics during the virus replication cycle. The results of spectroscopic studies show the critical role of protein incorporation into the membrane, both during the infectious entry of the virus into the host cell and during the assembly of a new virion in the host membrane. The protein is efficiently incorporated into the membrane using a strong C-terminal interfacial anchor. A simple tilt mechanism and subtle structural adjustment at the very end of its N-terminus provide a favorable thermodynamic association of proteins in the lipid bilayer [1, 2, 3].

Viruses, especially bacterial viruses or bacteriophages, play a key role in controlling biological systems. Advances in molecular biology over the last 50 years have been built largely due to the study of bacteriophages. Restriction endonucleases, which form the basis of molecular cloning, were developed after studies on phage infection. Many phage enzymes provide tools to the molecular biologists who study the pathways of replication, transcription, translation, and transport (Figure 1). Phage display techniques provide them with a powerful methodology for identifying and optimizing ligand antibodies and other biomolecules. Modern application of bacteriophages in engineering materials puts them in the foreground of new nano-technological devices [3, 4, 5]

BASIC CONSIDERATIONS

Filamentous bacteriophages in modern biophysics have served as a model system for the development and application of spectroscopic methods suitable for biological supramolecular assemblies. Filamentous phage envelope proteins, which can be easily synthesized in the laboratory, have two primary roles during the replication cycle as membrane proteins and as the main structural elements of phage particles, thus showing the astonishing possibility of finding two such different roles in the bacteriophage replication cycle. Learning about protein structures in different environments allows us to fully understand their latest nano-technological applications in nano-biology and bio-nanotechnology. Filamentous phages make up a family of viruses that have about ten genes. The relative simplicity of these viruses and the ease with which they can be genetically modified have made them extremely useful models for studying macromolecular structures and interactions. All filamentous strains of bacteriophages have a similar virion structure and life cycle. M13 filamentous phages are best-studied biochemically, genetically and biophysically.

M13 bacteriophage causes chronic infections, with infected cells continuing to grow and divide, albeit at a lower rate than normal. The phage is a long thread-shaped particle, 6.5 nm in diameter and 900 nm long. The flexible strand contains a circular, single-stranded viral DNA genome, protected by a long cylindrical protein coat. The major envelope proteins that make up the tube around the viral DNA, in the folding helix, are oriented so that the N end is on the outside of the envelope, and the C terminates by binding to the DNA inside the envelope. The major protein coat's hydrophobic domain is located in the central part of the protein and connects the protein coat to the adjacent protein coat to form viral particles. During reproduction, the main envelope protein is involved in

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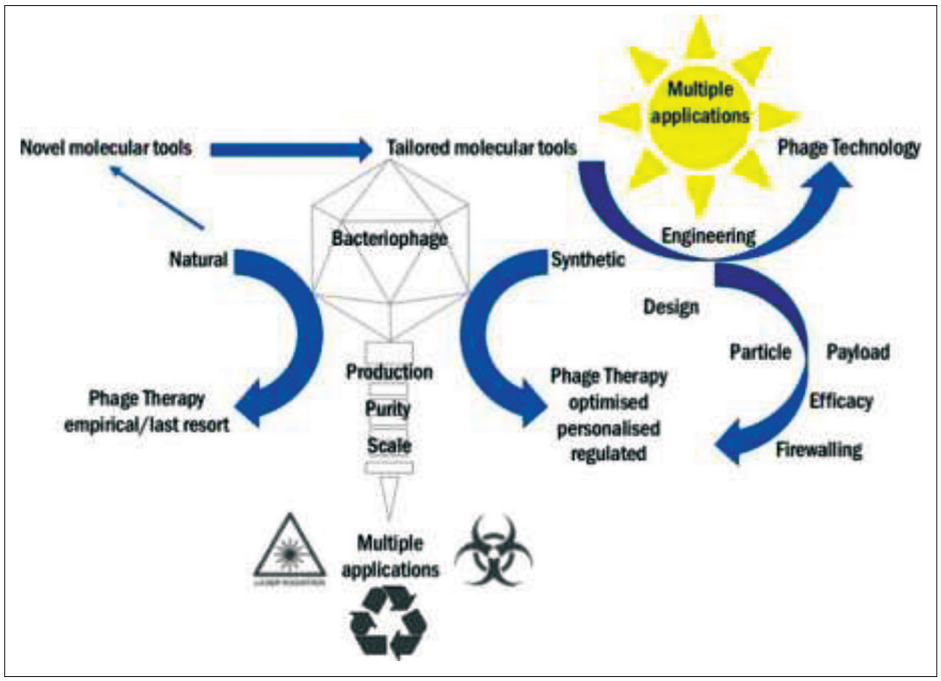

Figure 1. Bacteriophages can be envisaged as natural microbial control agents and machines for targeted synthetic genetic programming. The encoded proteins, as well as the structures of phages, offer a multitude of possibilities as outlined in this introductory chapter and detailed in the volume.
Slika 1. Bakteriofagi mogu služiti kao prirodni mikrobni kontrolni agenti, kao i nanomašine za targetirano sintetsko genetsko programiranje. Kodirani proteini, kao i struktura faga, nude mnogo mogućnosti, koje su opisane u uvodnom delu i daljem tekstu.

various molecular processes that take place in different cell environments. After entering the cell, the major envelope protein is removed from the phage particle and precipitated in the inner membrane of the host. Viral DNA enters the cell and converts to a double-stranded replicative form by the host enzyme. Progeny DNA is replicated by a cyclic mechanism, assembling with the gp5 protein to replicate and self-assemble the virus into an elongated intracellular nucleoprotein complex [5–8].

The replication of self-assembled protein (which covers and protects viral DNA inside the cell) is then replaced by envelope proteins (which cover and protect viral DNA outside the cell) in the cell membrane, where the virion is extruded through the membrane by viral and host proteins. The new envelope protein is synthesized as the envelope protein, which is the major envelope protein's precursor. It contains an additional leading amino acid sequence necessary for insertion into the cytoplasmic membrane. The envelope molecule is inserted into the membrane and then the additional leader sequence is cut off with the host peptidase. The resulting mature transmembrane protein coat is stored in the inner membrane before its use in the phage assembly process. Upon completion of the phage assembly, it is released into the medium and is ready to attack the new host cell. A key element in this whole process is the main envelope protein, which is responsible for protein-protein, protein-lipid and protein-DNA interactions during the assembly and disassembly of macromolecules [6–9].

The structure of the phage-bound coat protein has been known for many years, based on the results of fiber X-ray diffraction experiments: This structure is an almost perfect a-helix, with 4-5 flexible unstructured amino acid residues at the N-terminus exiting the phage sheath into the aqueous phase. The lysine-rich C terminus is linked to viral DNA phosphate groups, allowing phage particles to be elongated by simply inserting more DNA into the viral genome, with each protein coat subunit being approximated by a slightly curved α-helix measuring about 1 to 7 nm. Diffraction data show that the axis of the α-coil is set at a small angle to the axis of the virion, which is very precisely bounded by adjacent subunits. They form an overlapping spiral string with widely connected side chains. Envelope protein subunits possess continuous apolar domains that are held together in a virion by hydrophobic interactions between them [6–9].

The highly organized crystalline environment of the main envelope protein in the phage particle allows obtaining detailed structural and topological information about it. Based on NMR, the protein's determined structure showed that it possesses two helices in the lipid bilayer. The major coat protein is a single-coil monotopic protein that should be inserted into the membrane during the phage life cycle, have a stable thermodynamic connection to the membrane, and then leave the membrane during the assembly process. Assuming that the transmembrane coil is aligned with the normal lipid bilayer, then the L-shaped protein conformation is probably the best topology for the membrane-inserted protein due to optimal hydrophobic interactions. Because determining the topology of proteins in lipid bilayers is particularly important for virus self-assembly and for the ability of proteins to serve as a means of displaying peptides, new biophysical tools have been developed to study protein structure, topology, and lipid bilayer dynamics [5–9].

POTENTIAL PHAGES APPLICATIONS IN NANOTECHNOLOGIES

Phages could serve as probes in a new generation of real-time food safety control and environmental monitoring sensors. As detector elements, they are superior to polyclonal and monoclonal antibodies, because they are cheap, highly specific, selective and resistant to adverse environmental conditions. In addition, the unique structure of M13 bacteriophages has been used as a biological model for nanotechnology, such as the directed synthesis of semiconductor / magnetic nanowires and lithium-ion battery electrodes. Filamentous viruses enable the organization of various nanomaterials into periodically arranged hierarchical structures, such as viral rings and wires, which have an electronic, optical and biotechnological application. In materials science this approach is used to create new peptides that can bind to selected technical materials. Bacteriophages M13 with a protein coat, since they can bind to semiconductor and magnetic materials, were used as templates for the growth and organization of nanowires, serving as a template for the synthesizing of monocrystalline ZnS and CdS, and chemically arranged nanowires CoPt and FePt. The nuclear peptides incorporated into the M13 protein coat thus provide a pattern for the directed preparation of semiconductor and magnetic materials [4–7].

