YU ISSN 0042-8450

ВОЈНОСАНИТЕТСКИ ПРЕГЛЕД

Часопис лекара и фармацеута Војске Србије

Military Medical and Pharmaceutical Journal of Serbia

Vojnosanitetski pregled

Vojnosanit Pregl 2013; January Vol. 70 (No. 1): p. 1-144.

YU ISSN 0042-8450 vol. 70, br. 1, 2013.

VOJNOSANITETSKI PREGLED

Prvi broj *Vojnosanitetskog pregleda* izašao je septembra meseca 1944. godine

Časopis nastavlja tradiciju *Vojno-sanitetskog glasnika*, koji je izlazio od 1930. do 1941. godine

IZDAVAČ
Uprava za vojno zdravstvo MO Srbije

Adresa redakcije: Vojnomedicinska akademija, Institut za naučne informacije, Crnotravska 17, poštanski fah 33–55, 11040 Beograd, Srbija. Telefoni: glavni i odgovorni urednik 3609 311, glavni menadžer časopisa 3609 479, pretplata 3608 997. Faks 2669 689. E-mail (redakcija): **vsp@vma.mod.gov.rs**

Radove objavljene u „Vojnosanitetskom pregledu" indeksiraju: Science Citation Index Expanded (SCIE), Journal Citation Reports/Science Edition, Index Medicus (Medline), Excerpta Medica (EMBASE), EBSCO, Biomedicina Serbica. Sadržaje objavljuju Giornale di Medicine Militare i Revista de Medicina Militara. Prikaze originalnih radova i izvoda iz sadržaja objavljuje International Review of the Armed Forces Medical Services.

Časopis izlazi dvanaest puta godišnje. Pretplate: žiro račun kod Uprave za javna plaćanja u Beogradu br. 840-941621-02 – VMA (za Vojnosanitetski pregled), poziv na broj 1962-1. Za pretplatu iz inostranstva obratiti se službi pretplate na tel. 3608 997. Godišnja pretplata: 5 000 dinara za građane Srbije, 10 000 dinara za ustanove iz Srbije i 150 € (u dinarskoj protivvrednosti na dan uplate) za pretplatnike iz inostranstva. Kopiju uplatnice dostaviti na gornju adresu.

Štampa Vojna štamparija, Beograd, Resavska 40 b.

YU ISSN 0042-8450 vol. 70 No. 1, 2013

VOJNOSANITETSKI PREGLED

The first issue of *Vojnosanitetski pregled* was published in September 1944
The Journal continues the tradition of *Vojno-sanitetski glasnik* which was published between 1930 and 1941

PUBLISHER

Military Health Department, Ministry of Defence, Serbia

Editorial Office: Military Medical Academy, INI; Crnotravska 17, PO Box 33–55, 11040 Belgrade, Serbia. Phone: Editor-in-chief +381 11 3609 311; Main Journal Manager +381 11 3609 479; Fax: +381–11–2669–689; E-mail: **vsp@vma.mod.gov.rs**
Papers published in the Vojnosanitetski pregled are indexed in: Science Citation Index Expanded (SCIE), Journal Citation Reports/Science Edition, Index Medicus (Medline), Excerpta Medica (EMBASE), EBSCO, Biomedicina Serbica. Contents are published in Giornale di Medicine Militare and Revista de Medicina Militara. Reviews of original papers and abstracts of contents are published in International Review of the Armed Forces Medical Services.
The Journal is published monthly. Subscription: Account in Uprava za javna plaćanja in Belgrade. Giro Account No. 840-941621-02 – VMA (za Vojnosanitetski pregled), refer to number 1962-1. To subscribe from abroad phone to +381 11 3608 997. Subscription prices per year: individuals 5,000.00 Din, institutions 10,000.00 Din in Serbia, and foreign subscribers 150 €.

Printed by: Vojna štamparija, Beograd, Resavska 40 b.

SADRŽAJ / CONTENTS

Spomen-kosturnica u obliku bele, mermerne piramide podignuta je u Medžidiji, Rumunija, 1926. godine, u slavu palim borcima Prve srpske dobrovoljačke divizije, čuvene po hrabrosti i samopožrtvovanju tokom borbi u Dobrudži, u jesen 1916. godine, za vreme Prvog svetskog rata (vidi str. 90–101).

The mausoleum, in the form of white, marble pyramid, builded in Medgidia, Romania, in 1926 in honor of the fallen soldiers of the First Serbian Volunteer Division, famous for their courage and self-sacrifice during combats in the fall of the 1916 in Dobrudja, during the First World War (See p. 90–101).

E D I T O R I A L / U V O D N I K

Evergreen

Evergrin

Silva Dobrić

Institute for Scientific Information, Military Medical Academy, Belgrade, Serbia

The end of year and the beginning of another, although unintentionally, always reminds us of the inevitable passage of time, more pronounced in the elderly. However, there is something, very rare indeed, that regardless of the passing of time remains forever young. Scientific journals fall into that rare category of evergreens. Their "elixir of youth" are the papers published on their pages, i.e. their actuality and novelities content not allowing "ravages of time" to take its course. On the other hand, editorial boards and publishers also make efforts to maintain the vitality of their favourite. Therefore, it is not surprising that scientific journals published more than 100 years are still "IN"; and what's more, it is an honor and a privilege to publish in them.

The "Vojnosanitetski Pregled" (VSP) will be its respectable 70 years of existence in less than two years, and today it seems younger than ever. Joining the renowned citation database Science Citation Index Expanded in 2008 and getting the impact factors had a major influence on it. Since then, new manuscripts have been arriving almost everyday, so that they count between 300 and 350 on annual basis. Analysing the papers published in the VSP during 2012 made in the mid of December, just before writing this Editorial, showed 281 papers received from January 1 2012 to date (256 by Serbian and 25 by foreign authors), but it is expected to be closer to 300 or even more by the end of the year. However, a less number of manuscripts submitted in 2012 as compared to 2010 and 2011 when 340 and 324 manuscripts were sent, respectively, can be explained by the fact that now the manuscripts from the so-called border medical fields are no longer taken into account, which was not the case in the previous years, and that the manuscripts received have to be written in English, which can also be a limiting factor for some number of, primarily, local authors. However, the general assessment of the Editorial Board of the Journal is that the quality of the submitted papers is improving each year, that is the greatest guarantee for its survival and further advancement.

From the submitted manuscripts, by mid-December, the peer review process have been completed for 142 ones, of

Prelazak iz jedne u drugu godinu, i nehotice, uvek nas podseti na neumitnost prolaznosti, što je sve izraženije što je čovek stariji. Međutim, ima stvari, doduše malo, koje bez obzira na vreme trajanja ostaju večno mlade. Naučni časopisi spadaju u tu retku kategoriju evergrina. Njihov „eliksir mladosti" predstavljaju radovi objavljeni na njihovim stranicama, odnosno sadržaji tih radova jer oni, svojom aktuelnošću i novinama koje donose, ne dozvoljaju da „zub vremena" učini svoje. S druge strane, i izdavači, odnosno uredništva časopisa, nastoje da održe vitalnost svog pulena. Stoga ne čudi što su časopisi, koji su odavno prebacili 100 godina postojanja, i dalje „IN", štaviše, čast je i privilegija objaviti rad u njima.

„Vojnosanitetski pregled" (VSP) za nepune dve godine napuniće respektabilnih 70 godina postojanja, a danas, čini se, mlađi je nego ikada. Ulazak u poznatu citatnu bazu Science Citation Index Expanded 2008. godine i dobijanje impakt faktora imali su presudan uticaj na to. Od tada, priliv novih radova u Redakciju časopisa gotovo da je svakodnevan, tako da se na godišnjem nivou kreće između 300 i 350. Analiza pristiglih radova u toku 2012. godine, učinjena sredinom decembra, neposredno pred pisanje ovog Uvodnika, pokazala je da je od 1. januara 2012, do tog datuma, primljen 281 rad (256 iz Srbije i 25 iz inostranstva), ali za očekivati je da će se do kraja godine približiti broju 300 ili ga čak i premašiti. Ipak, nešto manji broj pristiglih radova u odnosu na 2010. godinu, kada je primljeno 340 radova, i 2011. sa 324 primljena rada, može se objasniti činjenicom da sada više ne uzimamo u razmatranje radove iz tzv. graničnih medicinskih oblasti, što prethodnih godina nije bio slučaj, i što radovi koje primamo moraju biti napisani na engleskom jeziku, što, takođe, može da bude ograničavajući faktor za izvestan broj, prvenstveno, domaćih autora. Međutim, opšta ocena Uredništva jeste da se kvalitet pristiglih radova iz godine u godinu poboljšava, a to je najveći garant daljeg opstanka i napredovanja časopisa.

Od pristiglih radova, do sredine decembra proces recenziranja završen je za 142 rada, od čega je njih 60% ko-

Correspondence to: Silva Dobrić, Military Medical Academy, Institute for Scientific Information, Crnotravska 17, 11 000 Belgrade, Serbia. Phone: +381 11 36 09 311. E-mail: vsp@vma.mod.gov.rs

which 60% were finally accepted for publishing, and 40% rejected, the general conclusion being that the number of rejected papers increases from year to year, directly indicating the raising criteria for papers acceptance by the Editorial Board and by the referees.

The number of papers published in 2012 is 186 being almost the same as in the previous year (in 2011 totally 187 different categories articles) (Table 1). However, it should be noted that since the early 2012, in addition to the printed version of each issue, 4−5 articles have been published every month, in electronic form OnLine-First with DOI numbers to be available through the website of the Journal and through the DOISerbia service and the Serbian Citation Index (SCIndex) and the website of the Consortium of Libraries of Serbia for Coordinated Acquisition (KoBSON). Thus, we want articles more to, be visible and available for citation especially those recommended by the reviewers to be published as high priority.

As it was earlier, the most numerous of the published articles are those befalling to the categories Original articles (50%), and Case reports (24.2%), which is the tendenly with the articles published OnLine-First: 33 original articles, 8 case reports, 6 current topics and 3 general reviews.

When analyzing the published articles by authors affiliation, the situation from the previous years repeats itself. Namely, the largest number of articles published on the pages of VSP still come from the authors of the so-called "civilian health sector". In 2012 there have been 75% of articles from civil medical and academic institutions, 18.5% articles written by authors from military medical institutions (mainly from the Military Medical Academy, Belgrade), and 6.5% were articles co-written by authors from both civil and military medical institutions.

As readers of the VSP already know, in the early 2012 on all the manuscripts that come to the Editorial Office have to be submitted electronically through the e-Ur system − the system for electronic editing of journals. From 24 July, 2012, we have been using the improved version of the system called ASEESTANT. It offers several benefits: checking manuscripts submitted to plagiarism/selfplagiarism, control of references accuracy, and the selection of appropriate keywords according to the thesaurus of the key words from the U.S. National Library of Medicine, which, as the standardized terms, are used in all medical scientific publications. This service, so, makes the work of the Editorial Staff and the Editorial Board of the Journal easier, the quality and regularity of the review process improved, and the protection against duplicate publication and/or plagiarism possible, making us believe all that contribute to a better quality and impact of the Journal.

In the appraching 2013 we plan to go on with the tradition of raising the Journals quality. In order to prevent authorship misuse, in the sense of undeserved authorship (adding in the byline names of those not contributing to all phases of scientific work), all authors of submitted manuscripts will have to sign the statement of contribution to the work. Also, they will have to sign the statement of conflicts making us of interests, as another important element of establishing thrue ethical principles in scientific publishing.

načno prihvaćeno za objavljivanje, dok je 40% odbijeno. Treba istaći da se iz godine u godinu broj odbijenih radova povećava, što je direktni pokazatelj podizanja kriterijuma za prihvatanje radova i od strane Uredništva i od strane recenzenata.

Što se tiče broja objavljenih radova u 2012. godini, on je gotovo isti kao i prethodne 2011. godine i iznosi 186 (u 2011. objavljeno je ukupno 187 radova iz različitih kategorija) (Tabela 1). Međutim, treba naglasiti da je od početka 2012. godine, svaki mesec, pored štampane verzije pojedinog broja, izlazilo 4−5 radova u elektronskom obliku kao OnLine-First sa DOI brojem, koji su bili dostupni preko sajta časopisa i servisa DOISerbia u Srpskom citatnom indeksu (SCIndeks), dostupnom preko sajta Konzorcijuma biblioteka Srbije za objedinjenu nabavku (KoBSON). Na ovaj način želeli smo da što veći broj radova, pogotovo onih koji dobiju preporuku od recenzenata za objavljivanje po prioritetu, bude što pre vidljivo i dostupno za citiranje.

Kao i proteklih godina, među objavljenim radovima najviše je bilo onih iz kategorije Originalni članci (50%), iza koji slede Prikazi bolesnika (24,2%). Slična struktura je i među radovima objavljenim OnLine-First. U 2012. godini na taj način objavljena su 33 originalna rada, osam prikaza bolesnika, šest aktuelnih tema i tri opšta pregleda.

Kada se analiziraju afilijacije autora objavljenih radova, ponavlja se situacija iz prethodnih godina da najveći broj radova objavljenih na stranicama VSP-a i dalje dolazi od autora iz tzv. civilnog sektora. U 2012. godini učešće ovih radova iznosilo je 75%, 18,5% su činili radovi autora iz vojnozdravstvenih ustanova (uglavnom iz Vojnomedicinske akademije, Beograd), dok se preostalih 6,5% odnosilo na radove koji su zajedničko delo autora iz vojnih i civilnih zdravstvenih ustanova.

Kao što je čitaocima VSP-a poznato, od početka 2012. godine, svi radovi koji dolaze u Redakciju časopisa predaju se elektronski kroz sistem e-Ur: Elektronsko uređivanje časopisa. Od 24. jula 2012. koristimo unapređenu verziju tog sistema pod nazivom ASEESTANT. Ona pruža nekoliko pogodnosti: proveru pristiglih rukopisa na plagijarizam/autoplagijaritzam, kontrolu ispravnosti referenci navedenih u prijavljenim radovima i izbor odgovarajućih ključnih reči prema tezaurusu ključnih reči američke Nacionalne medicinske biblioteke koje se, kao standardizovani termini, koriste u svim medicinskim naučnim publikacijama. Zahvaljujući ovom servisu olakšan je rad Redakcije i Uredništva VSP-a, poboljšan je kvalitet i regularnost recenzentskog postupka, obezbeđena zaštita od objavljivanja duplikata i/ili plagijata, što će, verujemo, doprineti boljem kvalitetu i uticajnosti časopisa.

U nastupajućoj 2013. godini nastavljamo sa podizanjem kvaliteta časopisa. U cilju sprečavanja zloupotrebe autorstva, u smislu nezasluženog dopisivanje među autore i onih koji nisu bitno doprineli u različitim fazama pripreme rada za objavljivanje, ubuduće će se od svih autora tražiti potpisana izjava o doprinosu radu. Takođe, tražiće se i potpisana izjava o nepostojanju konflikta interesa, kao još jednog značajnog elementa uspostavljanja pravih etičkih principa u naučnom publikovanju.

We keep trying to enlarge our reviewers team by a number of foreign reviewers which proved to be a very good shift in the past year. A good reviewer is of immeasurable value, and due to this, the Editorial Board and the Publisher of the VSP always show sincere appreciation of their efforts to raise the quality of articles, and, in so doing, the quality od the Journal.

The names of the reviewers who deserve, on this occasion, great recognition and appreciation for the cooperation during the 2012, with an invitation to join us in 2013, are listed in Table 2.

Takođe, nastojaćemo da naš recenzentski tim ojačamo sa većim brojem recenzenata iz inostranstva, što se pokazalo kao izuzetno dobar potez u protekloj godini. Dobar recenzent zlata vredi, i zato im Redakcija, Uredništvo i Izdavač VSP-a uvek izražavaju najiskreniju zahvalnost za sve napore koje ulažu u podizanju kvaliteta radova, a tome i samog časopisa.

Imena recenzenata, kojima ovom prilikom odajemo veliko priznanje i zahvalnost za saradnju tokom 2012. godine, sa pozivom da nam se pridruže i u 2013, navedena su u tabeli 2.

Table 1

Categories and the number of articles published in the Vojnosanitetski pregled in 2012 / Kategorije i broj članaka objavljenih u Vojnosanitetskom pregledu u 2012.

Category of an article/ Kategorija članka	Articles / Članci	
	n	%
Editorial/ Uvodnik	8	4.3
Original article/ Originalni članak	93	50.0
General review/ Opšti pregled	7	3.8
Current topic/ Aktuerlna tema	16	8.6
Practical advices for physicians/ Seminar praktičnog lekara	2	1.1
Case report/ Prikaz slučaja	45	24.2
History of medicine/ Istorija medicine	4	2.2
Letter to the editor/ Pismo uredniku	2	1.1
In focus/ U fokusu	6	3.2
Book review/ Prikaz knjige	2	1.1
Scientific meeting report/ Izveštaj sa stručnog skupa	1	0.5
Total	186	100.0

Tabela 2

Reviewers of the Vojnosanitetski pregled in 2012 / Recenzenti Vojnosanitetskog pregleda u 2012. godini

Aleksić Petar
Antić Branislav
Arsović Nenad

Baletić Nenad
Balint Bela
Bančević Vladimir
Berisavac Milica
Beutin Lothar
Bjegović Mikanović Vesna
Bogdanović Dragana
Bogovac Mirjana
Bokonjić Dubravko
Bouros Demosthenes
Božinović Prekajski Niveska
Brkić Snežana
Brkić Zlata

Čabarkapa Milanko
Čolović Radoje

Catalan Alfonso
Cerović Snežana
Cikota Bojana
Cohen Irun

Ćuk Vladimir
Ćirić Jasmina
Ćirić Zoran

Daković Dragana
Davidović Lazar
Dedić Gordana
Dimković Siniša
Dinčić Dragan
Dinčić Evica
Dobrić Silva
Dragović Simić Viktorija
Dragović Tamara
Duka Miloš

Đerić Dragoslava
Đurović Aleksandar
Đurović Branika
Đurović Branislav
Đurović Branka

Garović Vesna
Gazivoda Dragan
Glišić Branislav
Gržić Renata
Hajduković Zoran

Hrvačević Rajko

Igić Rajko
Ignjatović Svetlana
Ilić Radoje
Ilić Stojanović Olivera
Ilić Tihomir
Išpanović Radojković Veronika
Ivanovski Ninoslav

Jakovljević Vladimir
Janić Dragana
Janjić Zlata
Janković Borisav
Janković Radmilo
Janković Slobodan
Jauković Ljiljana
Jovanović Dragana
Jovanović Ida
Jović Jasna
Jović Nebojša

Kandolf -Sekulović Lidija
Kondo Akiko
Kostić Vladimir

Kovačević Nada
Kozarski Jefta
Kozomara Ružica
Kuljuć Kapulica Nada

Laaser Ulrich
Lakić Aneta
Lazić Srđan
Lazić Zoran
Lečić Toševski Dušica
Lepić Toplica
Lepšanović Zorica
Ljubić Aleksandar
Lučić Miloš

Magić Zvonko
Maksić Đoko
Mandić-Gajić Gordana
Marić Nađa
Marjanović Ivan
Marjanović Marjan
Marjanović Slobodan
Marković Dejan
Martinović Žarko
Matić Smiljana
Medenica Ivica

Micić Dragan
Mićić Sava
Mijušković Željko
Mikić Dragan
Mikov Momir
Miladinov-Mikov Marica
Milenković Dragica
Milenković Marina
Milenković Svetislav
Mileusnić Dušan
Milović Novak
Miodrag Čolić
Mirković Darko
Mirović Veljko

Nedeljković Nenad
Nežić Duško
Nikolić Branka
Nikolić Dragan
Nikolić Ljubiša
Nikolić Miloš
Nikolić Predrag
Nikolić-Đurović Marina
Novaković Marjan
Nožić Darko

Obradović Dragana
Obradović Miljana
Obradović Slobodan
Opinćal-Stošić Tatjana
Otašević Petar

Pašić Srđan
Pavlović Milorad
Pekmezović Tatjana
Pelemiš Milomir
Perišić Nenad
Perišić Živko
Pešut Dragica
Petković Stevan
Petronijević Milan
Petrović Bojan
Petrović Silvana
Petrović Zdravko
Pizzo Giuseppee
Plavec Goran
Popović Brkić Vera
Popović Zoran
Potpara Tatjana
Potthoff Andrej

Radak Đorđe
Radaković Sonja
Rađen Slavica
Radosavljević Tatjana
Radosavljević Vladan
Raičević Ranko
Rajšić Nenada
Rebić Predrag
Renn John
Ristić Anđelka
Ristić Ljubiša
Roganović Zoran

Romić Predrag
Šašić Mirjana
Šobić Šaranović Dragana
Špirić Željko
Šuljagić Vesna
Šurbatović Maja

Samardžić Radomir
Savić Slobodan
Sharma Amit
Simić Snežana
Slavković Slobodan
Sokić Dragoslav
Sood Sankalp
Srdić-Rajić Tatjana
Stamatović Dragana
Stamatović Novak
Stamenković Dragoslav
Stamenković Dušica
Stanković Goran
Stanković Nebojša
Stefanović Dara
Stefanović Dušan
Stolić Radojica
Stošić Srboljub
Strajnić Ljiljana
Subotić Dragan

Tadić Vanja
Tarabar Dino
Tatić Svetislav
Tatić Vujadin
Tavčiovski Dragan
Terzić Milan
Till Viktor
Todorić Milomir
Todorović Aleksandar
Todorović Ljubomir
Todorović Milena
Tomić Aleksandar
Tukić Ljiljana
Tulić Cane

Ušaj – Knežević Slavica

Vasić Jugoslav
Vasiljević Nađa
Vezmar Kovačević Sandra
Vučićević Katarina
Vučinić Slavica
Vučinić Žarko
Vučković Sonja
Vujić Dragana
Vukčević Vladan
Vukomanović Vladislav

Zečević Radoš
Zolatorevski Lidija

Živković Maja
Žunić Gordana

ORIGINALNI ČLANAK

UDC: 617-089.5:[615.816:616.366-089.87
DOI: 10.2298/VSP1301009S

Efekti mehaničke ventilacije kontrolisane pritiskom kod osoba sa oštećenjem respiratorne funkcije tokom laparoskopske holecistektomije

Effect of mechanical pressure-controled ventilation in patients with disturbed respiratory function during laparoscopic cholecystectomy

Maja Šurbatović*‖, Zoran Vesić†, Dragan Djordjević*, Sonja Radaković‡‖, Snježana Zeba*‖, Duško Jovanović*, Marijan Novaković§‖

***Klinika za anesteziologiju i intenzivnu terapiju, ‡Sektor za preventivnu medicinu, §Uprava, Vojnomedicinska akademija, Beograd, Srbija; †Ministarstvo odbrane Republike Srbije, Beograd, Srbija; ‖Medicinski fakultet Vojnomedicinske akademije, Univerzitet odbrane, Beograd, Srbija**

Apstrakt

Uvod/Cilj. Danas se laparoskopska holecistektomija (LH) smatra „zlatnim standardom" za laparoskopsku hirurgiju. Kod bolesnika sa pratećim oštećenjem respiratorne funkcije, međutim, pneumoperitoneum i položaj bolesnika neophodni za izvođenje procedure LH, dovode do dodatne intraoperativne respiratorne disfunkcije koja predstavlja izazov za anesteziologa. Cilj našeg istraživanja bio je da se utvrdi koji od dva primenjena modusa mehaničke ventilacije obezbeđuje bolje ventilatorne parametre i parametre oksigenacije tokom izvođenja anestezije za LH kod bolesnika koji pripadaju grupi III prema *American Society of Anaesthesiologists* (ASA) klasifikaciji zbog pratećih respiratornih oboljenja. **Metode.** Ispitivanjem su obuhvaćene dve grupe po 30 bolesnika podvrgnute LH. Prva grupa bila je ventilisana primenom tipa intermitentnog pozitivnog pritiska u vazdušnim putevima (grupa IPPV), a druga primenom tipa ventilacije kontrolisane pritiskom (grupa PCV). U četiri vremenska intervala praćeni su respiratorni parametri: respiratorni volumen (VT), vršni inspiratorni pritisak (PIP), komplijansa (C), parcijalni pritisak CO_2 na kraju ekspirijuma ($PETCO_2$), saturacija arterijske krvi kiseonikom (SpO_2), parcijalni pritisci kiseonika i ugljen-dioksida u arterijskoj krvi (PaO_2 i $PaCO_2$) i pH arterijske krvi. **Rezultati.** Nalazi VT, SpO_2, PaO_2, $PaCO_2$ i pH nisu se statistički značajno razlikovali ni unutar, ni između grupa. U vremenskom intervalu t_1 nije bilo statistički značajne razlike u vrednostima PIP, C, $PETCO_2$ između IPPV i PCV grupe. U sledeća tri vremenska intervala bilo je statitički značajne do visokoznačajne razlike u vrednostima ova tri respiratorna parametra između dve ispitivane grupe: PIP je bio manji, a C i $PETCO_2$ bili su veći u PCV grupi. **Zaključak.** Mehanička ventilacija tipa PCV obezbeđuje bolje intraoperativne parametre ventilacije tokom izvođenja LH kod bolesnika koji pripadaju grupi III prema ASA klasifikaciji zbog pratećih respiratornih oboljenja.

Ključne reči:
laparoskopija; disanje, veštačko; ventilacija po tipu intermitentnog pozitivnog pritiska.

Abstract

Background/Aim: Laparoscopic cholecystectomy is considered to be the gold standard for laparoscopic surgical procedures. In ASA III patients with concomitant respiratory diseases, however, creation of pneumoperitoneum and the position of patients during surgery exert additional negative effect on intraoperative respiratory function, thus making a higher challenge for the anesthesiologist than for the surgeon. The aim of this study was to compare the effect of intermittent positive pressure ventilation (IPPV) and pressure controlled ventilation (PCV) during general anesthesia on respiratory function in ASA III patients submitted to laparoscopic cholecystectomy. **Methods.** The study included 60 patients randomized into two groups depending on the mode of ventilation: IPPV or PCV. Respiratory volume (VT), peak inspiratory pressure (PIP), compliance (C), end-tidal CO_2 pressure ($PETCO_2$), oxygen saturation (SpO_2), partial pressures of O_2, CO_2 (PaO_2 and $PaCO_2$) and pH of arterial blood were recorded within four time intervals. **Results.** There were no statistically significant differences in VT, SpO_2, PaO_2, $PaCO_2$ and pH values neither within nor between the two groups. In time interval t_1 there were no statistically significant differences in PIP, C, $PETCO_2$ values between the IPPV and the PCV group. But, in the next three time intervals there was

Correspondence to: Maja Šurbatović, Vojnomedicinska akademija, Klinika za anesteziologiju i intenzivnu terapiju, Crnotravska 17, 11 040 Beograd, Srbija. Tel.: +381 11 3608 252. E-mail: maja.surbatovic@gmail.com

a difference in PIP, C, and $PETCO_2$ values between the two groups which ranged from statistically significant to highly significant; PIP was lower, C and $PETCO_2$ were higher in the PCV group. **Conclusion.** Pressure controlled ventilation better maintains stability regarding intraoperative ventilatory parameters in ASA III patients with concomitant respiratory diseases during laparoscopic cholecystectomy.

Key words:
laparoscopy; respiration, artificial; intermittent positive-pressure ventilation.

Uvod

Opšta anestezija dovodi do promena plućnih volumena usled čega nastaju i ostale promene plućne mehanike. Uvodom u opštu anesteziju snižava se vitalni kapacitet (VC), funkcionalni rezidualni kapacitet (FRC) i komplijansa grudnog koša i pluća (C). Najveći negativni uticaj na plućnu funkciju sniženjem VC, FRC, komplijanse i prouzrokovanjem alveolarne hipoventilacije i hipoksemije imaju hirurške intervencije u gornjim kvadrantima abdomena u koje spada i laparoskopska holecistektomija. Pored navedenih negativnih uticaja opšte anestezije na funkciju respiratornog sistema, tokom laparoskopskih intervencija ispoljava se dodatno negativno delovanje položaja bolesnika, povišenog intraabdominalnog pritiska i intratorakalnog pritiska usled nastanka pneumoperitoneuma. U toku laparoskopske holecistektomije najčešće se povećavaju vršni inspiratorni pritisak (PIP), intratorakalni pritisak (ITP), plućna rezistencija i parcijalni pritisak ugljen-dioksida u arterijskoj krvi ($PaCO_2$), a smanjuju se FRC, plućna komplijansa i pH arterijske krvi. Parcijalni pritisak kiseonika u arterijskoj krvi (PaO_2) može ostati isti ili se smanjuje [1].

Kod bolesnika sa preoperativnim oštećenjem plućne funkcije povećan je rizik od perioperativnog razvoja hipoksemije, hipoventilacije sa povećanim $PaCO_2$, plućne infekcije, potrebe za reintubacijom i postoperativnom mehaničkom ventilacijom [2, 3]. Kod bolesnika sa opstruktivnim oboljenjima, tokom hirurške intervencije i mehaničke ventilacije, postoji predispozicija ka pojačanom stvaranju i zastoju sekreta u disajnim putevima, poremećaju protoka gasova, razvoju atelektaza i pneumonija. Posebno su ugroženi bolesnici kod kojih se vrši hirurška intervencija u gornjim kvadrantima abdomena. Ventilacija pod pozitivnim pritiskom bolesnika sa restriktivnim oboljenjem pluća praćena je visokim vršnim pritiskom u vazdušnim putevima jer je viši pritisak potreban za širenje manje elastičnih pluća. Kod bolesnika sa hroničnim restriktivnim bolestima pluća preporučuje se ventilacija manjim respiratornim volumenom (ispod 10 mL/kg).

Po uvodu u anesteziju kranijalno pomeranje dijafragme i relaksacija respiratorne muskulature dovode do sniženja FRC i komplijanse i pluća i zida grudnog koša. Insuflacija CO_2 u peritonealnu duplju izaziva dalje pomeranje dijafragme ka glavi, dalje sniženje FRC i komplijanse, a povećanje rezistencije plućnog tkiva. Kada se FRC smanji u odnosu na *closing*-kapacitet (kapacitet zatvaranja malih vazdušnih puteva), kao rezultat atelektaze i intrapulmonalnog šanta, razvija se hipoksemija. Intraoperativna hipoksemija tokom laparoskopske holecistektomije je retka kod zdravih bolesnika, ali razvija se kod 50% gojaznih bolesnika i onih koji već imaju postojeća kardiovaskularna i respiratorna oboljenja. Prema tome, insuflacija abdominalne duplje sa CO_2 povezana je sa značajnim smanjenem FRC, velikim povećanjem vršnog inspiratornog pritiska u vazdušnim putevima i atelektazama.

Mehanička ventilacija tipa intermitentnog pozitivnog pritiska u vazdušnim putevima (IPPV) je kontrolisana (aparat potpuno preuzima ventilatornu funkciju bolesnika). Inspiratorna faza prestaje nakon postignutog određenog pritiska, volumena, protoka ili vremena što zavisi od cikliranja ventilatora. Tokom kontrolisanog disanja ventilator isporučuje zadati broj inspirijuma. Ekspiratorna faza je pasivna, a omogućena je elastičnošću grudnog koša i pluća.

Mehanička ventilacija kontrolisana pritiskom (*pressure controlled ventilation* – PCV) je oblik ventilatorne podrške tokom kojeg je ograničena vrednost maksimalnog pozitivnog pritiska koji se ostvaruje u toku inspirijuma. Da bi se smanjio negativni uticaj visokog pritiska u disajnim putevima (PIP i Paw), nađeno je rešenje primenom ventilacije tokom koje je ograničena vrednost izabranog pritiska. Vrednosti maksimalnog inspiratornog pritiska u disajnim putevima ne mogu preći vrednost određenog zadatog pritiska (Pset). Tokom ventilacije kontrolisane pritiskom, pritisak je nezavisna veličina, dok su volumen i protok zavisni od pritiska, plućne komplijanse i rezistencije plućnog tkiva. Kod PCV podešavaju se vrednosti selektovanog pritiska (Pset) koji se ostvaruje u toku inspirijuma, disajna frekvencija i inspiratorno vreme. Ovakva ventilacija je ciklirana vremenom, a vrednosti maksimalnog pritiska u disajnim putevima (PIP) i alveolarnog pritiska, koji su determinante plućne barotraume, ne mogu preći vrednost zadatog pritiska.

Tokom PCV, vršni pritisak u vazdušnim putevima održava se sve vreme inspirijuma što omogućava širenje svih jedinica pluća do stepena koji zavisi primarno od komplijanse. Veliki nedostatak PCV je činjenica da respiratorni volumen (V_T) varira u zavisnosti od komplijanse i otpora u vazdušnim putevima, tako da u toku opšte anestezije sa primenom PCV mora da se prati ostvareni respiratorni volumen da ne bi došlo do intraoperativne hipoventilacije i hipoksije.

Ključni momenti koji izazivaju različite patofiziološke promene kod laparoskopske holecistektomije su položaj bolesnika na operacionom stolu, stvaranje povećanog intraabdominalnog pritiska (IAP) i povećanog intratorakalnog pritiska (ITT) insuflacijom ugljen-dioksida u trbušnu duplju, što ima negativan efekat na respiratorni sistem, odnosno na već postojeće oštećenje plućne funkcije.

Jasno je da će već postojeća respiratorna insuficijencija, emfizem ili hronična opstruktivna bolest pluća, kod bolesnika grupe III po *American Society of Anaesthesiologists* (ASA) klasifikaciji biti pogoršana ekstenzivnošću i specifič-

nostima laparoskopske holecistektomije. Zbog toga je od izuzetnog značaja da se utvrdi koji tip mehaničke ventilacije (IPPV ili PCV), u toku anestezije za laparoskopsku holecistektomiju, dovodi do najmanjeg oštećenja respiratorne funkcije, o čemu po podacima iz literature još ne postoji saglasnost autora.

Metode

Ispitivanje je obavljeno na Klinici za anesteziologiju i intenzivnu terapiju Vojnomedicinske akademije (VMA) na ukupno 60 ispitanika, oba pola, kod kojih je postavljena indikacija za hirurško lečenje holelitijaze laparoskopskom hirurškom tehnikom.

Kriterijumi za izbor ispitanika bili su: dijagnostikovana hronična holelitijaza i postavljenja indikacija za njeno hirurško lečenje laparoskopskom operativnom tehnikom; pripadnost ispitanika grupi III po ASA klasifikaciji na osnovu prethodno izvršene procene opšteg zdravstvenog stanja (svi ispitanici pripadali su ovoj grupi zbog teškog poremećaja respiratorne funkcije, a neki od njih su, pored navedene, imali i poremećaje funkcije drugih organskih sistema).

Ispitanici su bili podeljeni prema tipu mehaničke ventilacije kojim su ventilirani tokom hirurške intervencije u dve grupe po 30. Podela ispitanika u grupe obavljena je metodom slučajnog izbora, neposredno po pozivanju bolesnika u operacionu salu: 1) bolesnici ventilisani primenom tipa IPPV – IPPV grupa; 2) bolesnici ventilisani primenom tipa PCV – PCV grupa.

U grupi IPPV, disajna frekvenca je bila 12/min, VT od 10 do 12 ml/kg TT, odnos inspirijuma i ekspirijuma je bio 1 : 2. U grupi PCV, disajna frekvenca bila je 12/min, maksimalni inspiratorni pritisak bio je ograničen na 25 cmH_2O, odnos inspirijuma i ekspirijuma bio je 1 : 2.

Kod ispitanika su praćene vrednosti respiratornih parametara: respiratorni volumen – VT, PIP, C, vrednost parcijalnog pritiska CO_2 na kraju ekspirijuma – $P_{ET}CO_2$, SpO_2, PaO_2, $PaCO_2$, pH iz arterijske krvi. Vrednosti VT, PIP, C i $P_{ET}CO_2$ registrovane su korišćenjem aparata za anesteziju Drëger-Julian u četiri vremenska intervala. Vrednost SpO_2 registrovana je korišćenjem Datex-Engstrom AS/3 monitora, a vrednosti PaO_2, $PaCO_2$, pH iz arterijske krvi određivane su korišćenjem gasnog analizatora ABL-520 Radiometar, takođe u četiri vremenskih intervala.

Merenja VT, PIP, C, $P_{ET}CO_2$, PaO_2, $PaCO_2$, pH iz arterijske krvi i SpO_2 izvedena su u sledećim vremenskim intervalima: t_1 – posle uvoda u anesteziju, a pre kreiranja pneumoperitoneuma; t_2 – 5 min posle stvaranja pneumoperitoneuma; t_3 – tokom pneumoperitoneuma; t_4 – 5 min posle oslobađanja od pneumoperitoneuma.

U statističkoj analizi primenjeni su jednosmerna analiza varijanse, Tukey-ov test, Studentov *t*-test i višestruka regresiona analiza.

Rezultati

Demografske karakteristike bolesnika i pridružena oboljenja respiratornog sistema prikazana su u tabeli 1.

Tabela 1

Demografske karakteristike bolesnika i prateća respiratorna oboljenja

Karakteristike bolesnika	IPPV	PCV
Broj (n)	30	30
Godine starosti: aritm. sredina (raspon)	57,1 (43–69)	57,3 (44–69)
Pol (n)		
muški	15	14
ženski	15	16
Prateća respiratorna oboljenja (n)		
hronični bronhitis	7	8
hronična opstruktivna bolest pluća	10	10
bronhijalna astma	6	5
emfizem pluća	6	6
sarkoidoza pluća	1	1

IPPV – mehanička ventilacija prema tipu intermitentnog pozitivnog pritiska u vazdušnim putevima; PCV –mehanička ventilacija kontrolisana pritiskom

Respiratorni volumen

Vrednosti VT, kao respiratornog parametra, merene su kontinuirano, a evidentirane su u vremenskim intervalima od t_1 do t_4. U svakoj grupi ponaosob testirana je značajnost razlike promena VT između dva uzastopna vremena (lančano) Tukey-ovim testom; u funkciji vremena unutar grupa nije bilo statističke značajnosti u promeni vrednosti VT. Između IPPV grupe i PCV grupe testirana je značajnost razlike promena VT u funkciji vremena od t_1 do t_4 Studentovim *t*-testom. Pokazalo se da između grupa nema statistički značajne promene VT. Postojala je tendencija većeg prosečnog respiratornog volumena u IPPV grupi (11,5 prema 8,5 mL/kgTT)

Vršni inspiratorni pritisak (PIP), komplijansa (C) i vrednosti parcijalnog pritiska CO_2 na kraju ekspirijuma ($P_{ET}CO_2$)

Vrednosti PIP, C i $P_{ET}CO_2$, kao respiratornih parametra, merene su kontinuirano, a evidentirane su u vremenskim intervalima od t_1 do t_4. U svakoj grupi ponaosob testirana je značajnost razlike promena svakog od ova tri respiratorna parametra između dva uzastopna vremena (lančano) Tukey-ovim testom. Što se tiče PIP-a, u grupi PCV u intervalu t_2–t_3 razlika je bila statistički značajna, u ostalim intervalima postojala je statistički visokoznačajna razlika u obe grupe. U

grupi IPPV PIP se kretao od 19 cmH_2O do 26,8 cmH_2O, dok se u grupi PCV održavao u značajno manjem rasponu od 19 cmH_2O do 22 cmH_2O. Kada je u pitanju komplijansa, u intervalu t_1–t_2 u IPPV grupi postojala je statistički visoko značajna razlika, a u grupi PCV statistički značajna razlika. U ostalim intervalima nije bilo statističke značajnosti razlike. U grupi IPPV komplijansa u intervalu od t2 do t4 bila je ispod 40 L/cmH_2O, dok je u grupi PCV bila preko 40 L/cmH_2O. Vrednosti $PETCO_2$ pokazale su da u grupi IPPV postoji statistički visokoznačajna razlika u intervalu t_1–t_2 i t_3–t_4, a u intervalu t_2–t_3 statistički značajna. Kod grupe PCV u t_1–t_2 i t_2–t_3 nije bilo statističke značajnosti, u intervalu t_3–t_4 razlika je bila statistički značajna. U grupi IPPV $PETCO_2$ bio je viši za 30 mmHg u svim vremenskim intervalima, dok je u PCV grupi bio niži od 30 mmHg. U tabeli 2 prikazano je poređenje PIP-a, C i $PETCO_2$ u okviru grupa u funkciji vremena.

Između grupa testirana je značajnost razlike u promeni PIP, C i $PETCO_2$ u funkciji vremena od t_1 do t_4 Studentovim *t*-testom. Primenom ovog testa zaključeno je da između grupa nema statistički značajne razlike u vrednostima PIP u t_1. U t_2, t_3 i t_4 postojala je statistički visokoznačajna razlika. Što se komplijanse tiče, pokazalo se da između grupa nema statistički značajne promene u t_1. Statistički značajna razlika između grupa postojala je u t_2, t_3 i t_4. Vrednosti $PETCO_2$ su pokazale da u t_1 između grupa nema statistički značajne razlike, dok je u ostalim vremenima posmatranja razlika bila visokoznačajna (t_2 i t_3) i značajna (t_n).

