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Contents / Sadržaj

Glupost je čudnija od svih bolesti.
Oboleli nikad ne pate zbog nje, ali pate drugi.
Pol Anri Spak

U svojim uredničkim komentarima obično apostrofiram značaj ličnog obrazovanja i učenja, jer je to jedini put ka najvažnijem životnom zakonu, ka istini. A koliko je taj put dalek i „neravan" svedoči svaki segment sadašnjeg trenutka. Iako je rad preduslov svakog uspeha, naš život neretko oblikuju ljudi sa sumnjivim diplomama, ljudi sa još sumnjivijim naučnim zvanjima, ljudi bez ikakvog iskustva u poslovima koje obavljaju, odnosno osobe čiji su ljudski i moralni kvaliteti na najnižoj lestvici. Da li je budućnost kojoj težimo moguća tamo gde neuki i nekompetentni ljudi odlučuju o svemu, pa čak i o nekim važnim naučnim i stručnim pitanjima?

Da li na univerzitetu postoji „kritička masa" i energija koja može da „resetuje" sistem koji je podlegao neprincipijelnosti i nepoštovanju osnovnih etičkih normi? Mora se ozbiljno poraditi na sprečavanju lažnih doktorata, plagijata, hiperprodukcije nastavnog kadra, ali i na valorizovanju elementarnih moralnih načela. Mora se u naše moralne kodekse utemeljiti da je cilj znanje a ne ocena. Mora se stalno naglašavati da su znanje, posvećenost, stručnost i odgovornost jedini preduslovi za bilo koju funkciju.

Jedan od ozbiljnih problema na našim univerzitetima je i činjenica što se nastavnička karijera obezbeđuje onog momenta kada neko postane asistent na nekom fakultetu. Do zvanja redovnog profesora se obično „lako" stiže bez obzira na to da li „kandidat" ima talenta, bez obzira na to što nije najbolji, odnosno što nema nikakve ni naučne ni stručne rezultate. Jedini kriterijum je da je kanditat „podoban", odnosno da je potomak neke poznate familije, a sve ostalo je splet okolnosti.

Zato su u nauci ali i u društvu neophodne prave i dobro osmišljene reforme, kako bi se brojne devijantne pojave mogle iskoreniti. Ali ne reforme koje će ostati samo kao zahtevi na papiru, već kao ozbiljni i suštinski sveobuhvatni kriterijumi bez ikakvog političkog uticaja. Ovi kriterijumi bi trebalo da u centar postave čoveka sa visokomoralnim ljudskim kvalitetima i visokim naučnim kompetencijama, jer će samo takve osobe moći da „maglovitu" budućnost učine dostojnijom i izvesnijom.

Iskreno se nadam da osim želje na univerzitetu postoji i kvalitet da se nešto promeni. Oni koji mogu, znaju i imaju energije da iznedre promene moći će to da učine jedino ako ne budu „sputani" od onih koji su za ovakvo stanje i najzaslužniji.

Ovaj urednički komentar ću završiti citatom Ive Andrića koji najbolje oslikava naš sadašnji trenutak u skoro svim segmentima života: „U vremenu kada su svi ljudi uplašeni i zbunjeni, ili uvređeni i oštećeni, zlo se lako prima i širi dalje".

Prof. dr Slavoljub Živković

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The use of essential oils based antiseptic solution in the treatment of denture stomatitis

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SUMMARY

Introduction Local therapy of denture stomatitis (DS) associated with *Candida* species fungi infection usually involves the application of nystatin and miconazole. Due to the fact that these drugs may be less efficient against biofilm and possible resistance development, a new approach in the treatment includes the use of antiseptic agents.
The aim of the study was to compare clinical and microbiological therapeutic outcomes of antiseptic solution *Listerine*® and *Daktanol*® antifungal oral gel in the treatment of DS associated with *Candida* species fungi.
Material and Methods The study included 30 patients affected by DS, divided into the two treatment groups, control group (n=15) treated by *Daktanol*® gel and experimental group (n=15) treated by the antiseptic solution *Listerine*®. Successful treatment was evaluated based on palatal mucosa inflammation reduction classified according to the Newton classification and the difference in the number of fungal colony- forming units (CFU) isolated by smears before and after the treatment that lasted 14 days.
Results Reduction in inflammation intensity and fungal CFU number on palatal mucosa ($p<0,01$) as well as on denture base ($p<0,01$) were observed in both groups of subjects after the treatment.
Conclusion Antiseptic solution *Listerine*® and *Daktanol*® antifungal gel both reduced palatal mucosal inflammation and CFU number of fungi in mouth without significant differences among them. CFU number of fungi isolated from denture base was significantly lower after the treatment with *Listerine*® ($p<0.05$).
Keywords: *Candida albicans*; denture stomatitis; Listerine, Daktanol gel

INTRODUCTION

Denture stomatitis (DS) is an inflammation of oral mucosa covered by the denture base with reported incidence of 15-70% [1]. It is linked to a number of non-infectious aetiologies such as poor quality of dentures, poor hygiene and nocturnal denture wearing as well as infectious etiological factor, *Candida* species fungi [2]. Number of fungal cells in the saliva and on dentures is leading etiological factor for DS development [3]. The degree of *Candida albicans (C.albicans)* denture base contamination is directly correlated to inflammation intensity [4].

This disease is characterized by erythema of oral mucosa covered by the denture base; it is usually asymptomatic and often associated with angular cheilitis [5]. Its intensity is clinically evaluated according to the Newton classification criteria [6,7]. DS treatment most frequently includes local and systemic administration of antifungals, reduction and eradication of biofilm, change of bad habits related to nocturnal denture wearing and poor denture hygiene as well as replacement of inadequate dentures. The most frequently used local antifungal drugs in dental practice are nystatin and miconazole [8,9]. Although effectively act against fungi, these drugs have no effect on biofilm matrix, therefore well-protected pathogens can survive in extracellular matrix. Also, their use comes with the risk of resistance development [10, 11]. Commercially available oral antiseptic *Listerine*® acts directly against fungal cell, chemically, causing damage to the cell wall structure and membrane permeability. Also, it disrupts metabolic processes dependant on microorganism membrane enzymes, and, as other phenolic products, exerts anti-inflammatory effect [12]. *Listerine*® acts against free unbound *Candida* cells as well as against formed biofilm and it is shown to be more efficient than the azole antifungal drugs [13]. This *Listerine*® solution property has not been studied in a clinical trial yet.

The aim of this study was to compare clinical and microbiological outcomes of antiseptic agent *Listerine*® and antifungal *Daktanol*® gel in the treatment of DS associated with *Candida* species fungi.

MATERIAL AND METHODS

This prospective clinical study was conducted at the Department of Dental Medicine in Foca and the Microbiological laboratory of the University Hospital Foca. The

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research was conducted in accordance with the principles of the Helsinki Declaration of 2008. Prior to inclusion in the study subjects were informed about the aim and the protocol of research and gave their consent to participation. The study included 30 patients, maxillary acrylic dentures wearers, affected by DS. Criteria for inclusion of patients in the study were good general health, or chronic well-controlled disease and dental acrylic dentures wearing for at least one year. Also, swabs of palatal mucosa and denture basal surface had to be positive on *Candida* species fungi. Exclusion criteria were as follows: the use of corticosteroids and immunosuppressive therapy, tumours of the maxillofacial region and radiotherapy in the head and neck area, chemotherapy in the last year, blood disorders, local surgical therapy in the last 3 months, local or systemic use of antibiotics and antifungal drugs in the last 3 months, the use of hormones for therapeutic purposes, the existence of other diseases in the oral cavity and also functionally, prophylactically and cosmetically inadequate dentures that are indicated for replacement with new dentures.

By the means of random numbers table, participants who joined the study under even-number were classified into the control group and received standard treatment with *Daktanol*® oral gel (2% miconazole, Galenika a.d. Belgrade, Serbia). The participants included in the study under odd-number were classified into the experimental group and were treated with antiseptic agent *Listerine*® *cool mint* ™ (Johnson&Johnson, S.p.A. Rome, Italy). Each group consisted of 15 patients, and therapy was administered according to the following protocol:

1. Control group of patients (n = 15) received standard local antifungal therapy in the form of *Daktanol*® gel containing 2% miconazole, used at the dose of 1-2 teaspoons, 4 times per day for 14 days. Basal surface of dentures were coated with gel overnight and rinsed with water in the morning prior to use.

2. Experimental group of patients (n = 15) used an antiseptic agent *Listerine*® also for 14 days as follows: 4 times a day 1 minute long mouth swish with antiseptic solution that was spitted afterward. Dentures were immersed in this solution overnight with base facing upward in a glass container, and completely overflowed by the *Listerine*® solution. In the morning dentures were rinsed with water.

Parameters used for the evaluation of inflammation and denture contamination, as well as the presence of expected improvement were clinical and microbiological. Clinical improvement implied palatal mucosa inflammation intensity reduction after the treatment compared to the inflammation intensity prior to the therapy.

1. Clinical parameters

The intensity of palatal mucosa inflammation was assessed by Newton classification which distinguishes three clinical types of denture stomatitis [6,7]: type *Newton I*: dotted hyperaemic shape and size of a pinhead lesion (pin-point) that present localized areas of poorly expressed inflammation; type *Newton II*: diffuse erythema of palatal mucosa covered by the denture base – generalized inflammation; type *Newton III*: granular surface of the mucosa- inflammatory papillary hyperplasia.

2. Microbiological parameters

Swabs of palatal mucosa surfaces and denture bases were taken without prior mouth and dentures rinsing, in the morning, before any food intake. Sabouraud dextrose agar was used to grow fungi and it was incubated at 37 ° C, for 48 hours. The number of fungal colonies (CFU-colony forming units) was counted after 48 hours. The following criteria were used for CFU quantification: <10 colonies present after incubation – smear is negative; 10-25 colonies present after incubation – fungi present in small numbers; >25 colonies present after incubation – fungi are present in large numbers [14]. Two days after the treatments, control examinations were conducted and swabs for samples cultivation were taken again.

The obtained data were statistically analyzed in the SPSS software (SPSS for Windows, version 11.5, Chicago, Ill.). Description of the sample was carried out by descriptive statistics methods. Therapeutic results within a group (paired samples) were evaluated by the Wilcoxon Signed Rank test. The effect of the therapy on the clinical improvement was assessed using the Fisher's Exact Test, and therapeutic results between observed groups were compared using χ^2 test. The relationship between certain characteristics and inflammation intensity as well as therapy outcomes were estimated using χ^2 test and Fisher's Exact Test. Results are presented in tables.

RESULTS

The study included 30 patients, 19 (63.3%) female and 11 (36.7%) male, with an average age of 56.1 years (SD = 7.126). The largest number of dentures, 17 (56.7%), was between 5 and 10 years old, 9 dentures (30%) were aged over 10 years, while 4 dentures (13.3%) were used less than 5 years. Most of patients, 27 (90%) had clinically present denture stomatitis classified as type II *Newton*, while 3 patients (10%) had denture stomatitis *Newton* type III.

In both groups, significant reduction in the CFU number at the palate ($p < 0.01$) as well as at the denture base was observed after the treatments ($p < 0.01$) (Table 1).

Inflammation intensity reduction was observed in most patients, but significant difference after treatments between the two applied therapeutic modalities was not observed (Table 2). There was no significant difference in palate smear CFU number reduction in relation to the applied therapy, but significant difference ($p < 0.05$) was observed in denture smear CFU number reduction in patients treated with antiseptic agent *Listerine*® compared to patients treated by *Daktanol*® oral gel (Table 3).

Gender, age of patients or age of dentures, had no statistically significant effect on clinical improvement after the treatment, as well as on the reduction in the CFU numbers isolated from palatal mucosa and denture basal surface after the treatment (Table 4)

Table 1. Reduction of isolated fungal colony forming unit (CFU) number after the treatment
Tabela 1. Smanjenje broja izolovanih gljivičnih kolonija nakon završene terapije

CFU number before and after the treatment Broj CFU pre i posle terapije	*Listerine®*		*Daktanol®*	
	Palate Nepce	Denture base Proteza	Palate Nepce	Denture base Proteza
Increased CFU number Povećan broj CFU	0	0	0	0
Decreased CFU number Smanjen broj CFU	15	15	14	15
Unchanged CFU Nepromenjen CFU	0	0	1	0
Z; p	-3.542; 0.000	-3.397; 0.001	-3.520; 0.000	-3.508; 0.000

Paired sample, Wilcoxon Signed Rank test
Vezani uzorak, Vilkoksonov test ekvivalentnih parova

Table 2. The influence of applied treatments on the clinical improvement
Tabela 2. Efikasnost primenjenog tretmana na kliničko poboljšanje

Clinical improvement Kliničko poboljšanje	Applied therapy		Total Ukupno (n; %)
	Daktanol®	*Listerine®*	
Yes Da	11; 60.0	13; 86.7	24; 80.0
No Ne	4; 40.0	2; 13.3	6; 20.0
Total Ukupno	15; 100.0	15; 100.0	30; 100.0

Fisher's Exact Test =0.241
Fišerov test tačne verovatnoće =0,241

Table 3. Reduction of fungal colonies number isolated from palate and denture base after the treatment
Tabela 3. Efikasnost primenjenih terapija na smanjenje broja gljivičnih kolonija izolovanih sa nepca i baze proteze nakon terapije

Reduction of the number of fungal colonies Smanjenje broja gljivičnih kolonija		Applied treatment Primenjena terapija		Total Ukupno (n; %)
		Daktanol®	*Listerine®*	
Palate* Nepce*	Yes Da	9; 60.0%	12; 80.0%	21; 70.0%
	Partially Delimično	5; 33.3%	3; 20.0%	8; 26.7%
	No Ne	1; 6.7%	0; 0.0%	1; 33%
Denture base** Baza proteze**	Yes Da	8; 53.3%	14; 93.3%	22; 73.3%
	Partially Delimično	7; 46.7%	1; 6.7%	8; 26.7%
	No Ne	0; 0.0%	0; 0.0%	0; 0.0%

*χ^2 = 1.929; df = 2; p = 0.381
** χ^2 = 6.136; df = 1; p = 0.013

DISCUSSION

The present study evaluated the effect of oral antiseptic *Listerine®* on palatal mucosa inflammation and *Candida* species CFU number isolated from palatal mucosa and denture base among denture wearers affected with DS.

Literature review couldn't identify other similar clinical studies. Yet, there are indirect evidences of *Listerine®* antifungal properties that justify its use in DS treatment. Meiller et al. conducted an *in vitro* study in which they observed the effect of Listerine® on clinically isolated *Candida* species, British and American strains of the same species. Fungal cells were incorporated into an experimental biofilm. Authors reported that after 60 seconds of experimental biofilm exposure to this antiseptic, no living fungal cells were observed in the sample [15]. In a study conducted with clinically isolated *Candida* species, *Listerine®* showed very good antimicrobial activity in laboratory testing. After 60 minutes, there was no living cell of *Candida* species in the sample [16]. *Listerine®* was also efficient against experimental biofilm composed of one laboratory and 34 clinically isolated *C. albicans* strains where it reduced metabolical activity of fungi for 75-80% [13]. The effect of *Listerine®* against *C.albicans* clinical isolates was confirmed in a recent study where *Listerine®* reduced *C.albicans* growth on Sabouraud dextrose agar [17]. The results presented in this study clinically confirmed findings of previous *in vitro* studies.

In the present study, *Listerine®* showed better efficacy in reducing the number of CFU isolated from denture basis compared to the *Daktanol®* gel. This finding could be the result of different viscosity of used agents. Miconazole was applied in a form of gel, what might hinder its denture base coating. However, due to its physical caracteristics, *Listerine®* solution, could reach rugged, porous acrylic surface more easily. Beside therapy, improved hygiene and avoiding nocturnal dentures wearing could have positive impact on palatal mucosa inflammation reduction.

CONCLUSION

Therapeutic outcomes after the use of antiseptic agent *Listerine®* in DS treatment are similar to the therapeutic outcomes obtained by standard *Daktanol®* oral gel therapy. Therefore, *Listerine®* can be used in the treatment of DS associated with *Candida* species.

Table 4. Influence of gender, age of respondents and age of dentures on the treatment outcome
Tabela 4. Uticaj pola, starosti ispitanika i starosti proteza na ishod terapije

	Clinical improvement Kliničko poboljšanje	Reduction of CFU in palate smear Smanjenje CFU izolovanih u brisu nepca	Reduction of CFU in denture base smear Smanjenje CFU izolovanih u brisu proteza
Gender Pol	Fisher's Exact Test =0.417	χ^2 = 0.600 p = 0.741	χ^2 = 0.695 p = 0.706
Age of respondents Starost ispitanika	χ^2 = 0.653 p = 0.884	χ^2 = 10.296 p = 0.113	χ^2 = 8.504 p = 0.203
Age of dentures Starost proteze	χ^2 = 2.995 p = 0.224	χ^2 = 6.622 p = 0.157	χ^2 = 4.451 p = 0.348

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Primena antiseptičnog sredstva na bazi esencijalnih ulja u terapiji proteznog stomatitisa

Ognjenka Janjić-Pavlović[1], Ivica Stančić[2], Smiljka Cicmil[1], Zorica Stojanović[1], Jelena Lečić[1], Sašo Elenčevski[3]

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KRATAK SADRŽAJ

Uvod Lokalna terapija proteznog stomatitisa (PS) udruženog sa infekcijom gljivicama iz roda *Candida* najčešće se sprovodi upotrebom nistatina i mikonazola. Zbog otežanog dejstva ovih lekova na biofilm, kao i zbog razvoja moguće rezistencije, pažnja se usmerava na terapijske efekte koji se mogu postići primenom antiseptičkih sredstava.

Cilj rada je da se uporede klinički i mikrobiološki ishodi primene antiseptičkog rastvora *Listerine*® i oralnog gela sa antimikotskim dejstvom *Daktanol*® u lečenju PS udruženog sa pojavom gljivica iz roda *Candida*.

Metode rada Studija je uključivala 30 ispitanika obolelih od PS, podeljenih u dve terapijske grupe: kontrolnu (n = 15) lečenu gelom *Daktanol*® i eksperimentalnu (n = 15) – antiseptikom *Listerine*®. Uspešnost terapije procenjivana je na osnovu smanjenja intenziteta zapaljenja palatinalne sluzokože procenjenog prema klasifikaciji *Newton* pre početka lečenja i razlike u broju brisom izolovanih gljivičnih kolonija (CFU) pre i posle terapije, koja je trajala 14 dana.

Rezultati Kod obe grupe ispitanika došlo je do smanjenja intenziteta zapaljenja i smanjenja CFU i na nepcu ($p < 0,01$) i na bazi proteze ($p < 0,01$) nakon terapije.

Zaključak Antiseptički rastvor *Listerine*® i antimikotik *Daktanol*® dovode do smanjenja intenziteta zapaljenja palatinalne sluzokože i do značajnog smanjenja CFU u terapiji PS, bez međusobno značajne razlike, osim u smanjenju broja CFU izolovanih sa baze proteza nakon terapije, gde je rastvor *Listerine*® pokazao veću efikasnost ($p < 0,05$).

Ključne reči: *Candida albicans*; protezni stomatitis; Listerine, Daktanol gel

UVOD

Protezni stomatitis (PS) jeste zapaljenje oralne sluzokože pokrivene bazom proteze sa prijavljenom učestalošću od 15 do 70% [1]. Povezan je sa brojnim neinfektivnim etiološkim faktorima kao što su loš kvalitet proteza, slaba higijena i noćno nošenje nadoknada, kao i sa infektivnim etiološkim faktorom, gljivicama iz roda *Candida* [2]. Broj gljivičnih ćelija u pljuvački i na samim protezama igra vodeću ulogu u razvoju PS [3]. Stepen kontaminacije baze proteze gljivicom *Candida albicans (C. albicans)* u direktnoj je korelaciji sa intenzitetom zapaljenja palatinalne sluzokože [4].

Oboljenje karakteriše eritem onih područja oralne sluzokože koja su pokrivena bazom proteze, često je udruženo sa angularnim heilitisom i najčešće je asimptomatsko [5]. Intenzitet oboljenja se klinički procenjuje klasifikacijom *Newton* [6, 7]. Terapija najčešće obuhvata lokalnu i sistemsku primenu antimikotika, redukciju i eradikaciju biofilma, promenu loših navika vezanih za noćno nošenje i slabu higijenu proteza, te zamenu postojećih proteza ukoliko su neadekvatne. Dva najčešće primenjivana lokalna antimikotika u stomatološkoj praksi su nistatin i mikonazol [8, 9]. Iako efikasno deluju na gljivice, ovi lekovi nemaju uticaj na sam matriks biofilma, te dobro zaštićeni patogeni u ekstracelularnom matriksu mogu preživeti, a moguć je i razvoj rezistencije [10, 11]. Komercijalno dostupan oralni antiseptik *Listerine*® na gljivičnu ćeliju deluje direktno, hemijski, dovodeći do oštećenja strukture ćelijskog zida i poremećaja kontrole permeabiliteta membrane. Takođe, prekida metaboličke procese koji zavise od enzima prisutnih u membrani mikroorganizma, te kao i ostali fenolni preparati ispoljava antiinflamatorno dejstvo [12]. *Listerine*® deluje i na slobodne nevezane ćelije *Candide*, kao i na formirani biofilm i to sa većom efikasnošću od azolnih antimikotika [13]. Ova dejstva rastvora *Listerine*® do sada nisu ispitivana u kliničkim uslovima.

Cilj rada bio je da se uporede klinički i mikrobiološki ishodi primene antiseptičkog sredstva *Listerine*® i antimikotika *Daktanol*® u lečenju PS udruženog sa pojavom gljivica iz roda *Candida*.

MATERIJAL I METODE RADA

Ova prospektivna klinička studija sprovedena je na Klinici za stomatologiju Medicinskog fakulteta u Foči i u Mikrobiološkoj laboratoriji Univerzitetske bolnice Foča. Istraživanje je sprovedeno u skladu sa principima Helsinške deklaracije iz 2008. godine, a pre uključivanja u studiju ispitanici su bili upoznati sa ciljem i protokolom istraživanja i dali svoju saglasnost za učestvovanje u istom. Studija je obuhvatala 30 ispitanika, nosilaca gornjih totalnih akrilatnih proteza, obolelih od proteznog stomatitisa. Kriterijumi za uključivanje ispitanika u studiju bili su dobro opšte zdravstveno stanje, odnosno dobro kontrolisano opšte oboljenje i nošenje totalne proteze bar jednu godinu. Takođe, brisevi palatinalne sluzokože i bazalne površine proteze morali su biti pozitivni na gljivice iz roda *Candida*. Kriterijumi za isključenje bili su sledeći: upotreba kortikosteroida i imunosupresivna terapija, tumori maksilofacijalne regije, zračna terapija u predelu glave i vrata, primenjena hemoterapija u poslednjoj godini dana, krvne diskrazije, lokalna hirurška terapija u protekla tri meseca, lokalna ili sistemska upotreba antibiotika i antimikotika u poslednja tri meseca, upotreba hormona u terapijske svrhe, postojanje drugog oboljenja u usnoj duplji, te neadekvatne proteze čija je funkcionalna, profilaktička i estetska vrednost ugrožena u tolikoj meri da je indikovana izrada novih nadoknada.

Pomoću tablice slučajnih brojeva određeno je da se ispitanici koji su se u studiju priključili pod parnim rednim brojem svrstaju u kontrolnu grupu i primaju standardnu terapiju oralnim gelom *Daktanol®* (2% mikonazol, Galenika a.d. Beograd, Srbija). Ispitanici uključeni u studiju pod neparnim rednim brojem svrstani su u eksperimentalnu grupu i primili su terapiju antiseptičnim sredstvom *Listerine® cool mint™* (Johnson & Johnson, S.p.A. Rim, Italija). Svaka grupa se sastojala od po 15 ispitanika, a terapija je primenjivana po sledećem protokolu:

1. kontrolna grupa ispitanika (n = 15) primala je standardnu lokalnu antifungalnu terapiju u vidu gela *Daktanol®*, koji sadrži 2% mikonazola, u dozi od 1-2 kafene kašičice, četiri puta dnevno, tokom 14 dana. Proteze su se preko noći sa bazalne strane premazivale gelom, a ujutro ispirale vodom pre upotrebe.

2. eksperimentalna grupa ispitanika (n = 15) koristila je antiseptično sredstvo *Listerine®* i to po sledećem režimu: četiri puta dnevno jednominutno mućkanje antiseptičnog rastvora, koji se nakon tog vremena ispljune, a proteze su preko noći potapali u ovaj rastvor tako što su ih postavljali u čašu, bazom okrenutom ka gore i prelivali rastvorom *Listerine®* do potpunog prekrivanja svih njenih površina. Ujutro su se proteze ispirale vodom, a terapija je takođe trajala 14 dana.

Parametri na osnovu kojih se procenjivao stepen inflamacije i kontaminacije nadoknada, kao i nastupanje očekivanog poboljšanja, bili su klinički i mikrobiološki. Kliničko poboljšanje podrazumevalo je smanjenje intenziteta zapaljenja sluzokože nakon terapije u odnosu na intenzitet utvrđen pre terapije.

1. Klinički parametri

Intenzitet zapaljenja palatinalne sluzokože procenjivao se klasifikacijom *Newton*, prema kojoj razlikujemo tri klinička tipa proteznog stomatitisa [6, 7]: tip *Newton I* – tačkaste hiperemične lezije oblika i veličine glave čiode koje predstavljaju lokalizovana područja slabo izraženog zapaljenja; tip *Newton II* – difuzni eritem cele nepčane sluznice koja je pokrivena bazom proteze – generalizovano zapaljenje; te tip *Newton III* – granularna površina mukoze – zapaljenska papilarna hiperplazija.