Some of their applications could be extremely important for tissue engineering and regenerative medicine, because such structures mimic the native extracellular matrix, which consists of a fibrous protein network and provides cells with physical support [5, 6, 7]. Recent findings indicate that the major coat proteins, which are the major building blocks of proteins, do not change their shape significantly during membrane self-organization, resulting in a very efficient mechanism for reducing energy expenditure. Any nanotechnology derived from M13 bacteriophages should take into account their basic physicochemical rules. Although many aspects of the biogeneration of nanostructures have not yet been comforted, it is expected that new advances in biophysical techniques will increase our molecular insight in the coming decades. If we understand that we are just at the beginning of a new era of bio-nanotechnology, it is clear that the future of nano-machines based on viruses is extremely bright [7, 8].

Large, multi-unit, protein complexes are initially assembled with weak interactions, which are very suitable for intracellular complexes because they are temporarily assembled and disassembled to activate mutually coordinated cellular functions. Virus particles, like other self-organizing systems, although initially formed by weak interaction, since part of their life cycle is extracellular, they must find a way of organization that guarantees them strong stability. Due to such conflicting requirements, the initial particle, usually called Procapsid or Provirion, matures over time, experiencing various conformational changes within the capsid. Some viruses, such as nodaviruses, undergo subtle autocatalytic cleavage of capsid subunits after leaving the cell, resulting in stabilizing virus particles and increasing their infectivity. In enveloped viruses, such as tetraviruses, there is a dramatic reorganization of the particle during maturation, resulting in smaller virus's autocatalytic cleavage [9, 10, 11].

DNA-BASED SYNTHETIC MOLECULAR MACHINES

DNA nano-machines are made by self-assembly, using techniques that rely on sequence-specific interactions that bind complementary oligonucleotides into a double helix. They are activated by interaction with certain signaling molecules or changes in their environment, triggering an appropriate response, to an external trigger, to serve as an intelligent molecularly sensitive drug delivery or controlled chemical synthesis. Biological molecular motors that carry cargo within cells have inspired the construction of rudimentary DNA walkers that stretch along self-installed pathways autonomously, gaining energy by catalyzing DNA or RNA fuel reactions [12, 13].

The exceptional specificity of interactions between complementary nucleotides makes DNA a useful building material, as due to the interaction between its short chains, it is possible to control the design of their basic sequences reliably. The construction of branched connections between the double helix enables the creation of complex three-dimensional objects. One way to take advantage of this extremely precise architectural control is to use self-assembled DNA patterns to position functional molecules, serving as molecular electronic circuits, optical devices, or enzyme networks. DNA is not a natural choice of material for building active structures, as it does not possess the structural and catalytic versatility of proteins and RNA, which are more suitable for that purpose [13, 14] (Figure 2).

MOLECULAR SWITCHES

The simplest active DNA nanostructures are switches or actuators, which act between two conformations, and their movement is stimulated by changes in temperature, ionic conditions or by binding of a signaling molecule, which is most often a DNA coil. Conformation changes are caused by changes in the environment, where the rotational movement is conditioned by a change in DNA twisting, in which double-stranded DNA with the sequence (CG) n is transferred from the usual right-handed helix (B-DNA) to the left-handed conformation (Z-DNA). These changes may be caused by high salt concentration and low temperature. Forster resonant energy transfer (FRET), which induced transition between the corresponding fluoro-phores and DNA helix separation on a nanometer scale with highly efficient energy transfer, mediated by dipole-dipole interaction, represents possible mechanism of the FRET based nano*machine [13–16]. Yang and co-workers converted the changes in DNA twisting into linear motion. Their device consisted of a closed loop of double-stranded DNA attached to the opposite arms of a four-legged holistic joint, whereby the holiday joint could migrate by breaking up identical base pairs in one pair of opposite arms and transforming them into another pair. The change in DNA conformation within the loop was initiated by the addition

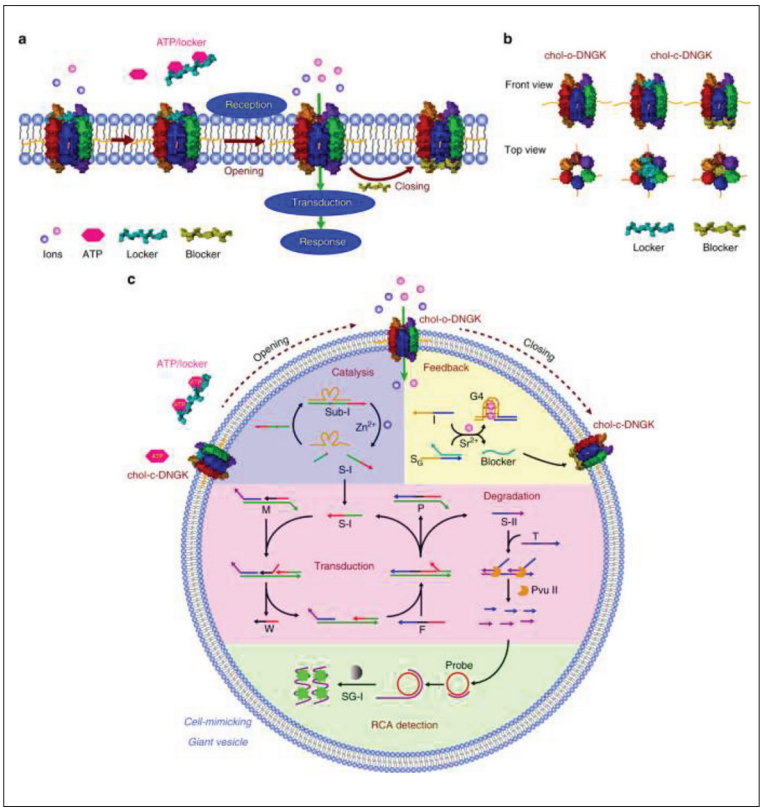

Figure 2. Biomimetic giant vesicle engineered for the construction of an artificial molecular signaling system (AMSsys)
Slika 2. Biomimetička velika vezikula stvorena za kreiranje veštačkog molekularnog signalnog sistema (AMSsys)

of ethidium bromide, which binds between adjacent base pairs, extending and partially unwinding the double helix. The resulting stress is alleviated by the migration of the node, ie the shortening of the protruding arms of the connections inside the loop, which allows its elongation without changing the total number of threads in it. A single chain of cytosine bends into an i-motif, a compact three-dimensional structure held by base pairs of protonated cytosines. In the presence of a second complementary strand of DNA, competition occurs between the i-motif and the elongated double helix formed by the hybridization of two strands [15, 16].

The transition of i-motif into a two-way transition produces mechanical work. If one surface of the silicon cantilever is coated with bonded coils containing cytosine, then a compressive surface stress bends that bends the cantilever. Such stress due to hybridization of the complementary coil with the cantilever is not expected, the effect is present at high salt concentrations, at which the interactions at a distance are comparable to the observed separation of the coils. Conformational change caused by pH-dependent binding of a single strand of DNA to a duplex to form a triple-helical structure, which is the basis of the design of a nano-mechanical actuator (Figure 2). Active DNA nanostructures are also evolving as sensors, which allow elementary logic operations to be performed on their outputs,

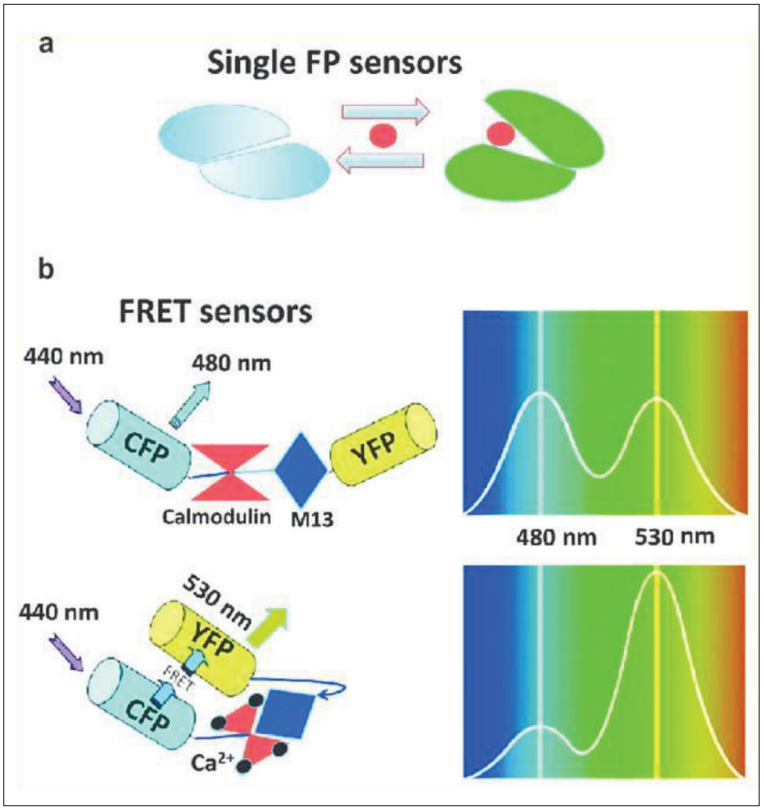

Figure 3. Basic principles of biosensors organization: (a) Single FP-based sensors consist of a protein changes its conformation and luminosity for specific binding to the ligand; (b) FRET sensors consist of two fluorescent proteins, connected by a linker containing in some cases a ligand receptor. In the absence of ligand, fluorescent proteins are far from each other and light releasing by the first protein does not excite the second one. Upon binding of the ligand linker changes its conformation and proteins come together, as a result, the radiation from the first protein excites the second one and its luminosity increases.
Slika 3. Osnovni principi organizacije biosenzora: a) Pojedinačni FP bazirani senzori sastoje se od proteina koji menja svoju konformaciju i luminoznost pri specifičnom vezivanju za ligand;
b) FRET senzori sastoje se od dva fluorescentna proteina, koja su vezana linkerom, u nekim slučajevima ligandom receptora. U odsustvu liganda, fluorescentni proteini su daleko jedan od drugog i oslobađanje svetlosti od strane jednog proteina ne rezultuje ekscitacijom drugog. Nakon vezivanja liganda, linker menja svoju konformaciju i proteini se vezuju. Kao rezultat, radijacija prvog proteina ekscitira drugi i povećava se njegova luminoznost.