U tabeli 3 prikazano je poređenje PIP, C i $PETCO_2$ između grupa po vremenima merenja.

Saturacija arterijske krvi kiseonikom utvrđena pulsnom oksimetrijom (SpO_2), parcijalni pritisci kiseonika i ugljen-dioksida u arterijskoj krvi (PaO_2 i $PaCO_2$) i pH arterijske krvi

Saturacija arterijske krvi kiseonikom utvrđene pulsnom oksimetrijom merena je kontinuirano, a evidentirana je u vremenskim intervalima od t_1 do t_4. Takođe, vrednosti parcijalnih pritisaka kiseonika i ugljen-dioksida u arterijskoj krvi i pH arterijske krvi izmerene su u vremenskim intervalima od t_1 do t_4.

U svakoj grupi ponaosob (IPPV i PCV) testirana je značajnost razlike promena navedenih parametara saturacije i gasnih analiza arterijske krvi (SpO_2, PaO_2, $PaCO_2$ i pH*)* između dva uzastopna vremena (lančano) Tukey-ovim testom; u funkciji vremena unutar grupa nije bilo statistički značajne razlike u vrednostima nijednog od navedenih parametara. Između grupa testirana je značajnost razlike promena navedenih parametara u funkciji vremena od t_1 do t_4 Studentovim *t*-testom. Primenom ovog testa zaključeno je da ni između grupa nema statistički značajne promene vrednosti parametara saturacije i gasnih analiza arterijske krvi.

Značajnost međusobnog uticaja PIP-a, C i $PETCO_2$

Značajnost međusobnog uticaja PIP, C i PetCO2 procenjivana je metodom višestruke regresione analize u obe posmatrane grupe. U grupi IPPV nađena je statistički visokoznačajna pozitivna korelacija između $PETCO_2$ i PIP (koeficijent B = 0,443; $p < 0,01$), i statistički značajna negativna ko-

Tabela 2
Poređenje vršnog inspiratornog pritiska (PIP), komplijanse (C) i vrednosti parcijalnog pritiska CO_2 na kraju ekspirijuma ($PETCO_2$) u okviru grupa u funkciji vremena

Vreme*	PIP		C		$PETCO_2$	
	IPPV	PCV	IPPV	PCV	IPPV	PCV
t_1–t_2	$p < 0,01$	$p < 0,01$	$p < 0,01$	$p < 0,05$	$p < 0,01$	n.s.
t_2–t_3	$p < 0,01$	$p < 0,05$	n.s.	n.s.	$p < 0,05$	n.s.
t_3–t_4	$p < 0,01$	$p < 0,01$	n.s.	n.s.	$p < 0,01$	$p < 0,05$

***t_1 – posle uvoda u anesteziju a pre kreiranja pneumoperitoneuma; t_2 – 5 min posle stvaranja pneumoperitoneuma; t_3 – tokom pneumoperitoneuma; t_4 – 5 min posle oslobađanja od pneumoperitoneuma.**
n.s. – nije značajno
IPPV – mehanička ventilacija prema tipu intermitentnog pozitivnog pritiska u vazdušnim putevima
PCV – mehanička ventilacija kontrolisana pritiskom

Tabela 3
Poređenje vršnog inspiratornog pritiska (PIP), komplijanse (C) i vrednosti parcijalnog pritiska CO_2 na kraju ekspirijuma ($PETCO_2$) između grupa po vremenima merenja

Vreme*	PIP		C		$PETCO_2$	
	IPPV	PCV	IPPV	PCV	IPPV	PCV
t_1	n.s.		n.s.		n.s.	
t_2	$p < 0,01$		$p < 0,05$		$p < 0,01$	
t_3	$p < 0,01$		$p < 0,05$		$p < 0,01$	
t_4	$p < 0,01$		$p < 0,05$		$p < 0,05$	

***t_1 – posle uvoda u anesteziju, a pre kreiranja pneumoperitoneuma; t_2 – 5 min posle stvaranja pneumoperitoneuma; t_3 – tokom pneumoperitoneuma;**
t_4 – 5 min posle oslobađanja od pneumoperitoneuma.
n.s. – nije značajno
IPPV – mehanička ventilacija prema tipu intermitentnog pozitivnog pritiska u vazdušnim putevima
PCV – mehanička ventilacija kontrolisana pritiskom

relacija između $P_{ET}CO_2$ i C (koeficijent B = -0,25; $p < 0,05$). U grupi PCV nije ustanovljena statistički značajna korelacija između $P_{ET}CO_2$ i PIP-a (koeficijent B = 0,037), kao ni između $P_{ET}CO_2$ i C (koeficijent B = -0,04).

Diskusija

Danas je sve više bolesnika sa postojećom preoperativnom respiratornom insuficijencijom, emfizemom ili hroničnom opstruktivnom bolešću pluća. Oni zbog toga pripadaju grupi III po ASA klasifikaciji. Ovakva konkomitantna respiratorna disfunkcija biće još više pogoršana ekstenzivnošću i specifičnostima laparoskopske holecistektomije. Zbog toga je od izuzetnog značaja da se utvrdi koji tip mehaničke ventilacije (IPPV ili PCV) u toku anestezije za laparoskopsku holecistektomiju dovodi od najmanjeg oštećenja respiratorne funkcije, o čemu, po podacima iz literature, još ne postoji saglasnost autora.

Naše istraživanje pokazalo je da u t_1 (posle uvoda u anesteziju, a pre kreiranja pneumoperitoneuma) nema statistički značajne razlike između grupa IPPV i PCV ni u jednom od ispitivanih parametara (PIP, C, $P_{ET}CO_2$). To je bilo očekivano zbog toga što je stvaranje pneumoperitoneuma insuflacijom ugljen-dioksida od strane hirurga jedan od ključnih elemenata specifičnih za laparoskopske intervencije koji negativno utiče na respiratornu funkciju, bez obzira na to koji tip mehaničke ventilacije prilikom opšte anestezije je primenjen.

U sledeća tri vremenska intervala (t_2 – 5 min posle stvaranja pneumo-peritoneuma; t_3 – tokom pneumoperitoneuma; t_4 – 5 min posle oslobađanja od pneumoperitoneuma) bilo je statitički značajne do visokoznačajne razlike u vrednostima ova tri respiratorna parametra između dve ispitivane grupe.

U grupi IPPV PIP se kretao od 19 cmH_2O do 26,8 cmH_2O, dok se u grupi PCV održavao u značajno manjem rasponu od 19 cmH_2O do 22 cmH_2O. Za bolesnike bez pratećih respiratornih oboljenja, a još više za bolesnike koji imaju izraženu respiratornu disfunkciju, bez obzira na poreklo, veoma je važno da maksimalni (vršni) pritisak u vazdušnim putevima i alveolama ne pređe određenu zadatu vrednost. Kada je ovaj pritisak povećan, a plućni parenhim već oštećen, postoji veća verovatnoća da će se razviti barotrauma pluća. Barotrauma praktično dovodi do kidanja zidova alveola. Ova disrupcija može progredirati do stvaranja bula i razvoja pneumotoraksa koji je komplikacija opasna po život. Model mehaničke ventilacije tipa PCV, dakle kontrolisane pritiskom, gde pritisak u vazdušnim putevima može da se ograniči, u našoj studiji pokazao se boljom varijantom.

U grupi IPPV komplijansa u intervalu od t_2 do t_4 bila je ispod 40 L/cmH_2O, dok je u grupi PCV bila preko 40 L/cmH_2O. Za sve bolesnike podvrgnute opštoj anesteziji bolje je da komplijansa pluća bude što veća. To omogućava bolju oksigenaciju krvi, razmenu gasova na alveolokapilarnoj membrani i usklađen odnos ventilacije i perfuzije u različitim regionima pluća što smanjuje intrapulmonalni šant (procenat krvi koji prolazi kroz neventilisane delove pluća i ostaje neoksigenisan). Naše istraživanje je pokazalo da je komplijansa pluća bila statistički značajno veća u grupi PCV, pa je i prema ovom respiratornom parametru to bolji modus mehaničke ventilacije kod bolesnika sa respiratornom disfunkcijom tokom laparoskopskih intervencija.

Parcijalni pritisak CO_2 na kraju ekspirijuma, koji se registruje kapnometrijom, veoma je značajan respiratorni parametar pri izvođenju opšte anestezije bez obzira na vrstu hirurške intervencije. Kapnometrija je jedan od najznačajnijih elemenata perioperativnog monitoringa. Mnogo je osetljivija od pulsne oksimetrije u detektovanju različitih poremećaja cirkulacije i ventilacije (npr. kod plućne tromboembolije promene očitavanja kapnometrije se javljaju pre promene očitavanja pulsne oksimetrije, a tada je za spašavanje bolesnika od ove smrtonosne komplikacije važan svaki sekund). Još veći značaj kapnometrija, odnosno registrovanje $P_{ET}CO_2$, ima u toku anestezije za laparoskopske intervencije gde se aktivno insuflira CO_2 zbog stvaranja pneumoperitoneuma. U našem istraživanju, u IPPV grupi $P_{ET}CO_2$ je bio viši 30 mmHg u svim vremenskim intervalima, dok je u PCV grupi bio niži od 30 mmHg. Sve navedene vrednosti su prihvatljive. Ono o čemu treba voditi računa je da vrednost ne bude veća od 40 mmHg ako peritonealna insuflacija CO_2 poveća produkciju a smanji funkcionalni rezidualni volumen [4].

Eren i sar. [5] istraživali su efekat modusa mehaničke ventilacije na repiratornu mehaniku tokom laparoskopskih holecistektomija. Modus IPPV smatraju konvencionalnim, a PCV alternativnim. To je u skladu sa praksom u našoj zemlji gde se najčešće primenjuje IPPV mehanička ventilacija. Njihova studija je obuhvatila 30 bolesnika podvrgnutih elektivnoj laparoskopskoj holecistektomiji koji su podeljeni u dve grupe prema načinu mehaničke ventilacije. U PCV grupi, PIP se nije promenio (povećao) nakon stvaranja pneumoperitoneuma za razliku od IPPV grupe što je u skladu sa našim rezultatima. Nasuprot našim rezultatima, komplijansa se u obe grupe smanjila nakon kreiranja pneumoperitoneuma i između grupa nije bilo statistički značajne razlike.

Danas mehanička ventilacija tokom anestezije za laparoskopske procedure dobija na značaju zbog toga što se sve više hirurških intervencija obavlja na ovaj način. Međutim, ono što hirurzima olakšava rad, anesteziolozima često otežava. Pneumoperitoneum je neophodan, ali kompromituje respiratornu funkciju i kod zdravih, često gojaznih bolesnika. Osim toga, većina hirurških procedura traje mnogo duže kada se izvode laparoskopski pa se bolesnik u dužem vremenskom periodu izlaže kombinovanim neželjenim efektima [6]. Stvaranje pneumoperitoneuma pri intra-abdominalnom pritisku (IAP) od 10 do 15 mmHg smanjuje komplijansu pluća i kod bolesnika bez respiratornih poremećaja [7].

U našem istraživanju, parametri saturacije krvi kiseonikom i elementi gasnih analiza arterijske krvi (saturacija arterijske krvi kiseonikom utvrđena pulsnom oksimetrijom – SpO_2, parcijalni pritisci kiseonika – PaO_2 i ugljen-dioksida – $PaCO_2$ u arterijskoj krvi i pH arterijske krvi) nisu se statistički značajno razlikovali ni unutar grupa, ni između grupa. Do respiratornog ili mešovitog acido-baznog disbalansa može doći zbog CO_2 pneumoperitoneuma, koji smanjuje pH ka acidozi. Jedna studija je pokazala da snižavanje insuflacionog pritiska sa 15 na 10 mmHg nije doprinelo eliminaciji acido-baznih poremećaja [8].

Jedno od mogućih objašnjenja za činjenicu da PCV modus mehaničke ventilacije, koji značajno popravlja respiratorne parametre, u našem istraživanju nije poboljšao parametre saturacije i oksigenacije u odnosu na konvencionalnu IPPV ventilaciju leži u tome što nijedan modus mehaničke ventilacije nije primenjen sa pozitivnim end-ekspiratornim pritiskom (PEEP).

Postoje fiziološke promene koje su posledica CO_2 pneumoperitoneuma i položaja bolesnika tokom laparoskopskih intervencija. Naime, CO_2 pneumoperitoneum ispoljava svoje fiziološke efekte putem dva različita mehanizma. Prvi je fokusiran na mehaničke efekte povezane sa povećanim intraperitonealnim pritiskom, a drugi za hemijske efekte CO_2 kao gasa koji se koristi za insuflaciju. Pneumoperitoneum dovodi do povećanja intra-abdominalnog pritiska koji ima za posledicu elevaciju dijafragme. Ovo rezultuje kolapsom alveola bazalnih delova pluća što dovodi do smanjenja funkcionalnog rezidualnog kapaciteta, nesklada ventilaciono-perfuzionog odnosa (V/Q) i povećanja intrapulmonalnog šantovanja krvi. Konačno, javlja se hipoksemija i povećan alveolarno-arterijski kiseonički gradijent – (A-a)DO_2. Sve ove promene su još mnogo izraženije kod bolesnika sa konkomitantnim respiratornim oboljenjima. Pozitivan pritisak u vazdušnim putevima na kraju ekspirijuma (PEEP) ima više korisnih efekata u ovom kliničkom scenariju: povećava funkcionalni rezidualni kapacitet tako što će povećati volumen alveola ("obnavlja" alveole); povećava komplijansu pluća; sprečava prevremeno zatvaranje malih vazdušnih puteva što je veoma bitno jer je potreban mnogo veći pritisak od normalnog da bi se oni ponovo otvorili; i smanjuje intrapulmonalni šant [9, 10].

U jednoj od najnovijih studija [11] istraživači su ispitivali dejstvo blagog PEEP-a od 5 cmH_2O, primenjenog uz PCV modus mehaničke ventilacije, na parametre ventilacije i oksigenacije tokom pneumoperitoneuma. Trideset bolesnika, podvrgnutih laparoskopskoj holecistektomiji, je randomizovano u dve grupe. Prva je ventilrana primenom modusa PCV, ali bez dodatnog PEEP-a, (PEEP je bio 0 cmH_2O), a druga po istom modusu ventilacije uz PEEP od 5 cmH_2O. Rezultati su pokazali da je indeks oksigenacije (PaO_2/FiO_2 – u ovom indeksu FiO_2 predstavlja inspiratornu frakciju kiseonika) bio značajno veći u grupi sa PEEP-om. Autori su zaključili da PEEP od 5 cmH_2O treba da se primenjuje uz PCV mehaničku ventilaciju tokom laparoskopskih procedura da bi se smanjila intraoperativna atelektaza pluća do koje dovodi pneumoperitoneum i da bi se poboljšala razmena gasova na alveolokapilarnoj membrani i oksigenacija krvi.

I druge studije su pokazale korisne efekte PEEP-a. Maracajá-Neto i sar. [12] su poredili respiratornu mehaniku kod dve grupe pacijenata podvrgnutih laparoskopskoj holecistektomiji: sa PEEP-om od 10 cmH_2O i bez PEEP-a. Autori su zaključili da PEEP od 10 cmH_2O ublažava efekte pneumoperitoneuma i poboljšava respiratornu mehaniku. Drugi autori su PEEP-u dodali manevre "obnavljanja" alveola i to tako što su bolesnici ventilisani manuelno do pritiska u vazdušnim putevima od 40 cmH_2O tokom deset respiratornih ciklusa u jednoj minuti, a onda su vraćani na mehaničku ventilaciju sa PEEP-om od 5 do 10 cmH_2O [13]. Zaključili su da je ovaj manevar koristan zbog toga što poboljšava arterijsku oksigenaciju. Ova saznanja su naročito korisna za modele ventilacije kod morbidno gojaznih bolesnika koji se podvrgavaju laparoskopskim barijatrijskim procedurama [14, 15].

Značaj ispitivanja modusa mehaničke ventilacije kod laparoskopskih intervencija sa vremenom biva sve veći, primarno zbog toga što se sve više hirurških procedura izvodi laparoskopski, što one sve duže traju i što im se podvrgavaju bolesnici sa sve težim konkomitantnim oboljenjima. To predstavlja veliki izazov za anesteziologa jer adekvatno izvedena mehanička ventilacija predstavlja jedan od vidova zaštite organizma od negativnih efekata pneumoperitoneuma koji ne ometa samo respiratornu funkciju nego deluje i na različite sisteme organa. Sistemsko dejstvo specifičnih elemenata laparoskopske hirurgije najbolje se ogleda preko smanjenja saturacije kiseonikom arterijske krvi. U nekim organima to može biti potencijalno veoma opasno npr, u slučaju gastrične mukoze, jer predstavlja okidač za sistemski inflamatorni odgovor koji je najčešće štetan po bolesnika [16].

Da je budućnost u laparoskopskoj hirurgiji za mnoge procedure potvrđuje i istraživanje koje se fokusiralo na mogućnost izvođenja ovakvih intervencija u beztežinskom stanju, u svemiru [17]. Od 1987. godine, kada je prvu uspešnu laparoskopsku holecistektomiju izveo Filip More, ove procedure su postale zlatni standard, koji bolesnicima pruža mnoge pogodnosti. Zbog toga je zadatak anesteziologa da obezbedi sigurnost bolesnika [18] i, koliko je god moguće neutrališe negativne efekte pneumoperitoneuma i položaja bolesnika koji su specifični za laparoskopske intervencije.

Zaključak

Mehanička ventilacija tipa PCV obezbeđuje bolje intraoperativne parametre ventilacije tokom izvođenja LH kod bolesnika koji pripadaju grupi III prema ASA klasifikaciji zbog pratećih respiratornih oboljenja.

L I T E R A T U R A

1. *Hedenstierna G.* Causes of Oxygenation Impairment During Anesthesia. In:Vincet JL, editor. Yearbook of Intensive Care and Emergency Medicine. Berlin, Heidelberg, New York: Springer; 2000; pp. 343−51.
2. *Bapoje SR, Whitaker JF, Schulz T, Chu ES, Albert RK.* Preoperative evaluation of the patient with pulmonary disease. Chest 2007; 132(5): 1637−45.
3. *Qaseem A, Snow V, Fitterman N, Hornbake ER, Lawrence VA, Smetana GW,* et al. Clinical Efficacy Assessment Subcommittee of the American College of Physicians. Risk assessment for and strategies to reduce perioperative pulmonary complications for patients undergoing noncardiothoracic surgery: a guideline from the American College of Physicians. Ann Intern Med 2006; 144(8): 575−80.
4. *Tanaka T, Satoh K, Torii Y, Suzuki M, Furutani H, Harioka T.* Arterial to end-tidal carbon dioxide tension difference during laparoscopic colorectal surgery. Masui 2006; 55(8): 988−91. (Japanese)
5. *Eren G, Cukurova Z, Hergunsel O, Tas B, Leblebici H, Emir N,* et al. Effects of modes of ventilation on respiratory mechanics

during laparoscopic cholecystectomies: A-263. Eur J Anaesthesiol 2006; 23: 69.
6. *Valenza F, Chevallard G, Fossali T, Salice V, Pizzocri M, Gattinoni L.* Management of mechanical ventilation during laparoscopic surgery. Best Pract Res Clin Anaesthesiol 2010; 24(2): 227–41.
7. *Rauh R, Hemmerling TM, Rist M, Jacobi KE.* Influence of pneumoperitoneum and patient positioning on respiratory system compliance. J Clin Anesth 2001; 13(5): 361–5.
8. *Sefr R, Puszkailer K, Jagos F.* Randomized trial of different intra-abdominal pressures and acid-base balance alterations during laparoscopic cholecystectomy. Surg Endosc 2003; 17(6): 947–50.
9. *O'Malley C, Cunningham AJ.* Physiologic changes during laparoscopy. Anesthesiol Clin North America 2001; 19(1): 1–19.
10. *Andersson LE, Bååth M, Thörne A, Aspelin P, Odeberg-Wernerman S.* Effect of carbon dioxide pneumoperitoneum on development of atelectasis during anesthesia, examined by spiral computed tomography. Anesthesiology 2005; 102(2): 293–9.
11. *Kim JY, Shin CS, Kim HS, Jung WS, Kwak HJ.* Positive end-expiratory pressure in pressure-controlled ventilation improves ventilatory and oxygenation parameters during laparoscopic cholecystectomy. Surg Endosc 2010; 24(5): 1099–103.
12. *Maracajá-Neto LF, Verçosa N, Roncally AC, Giannella A, Bozza FA, Lessa MA.* Beneficial effects of high positive end-expiratory pressure in lung respiratory mechanics during laparoscopic surgery. Acta Anaesthesiol Scand 2009; 53(2): 210–7.
13. *Pang CK, Yap J, Chen PP.* The effect of an alveolar recruitment strategy on oxygenation during laparascopic cholecystectomy. Anaesth Intensive Care 2003; 31(2): 176–80.
14. *Whalen FX, Gajic O, Thompson GB, Kendrick ML, Que FL, Williams BA,* et al. The effects of the alveolar recruitment maneuver and positive end-expiratory pressure on arterial oxygenation during laparoscopic bariatric surgery. Anesth Analg 2006; 102(1): 298–305.
15. *Talab HF, Zabani IA, Abdelrahman HS, Bukhari WL, Mamoun I, Ashour MA,* et al. Intraoperative ventilatory strategies for prevention of pulmonary atelectasis in obese patients undergoing laparoscopic bariatric surgery. Anesth Analg 2009; 109(5): 1511–6.
16. *Schwarte LA, Scheeren TW, Lorenz C, De Bruyne F, Fournell A.* Moderate increase in intraabdominal pressure attenuates gastric mucosal oxygen saturation in patients undergoing laparoscopy. Anesthesiology 2004; 100(5): 1081–7.
17. *Kirkpatrick AW, Keaney M, Hemmelgarn B, Zhang J, Ball CG, Groleau M,* et al. Intra-abdominal pressure effects on porcine thoracic compliance in weightlessness: implications for physiologic tolerance of laparoscopic surgery in space. Crit Care Med 2009; 37(2): 591–7.
18. *Srivastava A, Niranjan A.* Secrets of safe laparoscopic surgery: Anaesthetic and surgical considerations. J Minim Access Surg 2010; 6(4): 91–4.

Primljen 31. XII 2010.
Prihvaćen 15. VII 2011.

ORIGINAL ARTICLE

UDC: 355.23:[613.71/.73:796.015
DOI: 10.2298/VSP1301016M

The effectiveness of physical education of the Military Academy cadets during a 4-year study

Efikasnost fizičkog vaspitanja kadeta Vojne akademije tokom četvorogodišnjih studija

Lela Marić*, Branko Krsmanović†, Tatjana Mraović‡, Aleksandra Gogić§, Jelena Sente‖, Miroslav Smajić†

***Military Academy, University of Defence, Belgrade, Serbia; †Faculty of Sport and Physical Education, University of Novi Sad, Novi Sad, Serbia; ‡Institute of Hygiene, §Institute for Scientific Information, Military Medical Academy, Belgrade, Serbia; ‖Underwater Activities Club "Sirmium", Sremska Mitrovica, Serbia;**

Abstract

Background/Aim. The main role of physical education is health and educational practices of cadets and all-round personality development. Instruction executing is successful only when the set requirements are realized. The aim of this study was to evaluate the effectiveness of physical education in order to rise physical capabilities of the Military Academy cadets during a 4-year education. **Methods.** The study was conducted in the Military Academy, Belgrade. A total of 120 cadets who at the beginning of the study were 19 years ± 6 months and at the end 23 years ± 6 months were included in this study. The study used the following tests for verification and assessment of physical fitness: pull-ups, lifting the trunk from the ground, standing long jump seats, running at 1,600 m and overcoming the infantry obstacles. The data were analyzed using statistical programs to calculate the central and dispersion parameters. The difference in the achieved results in the individual variables were evaluated by the univariate analysis of variance (ANOVA), while the differences in the system variables by region were identified by the multivariate analysis of variance (MANOVA) and discriminant analysis. The group membership was determined using profile analysis. **Results.** There were statistically significant differences in all the tests to evaluate the effectiveness of physical education during a 4-year study, except in the standing long jump test. The best average results in motor capabilities tests, were achieved after two years of study, while in the endurance tests showed the best results achieved at the end of a 4-years studying. **Conclusion.** The results of overcoming specific tests for the physical abilities of the Military Academy cadets show that the physical education curriculum only slightly improves the development of physical skills of cadets during a 4-year study. The existing program shows the best results in the pull-ups test of the ground troops, and the worst in the multiple motor control tests (endurance, strength and speed).

Key words:
military personnel; education; physical education and training; program evaluation.

Apstrakt

Uvod/Cilj. Osnovna uloga fizičkog vaspitanja je ostvarivanje zdravstvenog i vaspitnog delovanja na kadete i formiranje svestrane ličnosti. Realizacije nastave je uspešna samo kada su ostvareni postavljeni zahtevi. Cilj ovog rada bio je procena efikasnosti nastavnog programa fizičkog vaspitanja u postizanju povećanja fizičke sposobnosti kadeta Vojne akademije u toku četvorogodišnjeg školovanja. **Metode.** Istraživanje je sprovedeno u Vojnoj akademiji u Beogradu i obuhvatilo je 120 kadeta koji su na početku školovanja imali 19 godina ± 6 meseci, a na kraju školovanja 23 godine ± 6 meseci. Testovi za proveru i ocenjivanje fizičke pripremljenosti bili su: zgibovi na vratilu, dizanje trupa sa tla, skok u dalj iz mesta, trčanje na 1 600 m i savladavanje pešadijskih prepreka. Podaci su obrađeni primenom statističkih programa za izračunavanje centralnih i disperzionih parametara. Za utvrđivanje razlika između postignutih rezultata tokom školovanja korišćena je univarijantna analiza varijanse (ANOVA), a razlike u sistemu varijabli po prostorima utvrđene su multivarijantnom analizom varijanse (MANOVA) i diskriminativnom analizom. Pripadnost grupi određena je analizom profila. **Rezultati.** Uočene su statistički značajne razlike u svim testovima za procenu efikasnosti nastave fizičkog vaspitanja tokom četvorogodišnjeg školovanja, osim u testu skok udalj iz mesta. Najbolji prosečni rezultati u testovima za procenu motoričkih sposobnosti postignut je posle druge godine stu-

Correspondence to: Lela Marić, Military Academy, University of Defence, Belgrade, Serbia; Phone: +381 64 217 63 90.
E-mail: mariclela28@yahoo.com

dija, dok su u testovima za procenjivanje izdržljivosti najbolji rezultati postignuti na kraju četvorogodišnjeg školovanja. **Zaključak.** Rezultati u savladavanju specifičnih testova za procenu fizičke sposobnosti kadeta Vojne akademije pokazuju da nastavni program fizičkog vaspitanja samo donekle poboljšava razvoj fizičkih sposobnosti kadeta tokom četvorogodišnjeg školovanja. Postojećim programom najbolji rezultati postižu se u testu dizanje trupa sa tla, a najslabiji u testovima koji obuhvataju više motoričkih sposobnosti (izdržljivost, snagu i brzinu).

Ključne reči:
vojni kolektiv; obrazovanje; fizičko vaspitanje i trening; procena, istraživanja.

Introduction

Human factor and physical fitness of the members of armed forces have always been and will remain one of the most important goals and qualities of the national defense in the armed forces worldwide, regardless high performance and development of weapons technology. Previous experience in our history suggests that physical training was of great importance, through training of both soldiers and officers for successful command and control [1–2]. This confirms that physical education now plays an important role in the education of Military Academy cadets, under the specific conditions of life and work in a military school. The curricula and other normative acts regulate the implementation of training and education, by setting goals and tasks of the subjects, contents, number of classes for each thematic area and the guidelines for implementation.

The main role of physical education is health and educational influence on the cadets and versatile personality development trained for combat operations. In the "Guidelines for physical training in the military", specific goals have been set: to achieve and maintain a high level of physical fitness of cadets, to train them to organize and perform physical training with soldiers and units, to build awareness of the importance of physical fitness of personnel in preparation and execution of combat operations, preservation of health and improvement of work activities during the service [3].

Teaching physical education in the Military Academy of the Armed Forces of Serbia has not so far been the subject of a more extensive research based on empirical methods. There have been some records on the officers in Serbia teaching gymnastics, the connection of gymnastics with military exercises and the like, but without finding a casual link of such a situation [4]. Physical education is reflected in the specific physical exercises, depending on the profession or workplace. Professional working ability is defined as the ability to perform different activities, determined by the requirements of a workplace. Each profession requires some knowledge and skills, and in some cases, predispositions [5]. The aim of this study was to evaluate the effectiveness of physical education in order to increase physical capabilities of the Military Academy cadets during a 4-year education.

Methods

In this longitudinal study the examinees were compared and monitored in terms of motor military skills during their 4-year training. When recruiting candidates to the Military Academy, all the examinees passed the appropriate medical and psychological examinations. Medical examinations and physical fitness tests were carried out each within the study.

The research was conducted in the Military Academy, Belgrade. A total of 120 cadets, aged 19 years ± 6 months at the beginning of training, were included and monitored throughout their studies up to the age of 23 years ± 6 months upon graduation. The effectiveness of teaching was evaluated by the level of achievement in doing specific tests arising from the content of the curriculum for physical education in the Military Academy. During the school year, physical education is taught by two regular physical education classes and 2 h in sports day. During a 4-year study, at the end of each school year, checking the physical fitness of students is done, based on five motor tests, to assess situational motor skills including: pull-ups, performed for 60 sec with a range from a minimum of 3 to a maximum of 15 repetitions sit-ups, for 60 sec a range of recurrence from 25 to 50 standing long jump, in three attempts to perform a jump in the range from 183 cm to 287 cm a 1,600-meter-run (1,600) need to run out in time of 320 sec to 450 sec and obstacle course (OC) to be overcome in time from 80 sec to 175 sec [3].

The obtained data were analyzed using statistical programs to calculate the central and dispersion parameters: arithmetic mean ($\bar{x}$), standard deviation (SD), variance (Sig), minimum score (Min), maximum score (Max), standard error (SE), coefficient of variation (CV%). The differences between individual years of training during the four years in some variables were determined by the use of univariate analysis of variance (ANOVA) and differences in the system of variables by regions were determined by the multivariate analysis of variance (MANOVA) and discriminant analysis. The group membership was estimated by profile analysis [6].

Results

The specific motor competence of the group of examinees after the first year of training is fairly homogeneous (Table 1), except for the obstacle course test. The scores tests ranged from a minimum of 89 sec to a maximum of 441 sec, which affected the normal distribution of values (CV 26.82%). The minimum and maximum scores in the events standing long jump (from 170 cm to 265 cm) and a 1,600-meter-run (from 332 sec to 490 sec) also indicated some differences, but they did not affect the normal distribution (CV%). Based on the observed individual differences, a statistically significant difference was found only in the variable obstacle course ($p = 0.000$).

Analyzing the results of the examinees by the use of mean values after the second year, there is a significant inch,

Table 1

Central and dispersion parameters per year of trening in the tests obstacle course (OC), standing long jump (SLJ) and 1,600-meter-run (1,600)

Years of training	OC (sec) $\bar{x} \pm SD$ (min–max)	CV%	SLJ (cm) $\bar{x} \pm SD$ (min–max)	CV%	1,600 m (sec) $\bar{x} \pm SD$ (min–max)	CV%
1st	139.7 ± 37.5 (89.0–441.0)	26.82	224.1 ± 17.9 (170.0–265.0)	8.00	405.7±30.4 (332.0–490.0)	7.49
2nd	131.2 ± 26.9* (93.0–320.0)	20.53	227.3 ± 17.7 (170.0–265.0)	7.79	385.0 ± 37.8 † (305.0–550.0)	9.82
3rd	143.7 ± 24.3 (95.0–270.0)	16.90	222.7 ± 17.4 (180.0–265.0)	7.81	410.1 ± 31.4 (336.0–530.0)	7.65
4th	140.2 ± 23.6 (91.0–235.0)	16.87	227.0 ± 18.3 (183.0–265.0)	8.09	407.3 ± 31.3 (322.0–450.0)	7.70

Set norms: OC to 176 sec; SLJ 183–287 cm; 1,600 320–450 sec; CV% – coefficient of variation

***p* < 0.007; †*p* < 0.000 (ANOVA)**

except for the variable standing long jump, where the minimum and maximum values are identical to those achieved after the first year of training. The coefficient of variation indicates no significant deviation from the arithmetic mean in the variables standing long jump (7.79%) and a 1,600-meter-run (9.82%). A slightly higher coefficient of variation in the obstacle course test indicates some differences in individual values, but they did not affect the normal distribution. Group heterogeneity in obstacle course is slightly lower than values after the first year of training (89–441 sec) and ranges from a minimum of 93 sec to a maximum of 320 sec.

Analyzing the results of the examinees by the use of mean values after the third year of training, shows a significant decrease in values as compared to the second year of training. Based on the coefficient of variation, heterogeneity of the group of examinees is slightly higher than in the previous two years in the obstacle course test (16.90%). But individual differences in the obstacle course do not affect the normal distribution of values after the third year of training, as well. The values range from a minimum of 95 sec to a maximum of 270 sec. Comparing these to the values after the second year of training, the maximum values are lower by 50 sec. The values of specific motor skills range within the limits of normal distribution in all of the three tests. The observed individual differences among examinees did not affect the normal distribution.

In terms of mean values after the fourth year of training a slight increase in all the three variables compared to the third year values is noticed. The homogeneity of the examinees was observed in all the three tests, and based on the coefficient of variation. Individual differences in the test obstacle course do not affect the normal distribution of values, ranging from a minimum of 91 sec to a maximum of 235 sec. Comparing these to the values after the first year of training, the maximum values are lower by about 200 sec. Minimum values in the standing long jump (183.0 cm) have higher values than in the previous three years. Physical competence during the 4-year training in the variables pull-ups and sit-ups was determined by measuring the number of repetitions achieved for one minute.

Table 2 shows the number and percentage of the examinees per year of training in relation to the pull-ups test. The largest number of the examinees in the first year of training, 45 (37.5%) of 120, were classified into the group with the number of repetitions from 4 to 6 pull-ups. However, after the third year of training, most examinees, 29 (24.2%) out of 120, were classified in the group with repetitions from 4 to 6 pull-ups. It is noticeable that 24 (20.0%) examinees were classified into the group with the number of repetitions over 14 pull-ups, which was higher than the values of the first and second year. In the fourth year of training, most of the examinees, 29 (24.2%) of them, were classified in the group with the number of repetitions over 14 pull-ups. By analyzing the number and the percentage of the examinees, an increase in dynamic strength of arms and shoulders after each year of training was observed.

In the sit-ups test within the time limit of 60 sec, the largest number of the examinees in the first year of training, 44 (36.7%) of 120, was classified into the group with the number of repetitions up to 49. In the fourth years of training, the majority of the examinees was in the group with 50 repetitions.

Analysis of the central and dispersion parameters of the examinees shows numerical differences in the average values in some variables for the assessment of motor competence during training. Multivariate analysis of variance showed a statistically significant difference among the examinees during training in relation to motor variables criterion (p = 0.000).

Analysis of individual values (Table 1), by the univariate analysis of variance shows a statistically significant difference among the examinees during training in the obstacle course variables (p = 0.007) and a 1,600-meter-run (p = 0.000). The results indicate that the examinees were at different levels of preparation in these two criterion variables that can be influenced by exercise. A statistically significant difference was not found in the test standing long jump during training.

Assessment of motor competence during training with no parametric values (Table 2), but with categorical data, in this part of the study shows numerical differences in average values per years, and were processed using the Roy's χ^2 test. The multivariate analysis of variance showed a statistically significant difference among the examinees during training (p = 0.000), in the pull-ups and sit-ups within the time limit of 60 sec. The estimating individual values by the univariate

Table 2

Distribution of cadets regarding the number of repetitions in the tests within the time limit of 60 sec pull-ups and sit-ups per year of trening

Number of repetitons	Years of trening			
	1st n (%)	2nd n (%)	3rd n (%)	4th n (%)
Pull-ups [1]				
up to 3	20 (16.7)	21 (17.5)	26 (21.7)	23 (19.2)
from 4 to 6	45 (37.5)	27 (22.5)	29 (24.2)	24 (20.0)
from 7 to 9	24 (20.0)	27 (22.5)	22 (18.3)	21 (17.5)
from 10 to 13	15 (12.5)	23 (19.2)	19 (15.8)	23 (19.2)
over 14	16 (13.3)	22 (18.3)	24 (20.0)	29 (24.2*)
Sit-ups [2]				
from 25 to 49	44 (36.7)	17 (14.2)	26 (21.7)	11 (9.2)
50	24 (20.0)	67 (55.8)	55 (45.8)	89 (74.2†)
from 51 to 53	19 (15.8)	13 (10.8)	12 (10.0)	8 (6.7)
from 54 to 56	17 (14.2)	14 (11.7)	14 (11.8)	8 (6.7)
over 57	16 (13.3)	9 (7.5)	13 (10.8)	4 (3.3)

[1] Requested norms: 3–15; [2] Requested norms: 25–50

***$p < 0.003$; †$p < 0.000$ (ANOVA)**

tests analysis of variance showed a statistically significant difference among the examinees in the pull-ups tests (p = 0.003) and sit-ups within the time limit of 60 seconds (p = 0.000). The results indicate that the examinees were at different levels of preparation and ability assessed by these tests, but this can be influenced by exercise. The homogeneity of the group of examinees was the largest and identical after finishing the first and fourth year of training, 89 examinees out of 120 had the characteristics of their group (74.17%). The lowest homogeneity was observed after the third year of training, 56 examinees out of 120 had the characteristics of their group (46.67%).

Discussion

The effectiveness of physical education of the Military Academy cadets was evaluated on the basis of their attainment of specific motor skills during a 4-year study, based on physical education curriculum, which develops explosiveness, strength and endurance. The effectiveness of this program is estimated at the end of each year during the training of cadets through the five tests, pull-ups, sit-ups, standing long jump, 1,600-meter-run and obstacle course. These tests assess motor abilities (strength, speed, explosiveness, endurance, agility and coordination).

Unlike our program physical abilities checks of the cadets in the Military Academy, the U.S. military use tests – APFT: push-ups with the time limit of 2 min (35–100), sit-ups with the time limit of 2 min (47–97) and a timed two-mile run (16:30) and IOCT test (indoor obstacle course test including 10 obstacles) [7]. This program is made in the Department of Physical Education in the United States Military Academy and is aimed at developing and maintaining a high standard of physical strength, agility and endurance of the cadets, necessary to meet the needs they faced in the military service.

Individual differences among the examinees in the values of specific motor skills obtained in our study, especially in the variable obstacle course, may be due to insufficient training to perform this complex test, and the lack of individual preparation of cadets over the previous period of training [8]. The cadets' results after the second year of training in the motor skills stated above indicate their better preparation and training [9]. The cause of small individual differences in the tests may be due to better attainment of motor skills which require a high level of ability. Poorer values achieved after the third year, compared to these of the second year, may be due to the development of motor skills that have certain regularities, such as heterochrony, stageness, phaseness and transfer in developing ability [10]. It is known that oriented development of motor skills with a relatively prolonged constant load leads to the reduced effects of actions. Analyzing the results of specific motor abilities of cadets after the fourth year of training, higher values compared to the previous year were observed. During training, the results of the test for the assessment of explosive power, standing long jump, indicate that there are no statistically significant differences in the values. On the basis of mean values, the ranging from 223 cm to 227 cm, it is possible that the explosive power is more genetically caused [11–12]. It is evident that the number of examinees with the maximum results in the sit-ups and pull-ups tests within the time limit of 60 sec varies by years of training. After the fourth year of training, over 74% of the examinees were in the group with 50 repetitions in the sit-ups tests, and in the pull-ups test, over 24% of the increased the number of those over 14 repetitions.

It should be noted that the program for any of the four years is the same. It should be noted, also that among the examinees there were those who did not meet the criteria of the tests at the end of the school year, but they managed to do that in the subsequent examination periods before the new school year. After the fourth year, the examinees achieved the required results in the period before their promotion to the rank of lieutenant. The reason may be a better psychophysical readiness and motivation for the final part of the exam, because after four years of training, within the next two months, the examinees graduate and are promoted into the professional members of the Armed Forces of Serbia.

Conclusion

The results obtained at the end of each year training vary within the norms required for the assessment of physical abilities of the Military Academy cadets. The planned program is satisfactory, as far as the set standards, but is insufficient to achieve maximal results. The values of the tests performed might be a consequence of genetic predisposition of cadets, less motor engagement in the last two years of the study or less motivation of cadets. With the existing program, the best results are achieved in the test for pull-ups of the ground troops, and the worst in the multiple motor control tests (endurance, strength and speed).