2. Mikrobiološki parametri

Uzimanje brisa inflamatorno promenjene nepčane sluzokože i bazne površine proteze vršeno je ujutro, natašte, bez prethodnog ispiranja usne šupljine i proteza, sterilnim štapićem vate. Materijal se zasijavao na saburo dekstrozni agar i inkubirao na 37°C, 48 časova, nakon čega su utvrđivani prisustvo i broj gljivičnih kolonija. Da bi se kvantifikovao broj gljivičnih kolonija (CFU – *colonie forming units*), korišćeni su sledeći kriterijumi: <10 kolonija nakon inkubacije – bris je negativan; 10–25 kolonija prisutnih nakon inkubacije – gljivice prisutne u manjem broju i >25 kolonija prisutnih nakon inkubacije – gljivice prisutne u većem broju [14]. Dva dana nakon isteka terapije vršeni su kontrolni pregledi i ponovo su uzimani brisevi za kultivaciju uzoraka.

Dobijeni podaci su statistički obrađeni u SPSS programu (*SPSS* za *Windows,* verzija *11.5, Chicago, Ill.*). Opis uzorka vršen je metodama deskriptivne statistike. Terapijski rezultati unutar jedne grupe (vezani uzorak) procenjeni su pomoću Vilkoksonovog testa ekvivalentnih parova. Uticaj terapije na kliničko poboljšanje procenjen je primenom Fišerovog testa tačne verovatnoće, a terapijski rezultati između ispitivanih grupa poređeni su pomoću χ^2 testa. Povezanost pojedinih obeležja sa intenzitetom zapaljenja i ishodom terapije procenjen je pomoću χ^2 testa i Fišerovog testa tačne verovatnoće. Rezultati su prikazani tabelarno.

REZULTATI

U studiju je bilo uključeno 30 ispitanika, 19 (63,3%) ženskog i 11 (36,7%) muškog pola, prosečne starosti 56,1 godina (SD = 7,126). Najveći broj proteza – 17 (56,7%) bio je star između pet i deset godina, devet proteza (30%) bile su stare više od deset godina, dok su četiri proteze (13,3%) korišćene kraće od pet godina. Najveći broj pacijenata – 27 (90%) imao je klinički izražen protezni stomatitis klasifikovan kao tip *Newton II*, dok su tri pacijenta (10%) imala protezni stomatitis tipa *Newton III*.

Kod obe grupe ispitanika došlo je do visoko značajnog smanjenja broja CFU i na nepcu ($p < 0,01$) i na bazi proteze nakon terapije ($p < 0,01$) (Tabela 1).

Smanjenje intenziteta zapaljenja nastupilo je kod većine ispitanika, bez uočenog postojanja značajne razlike nakon terapije između dva primenjena terapijska modaliteta (Tabela 2). Nije pronađena značajna razlika u smanjenju broja CFU dobijenih brisom nepca u odnosu na primenjenu terapiju, ali je nađena značajna razlika ($p < 0,05$) u smanjenju broja CFU izolovanih brisom baze proteze kod pacijenata lečenih antiseptičnim sredstvom *Listerine®* u odnosu na pacijente lečene oralnim gelom *Daktanol®* (Tabela 3).

Pol i starost pacijenata, kao ni starost proteza, nisu imali statistički značajnog uticaja na smanjenje intenziteta zapaljenja nakon terapije, kao ni na smanjenje CFU broja izolovanih sa palatinalne sluzokože i bazalnih površina proteza nakon terapije (Tabela 4).

DISKUSIJA

U našoj studiji procenjivan je uticaj oralnog antiseptika *Listerine®* na stepen inflamacije palatinalne sluzokože i broj kolonija gljivica iz roda *Candida* na nepcu i proteznoj ploči kod nosilaca totalnih proteza obolelih od proteznog stomatitisa.

Druge kliničke studije koje su koristile isti tretman, po istom ili sličnom protokolu kao u našem istraživanju, nisu nađene pregledom literature. Ipak, postoje posredni dokazi o antifungalnom dejstvu ovog sredstva koji opravdavaju njegovu primenu u terapiji proteznog stomatitisa. Meiller i saradnici su sproveli *in vitro* studiju u kojoj je proučavano dejstvo antiseptika *Listerine®* na kliničke izolate iz roda *Candida* i britanske i američke laboratorijske sojeve istog roda. Posmatrajući antimikrobno dejstvo na gljivične ćelije u sastavu eksperimentalnog biofilma, utvrđeno je da nakon 60 sekundi izlaganja ovom antiseptiku više nije bilo živih gljivičnih ćelija u uzorku [15]. U studiji u kojoj su uzorci mikroorganizama poticali od pacijenata sa *Candida*-pozitivnim brisom oralne sluzokože, *Listerine®* je pokazao veoma dobru antimikrobnu aktivnost u *in vitro* uslovima. Nakon 60-minutnog izlaganja, u uzorku nije bilo živih ćelija *C. albicans* [16]. U studiji u kojoj je ispitivano dejstvo antiseptika *Listerine®* na biofilm sastavljen od jednog laboratorijskog i 34 klinički izolovana soja vrste *C. albicans*, utvrđeno je da je *Listerine®* re-

dukovao postojeću metaboličku aktivnost gljivica za 75–80% [13]. Efekat sredstva *Listerine®* na kliničke izolate *C. albicans* u biofilmu laboratorijski je potvrđen i u skorijem istraživanju, gde je *Listerine®* inhibisao rast *C. albicans* na saburo agaru [17]. Rezultati prikazani u našem istraživanju klinički su potvrdili nalaze navedenih *in vitro* studija, što se ogleda u smanjenju broja brisom izolovanih gljivičnih kolonija sa nepca i baze proteza nakon terapije proteznog stomatitisa.

Listerine® je u našoj studiji pokazao veću efikasnost u smanjenju broja gljivičnih kolonija izolovanih brisom baze proteze u odnosu na rezultat dobijen u kontrolnoj grupi. Na ovakav nalaz je mogla uticati različita viskoznost primenjenih sredstava. Mikonazol je bio primenjen u formi gela, pa je samim tim teže oblagao površinu proteza. *Listerine®* je zbog fizičkih karakteristika rastvora lakše dopirao do neravnih, poroznih prostora i mikropukotina koje su prisutne na akrilatnoj bazalnoj površini zubnih nadoknada, što je mogući uzrok pronađene razlike. Pored korišćene terapije, smanjenju inflamacije palatinalne sluzokože mogli su doprineti poboljšani higijenski režim usne duplje i proteza, kao i noćno nenošenje proteza.

ZAKLJUČAK

Terapijski ishodi primene antiseptičkog sredstva *Listerine®* u lečenju proteznog stomatitisa slični su terapijskim ishodima dobijenim primenom standardne terapije oralnim gelom *Daktanol®*. Oralni antiseptik *Listerine®* može se koristiti u terapiji proteznog stomatitisa udruženog sa pojavom gljivica iz roda *Candida* kod nosilaca totalnih akrilatnih proteza.

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The cost of individual health care in Serbia according to the international classification of diseases in the period 2010–2015

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SUMMARY

Introduction There is growing interest in the world for estimating the cost for the treatment of a disease. This value can be used to determine to which extent a particular disease or group of diseases burden society in terms of the global crisis (Segel 2006). In 2000, Organization for Economic Countries Development (OECD) established a System of Health Accounts (SHA), and provided methodological guide for calculating the cost of treating the disease.
The aim of this study was to determine the cost of individual health care in the Republic of Serbia according to the major International Classification of Diseases (ICD) for the period 2010-2015.

Material and Methods A retrospective and comparative analysis of health statistics from the database of the Institute of Public Health of Serbia (IPHS) and financial information provided by the National Health Insurance Fund (NHIF) in the period 2010–2015 was performed. Financial information and data on hospital services, outpatient, home health care, auxiliary health care services, drug consumption and consumer goods in healthcare were analyzed using SHA methodology.

Results showed that during observation period the maximum cost of individual health care in Serbia by main classification ICD was achieved in 2015 and it was 194,128,864,011 RSD (€1,580,853,941; $1,764,807,854) and the minimal cost was achieved in 2010, 151,333,139,835 RSD (€1,434,464,541; $1,908,843,843).

Conclusion The cost of individual health care in the Republic of Serbia in the period 2010-2015 increased by thirty percent. The highest amount was allocated to treat people with diseases of the circulatory system.

Keywords: cost of the disease; health spending; health accounts; cardiovascular diseases

INTRODUCTION

Health care is one of the most important human activities and one of the most dynamic in terms of growth in expenses for its delivery. Numerous achievements in medicine, pharmaceuticals and medical technologies improve health status of people in most countries, but they also increase the cost. Therefore, the focus of health policy is to establish efficacy but also the effectiveness of health care. Health care costs are a burden that particular disease or group of diseases put on society [1].

Realistic presentation of financial data in health care, in particular those relating to the monitoring costs for the treatment of patients with certain diseases are absolutely necessary. In the recent years, there was growing interest for calculating the costs of treating patients [2, 3, 4], that have certain diseases [5-16], injuries [17] disorders [18] and conditions [19-24].

In 2000, Organization for Economic Development of countries (OECD) established the System of Health Accounts (SHA) that formulated indicative methodological guidance for calculating the cost of treating patients through the Table 6. Until the initiation of new Methodological guidelines SHA 2011, the Table 6 in SHA was used [25], as part of the National Health Accounts (NHA). That Table provides the current cost of health care according to the main groups of disorders by the International Classification of Diseases (ICD).

In the system of health accounts an individual health care costs are defined as those costs in the health system that do not involve shared health care like public health services, collective prevention, health insurance, health administration and costs for performing functions related to the health care.

The aim of this study was to perform a comparative analysis of the costs of individual health care in the Republic of Serbia by main groups of diseases (ICD) for the period 2010-2015.

MATERIALS AND METHODS

A comparative retrospective analysis of statistical data from the database of the Institute of Public Health of Serbia (IPHS) and financial data of the National Health Insurance Fund (NHIF) for the period 2010-2015 was performed.

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Table 1. Groups of diseases according to the International Classification of Diseases (ICD), and methodology for the preparation of the table 6 NHA based on the SHA (version 1)
Tabela 1. Grupa oboljenja prema Međunarodnoj klasifikaciji bolesti (MKB) i metodologija za izradu tabele 6 NZR na osnovu SZR (verzija 1)

Code Šifra	Groups of diseases Grupe oboljenja
A A00-B99	Infectious and parasitic diseases Infektivne i parazitske bolesti
C C00-D48	Neoplasms Tumori
D D50-D89	Diseases of the blood and blood-forming organs Bolesti krvi i krvotvornih organa
E E00-E90	Endocrine and metabolic diseases Endokrine bolesti i bolesti metabolizma
F F00-F99	Mental disorders Mentalni poremećaji
G G00-G99	Diseases of the nervous system Bolesti nervnog sistema
I I00-I99	Diseases of the circulatory system Bolesti cirkulatornog sistema
J J00-J99	Diseases of the respiratory system Bolesti disajnog sistema
K K00-K93	Diseases of the digestive system Bolesti digestivnog sistema
L L00-L99	Skin and subcutaneous tissue disorders Bolesti kože i potkožnog tkiva
M M00-M99	Diseases of the musculoskeletal system Bolesti mišićno-koštanog sistema
N N00-N99	Diseases of the genitourinary system Bolesti urogenitalnog sistema
O O00-O99	Complications of pregnancy and childbirth Komplikacije trudnoće i porođaja
P P00-P96	Perinatal conditions Perinatalna stanja
Q Q00-Q99	Congenital malformations Urođene anomalije
R R00-R99	Symptoms and pathologies Simptomi i patološka stanja
S S00-T98	Injuries, poisoning and consequences Povrede, trovanja i posledice
U U01-Z99	All other categories Sve ostale kategorije

Information about provided services for hospital, outpatient and home treatment, as well as day care, extra services, health care, drug consumption and consumer goods to health care, were analyzed and cross- compared with financial data from the Republic Health Insurance Fund after examination procedure of SHA (version 11).

The cost of individual health care in the Republic of Serbia, according to the main categories of the ICD was grouped into the following scheme and methodology of SHA 11:

- H.C.1. – Hospitalization services are financially expressed by the number of hospital days by groups of diseases (data source: NHIF [26]) multiplied by the cost of hospital day from the current price list of health services NHIF.
- H.C.1.2. – Day care services are registered by groups of diseases and multiplied by the price of the day from the current price list of health services NHIF.
- H.C.1.3. – Funds spent for the treatment of ambulatory patients are determined by the number of outpatient treatment services (data source: NHIF) multiplied by the current price list of health services NHIF.
- H.C.1.4. – Home health services cost were expressed by the services provided at home treatment (data source: planning table for health centers that handles NHIF) multiplied by the price of the current price list of health services NHIF.
- H.C.4. – Auxiliary health services (laboratory analysis, diagnostics and medical transportation) are financially expressed by the empirically estimated total number of these service multiplied by the corresponding prices of the current price list of health services NHIF.
- H.C.5.1. – Financial view of drugs and other consumer goods was obtained from the Agency for Medicines and Medical Devices Agency of Serbia.

The sum of items H.C.1, H.C.1.2, H.C.1.3, H.C.1.4, H.C.4. and H.C.5.1. by groups of diseases gave the estimated financial value for the total cost of health care of population in Serbia by groups of diseases ICD.

Retrospective and comparative research methods were applied in the analysis. Data obtained from the National Fund of Statistics (NFS) and the National Bank of Serbia (NBS) were used as well.

The following Tables present groups of diseases codes according to the International Classification of Diseases (Table 1), as well as Table 6 that was used to calculate the cost of individual health care (Table 2).

RESULTS

The results determined the cost of individual health care by main categories of the ICD The total sum (in RSD, Euros and US Dollars) that was spent for health care of population in Serbia during observed years, according to the ICD is shown in the Table 3.

Total cost for individual health care by major ICD classification in 2010 was 151,331,867,999 RSD. In regards to the groups of diseases the highest individual cost was allocated for the circulatory system diseases (19.58%), neoplasms (10.0%), diseases of the digestive system (9.75%), nervous system diseases (8.33%), infectious and parasitic diseases (8.99%), while the least was for congenital anomalies (0.27%) (Figure 1). In 2010, the total individual cost for health care by main categories ICD per capita was € 195.00, and in relation to the gross domestic product (GDP) in 2010 it was 5.20%.

151,614,999,373 RSD in 2011 was the total individual cost for health care by major ICD categories. In relation to the groups of diseases the highest individual cost was allocated for the circulatory system diseases (19.14%), diseases of the digestive system (9.71%), infectious and parasitic diseases (8.88%) and neoplasms (8.90%), while the least was for congenital anomalies (0.30%) (Figure 2). The total cost for individual health care by main categories of ICD in 2011 per capita was € 205.00, and it was 4.77% of the GDP for that year.

Total cost for individual health care by major ICD categories in 2012 was 166,575,285,407 RSD. The highest indi-

Table 2. Table 6 used for cost calculation
Tabela 2. Tabela 6 korišćena za izračunavanje

Individual health care **Individuala zdravstvena zaštita**	**HC.1-HC.3**				**HC.4**	**HC.5**	**HC.5.1**	**HC.5.2**	**HC.1-HC.5**
Table 6 Tabela 6	*Hospital treatment* *Bolničko lečenje*	*Day services* *Usluge dnevne nege*	*Ambulant treatment* *Ambulantno lečenje*	*Home treatment* *Kućno lečenje*	*Additional services* *Pomoćne usluge*	*Medical goods for non hospital treatment* *Medicinska roba za vanbolničko lečenje*	*Medications and additional medical goods* *Lekovi i druga potrošna medicinska dobra*	*Therapeuticals* *Terapeutska pomagala*	*Totoal cost* *Ukupni troškovi*
Infectious and parasitic diseases Infektivne i parazitske bolesti									
Neoplasms Tumori									
Diseases of the blood and blood-forming organs Bolesti krvi i krvotvornih organa									
Endocrine and metabolic diseases Endokrine bolesti i bolesti metaboliza									
Mental disorders Mentalni poremećaji									
Diseases of the nervous system Bolesti nervnog sistema									
Diseases of the circulatory system Bolesti cirkulatorog sistema									
Diseases of the respiratory system Bolesti disajnog sistema									
Diseases of the digestive system Bolesti digestivnog sistema									
Skin and subcutaneous tissue disorders Bolesti kože i potkožnog tkiva									
Diseases of the musculoskeletal system Bolesti mišićno-koštanog sistema									
Diseases of the genitourinary system Bolesti urogenitalnog sistema									
Complications of pregnancy and childbirth Komplikacije trudnoće i porođaja									
Perinatal conditions Perinatalna stanja									
Congenital malformations Urođene anomalije									
Symptoms and pathologies Simptomi i patološka stanja									
Injuries, poisoning and consequences Povrede, trovanja i posledice									
All other categories Sve ostale kategorije									

Table 3. Total cost of health care by groups of ICD in Serbia in the period 2010–2015
Tabela 3. Ukupni troškovi za zdravstvenu zaštitu prema grupama MKB u Srbiji u periodu 2010–2015. godine

Year **Godina**	**Totoal cost (dinars, RSD)** **Ukupni troškovi (dinari, RSD)**	**Totoal cost (euros, €)** **Ukupni troškovi (evri, €)**	**Totoal cost (dollars, $)** **Ukupni troškovi (dolari, $)**
2010	151,333,139,835	1,434,464,541	1,908,843,843
2011	151,614,999,374	1,488,057,711	2,076,022,877
2012	166,972,893,694	1,477,505,475	1,898,282,102
2013	176,734,078,012	1,563,050,128	2,081,183,208
2014	183,189,009,509	1,547,596,600	1,875,591,374
2015	194,128,864,011	1,580,853,941	1,764,807,854

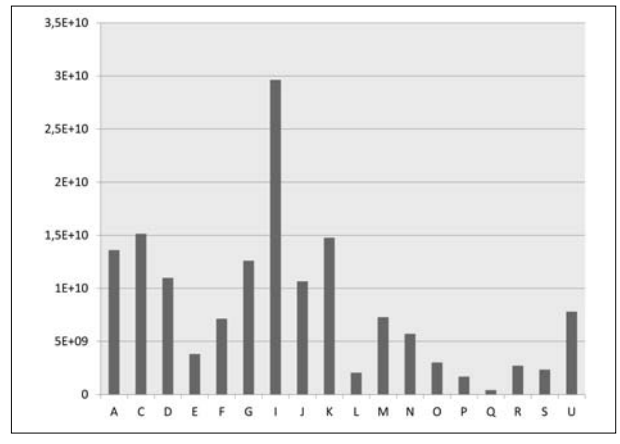

Figure 1. Individual costs for health care, according to the ICD classification in 2010.
Grafikon 1. Individualni troškovi za zdravstvenu zaštitu po MKB klasifikaciji u 2010. godini

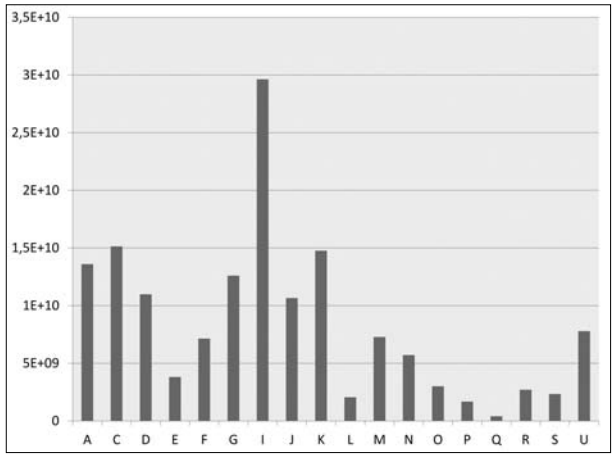

Figure 2. Individual costs for health care, according to the ICD classification in 2011.
Grafikon 2. Individualni troškovi za zdravstvenu zaštitu po MKB klasifikaciji u 2011. godini

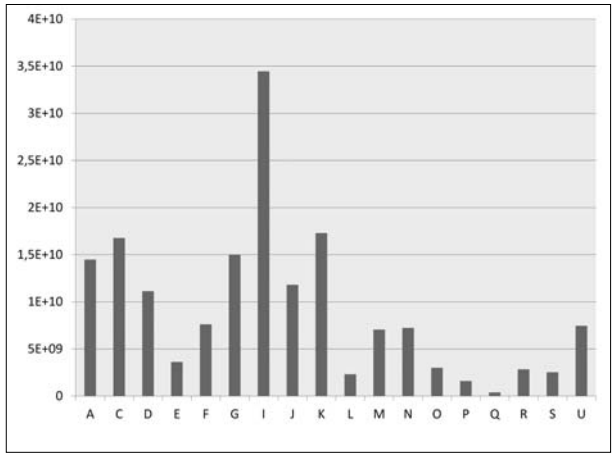

Figure 3. Individual costs for health care, according to the ICD classification in 2012.
Grafikon 3. Individualni troškovi za zdravstvenu zaštitu po MKB klasifikaciji u 2012. godini

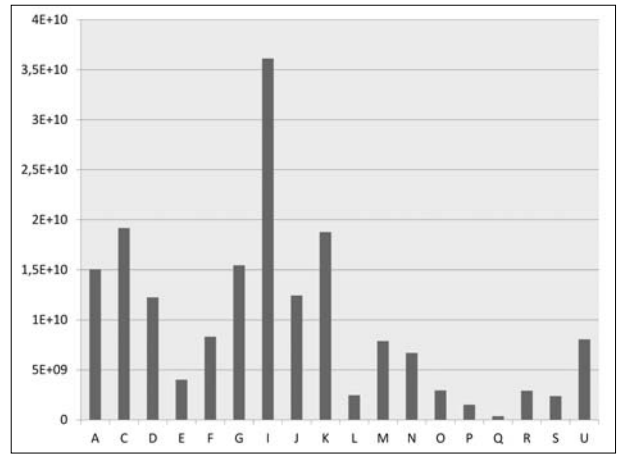

Figure 4. Individual costs for health care, according to the ICD classification in 2013.
Grafikon 4. Individualni troškovi za zdravstvenu zaštitu po MKB klasifikaciji u 2013. godini

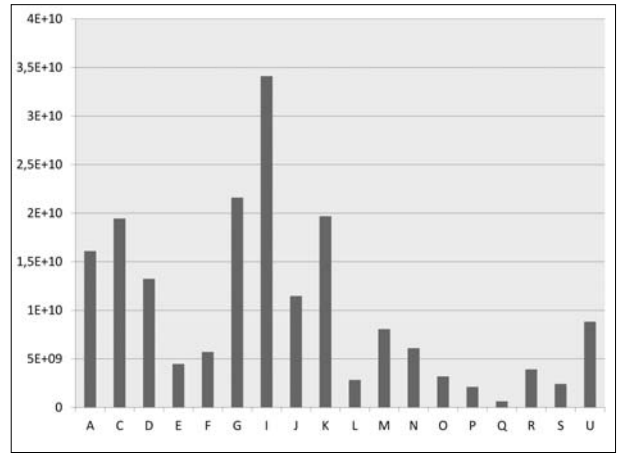

Figure 5. Individual costs for health care, according to the ICD classification in 2014.
Grafikon 5. Individualni troškovi za zdravstvenu zaštitu po MKB klasifikaciji u 2014. godini

vidual cost was observed by the following disease groups: circulatory system diseases (20.69%), diseases of the digestive system (10.38%), neoplasms (10.07%), nervous system (9.00%), and infectious parasitic diseases (8.69%), while the least was for congenital anomalies (0.23%) (Figure 3). In 2012, the total individual cost for health care by major ICD categories per capita was € 205.00. That represented 4.97% of the GDP for the year 2012.

In 2013, the total individual cost for health care by major ICD categories was 176,734,078,012 RSD. By the ICD classification, the greatest individual cost was allocated for the circulatory system diseases (20.44%), neoplasms (10.84%), diseases of the digestive system (10.62%), nervous system (8.74%), infectious parasitic diseases (8.51%), and the least for congenital anomalies (0.21%) (Figure 4). Individual cost for health care by main categories of ICD in 2013 per capita was € 219.00, being 4.88% of the GDP for 2013.

The total cost for individual health care by major ICD categories in 2014 was 183,189,009,508 RSD. The individual cost was allocated mostly for the circulatory system diseases (18.62%), nervous system (11,79), diseases of the digestive system (10.74%), neoplasms (10.62%), infectious and parasitic diseases (8.79%), and the least for congenital anomalies (0.34%) and perinatal conditions (1.15%) (Figure 5). In 2014, the total individual cost for health care by main categories ICD per capita was € 217.00. That was 4.72% of the GDP for 2014.

The highest cost of individual health care according to the ICD was recorded in 2015 and amounted to 194,128,864,011 RSD (€ 1.580.853.941; $ 1,764,807,854). In 2015, most money was spent on circulatory diseases (19.8%), diseases of digestive system (10.67%), neoplasms (10.13%), and the least for congenital anomalies (0.32%). Results are expressed in RSD and shown on the Figure 6. The total cost for individual health care by major ICD category in 2015 per capita was € 223.00. That was about 4.89% of the GDP for 2015.

Observed by years (2010-2015) the total cost of individual health care by major ICD categories shows increasing tendency (Figure 7). The highest cost was allocated for cardiovascular diseases. The percentage share of the total individual cost for health care in the period 2010-2015, per capita, expressed in RSD, euros and dollars is shown in the Table 4.