which in combination with computers, can be applied to design smart drug delivery systems [15, 16, 17].

CONCLUSIONS

The bacteriophage M13 protein coat is described as the extremely inspiring in nano-machine manufacturing, because it allows the synthesis of particles of the desired functionality, assuming that no basic biophysical principle is violated, especially the ability of the protein to form a stable symmetrical coating and the ability of the protein to insert, anchor and assemble into a new particle virus. This technology is evolving on its own, so it is expected that not only phages but also others similar viruses like smart machines can produce new proteins that could improve the technology itself. This paper also described DNA-based nano-machines, as potential actuators or switches and for intelligent molecularly sensitive drug delivery systems or systems for controlled chemical synthesis.

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Virusi kao potencijalne nanomašine

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KRATAK SADRŽAJ

U ovom radu virusi su predstavljeni kao vrlo efikasne nanomašine koje proizvode brojne sopstvene kopije. Posmatrajući ove nanoarhitekture, postavlja se pitanje koje molekularne sile i procesi čine skup takvih struktura, s obzirom na to da su izuzetno inspirativne za razvoj novih tehnologija na nanonivou. Potrebno je duboko razumevanje pojedinačnih molekularnih gradivnih blokova i njihovih struktura, svojstava njihovih sklopova i dinamičkog ponašanja.

Ključne reči: virusi; bakteriofagi; DNK nanomašine

UVOD

Visoka efikasnost terapije zasnovane na bakteriofagima u suzbijanju kolonija mnogih rezistentnih bakterija, kao što je to slučaj sa cističnom fibrozom pluća koja izaziva vrlo ozbiljnu infekciju, učinila je bakteriofage jednim od najvažnijih novih dostignuća u molekularnoj biologiji, biofizici i bionanotehnologiji. Filamentozni virusi, kao što su bakteriofagi M13, imaju virionsku arhitekturu koja im omogućava preciznu izgradnju uređenih dvodimenzionalnih i trodimenzionalnih struktura bez oštećenja na nivou nanometarskih dimenzija. To ne bi bilo moguće bez detaljnog poznavanja strukture i dinamike proteinskog omotača tokom ciklusa razmnožavanja virusa. Rezultati spektroskopskih studija pokazuju kritičnu ulogu ugradnje proteina u membranu, kako tokom infektivnog ulaska virusa u ćeliju domaćina, tako i tokom sklapanja novog viriona u membrani domaćina. Protein se efikasno ugrađuje u membranu zahvaljujući snažnom C-terminalnom interfacijalnom sidru, koje jednostavnim mehanizmom nagiba i suptilnim strukturnim prilagođavanjem na samom kraju svog N-terminusa obezbeđuje povoljno termodinamičko pridruživanje proteina u lipidnom dvosloju [1, 2, 3].

Virusi, posebno bakterijski ili bakteriofagi, igraju ključnu ulogu u kontroli bioloških sistema. Napredak u molekularnoj biologiji tokom poslednjih 50 godina bazira se velikim delom na proučavanju bakteriofaga. Restrikcijske endonukleaze, koje čine osnovu molekularnog kloniranja, razvijene su nakon studija o infekciji fagom. Mnogi enzimi faga pružaju molekularnom biologu alate koji pomažu u proučavanju puteva replikacije, transkripcije, translacije i transporta. Tehnika prikazivanja faga im pruža moćnu metodologiju za identifikaciju i optimizaciju liganda antitela i drugih biomolekula. Savremena primena bakteriofaga u inženjerskim materijalima stavlja ih u prvi plan kod novih nanotehnoloških dostignuća [3, 4, 5].

OSNOVNA RAZMATRANJA

Vlaknasti bakteriofagi u savremenoj biofizici služili su kao model sistema za razvoj i primenu spektroskopskih metoda pogodnih za biološke supramolekularne sklopove. Proteini ovojnice filamentnih faga, koji se lako mogu sintetisati u laboratoriji, imaju dve primarne uloge tokom ciklusa replikacije, kao membranski proteini i kao glavni strukturni elementi čestica faga, pokazujući tako začuđujuću mogućnost da se nađu dve tako različite uloge u bakteriofagnom ciklusu replikacije. Saznanja o strukturama proteina u različitim okruženjima omogućavaju nam da u potpunosti razumemo njihove najnovije nanotehnološke primene u nanobiologiji i bionanotehnologiji.

M13 bakteriofag uzrokuje hronične infekcije, pri čemu zaražene ćelije nastavljaju da rastu i da se dele, mada sa nižom stopom od normalne. Fag je dugačka čestica u obliku niti, prečnika 6,5 nm i dužine 900 nm. Fleksibilna nit sadrži kružni, jednolančani virusni DNK genom, zaštićen dugim cilindričnim proteinskim omotačem. Glavni proteini omotača čine cev oko virusne DNK, i u preklopnom spiralnom nizu su orijentisani tako da se N kraj nalazi na spoljnoj strani omotača, a C završava vezujući se sa DNK u unutrašnjosti omotača. Hidrofobni domen glavnog proteinskog omotača se nalazi u centralnom delu proteina, i povezuje omotač proteina sa susednim proteinskim omotačem formirajući virusne čestice. Tokom reprodukcije glavni protein omotača uključen je u različite molekularske procese koji se odvijaju u različitim okruženjima ćelije. Nakon ulaska u ćeliju, glavni protein omotača se uklanja iz čestice faga i taloži u unutrašnjoj membrani domaćina. Virusna DNK ulazi u ćeliju i pretvara se u dvolančani replikativni oblik od strane enzima domaćina. DNK se replicira cikličnim mehanizmom, pri čemu se sastavlja sa proteinom gp5 za replikaciju i samoasembliranje virusa u izduženi unutarćelijski nukleoproteinski kompleks [5–8].

Replikaciono-samoasemblirani protein (koji pokriva i štiti virusnu DNK unutar ćelije) zatim se zamenjuje proteinima omotača (koji pokrivaju i štite virusnu DNK izvan ćelije) u ćelijskoj membrani, gde se virion istiskuje kroz membranu uz pomoć virusnih proteina i proteina domaćina. Novi protein se sintetiše kao protein omotača, koji je prethodnik glavnog proteina omotača. On sadrži dodatnu vodeću sekvencu aminokiselina neophodnu za umetanje u citoplazmatsku membranu. Molekul omotača se ubacuje u membranu i potom se dodatna vodeća sekvenca odseca peptidazom domaćina. Nastali zreli transmembranski proteinski omotač se skladišti u unutrašnjosti membrane pre njegove uloge u procesu faga. Nakon kompletiranja faga, on se oslobađa u medijum i spreman je za napad na novu ćeliju domaćina. Ključni element u celom ovom procesu je glavni protein omotača, koji je odgovoran za interakcije protein-protein, protein-lipid i protein-DNA tokom sastavljanja i rastavljanja makromolekula.

Struktura proteina omotača vezanog za fage poznata je već dugi niz godina, na osnovu rezultata eksperimenata rendgenske

difrakcije vlakana. Ova struktura je gotovo savršena a-spirala, na čijim se krajevima nalazi 4-5 fleksibilnih nestrukturiranih aminokiselinskih ostataka koji na N kraju izlaze iz omotača faga u vodenu fazu. C-terminus bogat lizinom povezan je sa virusnim DNK fosfatnim grupama i omogućava izduživanje čestice faga jednostavnim umetanjem više DNK u virusni genom, pri čemu se svaka podjedinica proteinskog omotača može aproksimirati pomoću jedne blago zakrivljene a-spirale dimenzija oko 1 do 7 nm. Podaci difrakcije pokazuju da je osa a-spirale postavljena pod malim uglom u odnosu na osu viriona, koji je vrlo precizno ograničen susednim podjedinicama. One čine preklapajući spiralni niz sa široko povezanim bočnim lancima. Podjedinice proteina omotača poseduju kontinuirane apolarne domene koji se drže se zajedno u virionu hidrofobnim interakcijama među njima [6–9].