REFERENCES

1. *Rodić N.* The influence of basic training on physical fitness. Belgrade: Novi glasnik; 1993. (Serbian)
2. *Rodić N.* A new program for physical training of soldiers. Belgrade: Novi glasnik; 1994. (Serbian)
3. Instructions for physical training in the Armed Forces of Yugoslavia. Beograd: Army General Staff of Yugoslavia; 1995. (Serbian)
4. *Perić D.* Teaching physical education at the Military Academy of Land Forces from its establishment until disintegration of Yugoslavia (1850–1990) [thesis]. Belgrade: Faculty of Physical Education; 2000. (Serbian)
5. *Reljić J.* Physical education as training for professional occupations. Belgrade: Faculty of Physical Education; 1981. (Serbian)
6. *Perić D.* Statistics used in sport and physical education. Belgrade: Faculty of Physical Education: Idea Belgrade; 2001. (Serbian)
7. Department of Physical Education at the United States Military Academy 2007. Available from: http://www.usma.edu/physical.asp [cited 2009 December 5].
8. *Marić L.* The effectiveness of physical education at the Military Academy on various professional commitments with regard to the working conditions [thesis]. Novi Sad: Faculty of Physical Education; 2005. (Serbian)
9. *McKenzie TL, Sallis JF, Prochaska JJ, Conway TL, Marshall SJ, Rosengard P.* Evaluation of a two-year middle-school physical education intervention: M-SPAN. Med Sci Sports Exerc 2004; 36(8): 1382−8.
10. *Krsmanović B, Berković L.* Theory and methods of physical education. Novi Sad: Faculty of Physical Education; 1999. (Serbian)
11. *Marić L., Krsmanović B.* Anthropometric characteristics and motor abilities of the Military Academy cadets. Glasnik Antropološkog društva Jugoslavije; 2007. p. 199−206. (Serbian)
12. *Marić L., Krsmanović B.* The influence of motor abilities on motor efficiency of cadets of the Military Academy. Časopis Sportske akademije Crne Gore 2008. No 15, 16, 17/ IV, p. 317−22. (Serbian)

Received on January 14, 2011.
Revised on May 10, 2011.
Accepted on July 12, 2011.

ORIGINAL ARTICLE

UDC: 616.314-08-7:615.46
DOI: 10.2298/VSP1301021I

The flow of two zinc oxide-eugenol-based endodontic sealers

Napon tečenja dva cink-oksid eugenolna endodontska silera

Dragan V. Ilić

Faculty of Dental Medicine, Clinic of Restorative Dentistry and Endodontics, Belgrade, Serbia

Abstract

Background/Aim. Endodontic sealers (ES) for obturation are usually prepared with a slight variation of their components both on purpose or unintentionally. Considering that fact, as well as a frequent use of compaction techniques with the applied force to gutta-percha and ES of 1–3 kg, the aim of this study was to investigate the flow of two zinc-oxide eugenol ES in regard to the applied force and a variation of sealer's components. **Methods.** The experimental group samples of both ES were prepared according to the manufacturer's instructions, applied between pair of glass slabs and loaded by weights of 1 and 2 kg, respectively (American National Standard, Specification No. 57). Some samples of one ES were prepared as thick consistency with 10% more powder and some as thin mixture with 10% less powder than the standard prescription. These semples had been exposed to the load of 2 kg. The control group included samples of both ES prepared as standard prescription but exposed to the weight of one glass slab only. The spread ES appeared as a regular circle 10 min upon mixing and weighting. Measuring of the circle diameter was done by an orthodontic ruler. The flow of the used ES was considered the function of its spread diameter. **Results.** Application of 1 *vs* 2 kg load for both regularly mixed sealers in the scope of disk diameter (flow) was statistically insignificant ($p > 0.05$). This means that the stated null hypothesis that there would be no significant difference in flow rate among the regularly mixed sealers at the level of $\alpha = 0.05$ is accepted. The findings about difference in the disk diameter in regard to mixing variation of Endomethasone indicate that the null hypothesis that there would be no significant difference in flow rate between the regular and thick mixed mass at the level of $\alpha = 0.05$ is accepted. In the comparison of regular and thin mix a significant difference was noted and the null hypotesis is rejected ($p < 0.01$). The control group results displayed Roth 801 as less viscous than Endomethasone sealer ($p < 0.01$). **Conclusion.** Application of 1 or 2 kg pressure on the samples of both exposed sealers does not significantly affect the flow values as well as comparison of the regular to thick consistency of Endomethasone while comparison of its regular to thin mass shows a significant difference.

Key words:
root canal filling materials; zinc oxide-eugenol cement; rheology; viscosity.

Apstrakt

Uvod/Cilj. Endodontski sileri (ES) za opturaciju kanala korena zuba često se u praksi pripremaju sa varijacijama svojih komponenti. Uzimajući u obzir ovo, kao i činjenicu da se sve češće koriste metode kompakcije gutaperke i ES sa primenjenim pritiscima od 10 do 30 N, cilj ovog rada bio je ispitivanje napona tečenja (*flow*) dva ES na bazi cink-oksid eugenolnih silera. U tom smislu je planirano ispitivanje promene napona tečenja kod ES sa i bez primene opterećenja kao i sa minimalnim odstupanjima gustine materijala od regularno zamešane preskripcije (gušća i ređa konzistencija). **Metode**. U eksperimentalnoj grupi uzorci dva cink-oksid eugenolna ES pripremljeni su prema uputstvu proizvođača, a zatim nanešeni između staklenih pločica i opterećeni tegovima od 1 i 2 kg (American National Standard, Specification No.57). Deo uzorka jednog silera bio je pripremljen kao gušća i ređa konzistencija (mešavina sa ± 10% praha od preporučene razmere) izloženih sili od 2 kg. Kontrolnu grupu činili su uzorci oba ES zamešanih prema uputstvu proizvođača bez opterećenja, izloženi samo težini jedne staklene pločice. Veličina napona tečenja posmatrana je u funkciji prečnika razlivenog silera kao parametra napona tečenja između para pločica. **Rezultati.** Poređenjem uzoraka (prečnika razlivenih silera) opterećenih sa 1 prema 2 kg kod oba materijala, nađena je statistički neznačajna razlika ($p > 0,05$). Nalazi u vezi prečnika razlivenih silera u pogledu varijacije ± 10% praha kod ES *Endomethasone N* ukazuju na to da ne postoji značajna razlika u naponu tečenja između standardno i gušće zamešane mase ($p > 0,05$), dok je razlika bila značajna poređenjem uzoraka standardno zamešane mase prema onima saređom konzistencijom ($p < 0,01$). **Zaključak.** Promena sile sa 1 kg na 2 kg kod uzoraka oba silera ne utiče značajno na napon tečenja kao ni poređenje standardno i gušće zamešanog *Endomethasone*, dok je poređenjem njegove standardne i ređe zamešane mase utvrđena značajna razlika.

Ključne reči:
zub, materijali za punjenje korenskog kanala; cink oksid-eugenol pasta; reologija; viskoznost.

Correspondence to: Dragan Ilić, Faculty of Dental Medicine, Clinic for Restorative Dentistry and Endodontics, Rankeova 4/IV, Belgrade, Serbia. Phone.: +381 11 244 33 66. Mob.: +381 62 372 271. E-mail: dr.ilicb@gmail.rs

Introduction

The main function that a root canal sealer and gutta-percha should meet during obturation are lubrication and setting the master and auxiliary gutta-percha cones acting as intermediary and sticky-adhesive substance in a labyrinthine depulpated space. The outcome of endodontic therapy might depend on sealers' properties whether they are biological, chemical or physical ones. The flow of endodontic sealer (ES) is affected by its viscosity as well as temperature and humidity. By the way, it is obvious that sealer's flow depends on the shape, width and taperness of the root canal.

An adequate consistency is required whether to use a paste carrier (Lentulo spiral filler) or soaked gutta-percha point. The endodontists often adjust the powder/liquid ratio to the appropriate consistency of the sealer, usually in eugenate sealer materials. The most desirable consistency should be chosen considering the 2 aims: not to overfill the apical canal portion when thicker consistency is required (wide open apical foramen, unfinished root growth), and on the contrary, weak consistency, is desirable when last millimeter of canal is not to be well obturated, i.e. strongly curved/narrow canals.

Some authors advocate for cleaning of smear layer as the important condition for ES flow and its penetration into the dentine tubules [1]. On the other hand, during the use of compaction techniques by the instruments and devices for obturation mass, sealer or gutta-percha cones pressing, high values of exposed pressure act as hydrodynamic pump to the root-canal walls. A result of compaction forces should be visible in filling all the canal irregularities, accessory ones as well as apical delta due to high exposed values of lateral and vertical forces. Some authors apply the force of 2 kg (~19.6 N) imitating clinical compaction stress, while the others use zinc-oxide eugenol ES and real clinical obturation force of 10, 15, 20 and the 25 N in studies on the quality of apical seal [2–3]. Their exams were based on the study where the average manual force during obturation ranged 10–30 N among eight endodontists [4]. Application of heat in some obturation techniques may also influence the flow characteristic of a sealer [5].

Various devices have been used in evaluation the rheological properties of ES. This might be the reason for not having any important laws and conclusions about flow properties of sealers. In the study on temperature influence on sealer custom-made capillary rheometer [5, 6], and cone-and-plate geometry were used [5].

ES flow rate has been studied by some investigators using a vertical glass plate [7] or a 2-plate system [5, 8–12]. Some investigation of ES flow was done through notification of microleakage into the lateral canals or tubules [13–17]. The aspect of contact angles at 4 ES points out the correlation on their flow properties [18].

Japanese authors [19] compared the two testing device values (vertical plate and two-plate system) of the flow on the same sealers. Extrusion viscometer [20] or free extrusion of ES through the bore [21] has been used for research on the rheological characteristic.

Considering the aforementioned, the aim of this study was to investigate the rheology features by influence of powder: liquid ratio and the two forces applied to the zinc-oxide eugenol (ZOE) ES. The first null hypothesis was that there would be no significant difference in flow rate among the regularly prepared sealers at the level of $\alpha = 0.05$ and regardless the applied load of 1 and 2 kg at the level of $\alpha = 0.05$. The second null hypothesis was that there would be no significant difference in flow regardless the sealer consistency and in comarison to regularly prepared mix considering the level of $\alpha = 0.05$.

Methods

The root canal sealers

The two ZOE preparations as ES were tested for the study whose approximate contents according to the manufacturer are given in Table 1 [22, 23].

Table 1

The approximate composition /main ingredients/ of the used zinc oxide-eugenol (ZOE) endodontic sealers

ZOE endodontic sealers	Ingredients
Endomethasone N (Septodont) powder	zinc-oxide, magnesium stearate, thymol iodide, barium sulphate, hydrocortisone acetate, excipients liquid: eugenol, excipients [22]
Roth 801 (Roth Inter Limit.) powder	zinc-oxide, staybelite resin, bismuth subcarbonate, barium sulphate, sodium borate anhydrous liquid: pure eugenol [23]

The study groups

The study involved the experimental and the control group.

The experimental group involved the samples of a regular and variated mixture of the two aforementioned ZOE ES (Table 1). Regular mixtures of Endomethasone (12 samples) and Roth 801 (10 samples) were subjected to the load of 1 kg and 11 samples per each sealer to the load of 2 kg. The variated Endomethasone samples were prepared as thick and thin consistency (11 samples of each) and exposed to the load of 2 kg.

The control group included 3 samples of each used sealer regulary prepared. They were exposed to the weight of only one mixing slab (100 g).

The load exposure time was 10 min for all samples in the experimental and control groups.

The components ratio

Endomethasone N was prepared as regular prescription with ratio: one spoonful of powder to two drops of liquid [22]. Thick consistency contained 10% more powder (by weight)

than regular mix. Thin consistency had 10% less powder than regular mix. The reason to choose so minimal deviation of standard proportion (± 10% of powder) was reality of clinical situation where many times ES is prepared by such a variated proportion unintentionally or intentionally, as well as that no up-to-date literature data appeared of such study concept.

Roth's 801 sealer was prepared as regular prescription to the consistency of petrolatum gel by the powder liquid ratio of 0.13 g : 0.03 g [21].

The adjustment of components was done by digital scale device with accuracy of 0.001g (Mettler PE 360, Germany).

The protocol and the experimental device for the experimental and control groups

The experiment methodology was based mostly on the 2-glass-plate geometry system (ADA specification No 57).

The ES were mixed according to the directions of a corresponding manufacturer and variated for Endomethasone samples [24]. The same amount of sealer (0.06 mL) was placed immediately after mixing by graduated syringe to the center of glide mixing plate and spread for 1 min by dental probe forming the circle of approximately 10 mm diameter. Another glass plate weighing 100 g was then gently placed over the first 3 min after initiation of mixing. An extra load of 1 and 2 kg was added for the samples in the experimental group. A 2-glass-plate system was then fixed laterally to prevent minimal moving. All 4 brinks of the 2-glass-plate unit were fixed by ten tangentially placed 4 cylindrical metal weights of 1 kg. Measurements were done at the stable lab temperature of 22 ºC and 65% humidity.

There was no load application except the weight of mixing slab of around 100 g in the control group samples.

The measuring

Measuring of values of the two perpendicular diameters were done in both experimental and control group 10 min after sealer application by the help of an orthodontic ruler of 0.5 mm raster and error of 0.025 mm (Figure 1). The values were recorded as the average estimation of two measured diameters summing the maximal and minimal values by the help of 4 × magnifying glass. The sample was discarded if the difference in the two recorded diameters per sample was more than 1 mm.

The Student's t-test was used for recording the differences in disk diameters among experimental samples in regard to the applied pressures (weight), in the cases of variations of two components in Endomethasone N samples as well as for comparison between Endomethasone N and Roth 801 viscosity values for regular mix and load of 2 kg.

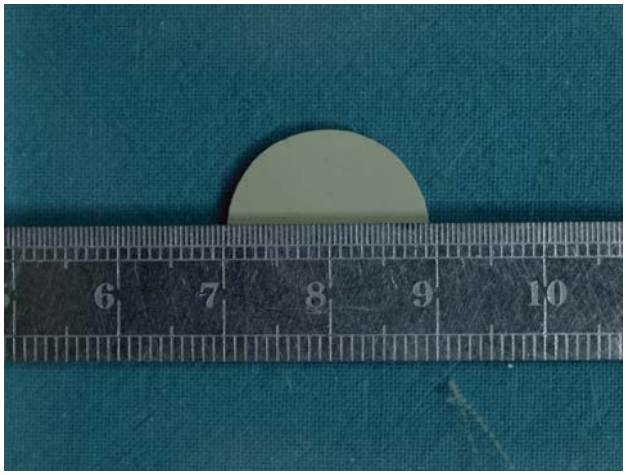


Fig.1 – The disk diameter of spread sealer measured by orthodontic ruler

Results

The mean disk diameter values of spreading regular and variated prepared mixed mass of sealers upon exposing the load of 1 and 2 kg are presented in Table 2 for the experimental and in Table 3 for the control group.

Table 2

The disk diameter mean and coefficient of variation CV (%) values of spread regular and varied mixtures of sealers between the glass slabs upon exposing to the extra load of 1 and 2 kg (experimental group)

Sealer	Mixtures			
	Regular prepared		Varied	
	1kg	2kg	thick (2kg)	thin (2kg)
	$\bar{x}$ (CV)	$\bar{x}$ (CV)	$\bar{x}$ (CV)	$\bar{x}$ (CV)
Endomethasone N	21.9 (8.5)	24.1 (8.7)	25.0 (11.6)	21.7 (9.1)
Roth 801	29.6 (12.0)	32.8 (17.7)	----------	----------

Table 3

The disk diameter and coefficient of variation (CV) values of spread sealer mass without extra load (control group)

Sealer	Mean diameter (mm)	CV (%)
Endomethasone N	15.7	8.2
Roth 801	22.4	2.5

The difference in disk diameters of the mixed mass spread by exposing the load of 1 kg weight vs. 2 kg for both regularly prepared sealer mixture was not statistically significant ($p > 0.05$).

A significant statistical difference in disk diameters values was found in comparison of Endomethasone N and Roth 801 sealer when regularly mixed and used 1 or 2 kg load ($p < 0.01$).

Comparison of disk diameters values for 2 kg load of the regular and thick Endomethasone N preparation failed to show statistically significant difference ($p > 0.05$).

A statistically significant difference in disk diameter was found in comparison of the regular Endomethasone N mixture to the thin mixed mass ($p < 0.01$), as well as in thick *vs* thin Endomethasone N mixed mass ($p < 0.01$).

The disk diameters in the control group within each sealer of no extra load were recorded as vary near values for both materials, thus provided the reliable parameters for statistical analysis (Table 3). Those mean values of disk diameters point out that the load of only one glass plate to exposed Roth 801 as sealer give provide statistically significantly bigger diameters than Endomethasone N ($p < 0.01$) cases.

Discussion

The obtained coefficient of variation (CV) values for samples in this study for both experimental and control group were far below 30%, *ie* in the ranges of 8.5%–17.7% and 2.5%–8.2%, respectively, what characterized them as statistically homogenous groups very suitable for a precise statistical analysis.

In this study two glass plates were used on the same way as Grossman did in his 1976 study [25]. The reason to choose this method is its simplicity as well as the presence of comparable literature data. The flow investigation under ISO specifications [26] on ZOE endodontic sealers (Canals, Showa) and sealer with other ingredients displayed diameter values higher than 20 mm (39.2–46.2 mm) thus satisfied ISO requirements [19]. A study on 3 sealer flow in a 2-plate system under ADA conditions exposed mean diameter values in the range of 32.7–37.9 mm whith the highest values for ZOE sealer [11]. In the flow study of Endomethasone authors did not required the ADA specification No. 57, although of limited value (20 mm disk diameter) and ISO standard by the value of only 11 mm diameter [13]. The present study results satisfied ADA specification No. 57 of sealer with the mean diameter values higher than 20 mm (21.9–32.8 mm).

One can say that all aforementioned results were in the range of $d \geq 20$ mm. The variation might be explained by a slightly performed deviation of the experimental conditions such as weight of a glass slab (30, 80, 100, 120, 450 or 500 gr), or extra load (1, 2 or 2.5 kg) the amount of the applied sealer on the plate (0.05, 0.06, 0.1 or 0.5 mL) and load exposure time (30 sec, 7 or 10 min).

In this study the comparison of Endomethasone N thick *versus* thin mix by glass plates revealed the significant difference in disk diameters. The difference of powder saturation between these two consistencies was around 20%. In comparison of the two ZOE ES with the deviation of around 30% of median consistency French authors noted the similar results [12].

Although some authors used the powder increase of 50% in Grossman ZOE ES they did not obtain a significant difference in diameter values among thick and thin mix, as well as to Tubliseal ZOE ES, most probably due to negligence of both ADA and ISO specification. However, it is amazing that their study noted disk diameters larger than 20 mm for all sealers thus required the ADA conditions [6]!

The hypothesis that the flow is comparable with the penetration degree into dentinal tubules checked by SEM stated the authors who noted the ZOE Pulp Canal Sealer as low potential sealer in regard to resin-based sealer [16].

The compaction force of 1.0 kg was applied in the study to imitate the Schilder plugger for compaction during obturation in test of various viscosity mixture [27]. According to the noted force values during compaction in the range of 8–35 N [2, 3] the aim of this study was to compare those authors' results themselves. This is the reason to chose the load for of 10.0 and 20.0 N in this study.

An increase in intracanal pressure during the rise in sealer viscosity is noted both with more or less thick consistency [27]. This might be of high importance in thin roots due to the possible fracture [28].

It is sometimes very difficult to make the accurate proportion of the powder and liquid or two-paste system because double-syringe or accurate spoonful or bottle for all brands of ES are sometimes missing. That is another reason to believe in unintentional variation of ingredients in the amount of ± 10% of one component during preparation. It is questionable if a slight variation of one component might significantly influence the rise or decrease in sealer flow and thus cause an unwanted change in planned clinical consistency up to the concerned clinical endodontic situation. The variation of sealer consistency was applied in this experiment due to the recommendation of the Endomethasone N manufacturer allowing regular ratio of 1 : 2 of powder and liquid with deviation of around ± 50% [22]. Actually, the manufacturer allows the mixing variation in the sense of thicker and thinner consistency depending on the clinical situation. Using the variations in powder of Endomethasone N of only ± 10% (clinical approved mixtur) and obtaining only a limited influence of the sealer's rheology, this study is characterized as the novelty in the literature data. The result of French authors about the influence of powder variation of around ± 30%–50% to the rheology of the two ZOE ES, Pulp Canal sealer and Cortisomol revealed a significant flow change [12]. Some authors obtained statistically significant differences in flow parameters comparing the viscosity of the mixture much thicker than much thinner (± 10% of powder) and than regular ZOE sealer mix [5].

The noted flow rate in vertical glass plate experiment of two ZOE sealers, Endomethasone and Procosol, revealed significant difference where both of them were exposed to significantly lower flow than resin-based and $Ca(OH)_2$ filler materials [7].

Sealer extrusion through the bore did not showed significant difference of flow between two ZOE sealers Roth 801 and Tubliseal EWT [21].

Although this study showed no influence of guttapercha on the flow of obturation mass, it was shown that the flow of filling material such as ZOE ES depends on guttapercha flow. It can be explained by the influence of the chemical compounds of gutta-percha cone that vary in different brands [29] what is advised to be studied in the next research.

Observing literature data sometimes presented as heterogeneous rheological characteristic of the ES obtained for the same materials but under slightly or largely changed conditions, point out to the need of strict following the standards in order to compare the results of investigators worldwide.

Conclusion

Application of 1 kg *versus* 2 kg load for both regularly mixed sealers in the scope of the obtained disk diameter (flow) was statistically insignificant ($p > 0.05$).

A significant statistical difference in disk diameters values (flow) was found in comparison of Endomethasone and Roth's 801 sealers both regulary mixed for application of 1 kg or 2 kg load ($p < 0.01$).

The obtained difference in disk diameter in mixing variation of Endomethasone N and 2 kg load points out statistical insignificance in flow rate between regular and thick mixtures ($p > 0.05$). A significant difference was found in comparison of regular and thin mixtures by the load of 2 kg ($p > 0.01$).

REFERENCES

1. *Calt S, Serper A.* Dentinal tubule penetration of root canal sealers after root canal dressing with calcium hydroxide. J Endod 1999; 25(6): 431−3.
2. *Hatton JF, Ferrillo PJ Jr, Wagner G, Stewart GP.* The effect of condensation pressure on the apical seal. J Endod 1988; 14(6): 305−8.
3. *Joyce AP, Loushine RJ, West LA, Runyan DA, Cameron SM.* Photoelastic comparison of stress induced by using stainless-steel versus nickel-titanium spreaders in vitro. J Endod 1998; 24(11): 714−5.
4. *Harvey TE, White JT, Leeb IJ.* Lateral condensation stress in root canals. J Endod 1981; 7(4): 151−5.
5. *Lacey S, Pitt Ford TR, Yuan XF, Sherriff M, Watson T.* The effect of temperature on viscosity of root canal sealers. Int Endod J 2006; 39(11): 860−6
6. *Lacey S, Pitt Ford TR, Watson TF, Sherriff M.* A study of the rheological properties of endodontic sealers. Int Endod J 2005; 38(8): 499−504.
7. *Kaplan AE, Ormaechea MF, Picca M, Canzobre MC, Ubios AM.* Rheological properties and biocompatibility of endodontic sealers. Int Endod J 2003; 36(8): 527–32.
8. *Bernardes RA, de Amorim Campelo A, Junior DS, Pereira LO, Duarte MA, Moraes IG,* et al. Evaluation of the flow rate of 3 endodontic sealers: Sealer 26, AH Plus, and MTA Obtura. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2010; 109(1): e47−9.
9. *Siqueira FJ Jr, Fraga RC, Garcia PF.* Evaluation of sealing ability, pH and flow rate of three calcium hydroxide-based sealers. Endod Dent Traumatol 1995; 11(5): 225−8.
10. *Siqueira JF Jr, Favieri A, Gahyva SM, Moraes SR, Lima KC, Lopes HP.* Antimicrobial activity and flow rate of newer and established root canal sealers. J Endod 2000; 26(5): 274−7.
11. *Gambarini G, Testarelli L, Pongione G, Gerosa R, Gagliani M.* Radiographic and rheological properties of a new endodontic sealer. Aust Endod J 2006; 32(1): 31−4.
12. *Camps J, Pommel L, Bukiet F, About I.* Influence of the powder/liquid ratio on the properties of zinc oxide-eugenol-based root canal sealers. Dent Mater 2004; 20(10): 915−23.
13. *Almeida JF, Gomes BP, Ferraz CC, Souza-Filho FJ, Zaia AA.* Filling of artificial lateral canals and microleakage and flow of five endodontic sealers. Int Endod J 2007; 40(9): 692−9.
14. *Weis MV, Parashos P, Messer HH.* Effect of obturation technique on sealer cement thickness and dentinal tubule penetration. Int Endod J 2004; 37(10): 653−63.
15. *Ordinola-Zapata R, Bramante CM, Graeff MS, del Carpio Perochena A, Vivan RR, Camargo EJ,* et al. Depth and percentage of penetration of endodontic sealers into dentinal tubules after root canal obturation using a lateral compaction technique: a confocal laser scanning microscopy study. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2009; 108(3): 450−7.
16. *Mamootil K, Messer HH.* Penetration of dentinal tubules by endodontic sealer cements in extracted teeth and in vivo. Int Endod J 2007; 40(11): 873−81.
17. *Karabucak B, Kim A, Chen V, Iqbal MK.* The comparison of gutta-percha and Resilon penetration into lateral canals with different thermoplastic delivery systems. J Endod 2008; 34(7): 847−9.
18. *Kontakiotis EG, Tzanetakis GN, Loizides AL.* A comparative study of contact angles of four different root canal sealers. J Endod 2007; 33(3): 299−302.
19. *Ono K, Matsumoto K.* Physical properties of CH61, a newly developed root canal sealer. J Endod 1998; 24(4): 244−7.
20. *Negm MM, Lilley JD, Combe EC.* A study of the viscosity and working time of resin-based root canal sealers. J Endod 1985; 11(10): 442−5.
21. *McMichen FRS, Pearson G, Rahbaran S, Gulabivala K.* A comparative study of selected physical properties of five root-canal sealers. Int Endod J 2003; 36(9): 629−35.
22. Endomethasone N Ciment d'obturation canglaire, Septodont. Available from: www.septodont.com.plat@EbookBrowse. (English)
23. *Mazinis E, Eliades G, Lambrianides T.* An FTIR study of the setting reaction of various endodontic sealers. J Endod 2007; 33(5): 616−20.
24. ANSI/ADA Standard No. 57—Endodontic Sealing Material: 2000 (Reaffirmed 2006). Available from: www.accessdata.fda.gov/.../cfstandards/detail [cited 2008 September 9]
25. *Grossman LI.* Physical properties of root canal cements. J Endod 1976; 2(6): 166−75.
26. International Organization for Standardization ISO-6876 Dental Root Sealing Materials. Geneva: International Organization for Standardization; 2001.
27. *Brooke KK, Grace MG.* Relationship of intracanal pressure with viscosity of endodontic sealer during warm Gutta-Percha vertical compaction. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2000; 89(5): 618−22.
28. *Holcomb JQ, Pitts DL, Nicholls JI.* Further investigation of spreader loads required to cause vertical root fracture during lateral condensation. J Endod 1987; 13(6): 277−84.
29. *Venturi M, Di Lenarda R, Breschi L.* An ex vivo comparison of three different gutta-percha cones when compacted at different temperatures: rheological considerations in relation to the filling of lateral canals. Int Endod J 2006; 39(8): 648−56.

Received on Januar 27, 2011.
Revised on May 10, 2011.
Accepted on May 20, 2011.

ORIGINAL ARTICLE

UDC: 617.7-089.87
DOI: 10.2298/VSP1301026K

Causes of eye removal – analysis of 586 eyes

Uzroci enukleacije očne jabučice – analiza 586 enukleisanih očnih jabučica

Miroslav Knežević, Jelena Paović, Predrag Paović, Vojislav Sredojević

Institute for Eye Diseases, Clinical Center of Serbia, Faculty of Medicine, Belgrade, Serbia

Abstract

Background/Aim. Eye enucleation is one of the oldest surgical procedures. The aim of the study was to determine the causes of enucleation as seen in a major reference eye center in Serbia. **Methods.** Retrospective case series involving a review of all enucleation procedures performed in the period between January 2000 and December 2008 at the Institute for Eye Diseases, Clinical Center of Serbia, Belgrade. The collected information included the basic demographic data and diagnosis of the affected eye. The diagnosis was made based on history, clinical and histological examinations. Clinical indications for enucleation were categorized as tumors, glaucoma, trauma, infections and other diseases. A statistical analysis was made using the Student's t-test. **Results.** There were 586 patients, 315 male and 271 female in our series. The mean age was 57.81, ranging from 3 months to 96 years. The most common cause of enucleations was tumor (76.11%), ($p < 0.05$). Choroid melanoma was the most common etiology leading to enucleation (81.18%), followed by retinoblastoma (12.34%). A total of 8.02% of enucleations were performed due to glaucoma that was primarily neovascular in 42.55% of cases or caused by trauma in 38.8% of cases. Trauma was the third common etiology of enucleation, and it was acute in 56.26% of cases or resulted in phthisis bulbi in 31.25% of cases. Enucleation caused by inflammation was performed in 2.90% of cases, out of which 52.94% of enucleations occurred after perforation of the cornea. In the group of other diseases the most common cause of enucleation was atrophy of the eye ball. **Conclusion.** Neoplasm, neovascular glaucoma, acute eye injury and atrophy of the eye ball are the most common causes of enucleation.

Key words:
eye enucleation; eye diseases; risk factors.

Apstrakt

Uvod/Cilj. Uklanjanje očne jabučice predstavlja jednu od najstarijih hirurških procedura. Cilj rada je bio ispitivanje razloga za uklanjanje očne jabučice. **Metode.** Retrospektivnom studijom obuhvaćene su sve enukleacije očnih jabučica koje su urađene u periodu između januara 2000. i decembra 2008. u Institutu za očne bolesti u Beogradu. Analizom su obuhvaćene demografske karakteristike bolesnika i dijagnoze oboljenja oka kod kojih je urađena enukleacija. Dijagnostika je bila zasnovana na kliničkom i histološkom nalazu. Kliničke indikacije za enukleaciju podeljene su na: tumore, glaukom, traumu, infektivne bolesti i druge bolesti oka. Statistička analiza je vršena korišćenjem Studentovog *t*-testa. **Rezultati.** Enukleacija je urađena kod 586 bolesnika, 315 muškaraca i 271 žene, prosečne starosti 57,81 (raspona od 3 meseca do 96 godina). Najčešći uzroci enukleacije bili su tumori (76,11%), ($p < 0,05$). Najčešći uzroci enukleacije u okviru tumora bolesnika bili su horoidalni melanomi (81,18%) i retinoblastomi (12,34%). Enukleacija zbog glaukoma urađena je kod 8,02% bolesnika, i to najčešće zbog neovaskularnog glaukoma (42,55%). Trauma je bila uzrok za enukleaciju kod 38,30% bolesnika. Po učestalosti trauma je treći najčešći razlog za enukleaciju, najčešće akutna trauma (56,26%), a zatim ftiza očne jabučice (31,25%). Inflamacija kao razlog za enukleaciju bila je prisutna kod 2,90% bolesnika, od čega kod 52,94% enukleacija je urađena posle perforacije rožnjače. U grupi drugih bolesti, najčešći razlog za enukleaciju bila je atrofija očne jabučice. **Zaključak.** Neoplazme, nevaskularni glaukom, akutna povreda oka i atrofija očne jabučice predstavljaju najčešće razloge za enukleaciju očne jabučice.

Ključne reči:
oko, enukleacija, oko, bolesti; faktori rizika.

Introduction

Enucleation is the removal of the eyeball, excluding the conjunctiva and the muscles. It is one of the oldest surgical procedures of the eye. Enucleation is performed in tertiary ophthalmological institutions when all treatment options are exhausted. Various eye diseases may lead to a blind and painful eye or phthisis bulbi, the diseases that are the most common causes of enucleation. The indications for enucleation are the same worldwide [1–8]. Differences between some regions depend on the development of their respective health protection systems. There are numerous studies about the changing patterns of diseases leading to enucleation [9–23]. According to these studies, the following are the causes of enucleation: neoplasm,

Correspondence to: Jelena Paović, Institute for Eye Diseases, Clinical Centre of Serbia, Pasterova 2, Belgrade, Serbia.
Phone: +381 63 245 552. E-mail: maliceda@eunet.rs

end-stage glaucoma, blunt or penetrating injuries of the eyeball, endophthalmitis, chronic uveitis, congenital glaucoma, etc. In developed countries, the most common causes of enucleation are ocular tumors, while in poor countries traumas of the eye are the most common etiology leading to enucleation. The introduction of new procedures in treatment of ocular tumors has significantly reduced the number of enucleations performed due to malignant choroidal melanoma [24]. One of the causes of enucleation is neovascular glaucoma. Panretinal photocoagulation and ligation of anterior ciliary arteries have significantly reduced the number of enucleations caused by neovascular glaucoma [25]. Endophthalmitis results in enucleation when other treatment options are exhausted. In the past few years serious intraocular infections led more often to evisceration than enucleation. The indications for enucleation and evisceration decreased in the last decade, most probably due to improved modalities of treatment [26]. Endophthalmitis after ulcerations and melting of the cornea may result in enucleation or evisceration [27].

The aim of this study was to determine the causes of enucleation as seen in the major reference eye center in Serbia.

Methods

This case series involved a review of patients hospitalized during the period between January 2000 and December 2008 at the Institute for Eye Diseases, Clinical Center of Serbia (CCS), Belgrade, Serbia. Histories of the disease and pathohystological findings of the enucleated eyes were used as the data source. The patients were divided into age groups, subdivided into groups encompassing 10 years, those who were less than 10-year old, and patients who were more than 90-year-old. The distribution of enucleation was performed per age. The primary clinical indications for enucleation were categorized into five groups: tumors, glaucoma, trauma, inflammation and other. Tumors were classified as benign and malignant. Malignant tumors were divided into primary and metastasizing. Glaucoma was divided into the following groups: absolute, congenital, neovascular and posttraumatic. Traumas were divided into acute, fresh traumas and posttraumatic conditions, such as retinal ablation and massive hemorrhage. Inflammations of the eye resulting in enucleation were divided into: uveitis, keratitis with perforation, endophthalmitis, posttraumatic uveitis and other inflammatory conditions. Other diseases leading to enucleation were divided into old detachments, phthisis bulbi, congenital anomalies of the eye, atrophy of the eyeball and Coats' disease.

The statistical analysis was made by using the Student's *t*-test with statistical significance of $p < 0.05$.

Results

Enucleation was performed in 586 patients, 315 men and 271 women. The youngest patient was 3-month-old, while the oldest was 96-year-old. The average mean age of enucleated patients was 57.81 ± 7.50 years, and the age group most commonly affected by enucleation ranged between 50 and 70 years (Figure 1).

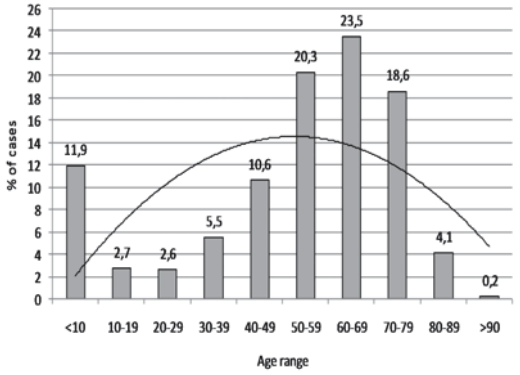


Fig. 1 – Age distribution of the patients with enucleated eyes

In the observed term the number of patients undergoing enucleation declined. In 70% of the patients, the difference in their respective age varied by 10%–15% (Figure 2).

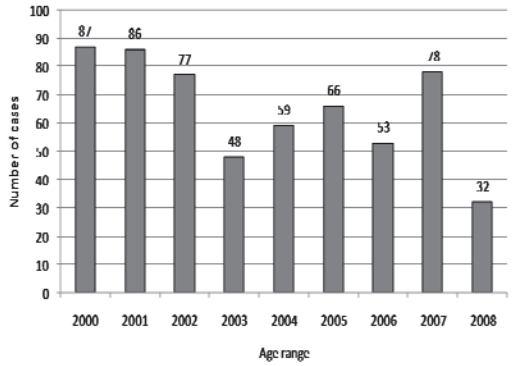


Fig. 2 – Distribution of the performed enculeations per year in the observed term

The most common indication for enucleation was tumour (76.11%) as compared to other causes with the respective share ranging between 2.73% and 10.24%. Tumors are significantly the most common cause of enucleation, ($p < 0.05$). Glaucoma accounted for 8.02% of enucleation cases, trauma for 2.73%, inflammation for 2.90% of the cases, while other diseases and conditions resulted in enucleation in 10.24% of cases (Figure 3).

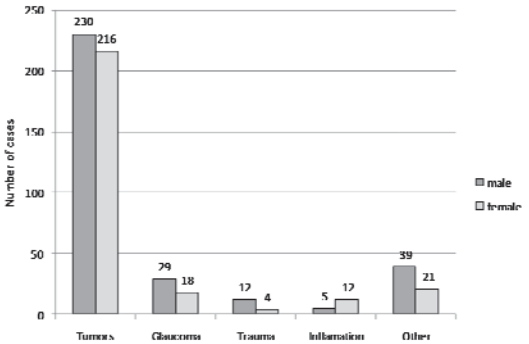


Fig. 3 – Diseases resulting in enucleation

The number of men and women affected by enucleation due to ocular tumour was approximately the same – 228 men and 212 women, while enucleation caused by eye trauma occurred more often in men than women, i.e. 2/3 of male versus 1/3 of female patients.

Out of the total number of enucleations, enucleation was performed in 76.11% of the patients due to tumour. Benign tumors (choroidal hemangioma and leiomyomata) occurred with significantly lower probability of 1.3% as compared to malignant tumors that occurred in 98.7% of the cases, ($p < 0.05$). Primary malignant tumors accounted for 97.7% of all malignant tumors. Among primary malignant tumors, malignant choroidal melanoma was the most common cause of enucleation (81.18% of all malignant tumors). Malignant choroidal melanoma was significantly more common cause of enucleation in general (61.77%), as well as in the group of primary malignant tumors (81.18%). The probability of enucleation due to malignant choroidal melanoma did not vary a lot between the two genders (Table 1).

Glaucoma as a cause of enucleation accounted for 8.02% of the cases. Two third of patients suffering from glaucoma were men and 1/3 women. Absolute glaucoma occurred in 10.64% of the cases, congenital glaucoma in 8.51% of the cases, neovascular glaucoma in 42.55% of the cases and glaucoma resulting from trauma in 38.30% of the cases. Significantly, the most common cause of enucleation in case of glaucoma was neovascular glaucoma, followed by glaucoma caused by trauma ($p < 0.05$) (Table 2).

Trauma leading to enucleation occurred in 2.73% of all enucleation cases, having significantly higher incidence in men than women, ($p < 0.05$). In 56.26% of the cases there was an acute injury ($p < 0.05$). Esthetic reasons relating to phthisis bulbi caused enucleation in 31.25% of the cases, which was not significantly less than acute injuries resulting in enucleation (Table 3).

Table 1

Number of enucleations caused by tumors

Tumors	Gender (n)		Total patients	
	Male	Female	n	%
Benign				
choroidal hemangioma	2	3	5	1.12
leiomyoma	0	1	1	0.22
Σ	2	4	6	1.3
Malignant				
Primary				97.7
penetration of conjunctival tumour into the eye	7	5	12	2.69
retinoblastoma	23	32	55	12.34
medulloepithelioma	0	1	1	0.22
uveal melanoma	192	170	362	81.18
Metastasizing				2.3
metastasizing tumour	6	4	10	2.23
Σ	228	212	440	98.7
Total	230	216	446	100

Table 2

The number of enucleations caused by glaucoma

Glaucoma	Gender (n)		Total patients	
	Male	Female	n	%
Absolute glaucoma	1	4	5	10.64
Congenital glaucoma	3	1	4	8.51
Neovascular glaucoma			20	42.55
due to diabetes	3	2	5	10.64
postoperative	1	1	2	4.26
due to retinal vein occlusion	6	3	9	19.14
other	2	2	4	8.52
Glaucoma caused by trauma	13	5	18	38.30
Total	29	18	47	100

Table 3

The number of enucleations caused by trauma

Trauma	Gender (n)		Total patients	
	Male	Female	n	%
Trauma –esthetic reasons	4	1	5	31.25
Acute trauma	6	3	9	56.25
Posttraumatic retinal detachment	1	0	1	6.25
Hemophtalmus	1	0	1	6.25
Total	12	4	16	100

Inflammation as the cause of enucleation occurred in 2.90% of all the enucleation cases. It was caused with a significant probability by keratitis leading to corneal perforation (52.94%) ($p < 0.05$), primarily in women. Other significant cause of enucleations resulting from inflammation was uveitis (23.53%), but it occurred with significantly lower incidence rate than keratitis with perforation. Other causes from the inflammation group such as endophthalmitis, exogenous postoperative uveitis and other inflammatory eye diseases were incidental events (Table 4).