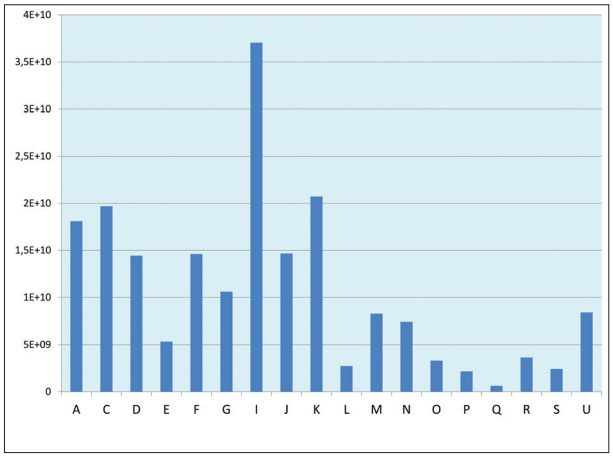

Figure 6. Individual costs for health care, according to the ICD classification in 2015.
Grafikon 6. Individualni troškovi za zdravstvenu zaštitu po MKB klasifikaciji u 2015. godini

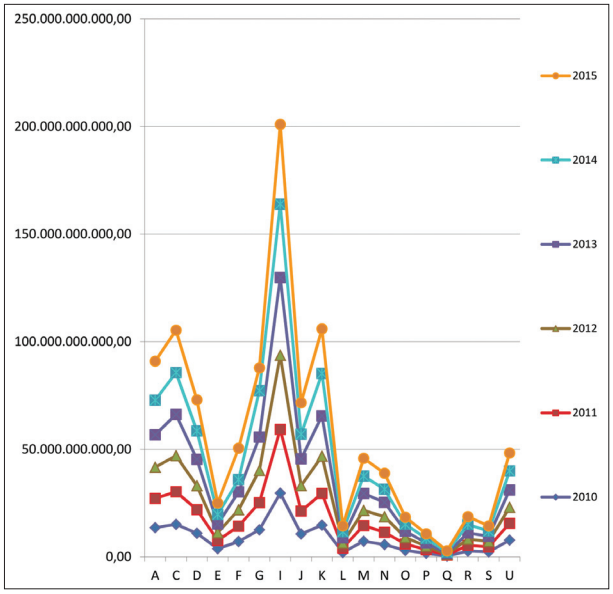

Figure 7. Growth trend of individual costs for health care, according to the ICD categories
Grafikon 7. Trend rasta individualnih troškova za zdravstvenu zaštitu po MKB kategorijama

Individual costs for health care by main categories ICD per capita are also trending upwards from € 195 in 2010 to 223 € in 2015. The percentage share of the total health care costs by major ICD categories in relation to the GDP in the period 2010-2015 had a decline from 5.20% in 2011 to 4.89% in 2015. In 2011 sharp decline was recorded with constant fluctuations in the period 2012-2015 (Table 5).

DISCUSSION

It has been shown that studies about the cost of health care of patients [2-4] usually focus on the particular disease [5-16], or individual injuries [17], disorders [18] or conditions only [19-24]. Current costs of health care for major groups of diseases ICD are presented in Table 6 of SHA, as part of the National Health account (NHA) [25]. Many users believe that the analysis of table 6 SHA,

Table 4. Individual cost for health care by main categories ICD per capita in the period 2010–2015.
Tabela 4. Individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama po stanovniku u periodu 2010–2015. godine

	2010	2011	2012	2013	2014	2015
Cost per capita (RSD) Troškovi po stanovniku u dinarima	20,178	20,887	23,251	24,669	25,686	27,360
Cost per capita (€) Troškovi po stanovniku u evrima	195	205	205	219	217	223
Cost per capita ($) Troškovi po stanovniku u dolarima	259	286	264	291	263	249

Table 5. Percentage of cost for health care by the GDP from 2010 to 2015
Tabela 5. Procenat troškova za zdravstvenu zaštitu u BDP-u od 2010. do 2015. godine

Year Godina	GDP (in millions RSD) BDP (u milionima RSD)	Percentage of cost from GDP Udeo troškova u BDP
2010	2,999,632.9	5.20
2011	3,268,315.6	4.72
2012	3,460,113.4	4.97
2013	3,715,738.8	4.88
2014	3,749,898.8	4.72
2015	3,842,439.3	4.89

entitled "Costs of health care by the main groups of the International Classification of Diseases" is the most important and useful for health politics decision-making. Although there is still no widely accepted clear methodological instructions for establishing table 6 (except the framework, that was used in the present study), international comparisons based on this table have started to perform now [27]. International comparability is actually major advantage of the Table 6. Its disadvantage, however, is that the cost estimates in different countries vary in scope and methodology of the assessment, so that they are based on a large set of assumptions or very small samples.

The total cost of health care for the residents of the Republic of Serbia by the ICD from 2010 to 2015 increased from about 151 billion to almost 194 billion RSD. Expenditure per capita expressed in euros has also increased during the period, from 195 euros in 2010 up to 223 euros in 2015. Expenditure per capita expressed in US dollars during this period recorded the same changes. By groups of diseases, the highest health care cost in the period 2010-2015 in the Republic of Serbia was used for the treatment of patients with cardiovascular diseases, and during this period increased thirty percent. Compared to the increase of funds for other disease groups, this was growth higher than average.

When this cost is compared with findings from Australia, Canada, France, Germany and the Netherlands [27], it is evident that in these countries the largest assets within the healthcare were allocated for cardiovascular diseases, which is most likely the result of today's rapid and stressful lifestyle.

Unlike Serbia, where funds allocated for neoplasm are on the second place, followed by diseases of the digestive

system, infectious and parasitic diseases, and diseases of the nervous system in other analyzed countries diseases of the nervous system are the second largest participation funding, followed by diseases of digestive system, musculoskeletal system and neoplasm.

At the end of this period (2015) after cardiovascular diseases (19.8%) that had the highest consumer funds, the second place was for diseases of the digestive system (10.67%), followed by neoplasm (10.13%). Cardiovascular diseases (CVD) represents a major economic burden to the health care system in Serbia in terms of direct costs (e.g., hospitalization, rehabilitation, doctor visits, drugs) and indirect costs related to the mortality and morbidity (e.g., productivity loss due to premature death, and short-term or long-term disability). When the results of the cost of CVD in Serbia is compared with the same cost from Australia, Canada and some European countries, it is evident that in these countries, the situation tends to be very similar in terms of health care costs.

A review of studies published in Australia, revealed that cardiovascular diseases had the highest level of health care expenditure compared to any group of diseases in Australia, and that puts them in front of the funds allocated for oral health, mental disorders, and musculoskeletal diseases. According to their research, spending on CVD has remained relatively stable at around 12% of all expenditures for the period 2000-2009, while the cost of CVD in Serbia is still increasing [28]. In the observed period the total cost reached 19% of all health care costs in 2015.

When compared by years, the total health care cost by major ICD categories showed tendency of growth, increasing by almost thirty percent. Rising costs in the Republic of Serbia for the treatment of diseases over the years suggest insufficient investment in prevention, public health services, capital investment and other functions related to the health care.

The percentage share of health care costs by major ICD categories by the GDP for the period 2010-2015 dropped from 5.20% to 4.89% in 2015 showing that there is room for more funds to be allocated to the health care. There are no available data for other countries and other years so data are not comparable.

CONCLUSION

Individual costs of treating patients in the period 2010-2015 increased by thirty percent and the largest funds in Serbia was allocated to treat people with cardiovascular diseases. The cost of treating cardiovascular diseases is likely to continue to grow due to increased stress, bad habits, increased rates of obesity and aging society. To minimize steadily increasing individual costs for cardiovascular diseases, it is crucial to work on prevention and promotion of CVD and development of a comprehensive study that will provide detailed understanding of the cost of CVD and its main drivers.

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Troškovi individualne zdravstvene zaštite u Srbiji po međunarodnoj klasifikaciji bolesti za period 2010–2015. godine

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KRATAK SADRŽAJ

Uvod U svetu raste interesovanje za izračunavanjem troškova lečenja određenih bolesti. Ova vrednost predstavlja breme kojim određena bolest ili grupa oboljenja opterećuje društvo u uslovima globalne krize (Segel 2006). Godine 2000. Organizacija za ekonomski razvoj država (OECD) formirala je Sistem zdravstvenih računa (SZR), okvirno metodološko uputstvo za izračunavanje troškova lečenja bolesti.

Cilj ovog rada bio je da se utvrde troškovi individualne zdravstvene zaštite u Srbiji prema Međunarodnoj klasifikaciji bolesti (MKB) u periodu 2010–2015. godine.

Materijal i metode rada Urađena je retrospektivna i komparativna analiza zdravstvenih statističkih podataka iz baze Instituta za javno zdravlje Srbije i finansijskih podataka Republičkog fonda za zdravstveno osiguranje za period 2010–2015. godine. Finansijski podaci sa podacima o uslugama bolničkog, ambulantnog i kućnog lečenja, pomoćnim uslugama zdravstvene zaštite, potrošnji lekova i potrošnih dobara u zdravstvu su analizirani primenom SZR metodologije.

Rezultati Tokom posmatranog perioda najveći trošak za lečenje bolesnika ostvaren je 2015. godine, a iznosio je 194.128.864.011 dinara (1.580.853.941 evra; 1.764.807.854 dolara), dok je najmanji ostvaren 2010. godine, a bio je 151.333.139.835 dinara (1.434.464.541 evra; 1.908.843.843 dolara).

Zaključak Komparativna analiza troškova lečenja bolesti je pokazala da su troškovi lečenja bolesnika u periodu 2010–2015. porasli trideset posto, a da su najveća novčana sredstva u Srbiji izdvojena za lečenje osoba sa bolestima cirkulatornog sistema.

Ključne reči: troškovi bolesti; zdravstvena potrošnja; zdravstveni računi; kardiovaskularne bolesti

UVOD

Zdravstvena zaštita je jedna od najvažnijih ljudskih delatnosti i jedna od najdinamičnijih u pogledu rasta troškova za njeno obezbeđenje. Brojna dostignuća u medicini, farmaceutici, medicinskim tehnologijama, utiču na poboljšanje zdravstvenog stanja ljudi u većini zemalja, ali i na rast troškova, pa je briga za uspostavljanje efikasnosti i efektivnosti zdravstvene zaštite sve više u fokusu zdravstvene politike. Troškovi zdravstvene zaštite predstavljaju teret kojim određena bolest ili grupa oboljenja opterećuje društvenu zajednicu [1].

Potreba za realnim prikazom finansijskih podataka u zdravstvenoj zaštiti, a posebno onih koji se odnose na praćenje troškova za lečenje bolesnika od određenih bolesti nameće se kao neophodnost. U skladu sa tim poslednjih godina raste interesovanje za izračunavanje troškova lečenja bolesnika [2, 3, 4], a studije širom sveta obrađuju kako pojedine bolesti [5–16], tako i povrede [17], poremećaje [18] i stanja [19–24].

Godine 2000. Organizacija za ekonomski razvoj država (*Organisation for Economic Co-operation and Development – OECD*) formirala je Sistem zdravstvenih računa (SZR), koji je formulisao okvirno metodološko uputstvo za izračunavanje cene lečenja bolesnika, kroz tabelu broj 6. Do pojave novog metodološkog uputstva SZR 2011. koristila se tabela broj 6 u SZR [25], kao deo Nacionalnog zdravstvenog računa (NZR). U pomenutoj tabeli prikazani su tekući troškovi zdravstvene zaštite prema glavnim grupama oboljenja Međunarodne klasifikacije bolesti (MKB).

U Sistemu zdravstvenih računa su kao troškovi INDIVIDUALNE zdravstvene zaštite definisani svi oni troškovi u zdravstvenom sistemu koji ne uključuju kolektivnu zdravtsvenu zaštitu, koju čine usluge javnog zdravstva, kolektivna prevencija, zdravstveno osiguranje, zdravstvena administracija i troškovi za vršenje funkcija u vezi sa zdravstvenom zaštitom.

Cilj ovog rada je bio da se izvrši komparativna analiza troškova individualne zdravstvene zaštite u Republici Srbiji prema glavnim grupama oboljenja MKB za period 2010–2015. godine.

MATERIJAL I METODE RADA

Urađena je komparativna retrospektivna analiza statističkih podataka iz baze Instituta za javno zdravlje Srbije (IZJZS) i finansijskih podataka Republičkog fonda za zdravstveno osiguranje (RFZO) za period 2010–2015. godine.

Podaci o izvršenim uslugama bolničkog, ambulantnog i kućnog lečenja, kao i dnevne nege, pomoćnim uslugama zdravstvene zaštite, potrošnji lekova i potrošnih dobara u zdravstvu analizirani su i ukršteni su sa finansijskim podacima iz Republičkog fonda za zdravstveno osiguranje po metodološkom uputstvu SZR (verzija 11).

Troškovi individualne zdravstvene zaštite u Republici Srbiji prema glavnim kategorijama MKB rađeni su po sledećoj shemi i metodologiji SZR 11:

- H.C.1. – usluge bolničkog lečenja su finansijski izražene tako što je broj bolničkih dana po grupama bolesti (izvor podataka: IZJZS [26]) pomnožen sa cenom bolničkog dana iz važećeg Cenovnika zdravstvenih usluga RFZO;
- H.C.1.2. – usluge dnevne nege su registrovane po grupama bolesti i pomnožene sa cenom dana iz važećeg Cenovnika zdravstvenih usluga RFZO;
- H.C.1.3. – finansijska sredstva utrošena za ambulantno lečenje bolesnika dobijena su tako što je broj usluga ambulantnog lečenja (izvor podataka: IZJZS) pomnožen sa cenom iz važećeg Cenovnika zdravstvenih usluga RFZO;
- H.C.1.4. – usluge kućnog lečenja finansijski su izražene tako što su usluge kućnog lečenja (izvor podataka: planske tabele za domove zdravlja koje obrađuje IZJZS) pomno-

žene sa cenom iz važećeg Cenovnika zdravstvenih usluga RFZO;
- H.C.4. – pomoćne usluge zdravstvene zaštite (laboratorijske analize, dijagnostika i prevoz bolesnika) finansijski su izražene tako što je iskustveno procenjen ukupan broj tih usluga pomnožen s odgovarajućim cenama iz važećeg Cenovnika zdravstvenih usluga RFZO;
- H.C.5.1. – finansijski prikaz utroška lekova i drugih potrošnih dobara dobijen je od Agencije za lekove i medicinska sredstva Srbije.

Zbir stavki H.C.1, H.C.1.2, H.C.1.3, H.C.1.4, H.C.4. i H.C.5.1. po grupama bolesti daje procenjenu finansijsku vrednost ukupne cene zdravstvene zaštite stanovnika Srbije prema grupama oboljenja MKB.

U analizi su primenjene komparativna i retrospektivna metoda istraživanja. Za analizu su korišćeni i podaci Republičkog fonda za statistiku (RZS) i Narodne banke Srbije (NBS).

U narednim tabelama prikazane su šifre grupa oboljenja prema Međunarodnoj klasifikaciji bolesti (Tabela 1), kao i Tabela 6, koja se koristi za izračunavanje troškova individualne zdravstvene zaštite (Tabela 2).

REZULTATI

Rezultati su odredili troškove individualne zdravstvene zaštite po glavnim kategorijama Međunarodne klasifikacije bolesti (MKB) (Tabela 3).

Ukupna novčana sredstva (izražena u dinarima, evrima i američkim dolarima) koja su utrošena na zdravstvenu zaštitu stanovnika Srbije tokom posmatranih godina, prema glavnim grupama oboljenja MKB, prikazana su u tabeli 3.

Ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama u 2010. godini bili su **151.331.867.999 dinara.** Posmatrano po grupama bolesti najveći individualni troškovi su izdvojeni za bolesti cirkulatornog sistema (19,58%), neoplazme (10%), bolesti digestivnog sistema (9,75%), bolesti nervnog sistema (8,33%), infektivne i parazitarne bolesti (8,99%), dok najmanje za urođene anomalije (0,27%) (Grafikon 6). U 2010. godini ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama po glavi stanovnika iznosili su 195,00 evra, dok je procenat ukupnih individualnih troškova u odnosu na GDP za 2010. godinu iznosio 5,20%.

151.614.999.373 dinara u 2011. godini bili su ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama. Posmatrano po grupama bolesti najveći individualni troškovi su izdvojeni za bolesti cirkulatornog sistema (19,14%), bolesti digestivnog sistema (9,71%), infektivne i parazitarne bolesti (8,88%) i za neoplazme (8,90%), dok najmanje za urođene anomalije (0,30%) (Grafikon 7). Ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama ukupno u 2011. godini po glavi stanovnika iznosili su 205,00 evra, dok je procenat ukupnih individualnih troškova u odnosu na GDP za 2011. godinu iznosio 4,77%.

Ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama u 2012. godini bili su **166.575.285.407** dinara. Najveći individualni troškovi posmatrano po grupama bolesti su izdvojeni za bolesti cirkulatornog sistema (20,69%), bolesti digestivnog sistema (10,38%), neoplazme (10,07%), bolesti nervnog sistema (9,00%), infektivne i parazitarne bolesti (8,69%), dok najmanje za urođene anomalije (0,23%) (Grafikon 8). U 2012. godini ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama ukupno po glavi stanovnika iznosili su 205,00 evra, dok je procenat ukupnih individualnih troškova u odnosu na GDP za 2012. godinu iznosio 4,97%.

U 2013. godini ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama bili su **176.734.078.012** dinara. Posmatrano po grupama bolesti, najveći individualni troškovi su izdvojeni za bolesti cirkulatornog sistema (20,44%), neoplazme (10,84%), bolesti digestivnog sistema (10,62%), bolesti nervnog sistema (8,74%), infektivne i parazitarne bolesti (8,51%), a najmanje za urođene anomalije (0,21%) (Grafikon 9). Individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama ukupno u 2013. godini po glavi stanovnika iznosili su 219,00 evra, dok je procenat ukupnih individualnih troškova u odnosu na GDP za 2013. godinu iznosio 4,88%.

Ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama u 2014. godini bili su **183.189.009.508** dinara. Najveći individualni troškovi posmatrano po grupama bolesti su izdvojeni za bolesti cirkulatornog sistema (18,62%), bolesti nervnog sistema (11,79), bolesti digestivnog sistema (10,74%), neoplazme (10,62%), infektivne i parazitarne bolesti (8,79%), a najmanje za urođene anomalije (0,34%) i perinatalna stanja (1,15%) (Grafikon 10). U 2014. godini ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama po glavi stanovnika iznosili su 217,00 evra, dok je procenat ukupnih individualnih troškova u odnosu na GDP za 2014. godinu iznosio 4,72%.

Najveći trošak za individualnu zdravstvenu zaštitu po Međunarodnoj klasifikaciji bolesti MKB-10 je ostvaren 2015. godine i iznosio je 194.128.864.011 dinara (1.580.853.941 evra; 1.764.807.854 dolara). U 2015. godini najviše je potrošeno na bolesti krvotoka (19,08%), bolesti digestivnog sistema (10,67%), neoplazme (10,13%), a najmanje za urođene anomalije (0,32%). Rezultati izraženi u dinarima prikazani su u grafikonu broj 11. Ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama u 2015. godini po glavi stanovnika iznosili su 223,00 evra, dok je procenat ukupnih individualnih troškova u odnosu na GDP za 2015. godinu iznosio 4,89%. Posmatrano po godinama (2010–2015), ukupni individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama (grupe bolesti po MKB-10 klasifikaciji) imaju tendenciju rasta, izraženu po tekućim cenama (Grafikon 12).

Posmatrano po grupama bolesti, najveći individualni troškovi u periodu od 2010. do 2015. godine su izdvojeni za kardiovaskularne bolesti. Procentualno učešće ukupnih individualnih troškova za zdravstvenu zaštitu u periodu od 2010. do 2015. godine izraženo po glavi stanovnika u dinarima, evrima i dolarima prikazano je u sledećoj tabeli (Tabela 4.).

Individualni troškovi za zdravstvenu zaštitu po glavnim MKB kategorijama po stanovniku imaju trend rasta od 195 evra u 2010. godini do 223 evra u 2015. godini. Procentualno učešće ukupnih troškova za zdravstvenu zaštitu po glavnim ICD-10 kategorijama u bruto domaćem proizvodu (BDP) u periodu od 2010. do 2015. imalo je pad od 5,20% do 4,89% 2015. godine. U 2011. godini je zabeležen nagli pad, sa stalnim fluktuacijama u periodu od 2012. do 2015. godine (Tabela 5.).

DISKUSIJA

Pregledom istraživanja koja se bave troškovima lečenja bolesnika [2, 3, 4] uvidelo se da većina studija obrađuje samo pojedine bolesti [5–16], odnosno pojedinačne povrede [17], poremećaje [18] ili stanja [19–24]. Tekući troškovi zdravstvene zaštite prema glavnim grupama bolesti MKB predviđeni su da budu prikazani samo u tabeli 6 SZR, kao deo Nacionalnih zdravstvenih računa (NZR) [25]. Mnogi korisnici analiza NZR smatraju da je upravo tabela 6 u SZR, pod nazivom „Troškovi za zdravstvenu zaštitu po glavnim grupama Međunarodne klasifikacije bolesti" (kategorije MKB), najvažnija i najkorisnija donosiocima zdravstvene politike. Premda još ne postoji jasno opšteprihvaćeno metodološko uputstvo za izradu tabele 6 (osim okvirnog, koje je korišćeno i u ovom istraživanju), međunarodna poređenja na osnovu ove tabele, iako i dalje vrlo retka, ipak su počela da se vrše [27]. Upravo ta međunarodna uporedivost je glavna prednost tabele 6. Njen nedostatak, međutim, jeste to što se procene troškova u različitim zemljama razlikuju u obuhvatu i metodologiji procene, tako da se zasnivaju na velikom skupu pretpostavki, odnosno veoma malim uzorcima.

Ukupni troškovi zdravstvene zaštite stanovnika Republike Srbije prema glavnim oboljenjima MKB su se od 2010. do 2015. godine povećali sa oko 151 milijardi na skoro 194 milijarde dinara. Troškovi po glavi stanovnika izraženi u evrima takođe su se tokom posmatranog perioda povećali, i to sa 195 evra u 2010. godini na 223 evra 2015. godine. Troškovi po glavi stanovnika izraženi u dolarima tokom posmatranog perioda beleže iste promene.

Posmatrano po grupama bolesti, najveći troškovi zdravstvene zaštite u periodu od 2010. do 2015. godine u Republici Srbiji izdvojeni su za lečenje osoba sa kardiovaskularnim oboljenjima, a tokom posmatranog vremenskog perioda povećani su za trideset procenata. To, u poređenju s rastom finansijskih sredstava za druge grupe bolesti, predstavlja rast koji je viši od prosečnog.

Kada se rezultati ovih troškova uporede sa nalazima iz Australije, Kanade, Francuske, Nemačke i Holandije [27], vidi se da se i u tim zemljama najveća sredstva u okviru zdravstvene zaštite izdvajaju za kardiovaskularna oboljenja, što je najverovatnije posledica današnjeg ubrzanog i stresnog načina života.

Za razliku od Srbije, gde se sredstva izdvojena za tumore nalaze na drugom mestu po potrošnji u posmatranom periodu, a prate ih bolesti digestivnog sistema, infektivne i parazitarne bolesti i bolesti nervnog sistema, u pomenutim zemljama bolesti nervnog sistema se nalaze na drugom mestu po učešću finansijskih sredstava, a slede ih bolesti digestivnog sistema, koštano-mišićnog sistema i tumori.

Na kraju posmatranog perioda, posle kardiovaskularnih bolesti (19,08%), koje beleže najveću potrošnju finansijskih sredstava, na drugom mestu sa najvećom potrošnjom finansijskih sredstava su bolesti digestivnog sistema (10,67%), a slede neoplazme (10,13%). Kardiovaskularne bolesti (KVB) predstavljaju veliki ekonomski teret za sistem zdravstvene zaštite u Srbiji u smislu direktnih troškova (npr. hospitalizacija, rehabilitacija, poseta lekaru, lekovi) i indirektnih troškova vezanih za mortalitet i morbiditet (na primer, gubici produktivnosti zbog prerane smrti i kratkoročni ili dugoročni invaliditet). Kada su rezultati troškova KVB u Srbiji porede sa pregledom studija i njihovih nalaza iz Australije, Kanade i nekih evropskih zemlja, evidentno je da u ovim zemljama situacija teži da bude veoma slična u smislu zdravstvenih troškova.

Pregled studija objavljenih u Australiji pokazuje da kardiovaskularne bolesti predstavljaju najviši nivo zdravstvene potrošnje u odnosu na bilo koju grupu bolesti u Australiji, što ih stavlja ispred potrošnje za oralno zdravlje, mentalne poremećaje i mišićno-skeletna oboljenja. Prema australijskom istraživanju, potrošnja za KVB je ostala relativno stabilna, na oko 12% svih rashoda za period 2000–2009. godine zdravstvene zaštite, dok su troškovi za KVB u Srbiji u stalnom porastu [28]. U posmatranom periodu su dostigli 19% svih troškova zdravstvene zaštite u 2015. godini.