Visoko organizovano kristalno okruženje glavnog proteina omotača u čestici faga omogućava dobijanje detaljnih strukturnih i topoloških informacija o njemu. Na osnovu NMR je pokazano da određena struktura proteina poseduje dve zavojnice u dvosloju lipida. Glavni protein omotača je monotopični protein koji pokriva membranu jednom zavojnicom i koji se tokom životnog ciklusa faga umeće u membranu; on ima stabilnu termodinamičku vezu sa membranom, koju napušta tokom procesa sklapanja. Ako se pretpostavi da je transmembranska zavojnica poravnata sa normalnim lipidnim dvoslojem, tada je konformacija proteina u obliku slova L verovatno najbolja topologija za membranski umetnuti protein zbog optimalnih hidrofobnih interakcija. Budući da je određivanje topologije proteina u dvoslojevima lipida posebno važno za samoasembliranje virusa i za sposobnost proteina da služi kao sredstvo za prikazivanje peptida, razvijeni su novi biofizički alati za proučavanje strukture proteina, topologije i dinamike dvosloja lipida [5–9].

POTENCIJALNA PRIMENA FAGA U NANOTEHNOLOGIJAMA

Fagi bi mogli da posluže kao sonde u novoj generaciji senzora za kontrolu bezbednosti hrane i nadzor nad životnom sredinom u realnom vremenu. Kao elementi detektora, oni su superiorniji od poliklonalnih i monoklonskih antitela, jer su jeftini, visoko specifični, selektivni i otporni na nepovoljne uslove okoline. Pored toga, jedinstvena struktura M13 bakteriofaga je iskorišćena kao biološki obrazac u nanotehnologiji, u usmerenoj sintezi poluprovodničkih / magnetnih nanožica i litijum-jonskih baterijskih elektroda. Filamentozni virusi omogućavaju organizaciju različitih nanomaterijala u periodično uređene hijerarhijske strukture, poput virusnih prstenova i žica, koje imaju elektronsku, optičku i biotehnološku primenu. U nauci o materijalima ovaj pristup se koristi za stvaranje novih peptida koji se mogu vezati za odabrane tehničke materijale. Bakteriofagi M13 sa proteinskim omotačem, budući da mogu da se vežu za poluprovodničke i magnetne materijale, korišćeni su kao predlošci za rast i organizovanje nanožica, služeći kao templejt za sintezu monokristalnih ZnS i CdS, i hemijski uređenih nanožica CoPt i FePt. Nuklearni peptidi ugrađeni u proteinski omotač M13 na taj način pružaju obrazac za usmerenu pripremu poluprovodničkih i magnetnih materijala [4–7].

Njihova primena mogla bi biti izuzetno značajna za tkivno inženjerstvo i regenerativnu medicinu, jer takve strukture imitiraju nativni ekstracelularni matriks, koji se sastoji od vlaknaste proteinske mreže i pruža ćelijama fizičku potporu [5, 6, 7].

Nedavna otkrića ukazuju da glavni proteini omotača, koji su glavni gradivni blokovi proteina, ne menjaju značajnije svoj oblik tokom membranske samoorganizacije, što ima za posledicu vrlo efikasan mehanizam za smanjenje potrošnje energije. Svaka nanotehnologija koja se bazira na bakteriofagu M13 treba da uzme u obzir njihova osnovna fizičko-hemijska pravila. I mada još uvek nisu rešeni mnogi aspekti biogeneracije nanostruktura, treba očekivati da će u narednim decenijama napredak u biofizičkim tehnikama povećati naša molekularna saznanja. Ako shvatimo da smo tek na početku nove ere bionanotehnologije, jasno je da je budućnost nanomašina na bazi virusa izuzetno svetla [7, 8].

Veliki, multijedinični, proteinski kompleksi se u početku spajaju slabim vezama, koje su vrlo pogodne za unutarćelijske komplekse, jer se privremeno sastavljaju i rastavljaju kako bi se aktivirale međusobno usklađene ćelijske funkcije. Čestice virusa, kao i drugi samoorganizujući sistemi, u početku se formiraju slabom vezom i, budući da je deo njihovog životnog ciklusa vanćelijski, oni moraju pronaći način organizacije koji im garantuje snažnu stabilnost. Zbog tako suprotstavljenih zahteva početna čestica koja se obično naziva Prokapsid ili Provirion vremenom sazreva, doživljavajući različite konformacione promene unutar kapsida. Neki virusi, kao nodavirusi, podvrgavaju se suptilnom autokatalitičkom cepanju kapsidnih podjedinica nakon izlaska iz ćelije, što dovodi do stabilizacije čestica virusa i povećanja njihove infektivnosti. Kod virusa bez ovojnice, poput tetravirusa, dramatično se reorganizuje čestica tokom sazrevanja, pri čemu dolazi do autokatalitičkog cepanja manjeg virusa [9, 10, 11].

SINTETIČKE MAŠINE ZASNOVANE NA DNK

DNK nanomašine se izrađuju samosastavljanjem, korišćenjem tehnika koje se oslanjaju na interakcije specifične za sekvence koje vezuju komplementarne oligonukleotide u dvostruku spiralu. Aktiviraju se interakcijom sa određenim signalnim molekulima ili promenama u njihovom okruženju, izazivajući odgovarajući odgovor na spoljni okidač, i služe za inteligentnu molekularno osetljivu isporuku leka ili kontrolisanu hemijsku sintezu. Biološki molekularni motori koji prenose teret unutar ćelija inspirisali su izgradnju rudimentarnih DNK šetača koji se protežu duž samoinstaliranih staza autonomno, dobijajući energiju katalizom reakcija DNK ili RNK [12, 13].

Izuzetna specifičnost interakcija između komplementarnih nukleotida čini DNK korisnim gradivnim materijalom, jer je interakcijom između njenih kratkih lanaca moguće pouzdano kontrolisati dizajniranje njihovih osnovnih sekvenci. Izgradnja razgranatih veza između dvostrukog heliksa omogućava stvaranje kompleksnih trodimenzionalnih objekata. Jedan od načina da se iskoristi ova izuzetno precizna arhitektonska kontrola je upotreba samoasembliranih DNK obrazaca za pozicioniranje funkcionalnih molekula, koji mogu da posluže kao molekularna elektronska kola, optički uređaji ili enzimske mreže. DNK nije prirodni izbor materijala za izgradnju aktivnih struktura, jer ne poseduje strukturnu i katalitičku svestranost proteina i RNK, koji su pogodniji za tu svrhu [13, 14].

MOLEKULARNI PREKIDAČI

Najjednostavnije aktivne DNK nanostrukture su prekidači ili aktuatori, koji deluju između dve konformacije, pri čemu je njihovo pomeranje podstaknuto promenama temperature, jonskih uslova ili vezivanjem signalnog molekula, koji najčešće predstavlja DNK zavojnica. Promene konformacije su izazvane promenama u okruženju, pri čemu je rotaciono kretanje uslovljeno promenom uvijanja DNK, pri kome se dvolančana DNK sa sekvencom (CG) n prebacuje iz uobičajene desnoruke zavojnice (B-DNK) u levoruku konformaciju (Z-DNK), u uslovima visoke koncentracije soli i niske temperature. Forsterov rezonantni prenos energije (FRET), koji predstavlja mogući mehanizam jedne takve biomašine, obezbeđuje prenos između odgovarajućih fluorofora i DNK zavojnica na nanometarskoj skali uz posredovanje dipol-dipol interakcije [13, 14, 15].

Jang i saradnici su pretvorili promene pri uvijanju DNK u linearno kretanje. Njihov uređaj se sastojao od zatvorene petlje dvolančane DNK pričvršćene na suprotne krakove četvorokrakog Holidejevog spoja, pri čemu Holidejev spoj može migrirati (izomerizirati) razbijanjem identičnih parova baza u jednom paru suprotnih krakova i njihovom transformacijom u drugi par. Promena konformacije DNK unutar petlje inicirana je dodavanjem etidijum-bromida, koji se veže između susednih baznih parova, produžavajući i delimično odmotavajući dvostruku spiralu. Rezultujući stres se ublažava migracijom čvora, odnosno skraćivanjem izbočenih krakova veza unutar petlje, što dopušta njeno izduženje bez promene ukupnog broja navoja u njoj. Ekološke promene u konformaciji jednolančane DNK mogu izazvati linearno kretanje, tako što se u blago kiselim uslovima jedan lanac citozina savija u i-motiv, kompaktnu trodimenzionalnu strukturu koju drže parovi baza protoniranih citozina. U prisustvu drugog komplementarnog lanca DNK javlja se konkurencija između i-motiva i produžene dvostruke zavojnice nastale hibridizacijom dve zavojnice [15, 16].