Other diseases and conditions, such as: conditions after retinal detachment, phthisis bulbi, congenital anomalies of the eye, Coats' disease, atrophy of the eyeball and other diseases and conditions leading to enucleation occurred in 10.24% of all the enucleation cases. In the group of other diseases, a significant, most common cause of enucleation was the atrophy of eyeball (56.67%) ($p < 0.05$) (Table 5).

Glaucoma caused enucleation in 8.02% of the cases, trauma in 2.73%, inflammation in 2.90% and other conditions in 10.24% of the cases. In comparing with the literature results, an increased number of enucleated eyes caused by malignant choroidal melanoma contrary to enucleations caused by retinoblastoma [24] can be explained by the fact that malignant tumors were diagnosed later, when the other methods of treatment could not be performed. Metastasizing tumors (2.3%) were diagnosed later because of secundary glaucoma. The ratio between men and women with the performed enucleation because of tumour was approximately the same (228 men and 212 women), while the number of enucleations due to trauma in men was higher than in women (2/3 men and 1/3 women). Contrary to our results, Gyasi et al. [22] revealed that the most often causes of enucleation were infections, in 47.9% of cases, followed by trauma, in 23.2% of cases, degenerations in 14.9% of cases, and other diseases in 8.9% of

Table 4

Number of enucleations caused by inflammation

Inflammation	Gender (n)		Total patients	
	Male	Female	n	%
Endogenous uveitis	2	2	4	23.53
Keratitis with perforation	1	8	9	52.94
Endophthalmitis	0	1	1	5.88
Exogenous posttraumatic uveitis	1	0	1	5.88
Other inflammatory conditions of the eye	1	1	2	11.77
Total	5	12	17	100

Table 5

Other diseases that caused enucleation

Eye diseases and conditions	Gender (n)		Total patients	
	Male	Female	n	%
Conditions after retinal detachment	4	6	10	16.67
Phthisis bulbi	1	1	2	3.32
Retinopathy pigmentosa	1	0	1	1.67
Congenital anomalies of the eye	3	1	4	6.67
Retinopathy of prematurity	2	2	4	6.67
Atrophy caused by trauma	25	9	34	56.67
Coats's disease	3	2	5	8.33
Total	39	21	60	100

Discussion

In a tertiary ophthalmological institution – the Institute of Eye Diseases, CCS in Belgrade, during the period of 8 years enucleations were performed in 586 patients. The youngest patient was 3-month old, while the oldest was 96. The average mean age of the patients was 56.81 ± 7.50 years, as opposed to Gyasi et al. [22] where the mean age of 336 patients with enucleation was 36.4. The distribution of patients according to their respective age reveals that in 70% of patients the affected age differs by 10%–15%. The distribution of patients per year shows that the number of the performed enucleations has declined during the course of time, which may be explained by the progress made in the treatment of eye diseases that may lead to enucleation.

The most common indication for enucleation was tumour (76.11%). Other causes had a share of 2.73%–10.24%. cases. Neoplasm was the fifth cause according to its incidence rate, i.e. 5.1%. Setlur et al. [23] in 2010 during a 60-year-follow-up enucleations found that neoplasm was still the most common cause of enucleation, while there was a fluctuation with age in terms of an increased number of enucleations due to retinoblastoma as compared to malignant choroidal melanoma. In our study malignant tumors occurred in 98.7% of the cases. Among malignant tumors, the most often were malignant choroidal melanoma, in 81.18% of the cases, and retinoblastoma in 12.34% of the cases. The ratio between men and women was approximately the same (43.05% men and 38.13% women), contrary to retinoblastoma where men and women were equally represented. Glaucoma was the second most common cause of enucleation, accounting for 8.02% of the cases. A similar percentage, 8% of all enucleations, was observed during a 60-year-follow-up term by Setlur et al. [23] in 2010, where the number of enucleations

during a longer follow-up caused by glaucoma declined and amounted to 23% in a period between 1950 and 1959 and 8% in a period between 2000 and 2006. Two third patients to whom enucleation was performed due to glaucoma were men and 1/3 were women. The most common cause of enucleation was neovascular glaucoma (42.55%), followed up by posttraumatic glaucoma (38.30%). The great number of enucleations caused by neovascular glaucoma compared with literature data [25] can be explained due to the fact that panretinal photocoagulation was not performed. Trauma as the cause of enucleation accounted for 2.73% of cases, occurring with a significantly higher probability in men than in women, while acute trauma occured with a significantly higher probability (56.25%). The second most common cause of enucleation after acute trauma was enucleation due to aesthetic reasons (31.25%). Inflammation as the fourth among the most common causes of enucleation accounted for 2.90% of all enucleations. It occurred with a significant probability due to some infection after perforation of the corneal ulcer (52.94%), and it occurred more often in women than in men, followed up by uveitis in 23.53% of cases. Perforation of the corneal ulcer and bacterial endophthalmitis, most frequently caused by *Pseudomonas aeruginosa* and other bacteria and fungi, if not treated, may lead to enucleation or evisceration [27]. In 10.24% of cases the cause of enucleation were other diseases and among them, atrophy of the eyeball had a significantly highest probability.

In summary, the most common indications for eye removal were: neoplasm, neovascular glaucoma, acute eye injury and atrophy of the eyeball.

Conclusion

The most common indication for enucleation was tumour (76.11%). Among malignant tumors, the most often were malignant choroidal melanoma, in 81.18% of cases, and retinoblastoma in 12.34% of cases.

Glaucoma was the second most common cause of enucleation, accounting for 8.02% of cases. The most common cause of enucleation was neovascular glaucoma (42.55%). Trauma as the cause of enucleation accounted for 2.73% of cases, occurring with a significantly higher probability in men than in women, while acute trauma occured with a significantly higher probability (56.25%).

Inflammation as the fourth among the most common causes of enucleation accounted for 2.90% of all enucleations.

REFERENCES

1. *Tahri H, Benatya AD, Chefchaouni CM, El Bakkali M, Berraho A.* Enucleations: epidemiologic investigation in Morocco, presentation of 183 cases. Bull Soc Belge Ophtalmol 2004; (292): 31−4. (French)
2. *Epee E, Masanganise R.* The rate of and indications for enucleations at Sekuru Kaguvi Eye Unit in Harare: a comparative analysis. Cent Afr J Med 2003; 49(1–2): 13–15.
3. *Kaimbo K.* Causes of enucleationin Zaire. J Fr Ophthalmol 1988; 11(10): 677–680. (French)
4. *Shapiro A, Monselise MB.* Destructive ophthalmic procedures, a comparison between a developed and a developing country. Albrecht Von Graefes Arch Klin Exp Ophthalmol 1978; 207(4): 271−3.
5. *Bekibele CO, Oluwasola AO.* A clinicopathological study of orbito-ocular diseases in Ibadan between 1991–1999. Afr J Med Med Sci 2003; 32(2): 197−202.
6. *Haile M, Alemeyehu W.* Causes of removal of the eye in Ethiopia. East Afr Med J 1995; 72(11): 735−8.
7. *Dada T, Ray M, Tandon R, Vajpayee RB.* A study of the indications and changing trends of evisceration in North India. Clin Experiment Ophthalmol 2002; 30(2): 120−3.
8. *Spraul CW, Grossniklaus HE.* Analysis of 24,444 surgical specimens accessioned over 55 years in an ophthalmic pathology laboratory. Int Ophthalmol 1997−1998; 21(5): 283−304.
9. *Gunalp I, Gunduz K, Ozkan M.* Causes of enucleation: a clinicopathological study. Eur J Ophthalmol 1997; 7(3): 223−8.
10. *Scat Y, Liotet S, Bellefqih S.* Etiology of enucleations.Apropos of 3.246 cases. J Fr Ophtalmol 1996; 19(4): 242−7.
11. *Obuchowska I, Sherkawey N, Elmdhm S, Mariak Z, Stankiewicz A.* Clinical indications for enucleation in the material of Department of Ophthalmology, Medical Academy in Białystok in the years 1982–2002. Klin Oczna 2005; 107(1−3): 75−9. (Polish)
12. *Hansen AB, Petersen C, Heegaard S, Prause JU.* Review of 1028 bulbar eviscerations and enucleations. Changes in aetiology and frequency over a 20-year period. Acta Ophthalmol Scand 1999; 77(3): 331−5.
13. *Saeed MU, Chang BY, Khandwala M, Shivane AG, Chakrabarty A.* Twenty year review of histopathological findings in enucleated/eviscerated eyes. J Clin Pathol 2006; 59(2): 153−5.
14. *Erie JC, Nevitt MP, Hodge D, Ballard DJ.* Incidence of enucleation in a defined population. Am J Ophthalmol 1992; 113(2): 138−44.
15. *Stiebel H, Sela M, Pe'er J.* Changing indications for enucleations in Hadassah University Hospital, 1960-1989. Ophthalmic Epidemiol 1995; 2(3): 123−7.
16. *Naumann GD, Portwich E.* Etiology and final clinical cause for 1000 enucleations. (A clinico-pathologic study) (author's transl). Klin Monbl Augenheilkd 1976; 168(05): 622−30. (German)
17. *Davanger M.* Causes of enucleation in Uganda. Br J Ophthalmol 1970; 54(4): 252−5.
18. *Lim JK, Cinotti AA.* Causes for removal of the eye: a study of 890 eyes. Ann Ophthalmol 1976; 8(7): 865−9.
19. *de Gottrau P, Holbach LM, Naumann GO.* Clinicopathological review of 1146 enucleations (1980–90). Br J Ophthalmol 1994; 78(4): 260−5.
20. *Cheng GY, Li B, Li LQ, Gao F, Ren RJ, Xu XL, Jonas JB.* Review of 1375 enucleations in the TongRen Eye Centre, Beijing. Eye (Lond) 2008; 22(11): 1404−9.
21. *Green MD, Apel AJ, Naduvilath T, Stapleton FJ.* Clinical outcomes of keratitis. Clin Experiment Ophthalmol 2007; 35(5): 421−6.
22. *Gyasi ME, Amoaku WM, Adjuik M.* Causes and incidence of destructive eye procedures in north-eastern ghana. Ghana Med J 2009; 43(3): 122−6.
23. *Setlur VJ, Parikh JG, Rao NA.* Changing causes of enucleation over the past 60 years. Graefes Arch Clin Exp Ophthalmol 2010; 248(4): 593−7.
24. *Frenkel S, Hendler K, Pe'er J.* Uveal melanoma in Israel in the last two decades: characterization, treatment and prognosis. Isr Med Assoc J 2009; 11(5): 280−5.

25. *Olinevich VB, Gantsovskiĭ PI, Mamikonian VR, Tsvetkova IV*. A surgical treatment for neovascular terminal glaucoma (preliminary communication). Vestn Oftalmol 2008; 124(4): 5−7. (Russian)
26. *Gaton DD, Ehrlich R, Muzmacher L, Hamel N, Lusky M, Weinberger D*. Enucleations and eviscerations in a large medical center between the years 1981 and 2007. Harefuah 2008; 147(10): 758−62, 840. (Hebrew)
27. *Constantinou M, Jhanji V, Tao LW, Vajpayee RB*. Clinical review of corneal ulcers resulting in evisceration and enucleation in elderly population. Graefes Arch Clin Exp Ophthalmol 2009; 247(10): 1389−93.

Received on January 28, 2011.
Revised on August 31, 2011.
Accepted on September 19, 2011.

ORIGINAL ARTICLE

UDC: 616.135-08
DOI: 10.2298/VSP1301032D

Endovascular treatment of thoracic aortic diseases

Endovaskularno lečenje oboljenja grudne aorte

Lazar Davidović*, Miodrag Jevtić†, Djordje Radak‡, Dragan Sagić‡, Ivan Marjanović†, Igor Končar*, Momčilo Čolić*, Siniša Rusović†, Želimir Antonić‡

***Clinic for Vascular and Endovascular Surgery, Clinical Center of Serbia, Belgrade, Serbia; †Clinic for Vascular Surgery, Military Medical Academy and Faculty of Medicine of the Military Medical Academy, University of Defence, Belgrade; Serbia; ‡Clinic for Vascular Surgery, Institute for Cardiovascular Disaeses "Dedinje", Belgrade, Serbia**

Abstract

Bacground/Aim. Endovascular treatment of thoracic aortic diseases is an adequate alternative to open surgery. This method was firstly performed in Serbia in 2004, while routine usage started in 2007. Aim of this study was to analyse initial experience in endovacular treatment of thoracic aortic diseses of three main vascular hospitals in Belgrade – Clinic for Vascular and Endovascular Surgery of the Clinical Center of Serbia, Clinic for Vascular Surgery of the Military Medical Academy, and Clinic for Vascular Surgery of the Institute for Cardiovascular Diseases "Dedinje". **Methods.** Between March 2004. and November 2010. 41 patients were treated in these three hospitals due to different diseases of the thoracic aorta. A total of 21 patients had degenerative atherosclerotic aneurysm, 6 patients had penetrating aortic ulcer, 6 had posttraumatic aneurysm, 4 patients had ruptured thoracic aortic aneurysm, 1 had false anastomotic aneurysm after open repair, and 3 patients had dissected thoracic aneurysm of the thoracoabdominal aorta. In 15 cases the endovascular procedure was performed as a part of the hybrid procedure, after carotid-subclavian bypass in 4 patients and subclavian artery transposition in 1 patient due to the short aneurysmatic neck; in 2 patients iliac conduit was used due to hypoplastic or stenotic iliac artery; in 5 patients previous reconstruction of abdominal aorta was performed; in 1 patient complete debranching of the aortic arch, and in 2 patients visceral abdominal debranching were performed. **Results.** The intrahospital mortality rate (30 days) was 7.26% (3 patients with ruptured thoracic aneurysms died). Endoleak type II in the first control exam was revealed in 3 patients (7. 26%). The patients were followed up in a period of 1–72 months, on average 29 months. The most devastating complication during a follow-up period was aortoesofageal fistula in 1 patient a year after the treatment of posttraumatic aneurysm. Conversion was performed with explantation of stent-graft and open aortic *in situ* recontruction, followed by esophagectomy and the creation of cervical and gastrical stoma. **Conclusion.** Having in mind initial results of the 3 main vascular clinics in Belgrade, Serbia, economical situation in our country, as well as the published international results, endovascular treatment of thoracic aortic diseases is indicated in hemodinamicaly unstable patients with acute traumatic aneurysm, or in stabile patients older than 65, as well as in case of chronic diseases of the thoracic aorta in patients with significant comorbid conditions or in patients older than 65 years. Endovascular procedures on the thoracic aorta could be performed, hower, only in high-volume centers with experience in routine open surgery of thoracic aorta.

Key words:
aorta, thoracic; aortic diseases; aortic aneurysm; vascular surgical procedures; treatment otucome; mortality.

Apstrakt

Uvod/Cilj. Endovaskularno lečenje oboljenja grudne aorte postaje adekvatna alternativa otvorenom hirurškom pristupu. Ova nova metoda u Srbiji je izvedena prvi put 2004. godine, a rutinski se izvodi od 2007. godine. Cilj ovog rada bio je prikaz zajedničkih početnih iskustava u endovaskularnom lečenju oboljenja grudne aorte Klinike za vaskularnu hirurgiju Vojnomedicinske akademije, Instituta za kardiovaskualrne bolesti „Dedinje" i Klinike za vaskularnu i endovaskularnu hirurgiju Kliničkog centra Srbije. **Metode.** Od marta 2004. do polovine novembra 2010. godine operisan je ukupno 41 bolesnik, zbog različitih oboljenja grudne aorte. Dvadeset jedan bolesnik imao je degenerativnu aneurizmu grudne aorte, šest bolesnika imalo je penetrantni aortni ulkus, šest traumatsku aneurizmu istmičnog dela grudne aorte, četiri rupturiranu aneurizmu grudne aorte, jedan anastomotičnu pseudoaneurizmu koja je nastala nakon klasičnog hirurškog lečenja aneurizme grudne aorte, a tri bolesnika disekantnu aneurizmu torakoabdominalne aorte. Kod 15 bolesnika endovaskularna procedura bila je moguća jedino u sklopu hibridne procedure – kod pet bolesnika sa kratkim

Correspondence to: Lazar Davidović, Clinic for Vascular and Endovascular Surgery, Clinical Center of Serbia, Belgrade, K. Todorovića 8, 11 000 Belgrade, Serbia. Phone: +381 11 3065 176. E-mail: davidovic.lazar@gmail.com

vratom učinjena je transpozicija potključne arterije (četiri bolesnika) i karotido-supklavijalni bajpas (kod jednog bolesnika); dva ilijačna konduita kod bolesnika sa neadekvatnim ilijačnim i/ili femoralnim arterijama; pet rekonstrukcija abdominalne aorte zbog udruženog oboljenja ovog segmenta; jedan „debrančing“ aortnog luka i dva „debrančinga“ abdominalne aorte. **Rezultati.** U prvih 30 dana zabeležena su tri (7,26%) smrtna ishoda. U sva tri slučaja radilo se o bolesnicima koji su imali rupturu aneurizme grudne aorte. Endolik tipa II zabeležen je kod tri (7,26%) bolesnika koji su lečeni konzervativno, s obzirom na to da nije bilo uvećanja aneurizmatske kese. Bolesnici su bili praćeni od 1 do 72 meseca, prosečno 29 meseci. Najozbiljnija komplikacija tokom perioda praćenja bila je aortoezofagealna fistula kod jednog bolesnika. Izvršena je konverzija tokom koje je odstranjen endovaskularni graft. U istom operativnom aktu urađena je ezofagektomija, rekonstrukcija aorte na standardan način, cerviko- i gastrostoma. **Zaključak.** Imajući u vidu početne rezultate, ekonomske mogućnosti našeg društva, kao i objavljene rezultate najvećih svetskih serija, endovaskularno lečenje oboljenja torakalne aorte indikovano je u slučaju akutne traumatske aneurizme hemodinamski nestabilnih bolesnika ili prisutne politraume, odnosno hemodinamski stabilnih bolesnika koji su stariji od 65 godina, kao i u slučaju hroničnih traumatskih ili degenerativnih oboljenja grudne aorte kod bolesnika sa značajnim komorbiditetom ili kod bolesnika starijih od 65 godina. Endovaskularne procedure na grudnoj aorti, međutim, mogu izvoditi samo ustanove koje se rutinski bave otvorenom hirurgijom grudne aorte u uslovima ekstrakorporalne cirkulacije.

Ključne reči:
aorta; aorta, bolesti; aorta, aneurizma; hirurgija, vaskularna, procedure; lečenje, ishod; mortalitet.

Introduction

In the last decades we have faced an increased incidence of all diseases of the thoracic aorta –degenerative, traumatic and dissected aneurysms, penetrating aortic ulcers (PAU). Some of them (dissected) are more frequent among middle-aged, patients or in very young patients (traumatic). Besides being medical burdery these diseases are the economic burden to society [1]. Early results of the treatment of these diseases has been improved with introduction of endovascular procedures [2].

First endovascular surgery on the thoracic aorta [thoracic endovascular aneurysn repair – (TEVAR)] in Serbia was performed in 2004 at the Institute for Cardiovascular Diseases (ICVD) “Dedinje”. However, these procedures have been routinly performed in Serbia since 2007.

The aim of this study was to present the first initial experience in thoracic aortic diseases treatment in the three main vascular hospitals in Belgrade – Clinic for Vascular Surgery of the Military Medical Academy, Clinic for Vascular Surgery of ICVB “Dedinje” and Clinic for Vascular and Endovascular Surgery of the Clinical Center of Serbia.

Methods

From 2007 to December 2010, 41 patients were treated with TEVAR due to different diseases of the thoracic aorta. The average age of the treated patients was 72.43 years. Twenty one (51.29%) patients had degenerative aneurysm of the thoracic aorta, 6 (14.63%) patients was operated for PAU, 6 (14.63%) patients had traumatic (acute 1 patient or chronic 5 patients). Four (9.75%) patients had ruptured thoracic aneurysm, and 1 (2.43%) patient had anastomotic aneurysm after open treatment, and 3 (7.26%) patients had dissected aneurysm of thoracoabdominal aorta (Table 1). Indications for endovascular treatment were significant cardiorespiratory comorbid condition, hostile thoracic cavity and older age.

General anesthesia was applyed in 10 (24.39%) patients, and epidural in 31 (75.61%) patients. Valiant® (Medtronic, Santa Rosa, CA, USA), TAG® (Gore), and Relay® (Bolton Medical) stent grafts were used in 36 (87.8%), 4 (9.75%) and 1 (2.43%) patient, respectively.

Table 1
Demographic characteristics, the types of thoracic aortic diseases and the treatment based on the two-stage hybrid procedures

Variables	Patients n (%)
Sex	
male	38 (92.74)
female	3 (7.26)
Average age (years)	72.43
Type of the disease	
degenerative aneurysm	21 (54.00)
penetrating aortic ulcer	6 (15.58)
traumatic aneurysm	6 (15.58)
anastomotic aneurysm	1 (2.43)
ruptured aneurysm	1 (2.43)
dissected aneurysm	3 (7.26)
The hybrid procedure	
subclavian transposition	4 (9.72)
carotid-subclavian bypass	1 (2.43)
iliac conduit	2 (4.86)
abdominal aortic reconstruction	5 (12.15)
aortic arch debranching	1 (2.43)
visceral debranching	2 (4.86)

Results

Figure 1 shows penetrating aortic ulcer before (A) and after TEVAR (B), Figure 2 aortic dissection type B before (A) and after TEVAR (B), and Figure 3 shows traumatic aneurysm of the isthmic segment of the thoracic aorta before (A) and after TEVAR (B), too.

The procedure TEVAR was performed as a part of the two-stage hybrid procedure in 15 (36.45%) patients (Table 1). Before TEVAR, due to the short aneurysmal neck, subclavian transposition was performed in 4 patients and carotid-subclavian bypass in the 1 patient; due to hypoplastic

or stenotic iliac or femoral artery iliac conduit was performed in two cases; 5 open reconstructions of the abdominal aorta; 1 aortic arch debranching and two visceral debranching procedures were also performed. Different kinds of two-stage hybrid procedures are shown in Figures 4–6.

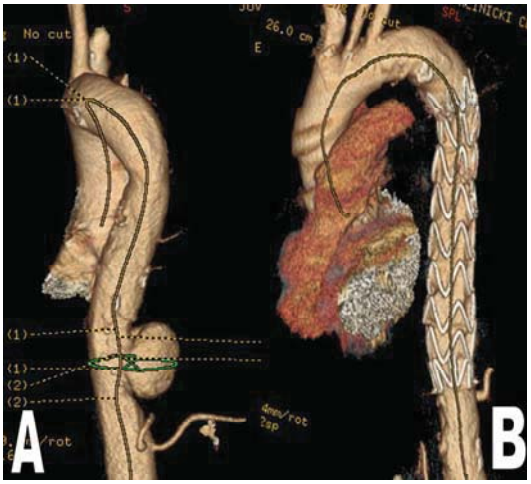


Fig. 1 – Penetrating aortic ulcer before (A) and after thoracic endovascular aneurysm repair (TEVAR) (B)

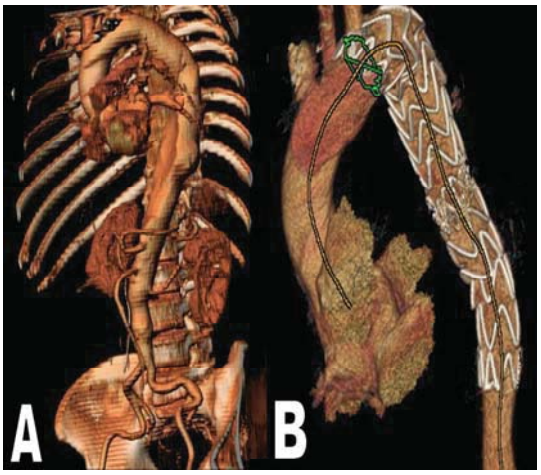


Fig. 2 – Aortic dissection type B before (A) and after thoracic endovascular aneurysm repair (TEVAR) (B)

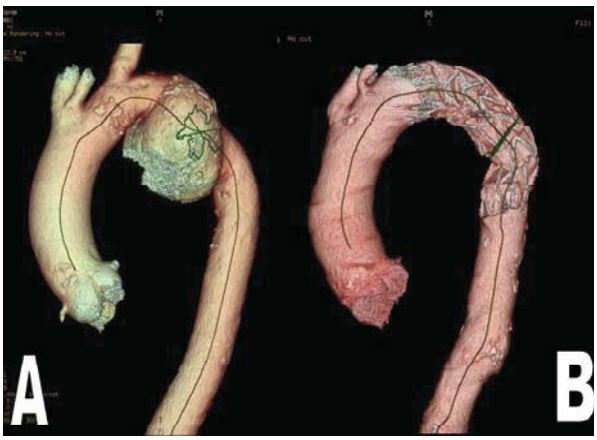


Fig. 3 – Traumatic aneurysm of the isthmic segment of thoracic endovascular aneurysm repair (TEVAR) of the thoracic aorta before (A) and after TEVAR (B)

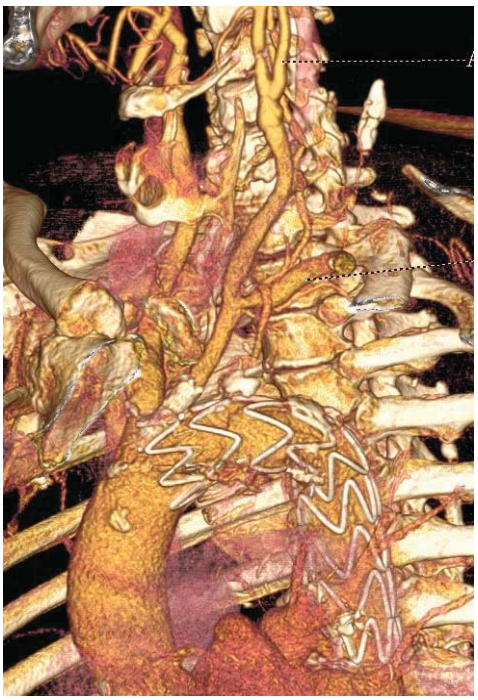


Fig. 4 – Multislice computed tomography (MSCT) angiography after thoracic endovascular aneurysm repair (TEVAR) – stenting of the left common carotid artery and left subclavian transposition

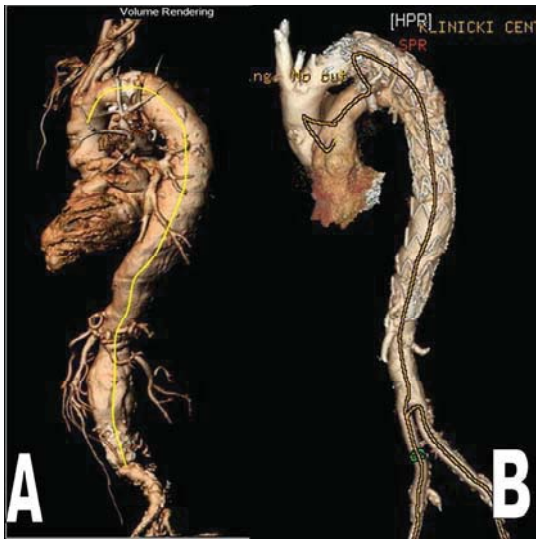


Fig. 5 – Multislice computed tomography (MSCT) angiograhy of a patient at high risk for complete open repair of thoracoabdominal aneurysm type II (Crawford classification) – the visceral part of the abdominal aorta repaired in the first stage (A), and proximal thoracic aneurysm repair with a stent-graft in the second stage (B)

In the first 30 postoperative days the 3 (7.26%) patients died. All these patients were treated for ruptured thoracic aneurysm. Endoleak type II was encountered in 3 (7.26%) patients with no other complications. All the patients were followed up 1–72 months, on average 29 months. Persistent endoleak type II was registered in 2 patients but without increasing aneurysm diameter. One patient had the most devas-

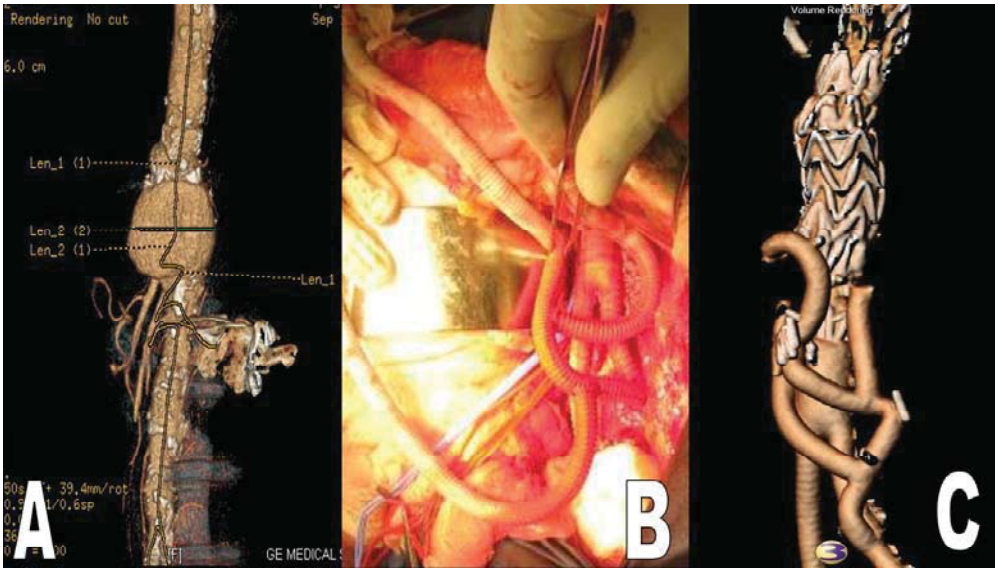


Fig. 6 – (A) Multislice computed tomography (MSCT) angiography shows a distal thoracic and suprarenal aneurysm in high risk patients; (B) The first stage of procedure was infrarenal aortic repair with bifurcated graft, with bypass from the left limb to all the four visceral branches; (C) Ten days later, aneurysm was excluded with a stent-graft.

tating complication – aortoesophageal fistula (AEF) developed 1 year after the TEVAR procedure due to posttraumatic thoracic aneurysm. AEF was treated with explantation of stent-graft and open *in situ* aortic reconstruction and omentoplasty, followed by esophagectomy with cervico– and gastrostoma. This procedure was complicated with aortobronchial fistula in the early postoperative recovery period treated with another stent-graft implantation. The patient was discharged after several months of care in order to be prepared for coloplasty, however, in the meantime he passed away in caohexic state due to malnutrition, with no signs of a new graft infection.

Discussion

Conventional open treatment of thoracic aortic disease comparing to abdominal aortic diseases is a far more complex procedure due to necessity to protect and perfuse the spinal cord and viscera [3, 4]. The TEVAR procedure brought a significant improvement in treatment of these pathology, especially in high risk patients [5–9]. However, TEVAR is limited by anatomical and morphological conditions or the thoracic aorta close to the aortic arch or to the visceral region of aorta [10]. Some of the limitations could be avoided, with some adjuvant procedures. In 4 patients we performed subclavian transposition, and carotid-subclavian bypass in 1 patient due to the short aneurysmal neck. Subclavian artery origin covering could cause arm, brain or spinal cord ischemia [10]. In patients with ruptured thoracic aneurysm covering of the subclavian artery origin was complicated by stroke, coma and death. In case of more proximal extention of aneurysm into the aortic arch, safe stent-graft implantation is possible only after previous "debranching" procedure (revascularization of the supraaortic branches with anatomical or extraanatomical reconstruction) [11]. A patient from our study suffered fatal stroke on the third postoperative day following the successfull anatomical debranching procedure.

Thoracic stent-graft safe implantation is possible if aortoiliac and femoral segments provide a diameter more than 7 mm, no sever tortuosity or anerysmatic dilatation with intraluminal thrombus at risk of embolization [12]. In 2 patients we performed iliac conduit, and in 5 patients we performed reconstrucion of the abdominal aorta in the first stage to secure safe passage of a delivery system.

Aortic infection is a contraindication for stent-graft implantation [13–15]. In 1 patient stent-graft infection was treated with open *in situ* reconstruction.

Inadequate endograft fixation can be the cause of endoleak type I [16]. The other types of endoleaks are the consequence of retrograde flow from the intercostal arteries, inadequate sealing between the graft components or fractures of stent-graft matherial or armature. Spinal cord ishemia is always a concern when thoracic aortic disease is to be treated. Risk increases with covering the subclavian artery, long segment of the thoracic aorta, if the abdominal aorta is already reconstructed or hypogastric and the lumbal arteries occluded [17]. In all our patients with these risk factors we performed preventive measures for keeping perfusion pressure with the middle systemic pressure above 100 mmHg, cerebrospinal fluid drainage and previous revascularization of vertebral or hypogastric bed. There were no episodes of spinal cord ishemia in our patients.

Long-term complications after TEVAR are still under investigations. One of the most devastating complication is aortic graft infection with fistulization to surrounding organs, the esophagus and the bronchus [15, 18]. Open treatment of these complications is one of the options and our patient suffered early aortobronchial fistula after the treatment. There is stil no consesus about the best treatment options.

Stent-graft migration is also a possible early or long-term complication requiring correction [19]. Younger patients with traumatic injuries are more prone to this complication because of their arch anatomy, and because of the estimated

long-term survival as well as due to aortic growth rate [20]. All these reasons should be kept in mind when selecting the method of treatment of acute aortic injury and stent-graft diameter because hypotension of these patients could reduce a measured aortic diameter. TEVAR of dissected aneurysm offers promising results only if treated by criteria that already exist for aneurysms – if there is a sufficient proximal and distal landing zone which is rare. For these purposes, the authors give algorithm of thoracic aortic disease treatment in Figure 7.

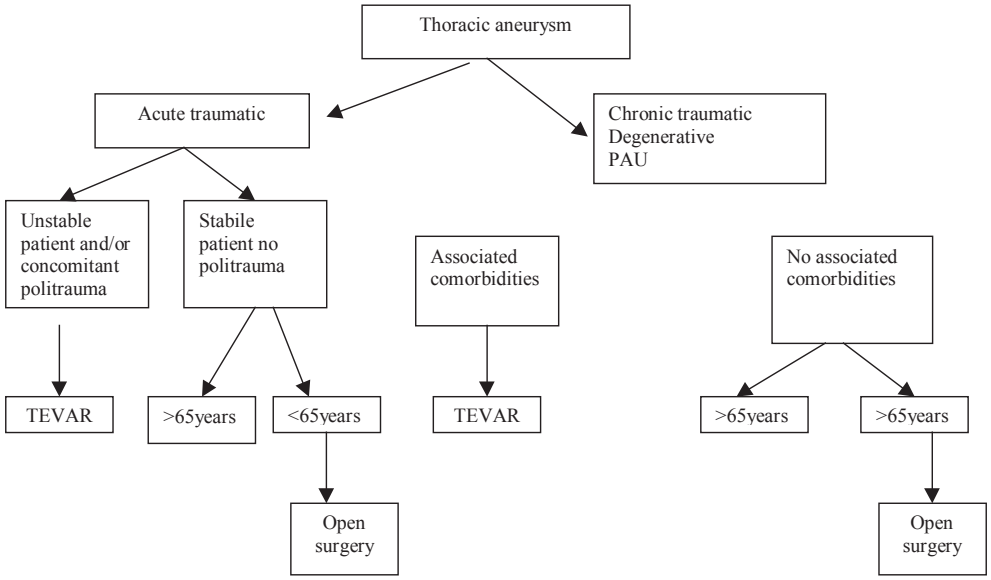


Fig. 7 – The algorithm for thoracic aneurysm treatment
PAU – penetrating aortic ulcer; TEVAR – thoracic endovascular aneurysm repair

Conclusion

In cases with acute traumatic injury of the thoracic aorta in hemodinamically unstable or politraumatized patients or patients older than 65 years, TEVAR is an acceptable method. In cases with chronic diseases of the thoracic aorta in high risk patients TEVAR is indicated, as well as in patients older than 65 years.

TEVAR safe and secure perfomance and its adjuvant procedures, as well as treatment of all complications is, however, possible only in high-volume centers with prevous experience in open treatment of thoracic aortic diseses.

R E F E R E N C E S

1. *Davidović L, Marković M, Čolić M, Ilić N, Končar I, Cvetković S*, et al. Treatment of traumatic rupture of the thoracic aorta. Srp Arh Celok Lek 2008; 136(9−10): 498−504. (Serbian)
2. *Volodos' NL, Karpovich IP, Shekhanin VE, Troian VI, Iakovenko LF.* A case of distant transfemoral endoprosthesis of the thoracic artery using a self-fixing synthetic prosthesis in traumatic aneurysm. Grudn Khir 1988; (6): 84−6. (Russian)
3. *Davidovic LB, Ilic N, Koncar I, Dragas M, Markovic M, Sindjelic R,* et al. Some technical considerations of open thoracoabdominal aortic aneurysm repair in a transition country. Vascular 2011; 19(6): 333−7.
4. *Kieffer E, Leschi JP, Chiche L.* Open repair of chronic posttraumatic aneurysms of the aortic isthmus: the value of direct aortoaortic anastomosis. J Vasc Surg 2005; 41(6): 931−5; discussion 935.
5. *Buz S, Zipfel B, Mulahasanovic S, Pasic M, Weng Y, Hetzer R.* Conventional surgical repair and endovascular treatment of acute traumatic aortic rupture. Eur J Cardiothorac Surg 2008; 33(2): 143−9.
6. *Marty-Ané CH, Berthet JP, Branchereau P, Mary H, Alric P.* Endovascular repair for acute traumatic rupture of the thoracic aorta. Ann Thorac Surg 2003; 75(6): 1803−7.
7. *Rousseau H, Dambrin C, Marcheix B, Richeux L, Mazerolles M, Cron C,* Watkinson A, et al. Acute traumatic aortic rupture: a comparison of surgical and stent-graft repair. J Thorac Cardiovasc Surg 2005; 129(5): 1050−5.
8. *Xenos ES, Abedi NN, Davenport DL, Minion DJ, Hamdallah O, Sorial EE,* et al. Meta-analysis of endovascular vs open repair for traumatic descending thoracic aortic rupture. J Vasc Surg 2008; 48(5): 1343−51.
9. *Demers P, Miller C, Scott Mitchell R, Kee ST, Lynn Chagonjian RN, Dake MD.* Chronic traumatic aneurysms of the descending thoracic aorta: mid-term results of endovascular repair using first and second-generation stent-grafts. Eur J Cardiothorac Surg 2004; 25(3): 394−400.
10. *Carroccio A, Ellozy S, Spielvogel D, Marin ML, Hollier L.* Endovascular stent grafting of thoracic aortic aneurysms. Ann Vasc Surg 2003; 17(4): 473−8.
11. *Drinkwater SL, Böckler D, Eckstein H, Cheshire NJ, Kotelis D, Wolf O,* et al. The visceral hybrid repair of thoraco-abdominal aortic aneurysms--a collaborative approach. Eur J Vasc Endovasc Surg 2009; 38(5): 578−85.
12. *Abu-Ghaida AM, Clair DG, Greenberg RK, Srivastava S, O'hara PJ, Ouriel K.* Broadening the applicability of endovascular aneurysm repair: the use of iliac conduits. J Vasc Surg 2002; 36(1): 111−7.
13. *Kieffer E, Chiche L, Gomes D.* Aortoesophageal fistula: value of in situ aortic allograft replacement. Ann Surg 2003; 238(2): 283−90.