Posmatrano po godinama, ukupni troškovi zdravstvene zaštite po glavnim MKB-10 kategorijama su pokazali tendenciju rasta i povećani su skoro trideset procenata. Ova činjenica govori u prilog rastu troškova u Republici Srbiji za lečenje bolesti tokom godina, i nedovoljnim ulaganjima u prevenciju, usluge javnog zdravlja, kapitalne investicije, kao i druge funkcije koje se odnose na zdravstvenu zaštitu.

Procenat učešća troškova za zdravstvenu zaštitu po glavnim MKB-10 kategorijama u BDP Srbije u periodu 2010–2015. smanjen je sa 5,20% na 4,89% u 2015. godini, i oni pokazuju da postoji prostor za više izdvojenih sredstava za zdravstvenu zaštitu. Ne postoje dostupni podaci za druge zemlje i druge godine, pa tako nisu uporedivi.

ZAKLJUČAK

Komparativna analiza troškova lečenja bolesti je pokazala da su individualni troškovi lečenja bolesnika u periodu 2010–2015 porasli trideset posto, da su najveća novčana sredstva u Srbiji izdvojena za lečenje osoba sa bolestima kardiovaskularnog sistema. Troškovi za kardiovaskularne bolesti će verovatno nastaviti da rastu, usled povećanog stresa, loših navika, povećane stope gojaznosti i starenja društva. Kako bi se smanjilo stalno povećanje individualnih troškova za kardiovaskularne bolesti, nameće se potreba rada na prevenciji i promociji KVB i izradi sveobuhvatne studije koja će omogućiti detaljno razumevanje troškova KVB i njenih glavnih pokretača.

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Dental and Jaws Status in Pre-historic Human Population of the Gomolava Site

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SUMMARY

Introduction Knowledge of biological and cultural heritage represents a significant basis for the advance of human civilization. The aim of this study was to determine and define dental health status of pre-historic people of the Vinca culture.

Material and method Study included twenty skeleton remnants of different gender and age from anthropological series of Gomolava. Skeletons originated from one necropolis and two individual graves dating from mid and early Neolithic periods of the Vinca culture. Based on radiocarbon analysis absolute skeletal age was determined to date within the periods from 5848±38 to 5739±35 BC. Preservation of skeletons varied from completely preserved skulls and jaws to fragmented parts of jaws. Data analysis was performed with methodology used in the research of human population teeth and jaws from the Lepenski Vir culture.

Results Results showed high level of teeth abrasion (98,1%), medium level of dental calculus deposits (44,9%), low level of tooth decay, significant number of retained roots, as well as the occurrence of periapical lesions and periodontal disease within the neolith population of Gomolava site.

Conclusion Taking into account absolute age of examined skeletons, collected data are very significant, from the perspective of its wide content. From a pathological perspective, teeth abrasion stands out as a dominant feature, while tooth decay fits within standard values for human population of the neolith period.

Keywords: the Vinca culture; Gomolava; teeth; abrasion; tooth decay

INTRODUCTION

Archaeological site Gomolava is located on the left bank of the river Sava in a wider region of Ruma municipality, the Republic of Serbia (Picture 1). First individual, amateur exploration of this site dates back as far as to the beginning of the 20th century, while first systematic ones were done on two occasions half a century later when larger sites were found and explored (over 3600m^2) [1-5]. First phase took place between 1954 and 1957. These explorations found that the site holds a significant cultural layer, which indicates continuity and length of colonisation to this area [3, 4, 6]. Through the second phase which went on for two decades (1965–1985) archaeologists like Brukner, Jovanovic and Tasic defined the stratigraphic site location and determined that the rich cultural layer, which thickness ranges from 5.5 to 6.5 m, contains remains of settlements from seven different cultural periods [1]. Based on material, cultural traces and remains of human skeleton found between the first cultural layer, Gomolava I, originating from late Neolith regarded as the period of Vinca culture, and the last one, Gomolava VII, belonging to the Roman period, there is possibility to explore human activity in a timeframe of over 6000 years [4, 5, 7-11]. Several necropolis and individual graves were found during excavation [2, 5, 8, 9, 12, 13]. Twenty human skeletons found at Gomolava I date back to late Neolith, e.g. the Vinca culture. These are significant and numerous findings with respect to soil characteristics these skeletons were excavated from. It is known that the soil, where skeletons were buried is very aggressive, leading to fast disintegration of biological remains. From this group, physical anthropologist Z. Zoffmann extracted and anthropologically processed fourteen adult skeletons, assorting them to a "very gracile type of atlanto-mediterranean taxons within the Carpathian basin", except two (2/75 & 16) which she assorted to the Cro-Magnon group. Anthropological analysis determined that the average height of pre-historic habitants of Gomolava ranged from 1576 to 1767 mm. Also, finding of pathological changes on skeletons is very deficient (occasional blow or arthrosis). Regarding dental parameters from that period, a relatively high rate of tooth decay was identified (3.7%) [12].

The aim of this study was to review and define dental health status of pre-historic people dating from the Vinca culture period, based on the analysis of teeth and jaws of preserved remains of human skeleton skulls from the anthropological group of Gomolava.

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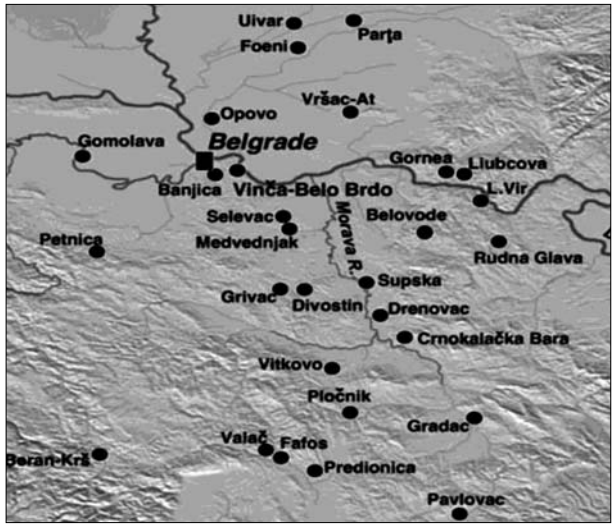


Picture 1. The map of the neolithic sites on the territory of Serbia (Borić D.)
Slika 1. Mapa neolitskih lokaliteta na tlu Srbije (Borić D.)

Picture 2. Original photo of the necropola from the Gomolava archeological site (exhibition of the Museum of Vojvodina Novi Sad)
Slika 2. Originalna fotografija nekropole sa lokaliteta Gomolava (postavka Vojvođanskog muzeja Novi Sad)

MATERIAL AND METHOD

Study included twenty skeleton remains of different gender and age. Skeletons originated from one necropolis and two individual graves dating from mid and early Neolithic periods of the Vinca culture (Picture 2). Based on radiocarbon analysis absolute skeletal age was determined to date within the periods from 5848±38 to 5739±35 BC [10, 11, 14]. Preservation of skeletons varied from completely preserved skulls and jaws to fragmented parts of jaws. Data analysis was performed with the methodology used in the research of human population teeth and jaws from the Lepenski Vir culture [15, 16, 17]. Table for basic information data included: skeleton number, individuals' age and gender (data taken from previously published anthropological analysis). In the Table for jaws preservation assessment number 1 stands for complete preservation, 0.5 for partial and 0 for the absence of examined jaw segment. Tables for lower and upper jaw generate a wide range of data (7 columns and 16 rows). First column shows data on dentition type. Second column generates information on teeth status and consists of 6 types of data: 1 – present tooth, 2 – tooth lost during lifetime, 3 – tooth lost after death, 4 – present tooth root, 5 – tooth root lost after death, 6 – tooth germ. Third column shows data on tooth surfaces affected with tooth decay numerically defined on a 1-5 scale, (1 – occlusal and incisal, 2 – mesial, 3 – distal, 4 – vestibular, 5 – oral). Fourth column consists of data assessing tooth decay depth and it is numerated 1-4, (1 – superficial, 2 – medium and deep, 3 – dental cavity trepanation, 4 – radix). Data on teeth abrasion were summerized up in the fifth table and expressed on a 0-4 scale (0 – tooth without abrasion, 1 – enamel abrasion, 2 – dentin abrasion, 3 – abrasion with dental cavity trepanation, 4 – radix). Sixth column shows data on existing levels of tooth tartar on a 0-3 scale, (0 – without tartar, 1 – 1/3 of tooth covered by tartar, 2 – 2/3 of tooth covered by tartar, 3 – tooth completely covered by tartar). Seventh column generates data on existing macroscopically visible periapical lesions (0 – not present, 1 – ≤5mm in diameter, 2 – >5mm in diameter).

Ordinary number of teeth [17, 18, 19] defined the number of rows in each of the two tables. All eventual findings on exceeded number of teeth were stated as anomalies in the space for additional comments.

RESULTS AND DISCUSSION

It was established after dental examination that there is not enough dental status data for the three individuals from graves 8, 14 and 15, and that the skeleton number 13 was on the display "in sittu" as a permanent museum exponent and not available for analysis. Therefore, the total number of samples included in this study was 16 skeletons, two children and 14 aduls. Adult group consisted of 13 male and 1 female individual.

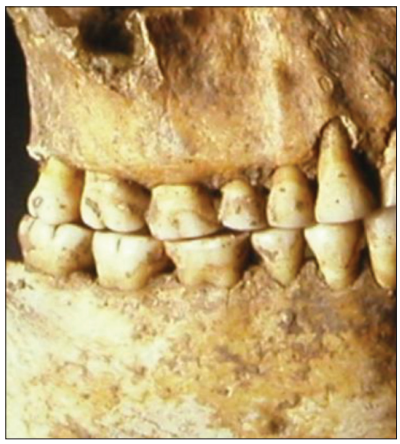
Oral status of 14 adult individuals is shown in Table 1. There were 256 present teeth, 28 have been lost during lifetime and 79 lost after death during excavation or storing in the museum collection. There were 28 tooth roots found due to pathological processes that occurred during life. It was found with high certainty, that for 6 teeth, roots existed in the moment of death that indicated their loss after death. Through macroscopic analysis data was gathered on 406 teeth in total.

Knowing that both aforementioned taxons that examined skeletons belong to have number of teeth and teeth formula similar to present-day humans (32 teeth: 8 incisors, 4 canines, 8 premolars and 12 molars), it can be said that the status of 90% of teeth was defined within this study, with respect to the initial assumption that ideally (completely preserved dentition) there would be a sample of 448 teeth present (14x32).

Beside aforementioned, it is also possible to add results on data from another 11 molars (5 upper and 6 lower), representing 2.9% of all teeth. It was determined with high certainty these teeth did not erupt before the mo-

Table 1. Oral status of adult individuals from neolith necropolis of Gomolava site
Tabela 1. Oralni status odraslih osoba iz neolitske nekropole sa lokaliteta Gomolava

Maxilla and mandible – adults Maksila i mandibula – odrasle osobe												
Type Tip	**ST.**	**ST %**	**K.Pov.**	**K.Pov %**	**K.D.**	**K.D.%**	**Abr.**	**Abr. %**	**Kam.**	**Kam. %**	**A.Par.**	**A.Par %**
1	265	65.27%	2	0.75%	2	0.75%	42	15.85%	106	40.00%	9	3.40%
2	28	6.90%	1	0.38%	1	0.38%	137	51.70%	12	4.53%	2	0.75%
3	79	19.46%	1	0.38%	1	0.38%	43	16.23%	1	0.38%	0	0.00%
4	28	6.90%	0	0.00%	0	0.00%	38	14.34%	0	0.00%	0	0.00%
5	6	1.48%	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
6	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Total Suma	406	100.00%	4	1.51%	4	1.51%	260	98.11%	119	44.91%	11	4.15%



Picture 3. Tooth abrasion
Slika 3. Abrazija zuba

Picture 4. Class II decay
Slika 4. Karijes II klase

Picture 5. Periapical lesions above the remaining tooth roots
Slika 5. Periapikalne lezije iznad zaostalih korenova

ment of death of an individual. It's valid to assume that in one sample (skeleton number 17) all four third molars were missing, due to the fact that the individual was less than 17 years old at the moment of death. Other seven third molars were probably left impacted because of their belonging to already mature individuals with formed dentition at the moment of death.

For the 29 teeth left (6.5%) it was not possible to collect data due to partial or complete devastation of jaws. In one case (skeleton number 13) maxillofacial segment was not complete, lacking the lower jaw with all 16 teeth (3.6%).

Optimal number of 448 teeth, which theoretically could have been present in all adult individuals at the moment of death, lacks another 13 teeth (2.9%) as it was not possible to determine their status due to skeletal devastation and loss of alveolar bone.

Teeth abrasion was dominant pathological finding in this neolith group. From 265 present teeth, this pathological process was found on 260 of them (98.1%). Different levels of tooth wearing were present. Most common was level 2 abrasion present on 137 (51.7%) of the total number of present teeth (Picture 3). There was similar number of teeth found with level 3 – 43 teeth (16.2%) and level 1 abrasion – 42 teeth (15.9%). The most severe case of abrasion (level 4) with certain dental cavity trepanation and total crown loss was identified on 38 teeth (14.3%). Similar findings for the neolith culture of Lepenski Vir were significantly lower, indicating that slightly more than half of total number of teeth was exposed to some level of abrasion (55%) [16]. Apart from that, abrasion was an eminent occurrence for all human populations of mesolith and neolith. Explanation to such tooth wear in the neolith population of Gomolava, can be found in the fact that 13 of 14 examined skeletons (93%) belong to mature and senile life chapters, meaning these individuals were in their fifth, sixth or seventh life decade at the moment of death.

Three male individuals belonging to mature age category were identified to have tooth decay. Tooth decay was found on 5 (1.9 %) of the total number of present teeth. Two lower incisors of the same skeleton were affected by tooth decay level 2 on their facing proximal surfaces. On the upper jaw of the same skeleton, level 1 tooth decay was identified on the occlusal surface of second molar. Other two teeth with decay were found on different skeletons. Both developed on molars. One affected two surfaces reaching as far as dentin (Picture 4) and the other one developed over three surfaces, causing dental cavity trepanation (level 3).

Dental calculus deposits were identified on 119 present teeth (44.9%). Most of them were level 1 deposit on 106 teeth (89%). One tooth was completely covered by calculus (level 3).

Due to resorption and perforation of alveolar bone during life, or devastation of porous bone under grave soil, it was sometimes possible to macroscopically detect

Table 2. Dental status of childhood individuals from neolith necropolis of Gomolava site
Tabela 2. Status zuba u dečjem uzrastu iz neolitske nekropole sa lokaliteta Gomolava

Skeletal number Red. br. skeleta	Permanent teeth Stalni zubi	Lost teeth during lifetime Izgubljeni tokom života	Lost teeth after death Izgubljeni posle smrti	Primary teeth Mlečni zubi	Lost teeth during lifetime Izgubljeni tokom života	Lost teeth after death Izgubljeni posle smrti	Permanent teeth buds Zametak stalnih zuba	Total Ukupno
4	9	0	3	10	0	2	2	26
8	7	0	4	9	1	3	0	24
Total Ukupno	16	0	7	19	1	5	2	50

Table 3. Oral status of childhood individuals from neolith necropolis of Gomolava site
Tabela 3. Oralni status osoba dečjeg uzrasta iz neolitske nekropole sa lokaliteta Gomolava

Maxilla and mandible – children Maksila i mandibula – deca												
Type Tip	ST.	ST %	K.Pov.	K.Pov %	K.D.	K.D.%	Abr.	Abr. %	Kam.	Kam. %	A.Par.	A.Par %
1	35	70.00%	0	0.00%	0	0	5	14.29%	0	0	0	0
2	1	2.00%	4	11.43%	4	11.43%	4	11.43%	0	0	0	0
3	12	24.00%	0	0.00%	0	0	0	0.00%	0	0	0	0
4	0	0.00%	0	0.00%	0	0	0	0.00%	0	0	0	0
5	0	0.00%	0	0.00%	0	0	0	0.00%	0	0	0	0
6	2	4.00%	0	0.00%	0	0	0	0.00%	0	0	0	0
Total Suma	50	100.00%	4	11.43%	0	0	9	25.71%	0	0	0	0

periapical changes on teeth. This study examined and identified such lesions in 12 cases. In 10 cases the extent of the lesion was level 1, and in other two level 2 lesions were found (Picture 5).

Changes to the alveolar bone were also found on some of the skeletal remains of human populations of Gomolava site, which indicated the presence of periodontal disease. Eight persons had alveolar bone resorption and roots exposed in the level of their furcation. These resorptions ranged from 3 to 7 mm.

This study also showed two significant findings on tooth roots status. The first one was the number of roots present (28), which was relatively high and equal to the number of teeth lost during life, especially when there are 6 samples of shallow and non-healed alveolus indicating roots came out just before, or more probably after death. Second one shows tooth crowns were completely devastated under some pathological process or trauma during life. It was not rare to detect periapical processes around tooth roots. Analysis of this osteological material showed the presence of 28 tooth roots within jaws of 8 individuals, as well as another 6 cases where root was lost after death. In the apical area of identified roots 5 periapical lesions were registered (four level 1 and one level 2). Etiological factor for this pathological occurrence on 31 teeth was most probably abrasion. Tooth decay was not present on other teeth, while surrounding ones had level 3 and level 4 abrasions. In three cases loss of tooth crown could be assigned to tooth decay, because other teeth are preserved, with low-level abrasion. In that case, percentage of tooth decay for the Gomolava human population would rise from 1.95 to 3%, which is still lower than the values determined by Zofmann [12]. In her studies tooth decay was found in 3.7% of cases that is significantly higher than average values for that period. Regarding tooth decay, it's incidence during neolith on the territory of modern Europe varied from 1.4% to 3.2% and that is in accordance with findings of the current study for the Gomolava site. Analysis of teeth from skeletons in the Iron Gate series from the territory of Iron Gate (modern day Serbia) found that tooth decay affected 1.28% teeth of 3.76% individuals from the neolith culture of Lepenski Vir [15, 17, 19-22].

Child skeletons belonged to the infants II age group, being at the time of death between 6-8 years of age, with presence of mixed dentition. It was concluded that the data in this study showed information on primary dentition and positions of first and second permanent molars and incisors. Permanent dentition of both individuals consisted of four first molars and two second molar germs in the upper jaw of one individual. Permanent dentition also included 8 incisors (5 upper and 3 lower). It was found that at the moment of death, additional seven permanent teeth were present, probably lost during excavation or storing. All permanent teeth were healthy, without any present tooth decay, abrasion, deposits, anomalies or periodontal changes (Table 2).

Primary dentition had 19 teeth present (47%) mostly molars (16) and canines (3). Remaining three canines were lost after death; one primary incisor was lost during life confirmed by the presence of agnate alveolus. One person was identified with class II tooth decay, which developed over occlusal and proximal surfaces of all four upper molars. Nine primary teeth were affected with abrasion (five level 1, four level 2). Deposits on teeth, tooth and jaw anomalies as well as changes on the periodontal tissues were not detected (Table 3).

This study showed with high certainty that after eight millenniums two children from the period of the Vinca culture, had completely preserved dentition that suits their age at the time of their death. Their skeletal remains showed they had all primary molars and canines, four permanent molars both, one primary and 15 permanent

incisors with one primary incisor being lost during life. Level 1 and level 2 abrasions were present on primary teeth, and in one case four teeth of class II level 2 tooth decay (caries media).

CONCLUSION

Taking into account the absolute age of examined skeletons, collected data are very significant, from the perspective of its wide content. Almost 8 millenniums ago human population had two dentitions. Tooth abrasion was dominant feature detected on most permanent teeth. In childhood it appeared in mixed dentition on primary teeth. Tooth decay affected both permanent and primary teeth, in low percentage, as typical for that period. Dental calculus was identified on almost half of teeth present. Signs of periodontal disease and periapical lesions were found.

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Status zuba i vilica praistorijske humane populacije sa lokaliteta Gomolava

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KRATAK SADRŽAJ

Uvod Poznavanje biološkog i kulturnog nasleđa je bitna pretpostavka daljeg civilizacijskog napretka. Cilj ovog rada je bio da se na osnovu očuvanih humanih skeletnih ostataka lobanja i vilica antropološke serije sa Gomolave, analizom zuba i vilica, sagleda i definiše zubno-zdravstveni status praistorijskih ljudi iz perioda Vinčanske kulture.

Materijal i metod Istraživanja su obuhvatila dvadeset skeletnih ostataka, različitog pola i individualne starosti. Skeleti potiču iz jedne nekropole i dva pojedinačna groba srednjeg i mlađeg neolita iz perioda Vinčanske kulture. Apsolutna starost skeleta je radiokarbonskom analizom ugljenika C14 datovana u periodu od 5848. ±38 do 5739. ±35 godina pre nove ere. Očuvanost skeleta je bila različita i kretala se od kompletno sačuvanih lobanja i vilica do fragmentovanih delova vilica. Podaci su analizirani metodologijom koja je primenjivana u istraživanjima zuba i vilica humane populacije Kulture Lepenskog Vira.

Rezultati Rezultati su ukazali na visok stepen abrazije zuba (98.1%) neolitske populacije sa lokaliteta Gomolava, umereni stepen naslaga (44,9%), nizak stepen karijesa, značajan broj zaostalih korenova, kao i pojavu periapikalnih procesa i parodontopatije.

Zaključak Uzimajući u obzir apsolutnu starost ispitivanih skeleta, prikupljeni podaci su izuzetno vredni po obimu građe i njenom sadržaju. Patološkom slikom dominira abrazija, dok je karijes prisutan u okviru standardnih vrednosti za humanu populaciju iz perioda neolita.

Ključne reči: Vinčanska kultura; Gomolava; zubi; abrazija; karijes

UVOD

Arheološki lokalitet Gomolava nalazi se na levoj obali reke Save u širem regionu grada Rume u Republici Srbiji (Slika 1). Prva pojedinačna, amaterska istraživanja ovog lokaliteta datiraju još sa početka 20. veka, a sistematskim istraživanjima se pristupilo pola veka kasnije kada su u dva navrata otkrivene i istražene velike površine (preko 3.600 m^2) [1–5]. Prva faza se odvijala od 1954. do 1957. godine, a istraživanja su otkrila da lokalitet sadrži značajan kulturni sloj koji ukazuje na kontinuitet i dužinu naseljavanja ovog prostora [3, 4, 6]. U drugoj fazi, koja je trajala dve decenije (1965–1985), istraživači poput Bruknera, Jovanovića, Tasića definisali su stratigrafski položaj lokaliteta i utvrdili da u veoma moćnom kulturnom sloju, čija se debljina kretala u rasponu od 5,5 do 6,5 metara, postoje ostaci naselja čak sedam različitih kulturnih perioda [1]. Između prvog kulturnog sloja, Gomolava I, koji potiče iz kasnog neolita, odnosno perioda Vinčanske kulture, i poslednjeg, Gomolava VII, koji pripada rimskom periodu, postoje tragovi materijalne kulture i ostaci humanih skeleta, na osnovu kojih se može pratiti kontinuitet života u vremenskom rasponu od preko 6.000 godina [4, 5, 7–11]. Tokom iskopavanja otkriveno je više nekropola i pojedinačnih grobova [2, 5, 8, 9, 12, 13]. Dvadeset skeleta pronađenih u horizontu Gomolava I datovano je u period kasnog neolita, odnosno Vinčanske kulture. Ovo je izuzetno značajan i brojan nalaz s obzirom na osobine tla u kome su skeleti pronađeni. Poznato je da je zemljište u kome su skeleti sahranjeni veoma agresivno i da dovodi do brze dezintegracije bioloških ostataka. Iz ove grupe fizički antropolog Z. Zoffmann je izdvojila i antropološki obradila četrnaest skeleta odraslih osoba koje je svrstala u „jednu gracilnu varijantu atlantomediteranskih taksona u Karpatskom basenu", sem dva skeleta (2/75 i 16) koja je opredelila u Kromanjonsku grupu. Antropološkom analizom je ustanovljeno da se prosečna visina praistorijskih stanovnika Gomolave kretala u rasponu od 1.576 do 1.767 mm, kao i da je nalaz patoloških promena na skeletima veoma oskudan (poneki udarac ili artroza). Od stomatoloških parametara, konstatovana je relativno visoka stopa karijesa za taj period (3,7%) [12].

Cilj ovog rada je bio da se na osnovu očuvanih humanih skeletnih ostataka lobanja i vilica antropološke serije sa Gomolave, analizom zuba i vilica sagleda i definiše zubno-zdravstveni status praistoriskih ljudi iz perioda Vinčanske kulture.

MATERIJAL I METOD RADA

Istraživanja su obuhvatila dvadeset skeletnih ostataka, različitog pola i individualne starosti. Skeleti potiču iz jedne nekropole, i dva pojedinačna groba srednjeg i mlađeg neolita iz perioda Vinčanske kulture (Slika 2). Apsolutna starost skeleta je radiokarbonskom analizom ugljenika C14 datovana u period od 5848. ±38 do 5739. ±35 godine pre nove ere [10, 11, 14]. Očuvanost skeleta je bila različita i kretala se od kompletno sačuvanih lobanja i vilica do fragmentovanih delova vilica.