Prelazak i-motiva u dvostrani prelaz proizvodi mehanički rad. Ako se jedna površina silicijumske konzole presvuče vezanim zavojnicama koje sadrže citozin, tada dolazi do kompresivnog površinskog naprezanja koje savija konzolu. Takvo naprezanje uslovljeno hibridizacijom komplementarne zavojnice sa konzolom nije očekivano, iako je taj efekat prisutan pri visokim koncentracijama soli, gde su interakcije na daljini uporedive sa uočenim razdvajanjem zavojnica. Konformaciona promena izazvana pH zavisnim vezivanjem pojedinačnog lanca DNK u dupleks i formiranje trostruko-spiralne strukture je osnova dizajna nanomehaničkog aktuatora. Aktivne DNK nanostrukture se razvijaju kao senzori, koji omogućavaju da se elementarne logičke operacije izvode na njihovim izlazima, koji u kombinaciji sa računarima mogu biti primenjeni za dizajniranje pametnih sistema za isporuku lekova [15, 16, 17].

ZAKLJUČCI

Proteinski omotač bakteriofaga M13 opisan je kao izuzetno inspirativan u proizvodnji nanomašina, jer omogućava sintezu čestica željene funkcionalnosti, pod pretpostavkom da nije povređen nijedan osnovni biofizički princip, a posebno sposobnost proteina da formira stabilnu simetričnu oblogu i da se ubaci, usidri i sklopi u novu česticu virusa.

Ova tehnologija se razvija samostalno, pa se očekuje da ne samo fagi već i drugi slični virusi poput pametnih mašina mogu da proizvode nove proteine koji bi mogli poboljšati samu tehnologiju.

Opisane nanomašine zasnovane na DNK mogu poslužiti kao potencijalni pokretači ili prekidači za inteligentne molekularno osetljive sisteme za isporuku lekova ili sisteme za kontrolisanu hemijsku sintezu.

CASE REPORT
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Multidisciplinary approach in treatment of spacing: orthodontic treatment and partial ve-neers using the injectable composite resin technique

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SUMMARY

Introduction Patients with orthodontic diagnosis of spacing most often require a multidisciplinary approach, which includes orthodontic and following restorative treatment to enhance the esthetic outcome. The aim of this case report is to present management of spacing in the anterior region by orthodontic treatment followed by partial veneers using the injectable composite resin technique.

Case report In this case, leveling and alignment of dental arches and stable occlusion was achieved during orthodontic treatment, with the correction of upper and lower incisor inclination and closing the diastemas. Intraoral and extraoral esthetic parameters were evaluated on the photographs, and partial veneers on lateral incisors and canines were designed for the wax-up model. In the injectable composite resin technique, the silicone index was used to transfer the wax-up to composite restorations. The highly filled injectable composite resin was injected through the silicone index and light-cured. The restoration required only gentle polishing due to the great precision of the index.

Discussion Post-orthodontic recontouring in the anterior region is mostly done by direct composite restorations because they are cost-effective, minimally invasive, and the procedure is relatively simple. The injectable resin composite technique can be a solution for the same indication since it is less technique sensitive and gives predictable and great esthetic results, mostly without tooth preparation.

Conclusion Management of anterior spacing in adult patients requires a comprehensive approach for optimal esthetic and functional results. In this case, orthodontic treatment was followed by partial veneers on lateral incisors and canines, using the injectable composite resin technique as a simple and predictable solution for minor restorative interventions to solve morphological tooth abnormalities in the esthetic smile zone.

Keywords: spacing; orthodontic treatment; composite; partial veneers; injectable technique

INTRODUCTION

Anterior spacing and tooth size discrepancy is one of the most common features in adult dentition. Meeting patient's demands and expectations is the first step in deciding which treatment option is the best for optimal esthetic results. Orthodontics alone can give a great esthetic improvement, but multidisciplinary treatment is often needed for excellent treatment outcomes. Most often, orthodontic treatment is followed by restorative treatment [1, 2]. Direct restorations are done in one session, applying layers of composite directly to the tooth surface. Indirect restorations are preferred in complex cases, and they require collaboration with dental technicians [3]. Direct restorations are practical and have several advantages, such as saving the tooth structure; reversibility of procedure, lower cost to the patient, and the material can be added or removed easily, if necessary [1, 4, 5].

The injectable resin composite technique is a predictable dental procedure where a diagnostic wax-up is translated into composite restorations. It is an indirect/ direct technique that can be used to repair fractured teeth and restorations, provisional restorations, veneers, resurfacing occlusal wear on posterior composite restorations, and also in primary dentition for teeth with multiple caries or fractures [6, 7]. This technique is minimally invasive and relatively inexpensive compared to conventional ceramic veneers procedure, and tooth preparation is mostly not required [8].

The aim of this case report is to present management of spacing in the anterior region by orthodontic treatment followed by partial veneers using the injectable composite resin technique.

CASE REPORT

A 32 years old male patient presented to the dental office complaining about his smile's esthetic appearance. He did not like the misalignment of the anterior teeth as well as the spaces between them.

The main cause of spacing in his case was a discrepancy of tooth size and arch length. In his case, the labial frenulum was not prominent. In occlusion, molar and canine relation

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Figure 1. Initial clinical situation – class I canine and molar relationship and spacing in anterior region of dental arch
Slika 1. Početna situacija – odnos I klase po Englu na očnjacima i molarima i rastresitost u prednjoj regiji zubnog niza

Figure 2. Esthetic fixed appliances in upper and lower dental arch
Slika 2. Estetski fiksni aparati u gornjem i donjem zubnom nizu

was class I on both sides. Cephalometric analysis did not show skeletal discrepancies, and both upper and lower incisors were proclined. Orthodontic treatment aimed to level and align dental arches, correct the upper and lower incisors' position, close the diastemas in the upper and lower dental arch, and achieve good occlusion. Since esthetics was very important for the patient even during the orthodontic treatment, ceramic braces (Radiance, American orthodontics, Roth prescription, slot 22) were chosen. Standard arch wire protocols were followed and power chains were used for closing the spaces between the teeth. (Figures 1, 2).

After 13 months of orthodontic treatment, all diastemas were closed and a stable static and functional occlusion achieved. Although all the spaces were closed in gingival areas of teeth, black spaces between upper lateral incisors and canines were visible in incisal parts. Recontouring of lateral incisors and canines was necessary to enhance the esthetic outcome (Figure 3). Direct composite buildups could be a solution in minimally invasive and non invasive cases, since they are esthetic, functional, and biologically sound treatment options for closing diastemas with clinically promising survival rates [1]. After explaining possible treatment options, the patient decided to take a restorative treatment based on the injectable resin composite technique because it offers esthetic and predictable results and no dental tissues preparation. For this technique, an adequate design of the restoration is needed. Intraoral and extraoral photos of the patient were taken with a digital camera (D3400, Nikon corporation) and esthetic parameters evaluated. The future shape of lateral incisors

Figure 3. Clinical situation after orthodontic treatment- stable occlusion, diastemas closed; spaces present between upper lateral incisors and canines (only incisal parts) on both sides due to irregularities of the tooth shape
Slika 3. Klinička situacija posle ortodontske terapije – stabilna okluzija I klase po Englu, zatvorene dijasteme; prisutni su prostori između gornjih lateralnih sekutića sa obe strane (samo u incizalnom delu) zbog nepravilnog oblika ovih zuba.

Figure 4. Future shape of lateral incisors and canines designed; future partial veneers wax-up
Slika 4. Dizajn budućeg oblika gornjih lateralnih sekutića i očnjaka; izgled budućih delimičnih faseta u vosku

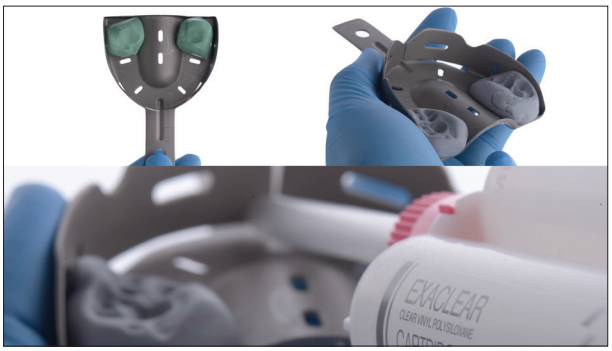

Figure 5. Preparation of impression tray using silicone stops to save the same thickness of the material
Slika 5. Priprema kašike za otisnu masu uz pomoć silikonskih stopera koji obezbeđuju istu debljinu materijala

and canines were designed in Keynote software (Apple Corporation). According to this design, partial veneers wax-up was made (Figure 4) Two veneers were planned on distal incisal surfaces of upper lateral incisors and two on mesial surfaces of canines to enhance the smile esthetics.