14. *Campagna AC, Wehner JH, Kirsch CM, Semba CP, Kagawa FT, Jensen WA,* et al. Endovascular stenting of an aortopulmonary fistula presenting with hemoptysis. A case report. J Cardiovasc Surg (Torino) 1996; 37(6): 643−6.
15. *Porcu P, Chavanon O, Sessa C, Thony F, Aubert A, Blin D.* Esophageal fistula after endovascular treatment in a type B aortic dissection of the descending thoracic aorta. J Vasc Surg 2005; 41(4): 708−11.
16. *Buth J, Harris PL, Hobo R, van Eps R, Cuypers P, Duijm L,* et al. Neurologic complications associated with endovascular repair of thoracic aortic pathology: Incidence and risk factors. a study from the European Collaborators on Stent/Graft Techniques for Aortic Aneurysm Repair (EUROSTAR) registry. J Vasc Surg 2007; 46(6): 1103−10; discussion 1110−1.
17. *Conrad MF, Cambria RP.* Contemporary management of descending thoracic and thoracoabdominal aortic aneurysms: endovascular versus open. Circulation 2008; 117(6): 841−52.
18. *Hinchliffe RJ, Krasznai A, Schultzekool L, Blankensteijn JD, Falkenberg M, Lönn L,* et al. Observations on the failure of stent-grafts in the aortic arch. Eur J Vasc Endovasc Surg 2007; 34(4): 451−6.
19. *Cheng D, Martin J, Shennib H, Dunning J, Muneretto C, Schueler S,* et al. Endovascular aortic repair versus open surgical repair for descending thoracic aortic disease a systematic review and meta-analysis of comparative studies. J Am Coll Cardiol 2010; 55(10): 986−1001.
20. *Mitchell RS, Ishimaru S, Ehrlich MP, Iwase T, Lauterjung L, Shimono T*, et al. First International Summit on Thoracic Aortic Endografting: roundtable on thoracic aortic dissection as an indication for endografting. J Endovasc Ther 2002; 9 Suppl 2: II98−105.

Received on February 2, 2011.
Accepted on May 5, 2011.

ORIGINAL ARTICLE

UDC: 615.035.4::616-006-085.065-084-092.9
DOI: 10.2298/VSP110905041D

Efficacy of amifostine in protection against doxorubicin-induced acute cardiotoxic effects in rats

Efikasnost amifostina u zaštiti od akutnih kardiotoksičnih efekata doksorubicina kod pacova

Viktorija Dragojević-Simić*†, Silva Dobrić†‡, Vesna Jaćević†‖, Dubravko Bokonjić†‖, Ivica Milosavljević†§, Aleksandra Kovačević*†, Dragan Mikić†¶

***Center for Clinical Pharmacology, ‡Institute for Scientific Information, ‖National Poison Control Center, §Center for Pathology and Forensic Medicine, ¶Clinic for Infectious and Tropical Diseases, Military Medical Academy, Belgrade, Serbia; †Faculty of Medicine of the Military Medical Academy, University of Defence, Belgrade, Serbia**

Abstract

Background/Aim. Amifostine (AMI) is a broad-spectrum cytoprotector which protects against variety of radio- and chemotherapy-related toxicities without decreasing their antitumor action. The aim of the study was to investigate the potential protective effects of AMI against acute cardiotoxic effects of doxorubicin (DOX) in male Wistar rats. **Methods**. AMI (300 mg/kg *ip*) was given 30 min before DOX (6 mg/kg and 10mg/kg b.w., *iv*). The evaluation of DOX-induced cardiotoxic effects, as well as cardioprotective efficacy of AMI was performed 48 h after their administration by determining serum activities of enzymes known to be markers of cardiac damage (creatine kinase – CK, aspartate aminotransferase – AST, lactate dehydrogenase – LDH, and its isoenzyme α-hydroxybutirate dehydrogenase – α-HBDH), as well as the histopathological and ultrastructural analysis of the heart tissue. **Results**. AMI successfully prevented a significant increase in serum activity of CK, AST, LDH and α-HBDH in animals treated with DOX in the dose of 6 mg/kg (121.14 ± 18.37 *vs* 167.70 ± 44.24; 771.42 ± 161.99 *vs* 1057.00 ± 300.00; 3230.00 ± 1031.73 *vs* 4243.10 ± 904.06; 202.57 ± 42.46 *vs* 294.90 ± 80.20 UI/l, respectively), and ameliorated DOX-induced structural damage of the rat myocardium. Pretreatment with AMI in rats given 10 mg/kg DOX reduced the cardiac damage score (CDS) from 2.62 ± 0.51 to 1.62 ± 0.51, i.e. to the CDS value obtained with the lower dose of DOX (6 mg/kg). The ultrastructural analysis of the rat myocardium showed that AMI successfully protected the sarcolemma of cardiomyocytes and reduced mitochondria damage induced by DOX given in the dose of 6 mg/kg. Besides, capillaries were less morphologically changed and apoptosis of endothelial cells was extremely rare in AMI-protected animals. AMI itself did not cause any prominent changes in the examined parameters in comparison with the control rats. **Conclusion**. AMI provided a significant protection against DOX-induced acute cardiotoxic effects in rats. This finding implies its potential to be a successful cardioprotector in patients treated with DOX due to malignant diseases.

Key words:
amifostine; doxorubicin; heart; drug toxicity; cytoprotection; rats, wistar.

Apstrakt

Uvod/Cilj. Amifostin (AMI) je citoprotektor širokog spektra koji može da spreči ispoljavanje toksičnih efekata radio- i hemioterapije bez smanjenja njihovog antitumorskog dejstva. Cilj ove studije bio je ispitivanje efikasnosti AMI u zaštiti od akutnih kardiotoksičnih efekata citostatika doksorubicina (DOX) kod mužjaka Wistar pacova. **Metode**. AMI (300 mg/kg *ip*) davan je 30 min pre DOX (6 mg/kg i 10 mg/kg *iv*). Ispitivanje toksičnih efekata DOX, kao i kardioprotektivne efikasnosti AMI sprovedeno je 48 sati nakon njihove primene. U tu svrhu određivana je serumska aktivnost enzima, koji su poznati kao markeri oštećenja miokarda (kreatin kinaze – CK, aspartat aminotransferaze – AST, laktat dehidrogenaze – LDH, i njenog izoenzima α-hidroksibutirat dehidrogenaze – α-HBDH), i izvršena je patohistološka i ultrastrukturna analiza tkiva miokarda. **Rezultati**. Amifostin je uspešno sprečio značajno povećanje aktivnosti enzima CK, AST, LDH i α-HBDH u serumu životinja kojima je dat DOX u dozi od 6 mg/kg (121,14 ± 18,37 *vs* 167,70 ± 44,24; 771,42 ± 161,99 *vs* 1057,00 ± 300,00; 3230,00 ± 1031,73 *vs* 4243,10 ± 904,06; 202,57 ± 42,46 *vs* 294,90 ± 80,20 UI/l, redom), dok je kod pacova koji su dobijali DOX u dozi od 10 mg/kg smanjio skor oštećenja miokada sa 2,62 ± 0,51 na 1,62 ± 0,51, odnosno na vrednost skora dobijenu u grupi pacova sa nižom dozom

Correspondence to: Viktorija Dragojević-Simić, Centre for Clinical Pharmacology, Military Medical Academy, Crnotravska 17, Belgrade, Serbia. Phone: +381 11 2663 329, +381 64 8743 066. E-mail: vdragsim@gmail.com

DOX (6 mg/kg). Ultrastrukturna analiza tkiva miokarda pokazala je da je prethodna primena AMI kod pacova koji su dobijali DOX u dozi od 6 mg/kg uspešno zaštitila sarkolemu kardiomiocita i smanjila oštećenje mitohondrija i kapilara, kao i pojavu apoptoze endotelnih ćelija. Sam AMI nije izazvao nikakve značajnije promene u ispitivanim parametrima u poređenju sa intaktnim (kontrolnim) pacovima. **Zaključak**. Amifostin ispoljava značajan kardioprotektivni efekat kod pacova u ranom periodu posle primene pojedinačnih visokih doza DOX. Ovaj nalaz ukazuje na potencijal AMI da bude uspešan kardioprotektor i kod onkoloških bolesnika koji primaju DOX.

Ključne reči:
amifostin; doksorubicin; srce; lekovi, toksičnost; ćelija, zaštita; pacovi, wistar.

Introduction

Doxorubicin (DOX), anthracycline antibiotic, is an important antineoplastic agent due to its high antitumor efficacy in haematological, as well as in solid malignancies. However, adverse effects such as myelosuppresion and development of irreversible cardiotoxicity, manifested as a dilated cardiomiopathy leading to congestive heart failure, limit the use of DOX [1–4].

Although the molecular pathogenesis of DOX cardiotoxicity is still controversial, oxidative stress-based hypothesis involving intramyocardial production of reactive oxygen species (ROS) has gained the widest acceptance [5, 6]. Namely, drug toxicity may ensue through free-radical formation and a subsequent redox cycle with O_2, resulting in the generation of ROS, such as superoxide anions ($O_2^{\cdot-}$), hydroxyl radicals ($OH^{\cdot}$) and hydrogen peroxide. The tissues with less developed antioxidant defenses, such as the heart, are particularly susceptible to injury by DOX-induced oxygen radicals [7, 8]. Cell membrane lipids are the most common substrates for oxidative attack. Once initiated, peroxidation continues and has a progressive course that results in structural and functional changes in the heart tissue.

Since treating cardiac complications is very troublesome and expensive, a variety of efforts have been made to reduce this cardiotoxicity without compromising the antitumor activity of DOX [9–11]. One of them is the administration of the agent that would protect the myocardium from DOX toxicity. Considering the aforementioned mechanism of that toxicity, the approach based on the use of antioxidants, including free radical scavengers, seems to be rational.

Amifostine (AMI) is a broad-spectrum cytoprotective agent, with numerous preclinical and clinical studies suggesting protection against a variety of radio- and chemotherapy-related toxicities, including myelotoxicity, neurotoxicity and nephrotoxicity, without decreasing the antitumor action [12–16]. It is actually a prodrug that cannot protect tissues until dephosphorylated by alkaline phosphatase in the plasma membrane to the active metabolite, WR-1065. Once inside the cell, its protective effects appear to be mediated by scavenging free radicals, hydrogen donation, induction of cellular hypoxia, the liberation of endogenous nonprotein sulfhydrils (mainly glutathione) from their bond with cell proteins, the formation of mixed disulphides to protect normal cells etc. Until now not too many reports have been published concerning the prevention of DOX-induced cardiotoxicity by AMI [17–20].

The present investigation extended these studies. Serum activity of enzymes, known to be markers of compromised cardiomyocyte integrity and histological as well as ultrastructural analysis (UA) of the myocardial tissue were used to estimate the protective efficacy of AMI against DOX-induced acute cardiotoxic effects in rats. High, single doses of DOX, 6 mg/kg and 10 mg/kg b.w., were chosen by taking into account the cumulative DOX dose (450 mg/m^2 body surface or 11 mg/kg b.w.), known to produce potentially lethal cardiomiopathy in humans [21].

Methods

Experimental animals and the protocol

Adult male Wistar rats weighing 200 g to 250 g were used. The animals were housed in plastic cages, five animals per cage, under standard laboratory conditions (room temperature, 12/12 h light/dark cycle, free access to a standard rodent chow and water).

The animals were divided into 6 experimental groups of animals treated as follows:

The group I was the control one (saline, 1 ml/kg *iv*); the group II was treated with AMI (300 mg/kg *ip* 30 min before saline (1 ml/kg *iv*); the group III was treated with 6 mg/kg *iv* of DOX; the group IV was treated with 300 mg/kg *ip* of AMI 30 min before DOX (6 mg/kg *iv*); the group V was treated with 10 mg/kg *iv* of DOX and group the VI was treated with 300 mg/kg *ip* of AMI 30 min before DOX (10 mg/kg *iv*).

The study was based on the Guidelines for Animal Studies no 282-12/2002 (Ethics Committee of the Military Medical Academy, Belgrade, Serbia).

Drugs

AMI was synthesized in the Chemical Department of Military Technical Institute, Belgrade, by original procedure based on the method described by Piper et al. [22], as already published [23]. AMI was prepared for administration by dissolving the substance in sterilized and apyrogenic 0.9% NaCl solution, *ex tempore*. DOX was obtained from commercial sources (Adriblastina®, Hemofarm, Vršac in colaboration with Farmitalia Carlo Erba, Milan, Italy) and was dissolved in the water supplied in the original drug package, immediately prior to injection.

Evaluation of myocardial toxicity and its prevention

Since earlier pathohistological studies have revealed that structural damage of the rat heart occurs within 48 h

after application of 6 and 10 mg/kg of DOX [9, 24] we evaluated the efficacy of the pretreatment with AMI on DOX-induced cardiotoxicity within this period after their administration, according to the study protocol. Blood samples were collected from the caudal vein, just before sacrifice by decapitation under light ether anaesthesia. Hearts were removed rapidly and utilized for histopathological analysis (HA). Each experimental group consisted of 8 animals.

Enzyme assays

Blood samples were centrifuged at 3.000 rpm for 10 minutes. The serum activity of creatine phosphokinase (CK), aspartate aminotransferase (AST), lactate dehydrogenase (LDH) and its isoenzyme α-hydroxybutyrate dehydrogenase (α-HBDH) was determined on an autoanalyser Express 550 (Ciba Corning, Gilford Systems) using the test reagents produced by Randox firm (United Kingdom) and the procedures recommended by the manufacturer.

Histopathological analysis

The removed hearts were fixed in 10% formalin. Transmural tissue samples from the left and right ventricular free walls were embedded in paraffin blocks. Tissue samples 5-μm thick were stained with haematoxylin & eosin (HE) and heart sections were analyzed (20 x and 40x; Olympus-2 microscope; Tokyo, Japan). Grading of the cardiac tissue damages and calculating the cardiac damage score (CDS) were performed by using 0–3 scale as previously described [18], taking into account only myocytes showing cytoplasmic vacuolisation and/or myofibrillar loss. The grading system was as follows: 0 = no damage; 1 = < 5% myocytes damaged, 2 = 16%–25% myocytes damaged; 3 = > 35% myocytes damaged. Per eight hearts from each group were available, and per 5 sections from each heart were analyzed. All morphological examinations were performed by 3 independent observers as a blind study with no prior knowledge of the treatment given to the animals.

Tissue Preparation and Electron Microscopy

Another experiment, according to the same study protocol, has been done for electron microscopy examination. Immediately after the animals were sacrificed sections of the myocardial tissue were taken from the free wall of the left ventricle of each heart and small cubes of tissue were fixed in cold 4% glutaraldehyde with 0.1M sodium cacodylate buffer, at pH 7.2. After washing in the same buffer, the samples were postfixed with 1% osmium tetroxide, during 1 h, on + 4C° and contrasted by uranyl acetate during 24 h. The tissue was dehydrated in graded ethanol, transferred to propylene oxide and embedded in Epon. Sections were cut at 40 - 50 nm with a diamond knife on an LKB ultramicrotome, stained with uranyl acetate and lead citrate, and examined with a Philips 201 C electron microscope. Each experimental group consisted of 5 animals.

Statistical analysis

The Student's *t*-test was used to asses differences in serum enzyme activity. Statistical evaluation of the difference in the severity of cardiac damage score among the various treatment groups was performed by using the Kruskal-Wallis rank test and Mann-Whitney U-test.

Results were considered significant when $p < 0.05$.

Commercial statistical software Stat for Windows, R.4.5., Stat Soft Inc., Tulsa, OK, USA, 1993, was used throughout the study.

Results

Effects of AMI on serum enzyme activity in DOX-treated rats

The assessment of cardiomyocytes integrity in the DOX-treated rats was done by determining the activity of AST, ALT, LDH and its isoenzyme α-HBDH in the serum. Serum activities of these enzymes were significantly increased in animals treated with both doses of DOX (6 and 10 mg/kg *iv*) comparing to those of the control group (Figure 1).

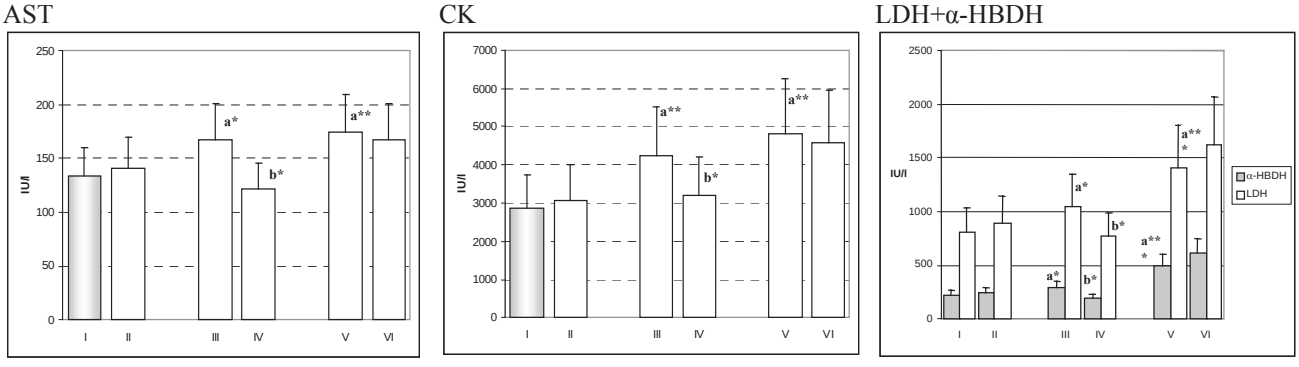


Fig. 1 – Influence of amifostine (AMI, 300 mg/kg *ip*) pretreatment on doxorubicin (DOX)-induced changes in aspartate aminotransferase (AST), creatine kinase (CK), lactate dehydrogenase (LDH) and α-hydroxybutyrate dehydrogenase (α-HBDH) serum activity in rats 48 h after their administration (AMI was given 30 min before *iv* injection of DOX, 6 mg/kg or 10 mg/kg)
I – the control (saline, 1 ml/kg *iv*); II – AMI; III – DOX (6); IV – AMI + DOX (6); V – DOX (10); VI – AMI + DOX (10)
a* , a, a*** – $p < 0.05$; $p < 0.01$; $p < 0.001$ *vs* I; b* - $p < 0.05$ *vs* III**

This increase was successfully prevented when animals were given AMI prior to being treated with DOX in a dose of 6 mg/kg. However, in the group of animals treated with 10 mg/kg of DOX, AMI failed to prevent DOX-induced increase of the serum activity of enzymes known to be markers of cardiomyocytes integrity damage.

On the other hand, AMI given before saline injection had no effect on the monitored parameters (Figure 1).

Effects of AMI on histopathological patterns of the hearts in DOX-treated rats

Light microscopic examination of the myocardium from DOX-treated rats in comparison with that of the control animals is shown in Figure 2. Histopathological analysis of the heart tissue of rats given both tested doses of DOX (6 mg/kg and 10 mg/kg) showed that most of the cardiac muscle cells were regularly arranged. However, in animals treated with 6 mg/kg of DOX a certain number of cardiomyocytes with fine granular cytoplasm, without clearly noticeable nuclei, was detected, some of which had small vacuoles and/or pale appearance of the cytoplasm. In animals pretreated with AMI just a small number of myocytes with fine granular cytoplasm was seen differing from surrounding normal myocardial tissue. The appearance of numerous vacuoles and segmental loss of normal tissue structure was seen in rats treated with 10 mg/kg DOX, while in animals pretreated with AMI more preserved myocardial structure was visible, with less extensive vacuolization of cardiomyocytes (Figure 2). Grading cardiac tissue damages by 0–3 scale in rats, treated with DOX in single doses of 6 and 10 mg/kg, revealed CDS of 1.62 ± 0.51 and 2.62 ± 0.51, respectively. The differences between the control and DOX treated groups were statistically significant (Table 1). In the group of rats treated with 6 mg/kg of DOX which had previously received AMI, myocyte alterations were significantly less severe than those observed in animals without pretreatment ($p < 0.01$). Pretreatment with AMI in rats given DOX in a dose of 10 mg/kg reduced CDS to the value obtained in the group of rats treated with 6 mg/kg of DOX (Table 1).

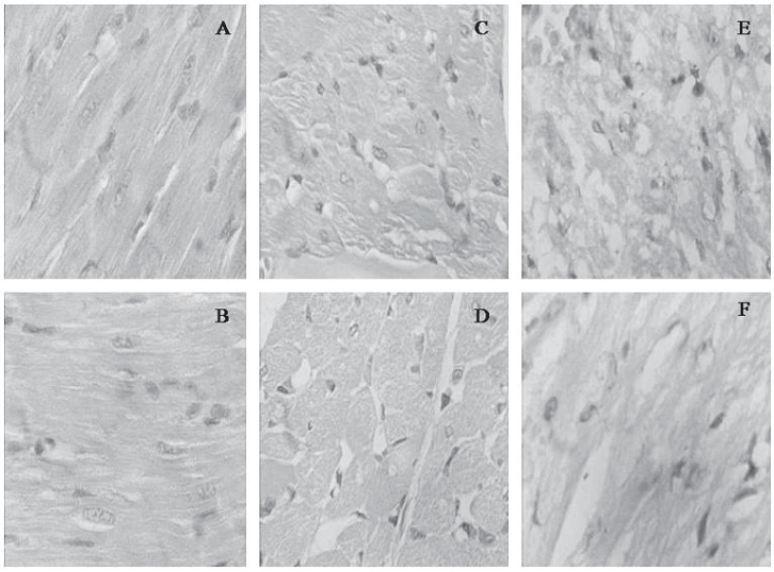


Fig. 2 – Light microscopy of the heart sections: (A) control group – myocardium of normal morphology, (B) group treated with amifostine (AMI) – no histological lesions found (H&E, ×40), (C) group treated with doxorubicin (DOX) 6 mg/kg – small number of myocites with discrete vacuolization, (D) group treated with AMI + DOX 6 mg/kg – a small number of cardiomyocytes with fine granular cytoplasm (H&E, ×20), (E) group treated with DOX 10 mg/kg – appearance of numerous vacuoles and segmental loss of normal tissue structure, (F) group treated with AMI + DOX 10 mg/kg – less extensive vacuolization of cardiomyocytes with more preserved myocardial structure (H&E, ×40)

Table 1

The influence of amifostine on cardiac damage scores (CDS) in rats treated with doxorubicin

Treatment (mg/kg)*	Cardiac damage score (CDS)** (8 hearts x 5 section)				Mean CDS ± SD
	0	1	2	3	
Control (saline, 1 ml/kg *iv*)	30	10	0	0	0.25 ± 0.46
AMI (300)	30	10	0	0	0.25 ± 0.46
DOX (6)	0	15	25	0	1.62 ± 0.51 [a]
AMI (300) + DOX (6)	25	10	5	0	0.50 ± 0.75 [b]
DOX (10)	0	0	15	25	2.62 ± 0.51 [a]
AMI (300) + DOX (10)	0	15	25	0	1.62 ± 0.51 [a b]

***Amifostine (AMI) was administered *ip* 30 min before doxorubicin (DOX) given *iv*; ** CDS: 0 – no damage; 1 – < 5% myocytes damaged; 2–16% to 25% myocytes damaged ; 3 – > 35% myocytes damaged; † Statistical evaluation was performed using Kruskal-Wallis test: [a] $p < 0.001$ *vs* control; Mann-Whitney U test: [b] $p < 0.01$ *vs* corresponding DOX group**

In animals sacrificed 48 h after giving AMI 30 min before saline (1 ml/kg *iv*) no any pathological changes were found, nor CDS was significantly different from that of the control group.

Effects of AMI on ultrastructural alterations of the hearts in DOX-treated rats

Ultrastructural analysis (UA) of the heart sections of rats treated with DOX in a dose of 6 mg/kg showed prominent alterations comparing to those of the control rats (Figure 3a). Cardiomyocytes were transparent, with preserved volume. Nuclei of the cardiomyocytes had an altered shape, with shallow invagination of nucleus membrane and enlarged perinuclear spaces. Mitochondria were numerous, hydropically degenerated with enlarged volume and light matrix. Their cristae were moved to periphery (Figure 3b). Sarcolemma of some cardiomyocytes were locally lysed and mitochondria could be seen out of the cell, in intercellular spaces. Endothelial cells in the capillaries between cardiomyocytes showed changes that could be described as the ones characteristic for programmed cell death – apoptosis. These cells became very thin, with condensed, dark cytoplasm and heavily condensed chromatin filling the majority of caryoplasma. (Figure 3c). In some biopsies rupture of capillary walls could be seen.

In the animals which received AMI before DOX injected in the same dose (6 mg/kg *iv*), structural changes were prominently less expressed, with no lysis of cardiomyocytes sarcolemma. Nuclei of myocytes were, most often, like those in the control animals, while the mitochondria damage was less prominent (Figure 3d). Capillaries were less morphologically changed and apoptosis of endothelial cells was extremely rare.

The application of AMI itself, without DOX, led to discrete changes of the cardiomyocytes comparing to the control animals. Shallow invagination of the nucleus membrane and marginal condensation of heterochromatin were most prominent. Mitochondria with lamellar cristae predominated in this group of animals.

Discussion

The results of this study showed that the serum activity of CK, AST, LDH and its isoenzyme α-HBDH, as the most characteristic marker for cardiac damage, was significantly increased in the animal groups treated with both doses of DOX comparing to the control rats, in a dose- dependant way. The elevation of serum concentrations of examined enzymes is a well-known quantitative index of compromised cellular integrity, and is also considered to be a good indicator of myocardial damage by DOX [25–27]. Formation of free radicals and peroxidation of lipids of cardiomyocyte membranes, including sarcolemma, caused by DOX, is thought to be followed by membrane permeability and other changes of membrane functions. Our findings are in accordance with the results of other authors who showed that increased serum activity of CK and LDH was detected in the period lasting between a few hours and 4 days after the administration of DOX doses ranging from 10 to 20 mg/kg, with the peak at day 2 [10, 27, 28]. It was considered that a damaged sarcolemma enables the enzymes to pass out of the cell, thus accounting for their prominent increase in the serum. This was actually

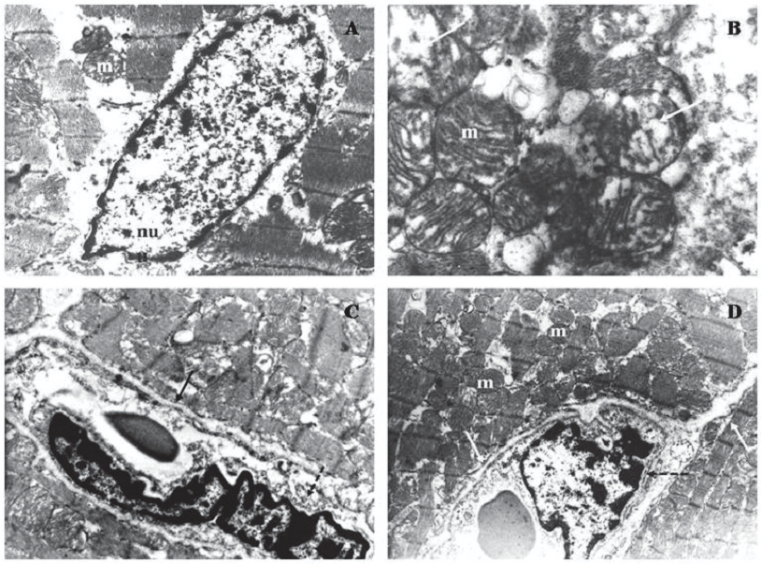


Fig. 3 – Electron micrograph of myocardium from: (A) control group of animals – control heart demonstrating normal peripheral distribution of nuclear chromatin (nu), sarcomeres, and mitochondria (original magnification ×36,000) (B) group of animals treated with doxorubicin (DOX) 6 mg/kg – mitochondria (m) hydropically degenerated with enlarged volume and light matrix (→). Cristae are moved to periphery (original magnification ×67,500). (C) group of animals treated with DOX 6 mg/kg – prominently thin capillary wall (→); endothelial cell nucleus irregularly shaped with increased quantity of heterochromatin (⇒) (original magnification ×23,850) (D) group of animals treated with amifostine (AMI) + DOX 6 mg/kg – mitochondria like in control animals, sarcolemma is preserved (→), capillary endothelial cell nucleus with marginally distributed heterochromatin (⇒) (original magnification ×30,000)
Note: AMI (300 mg/kg *ip*) was given 30 min before DOX

confirmed in our experiment in which the UA of the heart sections of the rats treated with 6 mg/kg of DOX showed that the sarcolemma of some cardiomyocytes was locally lysed and mitochondria could be seen out of the cell, *ie* in intercellular spaces.

On the other hand, HA revealed, taking into account only myocytes showing cytoplasmic vacuolization and/or myofibrillar loss, CDS of 1.62 ± 0.51 and 2.62 ± 0.51 in rats treated with 6 mg/kg and 10 mg/kg of DOX, respectively. The differences between the control and DOX-treated groups were dose-dependent and statistically significant. The myocardial cellular alterations associated with the administration of DOX in our experiments were similar to those reported in previous experimental studies [9, 24, 28, 29]. The affected myocytes displayed two characteristic light microscopic changes: cytoplasmic vacuolization and/or myofibrillar loss. The more myocytes showed these changes, the more pronounced the lesions became. UA of the rat heart 48 h after administration of 6 mg/kg of DOX revealed cardiomyocyte alterations described as oncosis. In parallel with the preserved volume and marginally condensed heterochromatin these cells had hydropically degenerated mitochondria with the light matrix and cristae moved to periphery. This was in accordance with the results of other authors who showed that the earliest and most often changes in the rat heart after application of DOX high doses were cellular oedema and swelling of the mitochondria in cardiomyocytes [28]. It is widely accepted that oncosis, as a type of prelethal changes, is characterized by the loss of cell volume control, typically resulting from adenosine triphosphate (ATP) deficiency and subsequent failure of Na^+-K^+ATPase at the plasmalemma, early clumping of nuclear chromatin, swelling of the mitochondria and dilatation of the endoplasmic reticulum (ER) and Golgi components [30–32]. On the other hand, apoptosis is characterized by cell shrinkage, accompanied by marked cell shape changes with multiple cytoplasmic protrusions and nuclear irregularities with intense chromatin clumping. The cytosol is electron-dense though some ER dilatation and mitochondrial condensation occur. Biochemically, there are both maintenance of ATP in the cell and the increased level of Ca^{2+}. In our experiments apoptotic cardiomyocytes were not observed. That can be explained by the fact that some special stainings, including TUNEL assay, are necessary for their detection. Also, Arola et al. [29] showed that 2 days after *ip* injection of DOX in the dose of 5 mg/kg only 0.033% of cardiomyocytes had TUNEL-positive nuclei (comparing with 0.0065% in control). The current understanding of molecular mechanisms underlying DOX-induced cardiomyocyte type of death, both apoptosis and necrosis, still imply excessive production of ROS. However, it is considered that predominant mechanism of cell death is determined by DOX dosage. Namely, low-dose DOX exposure induced apoptosis whereas high-dose exposure primarily induced oncosis of myocytes [5, 6, 33]. The latter corresponds to our experimental conditions. On the other hand, UA revealed some capillary endothelial cells with morphological changes characterizing apoptosis, in accordance with the results of other authors [34–36].

AMI successfully prevented significant increase of serum activity of all the examined enzymes in animals treated with DOX in a dose of 6 mg/kg. In AMI protected animals myocyte alterations were significantly less severe than those observed in animals without pretreatment. Moreover, the pretreatment with AMI in rats receiving higher dose of DOX (10 mg/kg) reduced CDS to the value obtained in the group of unprotected rats given 6 mg/kg of DOX. UA actually showed that the pretreatment with AMI in rats receiving 6 mg/kg of DOX protected the sarcolemma of cardiomyocytes, and significantly reduced mitochondria damage. Moreover, in the protected rats myocardial capillaries were less morphologically changed and apoptosis of endothelial cells was extremely rare. AMI itself did not cause any changes in all of the examined parameters in comparison with the control rats. Previous *in vitro* studies demonstrated that WR-1065, the active metabolite of AMI, was able to scavenge $OH^{\cdot}$ and $O_2^{\cdot -}$, including DOX-derived $O_2^{\cdot -}$ generated by NADH respiration of heart mitochondria particles [37]. Many studies still support the hypothesis that mitochondria are a primary target of DOX-induced oxidative stress. The fact that typical mitochondrial density per cell unit volume ranges from 25% to 35% in cardiomyocytes may partially explain why DOX is selectively toxic to the heart [38, 39]. AMI is a negative charged thiol which accumulates within the mitochondria and around DNA. These facts explain higher protective potential of AMI compared with that of neutral or positive charged thiols, taking into account some studies using perfused rat hearts which have shown that DOX is localized primarily arround the nucleus and within cell mitochondria [39, 40]. Also, both AMI and WR-1065 significantly reduce DOX-induced heart cell toxicity, measured by ATP content, normalised to the total cellular protein [37]. That can also be explained by their effective protection of mitochondria, as in our study, since oxidative phosphorylation is one of the functions of this organelae which provides a substantial portion of the ATP needed to meet energy demands of the heart. On the other hand, several lines of evidence suggest that AMI is presumably modified by membrane-bound alkaline phosphatase which is highly expressed in the endothelium and transferred into WR-1065. Then, WR-1065 quickly penetrates into cells, and acts as free-radical scavenger protecting them from oxidative damage [13, 14, 41]. Potent protective effects of AMI pretreatment in the model of pulmonary endothelial cell barrier dysfunction *in vitro* were shown. Owing to AMI the attenuation of oxidative stress, NF-κB inflammatory cascade and disruption of endothelial cell adhesions leads to the preservation of endothelial cell monolayer integrity [42]. On the other hand, marked elevation of the expression of antioxidant enzyme manganese superoxide dismutase (MnSOD) gene in human microvascular endothelial cells following their exposure to a WR-1065 can result in elevated resistance to the cytotoxic effects of ionizing radiation. Namely, MnSOD is nuclear-encoded mitochondrial enzyme that scavenges $O_2^{\cdot -}$ in mitochondrial matrix, and has been shown to be highly protective against radiation-induced ROS [43]. Based on the current data, the present authors speculate that succesful AMI protection of DOX-induced damage of heart capillaries,

whose endothelium as a rich source of oxidants contributes a lot to the oxidant-rich environment at that locus in this model, may be mediated by AMI antioxidant properties resulting in downregulation of oxidative stress and redox-sensitive signalling cascades. Bolman et al. [19, 44] have shown that AMI significantly decreases DOX-induced lipid peroxidation (evaluated by malondialdehyde level) and increases the levels of reduced gluthatione (GSH) and catalase activity in the hearts of rats treated by high doses of DOX. According to Luo et al. [26], after the application of DOX, ROS by inducing lipid peroxidation produce cytotoxic aldehydes resulting in inflammatory reactions. This eventually leads to increased synthesis of cytokines, infiltration of mononuclear cells and death of cardiomyocytes. In accordance with this, in our previous experiments the presence of mononuclear cells and fibroblasts was decreased in AMI-protected rats and necrotic myocytes were rare compared with DOX-only treated group [18]. However, the high dose of DOX was a cumulative one, given as a multiple, low, unitary dose regimen, with AMI always preceding DOX. According to that, our own results [18], as well as some others' [45, 46] support the statement that acute and chronic cardiac toxicity of DOX share the same mechanism, implying that chronic toxicity arises from repeated episodes of acute exposure which induces a cumulative damage. However, since single doses of DOX used in this experiment were very high, AMI might produce its cardioprotective effect by some other mechanisms, besides the antioxidative one. For example, it has recently been shown that AMI, given in doses similar to that used in this experiment, produced a strong anti-inflammatory activity [42, 47, 48] that might additionally offer protection against DOX-induced cardiac damage. However, further investigations are needed to confirm this hypothesis.

Conclusion

In summary, the present study demonstrates the potent protective effects of AMI pretreatment against acute cardiotoxic effects of DOX given in single high doses in rats. The obtained results imply the potential of AMI to be a successful cardioprotector in patients treated by DOX due to malignant diseases.

REFERENCES

1. *Sweetman SC*, editor. Martindale: The Complete Drug Reference 36 [CD-ROM]. London: Pharmaceutical Press; 2009.
2. *Chabner BA, Alegra CJ, Curt GA, Calabresi P.* Chemotherapy of neoplastic diseases. In: *Brunton LL, Lazo JS, Parker KL*, editors. Goodman & Gilman's the Pharmacological Basis of Therapeutics. New York: McGraw-Hill; 2006: p. 1315–404.
3. *Singal PK, Iliskovic N.* Doxorubicin-induced cardiomyopathy. N Engl J Med 1998; 339(13): 900–5.
4. *Pai VB, Nahata MC.* Cardiotoxicity of chemotherapeutic agents. Incidence, treatment and prevention. Drug Safety 2000; 22(4): 263–302.
5. *Šimunek T, Šterba M, Popelova O, Adamcova M, Hrdina R, Gerši V.* Anthracycline-induced cardiotoxicity: overview of studies examining the roles of oxidative stress and free cellular iron. Pharmacol Rep 2009; 61(1): 154–71.
6. *Zhang Y, Shi J, Li Y, Wei L.* Cardiomyocyte death in doxorubicin-induced cardiotoxicity. Arch Immunol Ther Exp (Warsz) 2009; 57(6): 435–45.
7. *Keizer HG, Pinedo HM, Schuurhuis GJ, Joenje H.* Doxorubicin (adriamycin): a critical review of free radical-dependent mechanisms of cytotoxicity. Pharm Ther 1990; 47(2): 219–31.
8. *Papadopoulou LC, Theophilidis G, Thomopoulos GN, Tsiftsoglou AS.* Structural and functional impairment of mitochondria in adriamycin-induced cardiomyopathy in mice: suppression of cytochrome C oxidase II gene expression. Biochem Pharmacol 1999; 57(5): 481–9.
9. *Tesoriere L, Ciaccio M, Valenza M, Bongiorno A, Maresi E, Albiero R,* et al. Effects of vitamin A administration on resistance on rat heart against doxorubicin-induced cardiotoxicity and lethality. J Pharm Exp Ther 1994; 269(1): 430–6.
10. *Dobrić S, Dragojević-Simić V, Bokonjić D, Milovanović S, Marinčić D, Jović P.* The efficacy of Selenium, WR-2721, and Their Combination in the Prevention of Adriamycin-Induced Cardiotoxicity in Rats. J Environment Pathol Toxicol Oncol 1998; 17(3–4): 291–9.
11. *Wouters K, Kremer L, Miller T, Herman E, Lipschultz S.* Protecting against anthracycline-induced myocardial damage: a review of the most promising strategies. Br J Haematol 2005; 131(5): 561–78.
12. *Dragojević-Simić V, Dobrić S.* The cytoprotective agent amifostine (WR-2721): current clinical use and trends in its development. Vojnosanit Pregl 1996; 53(4): 305–10 (Serbian)
13. *Spencer CM, Goa KL.* Amifostine: a review of its pharmacodynamic and pharmacokinetic properties, and therapeutic potential as a radioprotector and cytotoxic chemoprotector. Drugs 1995; 50(6): 1001–31.
14. *Capizzi RL.* The preclinical basis for broad-spectrum selective cytoprotection of normal tissue from cytotoxic therapies by amifostine. Semin Oncol 1999; 26(2 Suppl 7): 3–21.
15. *Culy CR, Spencer CM.* Amifostine: an update on its clinical status as a cytoprotectant in patients with cancer receiving chemotherapy or radiotherapy and its potential therapeutic application in myelodysplastic syndrome. Drugs 2001; 61(5): 641–84.
16. *Kouvaris J, Kouloullas V, Vlahos L.* Amifostine: The first Selective-Target and Broad-Spectrum Radioprotector. Oncologist 2007; 12(6): 738–47.
17. *Nazeyrollas P, Prevost A, Baccard N, Manot L, Devillier P, Millart H.* Effects of amifostine on perfused isolated rat heart and on acute doxorubicin-induced cardiotoxicity. Cancer Chemother Pharmacol 1999; 43(3): 227–32.
18. *Dragojević-Simić V, Dobrić S, Bokonjić D, Vucinic Z, Sinovec S, Jacevic V,* et al. Amifostine protection against doxorubicin cardiotoxicity in rats. Anticancer Drugs 2004; 15(2): 169–78.
19. *Bolman Z, Cicek C, Kadikoylu G, Barutca S, Serter M, Yeniseu C*, et al. The Protective Effects of Amifostine on Adriamycin-Induced Acute Cardiotoxicity in Rats. Tohoku J Exp Med 2005; 207(4): 249–53.
20. *Potermski P, Polakowski P, Wiktorowska-Owczarek A, Owczarek J, Pluzanska A, Orszulak-Michalak D.* Amifostine improves hemodynamic parameters in doxorubicin-pretreated rabbits. Pharmacol Rep 2006; 58(6): 966–72.
21. *Doroshow JH.* Doxorubicin-induced cardiac toxicity. N Engl J Med 1991; 324(12): 843–5.
22. *Piper JR, Stringfellow CRJr, Eliot RD, Johnston TP.* S-2-(omega-aminoalkyl amino)ethyl dihydrogen phosphorothioates and related compounds as potential antiradiation agents. J Med Chem 1969; 12(2): 236–43.