Podaci su analizirani metodologijom koja je primenjivana u istraživanjima zuba i vilica humane populacije Kulture Lepenskog Vira [15, 16, 17]. U tabeli za osnovne podatke generisani su: broj skeleta, individualna starost i pol individue (podaci preuzeti iz prethodno obavljenih antropoloških analiza). U tabeli za procenu očuvanosti vilica brojem 1 označavano je kompletno prisustvo ispitivanog dela vilice, sa 0,5 delimično, a 0 je označavala odsustvo tog dela. Tabele za donju i gornju vilicu generišu veliki broj podataka (sedam kolona i 16 redova). Prva kolona sadrži podatake o vrsti denticije. Druga kolona generiše podatke o statusu zuba i ona sadrži šest vrsta podataka: 1 – prisutan zub, 2 – izgubljen tokom života, 3 – izgubljen posle smrti, 4 – prisutan koren zuba, 5 – koren zuba izgubljen posle smrti,

6 – zametak zuba. U treću su unošeni podaci o površinama zuba zahvaćenih karijesom, i definisana je numerički od 1 do 5 u skladu sa brojem površina zuba (1 – okluzalno i incizalno, 2 – mezijalno, 3 – distalno, 4 – vestibularno, 5 – oralno). Četvrta kolona sadrži ocenu dubine karijesa i kreće se od 1 do 4 (1 – superficijalis, 2 – medija i profunda, 3 – trepanacija kavuma dentis, 4 – radiks). Podaci o abraziji zuba su unošeni u petu kolonu i stepenovani od 0 do 4 (0 – zub bez abrazije, 1 – abrazija gleđi, 2 – abrazija dentina, 3 – abrazija sa trepanacijom kavuma dentis i 4 – radiks). Šesta kolona sadrži podatke o prisustvu čvrstih zubnih naslaga kroz četiri stepena (0 – bez naslaga, 1 – prekrivena 1/3 zuba, 2 – prekrivene 2/3 i 3 – prekriven ceo zub). Kolona 8 generiše podatke o prisustvu makroskopski vidljivih periapikalnih lezija (0 – nema, 1 – do 5 mm u prečniku, 2 – preko 5 mm u prečniku).

Uobičajen broj zuba [17, 18, 19] definisao je i broj redova u svakoj od ove dve tabele. Svi eventualni nalazi o prekobrojnim zubima unošeni su kao konstantovane anomalije u prostor za posebne napomene.

REZULTATI I DISKUSIJA

Posle obavljenih stomatoloških pregleda ustanovljeno je da za tri individue iz grobova 8, 14 i 15 nema dovoljno stomatoloških podataka, a da je skelet br. 13 izložen *in situ* kao stalni muzejski eksponat, nedostupan za analizu, tako da je ukupan uzorak obuhvaćen ovom studijom iznosio 16 skeleta. Od toga, dva skeleta su pripadala dečjem uzrastu, a 14 odraslim osobama. U grupi odraslih 13 je bilo muškog, a jedan ženskog pola.

Kod 14 odraslih osoba ustanovljen je sledeći stomatološki status (Tabela 1): prisutno je bilo 265 zuba, izgubljeno u toku života 28, dok je 79 zuba izgubljeno posle smrti osobe, tokom iskopavanja ili čuvanja u zbirci muzeja. Očuvanih korenova zuba nastalih usled patoloških procesa bilo je 28, a za šest zuba je pouzdano ustanovljeno da su korenovi bili prisutni u trenutku smrti, odnosno da su izgubljeni posle egzitusa individue. Sumarno posmatrano, makroskopskom analizom dobijeni su podaci za 406 zuba.

S obzirom na to da oba pomenuta taksona kojima pripadaju ispitivani skeleti imaju broj i formulu zuba sličnu savremenom čoveku (32 zuba: osam inciziva, četiri očnjaka, osam premolara i 12 molara), može se reći da je ovim istraživanjima definisan status preko 90% zuba, uz početnu pretpostavka da bi se u idealnim uslovima sa kompletnim zubnim nizovima mogao očekivati uzorak od 448 zuba (14 × 32).

Pored navedenog, ovim rezultatima se mogu priključiti i podaci o još 11 trećih molara (pet gornjih i šest donjih), ili 2,9% svih zuba, za koje je pouzdano utvrđeno da do trenutka smrti osobe nije izniklo. Opravdano je pretpostaviti da jednoj osobi (skelet br. 17) nedostaju sva četiri umnjaka, zbog uzrasta u kom se nalazila – u trenutku smrti bila je mlađa od 17 godina. Ostalih sedam umnjaka je najverovatnije ostalo inpaktirano, jer su u pitanju osobe koje su u trenutku smrti bile u zreloj životnoj dobi, sa formiranim zubnim nizovima.

Za preostalih 29 zuba (6,5%) nije bilo moguće prikupiti podatke zbog delimične devastiranosti ili kompletnog gubitka viličnih kostiju. U jednom slučaju (skelet br. 13) maksilofacijalni segment nije bio kompletan, pošto je nedostajala mandibula sa svih 16 zuba (3,6%).

Do optimalnog broja od 448 zuba, koje su teoretski mogle imati ove odrasle osobe u trenutku smrti, nedostaje još 13 zuba (2,9%), za koje nije bilo moguće utvrditi status, usled devastiranosti skeleta i gubitka alveolarne kosti.

Proces abrazije je dominantna patološka pojava i u ovoj grupi skeleta iz perioda neolita. Od 265 prisutnih zuba na 260 (98,1%) uočava se ovaj patološki proces. Trošenje zuba je bilo različitog stepena. Najčešće je bila prisutna abrazija 2. stepena i to kod 137 zuba (51,7%) od svih prisutnih zuba (Slika 3). Približne vrednosti su ustanovljene za abraziju 3. stepena, kojom je zahvaćeno 43 zuba (16,2%) i abraziju 1. stepena, kojom je bilo zahvaćeno 42 zuba (15,9%). Najteži oblik abrazije (4. stepen), uz obavezno otvaranje komore pulpe i gubitak kompletne krunice, uočen je na 38 zuba (14,3%). Nalazi za neolitski deo Kulture Lepenskog Vira su znatno nižih vrednosti i ukazuju da je tek nešto više od polovine zuba bilo zahvaćeno nekim stepenom abrazije (55%) [16]. Inače, abrazija je imanentna pojava za sve humane populacije mezolita i neolita. Objašnjenje ovako velikog trošenja zuba u neolitskoj humanoj populaciji sa lokaliteta Gomolava leži u činjenici da od 14 istraženih skeleta 13 (93%) pripada životnoj dobi maturus i senilis, odnosno da su se osobe u trenutku smrti nalazile u starijem životnom dobu, odnosno u petoj, šestoj ili sedmoj deceniji života.

Karijes je otkriven kod tri osobe muškog pola, koje su svrstane u starosnu kategoriju maturus. Karijes se razvio na pet zuba (1,9%) od svih prisutnih zuba. Kod dva donja sekutića, na istom skeletu, zahvaćena je po jedna naspramna aproksimalna površina sa dubinom 2. stepena. Na istom skeletu u maksili je, na drugom molaru, otkriven karijes 1. stepena na okluzalnoj površini. Preostala dva zuba sa karijesom otkrivena su na različitim skeletima. Oba su se razvila na molarima. Jedan je zahvatio dve površine sa prodorom u dentin (Slika 4), a drugi se proširio na tri površine, i pri tome razorio zub uz trepanaciju komore pulpe (3. stepena).

Naslage na zubima u vidu zubnog kamenca konstatovane su na 119 prisutnih zuba (44,9%). Najviše zuba je bilo sa naslagama 1. stepena – 106 (89%). Uočen je jedan zub 3. stepena, koji je u potpunosti bio prekriven kamencem.

Zbog resorpcije i perforacije alveolarne kosti tokom života, ili usled brže devastacije poroznije kosti tokom boravka u zemlji, nekada je moguće makroskopski uočiti periapeksne promene na zubima. Ovim istraživanjem su uočene takve promene u 12 slučajeva. Veličina lezija u deset slučajeva je bila 1. stepena, a u dva slučaja uočene lezije su svrstane u 2. stepen (Slika 5).

Na skeletnim ostacima ljudi iz neolitske populacije sa lokaliteta Gomolava zapažene su promene na alveolarnoj kosti, koje su ukazivale na postojanje parodontopatije. Kod osam osoba je uočena resorpcija alveolarne kosti i eksponiranje korenova zuba i njhovih furkacija. Te resorpcije su se kretale od 3 do 7 mm.

Nalazi vezani za status korenova zuba su veoma zanimljivi i zato su zahtevali posebnu pažnju. Prvi razlog za toliku pažnju je broj prisutnih korenova (28), koji je relativno visok i identičan broju izgubljenih zuba tokom života. Posebno kada se tome priključi i šest nalaza plitke i nezarasle alveole, koja ukazuje da su korenovi iz njih ispali neposredno pred smrt, ili, što je verovatnije, tek posle smrti. Drugi značajan razlog za njihovu analizu proističe iz činjenice da su kompletne krunice tih zuba razorene usled nekog patološkog procesa ili zaživotne traume. Nije retka situacija da se iznad zaostalih korenova mogu uočiti i periapikalni procesi. Analizom ovog osteološkog materijala

ustanovljeno je da u vilicama osam osoba perzistira 28 radiksa, kao i još šest slučajeva kod kojih je koren izgubljen posle smrti individue. Iznad konstatovanih radiksa registrovano je pet periapeksnih promena (četiri 1. stepena i jedna 2. stepena). Etiološki faktor za nastanak ove patološke promene na 31 zubu je najverovatnije abrazija, jer karijes na ostalim zubima nije bio prisutan, a okolni zubi su imali izraženu abraziju 3. i 4. stepena. U tri slučaja gubitak krunice bi mogao da se pripiše karijesu, jer su ostali zubi očuvani, sa niskim stepenom abrazije. U tom slučaju procenat karijesa za humanu populaciju sa Gomolave bi se sa 1,9% povećao na 3%, što je i dalje ispod vrednosti koje je na sličnom uzorku ustanovila Zoffmann [12]. U njenim istraživanjima karijes se javlja u 3,7%, što je znatno iznad prosečnih vrednosti za pojavu karijesa toga doba. Inače, karijes incidenca tokom trajanja neolita na području današnje Evrope se kretala u rasponu od 1,4% do 3,2%, što je u skladu sa našim nalazima za lokalitet Gomolava. Analizom zuba skeleta Đerdapske serije sa područja Đerdapske klisure ustanovljeno je da je karijesom bilo zahvaćeno 1,28% zuba kod 3,76% osoba iz neolitskog perioda Kulture Lepenskog Vira [15, 17, 19–22].

Dečji skeleti su pripadali uzrastu infans II, individualne starosti između 6 i 8 godina sa prisutnom mešovitom denticijom. Ustanovljeno je da su prikupljeni podaci pružali informacije o mlečnoj denticiji, pozicijama prvih i drugih stalnih molara i sekutića. Od stalne denticije kod obe individue bila su prisutna sva četiri prva molara, a kod jedne i dva zametka drugih molara u gornjoj vilici. Od stalnih zuba prisutno je bilo osam sekutića (pet gornjih i tri donja). Utvrđeno je da je u trenutku smrti bilo prisutno još sedam stalnih zuba, koji su najverovatnije izgubljeni tokom iskopavanja ili skladištenja. Svi stalni zubi su zdravi, bez karijesa, abrazije, naslaga, anomalija i bez promena u parodoncijumu (Tabela 2).

Od mlečne denticije bilo je prisutno 19 zuba (47%) i to svi molari (16) i tri očnjaka. Preostala tri očnjaka su izgubljena posle smrti. Jedan mlečni sekutić je izgubljen tokom života, o čemu svedoči srasla alveola. Kod jedne osobe otkriven je karijes II klase, koji je zahvatao okluzalne i aproksimalne površine sva četiri gornja molara. Devet mlečnih zuba je bilo zahvaćeno abrazijom (pet 1. stepena, a četiri 2. stepena). Naslage na zubima, anomalije zuba i vilica, promene u parodoncijumu nisu ustanovljene (Tabela 3).

Dragocen je podatak da se iz današnje perspektive posle skoro osam milenijuma može pouzdano tvrditi da su dvoje dece iz perioda Vinčanske kulture imali potpuno očuvanu denticiju koja odgovara uzrastu u kome su se nalazili u trenutku smrti. Njihovi skeletni ostaci pokazuju da su imali sve mlečne molare i očnjake, po četiri stalna molara, jedan mlečni i 15 stalnih sekutića uz jedan i mlečni sekutić izgubljen tokom života, o čemu svedoči srasla alveola. Imali su abraziju na mlečnim zubima 1. i 2. stepena, i u jednom slučaju četiri karijesa II klase, srednje dubine (karijes medija).

ZAKLJUČAK

Uzimajući u obzir apsolutnu starost ispitivanih skeleta, prikupljeni podaci su izuzetno vredni po obimu građe i njenom sadržaju.

Pre skoro osam milenijuma humana populacija je imala dve denticije. Abrazija zuba je bila dominantna pojava i zahvatala je veliku većinu stalnih zuba. U dečjem uzrastu pojavljivala se u mešovitoj denticiji na mlečnim zubima. Od karijesa su obolevali stalni i mlečni zubi, u niskom procentu, tipičnom za taj period. Na gotovo polovini prisutnih zuba konstatovane su tvrde zubne naslage. Uočeni su i znaci parodontopatije i periapikalnih oboljenja.

ZAHVALNOST

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Scaffold in bone tissue engineering

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SUMMARY

Treatment of bone tissue injuries and diseases is still a great challenge for surgeons, but also for researchers who work with materials. Today stem cells are commonly used in bone tissue engineering. However, advances in biocompatible materials design, especially biodegradable porous structure (scaffold) is gaining an important role in the treatment of diseased bone tissue. The basic advantage of these carriers is specifically designed scaffold with defined porosity and pore structure that is favourable for cells settlement. Scaffolds are most commonly used as ceramic brackets because they have excellent characteristics in biodegradation and bioactivity. The process of scaffold production is important because the appropriate technology must ensure control of liquids and reproducibility of scaffold production through standardized process.

The aim of this study was to present some of different procedures of scaffold production in bone tissue engineering and point out the advantages and disadvantages of these methods.

Keywords: scaffold; bone tissue engineering; the polymer matrix; scaffold prototype

INTRODUCTION

The treatments of trauma and bone tissue diseases, as well as reconstruction of bone defects represent a great challenge for orthopaedic surgeons and engineers [1]. The most common procedures in tissue engineering involve the use of stem cells or differentiated adults cells that are plated in a biodegradable scaffold and cultured in a bioreactor, before implanting in the defective area. Advances in design and functionalization of biocompatible materials and the progress of processing, allow development of biodegradable porous structure with well-designed architecture – scaffold for tissue engineering [1, 2].

Scaffold should be designed in a specific way, with appropriate porosity and biodegradability, and meet specific requirements for individual defects, such as their shape and size. From the technological aspect, design and production of biodegradable scaffold represent great challenge in skeletal tissue engineering, due to defined porosity and pore structure, that are suitable for settling cells and can be maintained for a long time [3].

Ceramic brackets can be used as scaffold with well degradation properties and bioactivity. With such mechanical properties, they are used during new bone forming as polymeric carriers, which hydrophilicity encapsules cells (similar to the natural extracellular matrix) [4]. Procedural processing techniques of conventional polymer materials are popular, tailored and extended to the installation of bioactive inorganic phase in a porous 3D polymer network.

In addition, implantation of bioactive molecules into biodegradable scaffold promotes bone regeneration with many positive effects [4, 5]. Big challenge in materials science and tissue engineering technology is control of accuracy and reproducibility of scaffold production through standardized process. Different techniques of scaffold producing, which include processing of various polymeric and composite materials and development of different microstructures are very current topics in research. Despite numerous techniques applied, each of them has certain disadvantages in the control of scaffold porosity, pore size and distribution, as well as the presence of residues of toxic solvents in the scaffold.

The aim of this study was to present some of different methodological procedures used in scaffold production in bone tissue engineering.

METHODS OF SCAFFOLD PRODUCTION

Scaffold producing process must ensure high level of control of their macro- and micro-structural properties. Depending on scaffold's material and strategies of tissue engineering, there are different methodologies and conditions of scaffold processing to optimize predetermined purpose. Correspondingly, the process procedure, in any of the specific cases should be selected not to change chemical and biocompatible properties of the material, and not to limit its clinical effects. In addition, scaffold should have interconnected pores and sufficiently high density of pores with proper morphology, size and distribution, and its quality should be highly reproducible.

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The method of polymer matrix

Among many methods of designing scaffold structure, one of the most commonly used methods is the polymer matrix (foam) that is used as a model system for designing ceramic scaffold structure. This method comprises applying suspension of ceramic powder through the matrix and, after drying and solidification of the suspension, burning of polymeric foam to provide porous ceramic with a porosity that depends on matrix [6, 7].

In our research, we used matrix of polyurethane foam as a model system for obtaining and formatting internal geometry of hydroxyapatite (HAP) scaffold [8, 9]. After the combustion process of the polymer phase and the ceramic phase sintering at 800 °C, the resulting structure of the scaffold is obtained (Figure 1).

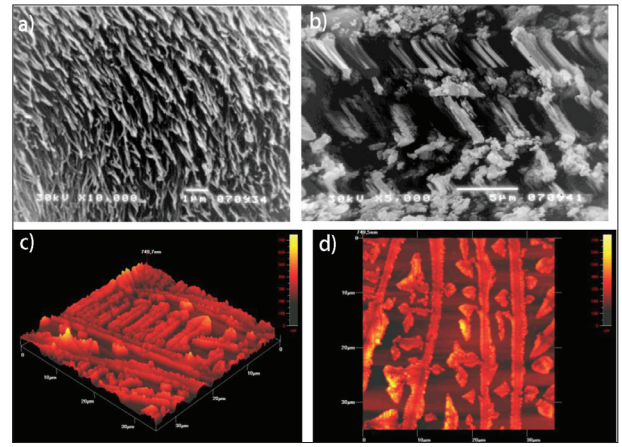

Figure 1. A typical layout structure HAP scaffold obtained by the polymer foam method: a) SEM recordings; b) AFM recordings
Slika 1. Tipičan izgled strukture HAP skafolda dobijenog metodom polimerne pene: a) SEM snimci; b) AFM snimci

The combination methods of polymer matrices and biomimicry

To functionalize the interior walls of scaffold obtained with polymeric matrix method, in our experiments, we used different types of polymers, such as poly (lactic-co-glycolic) acid (PLGA), hydroxyethyl cellulose, modified starch, alginate, etc. [10]. Subsequent biomimicry treatment in simulating body fluid allowed us to obtain active scaffold structure with specific structural design (Figure 2).

Beside these polymers, and other polymers, such as polycaprolactone, hydroxyethyl chitosan, biopolymers with RGD motifs, silk fibroin, as well as various growth factors may be involved in the functionalization of interior walls of scaffold. In our experiments we used materials with specific physical properties, such as super-paramagnetic materials-based ferro-fluid magnetite, maghemite, cobalt-ferrite, gadolinium oxide, etc. [10].

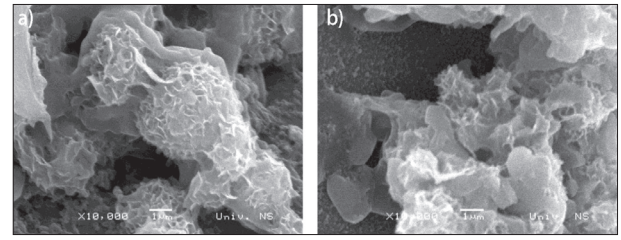

Figure 2. Scaffold obtained by combining the methods of polymer matrix, deposition of a thin polymeric film and biomimicry: a) a polymer film, PLGA; b) polymeric film hydroxy-ethyl cellulose
Slika 2. Skafoldi dobijeni kombinacijom metode polimerne matrice, depozicije tankog polimernog filma i biomimične metode: a) polimerni film, PLGA; b) polimerni film hidroksietilceluloze

Gas-foam method with the effect of supercritical CO_2

The supercritical fluid of substance is obtained when the substance is exposed to an environment where the temperature and pressure are above the critical pressure and temperature for that substance. Under these conditions, liquid and gaseous material components become identical, and further compression of the fluid phase will not result in dissolution [11, 12, 13].

CO_2 is most frequently used as the supercritical fluid, because it is low toxic, non-flammable, inexpensive, stable, and environmentally acceptable. It is used in foam-gas technique, where CO_2 under high pressure is used for scaffold processing (critical temperature of 31°C and pressure of 73.8 bar). In order to be properly processed by a mesoporous structure, a polymer disk is exposed to high pressure of (1-6 MPa) at room temperature at the beginning of the process, to allow saturation of the gas in the polymer and formation of a single phase between the gas and the polymer. Gas solubility in the polymer rapidly decreases with a decrease in pressure and CO_2 leads to nucleation and growth of gas bubbles, leading to the formation of pores larger than 500 mm. Macroporous foam poly-L-lactic acid (PLLA), polyglycolic acid (PGA) and poly-lactic-co-glycolic acid (PLGA) is obtained from such polymer discs inside which at high pressures of CO_2 an appropriate mixture of the polymer-gas is formed. Then comes a phase where gas molecules cluster, forming gas nuclei which diffuse, while inside of polymer discs macropores are left behind. Porosity and structure depend on the quantity of gas dissolved in the polymer, gas nucleation velocity and diffusion rate of gas molecules through the polymer (Figure 3) [12].

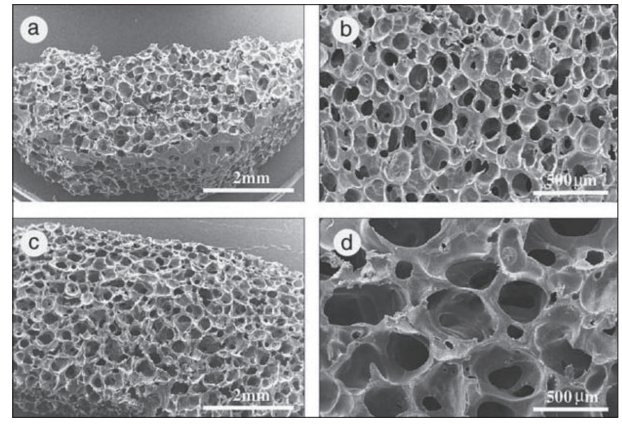

Figure 3. PLLA polymer after processing with supercritical CO2 at 240 bar, and 35°C for 12 minutes (a, b) and 60 min. (c, d) [12]
Slika 3. PLLA polimer posle procesiranja sa superkritičnim CO2 na 240 bari i 35°C u toku 12 minuta (a, b) i 60 minuta (c, d) [12]

Method of phase separation

During the phase separation, thermodynamic instability is established in a homogeneous multicomponent polymer solution, which under appropriate conditions indicates a tendency to split into more than one phase to reduce its total free energy. The polymer solution is separated into two phases, the phase rich in polymer and the phase poor in polymer. A polymer rich phase solidifies and the polymer poor phase is removed forming highly porous polymer network. Method of phase separation is known as thermal induced phase separation (TIPS). The basic principle of TIPS is to move polymer solution from its homogeneous single-phase region in the spinodal region of its binary system phase using rapid temperature change of the solution. This can be done by rapidly cooling polymer solution at the upper critical temperature or rapid heating at the lower critical temperature. When polymer solution is moved to the spinodal region, it becomes unstable and separates spontaneously into polymer-rich and solvent-rich region [14, 15, 16].

After thermodynamic separation of the homogenous solution of polymer-solvent, in the polymer rich and polymer-poor phase, the solution is usually exposed to another solvent or cooling below its binodal solubility. Then the solvent is removed by freeze drying, leaving the polymer in the form of foam. Morphology of the resulting scaffold is controlled with the phase transition that occurs during cooling, i.e. liquid-liquid phase separation, while in the solid-liquid phase anisotropic foam in the specific leaf form is obtained.

In general, micro and macrostructure of the polymer, obtained by the method of phase separation is possible to control with optimal selection of the process parameters such as polymer concentration, temperature and speed of separation. Incomplete solvent removal, especially in thicker constructs, results in reduced compatibility, and a change of the built-active factors. Although this method results in highly interconnected porous matrix, limited pore size obtained by this method causes serious problems related to the establishment of controlled macro- and microstructure scaffold (Figure 4) [17].

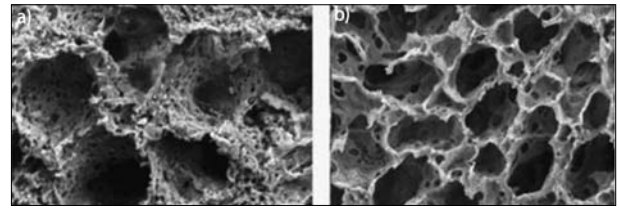

Figure 4. PLGA / nano-hydroxyapatite (95 : 5) scaffold prepared from a 10% (w/v) of a mixture of PLGA / dioxane / water with different solvent systems: a) water, 1,4-dioxane 85:15 and b) 1,4-dioxane water 87 : 13 [17]
Slika 4. PLGA/nanohidroksiapatit (95 : 5) skafold pripremljen iz 10% (w/v) smeše PLGA/ dioksan/ voda sa različitim sastavima rastvora: a) 1,4-dioksan – voda 85 : 15 i b) 1,4-dioksan – voda 87 : 13 [17]

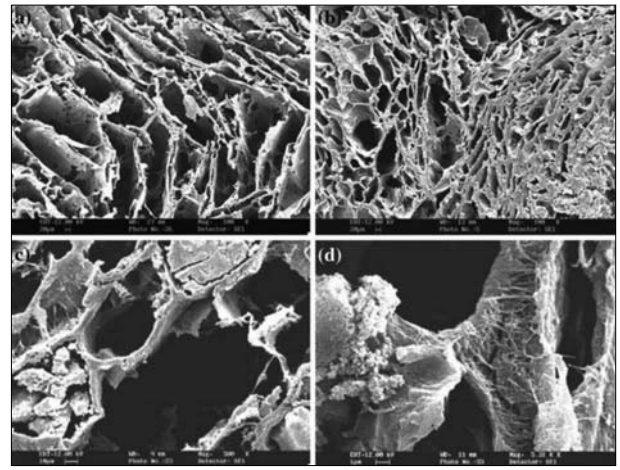

Figure 5. Scaffold obtained with sumblimation technique at least 10% (w/v) of emulsion: a) pure poly hydroxybutyrate-co-valerate (PHVB); b) 10% hydroxyapatite-HA: 90% PHVB; c and d) 20% HA: 90% PHVB [22]
Slika 5. Skafoldi dobijeni tehnikom sumblimacionog sušenja 10% (w/v) emulzije: a) čisti polihidroksibutirat-ko-valerat (PHVB); b) 10% hidroksiapatit-HA: 90% PHVB; c i d) 20% HA: 90% PHVB [22]

Method of sublimation drying

Sublimation drying (freeze drying) involves solvent removal (usually water), first with sublimation of frozen samples and then desorption of non frozen absorbed solvent under reduced pressure [18]. In the stage of freezing, polymer solution or dispersion is cooled below the temperature of solvent solidification. This leads to the formation of ice crystals and deployment of polymer molecules in the spaces between the ice crystals. In the second stage, the solvent is removed by applying pressure that is lower than the equilibrium vapour pressure of the frozen solvent. In the meantime, non-frozen water absorbed in the dried layer is desorbed. When frozen solvent is completely sublimated, the process continues with a slight heating of the sample until it is completely dried. Sublimation of ice crystals causes formation of highly porous sponge-like scaffold structure.