Based on the wax-up, a transparent silicone index was made using a clear polyvinyl siloxane (Exaclear,GC Corp.,Tokyo, Japan). Before making the silicone index, in order to hydrate, the plaster model was soaked in the cold water and left in for 5 minutes. Impression tray was

Figure 6. Silicone index with incisal perforations for composite injection
Slika 6. Silikonski ključ sa perforacijama u incizalnom delu koje će služiti za injektiranje kompozita

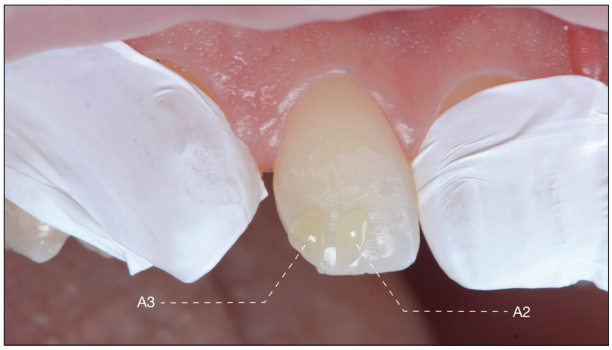

Figure 7. Determining the colour for the restoration using small amounts of composite; A2 chosen for the restoration
Slika 7. Određivanje boje budućih nadoknada korišćenjem male količine kompozita; A2 izabrana za buduće nadoknade

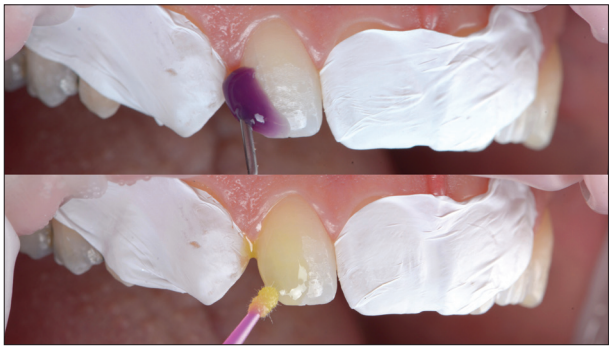

Figure 8. Phosphoric acid etching and bonding procedure
Slika 8. Priprema gleđi ortofosfornom kiselinom i nanošenje bonda

Figure 9. Injection of highly filled flowable composite through perforations on silicone index
Slika 9. Injektiranje tečnog kompozita kroz perforacije na silikonskom ključu

Figure 10. Final clinical situation
Slika 10. Klinička situacija na kraju procedure

Figure 11. Patient's smile after orthodontic and restorative procedure
Slika 11. Osmeh pacijenta posle završene ortodontske i restaurativne procedure

prepared using stoppers made of C - silicone (Zeta plus putty, Zhermack) to save the same silicone thickness in every part of the silicone key (Figure 5). Small perforations through the silicone index were made using the syringe of flowable resin composite (Figure 6). The perforations were made to the distal parts on the incisal edges of lateral incisors and canines' mesial parts. The material chosen for this intervention was a highly filled flowable resin composite (G-ænial Universal Injectable, GC corporation). The teeth were cleaned using fluoride-free polishing paste. Choosing the right color was done at the very beginning of the procedure, using a small amount of the material (composite buds) on the lateral incisors and canines, which were then light-cured. Shade A2 was selected for the procedure (Figure 7). The adjacent teeth were isolated using Teflon tape. One lateral incisor's polished enamel surface was etched with 37% phosphoric acid (37.5% Phosphoric Acid Gel, Kerr) for 40s, rinsed with water and air-dried. The universal adhesive (GC G-Premio Bond, GC Corporation) was applied to pre-etched surfaces with a micro brush for 10 sec, then air blown for 5 sec, and polymerized using a LED light-curing unit (3M Elipar™ DeepCure-S LED Curing Light) for 10 sec, according to manufacturers' instructions (Figure 8). The silicone index was positioned carefully and flowable resin composite injected through the perforation made on the incisal part of the silicone index for right lateral incisor (Figure 9). The restoration was light-cured for 40 sec from labial, occlusal and palatal direction. The

silicone index was removed, and the rest of the material was cut off with a scalpel. The procedure was repeated for right canine, and left lateral incisor and canine, one tooth at a time, protecting the finished restorations with the Teflon tape. Due to incredible precision of silicone index, the restorations required only simple and gentle polishing and finishing with finishing discs and silicone points. Proximal surfaces were smoothened with polishing strips. After polishing the restorations, new thermoplastic retainers were made to prevent tooth alignment changes after orthodontic treatment (Figures 10, 11).

DISCUSSION

Management of spacing in adults often requires a multidisciplinary approach for optimal results. Tooth alignment and stable static and functional occlusion can be achieved with orthodontic treatment, but the tooth shape abnormalities can be solved with indirect restorations (ceramic or composite) or by direct composite restorations.

Ceramics has always been a material of choice for anterior restorations because it is biocompatible, chemically stable, and effective in reproducing the tooth's natural translucency and structure. On the other hand, ceramic veneers require precise preparation since the preparation, among other causes, can be the reason for veneer fracture [3].

The injectable technique is relatively simple and gives a predictable outcome. Compared to direct composite restorations, this technique's main advantage is that it is less challenging and time-consuming. The injectable technique doesn't require preparation, which is very important in preserving sound dental tissues, especially in young patients [6–10]. In our case, the material used was G-aenial universal Injectable (GC Corporation), a highly filled injectable composite resin with improved mechanical properties and esthetics. The same material was used in the report of Hosaka et al. [10], whereas a group of authors in another study [9] used different, but also a highly filled flowable resin composite, G-aenial Universal Flo (GC Corporation).

The clinical effectiveness of these highly filled flowable materials was similar to paste-type composite in 36 months follow-up, in posterior restoration [11]. The study of Lai et al., who evaluated the surface gloss, roughness, and colour change of six different flowable composites, found that G-aenial universal Flo, termed as universal injectable composite by manufacturer, showed better surface properties after tooth abrasion than other composites tested [12].

CONCLUSION

Management of anterior spacing, as one of the most common dental features in adult patients, requires a comprehensive treatment for optimal esthetic and functional results. In the presented case, orthodontic treatment was followed by partial veneers on lateral incisors and canines, using the injectable composite resin technique as a simple and predictable solution for minor restorative interventions in the esthetic smile zone.

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Multidisciplinarni pristup u terapiji rastresitosti: ortodontska terapija i delimične fasete tehnikom injektiranja kompozita

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KRATAK SADRŽAJ

Uvod Kod pacijenata sa ortodontskom dijagnozom rastresitosti u zubnom nizu najčešće je potreban multidisciplinarni pristup, koji podrazumeva ortodontsku terapiju i posle nje restaurativnu proceduru kako bi se poboljšali estetski rezultati.

Cilj ovog rada bio je da se predstavi slučaj uspešnog rešavanja rastresitosti u prednjoj regiji zubnog niza uz pomoć ortodontske terapije i delimičnih faseta izrađenih tehnikom injektiranja kompozita.

Prikaz slučaja U ovom slučaju, ortodontskom terapijom postignuta je nivelacija zubnih nizova i stabilna okluzija, kao i korekcija inklinacije gornjih i donjih sekutića i zatvaranje dijastema. Nakon toga, analizirani su intraoralni i ekstraoralni parametri na fotografijama pacijenta, i dizajniran izgled delimičnih faseta u vosku na lateralnim sekutićima i očnjacima. U tehnici injektiranja kompozita koja je korišćena za izradu delimičnih faseta silikonski ključ poslužio je za prenošenje izgleda faseta u vosku u kompozitne ispune. Zatim je kroz perforacije na silikonskom ključu ubrizgan tečni kompozit poboljšanih estetskih i mehaničkih karakteristika i svetlosno polimerizovan. Zahvaljujući velikoj preciznosti silikonskog ključa, bilo je potrebno samo blago ispolirati ispune.

Diskusija Preoblikovanje zuba posle ortodontske terapije najčešće se radi metodom direktnog slojevanja kompozita jer procedura nije skupa, minimalno je invazivna i relativno je jednostavna. Tehnika injektiranja kompozita može biti rešenje za istu indikaciju jer tehnika manje zavisi od angažovanja terapeuta, i daje predvidive i dobre estetske rezultate, najčešće bez preparacije zubnih tkiva.

Zaključak Rešavanje rastresitosti u prednjoj regiji zubnih nizova kod odraslih pacijenata zahteva složen terapijski pristup za optimalne estetske i funkcionalne rezultate. U ovom slučaju ortodontska terapija bila je praćena izradom delimičnih faseta tehnikom injektiranja kompozita, kao jednostavnom procedurom za minimalne restaurativne intervencije kako bi se poboljšale nepravilnosti morfologije zuba u estetskoj zoni osmeha.

Ključne reči: rastresitost; kompozit; delimične fasete; tehnika injektiranja kompozita

UVOD

Rastresitost u prednjoj regiji zubnog niza i neusklađenost u veličini zuba i alveolarnog grebena predstavljaju veoma čestu pojavu kod odraslih pacijenata. Procena pacijentovih zahteva i očekivanja je prvi korak u odlučivanju koji je metod lečenja najbolji za postizanje optimalnih estetskih rezultata. Sama ortodontska terapija može dati značajna estetska poboljšanja, ali često je neophodan multidisciplinarni pristup za bolji konačan ishod terapije. Nakon ortodontske terapije najčešće je indikovana neka od restaurativnih procedura [1, 2]. Direktne restauracije se izvode u jednoj poseti, nanošenjem slojeva kompozita direktno na površinu zuba. Indirektne restauracije su poželjne kod složenijih slučajeva i zahtevaju saradnju sa zubnom laboratorijom [3]. Direktne restauracije su praktične i imaju nekoliko prednosti, kao što su minimalno uklanjanje zubnog tkiva, reverzibilnost postupka, niže cene intervencije, kao i to da se, ukoliko je potrebno, materijal može jednostavno dodati ili ukloniti [1, 4, 5].