23. *Trajković S, Dobrić S, Jaćević V, Dragojević-Simić V, Milovanović Z, Đorđević A.* Tissue-protective effects of fullerenol $C_{60}(OH)_{24}$ and amifostine in irradiated rats. Colloids Surf B Biointerfaces 2007; 58(1): 39–43.
24. *Griesser-Aleksić N, Nikolić V, Bogdanović G, Baltić V, Spasić M.* Prevention of doxorubicin cardiotoxicity with selenium. Onkološki arhiv 1994; 2: 195–7 (Serbian)
25. *Olson HM, Shannon CF.* Alterations of lactate dehydrogenase isoenzymes with subacute adriamycin toxicity. Cancer Treat Rep 1979; 63(11–12): 2057–9.
26. *Luo X, Evrovsky Y, Cole D, Trines J, Benson LN, Lehotay DC.* Doxorubicin-induced acute changes in cytotoxic aldehydes, antioxidant status and cardiac function in rats. Biochem Acta 1997; 1360: 45–52.
27. *Milic-Tores V, Srdjenovic B, Jacevic V, Dragojevic-Simic V, Djordjevic A, Luisa Simplicio A.* Fullerenol $C_{60}(OH)_{24}$ prevents doxorubicin-induced acute cardiotoxicity in rats. Pharmacol Rep 2010; 62(4): 707–18.
28. *Olson HM, Capen CC.* Subacute cardiotoxicity of adriamycin in the rat. Biochemical and ultrastructural investigations. Lab Invest 1977; 37(4): 386–94.
29. *Arola OJ, Saraste A, Pulkki K, Kallajoki M, Parvinene M, Voipio-Pulkki LM.* Acute doxorubicin cardiotoxicity involves cardiomyocyte apoptosis. Cancer Res 2000; 60(7): 1789–92.
30. *Trump BF, Berezesky IK.* Oncosis, apoptosis and necrosis: The role of [Ca2+]i deregulation. Electron Microscopy 1998; IV: 811–2.
31. *Maisch B.* How cardiac cell die-necrosis, oncosis and apoptosis. Herz 1999; 24(3): 181–8.
32. *Buja LM, Vela D.* Cardiomyocyte death and renewal in the normal and diseased heart. Cardiovasc Pathol 2008; 17(6): 349–74.
33. *L'Ecuyer T, Allebban Z, Thomas R, Vander Heide R.* Gluthatione S-transferase overexpression protects against anthracycline-induced H9C2 cell death. Am J Physiol Heart Circ Physiol 2004; 286(6): H2057–64.
34. *Zhang J, Clark JR, Herman EH, Ferrans VJ.* Doxorubicin-induced apoptosis in spontaneously hypertensive rats: differential effects in heart, kidney and intestine, and inhibition by ICRF-187. J Mol Cell Cardiol 1996; 28(9): 1931–43.
35. *Kotamraju S, Konorev EA, Joseph J, Kalayanaraman B.* Doxorubicin-induced apoptosis in endothelial cells and cardiomyocytes is ameliorated by nitrone spin traps and ebselen. J Biol Chem 2000; 275(43): 33585–92.
36. *Warren MC, Bump EA, Medeiros D, Braunhut SJ.* Oxidative stress-induced apoptosis of endothelial cells. Free Rad Biol Med 2000; 29(6): 537–47.
37. *Marzatico F, Porta C, Moroni M, Bertorelli L, Borasio E, Finotti N,* et al. In vitro antioxidant properties of amifostine (WR-2721, Ethyol). Cancer Chemother Pharmacol 2000; 45(2): 172–6.
38. *Gille L, Nohl H.* Analyses of the molecular mechanism of adriamycin-induced cardiotoxicity. Free Rad Biol Med 1997; 23(5): 775–82.
39. *Berthiaume JM, Wallace KB.* Adriamycin-induced oxidative mitochondrial cardiotoxicity. Cell Biol Toxicol 2007; 23(1): 15–25.
40. *Grdina DJ, Murley JS, Kataoka Y.* Radioprotectants: current status and new directions. Oncology 2002; 63(Suppl. 2): 2–10.
41. *Dragojević-Simić V, Dobrić S, Kilibarda V, Bokonjić D, Milovanović S.* Cytoprotector amifostine (WR-2721): pharmacokinetic properties and analytical methods for drug determination. Arch Toxicol Kinet Xenobiot Metab 1996; 4: 185–97.
42. *Fu P, Birukova AA, Xing J, Sammani S, Murley JS, Garcia JG,* et al. Amifostine reduces lung vascular permeability via suppresion of inflammatory signalling. Eur Respir J 2009; 33(3): 612–24.
43. *Murley JS, Kataoka Y, Weydert CJ, Oberley LW, Grdina DJ.* Delayed radioprotection by nuclear transcription factor κB-mediated induction of manganese superoxide dismutase in human microvascular endothelial cells after exposure to the free radical scavenger WR1065. Free Radic Biol Med 2006; 40(6): 1004–16.
44. *Bolman Z, Akalin N, Koseogly MH, Demir S, Kadikoylu G, Atalay H,* et al. Effects of amifostine pre-treatment against adriamycin-induced lipid peroxidation in rat heart. Haema 2003; 6: 61–4.
45. *Jensen RA.* Doxorubicin cardiotoxicity: contractile changes after long term treatment in the rat. J Pharmacol Exp Ther 1986; 236(1): 197–203.
46. *Pelikan PCD, Weisfeld ML, Jacobus WE, Miceli MV, Bulkey BH, Gerstebnlith G.* Acute doxorubicin cardiotoxicity: functional metabolic and morphologic alterations in the isolated, perfused rat heart. J Cardiovasc Pharmacol 1986; 8(5): 1058–66.
47. *Bhutia YD, Vijayaraghavan R, Pathak U.* Analgesic and antiinflamatory activity of amifostine, DRDE-07, and their analogs in mice. Indian J Pharmacol 2010; 42(1): 17–20.
48. *Dragojevic-Simic V, Jacevic V, Dobric S, Djordjevic A, Bokonjic D, Bajcetic M,* et al. Anti-inflammatory activity of fullerenol $C_{60}(OH)_{24}$ nano-particles in a model of acute inflammation in rats. Digest J Nanomater Biostr 2011; 6(2): P819–27.

Received on May 9, 2011.
Accepted on July 12, 2011.
OnLine-first November, 2012.

ORIGINAL ARTICLE

UDC: 577.1:618.3-036/-037
DOI: 10.2298/VSP110530023L

Significance of pregnancy-associated plasma protein A (PAPP-A) concentration determination in the assessment of final outcome of pregnancy

Značaj određivanja koncentracije plazma proteina trudnoće A (PAPP-A) u proceni konačnog ishoda trudnoće

Dragan Lončar, Mirjana Varjačić, Slobodan Arsenijević

Gynecology and Obstetrics Clinic, Clinical Centre Kragujevac, Kragujevac, Serbia

Abstract

Background/Aim. Pregnancy-associated plasma protein A (PAPP-A) is high molecular matrix metalloproteinase originally isolated in the serum of pregnant women. The aim of this study was to analyze the values of concentration of PAPP-A in assessment of progress and outcome of pregnancy in pregnant women diagnosed with threatening preterm delivery, preeclampsia and fetal growth restriction in relation to physiological pregnancy of the same gestational age. **Methods.** The study included 60 pregnant women that were divided into three groups according to gestational age and the diagnosis of imminent premature birth upon reception, preeclampsia and fetal growth restriction as follows: the group I from 28 to 32 weeks of gestation, a total of 25 pregnant women, the group II from 33 to 36 weeks of gestation, a total of 23 pregnant women, and the group III from 37 to 41 weeks of gestation, a total of 12 pregnant women. The control group consisted of 60 pregnant women without complications of pregnancy that were identically divided into three groups according to gestational age as in the sample. We performed quantitative determination of PAPP-A from the venous blood of patients by using commercial tests of the company Diagnostics Product Corporation (DPC), Los Angeles, California, USA. **Results.** There was a statistically significant difference in PAPP-A values in the examined groups in all gestational ages ($p < 0.01$). The value of the PAPP-A concentration in different gestational ages with equal statistical significance indicated the possibility of complications, which was examined during pregnancy in relation to the control group of pregnant women with physiological pregnancies. This study confirmed that there was a statistically significant difference in fetal body weight at birth ($p < 0.05$), Apgar score in 5 min after birth ($p < 0.05$), and gestational age at birth ($p < 0.05$), as parameters of the outcome of pregnancy course, between the examined groups of pregnant women in relation to the value of PAPP-A concentration. The age of pregnant women was not statistically different in the examined groups ($p > 0.05$). **Conclusion.** Differences in PAPP-A concentration should point out to the obstetrician the need for more intensive *antepartum* fetal surveillance in order to increase the chances of favorable perinatal outcome, regardless gestational age.

Key words:
pregnancy outcome; premature birth; pregnancy-associated plasma protein A; pre-eclampsia; fetal growth retardation; apgar score; gestational age.

Apstrakt

Uvod/Cilj. Plazma protein A povezan sa trudnoćom *pregnancy-associated plasma protein A* (PAPP-A) je visokomolekularna matriks metaloproteinaza koja je prvobitno izolovana iz seruma trudnih žena. Cilj istraživanja bio je analiza vrednosti koncentracije PAPP-A u proceni toka i ishoda trudnoće kod trudnica sa dijagnozom pretećeg prevremenog porođaja, preeklampsije i zastoja u rastu ploda u odnosu na fiziološke trudnoće iste gestacijske starosti. **Metode.** U studiju je bilo uključeno 60 trudnica koje su bile podeljene u tri grupe prema gestacijskoj starosti i prijemnoj dijagnozi pretećeg prevremenog porođaja, preeklampsije i zastoja u rastu ploda: grupa I od 28 do 32 nedelje gestacije imala je ukupno 25 trudnica, grupa II od 33 do 36 nedelja gestacije, ukupno 23 trudnice, i grupa III od 37 do 41 nedelje gestacije, ukupno 12 trudnica. Kontrolnu grupu činilo je 60 trudnica bez ispitivanih komplikacija podeljenih prema gestacijskoj starosti identično kao i u eksperimentalnoj grupi. Kvantitativno određivanje PAPP-A vršeno je iz venske krvi bolesnice primenom komercijalnih testova firme Diagnostics Product Corporation (DPC), Los Anđeles, Kalifornija, USA. **Re-**

Correspondence to: Dragan Lončar, Gynecology and Obstetrics Clinic, Clinical Centre Kragujevac, Vojislava Kalanovića 1A/3, 34 000 Kragujevac, Serbia. Phone: +381 64 616 8999, Fax: +381 34 370 151. E-mail: drloncar@sezampro.rs

zultati. Postojala je statistički značajna razlika u vrednostima PAPP-A u ispitivanim grupama u svim gestacijskim starostima ($p < 0,01$). Pokazano je da vrednost koncentracije PAPP-A, u različitim gestacijskim starostima, sa podjednakom statističkom značajnošću ukazuje na mogućnost komplikacija koje su ispitivane u toku trudnoće, u odnosu na kontrolnu grupu trudnica sa fiziološkim trudnoćama. Istraživanje je potvrdilo da je postojala statistički značajna razlika u telesnoj masi ploda na rođenju ($p < 0,05$), Apgar skoru nakon 5 minuta od rođenja ($p < 0,05$) i gestacijskoj starosti na rođenju ($p < 0,05$), kao parametara krajnjeg ishoda toka trudnoće, između ispitivanih grupa trudnica u odnosu na vrednost koncentracije PAPP-A. Starost trudnica nije se statistički razlikovala u ispitivanim grupama ($p > 0,05$). **Zaključak.** Razlike u koncentraciji PAPP-A treba da ukažu akušerima na potrebu intenzivnije kontrole fetusa pre porođaja, kako bi se povećale šanse za povoljan perinatalni ishod, bez obzira na gestacijsku starost.

Ključne reči:
trudnoća, ishod; porođaj, prevremeni; plazma protein-A, udružen sa trudnoćom; preeklampsija; fetus, zaostajanje u rastu; apgar skala; trudnoća, razvoj fetusa.

Introduction

Pregnancy-associated plasma protein A (PAPP-A) is high molecular matrix metalloproteinase originally isolated in serum of pregnant women. PAPP-A is a glycoprotein, macroglobulin, of molecular weight of 800,000 with alpha 2-electrophoretic mobility, and it is produced in syncytiotrophoblast cells of the placenta [1]. Determination of PAPP-A is performed by radioimmunoassay method (immune test with isotope). The first radioimmunoassay determination of PAPP-A was carried out in 1980. Using RIA method it is possible to determine its presence already 3–4 weeks after conception and no later than the 6th week of gestation. The maximum level PAPP-A has at the term delivery. PAPP-A exerts an inhibitory effect on the enzyme elastase, a protease located in the granules of neutrophils granulocytes and participates in processes that lead to the destruction of proteins. By direct immunofluorescence, the presence of PAPP-A in spermatozoid's heads is determined at about 2%. PAPP-A exerts an inhibitory effect on fixation of both complements and coagulation system, as well as on the affinity to heparin. It is assumed that suppressed level of PAPP-A reduces the zinc ion that is required in the fetal organogenesis, which represents one of the factors for the occurrence of congenital malformations. During pregnancy, PAPP-A concentration in maternal blood increases. Decreased concentration is related to increased incidence of chromosomal abnormalities in early gestation and in later pregnancy course because of the associated placental insufficiency. It is characterized by the appearance of fetal growth restriction, preeclampsia, preterm delivery and stillbirth. PAPP-A is a regulator of bioactivity of insulin-like growth factor [2]. Testing of the role of PAPP-A in other tissues of the organism has started recently. Increased values of PAPP-A were found in patients with acute coronary syndrom in contrast to healthy population and those with a diagnosis of stable angina pectoris. It is important to mention that PAPP-A, which is in circulation of patients with coronary disease is circulating in free form, whereas in pregnant women a complex of PAPP-A and the proform of eosinophil major basic protein is present [3–5]. This brings into question the adequacy of the used substrates that were synthesized for the detection of complex form of PAPP-A. PAPP-A represents a useful biomarker in clinical monitoring of pregnancy course. However, new prospective studies are needed by using appropriate substrates for the detection of PAPP-A in order to assess the proper role of metalloproteinase in clinical practice. The aim of this study was to analyze the value of PAPP-A concentration in assessing the final outcome of pregnancy in pregnant women diagnosed with threatening preterm delivery, preeclampsia and the fetal growth restriction in relation to physiological pregnancies of the same gestational age.

Methods

A prospective, observational study was conducted at the Gynecology and Obstetrics Clinic, Clinical Center Kragujevac, Kragujevac, Serbia, in 2010. During examination the clinical–experimental model of study was used. Quantitative measurements of PAPP-A levels were determined from venous blood of patients using the commercial tests of the company Diagnostic Product Corporation (DPC), Los Angeles, California, USA (DPC-USA). The tests, based on an analytical principle of immunochemiluminescence, were implemented using the automated analyzer Immulite 2000. The manufacturer of the analyzer is also DPC-USA.

The study included 60 pregnant women that were divided into three groups according to gestational age and the diagnosis of imminent premature birth upon reception, pre - eclampsia and fetal growth restriction as follows: the group I, from 28 to 32 weeks of gestation, a total of 25 pregnant women; the group II, from 33 to 36 weeks of gestation, a total of 23 pregnant women; the group III, from 37 to 41 weeks of gestation, a total of 12 pregnant women.

The criterion for inclusion of pregnant women in the study included the previously established all three diagnoses that were listed as complications of pregnancy course according to the following standards: preterm delivery before the end of 37th week of pregnancy; the diagnosis of preeclampsia based upon the blood pressure above 140/90 mmHg, proteinuria in 24 hour urine of $\geq$ 0.3 g / per day; intrauterine growth restriction (IUGR) of fetus was diagnosed on the basis of ultrasonographic growth parameters: biparietal diameter (BPD), transverse trunk diameter (TTD), head circumference (HC), abdominal circumference (AC), femur length (FL) and differences in the measured parameters below the 10th percentile than expected for gestational age.

The control group consisted of 60 pregnant women without complications of pregnancy that were identically divided into three groups according to gestational age as in the sample. All the obtained results of research were entered into a single database with valid logic control. Statistical analysis included calculating the average values and standard deviations (SD) for each numerical parameter and analysis of the obtained value in relation to the subgroups (*t*-test, Mann-Whitney) by using the statistical software SPSS 17.

Results

There was a statistically significant difference of PAPP-A values in the examined groups in all gestational ages ($p < 0.01$) (Table 1 and 2 and Figure 1). The mean values and standard deviations of PAPP-A concentration (mU/mL) in a total sample of pregnant women diagnosed with threatening preterm delivery, preeclampsia and intrauterine growth restriction were shown in Table 3.

It is found that pregnant women of 28–32 gestational weeks diagnosed with threatening preterm delivery and preeclampsia, showed significantly lower values PAPP-A than in healthy pregnant women ($p = 0.001$).

Pregnant women of 33–36 gestational weeks diagnosed with threatening preterm delivery and preeclampsia, showed significantly lower values of PAPP-A than in healthy pregnant women ($p = 0.01$) (Table 3 and 4).

Pregnant women at term and the diagnosis of preeclampsia, show significantly lower values of PAPP-A than healthy pregnant women ($p = 0.01$). Healthy pregnant women at 28–32 gestational weeks, showed significantly higher values than pregnant women diagnosed with preeclampsia and intrauterine growth restriction in the same gestational age. The same comment goes for the *t*-test in pregnant women of 33–36 gestational weeks and for a group of pregnant women with normal term pregnancies (Tables 3 and 4).

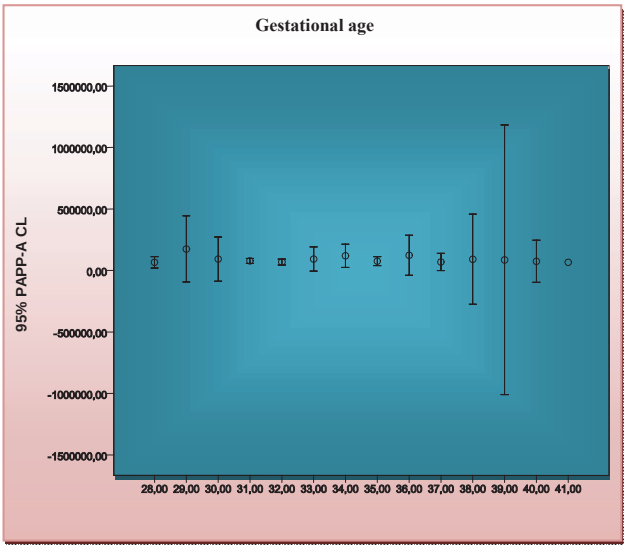


Fig. 1 – Distribution display of pregnancy-associated plasma protein A (PAPP-A) values in the total sample of pregnant women in relation to the weeks of gestation

Table 1

Pregnancy-associated plasma protein A (PAPP-A) in the examined pregnant women according to fetal age

Weeks of gestation (wg)	Number of women	PAPP-A concentration (mU/mL)		
		Min	Max	$\bar{x} \pm SD$
28–32	25	9,353	304,789	65,930 ± 62,095
33–37	23	424	357,207	103,601 ± 83,987*
> 37	12	37,352	276,849	129,827 ± 60,983*

***p* < 0.01 *vs* group 28–32 wg**

Table 2

The mean values and standard deviations of the concentration of pregnancy-asociated plasma protein (PAPP-A) (mU/mL) in the total sample of pregnant women

Weeks of gestation	Number of women	$\bar{x} \pm SD$
28–32	25	91,432 ± 48,121
33–36	23	135,061 ± 65,089
> 37	12	154,287 ± 43,458

Table 3

Obstetrics parameters and age in the group of pregnant women with preterm delivery, preeclampsia and intrauterine growth restriction (n = 60)

Weeks of gestation (number of women)	Fetal body weight (g) $\bar{x} \pm SD$	Apgar score/after 5 min $\bar{x} \pm SD$	Gestational fetal age at birth (ng) $\bar{x} \pm SD$	Age of the pregnant woman (year) $\bar{x} \pm SD$
28–32 (n = 25)	2,640 ± 110	7.2 ± 0.9	36.0 ± 2.2	26.4 ± 3.1
33–36 (n = 23)	2,750 ± 205	8.0 ± 1.8	38.3 ± 1.8	25.0 ± 2.8
> 37 (n = 12)	3,040 ± 180	8.3 ± 1.4	39.2 ± 3.0	28.2 ± 3.3

Table 4

Obstetrics parameters and age in the group of pregnant women with normal pregnancies

Weeks of gestation (number of women)	Fetal body weight (g) $\bar{x} \pm SD$	Apgar score/after 5 min $\bar{x} \pm SD$	Gestational fetal age at birth (ng) $\bar{x} \pm SD$	Age of the pregnant woman (year) $\bar{x} \pm SD$
28–32 (n = 20)	3,640 ± 210	9.2 ± 0.8	39.1 ± 2.6	27.1 ± 4.4
33–36 (n = 20)	3,550 ± 305	9.0 ± 1.0	38.8 ± 2.9	26.2 ± 1.7
> 37 (n = 20)	3,660 ± 290	9.3 ± 0.7	39.4 ± 2.7	27.3 ± 4.5

Discussion

The aim of this study was to determine the relative risk for preeclampsia and intrauterine growth restriction at different PAPP-A levels in different gestational ages. According to the literature data, low levels of PAPP-A during the first trimester are associated with the occurrence of preeclampsia later in the pregnancy [6]. PAPP-A levels in maternal serum between 11th and 13th week of gestation in 224 singleton pregnancies, which later developed preeclampsia, were compared to those of 47,770 normal singleton pregnancies that resulted in live born children after 37 weeks of gestation with body weights at birth greater or equal to the 10th percentile in physiological pregnancy [7]. Correlation between the level of this enzyme and the incidence of preeclampsia was estimated by comparing the relative concentration of PAPP-A at different gestation.

In the preeclampsia group, the median PAPP-A MoM was significantly reduced (0.772 MoM, $p < 0.0001$). With decreasing level of PAPP-A, a probability ratio for preeclampsia was growing. At the 5th percentile of the normal (PAPP-A MoM 0.415), the probability rate was increased 4 times [8, 9]. In our sample there was a statistically significant difference in values of PAPP-A in the examined groups at all gestational ages ($p < 0.01$). We showed that the value of PAPP-A concentration in different gestational ages with equal statistical significance indicates the possibility of complications examined during pregnancy course in relation to the control group of pregnant women with normal pregnancies. A probability factor of preeclampsia on any of PAPP-A MoM levels we consider useful in advising women with low levels of PAPP-A. The use of low PAPP-A in the prediction of preeclampsia and growth restriction for selection of women who will be suggested an intensive surveillance of pregnancy and therefore significantly reduce the incidence and mortality morbidity of mother as well as fetus. PAPP-A is a "protease" for insulin-like growth factor binding proteins 4 and 5. This means that it has the ability to help release insulin-like growth factor from these proteins so that they can freely interact with their cellular receptors. It is considered that insulin-like growth factor plays an important role in trophoblast invasion and hence in the early development and vascularization of the placenta [10, 11]. These early events in the formation of the placenta are extremely important for the outcome of pregnancy, and when abnormal, they are associated with miscarriage, fetal growth restriction, hypertensive disorders induced by pregnancy (preeclampsia), fetal death or preterm delivery. It is assumed that low levels of PAPP-A, leading to reduced release of insulin-like growth factor, could be a path to placentation abnormalities, culminating in the adverse outcomes of pregnancy. Spencer et al [8] in their study on 54,722 normal singleton pregnancies examined the role of PAPP-A in the course of pregnancy. At the 5th percentile of PAPP-A (0.415 MoM), the probability rate for the fetus loss before 24 weeks was increased 3.3 times and above 24 weeks 2.8 times. In other words, there was three times increased risk of fetal loss with low levels of PAPP-A. Cowans and Spencer [11] have recently confirmed a link between low PAPP-A and low fetal weight at birth in relation to the expected for gestational age. In their research they found a linear association of fetal growth restriction and reduced level of PAPP-A, in other words, the lower level of PAPP-A, the lower level of fetal birth weight of any gestational age [12].

Several other studies confirm the association of other "complications of pregnancy" listed above with low levels of PAPP-A [13–15]. For example, as additional results of risk assessment in the first and second trimester (FASTER) study, it was found that women with concentration of PAPP-A below the 5th percentile" were significantly more likely to experience fetal loss before or at the 24th week, low fetal weight at birth, preeclampsia, gestational hypertension, preterm delivery ($p < 0.001$), stillbirth, preterm premature rupture of fetal membranes and placental abruption ($p < 0.02$) [16].

Our research confirmed the allegations of these studies since we found statistically significant difference in body weight of the fetus at birth ($p < 0.05$), Apgar score 5 minutes after birth ($p < 0.05$), and gestational age at the time of delivery ($p < 0.05$), as parameters of the final pregnancy outcome between these groups of pregnant women in relation to the value of the concentration of PAPP-A The age of pregnant women in our study was not statistically different in the examined groups ($p > 0.05$). Despite this association, the positive predictive value of low level of PAPP-A for one of these outcomes is still relatively low.

Conclusion

PAPP-A concentration in the pregnant women of 28–36 gestational weeks had significantly lower values with the diagnosis of preterm delivery and preeclampsia, than in the control group. PAPP-A concentration in the pregnant women diagnosed with preeclampsia in term pregnancy was significantly lower than in he healthy pregnant women at term delivery. PAPP-A concentration is significantly higher in physiological pregnancies of 28–36 gestational weeks compared to the concentrations in pregnant women diagnosed

with preeclampsia and intrauterine growth restriction, of the same gestation age. PAPP-A concentration was significantly higher in physiological pregnancies term gestation in relation to the concentration in pregnant women diagnosed with preeclampsia and intrauterine growth restriction, of the same gestation age.

PAPP-A concentration in the examined groups of our sample had normal distribution due to inhomogeneity of samples and physiological differences in secretion of enzymes in different periods of pregnancy. The pathologic conditions that we examined additionally influenced the irregularity of PAPP-A distribution.

Considering these limiting parameters, the results of PAPP-A levels in serum of pregnant women can only have the screening value, and on the basis of these results, intensive antenatal care should be undertaken.

Acknowledgement

The authors would like to express their gratitude to the Ministry of Science and Technological Development of the Republic of Serbia for the Grant N°175014, out of which the clinical trial that served as the basis for this paper was partially financed.

REFERENCES

1. *Farr M, Strübe J, Geppert HG, Kocourek A, Mahne M, Tschesche H.* Pregnancy-associated plasma protein-E (PAPP-E). Biochim Biophys Acta 2000; 1493(3): 356−62.
2. *Bayes-Genis A, Conover CA, Overgaard MT, Bailey KR, Christiansen M, Holmes DR Jr,* et al. Pregnancy-associated plasma protein A as a marker of acute coronary syndromes. N Engl J Med 2001; 345(14): 1022−9.
3. *Boldt HB, Conover CA.* Pregnancy-associated plasma protein-A (PAPP-A): a local regulator of IGF bioavailability through cleavage of IGFBPs. Growth Horm IGF Res 2007; 17(1): 10−8.
4. *Loechel F, Fox JW, Murphy G, Albrechtsen R, Wewer UM.* ADAM 12-S cleaves IGFBP-3 and IGFBP-5 and is inhibited by TIMP-3. Biochem Biophys Res Commun 2000; 278(3): 511−5.
5. *Yan X, Baxter RC, Firth SM.* Involvement of pregnancy-associated plasma protein-A2 in insulin-like growth factor (IGF) binding protein-5 proteolysis during pregnancy: a potential mechanism for increasing IGF bioavailability. J Clin Endocrinol Metab 2010; 95(3): 1412−20.
6. *Bersinger NA, Ødegård RA.* Second- and third-trimester serum levels of placental proteins in preeclampsia and small-for-gestational age pregnancies. Acta Obstet Gynecol Scand 2004; 83(1): 37−45.
7. *Duckitt K, Harrington D.* Risk factors for pre-eclampsia at antenatal booking: systematic review of controlled studies. BMJ 2005; 330(7491): 565.
8. *Spencer K, Yu CK, Cowans NJ, Otigbah C, Nicolaides KH.* Prediction of pregnancy complications by first-trimester maternal serum PAPP-A and free beta-hCG and with second-trimester uterine artery Doppler. Prenat Diagn 2005; 25(10): 949−53.
9. *Winn VD, Gormley M, Paquet AC, Kjaer-Sorensen K, Kramer A, Rumer KK,* et al. Severe preeclampsia-related changes in gene expression at the maternal-fetal interface include sialic acid-binding immunoglobulin-like lectin-6 and pappalysin-2. Endocrinology 2009; 150(1): 452−62.
10. *Schneider MR, Wolf E, Hoeflich A, Lahm H.* IGF-binding protein-5: flexible player in the IGF system and effector on its own. J Endocrinol 2002; 172(3): 423−40.
11. *Cowans NJ, Spencer K.* First-trimester ADAM12 and PAPP-A as markers for intrauterine fetal growth restriction through their roles in the insulin-like growth factor system. Prenat Diagn 2007; 27(3): 264−71.
12. *Fowden AL.* The insulin-like growth factors and feto-placental growth. Placenta 2003; 24(8−9): 803−12.
13. *Baxter RC.* Insulin-like growth factor (IGF)-binding proteins: interactions with IGFs and intrinsic bioactivities. Am J Physiol Endocrinol Metab 2000; 278(6): E967−76.
14. *Overgaard MT, Boldt HB, Laursen LS, Sottrup-Jensen L, Conover CA, Oxvig C.* Pregnancy-associated plasma protein-A2 (PAPP-A2), a novel insulin-like growth factor-binding protein-5 proteinase. J Biol Chem 2001; 276(24): 21849−53.
15. *Loechel F, Fox JW, Murphy G, Albrechtsen R, Wewer UM.* ADAM 12-S cleaves IGFBP-3 and IGFBP-5 and is inhibited by TIMP-3. Biochem Biophys Res Commun 2000; 278(3): 511−5.
16. *Smith GC, Stenhouse EJ, Crossley JA, Aitken DA, Cameron AD, Connor JM.* Early pregnancy levels of pregnancy-associated plasma protein a and the risk of intrauterine growth restriction, premature birth, preeclampsia, and stillbirth. J Clin Endocrinol Metab 2002; 87(4): 1762−7.

Received on May 30, 2011.
Accepted on September 21, 2011.
OnLine-first July, 2012.

ORIGINAL ARTICLE

UDC: 617.7-007.681:617.735.2
DOI: 10.2298/VSP111229024G

Morphometric characteristics of optic disc in patients with myopia and primary open-angle glaucoma

Morfometrijske karakteristike optičkog diska kod bolesnika sa miopijom i primarnim glaukomom otvorenog ugla

Ranko Gvozdenović*, Dušica Risović*†, Ivan Marjanović*‡, Dragan Vuković*‡, Branislav Stanković*‡

***Faculty of Medicine, University of Belgrade, Belgrade, Serbia, ‡Institute of Ophthalmology, Clinical Center of Serbia, Belgrade, Serbia, †Eye Clinic, University Medical Center Zvezdara, Belgrade, Serbia**

Abstract

Background/Aim. Primary open-angle glaucoma is a multifactorial and progressive neuropathy, characterised by the acquired loss of ganglion cells of the retina and their axons. One of the risk factors for primary open-angle glaucoma is myopia over 5 diopters (D). The aim of our work was to investigate two groups of patients with primary open-angle glaucoma and myopia by using confocal scanning laser ophthalmoscopy, and to find out if the size of refractive error influences optic disk morfometric characteristics. **Methods.** One hundred eyes of one hundred patients with primary open-angle glaucoma and myopia were involved in our study. All the patients were classified into two groups, the first one with myopia < 5 D, and the second one with myopia ≥ 5 D. The Heidelberg retina tomograph is a technique we used in our study. We analized morfometric parameters of patients optic discs, with the aim to find a correlation between the parameters in each group separeatly, and also to find differences between the same parameters from both groups. **Results.** There were significant differences in disc area, cup area, rim area and mean RNFL thickness between the two groups. The size of damage of neuroretinal rim in the group with high myopia was 27%, and in the group with lower myopia 14%. The most frequently damaged segment of neuroretinal rim in the patients with high myopia was nasal segment and in the patients with low myopia infero-temporal one. The least frequently damaged segment of neuroretinal rim in both groups was temporal one. **Conclusion.** Optic discs of glaucomatous patients with high myopia have bigger diameter, also bigger and more irregularly distributed damaged zone of neuroretinal rim, and also thinner retinal nerve fiber layer compared to glaucomatous patients with lower myopia.

Key words:
myopia; glaucoma, open-angle; optic disk; tomography, optical coherence, prognosis.

Apstrakt

Uvod/Cilj. Primarni glaukom otvorenog ugla je multifaktorijalna i progresivna neuropatija koja se karakteriše stečenim gubitkom ganglijskih ćelija retine i njihovih aksona. Jedan od faktora rizika od primarnog glaukoma otvorenog ugla je miopija preko 5 D. Cilj našeg rada bio je da procenimo da li veličina refrakcione greške utiče na morfometrijske karakteristike optičkog diska koristeći konfokalnu skening laser oftalmoskopiju u ispitivanju dve grupe pacijenata sa dijagnostikovanim primarnim glaukomom otvorenog ugla koji istovremeno imaju miopiju. **Metode.** Stotinu očiju od stotinu bolesnika koji imaju dijagnostikovan primarni glaukom otvorenog ugla i istovremeno miopiju bili su uključeni u našu studiju. Bolesnici su bili podeljeni u dve grupe: prva, sa miopijom < 5 D, a druga sa miopijom ≥ 5 D. Heidelberg retina tomografom analizirani su morfometrijski parametri optičkih diskova bolesnika u cilju utvrđivanja postojanja međusobne povezanosti između parametara u svakoj grupi posebno, kao i postojanja statistički značajne razlike među istoimenim parametrima obe grupe. **Rezultati.** Između dve grupe ispitanika utvrđeno je postojanje statistički značajnih razlika u sledećim parametrima: prečniku diska, površine ekskavacije, površini neuroretinlnog oboda i srednje RNFL debljine. Oštećenje neuroretinalnog oboda u grupi bolesnika sa visokom miopijom bilo je 27%, dok je u grupi bolesnika sa niskom miopijom bilo 14%. Najčešće oštećen segment neuroretinalnog oboda bolesnika sa visokom miopijom bio je nazalni, a kod bolesnika sa niskon miopijom donji temporalni. Najređe oštećen segment neuroretinalnog oboda u obe grupe bio je temporalni. **Zaključak.** Optički diskovi glaukomnih bolesnika sa visokom miopijom imaju veći prečnik, veću i iregularnije raspoređenu zonu oštećenja neuroretinalnog oboda, kao i tanji retinalni sloj nervnih vlakana od glaukomnih bolesnika sa niskom miopijom.

Ključne reči:
miopija; glaukom, otvorenog ugla; optički disk; tomografija, optička, koherentna; prognoza.

Correspondence to: Ranko Gvozdenović, Institute of Ophthalmology, Clinical Center of Serbia. Belgrade, Serbia. Phone.: +381 64 211 42 20. E-mail: ranko2010@gmail.com

Introduction

Glaucoma is an eye disease characterized by the increase of intraocular pressure, increase of excavation of the optic disc and paracentral scotomas in visual field. According to the etiopathogenesis, it can be primary, secundary and congenital. Primary open-angle glaucoma is multifactorial and progressive neuropathy, characterised by the acquired loss of ganglion cells of the retina and their axons. Together with the loss of nerve fibers typical changes occur on the optical disc, as well as changes in the visual field [1, 2]. Clinical evaluation of optic disc is an absolutely necessary as the basis for the diagnosis and monitoring of patients with glaucoma [3]. One of the risk factors for primary open-angle glaucoma is myopia over the 5 diopters (D) [4]. Myopia is a defect of the eye that causes light to focus in front of the retina instead of directly on it, resulting in an inability to see distant objects clearly. Benign myopia is a refraction mistake that appears in the puberty period and reaches values of maximally -6 or -7 D, but with the correctional lenses normal visual acquity can be reached. Characteristics of benign myopia is that the retina has no pathological changes. Malignant myopia is a pathological condition of the eye and occurs in early childhood, progressively develops over lifetime and reaches values up to -15 D to -20 D. Malignant myopia is a degenerative eye disease, and changes in the retina are characteristic findings of malignant myopia [4]. The Heidelberg Retina Tomography II (HRT II) represents confocal scanning laser ophtalmoscopy which provides precise topographical mapping of the optic disc and peripapillar retina. It enables obtaining series of global morphometrical parametres, and parametres for each one of the six segments at which the neuroretinal rim is devided, based on three-dimensional reconstruction [5]. Various studies have shown the importance of HRT in the diagnosis and monitoring of patients with glaucoma, and detection of degenerative changes at the neuroretinal rim of eyes of patients with myopia [6, 7].

The aim of our study was to use HRT in two groups of patients diagnosed with primary open-angle glaucoma and myopia, too, to assess whether the size of refractive errors affect the morphometric characteristics of optic disc, and to determine the relationships and connections between the morphometric parameters obtained within the groups themselves and between two groups of patients. The aim of our study was also to determine if the Heidelberg Retinal Tomography instrument can distinguish between morphological characteristics of glaucomatous eyes with low myopia and glaucomatous eyes with high myopia.

Methods

The study included topographic data of 100 eyes of 100 patients from the data base in the cabinet for HRT, at the Ophtalmological Institute of the Faculty of Medicine the Belgrade University. The included patients had the diagnosis of primary open-angle glaucoma and myopia (≥ 1 D or ≤ 12 D). The included patients data on previous operative procedures, as well as eye trauma. The whole group was divided into eyes with a myopic refractive error less than -5 D (n = 50), and eyes with a refractive error equal to or higher than –5 D (n = 50) (Table 1).

Table 1

Basic data on the studied patients

Patients' data	Glaucoma and myopia	
	< 5 D	≥ 5 D
Number (n)	50	50
Male/female (n)	20/30	23/27
Age (years), $\bar{x} \pm SD$	55.56 ± 13.53	50.64 ± 14.46
Refraction error (D), $\bar{x} \pm SD$	2.22 ± 1.05	7.18 ± 2.29

Scanning confocal laser, Heidelberg Retina Tomograph (HRT II, Heidelberg Engineering Inc. Heidelberg, Germany) was used in this study for the collection of any necessary data (Figure 1). HRT II uses confocal scanning laser ophtalmoscopy by which we can get a series of photographies of the cross section of the optical nerve head of different deepness and after 3 D reconstruction it produces topographical photographies of the papilla and peripapillar retina. After it does that, HRT II, by the analysis aplication (Heidelberg Eye Explorer) summs up different structural parameters of the optical nerve [8]. To quantify morphometric rim and cup parameters in optic disc topography, a reference plane is defined. The reference plane is parallel to the retinal surface. It needs to be stable so that the parameters change only when true structural changes in the optic disc occur. Within the disc margin, the retinal surface located above the reference plane is defined as rim and below the reference level as cup

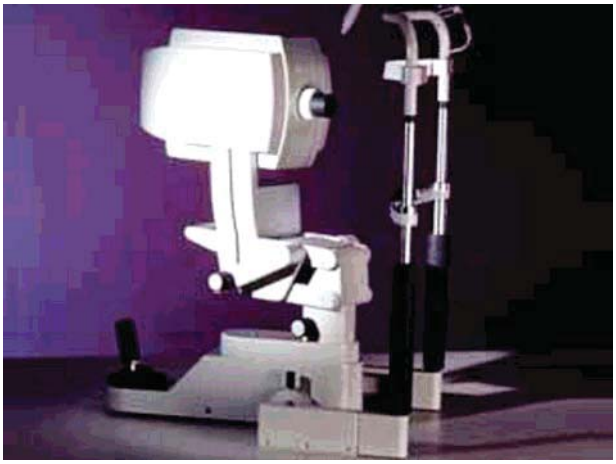


Fig. 1 – Scanning Confocal Laser, Heidelberg Retina Tomograph II (HRT II)

(Figure 2). In order to verify the quality of topographic images we used topographic images with standard deviation

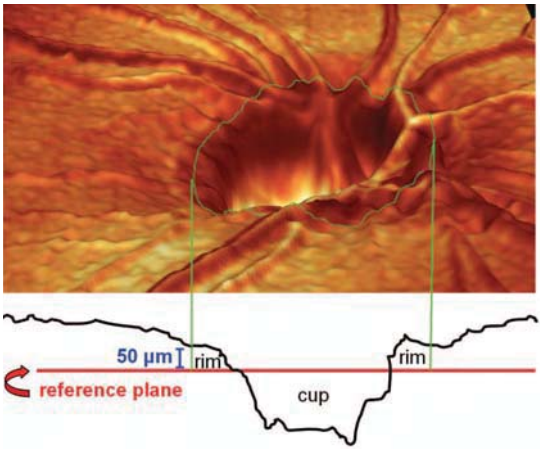


Fig. 2 – A three-dimensional Heidelberg Retina Tomograph (HRT) image of the optic disc.
The margin of the optic disc is defined by the contour line. The cross-sectional image below demonstrates the position of the standard reference plane. The reference plane is needed to distinguish between the cup and the rim.

less than 40 µm. Twelve morphometric parametres were taken into consideration in this study: disc area (mm^2), cup area (mm), rim area (mm^2), cup-to-disc area ratio (C/D ratio), cup volume (mm^3), rim volume (mm^3), mean cup depth (mm), maximum cup depth (mm), height variation contour (mm), cup shape measure (mm), mean retinal nerve fiber layer (mRNFL) thickness (mm) and RNFL cross-sectional area (mm^2).