The final pore structure depends on conditions during the preparation of scaffold such as pH value, the speed of freezing and partial pressure. Rapid uncontrolled cooling leads to different nucleation and grow of ice crystals, and hence different morphological heterogeneity within the scaffold, caused by physical and temporal variables during the transfer of heat through the system. It has been shown that it is possible to obtain substantially uniformly porous scaffold at a constant cooling rate during the freezing process. Highly porous and interconnected structure with small pore size is achieved using the freeze-drying technique [18].

Sublimation method of drying the emulsion, as modified by the phase separation method is used in the production of aliphatic polyester scaffold. Scaffolds obtained by this method have porosity above 90%, while the mean pore size is between 15 and 35 microns, with the largest pores larger than 200 microns (Figure 5). Scaffold porosity primarily depends on the volume fraction of the disperse phase, polymer concentration and its molecular weight. Technique of sublimation drying of emulsion is used for the installation of proteins into the polymer scaffold. Sublimation drying of aqueous solutions of biopolymers such

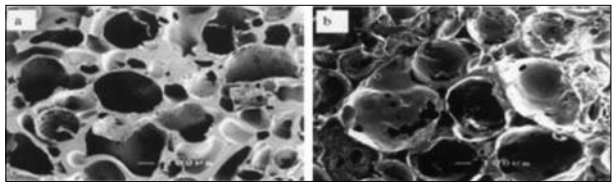

Figure 6. SEM: intersection of PLGA sponge obtained by the method of leaching (a-PLGA, b-hybrid PLGA-collagen) [17]
Slika 6. SEM: presek PLGA sunđera dobijenih metodom luženja (a-PLGA, b-hibrid PLGA-kolagen) [17]

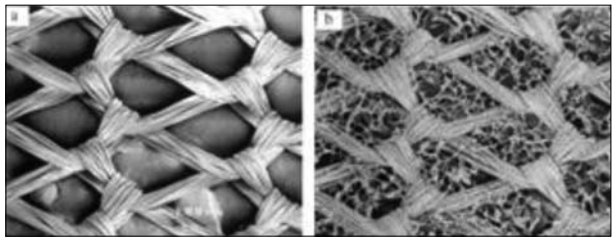

Figure 7. SEM: cross-section of the PLGA-collagen-hydroxy-apatite hybrid foam after 2 (a) and 4 (b) alternating cycles of dipping [24]
Slika 7. SEM: presek PLGA-kolagen-hidroksiapatit hibridnog sunđera posle dva (a) i četiri (b) alternirana ciklusa umakanja [24]

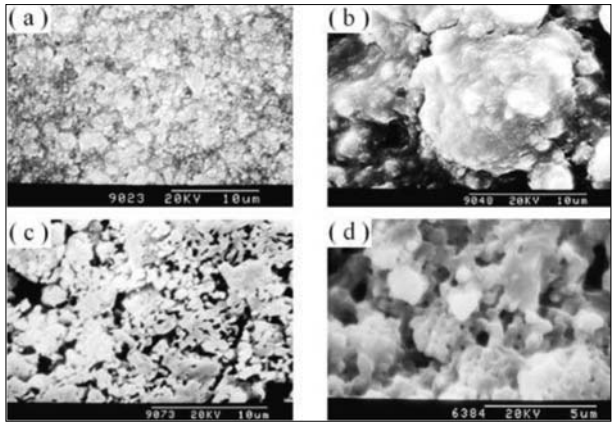

Figure 8. SEM: sintered coatings obtained with EPD in various dispersion media at different temperatures and vacuum sintering: a) ethanol, 800°C; b) glycol, 800°C; c) ethanol, 1000°C; d) glycol, 1000°C [26]
Slika 8. SEM: sinterovane prevlake dobijene sa EPD u raznim disperzionim medijumima pri različitim temperaturama vakumskog sinterovanja: a) etanol, 800°C; b) glikol, 800°C; c) etanol, 1000°C; d) glikol, 1000°C [26]

as collagen is also used in obtaining well-defined porous matrices (well-defined pore size and orientation) [19-22].

The method of casting / leaching of porogen

Casting solvent allows preparing porous structure with normal porosity but of limited thickness [17, 23]. A method of casting / leaching of porogen involves casting of the mixture of polymer and porogen into a mold, drying the mixture, followed by leaching of porogens with water and finally obtaining the pores. Salts are most commonly used porogens. Pore structure is most easily controllable with porogens contribution. Highly porous PLLA membrane with controlled porosity, the ratio of area:volume, and crystallinity are prepared by casting a dispersion of crystalline salts in the organic solution of PLLA, and then exposure to water. NaCl, Na-tartrate and Na-citrate with different particle sizes are used as materials for pore forming by leaching. Properties of obtained foam depend exclusively on fractions of salt and size of its particles, and not on the type of polymer solvent used. Therefore, it is used for the preparation of porous three-dimensional scaffold suitable for tissue engineering (Figure 6). With this technique it is possible to obtain highly porous scaffold structure with a porosity of over 93% and an average diameter of pores up to 500 microns.

The main advantage of this method is easy production of scaffold, without need for any expensive special equipment. This technique allows obtaining wide range of pore size and porosity that is easily controlled. The disadvantage of this method is the necessity to remove solvent particles inside the polymer. Casting method combined with leaching can be used to create a thin membrane or 3D samples of small thickness (up to 2 mm). One of the main disadvantages of this method is porogen particle agglomeration during the process of obtaining scaffold, causing uneven distribution of scaffold pores. Another drawback is linked to the use of organic solvents. They must be removed in order to avoid possible damage of seeded cells and proteins or other active molecules that are built into it [23].

Combined method of sublimation drying and casting porogen

Hybrid sponge of synthetic polymer, collagen and hydroxyapatite is suitable for the deposition of hydroxyapatite on its surface, because it mimics the structure of collagen and hydroxyapatite of the natural bone (Figure 7) [24]. Deposition of hydroxyapatite takes place through the process of alternating immersion PLGA-collagen sponge in aqueous $CaCl_2$ and Na_2HPO_4. PLGA-collagen sponge is first immersed in a solution of $CaCl_2$ in vacuum at 37°C for 12h, so that the space within the sponge is soaked with the best possible solution. After removing from $CaCl_2$ solution, the sponge is centrifuged to remove excess solution of Na_2HPO_4. This process of alternating immersion and centrifugation defines a cycle of depositing hydroxyapatite within the foam structure of PLGA-collagen scaffold. The deposition process is usually performed in a series of repeated cycles.

Electrophoretic deposition method

The method of electrophoretic deposition (EPD) is one of the most effective techniques in assembling fine particles of hydroxyapatite. It is very simple and it is possible to obtain very complex forms of particles. The equipment for performing EPD is cheap. At first, this method was mainly used for the deposition of hydroxyapatite coatings on metal or ceramics, while the deposition of sufficiently thick layers of apatite of adequate porosity is still great challenge [25]. Therefore, attention is on obtaining stable dispersions of hydroxyapatite, with high values of zeta potential and an appropriate concentration, that would in suitably selected conditions of depositing be able to give appropriate scaffold structure (Figure 8) [26].

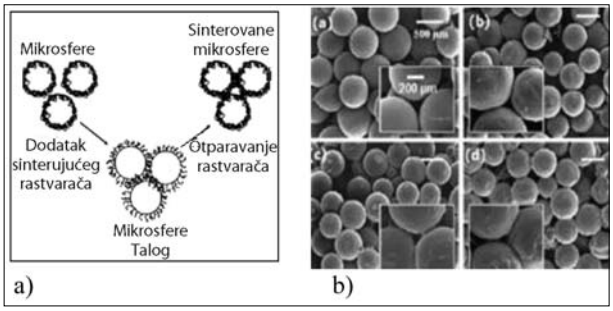

Figure 9. a) Schematic representation of sintering microspheres; b) scaffold morphology obtained by sintering microspheres [27]
Slika 9. a) Šematski prikaz sinterovanja mikrosfera; b) Morfologija skafolda dobijenog sinterovanjem mikrosfera [27]

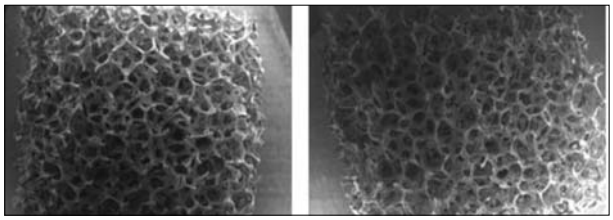

Figure 10. SEM a) Sintered compact on the surface obtained by dipping (18% paste) and b) combined method of dipping and spraying (18% paste, 6% of a sprayed suspension for 30 minutes) [30]
Slika 10. SEM a) Sinterovani kompakt na površini dobijen umakanjem (18% pasta) i b) kombinovanim postupkom umakanja i sprejovanja (18% pasta, 6% suspenzija za sprejovanje, 30 minuta vreme sprejovanja) [30]

Method of sintering microspheres

Non-toxic, neutral degradation products of amino acid esters (polyphosphazene) are ideal candidates for the use in orthopedic applications *in vivo.* Esters of amino acids (substituted poly polyphosphazenes) are used often due to good mechanical properties as scaffold is exposed to heavy loading. The most interesting are leucine, valine, and ethyl ester phenylaniline. Among these esters, ethyl ester phenyl amine shows the highest glass transition temperature (41.6°C). Therefore, it is used as a composite, in the mixture with hydroxyapatite whose particle size is in 100 nm order [27, 28, 29]. Composite first forms geometry in the form of microspheres, which is then sintered in a 3-dimensional porous scaffold, using so-called dynamic sintering of the solvent, as schematically presented in Figure 9.

Composite microspheres obtained by this method have pressure modulus of 46-81 MPa and mean pore size between 86 and 145 micrometers. 3D polyphosphazene-HA composite possesses good adhesion of osteoblasts, enables their optimal proliferation, and expression of alkaline phosphatase, what makes it suitable for use in the reconstruction of bone tissue.

Electrospray method

Electrospray method is typical replication method. It is used to obtain a scaffold without internal voids and limited number of micro cracks. In this method, ceramic suspension is pumped between the needle and the outer electrode while electric field breaks liquid into the little mono-disperse droplets [30]. Little particles of hydroxyapatite that are deposited this way provide good attachment for cells (Figure 10). It has been shown that foam coating with ceramics is better than foam obtained by the traditional method of soaking [30].

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Skafoldi u inženjerstvu koštanog tkiva

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KRATAK SADRŽAJ

Terapija i lečenje brojnih povreda i oboljenja koštanog tkiva je još uvek veliki izazov za hirurge, ali i za one istraživače koji se bave materijalima. Na polju inženjerstva koštanog tkiva danas se najčešće koriste matične ćelije. Međutim, napredak u dizajniranju biokompatibilnih materijala, a posebno biodegradibilnih poroznih struktura (skafolda) sve više dobija vrlo značajnu ulogu u lečenju obolelih koštanih tkiva. Specifično dizajnirani skafoldi sa definisanom poroznošću i strukturom pora koja je povoljna za naseljavanje ćelija osnovna je prednost ovih nosača. Skafoldi se najčešće koriste kao keramički nosači jer imaju izvanredne osobine vezane za biodegradaciju i jako izraženu bioaktivnost. Postupak izrade skafolda je vrlo važan jer se odgovarajućom tehnologijom mora obezbediti kontrola tečnosti i reproduktivnost izrade skafolda kroz standardizaciju procesa.

Cilj ovog rada je bio da se predstave različiti metodološki postupci izrade skafolda u inženjerstvu koštanog tkiva i ukaže na određene prednosti i nedostatke tih metoda.

Ključne reči: skafold; inženjerstvo koštanog tkiva; polimerne matrice; prototip skafolda

UVOD

Lečenje trauma i oboljenja koštanog tkiva, kao i rekonstrukcija koštanih defekata predstavljaju veliki izazov kako za ortopedske hirurge tako i za inženjere [1]. Najčešći pristupi na polju inženjerstva tkiva uključuju korišćenje matičnih ili diferenciranih ćelija odraslih, koje se zasejavaju u biodegradibilne skafolde i kultivišu u biorekatorima, pre implantiranja na defektno mesto. Napredak u dizajnu i funkcionalizaciji biokompatibilnih materijala, kao i napredak tehnika procesiranja omogućavaju razvoj biodegradibilnih poroznih struktura sa dobro dizajniranom arhitekturom, skafolda za tkivno inženjerstvo [1, 2].

Skafold treba da bude dizajniran na specifičan način, tako da poseduje odgovarajuću poroznost i biodegradibilnost i da ispunjava specifične zahteve vezane za individualne defekte, kao što je njihov oblik i veličina. Sa tehnološke tačke gledišta, veliki izazov kod skeletnog tkivnog inženjerstva je dizajniranje i izrada biodegradibilnog skafolda, koji ima definisanu poroznost i strukturu pora, koja je pogodna za naseljavanje ćelija i koja može da se održi u dovoljno dugom vremenu [3].

Kao skafoldi mogu da se koriste keramički nosači sa dobro prilagođenim osobinama degradacije i bioaktivnosti. Sa takvim mehaničkim osobinama oni služe kao potpora tokom stvaranja novog koštanog tkiva ili kao polimerni nosači zbog svoje hidrofilnosti pogoduju inkapsulaciji ćelija (slično prirodnom ekstracelularnom matriksu) [4]. Veoma su popularne procesne tehnike obrade konvencionalnih polimernih materijala, prilagođene i proširene na ugradnju neorganskih bioaktivnih faza u poroznu 3D polimernu mrežu.

Pored toga, ugradnja bioaktivnih molekula u biodegradibilne skafolde pospešuje regeneraciju kosti sa mnogim pozitivnim efektima [4, 5]. Veliki izazov u nauci o materijalima i tehnologiji na području tkivnog inženjerstva je kontrola tačnosti i reproduktivnosti izrade skafolda kroz standardizaciju procesa. Razne tehnike izrade skafolda, koje uključuju procesiranje različitih polimernih i kompozitnih materijala i razvoj različitih mikrostruktura danas su veoma aktuelne teme istraživanja. Ipak, još uvek i pored brojnih tehnika koje se primenjuju, svaka od njih poseduje određene nedostatke sa stanovišta kontrole poroznosti skafolda, veličine pora i distribucije, kao i prisustva ostataka toksičnih rastvarača u skafoldu.

Cilj ovog rada je bio da se predstave različiti metodološki postupci izrade skafolda u inženjerstvu koštanog tkiva.

METODE IZRADE SKAFOLDA

Proces izrade skafolda mora obezbediti visok nivo kontrole njihovih makro i mikrostrukturnih osobina. Zavisno od materijala od kog se skafold izrađuje i strategije inženjerstva tkiva razrađene su i različite metodologije i uslovi procesiranja skafolda radi optimizacije za unapred određenu namenu. U saglasnosti sa tim, procesne procedure, u svakom od konkretnih slučajeva, treba da budu odabrane tako da ne menjaju hemijske i biokompatibilne osobine materijala, da ne bi tako ograničili efekte njegove kliničke primene. Pored toga, skafold treba da da ima međusobno povezane pore i dovoljno visoku gustinu pora sa pravilnom morfologijom, veličinom i distribucijom i da uz to njegov kvalitet bude visokoreproduktivan.

Metoda polimerne matrice

Među brojnim metodama dizajniranja strukture skafolda jedna od najčešće korišćenih je metoda polimerne matrice (pene), koja se koristi kao model sistem pri oblikovanju keramičke strukture skafolda. Postupak se sastoji u nanošenju suspenzije praha keramičkog sistema preko matrice i, nakon sušenja i očvršćavanja suspenzije, spaljivanja polimerne pene pri čemu se dobija porozna keramička strukrura čija je poroznost dirigovana matricom koja je korišćena [6, 7].

U našim istraživanjima smo koristili matricu od poliuretanske pene kao model sistem za dobijanje i oblikovanje unutrašnje geometrije hidroksiapatitnog (HAP) skafolda [8, 9]. Nakon procesa izgaranja polimerne faze i sinterovanja keramičke faze na 800°C dobijena je struktura skafolda data na slici 1.

Kombinacija metode polimerne matrice i biomimične metode

Za funkcionalizaciju unutrašnjih zidova skafolda dobijenih metodom polimerne matrice u našim eksperimentima korišćene su različite vrste polimera, kao što su: poli(mlečna-ko-glikolna) kiselina (PLGA), hidroksietil celuloza, modifikovani skrob, alginat i dr. [10]. Naknadnim biomimičnim tretmanom u simulirajućem telesnom fluidu dobijene su visokoaktivne strukture skafolda vrlo specifičnog strukturnog dizajna (Slika 2).

Pored ovih polimera, i niz drugih polimera, kao što su: polikaprolakton, hidroksietilmetakrilat, hitozan, biopolimeri sa RGD motivima, fibroin svile, kao i različiti faktori rasta mogu biti uključeni u funkcionalizaciju unutrašnjih zidova skafolda. U našim eksperimentima korišćeni su i materijali koji pokazuju specifična fizička svojstva, kao što su superparamagnetni materijali – ferofluidi na bazi magnetita, maghemita, kobaltferita, gadolinijum-oksida itd. [10].

Metoda gas–pena uz dejstvo superkritičnog CO_2

Superkritični fluid date supstance dobija se kad se ta supstanca izloži dejstvu okoline u kojoj su pritisak i temperatura iznad kritičnog pritiska i kritične temperature za datu supstancu. Pod ovim uslovima tečne i gasne komponente materijala postaju identične i dalja kompresija ove fluidne faze neće imati za rezultat rastapanje [11, 12, 13].

CO_2 se najčešće koristi u svojstvu superkritičnog fluida, jer je slabo toksičan, nezapaljiv i jeftin, stabilan i prihvatljiv za okolinu. Koristi se kao kod tehnike pena–gas, gde se za procesiranje skafolda koristi CO_2 pod visokim pritiskom (kritična temperatura od 31°C i pritisak 73,8 bara). Da bi se pravilno procesirala mezoporozna struktura, polimerni disk se u polaznoj fazi procesa izlaže visokom pritisku, od 1 do 6 MPa na sobnoj temperaturi, kako bi se omogućilo zasićenje gasa u polimeru i formiranje jedinstvene faze između polimera i gasa. Rasvorljivost gasa u polimeru opada brzo pa sa smanjenjem pritiska CO_2 dolazi do nukleacije i rasta gasnih mehurova, koji dovode do formiranja pora većih od 500 μm. Makroporozne pene poli-L-mlečne kiseline (PLLA), poliglikolne kiseline (PGA) i poli-mlečne-ko-glikolne kiseline (PLGA) dobijaju se iz takvih polimernih diskova unutar kojih pri visokim pritiscima CO_2 nastaje odgovarajuća smeša polimer–gas. Potom sledi faza gde se gasni molekuli klasteruju, formirajući gasne nukleuse, koji potom difunduju, a unutar diskova polimera zaostaju makropore. Poroznost i struktura pora zavise od količine gasa rastvorenog u polimeru, brzine gasne nukleacije i brzine difuzije molekula gasa kroz polimer (Slika 3) [12].

Metoda fazne separacije

Tokom fazne separacije termodinamička nestabilnost se uspostavlja u homogenom višekomponentnom polimernom rastvoru, koji pod odgovarajućim uslovima pokazuje težnju da se razdvoji u više od jedne faze kako bi se smanjila njegova ukupna slobodna energija. Polimerni rastvor se razdvaja na dve faze – fazu koja je bogata polimerom i fazu koja je siromašna polimerom. Polimerom bogata faza očvršćava, a polimerom siromašna faza se uklanja ostavljajući za sobom visoko poroznu polimernu mrežu. Metoda fazne separacije je poznata pod nazivom termički indukovana fazna separacija (TIPS). Osnovni princip TIPS-a je da se pomeri polimerni rastvor iz svoga homogenog jednofaznog regiona u spinodalni region njegovog binarnog faznog sistema brzom promenom temperature rastvora. To se može izvesti brzim hlađenjem polimernog rastvora na njegovoj gornjoj kritičnoj temperaturi ili brzim zagrevanjem na donjoj kritičnoj temperaturi. Kada se polimerni rastvor pomeri u spinodalni region, on postaje nestabilan i razdvaja se spontano u polimerom bogati i rastvaračem bogati region [14, 15, 16].

Nakon termodinamičkog razdvajanja homogenog rastvora polimer–rastvarač u polimerom bogatu i polimerom siromašnu fazu, rastvor se najčešće izlaže drugom rastvaraču ili hlađenju ispod tačke njegove binodalne rastvorljivosti. Rastvarač se potom uklanja sumblimacionim sušenjem (freeze drying), ostavljajući polimer u formi pene. Morfologija tako dobijenog skafolda kontroliše se faznim prelazom koji se dešava tokom hlađenja, tj. tečno-tečne fazne separacije, dok unutar faze čvrsto–tečnost nastaje anizotropna pena specifične lisnate forme.

Uopšteno govoreći, kontrolu mikro i makrostrukture skafolda datog polimera dobijenu metodom fazne separacije moguće je kontrolisati optimalnim izborom procesnih parametara kao što su koncentracija polimera, temperatura i brzina separacije. Nekompletno uklanjanje rastvarača, posebno kod debljih konstrukata, rezultuje smanjenoj kompatibilnosti i mogućoj promeni ugrađenih aktivnih faktora. Iako su pomoću ove metode dobijene visoko međusobno povezane porozne matrice, ograničena veličina pora koja se dobija korišćenjem takve metode uslovljava ozbiljne probleme vezane za uspostavljanje kontrolisane makro i mikrostrukture skafolda (Slika 4) [17].

Metoda sublimacionog sušenja

Sublimaciono sušenje (freeze drying) uključuje uklanjanje rastvarača (najčešće vode) prvo sumblimacijom zamrznutih uzoraka, a potom desorpcijom nezaleđenog apsorbovanog rastvarača pod smanjenim pritiskom [18]. U stadijumu zamrzavanja polimerni rastvor ili disperzija se hlade ispod temperature na kojoj rastvarač prelazi u čvrsto stanje, usled čega dolazi do formiranja kristala leda i razmeštanja polimernih molekula u prostore između kristala leda. U drugoj fazi rastvarač se uklanja primenom pritiska koji je niži od ravnotežnog pritiska pare zamrznutog rastvarača. U međuvremenu nezaleđena apsorbovana voda u sušenom sloju se desorbuje. Kada je zamrznuti rastvarač kompletno sumblimovan, proces se nastavlja dalje uz blago zagrevanje uzorka sve dok ne bude kompletno osušen. Sumblimacija kristala leda uzrokuje formiranje visoko porozne sunđeraste strukture skafolda.

Konačna struktura pora zavisi od procesnih uslova tokom izrade skafolda kao što su pH, brzina zamrzavanja i parcijalni pritisak. Brzo, nekontrolisano hlađenje vodi neuniformnoj nukleaciji i rastu kristala leda, a samim tim i različitim morfološkim heterogenostima unutar skafolda, koje su uslovljene prostornim i vremenskim promenljivima tokom transfera toplote kroz sistem. Pokazano je da je moguće dobiti znatno uniformnije porozne skafolde pri konstantnoj brzini hlađenja tokom procesa zamrzavanja. Tehnikom sušenja zamrzavanjem dobijaju se visoko porozne i međusobno povezane strukture sa malim veličinama pora [18].

Metoda sumblimacionog sušenja emulzije, kao modifikovana metoda fazne separacije, koristi se u proizvodnji alifatičkih

poliesterskih skafolda. Skafoldi dobijeni ovom metodom imaju poroznost iznad 90%, dok je srednja veličina pora između 15 i 35 μm, sa najvećim porama većim i od 200 μm (Slika 5). Poroznost skafolda prvenstveno zavisi od zapreminske frakcije dispergovane faze, koncentracije polimera i njegove molekulske mase. Tehnika sumblimacionog sušenja emulzije koristi se i za ugradnju proteina u polimerni skafold. Sumblimaciono sušenje vodenih rastvora biopolimera, kao što je kolagen, koristi se i kod dobijanja dobro definisanih poroznih matrica (dobro definisane veličine i orijentacije pora) [19–22].