Tehnika injektiranja kompozita je predvidiva stomatološka procedura gde se dijagnostički voštani model prevodi u kompozitne nadoknade uz pomoć silikonskog ključa. Predstavlja indirektno/direktnu tehniku koja se može koristiti za restauraciju karijesom destruisanih ili frakturiranih zuba, za izradu privremenih nadoknada i faseta, obnavljanje okluzalnih površina na bočnim zubima, kao i za mlečne zube sa višestrukim karijesima ili prelomima [6, 7]. Predstavlja minimalno invazivnu tehniku, relativno jeftinu u poređenju sa postupkom izrade keramičkih faseta, u kojoj preparacija zuba najčešće nije potrebna [8].

Cilj ovog prikaza slučaja je da predstavi rešavanje rastresitosti u prednjoj regiji zubnog niza ortodontskom terapijom, praćenom izradom delimičnih faseta, tehnikom injektiranja kompozita.

PRIKAZ SLUČAJA

Pacijent starosti 32 godine, muškog pola, dolazi u stomatološku ordinaciju nezadovoljan estetikom svog osmeha, odnosno izgledom i rasporedom zuba, kao i razmacima između njih.

Glavni razlog postojanja dijastema u ovom slučaju bila je neusklađenost veličine zuba i dužine alveolarnog grebena. U njegovom slučaju, labijalni frenulum nije bio izražen. U okluziji, odnos očnjaka i molara bio je klasa I po Englu, i na levoj i desnoj strani (Slika 1). Kefalometrijska analiza nije pokazala veće skeletne diskrepance, a gornji i donji sekutići bili su proklinirani. Cilj ortodontske terapije bio je iznivelisati zube u gornjem i donjem zubnom nizu, ispraviti položaj gornjih i donjih sekutića, zatvoriti dijasteme u gornjem i donjem zubnom luku i postići dobru okluziju. Budući da je estetika bila vrlo važna za pacijenta čak i tokom ortodontske terapije, odabrane su keramičke bravice (Radiance, American Orthodontics, preskripcija Roth, slot 22). Praćeni su standardni protokoli u nivelaciji uz pomoć ortodontskih lukova, a za zatvaranje dijastema korišćeni su gumeni lanci (Slika 2).

Nakon 13 meseci ortodontske terapije, sve dijasteme su bile zatvorene i postignuta je stabilna statička i funkcionalna okluzija. Iako su svi prostori bili zatvoreni u gingivalnim delovima krunica zuba, u incizalnim delovima su bili vidljivi crni prostori između gornjih lateralnih sekutića i očnjaka. Bilo je indikovano preoblikovanje lateralnih sekutića i očnjaka kako bi se poboljšao estetski ishod (Slika 3).

Direktno slojevanje kompozita moglo bi biti rešenje kod minimalno invazivnih i neinvazivnih procedura, budući da se njima dobijaju dobri estetski i funkcionalni rezultati pri zatvaranju dijastema, sa klinički dobrim stopama trajanja ispuna [1].

Za injekcionu tehniku potreban je odgovarajući dizajn restauracije. Intraoralne i ekstraoralne fotografije pacijenta napravljene su digitalnim fotoaparatom (D3400, Nikon corporation) i na njima su analizirani estetski parametri. Budući oblik lateralnih sekutića i očnjaka dizajniran je u softveru Keynote (Apple Corporation) (Slika 4). Prema ovom dizajnu napravljen je voštani model u zubnoj laboratoriji. Planirane su dve delimične fasete na distalnim incizalnim površinama gornjih lateralnih sekutića i dve na mezijalnim površinama očnjaka.

Prema dizajnu voštanog modela napravljen je silikonski ključ od prozirnog polivinil-siloksana (Exaclear, GC Corporation, Tokio, Japan). Pre izrade silikonskog ključa, u cilju hidratacije, gipsani model je potopljen u hladnu vodu i ostavljen pet minuta. Kašika za otisak je pripremljena pomoću stopera izrađenih od c-silikona (Zetaplus putty, Zhermack) kako bi se ostvarila jednaka debljina silikona u svakom delu silikonskog ključa (Slika 5). Uz pomoć kanile kojom će biti ubrizgan kompozit napravljene su male perforacije kroz silikonski ključ (Slika 6). Perforacije su napravljene na distalnim delovima incizalnih rubova lateralnih sekutića i mezijalnih delova očnjaka.

Materijal izbora za ovu intervenciju bio je tečni kompozit (G-ænial Universal Injectable, GC corporation). Zubi su očišćeni pastom za poliranje bez fluorida. Odabir boje kompozita obavljen je na samom početku intervencije, korišćenjem male količine materijala polimerizovane lampom na lateralnim sekutićima i očnjacima. Odabrana je boja A2 (Slika 7). Susedni zubi izolovani su pomoću teflon trake. Gleđ zuba predviđenog za restauraciju tretirana je 37,5% ortofosfornom kiselinom (37,5% gel ortofosforne kiseline, Kerr) u trajanju od 40 s, isprana vodom i posušena vazduhom iz pustera. Univerzalni adheziv (GC G-Premio Bond, GC Corporation) nanet je na prethodno tretiranu površinu gleđi aplikatorom za bond u trajanju od 10 s, zatim je vazduhom iz pustera posušen maksimalnom snagom u trajanju od 5 s, i polimerizovan pomoću LED lampe za polimerizaciju (3M Elipar™ DeepCure-S LED Curing Light) u trajanju od 10 s, prema uputstvu proizvođača (Slika 8). Zatim je pažljivo postavljen silikonski ključ i kroz perforaciju na incizalnom delu silikonskog ključa ubrizgan tečni kompozit G-ænial Universal Injectable nijanse A2 (Slika 9). Restauracija je polimerizovana u trajanju od 40 s sa labijalne i okluzalne strane. Nakon uklanjanja silikonskog ključa ostatak materijala uklonjen je skalpelom. Postupak je ponovljen za desni očnjak i levi lateralni sekutić i očnjak, jedan po jedan zub, štiteći gotove ispune i susedne zube teflon trakom. Zahvaljujući velikoj preciznosti silikonskog ključa, restauracije je bilo dovoljno jednostavno i nežno ispolirati diskovima i silikonskim polirerima. Aproksimalne površine ispolirane su trakama za poliranje. Posle poliranja restauracije napravljene su nove retencionalne folije za pacijenta, da bi se sprečila pomeranja zuba posle ortodontske terapije (slike 10 i 11).

DISKUSIJA

Rešavanje rastresitosti zubnih nizova kod odraslih pacijenata često zahteva multidisciplinarni pristup za postizanje optimalnih rezultata. Nivelacija zuba i stabilna statička i funkcionalna okluzija postignute su ortodontskom terapijom, dok se anomalije oblika zuba mogu rešiti indirektnim restauracijama (keramičkim ili kompozitnim) ili direktnim kompozitnim restauracijama.

Keramika je uvek bila materijal izbora za nadoknade u prednjoj regiji zubnog niza jer je biokompatibilna, hemijski stabilna i efikasna u reprodukciji prirodne translucentnosti strukture zuba. S druge strane, keramičke fasete zahtevaju preciznu preparaciju, jer preparacija, između ostalih uzroka, može biti razlog loma nadoknada [3].

Tehnika injektiranja kompozita je relativno jednostavna i daje predvidiv ishod. U poređenju sa direktnim kompozitnim restauracijama, glavna prednost ove tehnike je to što je manje izazovna u smislu angažovanja terapeuta i kraće traje. Tehnika injektiranja kompozita najčešće ne zahteva preparaciju zubnih tkiva, što je vrlo važno u kontekstu očuvanja zdravih zubnih tkiva, posebno kod mlađih pacijenata [6–10]. U ovom slučaju je korišćen G-aenial Universal Injectable, univerzalni restaurativni kompozit visoke čvrstoće sa poboljšanim mehaničkim svojstvima i estetikom. Isti materijal korišćen je u studiji Hosaka et al. [10] , dok je grupa autora u drugoj studiji [9] koristila drugi tečni kompozit G-aenial Universal Flo (GC Corporation). Klinička efikasnost ovih tečnih kompozita pokazala se sličnom efikasnosti pastastih kompozita u studiji koja je pratila efikasnost kod ispuna u bočnoj regiji, u trajanju od 36 meseci [11]. U studiji Lai i saradnika, koji su procenjivali sjaj površine, hrapavost i promenu boje šest različitih tečnih kompozita, G-aenial universal Flo pokazao je bolja površinska svojstva posle abrazije zuba od ostalih testiranih kompozita [12].