Moorfields regression analysis (MRA) is a part of HRT programe, and represents method for detecting glaucomatous damage with the HRT. The MRA analyses the regression of the logarithm of the global and six sectoral rim areas (1. temporal, 2. supero-temporal, 3. infero-temporal, 4. nasal, 5. supero-nasal, 6. infero-nasal) to the matching disc areas and compares the results to a normative database. It defines these areas as damaged, borderline and normal based on the 95% and 99.9% confidence intervals (Figure 3). The method accurately discriminates between healthy controls and early glaucoma patients diagnosed using stereoscopic optic disc photography [9]. In our study we had to determine the group with larger damage of the neuroretinal rim (in percent) and which segment of the neuroretinal rim the most frequently and the least frequently often represented as the damaged for each group separately.

Statistical analysis included parameters data incorporated into the software program SPSS version 19.0 for Windows XP. We analysed the examined morphometrical parametres of the optical disc (disc area, cup area, rim area, cup volume, rim volume, cup/disc area ratio, mean cup depth, maximum cup depth, height variation contour, cup shape measure, mean RNFL thickness, RNFL cross sectional area) of the patients from both groups, with the aim to establish the existance of mutual correlation between the mean RNFL thickness and other parametres in each of the groups separately, and the existence of statistically significant difference between the same parametres between the groups (statistically significant difference was when $p < 0.05$). First, we calculated basic statistical parametres (minimal value – MIN, maximal value – MAX, middle – $\overline{X}$, standard deviation – SD, coefficient of the variation – CV). We used the method of linear correlation (statistically significant correlation was when $p < 0.05$) by which we examined the correlation between the examined morphometrical parametres, then parametric and non-parametric tests for the evaluation of the significance of the difference (*t*-test and Mann–Whitney U-test).

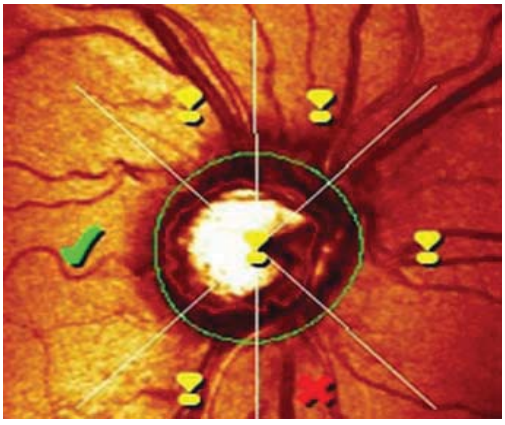


Fig. 3 – The figure shows six segments of neuroretinal rim with labels of Morfields classification
(green sign –normal, yellow sign – borderline, red – damaged)

Results

We calculated average values, standard deviations, minimal and maximal values of all the examined parametres for both groups of the patients (Table 2).

In the group with low myopia (< 5D), we established the existence of statistically significant correlation between mRNFL and disc area, cup area, rim area, cup volume, rim volume, cup/disc area ratio, height variation contour, cup shape measure, RNFL cross sectional area (Table 3).

In the group with high myopia (≥ 5 D) we established the existence of some statistically significant correlation between mRNFL and cup area, rim area, RNFL cross-sectional area, cup/disc area ratio, maximum cup depth, rim volume (Table 3).

According to the distribution of parameters values by the use of statistical tests for the evaluation of significance difference (*t*-test or Mann–Whitney U-test) among the same examined parameters from both groups, we established the existence of statistically significant differences between: disc area, cup area, rim area and mean RNFL thickness (Table 4).

By reading Moorfields analysis of HRT findings of both groups, we found that: the damage size of neuroretinal rim was higher in the group with high miopia (27%) than in the group of the patients with low miopia (14%). We also found that in the group with low miopia (< 5 D) the segment most often classified as damaged was infero-temporal, and the least often temporal one, untill in the group with high miopia (≥ 5 D) the segment most often classified as damaged was nasal segment, and the least often temporal one (Table 5).

Table 2

The basic statistical data

Parameters	Myopia [$\bar{x}$ ± SD (min, max)]	
	≥ 5	< 5
Disc area (mm ^{2})	3.278 ± 1.151 (1.370 – 7.662)	2.550 ± 0.545 (1.333 – 4.562)
Cup area (mm ^{2})	1.402 ± 0.943 (0.025 – 4.067)	1.003 ± 0.733 (0.084 – 3.560)
Rim area (mm ^{2})	1.876 ± 0.863 (0.294 – 5.297)	1.545 ± 0.492 (0.272 – 2.542)
Cup volume (mm ^{3})	0.371 ± 0.373 (0.002 – 1.653)	0.307± 0.363 (0.004 – 1.724)
Rim volume (mm ^{3})	0.452 ± 0.346 (0.027 – 1.439)	0.357 ± 0.194 (0.026 – 0.940)
Cup/disc area ratio	0.403 ± 0.230 (0.013 – 0.905)	0.371 ± 0.205 (0.032 – 0.900)
Mean cup depth (mm)	0.237 ± 0.137 (0.025 – 0.688)	0.263 ± 0.122 (0.065 – 0.723)
Maximum cup depth (mm)	0.621 ± 0.314 (0.071 – 1.583)	0.679 ± 0.220 (0.175 – 1.166)
Height variation contour (mm)	0.407 ± 0.178 (0.085 – 0.914)	0.369 ± 0.125 (0.159 – 0.629)
Cup shape measure (mm)	-0.145 ± 0.076 (-0.302 – -7.278)	-0.155 ± 0.102 (-0.376 – 0.167)
Mean RNFL thickness (mm)	0.110 ± 0.191 (-0.810 – 0.393)	0.189 ± 0.097 (-0.050 – 0.376)
RNFL cross sectional area (mm ^{2})	0.784 ± 0.904 (-1.045 – 2.8199)	1.047 ± 0.538 (-0.292 – 2.158)

RNFL – retinal nerve fiber layer

Table 3

Correlation between the mean retinal nerve fiber layer (RNFL) thickness and other parameters in both groups of patients

Parameters	Mean RNFL thickness	
	Myopia < 5 D	Myopia ≥ 5 D
Disc area	-0.310 *	-0.113
Cup area	-0.582 **	-0.426 *
Rim area	0.524 **	0.315 **
Cup volume	-0.404 **	-0.205
Rim volume	0.797 **	0.251 **
Cup/disc area ratio	-0.645 **	-0.471 **
Mean cup depth	-0.126	0.210
Maximum cup depth	0.155	0.176 **
Height variation contour	0.443 **	0.053
Cup shape measure	-0.574 **	0.119
RNFL cross sectional area	0.973 **	0.834 **

*** $p < 0,05$; ** $p < 0,01$**

Table 4

The difference between the same parameters in both groups of patients

Parameters	Z test
Disc area	-3,673 **
Cup area	-2,215 *
Rim area	-2,249 *
Cup volume	-1,284
Rim volume	-1,405
Cup/disc area ratio	0,739
Mean cup depth	-0,961
Maximum cup depth	-1,12
Height variation contour	-1,052
Cup shape measure	-0,676
Mean RNFL thickness	-2,761 **
RNFL cross sectional area	-1,94

*** $p < 0,05$; ** $p < 0,01$**

Table 5

Distribution of damaged segments of neuroretinal rim in both groups of patients

Myopia	Number of damaged segments (n)					
	Temporal	Supero-temporal	Infero-temporal	Nasal	Supero-nasal	Infero-nasal
< 5 D (n = 50 eyes)	3	8	10	8	6	8
≥ 5 D (n = 50 eyes)	5	7	8	20	13	13

Discussion

Our study shows statistical analysis of HRT topographical parameters of the optic disc in patients with primary open-angle glaucoma and myopia, too. Myopia is one of the most common ocular abnormalities reported worldwide, and its association with glaucoma is well-recognized. The prevalence of myopia is high in patients with ocular hypertension, primary open-angle glaucoma, and normal-tension glaucoma [10–13]. The risk of developing glaucoma is two to

three times higher in myopic individuals than in nonmyopic individuals, and this risk factor is independent of other risk factors for glaucoma [13]. Currently, glaucoma is diagnosed by considering the appearance of the optic disc and retinal nerve fiber layer and by standard achromatic perimetry [14]. However, myopic individuals often have enlarged optic discs with a more oval configuration and larger areas of peripapillary atrophy [15, 16]. Because of these features, glaucomatous changes cannot be easily interpreted in myopic discs, possibly leading to a misdiagnosis of glaucoma. In early glaucoma, structural change is known to precede functional damage [17, 18]. The RNFL is a sensitive indicator for predicting early glaucomatous changes [19–20], and the extent of RNFL damage correlates with the severity of functional deficit in the visual field [21, 22]. Thus, RNFL assessment may be more valuable than optic disc assessment in the case of myopic subjects. We compared the obtained results by the use of tests for the examination of the correlation and establishing differences between the examined parameters with the findings of other authors. The relationship between RNFL thickness and myopia has been extensively investigated [23–30]. However, whether RNFL thickness could vary with the refractive status of the eye remains unclear. It is therefore important to investigate whether there is any correlation between RNFL measurements and the axial length/refractive error in myopic patients, considering that the risk of developing glaucoma increases with the severity of myopia. Considering different approaches of various studies to the connection between myopia and glaucoma we examined the morphological characteristics of optical discs of patients diagnosed with primary open-angle glaucoma and high and low refractive errors in order that the results obtained in our study help ophthalmologists in routine examinations of the HRT findings, to help them to avoid errors in diagnosis of glaucomatous or myopic damage to the optic disk and its surroundings. We established highly statistically significant correlations between different parameters of both groups. Similar results were reported in studies of Adegbehingbe and Ouertani [31], and in the studie of Eid et al. [32]. Between the same examined parametres from both groups we established statistically significant differences in the following parametres: disc area, cup area, rim area i mean RNFL thickness. Similar results can be found in the study of Dichtl et al. [33].

The damage size of neuroretinal rim is higher in the group with high myopia (≥ 5 D) than in the group with low miopia, similar as the findings of Dichtl et al. [33]. In the group with low miopia (< 5 D) the segment most often classified as damaged was infero-temporal, and the least often temporal one, the finding reported also by Jonas et al. [34] while in the group with high miopia (≥ 5 D) the segment most often classified as damaged was nasal segment and the least often, temporal one. Limitation of the study was conditioned not just by its retrospective nature, but also by the lack of other clinical information on the patients included in the study, for the sake of the comparison with the findings of HRT, which is significant in the glaucoma diagnostic.

Conclusion

Glaucomatous eyes with high myopia (≥ 5 D) have larger diameters of optic disc, also larger cup and thinner layer of retinal nerve fibers, compared with the glaucomatous patients with low miopia (< 5 D), while the cup to disc area ratio has no significant difference between these two groups of patients. The increase of the cup and cup to disc area ratio leads to the reduction in average retinal nerve fiber layer thickness was a common morphometric characteristic of optic discs in both groups of patients. Probability of neuroretinal rim damage was 93% higher within the glaucomatous eyes with high, than within the glaucomatous eyes with low myopia. The most ofted damaged segment of the neuroretinal rim in patients with high myopia was nasal segment while in those with lower myopia was infero-temporal one. The rarest damaged segment of the neuroretinal rim in both groups of patients was temporal segment. The findings of Heidelberg Retina Tomography II make it possible to distinguish morphological characteristics of optic discs in both groups of patients.

REFERENCES

1. *Quigley HA.* Neuronal death in glaucoma. Prog Retin Eye Res 1999; 18(1): 39−57.
2. *Airaksinen PJ, Drance SM, Schulzer M.* Neuroretinal rim area in early glaucoma. Am J Ophthalmol 1985; 99(1): 1−4.
3. *Tuulonen A, Airaksinen PJ.* Initial glaucomatous optic disk and retinal nerve fiber layer abnormalities and their progression. Am J Ophthalmol 1991; 111(4): 485−90.
4. *Golubović S.* Ophthalmology. Belgrade: Medical Faculty; 2010. (Serbian)
5. *Wollstein G, Garway-Heath DF, Hitchings RA.* Identification of early glaucoma cases with the scanning laser ophthalmoscope. Ophthalmology 1998; 105(8): 1557−63.
6. *Durukan AH, Yucel I, Akar Y, Bayraktar MZ.* Assessment of optic nerve head topographic parameters with a confocal scanning laser ophthalmoscope. Clin Experiment Ophthalmol 2004; 32(3): 259−64.
7. *Uchida H, Yamamoto T, Araie M, Tomita G, Shirakashi M, Yoshikawa K.* Topographic characteristics of the optic nerve head measured with scanning laser tomography in normal Japanese subjects. Jpn J Ophthalmol 2005; 49(6): 469−76.
8. *Cioffi GA, Robin AL, Eastman RD, Perell HF, Sarfarazi FA, Kelman SE.* Confocal laser scanning ophthalmoscope. Reproducibility of optic nerve head topographic measurements with the confocal laser scanning ophthalmoscope. Ophthalmology 1993; 100(1): 57−62.
9. *Vernon SA, Hawker MJ, Ainsworth G, Hillman JG, Macnab HK, Dua HS.* Laser scanning tomography of the optic nerve head in a normal elderly population: the Bridlington eye assessment project. Invest Ophthalmol Vis Sci 2005; 46(8): 2823−8.
10. *Fong DS, Epstein DL, Allingham RR.* Glaucoma and myopia: are they related? Int Ophthalmol Clin 1990; 30(3): 215−8.
11. *Perkins ES, Phelps CD.* Open angle glaucoma, ocular hypertension, low-tension glaucoma, and refraction. Arch Ophthalmol 1982; 100(9): 1464−7.
12. *Seddon JM, Schwartz B, Flowerdew G.* Case-control study of ocular hypertension. Arch Ophthalmol 1983; 101(6): 891−4.

13. *Mitchell P, Hourihan F, Sandbach J, Wang J*J. The relationship between glaucoma and myopia: the Blue Mountains Eye Study. Ophthalmology 1999; 106(10): 2010−5.
14. *American Academy of Ophthalmology*. Glaucoma Panel. Primary open-angle glaucoma. Preferred practice pattern. San Francisco: American Academy of Ophthalmology; 2000. p. 1–36.
15. *Tay E, Seah SK, Chan SP, Lim AT, Chew SJ, Foster PJ,* et al. Optic disk ovality as an index of tilt and its relationship to myopia and perimetry. Am J Ophthalmol 2005; 139(2): 247−52.
16. *Jonas JB, Gusek GC, Naumann GO*. Optic disk morphometry in high myopia. Graefes Arch Clin Exp Ophthalmol 1988; 226(6): 587−90.
17. *Quigley HA, Katz J, Derick RJ, Gilbert D, Sommer A*. An evaluation of optic disc and nerve fiber layer examinations in monitoring progression of early glaucoma damage. Ophthalmology 1992; 99(1): 19−28.
18. *Sommer A, Katz J, Quigley HA, Miller NR, Robin AL, Richter RC, Witt KA*. Clinically detectable nerve fiber atrophy precedes the onset of glaucomatous field loss. Arch Ophthalmol 1991; 109(1): 77−83.
19. *Quigley HA, Dunkelberger GR, Green WR*. Chronic human glaucoma causing selectively greater loss of large optic nerve fibers. Ophthalmology 1988; 95(3): 357−63.
20. *Quigley HA, Dunkelberger GR, Green WR*. Retinal ganglion cell atrophy correlated with automated perimetry in human eyes with glaucoma. Am J Ophthalmol 1989; 107(5): 453−64.
21. *Leung CK, Chan WM, Yung WH, Ng AC, Woo J, Tsang MK,* et al. Comparison of macular and peripapillary measurements for the detection of glaucoma: an optical coherence tomography study. Ophthalmology 2005; 112(3): 391−400.
22. *Hoffmann EM, Medeiros FA, Sample PA, Boden C, Bowd C, Bourne RR*, et al. Relationship between patterns of visual field loss and retinal nerve fiber layer thickness measurements. Am J Ophthalmol 2006; 141(3): 463−71.
23. *Medeiros FA, Zangwill LM, Bowd C, Weinreb RN*. Comparison of the GDx VCC scanning laser polarimeter, HRT II confocal scanning laser ophthalmoscope, and stratus OCT optical coherence tomograph for the detection of glaucoma. Arch Ophthalmol 2004; 122(6): 827−37.
24. *Budenz DL, Anderson DR, Varma R, Schuman J, Cantor L, Savell J,* et al. Determinants of normal retinal nerve fiber layer thickness measured by Stratus OCT. Ophthalmology 2007; 114(6): 1046−52.
25. *Leung CK, Mohamed S, Leung KS, Cheung CY, Chan SL, Cheng DK,* et al. Retinal nerve fiber layer measurements in myopia: An optical coherence tomography study. Invest Ophthalmol Vis Sci 2006; 47(12): 5171−6.
26. *Hoh ST, Lim MC, Seah SK, Lim AT, Chew SJ, Foster PJ,* et al. Peripapillary retinal nerve fiber layer thickness variations with myopia. Ophthalmology 2006; 113(5): 773−7.
27. *Hougaard JL, Ostenfeld C, Heijl A, Bengtsson B*. Modelling the normal retinal nerve fibre layer thickness as measured by Stratus optical coherence tomography. Graefes Arch Clin Exp Ophthalmol 2006; 244(12): 1607−14.
28. *Choi SW, Lee SJ*. Thickness changes in the fovea and peripapillary retinal nerve fiber layer depend on the degree of myopia. Korean J Ophthalmol 2006; 20(4): 215−9.
29. *Vernon SA, Rotchford AP, Negi A, Ryatt S, Tattersal C*. Peripapillary retinal nerve fibre layer thickness in highly myopic Caucasians as measured by Stratus optical coherence tomography. Br J Ophthalmol 2008; 92(8): 1076−80.
30. *Schweitzer KD, Ehmann D, García* R. Nerve fibre layer changes in highly myopic eyes by optical coherence tomography. Can J Ophthalmol 2009; 44(3): e13−6.
31. *Adegbehingbe BO, Ouertani AM*. Confocal scanning laser tomography of the optic nerve head of glaucoma patients: intercorrelation of disc parameters. Ghana Med J 2009; 43(4): 150−6.
32. *Eid TE, Spaeth GL, Moster MR, Augsburger JJ*. Quantitative differences between the optic nerve head and peripapillary retina in low-tension and high-tension primary open-angle glaucoma. Am J Ophthalmol 1997; 124(6): 805−13.
33. *Dichtl A, Jonas JB, Mardin CY*. Comparison between tomographic scanning evaluation and photographic measurement of the neuroretinal rim. Am J Ophthalmol 1996; 121(5): 494−501.
34. *Jonas JB, Fernández MC, Stürmer J*. Pattern of glaucomatous neuroretinal rim loss. Ophthalmology 1993; 100(1): 63−8.

Received on December 29, 2011.
Accepted on January 31, 2012
OnLine-first July, 2012.

ORIGINAL ARTICLE

UDC: 577.112.38::616.15]::616.36-004
DOI: 10.2298/VSP110823032C

Plasma homocysteine levels in patients with liver cirrhosis

Nivo homocisteina u plazmi bolesnika sa cirozom jetre

Djordje M. Ćulafić*, Miroslav Lj. Marković†, Radmila Ž. Obrenović‡, Dragana D. Mijač*

***Clinic for Gastroenterology and Hepatology, ‡Institute of Medical Biochemistry, Clinical Center of Serbia, Faculty of Medicine, University of Belgrade, Belgrade, Serbia; †Clinic of Surgery, Military Medical Academy, Belgrade, Serbia**

Abstract

Background/Aim. Homocysteine (2-amino-4-mercaptobutyric acid) is an amino acid that may be found in small quantities in all cells, and is quantitatively the major methionine metabolite. The most prevalent form is protein-bound homocysteine (about 80%), mostly to albumins. If catabolism of homocysteine is impaired either due to enzyme defect or deficiency of required intracellular cofactors, homocysteine accumulates in cells and reaches the circulation. The aim of our study was to determine homocysteine values and factors affecting homocysteine metabolism in patients with liver cirrhosis. **Methods.** The prospective study included 35 patients with liver cirrhosis and 30 age and sex matched healthy controls. All the examinations were based on: medical history, physical examination, laboratory tests including serum homocysteine levels and liver biopsy. The degree of liver failure was assessed according to the Child-Pugh classification. **Results.** The mean plasma homocysteine levels were much higher in the patients with liver cirrhosis than in the healthy controls (*t*-test, $p < 0.001$). There was no significant difference between the plasma homocysteine concentration and etiology of liver cirrhosis (ANOVA, $p > 0.05$). Correlation analysis showed a positive correlation between the homocysteine and creatinine concentrations and between the serum albumin and homocysteine values, (Pearson's correlation, $p < 0.01$, and $p < 0.05$ respectively). **Conclusion.** In liver cirrhosis, the genesis of homocysteinemia is multifactorial, influenced significantly by impaired catabolic liver function, renal failure and hypoalbuminemia.

Key words:
hemocysteine; liver cirrhosis; hypoalbuminemia; creatinine.

Apstrakt

Uvod/Cilj. Homocistein (2-amino-4-merkapto-buterna kiselina) je aminokiselina koja se nalazi u malim količinama u svim ćelijama, i predstavlja glavni metabolit metionina. Homocistein se u serumu nalazi najčešće vezan za albumin (oko 80%). U stanjima u kojima dolazi do poremećaja katabolizma homocisteina, zbog enzimskog defekta ili nedostatka intracelularnih kofaktora neophodnih za njegov metabolizam, homocistein se skladišti u ćelijama i dospeva u cirkulaciju u slobodnom obliku. Cilj ove studije bio je utvrđivanje nivoa homocisteina kod bolesnika sa cirozom jetre i faktora koji mogu da utiču na njegov metabolizam. **Metode.** Ovom prospektivnom studijom bilo je obuhvaćeno 35 bolesnika sa cirozom jetre i 30 zdravih osoba koje su činile kontrolnu grupu. Grupe zdravih ispitanika i bolesnika bile su podudarne prema polu i uzrastu. Kod svih ispitanika ispitivanje je obuhvatilo anamnestičke podatke, fizikalni pregled, laboratorijske analize uključujući i nivo homocisteina u serumu, kao i biopsiju jetre u grupi bolesnika sa cirozom jetre. Stepen oštećenja jetre procenjivan je primenom Child-Plugh klasifikacije. **Rezultati.** Bolesnici sa cirozom jetre imali su u proseku više vrednosti homocisteina nego zdravi ispitanici (*t*-test, $p < 0,001$). Etiološki faktori za nastanak ciroze jetre (alkohol, virusi, autoimunski, kriptogena ciroza) nisu uticali na nivo homocisteina u serumu (ANOVA, $p > 0,05$). Takođe, utvrđeno je postojanje pozitivne korelacije između nivoa homocisteina i kreatinina, odnoso albumina u krvi kod bolesnika sa cirozom jetre (Pearson-ov koeficijent korelacije, $p < 0,01$ i $p < 0,05$ respektivno). **Zaključak.** Bolesnici sa cirozom jetre imaju više vrednosti homocisteina u krvi, što je posledica brojnih faktora kao što su oštećena funkcija jetre, bubrežna insuficijencija i hipoalbuminemija.

Ključne reči:
homocistein; jetra, ciroza; hipoalbuminemija; kreatinin.

Correspondence to: Dragana Mijac, Dr Koste Todorović 2, 11 000 Belgrade, Serbia. Phone: +381 11 366 3346.
E-mail: dmijac@open.telekom.rs

Introduction

Homocysteine (2-amino-4-mecarpto-butinic acid) is an amino acid that may be found in small quantities in all cells, and it is quantitatively the major methionine metabolite. Moreover, homocysteine can be found either free in a body or in the form of disulfide and proteins. In relation to a total homocysteine quantity, free or reduced one accounts for only 1%–2%. However, the most prevalent form is protein-bound homocysteine (about 80%), mostly to albumins [1].

It is well-known, that a high level of blood serum homocysteine is a powerful risk factor for cardiovascular disease [2]. On the other hand, elevated levels of homocysteine have been linked to increased fractures in elderly persons [3].

The liver has an important role in metabolism of homocystine. It is condensed with serine and upon separation of molecule of water and cystathionine-β-synthetase (vitamin B6 dependent enzyme) it yields cystathionine. Cystathionine, breaks down to homoserine and cysteine, by the action of cystathionase (vitamin B6 dependent enzyme). Homoserine is transformed into α-ketobutyric acid under the action of homoserine desaminase, when ammonium hydroxide and hydrogen sulfide are being separated from it. Homocysteine may in one part, oxidize to homocystine. Methionine may be resynthesized from homocysteine and methyl-tetrahydrofolic acid [4].

If catabolism of homocysteine is impaired either due to enzyme defect or deficiency of required intracellular cofactors, homocysteine accumulates in cells and reaches the circulation.

The aim of our study was to determine homocysteine values and factors affecting the homocysteine metabolism in patients with liver cirrhosis.

Methods

In the period from August 2008 to April 2009, the prospective study included 35 patients with liver cirrhosis and 30 age and sex matched healthy controls examined at the Clinic of Gastroenterology and Hepatology, Clinical Center of Serbia, Belgrade. Written informed consent was obtained from each subject.

Inclusion criterion was the patients' diagnosis of liver cirrhosis as an underlying disease. All the examinations were based on: medical history, physical examination, laboratory tests and liver biopsy. Laboratory tests included: liver functional tests as well as specific (etiological) tests. Puncture liver biopsy was performed in 7 (20%) patients, using the 1.4 mm Menghini needle. The degree of liver failure was assessed according to Child-Pugh classification.

A day after an overnight fast, fasting blood samples were drawn to determine biochemical indices and homocysteine determination were collected in additive-free Vacutainers (BD), centrifuged at 3,500 rpm (≈ 2,000 g) 10 minutes, using the "Heraeus Digifuga GL" centrifuge. After separation, serum was stored at -20°C until measurement. Blood was drawn between 8 and 9 a.m. Homocysteine was measured by commercial test kits by means of Immulite, (Siemens, USA) and expressed in μmol/L.

The results were expressed as mean ± SD or as stated. Distribution data were compared by χ^2-test. The differences between the groups were analysed using a paired Student's *t*-test and correlation coefficients (Spearman [1], correlation and Person's correlation test). All statistical analyses were performed with the SPSS 10.0 for Windows package (SPSS Inc., Chicago, IL). The values at the $p = 0.05$ level were considered statisticaly significant.

Results

The most common cause of liver cirrhosis was alcohol – 21 (60%) patients. The incidence of posthepatitis liver cirrhosis was lower – 7 (20%) of the patients, autoimmune liver diseases were quite rare – 4 (11.4%) of the patients, while etiology was unknown in 3 (8.6 %) of the cases.

The patients were classified into 3 classes according to generally accepted Child's system. Child-Pugh class A included 19 (54.2%), class B 8 (22.9%), and class C 8 (22.9%) of the patients.

The mean plasma homocysteine levels were much higher in the patients with the cirrhosis than in healthy controls. A statistically significant difference was found between homocysteine plasma values in patients with cirrhosis and healthy subjects (14.85 ± 5.40 versus 9.17 ± 1.99 μmol/L, *t*-test, $p < 0.001$).

On the other hand, there was no significant difference between the plasma homocysteine concentration and etiology of the cirrhosis (ANOVA, $p > 0.05$).

In relation to creatinine concentration, the patients were divided into two subgroups, *i.e.* with normal (< 120 μmol/L) and higher creatinine levels (> 120 μmol/L). The normal creatinine values were reported in 28 (80%) of the patients and higher levels were recorded in 7 (20%) of the cases.

Correlation analysis showed a positive correlation between homocysteine and creatinine concentrations (Pearson's test, r = 0.4622; $t = 2.994$, $p < 0.01$). In addition, a positive correlation between the serum albumin and homocysteine values was also established (Pearson's correlation, $p < 0.05$).

Discussion

Patients with cirrhosis develop a hyperdynamic state of circulation, with high cardiac output, increased blood volume, reduced systemic vascular resistance, and they are prone to arterial hypotension. Increased hepatic and collateral resistances as well as portal blood flow maintain portal hypertension [5, 6].

It is considered that bacterial lipopolysaccharide endotoxins cause multiple-hour release of nitric oxide (NO) from vascular endothelium, what leads to peripheral vasodilatation, hypotension and tachycardia. *In vitro* effect of endotoxin and cytokine on NO synthesis induction has been proved in endothelium and smooth muscles with progressive vascular relaxation and poorer response to vasoconstrictors [7].

High circulating endotoxin concentrations were found in cirrhosis, which may persist even without evident clinical

signs of infection. The endotoxins in cirrhosis are supposed to induce, directly or indirectly, the increase of NO release and synthesis, which causes the methionine-synthase inactivation, giving rise to accumulation of homocysteine in cells and extracellular space [8].

Homocysteine and related biogenic thiols produce chemically and physiologically specific products in reactions with nitric oxides: nitrogen dioxide, dinitrogen trioxide and dinitrogen tetroxide. A tendency towards interaction with metal nitrosyl complexes is also manifested. In both cases, reaction products are S-nitrosothiol or thionitrites. These substances strongly activate the enzyme guanylate cyclase and are an important intermediary agent in metabolism of the endothelium-relaxing factor (EDRF) [9].

Elevated homocysteine levels in cells and extracellular space, by inducing the synthesis of vasoactive EDRF, are involved in the pathogenesis of hyperdynamic circulation.

The study by Woitas et al. [10] found significantly higher homocysteine concentrations in cirrhotic patients in relation to the controls ($p = 0.0002$), and a non-significant correlation between homocysteine concentration and degree of liver insufficiency according to the Child-Pugh classification ($p = 0.1$).

Ferre et al. [11] analyzed 76 patients with cirrhosis. Alcoholic cirrhosis was diagnosed in 48 (63%) of the patients and non-alcoholic in 28 (37%) of the cases. They verified higher homocysteine concentrations in cirrhotic patients in relation to the healthy controls which depended upon the extent of liver impairment. No difference in homocysteine concentrations between alcoholic and nonalcoholic cirrhosis was found.

Our studies are compatible with those of Woitas et al. [10] and Ferre et al. [11]. In our study as in the study by Woitas et al. [10] there was no correlation between homocysteine concentrations and liver insufficiency according to the Child-Pugh classification ($p > 0.05$). It differs from the study by Ferre et al. [11] which noted that higher homocysteine concentrations correlated with more severe degree of liver insufficiency.

The kidneys are considered to have a major role in homocysteine metabolism and are involved in about 70%-elimination of homocysteine from plasma, by glomerular filtration and metabolism in tubular cells, breakdown through transsulfuration or remethylation into methionine. Some articles report that homocystinemia directly correlates with serum creatinine and glomerular filtration [12].

A significance of tubular metabolism of homocysteine has been corroborated by clinical studies. Renal posttransplantation levels of homocysteine are much higher in comparison to patients with end-stage renal failure who had not undergone transplantation. During transplantation, a kidney is subjected to ischemic injury of tubular cells, which may be additionally damaged by immunological reactions and immunosuppressive drugs. Lower homocysteine metabolism in tubules causes higher homocysteine values [13].

Our study found a positive correlation between homocysteine and creatinine levels ($p < 0.01$). Given that in our study only 7 (20%) of the patients had impaired renal function, evaluated on the basis of higher creatinine values, homocystinemia in cirrhosis could not be accounted for disordered glomerular filtration and tubular metabolism only.

Suliman et al. [14] indicated that blood homocysteine level was lower in patients with end-stage renal failure with cardiovascular diseases and they associated such paradox with hypoalbuminemia.

It is well-known that plasma homocysteine may be found in several forms. The majority, about 70%, is bound to plasma proteins, *ie*. to albumin, mostly *via* cysteine, while the remaining free homocysteine is, due to high reactivity of the thiol group, susceptible to autooxidation and formation of disulfide bonds; the rest is composed of free, reduced form of homocysteine (only 1% of total plasma homocysteine) [14, 15]. Our study found a positive correlation between albumin and homocysteine, what is in compliance with the past results.

On the other hand, it is well known that vitamin B6 deficiency is usually the result of malabsorption syndrome, uremia, cancer, cirrhosis, alcoholism, old age, and pregnancy [16]. Moreover, it is showed that plasma levels of pyridoxal-5'-phosphate (PLP) in cirrhotic patients were significantly lower than in healthy control subjects [17].

Nutrition regime significantly interferes with the level of homocysteine that may vary in relation to methionine intake by food. Animal food is richer in methionine than plant one. Meat and fish contain 2.7 g/100 g, eggs 3.2g/100 g, cow milk 2.9 g/100 mL of methionine versus fruit and vegetables containing only 0.9–1.2 g/100g [15, 18]. Although the nutritive status was not the subject of analysis in our study, it is well-known that cirrhotic patients have poor appetite and reduced intake of proteins in relation to healthy subjects [19].

Conclusion

In liver cirrhosis, the genesis of homocystinemia is multifactorial, influenced significantly by impaired catabolic liver function, renal failure and hypoalbuminemia.

REFERENCES

1. *Refsum H, Fiskerstrand T, Guttormsen AB, Ueland PM.* Assessment of homocysteine status. J Inherit Metab Dis 1997; 20(2): 286−94.
2. *Martí-Carvajal AJ, Solà I, Lathyris D, Salanti G.* Homocysteine lowering interventions for preventing cardiovascular events. Cochrane Database Syst Rev 2009; (4): CD006612.
3. *McLean RR, Jacques PF, Selhub J, Tucker KL, Samelson EJ, Broe KE,* et al. Homocysteine as a predictive factor for hip fracture in older persons. N Engl J Med 2004; 350(20): 2042−9.
4. *Mirković D, Majkić-Singh N, Ignjatović S.* Homocystine: chemistry, metabolism and role in pathophysiological processes. Jugoslov Med Biohem 2003; 22(2): 127−40.

5. *Bosch J, Pizcueta P, Feu F, Fernández M, García-Pagán JC.* Pathophysiology of portal hypertension. Gastroenterol Clin North Am 1992; 21(1): 1−14.
6. *La Villa G, Gentilini P.* Hemodynamic alterations in liver cirrhosis. Mol Aspects Med 2008; 29(1−2): 112−8.
7. *Clària J, Jiménez W, Ros J, Asbert M, Castro A, Arroyo V,* et al. Pathogenesis of arterial hypotension in cirrhotic rats with ascites: role of endogenous nitric oxide. Hepatology 1992; 15(2): 343−9.
8. *Nagasue N, Dhar DK, Yamanoi A, Emi Y, Udagawa J, Yamamoto A,* et al. Production and release of endothelin-1 from the gut and spleen in portal hypertension due to cirrhosis. Hepatology 2000; 31(5): 1107−14.
9. *Hankey GJ, Eikelboom JW.* Homocysteine and vascular disease. Lancet 1999; 354(9176): 407−13.
10. *Woitas RP, Stoffel-Wagner B, Poege U, Sauerbruch T.* Increased homocysteine in liver cirrhosis: a result of renal impairment? Clin Chem 2002; 48(5): 796−7; author reply 797−8.
11. *Ferré N, Gómez F, Camps J, Simó JM, Murphy MM, Fernández-Ballart J,* et al. Plasma homocysteine concentrations in patients with liver cirrhosis. Clin Chem 2002; 48(1): 183−5.
12. *Brattström L, Lindgren A, Israelsson B, Andersson A, Hultberg B.* Homocysteine and cysteine: determinants of plasma levels in middle-aged and elderly subjects. J Intern Med 1994; 236(6): 633−41.
13. *Simić-Ogrizović S, Radivojević D, Radovic M, Lezaic V, Mirković D, Babić D,* et al. Factors associated with hyperhomocysteinemia after renal transplantation. Ren Fail 2006; 28(1): 57−62.
14. *Suliman ME, Qureshi AR, Bárány P, Stenvinkel P, Filho JC, Anderstam B,* et al. Hyperhomocysteinemia, nutritional status, and cardiovascular disease in hemodialysis patients. Kidney Int 2000; 57(4): 1727−35.
15. *Guttormsen AB, Schneede J, Fiskerstrand T, Ueland PM, Refsum HM.* Plasma concentrations of homocysteine and other aminothiol compounds are related to food intake in healthy human subjects. J Nutr 1994; 124(10): 1934−41.
16. *Refsum H, Fiskerstrand T, Guttormsen AB, Ueland PM.* Assessment of homocysteine status. J Inherit Metab Dis 1997; 20(2): 286−94.
17. *García-Tevijano ER, Berasain C, Rodríguez JA, Corrales FJ, Arias R, Martín-Duce A,* et al. Hyperhomocysteinemia in liver cirrhosis: mechanisms and role in vascular and hepatic fibrosis. Hypertension 2001; 38(5): 1217−21.
18. *Merrill AH Jr, Henderson JM.* Diseases associated with defects in vitamin B6 metabolism or utilization. Annu Rev Nutr 1987; 7: 137−56.
19. *Kharbanda KK.* Alcoholic liver disease and methionine metabolism. Semin Liver Dis 2009; 29(2): 155−65.

Received on Avgust 23, 2011.
Revised on October 31, 2011.
Accepted on November 2, 2011.
OnLine-first September 2012.

PRACTICAL ADVICES
FOR PHYSICIANS

UDC: 616-072.1::617-089.5-089.819.3
DOI: 10.2298/VSP1301061S

The present and future of fiberoptic intubation

Sadašnjost i budućnost fiberoptičke intubacije

Zoran Slavković*†, Dušica M. Stamenković†, Predrag Romić*, Aleksandar Tomić*‡, Novak Milović*‡, Mirko Jovanović§, Menelaos Karanikolas‖

***Faculty of Medicine of the Military Medical Academy, University of Defence, Belgrade, Serbia; †Clinic of Anesthesiology and Intensive Care, ‡Clinic of Vascular Surgery, §Clinic of Urology, Military Medical Academy, Belgrade, Serbia; ‖Department of Anesthesiology, Washington University School of Medicine, St. Louis, Missouri, USA**

Key words:
fiber optic technology; intubation, intratracheal; laryngeal masks; bronchoscopes; endoscopes; anaesthesia.

Ključne reči:
fiberoptička tehnologija; intubacija, intratrahejna; laringealne maske; bronhoskopi; endoskopi; anestezija.

Introduction

Difficult airway management is critical during induction of anesthesia, and airway-related complications are the most frequent causes of morbidity, mortality and litigation against anesthesiologists [1–3]. Fiberoptic intubation (FOI) is the gold standard for endotracheal intubation in difficult or compromised airway situations when preservation of spontaneous breathing is detrimental [4, 5].

However, FOI is still not a routine technique due to lack of education and practice [6]. In this paper we presented basic principles of fiberoptic intubation, together with the recent developments in this field.

Fiberoptic bronchoscope

Although numerous types of supraglottic devices, rigid fiberoptic scopes and video-laryngoscopes were developed in recent years, fiberoptic bronchoscope is the only available device for nasal intubation, and is the recommended device for tracheal intubation under topical anesthesia in awake patients.

A part of light waves is transmitted, absorbed or reflected. The light is reflected along glass fibers and leaves them on the other end. A group of optic fibers forms a bundle. A typical bundle consists of 10,000 fibers, each of them has an 8 to 10 μm diameter. In order to minimize light deflection, each fiber is surrounded with 1 μm of isolator. Picture transmitting fibers known as coherent bundle consist of specifically arranged fibers.

A fiberoptic bronchoscope (FOB) consists of the body, an endoscope, universal light transmitter, camera and monitor or eye-piece (Figure 1). The FOB body has a handle for

Fig. 1 – The fiberoptic bronchoscope designed specifically for tracheal intubation (LF-V Tracheal Intubation Videoscope, Olympus Medical Systems Corporation, Tokyo, Japan).

moving the end of the endoscope in the vertical (up and down) plain, an eye-piece with a knob or button for adjusting the focus of the image, and a working channel that can be used for aspiration of secretions or delivery of oxygen or drugs. The endoscope, which is protected with waterproof plastic, contains optic fibers which transmit images from the end of the scope to the eye-piece or LCD screen, and also

Correspondence to: Zoran Slavković, Clinic of Anesthesiology and Intensive Care, Military Medical Academy, Crnotravska 17, 11 000 Belgrade, Serbia. Phone. +381 11 661 003. E-mail: zoran.slavkovic@vma.mod.gov.rs

contains a non-coherent bundle for transmitting light from the light source to the object of interest.