Metoda livenja / luženja porogena

Livenje rastvarača omogućava pripremu poroznih struktura sa normalnom poroznošću, ali sa ograničenom debljinom [17, 23]. Metoda livenja / luženja porogena uključuje u sebe livenje smeše polimera i porogena u kalup, sušenje smeše, potom luženje porogena sa vodom, da bi se napokon dobile pore. Kao porogeni najčešće se koriste soli. Strukturu pora je najlakše kontrolisati udelom porogena. Visoko porozne PLLA membrane kontrolisane poroznosti, kao i odnosa površina : zapremina, te kristaliničnosti pripremaju se livenjem disperzije kristalne soli u organski rastvor PLLA, da bi nakon toga so bila izlužena vodom. NaCl, Na-tartarat i Na-citrat raznih veličina čestica koriste se kao materijali za stvaranje pora luženjem. Osobine dobijenih pena zavise isključivo od frakcije soli i veličine njenih čestica, a ne zavise od vrste izabranog polimernog rastvarača. Zbog toga se ona koristi se za pripremu poroznih trodimenzionalnih skafolda pogodnih za inženjerstvo tkiva (Slika 6). Ovom tehnikom moguće je dobiti visoko porozne strukture skafolda sa poroznošću iznad 93% i srednjim prečnikom pora do 500 μm.

Glavna prednost ove metode je laka izrada skafolda, bez neophodnosti za nekim skupim specijalnim uređajima. Takva tehnika dopušta da se realizuju pore u širokom opsegu veličine i da se poroznost i veličina pora mogu kontrolisati. Nedostatak ove metode vezan je prvenstveno za neophodnost uklanjanja rastvornih čestica unutar polimera. Metoda livenja kombinovana sa luženjem može da se iskoristi za dobijanje tankih membrana ili 3D uzoraka male debljine (do 2 mm). Jedan od bitnih nedostataka metode je aglomeracija čestica porogena tokom procesa dobijanja skafolda, zbog čega pore skafolda imaju neujednačenu raspodelu. Drugi nedostatak je vezan za korišćenje organskih rastvarača. Oni moraju biti uklonjeni da bi se izbegla moguća oštećenja zasejanih ćelija i proteina ili drugih aktivnih molekula koji su u njega ugrađeni [23].

Kombinovana metoda sublimaciong sušenja i livenja porogena

Hibridni sunđer sintetičkog polimera, kolagena i hidroksiapatita pogodan je za deponovanje hidroksiapatitnih čestica na površini istog, jer oponaša strukturu kolagena i hidroksiapatita u prirodnoj kosti (Slika 7) [24]. Depozicija hidroksiapatita odvija se kroz proces alternirane imerzije PLGA-kolagenskog sunđera u vodenom rastvoru $CaCl_2$ i Na_2HPO_4. PLGA-kolagenski sunđer prvo se uranja u rastvor $CaCl_2$ pod vakuumom, na 37°C tokom 12 h, da bi se prostor unutar sunđera što bolje natopio rastvorom. Posle izvlačenja iz $CaCl_2$ rastvora sunđer se centrifugira da bi se odstranio višak Na_2HPO_4 rastvora. Ovaj proces alternirane imerzije i centrifugiranja definiše jedan ciklus deponovanja hidroksipatita unutar sunđeraste strukture PLGA-kolagen skafolda. Proces deponovanja najčešće se izvodi u nizu ponovljenih ciklusa.

Metoda elektroforetske depozicije

Metoda elektroforetske depozicije (EPD) jedna je od najefikasnijih tehnika asembliranja hidroksiapatitnih finih čestica. Vrlo je jednostavna i njome je moguće dobiti vrlo kompleksne forme čestica. Aparatura za izvođenje EPD je vrlo jeftina. U prvo vreme ova metoda je pretežno korišćena za deponovanje hidroksiapatitnih prevlaka na metal ili keramiku, dok je deponovanje (dovoljno debelih slojeva) apatita odgovarajuće poroznosti još uvek veliki izazov [25]. Zbog toga se velika pažnja posvećuje dobijanju hidroksiapatitnih stabilnih disperzija, sa visokim vrednostima zeta potencijala i odgovarajućom koncentracijom, koja bi u pogodno odabranim uslovima deponovanja mogla dati odgovarajuću strukturu skafolda (Slika 8) [26].

Metoda sinterovanje mikrosfera

Netoksični, neutralni degradacioni produkti estara amino kiselina (polifosfazena) idealni su kandidati za ortopedske primene *in vivo*. Estri aminokiselina (supstituisani polifosfofazeni) koriste se najčešće zbog svojih dobrih mehaničkih osobina kao skafoldi za tkiva koja trpe opterećenje. Među njima najzanimljiviji su leucin, valin i etil-estar fenilanilina. Među svim ovim estrima, etil-estar fenilamina pokazuje najvišu temperaturu staklastog prelaza (41,6°C). Zbog toga se koristi kao kompozit u smeši sa hidroksiapatitom, čija je veličina čestica reda 100 nm [27, 28, 29]. Kompozit prvo formira geometriju u formi mikrosfera, koje se potom sinteruju u 3-dimenzionalni porozni skafold, pomoću tzv. dinamičkog sinterovanja rastvarača, što je je šematski predstavljeno na slici 9.

Mikrosfere kompozita dobijene ovom metodom imaju modul pritiska 46–81 MPa i srednju veličinu pora između 86 i 145 μm. 3D polifosfazen-HA kompozit poseduje izuzetno dobru adhezivnost osteoblasta i omogućuje njihovu optimalnu proliferaciju i ekspresiju alkalne fosfataze, zbog čega je veoma pogodan za primenu u rekonstrukciji koštanih tkiva.

Elektrosprej metoda

Elektrosprej metoda je tipična replikaciona metoda. Koristi se da bi se dobio skafold bez unutrašnjih šupljina i da bi se sasvim limitirao broj mikropukotina. Kod ove metode keramička suspenzija se pumpa između igle i spoljašnje elektrode, pri čemu se zbog dejstva odgovarajućeg električnog polja tečnost kida u fine monodisperzne kapljice [30].

Fine čestice hidroksiapatita koje se deponuju na ovaj način obezbeđuju dobro kačenje ćelija (Slika 10). Pokazalo se da su dobijene prevlake pene sa keramikom kvalitetnije nego pene dobijene tradicionalnom metodom umakanja [30].

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CASE REPORT
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Endodontic treatment of lower molar in a patient with paraesthesia of inferior alveolar nerve – A Case Report

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SUMMARY

Root canal (endodontic) treatment is demanding and complex procedure. A variety of difficulties can occur in different phases of endodontic procedure. Complex anatomorphological tooth structure, curved canals, close proximity of lower molars and premolars to inferior alveolar nerve make endodontic treatment even more challenging. During endodontic treatment, an inferior alveolar nerve may become traumatized and symptoms may vary from mild neurosensory dysfunction to a complete loss of sensation in the innervation area of damaged nerve.

The aim of this paper is to present a clinical case of endodontic treatment of lower second molar with C-shaped root canal in a patient with paraesthesia of inferior alveolar nerve due to endodontic origin.

Keywords: paraesthesia; C-shaped canal; Guttaflow paste

INTRODUCTION

Successful endodontic treatment implies complete removal of microorganisms and their products as well as organic and inorganic contents from root canal space. Removal of pathologically changed pulp and contaminated dentin, instrumentation and irrigation of root canal and finally, adequate tridimensional obturation of endodontic space are basic principles of endodontic treatment. Modern concepts of biological endodontic treatment include instrumentation, irrigation, medication and obturation only within the root tooth canal space, without any contact with periapical and other surrounding tissues [1]. Inadequate application of anesthetic solution, irrigation and activation of hand and mechanical instruments as well as anatomical proximity of these structures may cause perforation of mandibular canal. This can lead to extrusion of irrigation solution, obturation sealers, numbness of mandibular nerve or its branches and contamination of mandibular canal with microorganisms from infected root canal [2]. Neurological symptoms and disorders may appear as intense pain, hyperesthesia, hypoesthesia, anesthesia, dysesthesia and paraesthesia. Symptoms may differ from mild neurological dysfunction to a complete loss of sensation in the innervation area of damaged nerve [2, 3]. Paraesthesia may occur as a consequence of local endodontic or even systemic factors. Local endodontic factors may be chemical (local anaesthetics, irrigation solutions, intersession medicaments), mechanical (over instrumentation) thermal (heated gutta-percha) and pressure factors on certain structures [2, 3]. Other local factors may be trauma (jaw fractions, contusions etc.), local infections (osteomyelitis, peri-implant infections), compressive lesions (benign and malignant neoplasms and cysts), tooth impactions, iatrogenic lesions upon tooth extraction and implantation techniques (in most cases, there is a swelling which causes compression and leads to the loss of sensitivity) [4, 5]. Systemic factors that cause paraesthesia might be multiple sclerosis, sarcoid, metastatic changes, viral and bacterial infections, leukemia, lymphoma, diabetes mellitus [5, 6].

Consequences of peripheral nerve injury and prognosis depend on proper and accurate diagnosis. **Neuropraxia**, or irritation is a first degree injury and it represents only physiological block of conduction, without interruption of axon continuity. The cause of conduction interruption is probably of biochemical origin on myelin sheath level. **Axonotmesis**, second degree injury, represents an injury with the loss of axon continuity and myelin sheath. **Third degree injury** is characterized by damaged endoneurium with scarring that supports axon regeneration. In the case of **fourth degree injury**, nerve continuity is preserved even though it is maintained by scarring tissue. **Neurotmesis, fifth degree**, is an injury with a complete disruption of nerve continuity [7]. It is important to mention that early stages of neurotmesis and axonotmesis are difficult to differ. In such case, only clinical examination may show which of the two injuries will spontaneously heal and which injury requires surgical intervention. Complications of endodontic treatment greatly depend on the complexity of canal configuration. In certain situations, lower second molar may have merged roots with C-shaped canal which usually implies deeper localization of pulp space and atypical anatomical openings [8].

The aim of this paper was to present the clinical case of endodontic treatment of lower second molar with C-shaped root canal in a patient with paraesthesia of inferior alveolar nerve due to endodontic origin.

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CASE REPORT

A 38-year old female patient was referred to the Department of Restorative Dentistry and Endodontics, School of Dentistry, University of Belgrade in February 2012. The patient suffered from intense pain in the right lower lip and mental region. During the tooth drilling (#47), the pulp was exposed and upon the application of anaesthesia (Ubistesin tm forte 4%, 3M Espe, Germany), her dentist initiated endodontic treatment. After initial exploring of the canal, symptoms of sensitivity of inferior alveolar nerve in the right chin area and right half of the lip appeared. The patient was then referred to the Department of Restorative Dentistry and Endodontics, School of Dentistry, University of Belgrade.

Upon the patient's arrival at the clinic, dental examination showed the presence of temporary crown on the tooth 47 and excellent oral hygiene. At that moment, the patient suffered from severe pain in the right part of her lower lip and mandible from the tooth 47 to incisal region. The pain was intensified in response to thermal stimuli (especially to cold) and percussion test. The patient was taking 3x1 tablet of 400 mg Ibubrufen per os in order to ease the pain. Periapical radiography showed possible close contact between neurovascular bundle of the tooth 47 and inferior alveolar nerve, and possible irritation of this nerve caused by endodontic treatment. Temporary crown, filling and intersession medicament were removed. The working length was determined using apex locator followed by carefully performed endodontic treatment of C-shaped canal using manual endodontic instruments. Preparation of the apical third was performed to the size of ISO # 25. It was intermittently irrigated with 1% solution of NaOCl, heated up to the body temperature. During the procedure, the patient suffered from pain and discomfort. When the treatment was finished a medicament dressing in the form of pad soaked in chlor-phenol camphor solution was administered to the patient. Painkillers were also prescribed, if necessary. After a week, intense pain was gone and the patient suffered only from the symptoms of right inferior alveolar nerve paraesthesia (tingling sensation, mild numbness and insensitivity of lower right lip and mental region). Canal obturation was performed with Guttaflow paste and adequate gutta-percha points with improved retention characteristics (Coltene, Whaledent AG, Switzerland). Upon obturation, the patient was referred to 3D orthopantomography of appropriate region that showed good canal obturation in all dimensions. The patient kept doing check-ups in the following period. Symptoms slowly eased and relieved and finally disappeared 8 months after their first occurrence.

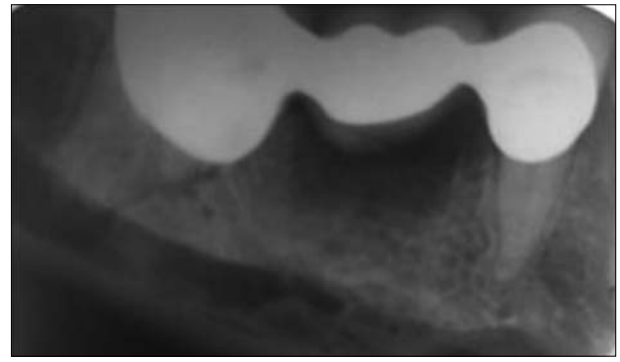

Figure 1. Preoperative radiography of the right side of the molar region of mandible
Slika 1. Preoperativna radiografija molarne regije desne strane donje vilice

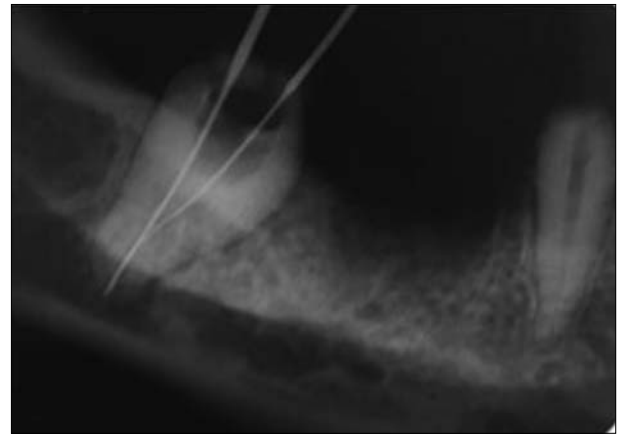

Figure 2. Periapical radiography with hand files showing close contact between neurovascular bundle of tooth 47 and mandibular nerve
Slika 2. Radiografija sa ručnim endodontskim instrumentima koja pokazuje blizak odnos neurovaskularnog snopa zuba 47 i mandibularnog kanala

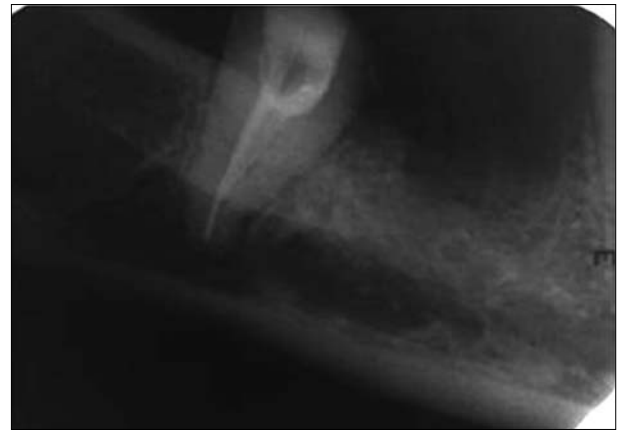

Figure 3. Postoperative radiography of tooth 47 shows hermetic canal opturation
Slika 3. Postoperativna radiografija zuba 47 koja pokazuje dobru opturaciju

DISCUSSION

Treatment of the tooth that caused neurosensory dysfunction depends on the type and severity of injury. The most common injuries of inferior alveolar nerve occur during surgical interventions, especially extraction of lower wisdom teeth and poorly planned implant placement [9, 10, 11]. Many authors reported occurrence of paraesthesia caused by endodontic treatment of root canal [5, 12–15]. Almost all of the materials used in endodontic treatment are neurotoxic at some level and can cause various inflammatory reactions that can lead to cell damage, ulceration, hemolysis and necrosis in contact with periapical tissues [4, 5, 6, 16]. Irrigation solutions (NaOCl and EDTA) can

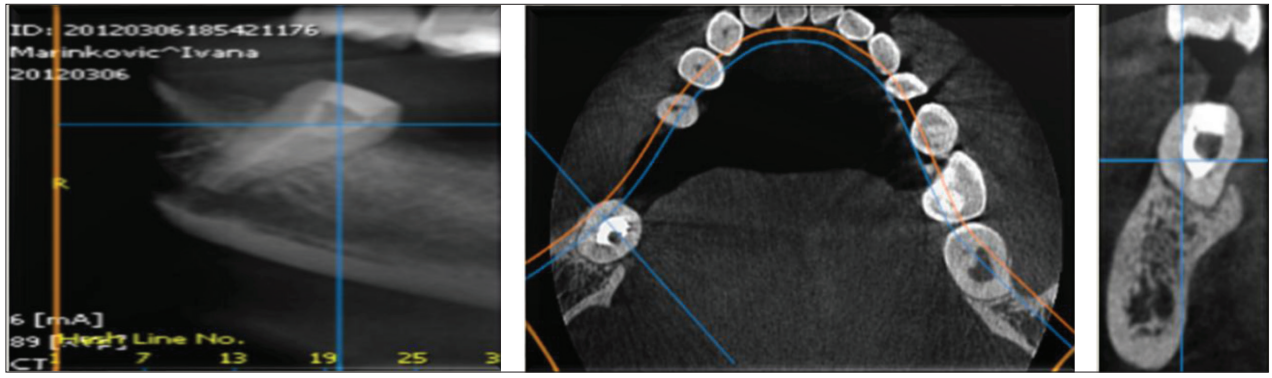

Figure 4. 3D OTP shows adequate tridimensional opturation of endodontic space of tooth 47 with characteristic C-shape
Slika 4. 3D OTP na kojoj se vidi trodimenzionalna opturacija zuba 47 sa karakterističnim c-oblikom komore pulpe

reach mandibular canal and cause chemical nerve damage [17, 18]. NaOCl is an endodontic irrigant of choice since it has excellent antimicrobial effect and ability to dissolve tissue. It is cytotoxic even in low concentrations and causes protein denaturation, releases chlorine gas and draws fluids osmotically into periapical space [12]. Ethanol, irrigant as well, may cause oversensitivity of apical tissue since it has very strong dehydration potential [12]. Hulsmann and Hahn studied the application of different concentration of NaOCl (3% and 5,25%) for irrigation during endodontic treatment of mandibular premolars and molars and found in some cases paraesthesia combined with sensitivity dysfunction in lower lip [19]. They determined that toxicity of this irrigant caused chemical damage to neurovascular bundle [19]. An adequate irrigation technique with the application of small pressure and use of special needles with lateral perforations will decrease the possibility for periapical irritation. Damage of inferior alveolar nerve may also be caused by inadequate instrumentation of root canals of lower molars and premolars, mechanical irritation of apex or even separated instrument in mandibular canal [11, 16, 20]. $Ca(OH)_2$ as an intersessional medicament and a strong base that may cause nerve damage even though such cases are rarely described in literature. [2] The literature most often describes compression syndrome caused by extrusion of endodontic sealers beyond apex [12, 14]. Experimental studies confirmed the role of eugenol and paraformaldehyde in neurotoxic reactions [1, 4, 12]. Kozam and Trowbridge reported in 1977 that eugenol has neurotoxic effect that can cause paraesthesia of inferior alveolar nerve. Eugenol causes chemical destruction of axon by protein coagulation [21, 22]. Canal sealers, AH 26 and AH 26 plus, also have cytotoxic potential [23]. AH 26, a synthetic resin, together with formaldehyde causes tissue necrosis and inflammation [12]. Ehrmann was the first to report paraesthesia case caused by overfilling with N2 paste [11]. Gutta-percha is the material of choice for root canal obturation. It is inert material but it may cause paraesthesia if mechanically irritates the nerve [24]. Vertical condensation technique and other obturation techniques that require heated gutta-percha may also cause nerve damage [24]. Block anesthesia of inferior alveolar nerve may as well cause paraesthesia. Injury is mostly provoked by nerve damage with injection needle, compression effect or even combination of the two [14].

It is very important to understand variations of anatomorpholocigal characteristics of certain teeth groups since deviations from the average morphological characteristics are most common reasons for failure of endodontic treatment. One of the most interesting anatomical variations is C-shaped root and canal system. The shape and number of roots are defined by Hertwig's epithelial sheath that bends in horizontal dimension below cementoemanel junction and fuses in the center leaving the openings of the canals. C-shaped root may be formed due to constant deposition of cement over time [8]. Studies on lower second molar have shown high incidence of C-shaped roots and canals (10-31,5%) in Japanese, Chinese, Hong Kong Chinese, Lebanese and Thai populations [8]. Clinical recognition of C-shaped canals is based on definition of observable criteria (anatomy of the floor of pulp chamber and persistent haemorrhagia or pain when separate canal orifices are found). Pulp chamber in teeth with C-shaped canals may be large in occlusoapical dimension with low bifurcation. Sometimes, the canal can be calcified thus masking its C-shape configuration.

Nerve recovery after its damage depends on the severity of damage and rapidity of cause removal. Often, after the removal of cause, symptoms of paraesthesia continue to exist since the injury was not just mechanical but chemical as well. Endodontic material can spread to periapex in four different ways (through the nerve bundle, by drainage through lymphatic vessels, periapical capillary system and diffusion between the bones and mucosal membrane toward soft tissues) [2, 25]. The anatomy of lower jaw favors diffusion of endodontic material, especially in the posterior area of lower jaw due to trabecular properties of cancellous bone that facilitates diffusion of different materials into the surrounding tissues. Special attention should be paid to the distance between anatomical openings of mandibular molars and mandibular canal. According to one study, this distance varies from 1 and 4 mm in the case of first lower molar, while it is less than 1 mm with second and third molar [25]. Cone Beam computed tomography- modern diagnostic method could help with therapy planning and prevention of paraesthesia occurrence as a complication of endodontic therapy [1].

Adequate endodontic treatment of the tooth that caused neurosensory dysfunction is important with additional application of cold packs, analgesics, antibiotic therapy,

nonsteroidal anti-inflammatory drugs, synthetic corticosteroids (dexamethasone), proteolytic enzymes (which disintegrate coagulum), vitamin B complex, vitamin C (it has antioxidative action which reduces ischemic effects), and adenosine thriphosphate that regenerates tissues for restoring nerve function [15, 16]. It is important to diagnose paraesthesia as soon as possible and remove potential causes of this dysfunction, preferably within 48 hours [12, 27]. Surgical treatment includes extraction of causal tooth, apicoectomy and surgical removal of foreign body [27].

Based on the patient history and clinical findings, nerve damage in our study was classified as second-degree damage by Seddon: recognized axonotmesis manifested by paralysis of motor and sensitive nerve function. Due to the close connection between anatomical opening and mandibular canal, endodontic hand instrument most probably injured the axon and myelin sheath while NaOCl caused chemical irritation. Healing occurred spontaneously after adequate endodontic treatment and disinfection of root canal that was of utmost importance. Healing conditions were improved by low concentration of NaOCl used as an irrigant and the use of Guttaflow paste for final obturation.

The key to successful endodontic treatment of complex canal configurations is to know dental anatomy and apply adequate instrumentation and obturation techniques. This case report shows, apart from properly conducted endodontic treatment, positive features of guttaflow paste that is the sealer of choice in cases of close relation between the tooth apex and mandibular canal.

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Endodontska terapija donjeg molara kod pacijenta sa parestezijom donjeg alveolarnog nerva – prikaz slučaja

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KRATAK SADRŽAJ

Endodontska terapija kanala korena zuba je težak i kompleksan zahvat, pa su zato česti i problemi u različitim fazama endodontskog postupka. Komplikovana anatomomorfološka struktura zuba, povijenost kanala, blizak odnos donjih molara i premolara i alveolarnog nerva dodatno komplikuje endodontsku proceduru. Tokom realizacije endodontske terapije može doći do traume donjeg alveolarnog nerva, a simptomi oštećenja alveolarnog nerva mogu da variraju od blage neurosenzorne disfunkcije do potpunog gubitka senzacija u predelu inervacionog područja oštećenog nerva.

Cilj ovog rada je bio da se na jednom slučaju iz kliničke prakse predstavi endodontski postupak lečenja kanala korena donjeg drugog molara, karakterističnog c-oblika, kod pacijenta sa parestezijom donjeg alveolarnog nerva endodontskog porekla.