ZAKLJUČAK

Rešavanje rastresitosti u zubnom nizu kod odraslih pacijenata zahteva složen tretman za postizanje optimalnih estetskih i funkcionalnih rezultata. U ovom slučaju ortodontska terapija je bila praćena izradom delimičnih faseta na lateralnim sekutićima i očnjacima, korišćenjem tehnike injektiranja kompozita kao jednostavnog i predvidivog rešenja za manje restaurativne procedure u estetskoj zoni osmeha.

Da li ste pažljivo čitali radove?

1. O statusu oralnog zdravlja mladih od 19 do 26 god. u Srbiji
   a) ima malo informacija
   b) nema uopšte informacija
   c) ima dosta informacija

2. Struktura virusa je slična arhitekturi nanomašina?
   a) Da
   b) Ne
   c) Nema nikakve sličnosti

3. Rastresitost u fontalnj regiji zubnog niza je:
   a) retko moguća kod odraslih
   b) česta pojava kod odraslih
   c) česta samo kod dece

4. Prisustvo debrisa i metalnih opiljaka je uočeno na svim instrumentima?
   a) Da
   b) Ne
   c) Samo kod dva seta

5. Studija koja se bavila oralnim zdravljem studenta realizovana je:
   a) 2010.
   b) 2012.
   c) 2020.

6. Bakteriofagi su jedno od najvažnijih dostignuća u molekularnoj biologiji, biofizici i bionanotehnologiji?
   a) Da
   b) Ne
   c) Postoje druga mnogo značajnija dostignuća

7. Kompletna terapija rastresitosti u frontalnoj regiji najčešće uključuje:
   a) ortodonsku i restaurativnu terapiju
   b) ortodonsku i implantološku terapiju
   c) ortodonsku i protetsku terapiju

8. Najmanje defekata NITI instrumenata je uočeno kod:
   a) Bio RaCe
   b) MTWO
   c) K3

9. Oralno zdravlje je ispitivano kod:
   a) studenata osnovnih studija
   b) kod svih mladih u Beogradu
   c) kod mladih u Srbiji

10. Terapija rastresitosti u frontalnoj regiji pored ortodonske najčešće uključuje:
    a) direktne restauracije
    b) indirektne restauracije
    c) izbeljivanje zuba

11. Na svim NITI instrumentima su uočeni proizvodni defekti?
    a) Da
    b) Ne
    c) Samo na dva seta

12. Studija o oralnom zdravlju je obuhvatila:
    a) 499 studenata
    b) 599 studenata
    c) 699 studenata

13. Tehnika injektiranja kompozita se realizuje tehnikom:
    a) silikonskog ključa
    b) pomoću prsta
    c) pomoću matrice

14. Korozija radnog dela NITI je uočena kod:
    a) MTWO
    b) HY FLEX
    c) Bio RaCe

15. Anketu o oralnom zdravlju je popunilo:
    a) 230 studenata
    b) 530 studenata
    c) 730 studenata

16. Bakteriofagi M13 imaju:
    a) bakterijsku arhitekturu
    b) virionsku arhitekturu
    c) kombinaciju bakterijske i virionske

17. Kod studenta iz Istočne i Južne Srbije vrednost KIP-a je iznosila:
    a) 7,43
    b) 8,69
    c) 9,84

18. Najučestaliji defekti na NiTi instrumentima su uočeni kod:
    a) PRO TAPER UNIVERZAL
    b) Bio RaCe
    c) K3

19. Parodontalni status kod studenta je evaluiran:
    a) kod 275 studenata
    b) kod 375 studenata
    c) kod 475 studenata

20. Klinička procena statusa oralnog zdravlja je realizovana primenom:
    a) KIP indeksa
    b) indeksa lo zuba
    c) indeksa ekstri zuba

21. Tehnika injektiranja kompozita predstavlja:
    a) klasičnu restaurativnu tehniku
    b) minimalno invazivnu tehniku
    c) vrlo agresivnu tehniku

22. Najzastupljeniji defekt na radnom delu NiTi je bio:
    a) prisustvo žlebova
    b) prisustvo debrisa
    c) prisustvo korozije

23. Anketni upitnik u studiji se sasoji iz:
    a) 4 odeljka
    b) 5 odeljaka
    c) 6 odeljaka

24. Samo procena 02:
    a) razlikuje se od vrednosti K s 02
    b) korelira sa vrednostima K s 02
    c) korelira samo kod studenta stomatologije K s 02

25. Struktura bakteriofaga M13 omogućava preciznu izgradnju uređenih:
    a) samo dvodimenzionalnih nanostruktura
    b) samo trodimenzionalnih nanostruktura
    c) i dvo i trodimenzionalnih nanostruktura

26. Pacijent gde je rešavana rasprostranjenost u frontalnoj regiji je:
    a) uzrasta 20 godina
    b) uzrasta 32 godine
    c) uzrasta 40 godina

27. Zatečene nečistoće na radnom delu NiTi su utvrđene:
    a) SEM-EDS analizom
    b) SEM analizom
    c) EDS analizom

28. Anketni upitnik je uključivao i odeljak o samoproceni 02?
    a) Da
    b) Ne
    c) Samo kod pojedinih studenata

29. Vrednost KIP-a kod pregledanih studenta je iznosila:
    a) 8,42
    b) 9,32
    c) 10,24

30. Samoprocena 02:
    a) značajno korelira sa vrednostima KiP-a
    b) ne korelira sa vrednostima KIP-a
    c) korelira samo kod studenta ženskog pola

31. M13 bakteriofag je uzročnik:
    a) akutnih infekcija
    b) hroničnih infekcija
    c) subakutnih infekcija

32. Ortodonska terapija rastresitosti je trajala:
    a) 12 meseci
    b) 13 meseci
    c) 18 meseci

33. SEM analiza NiTi instrumenta je realizovana:
    a) na uvođenju 100-1000
    b) na uvođenju 150-1500
    c) na uvođenju 150-2000

34. Vrednost KIP-a kod studenata tehničkih nauka je iznosila:
    a) 6,94
    b) 8,84
    c) 10,11

35. Bakteriofagi imaju ključnu ulogu u kontroli bioloških sistema?
    a) Da
    b) Ne
    c) Samo ponekad

36. Materijal za restauraciju zuba frontalne regije je uključivao:
    a) G-AENIAL UNIVERSAL INJECTABLE
    b) G-AENIAL UNIVERSAL FLO
    c) TETRIC EVO FLOW

37. Radni deo NiTi instrumenata je podvrgnut:
    a) samo SEM analizi
    b) samo EDS analizi
    c) SEM-EDS analizi

38. Vrednost KIP-a kod studenta čija majka poseduje fakultetsku diplomu je iznosila:
    a) 8,35
    b) 9,13
    c) 9,33

39. Studija o oralnom zdravlju studenta je obuhvatila:
    a) 173 studenta medicinskih nauka
    b) 173 studenta društvenih nauka
    c) 173 studenta tehničkih nauka

40. Protein se efikasno ugrađuje u membranu bakteriofaga:
    a) zahvaljujući C terminalnom interfacijalnom sidru
    b) zahvaljujući D terminalnom interfacijalnom sidru
    c) zahvaljujući M terminalnom interfacijalnom sidru

41. U istraživanju NiTi instrumenta je korišćeno:
    a) pet novih setova
    b) šest novih setova
    c) deset novih setova

42. Kod studenta koji svoje zdravlje smatraju zadovoljavajućim vrednost KIP-a je iznosila:
    a) 7,42
    b) 8,94
    c) 10,11

43. U studiji o oralnom zdravlju učestvovali su i studenti stomatologije?
    a) Da
    b) Ne
    c) Samo studenti završnih godina

44. Primena bakteriofaga je najznačajnija kod:
    a) inženjerskih materijala
    b) kod svih biomaterijala
    c) kod svih cementa

45. Delimične fasete su jednostavno i predvidljivo rešenje:
    a) za velike restaurativne procedure
    b) za manje restaurativne procedure
    c) za procedure u bočnoj regiji

46. Procena zdravlja potpornog aparata zuba kod studenta:
    a) nije pokazala značajne razlike u vrednostima analiziranih indeksa
    b) pokazala je značajne rezultate u vrednostima analiziranih indeksa
    c) značajne razlike su uočene samo kod studenata muškog pola

47. Enzimi bakteriofaga obezbeđuju alate za proučavanje puteva:
    a) replikacije, transkripcije i transporta
    b) isključivo translacije
    c) isključivo transkripcije

48. Vrednosti KIP-a kod studenta 2012. godine su bile:
    a) niže nego 1987.
    b) više nego 1987.
    c) identične

49. Tehniha prikazivanja faga je moćna metodologija za identifikaciju i optimizaciju liganda antitela i drugih biomolekula?
    a) Da
    b) Ne
    c) Samo u određenim situacijama

50. Materijal G-AENIAL UNIVERSAL INJECTABLE obezbeđuje:
    a) izuzetne estetske osobine
    b) dobre estetske osobine
    c) neadekvatne estetske osobine

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