Ključne reči: parestezija; c-oblik kanala; guttaflow pasta

UVOD

Uspešna endodontska terapija podrazumeva obradu kanalnog sistema, sa potpunim uklanjanjem mikroorganizama i njihovih produkata, kao i organskih i neorganskih komponenata iz prostora kanala korena. Uklanjanje patološki promenjene pulpe i kontaminiranog dentina, instrumentacija i irigacija kanala korena, i na kraju adekvatna trodimenzionalna opturacija endodontskog prostora predstavljaju osnovne principe endodontskog zahvata. Savremeni koncepti biološke endodontske terapije podrazumevaju instrumentaciju, irigaciju, medikaciju i opturaciju isključivo u okviru prostora kanala korena, bez kontakta sa periapeksnim i drugim okolnim tkivima [1]. Neadekvatna aplikacija anestetičkog rastvora, irigacija i aktivacija ručnih i mašinskih instrumenata, ali i anatomska bliskost ovih struktura, mogu usloviti perforaciju mandibularnog kanala. To može dovesti do potiskivanja irigacionih rastvora, opturacionih silera, lediranja mandibularnog nerva ili njegovih grana, ali i kontaminacije mandibularnog kanala prodorom mikroorganizama iz inficiranog kanala korena [2]. Neurološki simptomi i smetnje koje se javljaju mogu se ispoljiti u vidu intenzivnog bola, hiperestezije, hipoestezije, anestezije, dizestezije i parestezije. Simptomi mogu da variraju od blage neurosenzorne disfunkcije do potpunog gubitka senzacija u predelu inervacionog područja oštećenog nerva [2, 3]. Parestezija može nastati kao posledica lokalnih, endodontskih, ili pak sistemskih faktora. Lokalni endodontski faktori mogu biti hemijski (lokalni anestetici, rastvori za irigaciju, interseansni medikamenti), mehanički (instrumentacija preko vrha korena), termički (zagrejana gutaperka) i faktori koji svojim pritiskom deluju na određene strukture [2, 3]. Ostali lokalni faktori mogu biti traume (prelomi vilica, kontuzije itd.), lokalne infekcije (osteomijelitis, periimplantne infekcije), kompresivne lezije (benigne i maligne neoplazije i ciste), impakcije zuba, jatrogene lezije nakon ekstrakcije zuba i implantoloških tehnika (najčešće se radi o otoku koji je vršio kompresiju i uslovio gubitak senzibiliteta) [4, 5]. Sistemske faktore nastanka parestezije mogu usloviti multipla skleroza, sarkoidoza, metastatske promene, virusne i bakterijske infekcije, leukemija, limfomi, dijabetes melitus [5, 6].

Posledice povreda perifernih nerava, kao i prognoza njihovog izlečenja uslovljene su pravilnom, tačnom dijagnozom. **Neuropraksija,** ili iritacija, povreda prvog stepena, predstavlja samo fiziološki blok sprovođenja, bez prekida kontinuiteta aksona. Uzrok prekida sprovođenja je verovatno biohemijske prirode na nivou mijelinskog omotača. **Aksonotmeza,** povreda drugog stepena, predstavlja tip povrede sa prekidom kontinuiteta aksona i mijelinskog omotača. Povredu **trećeg stepena** karakteriše oštećenje endoneurijuma sa ožiljnim promenama kroz koje akson regeneriše. Kod povrede **četvrtog stepena** kontinuitet nerva je fizički očuvan, ali se održava samo ožiljnim tkivom. **Neurotmeza, peti stepen,** povreda je sa potpunim prekidom kontinuiteta nerva [7]. Važno je napomenuti da se neurotmeza i aksonotmeza u ranim fazama ne mogu razlikovati. U tom slučaju jedino klinička ispitivanja mogu ukazati koja će se od ovih povreda spontano oporaviti, a koja povreda zahteva hirurišku intervenciju. Komplikacije endodontske terapije povećava komplikovanost kanalne konfiguracije. U pojedinim slučajevima donji drugi molar može imati spojene korenove, pri čemu se onda javlja c-oblik kanala, koji najčešće uslovljava dublju lokalizaciju komore pulpe i atipične anatomske otvore [8].

Cilj ovog rada je da se na jednom slučaju iz kliničke prakse predstavi postupak terapije kanala korena drugog donjeg molara, karakterističnog c-oblika, kod pacijenta sa parestezijom donjeg alveolarnog nerva koja je uzrokovana endodontskom terapijom.

PRIKAZ SLUČAJA

Pacijentkinja starosti 38 godina javila se na Kliniku za bolesti zuba i endodonciju Stomatološkog fakulteta Univerziteta u Beogradu u februaru 2012. godine, po uputu svog stomatologa. Pacijentkinja je imala simptome intenzivnog bola u predelu desne strane donje usne i mentalnog predela. Tokom brušenja zuba (47) eksponirana je pulpa i stomatolog je posle aplikacije anestezije (Ubistesin tm forte 4%, 3M ESPE, Germany) započeo endodontski tretman. Posle ispitivanja inicijalne prohodnosti kanala javili su se simptomi nadraženosti donjeg alveolarnog nerva u predelu brade desne strane i odgovarajuće polovine usne. Pacijentkinja je upućena na Kliniku za bolesti zuba i endodonciju Stomatološkog fakulteta u Beogradu.

Po prijemu pacijenta na kliniku, stomatološkim pregledom su utvrđeni prisustvo privremene krunice na zubu 47 i odlična oralna higijena. Pacijentkinja je u tom trenutku imala intenzivan

bol u predelu desne strane donje usne i predela mandibule od zuba 47 pa do incizalnog dela. Bol se intenzivirao na termičke nadražaje (posebno na hladno), kao i na perkusiju zuba u toj regiji. Pacijentkinja je *per os* unosila 3 × 1 tabletu ibubrufena od 400 mg da bi smanjila intenzitet bola. Po urađenoj dentoalveolarnoj rendgengrafiji ovog predela posumnjalo se na mogućnost bliskog kontakta neuraovaskularnog snopa zuba 47 i mandibularnog nerva, te na moguću provokaciju ovog nerva izazvanog endodontskim tretmanom. Uklonjena je privremena krunica, privremeni ispun i uložak dezificijensa. Radna dužina je utvrđena elektrodontometrijskim postupkom i potom je usledila veoma pažljiva endodontska obrada kanala c oblika ručnim endodontskim instrumentima. Preparacija apeskne trećine kanala korena je urađena do veličine ISO #25, uz stalnu irigaciju 1% rastvorom NaOCl, zagrejanog na temperaturu tela. Sve vreme rada pacijentkinja je osećala bol i neprijatnost. Posle završene obrade kanala korena I aplikacije medikamentnog uloška u vidu vatice natopljene u rastvor hlorfenolkamfora pacijentu su ordinirani analgetici po potrebi. Nakon nedelju dana intenzivan bol je nestao i pacijent je imao samo simptome parestezije donjeg desnog alveolarnog nerva, u vidu peckanja, blagog trnjenja i neosetljivosti u predelu desne strane donje usne i mentalnog predela. Tada je izvršena opturacija kanala guttaflow fast pastom i ogovarajućim gutaperka kočićima sa poboljšanim retencionim karakteristikama (Coltene, Whaledent AG, Switzerland). Nakon opturacije pacijentkinja je upućena na 3D ortopantomografsko snimanje odgovarajućeg predela, gde je potvrđena dobra opturacija kanala u svim dimenzijama. Pacijentkinja je u narednom vremenskom periodu dolazila na kontrolne preglede. Simptomi su se polako smanjivali i smiravali da bi svi nestali tačno osam meseci od njihove pojave.

DISKUSIJA

Terapija zuba uzročnika neurosenzitivnih disfunkcija zavisi od vrste i težine povrede. Iako su najčešće povrede donjeg alveolarnog nerva usled hirurških intervencija, posebno donjih umnjaka, i loše isplanirane implantološke intervencije [9, 10, 11], mnogi autori iznose podatke o parestezijama koje su uzrokovane endodontskom terapijom kanala korena [5, 12–15]. Skoro svi materijali koji se koriste u endodonciji imaju neurotoksični efekat u određenom stepenu i mogu uzrokovati različite inflamatorne reakcije, koje dovode do oštećenja ćelija, ulceracije, hemolize i nekroze u kontaktu sa periapeksnim tkivima [4, 5, 6, 16]. Rastvori za irigaciju (NaoCl i EDTA) mogu dospeti do mandibularnog kanala i izazvati hemijsko oštećenje nerva [17, 18]. NaOCl je irigans izbora zbog svog odličnog antimikrobnog i organolitičnog efekta. On je citotoksičan i u niskim koncentracijama i izaziva denaturaciju proteina, oslobađanje gasa hlora i osmotsko izbacivanje fluida u periapsne prostore [12]. Etanol kao irigans može uzrokovati prenadraženost apeksnog tkiva zbog svog jakog dehidratacionog potencijala [12]. Hulsmann i Hahn su pratili primenu različite koncentracije NaOCl (3% i 5,25%) tokom irigacije mandibularnih premolara i molara i uočili pojavu parestezije kombinovane sa disfunkcijom senzibiliteta na donjoj usni [19]. Utvrdili su da toksičnost ovog irigansa izaziva hemijska oštećenja neurovaskularnog snopa [19]. Svakako, adekvatna tehnika irigacije sa primenom malog pritiska i korišćenjem specijalnih igala sa lateralnim perforacijama smanjiće mogućnost iritacije periapeksa. Oštećenje donjeg alveolarnog nerva može nastati i neadekvatnom instrumentacijom kanala korena donjih molara i premolara, mehaničkom iritacijom apeksa ili pak zalamanjem instrumenta u mandibularnom kanalu [11, 16, 20]. $Ca(OH)_2$ kao interseansni medikament, kao jaka baza može izazvati oštećenje nerva, iako je to jako retko prikazano u literaturi [2]. U literaturi je najčešće opisana pojava kompresivnog sindroma nastalog prebacivanjem endodontskih silera za opturacije [12, 14]. U eksperimentalnim studijama je potvrđena uloga eugenola i paraformaldehida kod neurotoksičnih reakcija [1, 4, 12]. Kozam i Trowdridge su još 1977. godine dokazali neurotoksični efekat eugenola kao uzročnika parestezija donjeg alveolarnog nerva. Koagulacijom proteina eugenol uzrokuje hemijsku destrukciju aksona [21, 22]. Kanalni sileri, AH 26 i AH 26 plus, takođe mogu imati citotoksični potencijal [23]. AH26, kao sintetska smola, zahvaljujući formaldehidu, izaziva nekrozu tkiva i inflamaciju [12]. Ehrmann je prikazao slučaj parestezije uzrokovane prepunjavanjem N2 pastom [11]. Gutaperka, kao materijal izbora u endodontskom tretmanu, kao inertan materijal, uzrokuje paresteziju [24] samo ukoliko mehanički nadraži nerv. Najčešće tehnika vertikalne kondenzacije i druge opturacione tehnike koje zahtevaju zagrejani gutaperka poen mogu usloviti oštećenje nerva [24].

Blok anestezija donjeg alveolarnog nerva takođe može biti uzrok nastanka njegove parestezije. Povreda najčešće nastaje hemoragijom zbog lediranja nerva injekcionom iglom, kompresivnim efektom ili pak kombinacijom oba [14].

Veoma je važno poznavati varijacije anatomomorfoloških karakteristika određenih grupa zuba, jer su odstupanja od prosečnih morfoloških karakteristika jedan od češćih uzroka neuspeha endodontskog tretmana. Jedna od najzanimljivijih anatomskih varijacija je sigurno c-oblik korena i kanalnog sistema. Oblik i broj korenova je određen Hertvigovom epitelijalnom membranom koja se savija u horizontalnoj dimenziji ispod cementnogleđne granice i spajajući se u centru ostavlja otvore kanala. c-oblik korena može biti formiran zbog konstantne depozicije cementa tokom vremena [8]. Studije na donjem drugom molaru pokazuju veliku incidenciju c-oblika korena i kanala (10–31,5%) kod Japanaca, Kineza, Hongkong Kineza, Libanske i Thai populacije [8]. Kliničko prepoznavanje c-oblika kanala bazira se na definisanju vidljivih kriterijuma (anatomiji poda pulpne komore, perzistentnoj hemoragiji ili bolu kada se pronađu odvojeni ulazi kanala). Pulpna komora kod zuba sa c-oblikom kanala može biti prostrana u okluzoapikalnoj dimenziji sa malom bifurkacijom, a nekad kanal može biti kalcifikovan, čime se može maskirati ova konfiguracija.

Oporavak nerva zavisi od težine njegovog oštećenja, ali i od brzine uklanjanja uzroka koji je to oštećenje izazvao. Često i posle uklanjanja uzroka perzistiraju simptomi parestezije jer povreda nije samo mehanička nego i hemijske prirode. Endodontski marerijali mogu dospeti do periapeksa na četiri različita načina (preko nervnog snopa, drenažom kroz limfne sudove, periapikalnim kapilarnim sistemom i difuzijom između kosti i sluzokože kroz membranu prema mekim tkivima) [2, 25]. Sama anatomija donje vilice favorizuje difuziju endodontskih materijala, posebno u zadnjoj oblasti donje vilice zbog trabekularne građe spongiozne kosti, koja olakšava širenje različitih materijala u okolna tkiva. Posebnu pažnju treba obratiti na udaljenost između anatomskih otvora mandibularnih molara i mandibularnog kanala. Prema jednoj studiji ovo rastojanje varira između 1 i 4 mm u slučaju prvog donjeg molara, dok je manje od 1

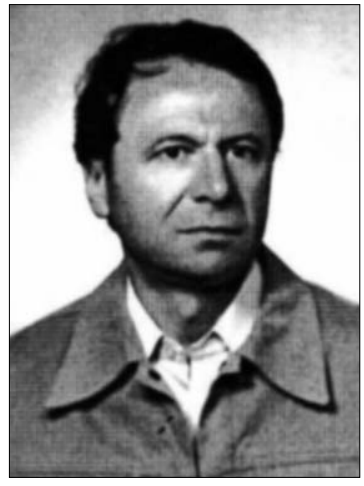
mm za drugi i treći donji molar [25]. Cone Beam kompjuterizovana tomografija, kao savremena dijagnostička metoda, može pomoći u izradi dobrog plana terapije i u preventivi nastanka parestezija kao komplikacija endodontske terapije [1].

Terapija zuba uzročnika neurosenzitivnih disfunkcija uz adekvatan endodontski tretman zahteva primenu hladnih obloga, analgetike, antibiotsku terapiju, nesteroidne antiinflamatorne lekove, sintetske kortikosteroide (dexamethasone), proteolitičke enzime (razgrađuju koagulum), vitamine B kompleksa, vitamin C (antioksidativnim dejstvom redukuje efekte ishemije) i adenozin-trifosfat, koji potpomaže regeneraciji tkiva [15, 16]. Najvažnije je što pre dijagnostifikovati parasteziju i ukloniti eventualne uzročnike ove disfunkcije, poželjno je za 48 sati [12, 27]. Hirurške metode terapije podrazumevaju ekstrakciju zuba uzročnika, apikotomiju, hirurško uklanjanje stranog tela [27].

Na osnovu anamnestičkih podataka i kliničkog nalaza donji drugi molar u ovom prikazu je svrstan u drugu grupu oštećenja po Seddonovoj klasifikaciji, tj. prepoznat je kao aksonotmeza, koja se manifestuje oduzetošću motorne i senzibilne funkcije nerva. Usled bliske veze anatomskog otvora i mandibularnog kanala najverovatnije je endodontski ručni instrument izazvao povredu aksona i mijelinske ovojnice, a NaOCl koji je korišćen kao irigans izazvao hemijsku iritaciju. Izlečenje je nastalo spontano, adekvatnim endodontskim tretmanom, sa dezinfekcijom kanala korena, koja je od suštinskog značaja. Povoljne uslove za izlečenje svakako su pomogli i niska koncentracija NaOCl kao irigansa i izbor gutaflow paste za definitivnu opturaciju.

Ključ uspešne endodonske terpije ovakvih komlikovanih kanalnih konfiguracija je u poznavanju dentalne anatomije i primeni odgovarajućih tehnika instrumentacije i opturacije. U prezentovanom slučaju je pored korektno sprovedene endodontske terapije iskorišćeno i pozitivno svojstvo guttaflow paste, koja je siler izbora u slučajevima bliskog odnosa apeksa zuba i mandibularnog kanala.

VNS dr sc. Dragutin Cekić (1931–2017)



Dr sc. Dragutin Dragan Cekić preminuo je 6. januara 2017. godine.

Dr Dragutin Cekić, viši naučni saradnik Klinike za dečju i preventivnu stomatologiju Stomatološkog fakulteta u Beogradu, rođen je 25. 8.1931. godine u Pirotu, u Srbiji. Osnovnu školu i gimnaziju završio je u mestu rođenja, a novoosnovani Stomatološki fakultet Beogradskog univerziteta, tada jedini te vrste na Balkanu i Jugoistočnoj Evropi, upisao je 1950. godine, a isti završio 1958. godine. Bila je to treća redovna generacija najboljih kandidata iz R. Srbije i susednih republika SFRJ-a.

Nakon završenog vojnog roka zaposlio se u Domu zdravlja u Pirotu, gde je proveo godinu dana, a potom 1961. godine prelazi u Beograd, gde nekoliko meseci radi u VI dečjem dispanzeru kao dečji zubni lekar. Aprila 1962. godine primljen je za kliničkog lekara na Klinici za dečju i preventivnu stomatologiju Stomatološkog fakulteta u Beogradu, gde je ostao do odlaska u starosnu penziju. Na klinici za dečju i preventivnu stomatologiju Stomatološkog fakulteta u Beogradu prošao je sve faze počev od kliničkog lekara i asistenta-pripravnika, potom asistenta i višeg stručnog saradnika pa sve do naučnog saradnika i višeg naučnog saradnika.

Kao odličan student bio je demonstrator na predmetima Fiziologija sa biohemijom i Bolesti zuba. Bio je nadaren i muzički školovan i za vreme studija je bio aktivan član kulturno-umetničkih društava Beogradskog univerziteta „Mika Mitrović“ i „Branko Krsmanović“, sa kojima je gostovao u mnogim evropskim i vanevropskim zemljama (Sovjetski Savez, Kina, Japan), u kojima su postigli velike uspehe i nepodeljene simpatije.

Tokom svoje uspešne karijere boravio je na prestižnom lstmanovom institutu u Stokholmu (Švedska) i klinikama i institutima u Engleskoj (London, Birmingen,Njukastl i dr.). Govorio je tečno engleski i francuski.

Od dolaska na kliniku pa sve do odlaska u starosnu penziju uspešno se bavio zdravstveno-vaspitnim radom (prevencijom i lečenjem različitih bolesti usta i zuba predškolske i školske dece i školske omladine), nastavno-obrazovnim radom u redovnom i poslediplomskom školovanju studenata, lekara na stažu i specijalizaciji i naučnoistraživačkim radom (terenska i klinička istraživanja najčešćih bolesti usta i zuba, karijesa, gingivoparodontalnih oboljenja i ortodontskih anomalija).

U toku svog dugogodišnjeg rada na predmetu i klinici napisao je značajan broj stručnih i naučnih radova, referata, studija i projekata, koji su izlagani na brojnim stručnim i naučnim sastancima, simpozijumima i kongresima i štampani u eminentnim stručno-naučnim publikacijama u zemlji i inostranstvu.

Učestvovao je u pisanju odabranih poglavlja zajedničkog udžbenika Dečja i preventivna stomatologija, kao i u pisanju različite pomoćne udžbeničke i zdravstveno-vaspitne literature u cilju poboljšanja zdravstvene zaštite, a time i zdravlja stanovništva Srbije, posebno dece i omladine.

Posebno treba istaći da je kruna njegovog dugogodišnjeg stvaralačkog, kliničkog, lekarskog rada bio i veliki doprinos u obradi i lečenju hendikepirane, autistične, zaostale u fizičkom i psihičkom razvoju dece i omladine, kao i lečenju dece i omladine od različitih, nekad i najtežih bakterijskih i virusnih bolesti (sida, infektivni hepatitis i dr.).

Na osnovu svega ovoga može da se zaključi da je ličnost i dugogodišnji trud i rad dr sc. Dragutina Cekića ušao u anale dostignuća vrednog nastavno-obrazovnog kolektiva Dečje i preventivne stomatologije, a time i podigao ugled i renome klinike i fakulteta.

Bio je član mnogih društava, organizacija i asocijacija: Srpskog lekarskog društva (Stomatološka sekcija i Sekcija dečje i preventivne stomatologije), Crvenog krsta, umetničkih društava „Mika Mitrović“ i „Branko Krsmanović“ i mnogih drugih, a njegove kućne vitrine krase mnoge pohvale, zahvalnice, diplome, povelje i nagrade.

O našem dragom drugu, kolegi, prijatelju i saborcu Dragutinu Draganu Cekiću, od milošte „Celetu“, moglo bi se mnogo, mnogo lepog govoriti, a još više pisati i sve bi bilo nedovoljno izrečeno i izgovoreno.

Svako od nas koji je sa njim dugo proveo u životu, radu i druženju nosi sa sobom poseban osećaj o njegovoj ličnosti i delu.

Neka osobeni epitaf sa čitulje kolektiva Stomatološkog fakulteta u Beogradu bude oličenje svega toga:

„Opraštamo se od našeg dragog prijatelja i saradnika, ličnosti koja je plenila svojom dobrotom, vedrinom, otvorenošću i iskrenošću.“

Ostaje velika praznina zbog odlaska sjajnog čoveka i prijatelja.

Neka mu je večna slava i hvala!

U Beogradu,
14. februar 2017. god.

Prof. dr sc.Vojislav Popović

Da li ste pažljivo čitali radove?

1. Terapija proteznog stomatitisa se sprovodi:
   a) upotrebom nistatina i mikonazola
   b) antibiotskom terapijom
   c) antivirusnom terapijom

2. Slučaj iz kliničke prakse je predstavio endodontsko lečenje drugog donjeg molara:
   a) sa uobičajenim kanalskim sistemom
   b) sa dva kanala
   c) sa kanalom „S“ oblika

3. Najčešći pristupi na polju inženjerstva tkiva uključuju:
   a) matične ćelije koje se zasejavaju na skafolde
   b) primenu različitih membrana
   c) primenu metalnih struktura

4. Rezultati analize skeletnih ostataka su ukazali na veliki stepen:
   a) karijesnih zuba
   b) abrazije zuba
   c) izvađenih zuba

5. Oboljenje protezni stomatitis je obično udružen sa:
   a) virusnom infekcijom
   b) gljivičnom infekcijom
   c) bakterijskom infekcijom velikog broja mikroorganizama

6. Kao skafoldi mogu da se koriste:
   a) metali
   b) keramički nosači
   c) tečne supstance

7. Analiza skeletnih ostataka je ukazala na značajan broj:
   a) zaostalih korenova
   b) periapikalnih procesa
   c) parodontopatičnih zuba

8. Kod endodontskog slučaja drugog donjeg molara postojala je parestezija:
   a) donjeg alveolarnog nerva
   b) palatinalnog nerva
   c) mentalnog nerva

9. Antiseptično sredstvo listerin može da se koristi u terapiji proteznog stomatitisa
   a) Da
   b) Ne
   c) Samo kad nema gljivične infekcije

10. Simptomi kod parestezije mogu biti u vidu:
    a) neurosenzitivne disfunkcije
    b) jakih bolnih senzacija
    c) blagih senzacija koje se ponavljaju

11. Od analiziranih skeleta dečjem uzrastu su pripadala:
    a) dva skeleta
    b) tri skeleta
    c) pet skeletova

12. Daktanol gel je korišćen u:
    a) kontrolnoj grupi
    b) eksperimentalnoj grupi
    c) u obe terapijske grupe

13. Metoda polimerne matrice je jedan od postupaka izrade skafolda
    a) Da
    b) Ne
    c) Da, ali ne zadovoljava sve zahteve

14. Lokalni faktor za nastanak parestezije tokom endodontskog zahvata može biti rastvor za irigaciju
    a) Da
    b) Ne
    c) Zavisi od zuba

15. Antiseptički rastvor listerin kod proteznog stomatitisa dovodi do:
    a) povećanja intenziteta zapaljenja
    b) smanjenja intenziteta zapaljenja
    c) nema efekta na zapaljenje

16. Aksonotmeza je:
    a) povreda prvog stepena
    b) povreda drugog stepena
    c) povreda trećeg stepena

17. Analiza skeletnih ostatka antropološke serije Gomolova je obuhvatila:
    a) 20 skeletnih ostataka
    b) 30 skeletnih ostataka
    c) 40 skeletnih ostataka

18. Veću efikasnost u lečenju proteznog stomatitisa pokazao je:
    a) daktanol gel
    b) listerin
    c) mikonazol

19. Dizajn skafolda treba da obezbedi:
    a) poroznost i biodegradibilnost
    b) dobre mehaničke karakteristike
    c) dobru kompaktnost

20. Status zuba i vilica praistorijske humane populacije je analiziran sa lokaliteta:
    a) Vinča
    b) Gomola
    c) Smederevo

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Dental links / Stomatološki linkovi

MEDLINE
http://www.nlm.nih.gov/databases/databases_medline.html

OPERATIVE DENTISTRY
http://www.jopdent.org/journal/journal.php

JOURNAL OF CONTEMPORARY DENTAL PRACTICE
http://thejcdp.com/

JOURNAL OF THE AMERICAN DENTAL ASSOCIATION (JADA)
http://jada.ada.org/

BRITISH DENTAL JOURNAL
http://www.nature.com/bdj/index.html

JOURNAL OF DENTAL RESEARCH
http://www.iadr.com/

CANADIAN MEDICAL ASSOCIATION JOURNAL (CMAJ)
http://www.cmaj.ca/

JOURNAL OF THE AMERICAN MEDICAL ASSOCIATION (JAMA)
http://jama.ama-assn.org/

JOURNAL OF THE CANADIAN DENTAL ASSOCIATION (JCDA)
http://www.cda-adc.ca/jcda/

JOURNAL OF PERIODONTOLOGY
http://www.joponline.org/loi/jop

JOURNAL OF ORTHODONTICS
http://jorthod.maneyjournals.org/

INTERNATIONAL JOURNAL OF PAEDIATRIC DENTISTRY
http://www.wiley.com/bw/journal.asp?ref=0960-7439

AUSTRALIAN DENTAL JOURNAL
http://www.ada.org.au/Publications/adj.aspx

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