FOBs designed for tracheal intubation newer, in addition to the work channel for aspiration of secretions, an incorporated antifogging system. The latest FOB model (Olympus MAF type include TM Olympus Medical Systems Corporation, Tokyo, Japan) has an incorporated camera, light source, small monitor and recording system (Figure 2). This type of FOB enables FOI in emergency cases and on the field, lowers the cost of equipment and is suitable for education. Tele-anesthesia is a recent, interesting development, whereby developments in robotic surgery are applied in anesthesia. As a result, a successful nasal and oral FOI has been performed on an airway simulation mannequin, using the multipurpose DaVinci Surgical System type S (DVS), (Intuitive Surgical, Sunnyvale, California, USA) [7].

Fig. 2 – The recently presented fiberoptic bronchoscope with a built-in camera, light source and 2.5 inch monitor (Olympus MAF type TM Olympus Medical Systems Corporation, Tokyo, Japan)

Basic principles of FOI

The first case of FOI was described in 1973 [8]. Today FOI is considered a basic anesthesia skill and new generations of anesthesiologists are expected to learn the techniques of nasal and oral FOI [6, 9, 10]. Education on FOI is a complex process, because data show that courses and practice on mannequins are not as effective as expected. In addition, real-life problems like secretions and airway dynamics cannot be effectively simulated, but can be seen on previously made recordings. The new FOI education system is based on American Heart Association echosonographic teaching known as "practice while watching" [9–11].

FOI is indicated when airway management with direct laryngoscopy is expected to be difficult or impossible (Table 1). Contraindications to FOI are relative and are related to the skills of the anesthesiologist (Table 2). Skills with FOI, thorough knowledge of the airway anatomy and the ability to obtain clear views of the field are the cornerstones for effective fiberoptic endoscopy.

Additional equipment essential for safe and successful FOI is listed in Table 3 (see also Figure 3).

Six movements are possible with FOB: forward or backward, up or down in the vertical plain and left/right rotation. The easiest FOI technique is described as "up-down-up", which means advancing the FOB forward, ante flexion under the epiglottis and upward at the level of the anterior commissure [12]. FOB with diameter of 4.1 mm can be used

Table 1

Indications for fiberoptic intubation

Anticipated difficult airway in cooperative and non coopertive patients
Unexpectedly difficult or unsuccesful intubation
Tracheal intubation in a patient with:
• Limited neck range of motion
• Cervical spine pathology (rheumatoid arthritis, trauma)
• Upper airway edema (inhalation injury)
• Mandibular or pharyngeal pathology
• Tracheal pathology
Physician education (in patients with normal airway anatomy)
Physician practice for skill improvement

Table 2

Contraindications to fiberoptic intubation

Expected difficult mask ventilation (if a user is not familiar with extraglottic device which can be used for ventilation)
Massive upper airway hemorrhage
Upper airway obstruction
Risk for regurgitation or vomiting (if a user is not familiar with the procedure for awake intubation)
Inexperienced anesthesiologist

Table 3

Equipment for fiberoptic intubation

Fiberoptic bronchoscope
Oropharyngeal airway: Ovassapian, Berman, Williams
Nasopharyngeal airway, standard tracheal tube connector, ventilation mask
Laryngeal mask, Intubation laryngeal mask
Stylet, bougie, Magill forceps
Laryngoscope with different blade types and sizes
Two suctions: one for the fiberoptic bronchoscope and another one for oroparyngeal suctioning, suction catheters
Endotracheal tubes in different sizes, from the size 5 to the size 8
Local anesthetics
Resuscitation drugs
Lubricating gel, lidocaine gel
Ambu bag
Fixation band
Anti-fogging substance
Cricothyrotomy and tracheostomy sets
Oxygen source
Monitoring equipment (electrocardiography, pulse oximetry, noninvasive blood pressure, capnography)

Fig. 3 – The fiberoptic bronchoscope and the accompaning airway equipment for difficult airway management

for placement of an endotracheal tube (ETT) with diameter ≥ 5 mm or a double lumen tube size 37 or greater. A smaller diameter FOB should be used for placement of ETT ≥ 4.5, in cases of specific pathological changes or in pediatric patients.

The standard FOB is ergonomically constructed for handling with the right hand. It is advised that proximal part of FOB is handled with the third, fourth and fifth finger of the right hand, while the second finger is on the suction port and the first finger is on the FOB commands. The left hand lies on the face of the patient and handles the distal part of the FOB like a pen. The FOB needs to remain straight, and bending should be avoided. The left hand is used to advance the FOB along the airway, but FOB movement should be slow, to allow recognition of the relevant airway structures.

In order to facilitate smooth movement, the outer surface of the FOB should be lubricated, and an anti-fogging substance should be used for lens clearance. In addition, the FOB should be connected to an external oxygen source, so that oxygen can be insufflated during FOI. Ideally, the ETT should be somewhat wider (but not much wider) than the bronchoscope, for optimal maneuverability. An ETT approximately 1.5 mm wider than the FOB is ideal, whereas a much wider ETT (i.e. 9.0 ETT with 4.5 FOB) would make placement of the ETT more difficult.

Patient preparation

Depending on the clinical situation, patient age, comorbidities and specific airway problems, FOI can be performed with a patient being awake, sedated or completely anesthetized. Maintenance of spontaneous breathing is recommended in patients with difficult airway, whereas FOI under general anesthesia is generally preferred when FOI is used for education, in patients with normal airway anatomy.

Adequate oxygenation, provision of sedation without compromising spontaneous breathing and attenuation of laryngeal reflexes are important for successful FOI. Patients need to be informed about the FOI procedure, potential complications, and the possibility that tracheostomy may be needed [13].

Preparation for FOI includes administration of an antisialogogue 20 min before the procedure (Table 4), in order to minimize secretions that can compromise the fiberoptic view. In our experience, scopolamine is an excellent antisialogogue.

Table 4

Antisialogogues

Drug	Dose	Route
Glycopyrrolate	0.2–0.4 mg	IV, IM
Atropine	0.5–1 mg	IV, IM
Scopolamine	0.2–0.4 mg	IM

IV: intravenous
IM: intramuscular

Topical anesthesia

Lidocaine 4% can be used for topical anesthesia of the nose, mouth, pharynx and larynx (max dose 4 mg/kg) [14]. Absorption of nebulized lidocaine is 25%, and the peak effect occurs after 30 min [15].

The trigeminal nerve innervates the nasal and oral mucosa, the upper part of the nasopharynx, the soft palate and the tonsils. Topical anesthesia of the mouth and oropharynx can be accomplished using a combination of benzocaine, tetracaine and butamben. A tongue blade can be used to apply pressure on the tongue and posterior pharynx, in order to evaluate whether topical anesthesia is adequate on all structures of interest [16]. Topical Lidocaine 4% has a rapid onset of effect (within one minute), reaches the peak effect after two to five minutes, and its duration of action is 30–45 minutes [17].

Three types of regional blocks can be used in anesthesia of the upper airway [18]. The glossopharyngeal nerve innervates the posterior third of the tongue, the part of the soft palate, oropharynx and upper part of epiglottis [16]. Bilateral injection 2 ml of lidocaine 2% in the basal part of the tonsilar arch blocks the glossopharyngeal nerve and provides anesthesia of the oropharynx. The superior laryngeal nerve innervates the base of the tongue, the epiglottis, the piriformis fossa and the vallecula. It can be blocked with bilateral injection 2 ml of lidocaine 2%, caudal from the hyoid bone horns and the medial to the external carotid artery [16]. Laryngeal stimulation causes glottis closure. Superior laryngeal nerve block provides anesthesia of the larynx and trachea under the vocal cords. Translaryngeal block is performed with injection of 4 ml lidocaine 4% with a 22 G needle through the cricothyroid membrane [16]. After aspiration of air confirms that the needle is in the trachea, lidocaine can be injected to block the recurrent laryngeal nerve and the superior laryngeal nerve [15]. However, because of concerns about complications such as tracheal injury, bleeding and subcutaneous emphysema, translaryngeal blocks are rarely used.

If the nasal route is used for FOI, vasoconstrictor and application of local anesthetic in the nasopharynx is also necessary.

The standard monitoring is mandatory during FOI (Table 3), but additional monitoring may be needed, depending on patient condition and comorbidities. During FOI, the patient can be in the sitting, semi-sitting or supine position. FOB can also be used with the patient in the prone or lateral decubitus position [19] in rare situations where urgent intubation is necessary. Chin lift, jaw thrust or pulling of the tongue may be necessary in order to facilitate visualization of the vocal cords in such cases.

Sedation

Adequate (but not excessive) sedation is important, in order to safely provide good intubating conditions with minimal cough or patient movement, while maintaining patient comfort, amnesia and anxiolysis. A variety of newer and older sedative agents can be used for sedation during FOI (Table 5), and the depth of sedation can be monitored using the Ramsay Sedation Scale (aiming for scores ≥ 2), entropy or BIS monitoring.

Table 5

Sedative / Hypnotic medications

Drug	Dose for intravenous use
Dexmedetomidine	1 μg/kg during 10 min 0.2–0.7 μg/kg/h
Midazolam	0.01–0.03 mg/kg 0.25–1 μg /kg/min
Diazepam	0.01–0.04 mg/kg
Meperidine	0.5 mg/kg
Propofol	25–100 μg /kg/min
Remifentanil	0.025–0.1 μg /kg/min 1–3 μg /kg
Fentanyl	25–50 μg slowly 0.01–0.04 μg /kg/min
Alfentanyl	10–25 μg /kg 0.25–1 μg /kg
Ketamine	0.2–0.5 mg/kg 20–40 mg 5–20 μg /kg/min
Haloperidol*	2–5 mg

***Haloperidol use is indicated in intoxicated or agitated patients**

Fentanyl and alfentanil should be used with great caution as sedative agents, because they can cause respiratory depression [20, 21]. Remifentanil, having very favorable characteristics for sedation during FOI [22], can be administered by target controlled infusion (TCI) (0.8 ng/mL) or manual controlled infusion (0.75 μg/kg and 0.075 μg/kg/min). Published data suggest that remifentanil can confer better hemodynamic stability and intubating conditions when administered by TCI [22].

Propofol administered by TCI and sevoflurane can also provide high degree of success during FOI [23]. However, compared to propofol, remifentanil seems to confer better conditions for endoscopy and intubation [24].

Dexmedetomidine, combined with small doses of midazolam, is particularly effective in providing adequate sedation while preserving spontaneous respiration during FOI in patients

with difficult airway [25, 26]. A combination of dexmedetomidine with midazolam (0.02 mg/kg) can achieve better patient cooperation compared with midazolam only (0.05 mg/kg) [27].

Orotracheal FOI

FOI can be performed through the mouth with use of specially designed oropharyngeal airways. The oropharyngeal airway can be placed when the patient is sedated, and can serve as the conduit for passage of the FOB during FOI. Several types of oropharyngeal airways are commercially available for FOI, and their role is to provide better control of FOB position and protect the FOB and patients' teeth (Figure 4). As the FOB advances, the base of the tongue is

Fig. 4 – The Berman oropharyngeal airway for fiberoptic intubation enables easier ETT placement, protecting the fiberoptic bronchoscope from patient's teeth

the first visible structure, and then the epiglottis can be visualized. Then, careful advancement of the FOB under the epiglottis reveals the anterior commissure. With the glottis in the center of the picture, the aryepiglottic folds, vestibular folds, vocal chords and opening of the larynx into the trachea can be clearly seen (Figure 5). Then, as the FOB advances through the vocal chords, visualization of the tracheal rings confirms that the FOB has entered the trachea. The FOB should be advanced, until the tip of the FOB is located three to five centimeters above the carina, in order to facilitate appropriate placement of the ETT. The ETT is then advanced gently over the FOB, using the right hand. The cuff of the ETT is inflated after visual confirmation of proper ETT position above the carina, and the FOB is removed. Finally, the ETT is connected to the circuit of the anesthesia machine,

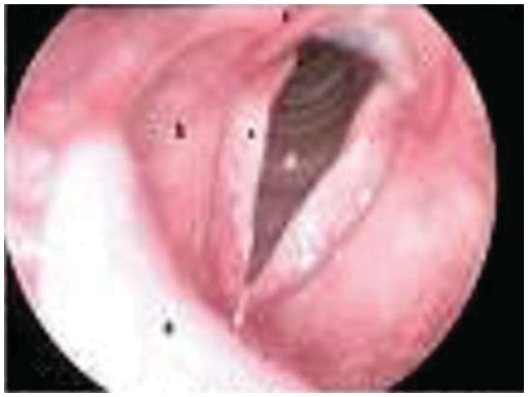


Fig. 5 – View through the fiberoptic bronchoscope video laryngoscope: the glottis can be seen at the center of the picture

and endotracheal placement is also confirmed by capnography and by auscultation for bilateral breath sounds.

Nasal FOI

Nasal FOI conducted through a nasopharyngeal tube, and is probably the easiest method for reaching the vocal cords. Nasal intubation is contraindicated in the presence of base of the skull fractures, brain tumors, coagulopathy, nasal obstruction, tumors localized in the epiglottis and bacteremia [28].

During nasal FOI, the ETT is lubricated with gel and is then advanced through the nostril until the cuff of the ETT disappears. Then, the ETT serves as a conduit for advancing the FOB. After the FOB passes through the nasopharynx pass, the uvula and the base of the tongue can be seen. Then, as the FOB advances further, the epiglottis, the ariepiglotic folds and parts of the glottis parts can be seen.

Additional equipment to facilitate FOI

FOI is considered a "low complexity skill" in modern anesthesia practice: it is a critical part of difficult airway guidelines [29], and its failure rate in experienced hands is around 1.2% [30]. There are several different techniques for "low complexity skill" FOI. Low skill FOI is achieved by passing a FOB loaded with an aintree intubation catheter (AIC, Cook UK, Letchworth, Herts, UK) through laryngeal mask (LMA) which had already been placed to maintain the patient's airway [31, 32]. After FOI, the removal of the LMA over the ETT is impaired by the short length of the ETT, easily resulting in tube dislocation from the trachea. Among several techniques to overcome this problem, Arndt tube exchanger (Arndt Airway Exchange Catheter Set, Cook Critical Care, Bloomington, Indiana) offers a reliable method not only for safe removal of the laryngeal mask over the tracheal tube but also for insertion of an adequate tracheal tube, particularly in pediatric patients [32, 33].

Placement of LMA without grids or intubating LMA (ILMA) provides oxygenation, ventilation and guides ETT. Use of ILMA (LMA-Fastrach™ , LMA North America, Inc., San Diego, CA) is the best for FOI. The first step is to remove a connector from the ETT. Then, an ETT size 6.0 can

be placed through LMA size three or four. FOI through LMA involves several steps [34]: First, the ETT is placed inside the LMA. When the ETT reaches the epiglottis elevator, the FOB is advanced through the ETT, and then through the vocal cords, into the trachea. Then, the ETT is advanced over the FOB to the trachea. The size of the ETT is important during this procedure: the ETT should be at least 6 mm, because a smaller ETT cannot lift the epiglottis elevator [34].

A rigid video laryngoscope (RVL) can also be very useful when used in combination with the FOB during FOI, because it helps open the oropharynx and minimize FOB lateralization [35]. After anesthesia induction, the RVL is used to visualize the upper part of the larynx, and then the FOB is placed in front of the glottis [36]. The assistant holds the FOB, which is then advanced through the vocal cords, and then the ETT is advanced over the FOB into the trachea. This technique combines two fiberoptic views: an external view through the RVL and an internal view through the FOB.

FOB for single lung ventilation

Intubation with a double-lumen endotracheal tube using FOB is possible, but extremely difficult, due to the construction and the small size of the double-lumen tube [37, 38]. Therefore, when lung isolation is needed, airway experts recommend placing a regular ETT through the FOB and then either use a bronchial blocker, or exchange from a regular ETT to a double-lumen ETT using a tube exchanger [38–40].

FOI complications

FOI complications are usually the consequence of excessive sedation or inadequate technique [41]. The most important complications are listed in Table 6.

Table 6

Complications of fiberoptic intubation

Epistaxis
Laryngeal trauma
Laryngospasm
Tracheal tube cannot pass between vocal cords
Pneumothorax
Endobronchial intubation
Toxicity from local anesthetic overdose
Respiratory depression

Extubation after FOI

Criteria for extubation of the trachea include hemodynamic stability, the return of protective airway reflexes, adequate spontaneous respiration, and the absence of significant airway edema. However, when extubating a patient with a difficult airway, the FOB should be readily available, in case an airway emergency occurs. Placement of an endotracheal tube exchanger through the ETT, so that the exchanger remains in place for some time after extubation, can be extremely helpful if reintubation is needed.

FOB maintenance

Endoscopes used for FOI are potential sources of infection, and therefore should be cleaned thoroughly after each use. Proper cleaning of a FOB with a special brush and water can effectively remove 99.9% of microorganisms. Glutaraldehyde is the agent most frequently used for sterilization and removal of spores. Following proper sterilization, a FOB can be stored in a straight hanging position or in special boxes.

Conclusion

Because of insufficient training, anesthesiologists rarely use FOI, even in situations when the use of FOI is clearly indicated. Proper patient and airway preparation enables fast and easy recognition of airway structures, followed by safe, timely ETT placement with minimal patient discomfort. Ongoing education and practice with FOI are necessary in order to enhance safety while managing patients with difficult airways.

Acknowledgment

We thank Mr. Goran Bukonja from Olympus d.o.o. Belgrade for providing original photos.

Conflict of interest statement

This work was supported solely by department funds. All authors state they have not received any financial support from any of the companies mentioned in this manuscript, and, therefore, they have no conflicts of interest to report.

REFERENCES

1. *Peterson GN, Domino KB, Caplan RA, Posner KL, Lee LA, Cheney FW*. Management of the difficult airway: a closed claims analysis. Anesthesiology 2005; 103(1): 33−9.
2. *Chenny FW*. Changing trends in anesthesia related death and permanent brain damage. ASA closed claims project. ASA Newsletter 2002; 66 (6): 6−8.
3. *Cheney FW, Posner KL, Lee LA, Caplan RA, Domino KB*. Trends in anesthesia-related death and brain damage: A closed claims analysis. Anesthesiology 2006; 105(6): 1081−6.
4. *American Society of Anesthesiologists Task Force on Management of the Difficult Airway*. Practice guidelines for management of the difficult airway: an updated report by the American Society of Anesthesiologists Task Force on Management of the Difficult Airway. Anesthesiology 2003; 98(5): 1269−77.
5. *Koerner IP, Brambrink AM*. Fiberoptic techniques. Best Pract Res Clin Anaesthesiol 2005; 19(4): 611−21.
6. *Agrò FE, Cataldo R*. Teaching fiberoptic intubation in Italy: state of the art. Minerva Anestesiol 2010; 76(9): 684−5.
7. *Tighe PJ, Badiyan SJ, Luria I, Lampotang S, Parekattil S*. Robot-assisted airway support: a simulated case. Anesth Analg 2010; 111(4): 929−31.
8. *Boggan E, Trudnowski RJ*. Endotracheal intubation using a fiberoptic bronchoscope. J Am Assoc Nurse Anesth 1973; 41(1): 61−2.

9. *Petrini F.* Airway management and guideline implementation: the gap from research to daily practice. Minerva Anestesiol 2008; 74(9): 449−50.
10. *Guglielmi M, Urbaz L, Tedesco C, Pusceddu A, Sogni A, Ronzoni G.* A structured training program for awake fiber optic intubation: teaching the complete package. Minerva Anestesiol 2010; 76(9): 699−706.
11. *Stringer KR, Bajenov S, Yentis SM.* Training in airway management. Anaesthesia 2002; 57(10): 967−83.
12. *Diemunsch P, Joshi GP, Collange O.* Fiberoptic intubation: skills and alternative techniques. ESA Copenhagen 2009; 19RC1; 207−10.
13. *Stoelting RK, Hillier SC.* Anticholinergic drugs. In: *Stoelting RK, Hillier SC*, editors. Handbook of Pharmacology and Physiology in Anesthetic Practice. 2nd ed. New York: Lippincott Williams&Wilkins, 2005. p. 267−75.
14. *Cox B, Durieux ME, Marcus MA.* Toxicity of local anaesthetics. Best Pract Res Clin Anaesthesiol 2003; 17(1): 111−36.
15. *Parkes SB, Butler CS, Muller R.* Plasma lignocaine concentration following nebulization for awake intubation. Anaesth Intensive Care 1997; 25(4): 369−71.
16. *Sutherland L, Misita D.* Regional and topical anesthesia for endotracheal intubation. In: *Hadzic A* editor. Textbook of regional anesthesia and acute pain management. 1st ed. New York: McGraw Hill Companies, Inc; 2007. p. 331−41.
17. *Ritchi JM, Greene NM.* Local Anesthetics. In: *Gilman AG, Goodman LS, Gilman A,* editors. Goodman and Gilman's the Pharmacological Basis of Therapeutics. 6th ed. New York: MacMillan Publishing Co; 1980. p. 300−20.
18. *Simmons ST, Shawn T.; Schleich, Arno R.* Airway Regional Anesthesia for Awake Fiberoptic Intubation. Reg Anesth Pain Med 2002; 27(2): 180−92.
19. *Hung MH, Fan SZ, Lin CP, Hsu YC, Shih PY, Lee TS.* Emergency airway management with fiberoptic intubation in the prone position with a fixed flexed neck. Anesth Analg 2008; 107(5):1704−6.
20. *Cohen LB, DeLegge MH, Aisenberg J, Brill JV, Inadomi JM, Kochman ML,* et al. AGA Institute review of endoscopic sedation. Gastroenterology 2007; 133(2): 675−701.
21. *Sternlo JE, Sandin RH.* Recurrent respiratory depression after total intravenous anaesthesia with propofol and alfentanil. Anaesthesia 1998; 53(4): 378−81.
22. *Yeganeh N, Roshani B, Azizi B, Almasi A.* Target-Controlled Infusion of Remifentanil to Provide Analgesia for Awake Nasotracheal Fiberoptic Intubations in Cervical Trauma Patients. J Trauma 2010; 69(5): 1185−90.
23. *Péan D, Floch H, Beliard C, Piot B, Testa S, Bazin V,* et al. Propofol versus sevoflurane for fiberoptic intubation under spontaneous breathing anesthesia in patients difficult to intubate. Minerva Anestesiol 2010; 76(10): 780−6.
24. *Rai MR, Parry TM, Dombrovskis A, Warner OJ.* Remifentanil target-controlled infusion vs propofol target-controlled infusion for conscious sedation for awake fibreoptic intubation: a double-blinded randomized controlled trial. Br J Anaesth 2008; 100(1): 125−30.
25. *Bergese SD, Khabiri B, Roberts WD, Howie MB, McSweeney TD, Gerhardt MA.* Dexmedetomidine for conscious sedation in difficult awake fiberoptic intubation cases. J Clin Anesth 2007; 19(2): 141−4.
26. *Bergese SD, Candiotti KA, Bokesch PM, Zura A, Wisemandle W, Bekker AY.* Phase IIIb, randomized, double-blind, placebo-controlled, multicenter study evaluating the safety and efficacy of dexmedetomidine for sedation during awake fiberoptic intubation. Am J Ther 2010; 17(6): 586−95.
27. *Bergese SD, Patrick Bender S, McSweeney TD, Fernandez S, Dzwonczyk R, Sage K.* A comparative study of dexmedetomidine with midazolam and midazolam alone for sedation during elective awake fiberoptic intubation. J Clin Anesth 2010; 22(1): 35−40.
28. *Roppolo LP, Vilke GM, Chan TC, Krishel S, Hayden SR, Rosen P,* et al. Nasotracheal intubation in the emergency department, revisited. J Emerg Med 1999; 17(5): 791−9.
29. *Pearce AC.* Fiberoptic intubation: modern clinical practice. Fiberoptic intubation: skills and alternative techniques. ESA Copenhagen 2009; 19RC2; 211−6.
30. *Ovassapian A, Dykes MH, Golmon ME.* A training programme for fiberoptic nasotracheal intubation. Use of model and live patients. Anaesthesia 1983; 38(8): 795−8.
31. *Higgs A, Clark E, Premraj K.* Low-skill fibreoptic intubation: use of the Aintree Catheter with the classic LMA. Anaesthesia 2005; 60(9): 915−20.
32. *Joffe AM, Liew EC.* Intubation through the LMA-Supreme: a pilot study of two techniques in a manikin. Anaesth Intensive Care 2010; 38(1): 33−8.
33. *Weiss M, Mauch J, Becke K, Schmidt J, Jöhr M.* Fibre optic-assisted endotracheal intubation through the laryngeal mask in children. Anaesthesist 2009; 58(7): 716−21. (German)
34. *Ferson DZ, Rosenblatt WH, Johansen MJ, Osborn I, Ovassapian A.* Use of the intubating LMA-Fastrach in 254 patients with difficult-to-manage airways. Anesthesiology 2001; 95 (5): 1175−81.
35. *Diemunsch PA, Noudem Y.* Virtual and real imaging in the management of the difficult airway. Acta Anaesthesiol Belg 2008; 59(3): 179−80.
36. *Greib N, Stojeba N, Dow WA, Henderson J, Diemunsch PA.* A combined rigid videolaryngoscopy-flexible fibrescopy intubation technique under general anesthesia. Can J Anaesth 2007; 54(6): 492–3.
37. *Nakanishi K, Terao K, Nabeta T, Yorozuya T, Kuzume K, Nagaro T.* Successful oral fiberoptic intubation by a double-lumen tube under the ventilation only via nostrils with a child size mask in a patient with limited mouth opening. Masui 2009; 58(5): 616−9. (Japanese)
38. *Cohen E, Benumof JL.* Lung separation in the patient with a difficult airway. Curr Opin Anaesthesiol 1999; 12(1): 29−35.
39. *Cohen E.* Methods of lung separation. Minerva Anestesiol 2004; 70(5): 313−8.
40. *Cohen E.* Recommendations for airway control and difficult airway management in thoracic anesthesia and lung separation procedures. Are we ready for the challenge? Minerva Anestesiol 2009; 75(1−2): 3−5.
41. *Heidegger T, Starzyk L, Villiger CR, Schumacher S, Studer R, Peter B,* et al. Fiberoptic intubation and laryngeal morbidity: a randomized controlled trial. Anesthesiology 2007; 107(4): 585−90.

Received on April 8, 2011.
Accepted on June 3, 2011.

CASE REPORT

UDC: 616.61-006-07/-08
DOI: 10.2298/VSP1301068S

Giant renal oncocytoma

Džinovski onkocitom bubrega

Nebojša Stojanović*, Ivan Ignjatovic†, Miloš Kostov‡¶, Žaklina Mijović§, Sladjana Živković*, Branko Košević‖

***Department of Urology, ‡Department of Pathology, Military Hospital Niš, Niš, Serbia; †Clinic of Urology, Clinical Center Niš, Niš, Serbia; §Institute of Pathology, Faculty of Medicine, Niš, Serbia; ‖Urology Clinic, Military Medical Academy, Belgrade, Serbia, ¶Faculty of Medicine of the Military Medical Academy, University of Defence, Belgrade, Serbia**

Abstract

Background. Renal onkocytoma is a distinctive benign tumor derived from epithelial cells of the distal renal tubules. These tumors are often clinically asymptomatic, diagnosed accidentally and difficult to distinguish from renal cell carcinoma. **Case report.** We presented a giant renal onkocytoma in a man aged 64, without any signs or symptoms of the urogenital system disorder. The preoperative diagnosis described the tumor mass of the right kidney, size 16 × 14 cm, and indicated a malignant tumor of kidney. The patient underwent radical nephrectomy. The tumor was encapsulated at the intersection with the characteristic central hyaline scar. Microscopically, it was built of uniform polygonal cells with abundant eosinophilic cytoplasm. Immunohystochemiclly, tumor cells were immunoreactive to CK AE1/AE3 and CD 117, but showed negative immunoreactivity to CK 7, RCC marker and Vimentin. **Conclusion.** Giant renal oncocytomas are rare tumors with benign clinical course. As a rule, they are discovered by accident. Clinical differentiation from malignant tumors of the kidney is not possible. They are treated surgically, mainly by radical nephrectomy. A definitive diagnosis is made only by histopathological examination of tumors using immunohistochemical marker panels.

Key words:
kidney neoplasms; diagnosis, differential; adenoma, oxyphilic; diagnosis; histological tehniques; surgical procedures, operative.

Apstrakt

Uvod. Onkocitom bubrega je karakterističan benigni tumor, poreklom od epitelnih ćelija distalnih bubrežnih tubula. Klinički su često asimptomatski, dijagnostikuju se slučajno i teško ih je razlikovati od karcinoma bubrežnih ćelija. **Prikaz bolesnika.** Prikazali smo gigantski onkocitom bubrega kod muškarca, starog 64 godine, bez znakova i simptoma od strane urogenitalnog sistema. Preoperativnom dijagnostikom opisana je tumorska masa desnog bubrega veličine l6 × 14 cm, koja je ukazala na maligni tumor bubrega. Bolesnik je podvrgnut radikalnoj nefrektomiji. Tumor je na preseku bio inkapsuliran sa karakterističnim centralnim hijalinim ožiljkom. Mikroskopski, bio je građen je od uniformnih poligonalnih ćelija, sa obilnom eozinofilnom citoplazmom. Imunohistohemijski, tumorske ćelije su bile imunoreaktivne na CK AE1/AE3 i CD 117, a negativnu imunoreaktivnost pokazivale su na CK 7, RCC marker i Vimentin. **Zaključak.** Gigantski onkocitomi bubrega su retki tumori sa benignim kliničkim tokom. Po pravilu se otkrivaju slučajno. Klinička diferencijacija od malignih tumora bubrega nije moguća. Lečenje je operativno, uglavnom radikalnom nefrektomijom. Definitivna dijagnoza se postavlja isključivo histopatološkim pregledom tumora, primenom panela imunohistohemijskih markera.

Ključne reči:
bubreg, neoplazme; dijagnoza, diferencijalna; adenom, oksifilni; dijagnoza; histološke tehnike; hirurgija, operativne procedure.

Introduction

Oncocytoma is a benign epithelial tumor that consists of oncocytes, large cells characterized by the profusion of mitochondria with eosinophilic granular cytoplasm. Renal oncocytoma suggests the origin of distal renal tubule cells and accounts for about 3%–7% of primary renal tumors [1]. The majority of them are asymptomatic and are accidentally discovered [2]. Renal oncocytoma was first described in 1942 by Zippel [3] as a unique pathological entity. We presented a giant renal onkocytoma as a very rare entity.

Correspondence to: Nebojša Stojanović, Department of Urology, Military Hospital Niš, Bulevar dr Zorana Đinđića bb, Niš, Serbia. Phone.: +381 18 508 849, Fax.: +381 18 238 191. E-mail: nesha.stojanović@gmail.com

Case report

A 64-year-old male patient after being examined for hypertension, was referred to the urologist due to the initial stage of renal failure. He was without any symptoms regarding the urogenital tract. On physical examination, by bimanual palpation of the flank area a painless mass the size of an adult male fist was palpated in the right loin. Ultrasonography followed by multislice computed tomography (MSCT) with intravenous urography (IVU) described a totally anatomical alteration of the right kidney with an expansive encapsulated nodule 16 × 14 × 13 cm with central zones of necrosis (Figure 1) with a

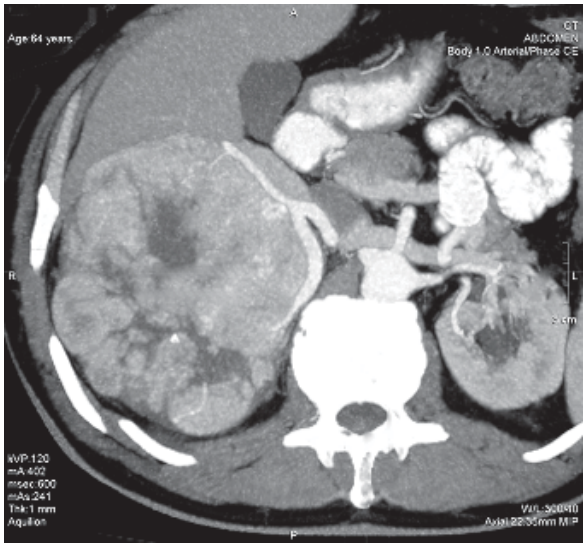


Fig. 1 – Multislice computed tomography of the abdomen – an expansive tumor of the kidney

partially preserved parenchyma in the upper pole of the kidney. The renal pelvis and ureter of the right kidney were not shown on IVU. Angiography described a hypervascular tumor mass in the arterial phase with pathological accumulation of contrast in the parenchyma phase (Figure 2). The complete radiologic imaging suggested renal cell carcinoma (RCC). The patient underwent radical nephrectomy employing transabdominal subcostal approach. During the course of an almost three hour surgery, there was no significant intraoperative hemmorhage as renal hilus blood vessels were timely tied off. The postoperative course was regular. The patient was discharged on the 9th postoperative day fully recovered.

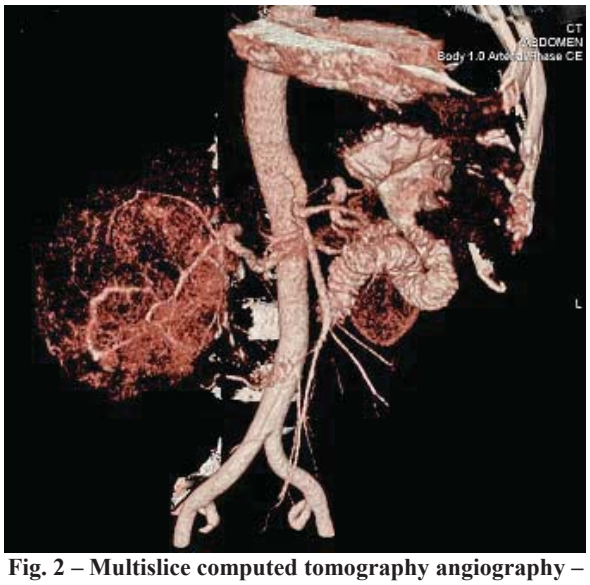


Fig. 2 – Multislice computed tomography angiography – pronounced pathological vascularization

Macroscopically, the kidney was deformed by the tumor and weighed around 900 g. On its section, there was a solid oval encapsulated tumor of 16 cm in diameter, lobular structure, tan brown in color with a central dense fibrous band with fibrous trabeculae extending out in a stellate pattern to the margins of the tumor and infrequent dark brown areas (Figure 3).

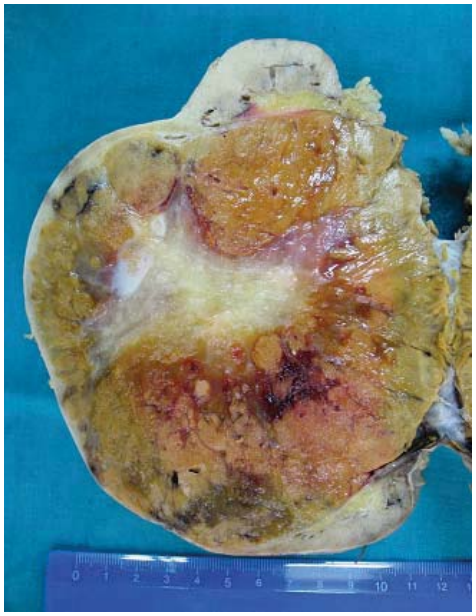


Fig. 3 – Macroscopic appearance of renal oncocytoma

For the purpose of routine histopathological examination, the material was fixated in 10% buffered, neutral formalin, embedded in paraffin, cut with a microtome 5 microns in depth and treated by the standard hematoxillin-eosin (HE) method. For the purpose of immunohistochemical analysis, streptavidin-biotin peroxidase technique was used in the standard procedure (DAKO, LSAB Kit), using monoclinic antibodies for cytokeratin AE1/AE3 (CK AE1/AE3), cytokeratin 7 (CK 7), RCC and Vimentin and polyclinic antibodies for CD117 (c-kit), (DAKO, Denmark). 3,3' – diaminobenzedin (DAB) was used as a chromogenic substrate and the slides were contrasted with Mayer hematoxillin.

Microscopically, the tumor was formed of uniformed polygonal cells with distinct cellular margins with rich eosinophilic granular cytoplasm, without nuclear atypia, with noticeable nucleoli arranged in an alveolar, solid or in a rare tubular manner (Figure 4). Mitoses were not seen. Immunohistochemically, the tumor cells showed weak cytoplasmatic immunoreactivity to CD 117 (Figure 5) and distinct diffuse cytoplasmatic immunoreactivity to CK AE1/AE3 (Figure 6) and the absence of immunoreactivity to CK 7, RCC marker and Vimentin.

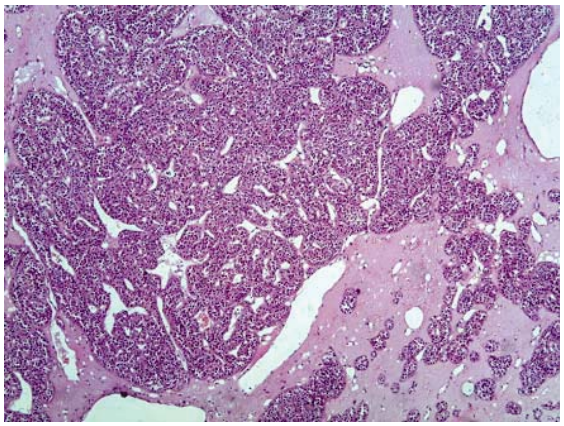


Fig. 4 – Aggregates of small eosinophilic cells – oncocytes (hematoxillin-eosin, × 50)

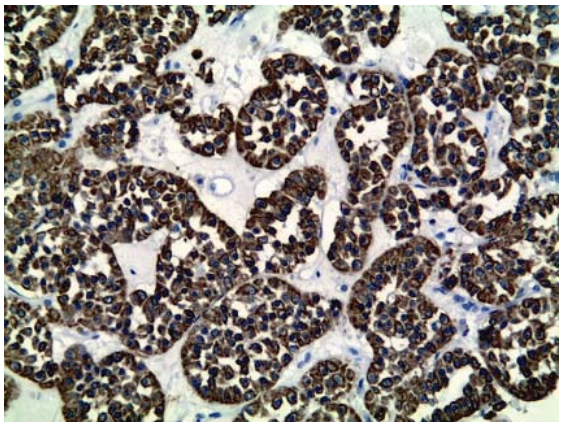


Fig. 5 – Weak focal cytoplasmic immunoreactivity of tumor cells to CD 117 (L SAB$^+$, × 200).

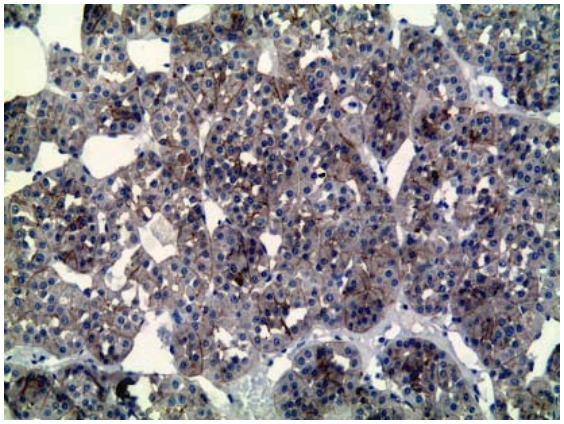


Fig. 6 – Diffuse cytoplasmic immunoreactivity expressed by tumor cells to CK AE1/AE3 (L SAB$^+$, × 200).

Discussion

Renal oncocytomas are grossly round, tan or light brown in color, encapsulated, well circumscribed and have the average size of 4–6 cm. Oncocytes are large epithelial cells with minimal nuclear atypia, with a developed eosinophilic cytoplasm and uniform nuclei. Oncocytomas built of well differentiated cells are benign nonaggressive tumors, which do not give metastasis and have a favorable clinical course [4].

Oncocytomas do not have a characteristic clinical presentation and are frequently diagnosed accidentally by using radiological imaging techniques such as ultrasound computed tomography (CT) or magnetic resonance imaging (MRI), because of other problems. Preoperative diagnostic methods are unable to differentiate oncocytomas from RCC. Oncocytomas are presented as solid, homogenous well circumscribed tumor formations with different attenuations similar to RCC. The central stellate pattern scar that is often imaged on the CT scan and a typical spoke-wheel pattern on the angiogram may suggest the diagnosis of oncocytoma but previous experiences have proved them unreliable and of insufficient predictive valuae [5]. Aspiration biopsy may give a preoperative diagnosis but is unreliable. A sufficient tumor specimen is not always obtained and there is a risk of hemorrhage from a hypervascular tumor. The limited value of biopsy is a confirmed presence of RCC and oncocytoma in the same lesion or in the different area of the same kidney. Due to the preoperative suspicion of RCC and the unreliable diagnosis by frozen section, radical nephrectomy is the safest method of therapy unless contraindicated by other factors (solitary kidney, bilateral tumors or poor renal function).

In a case report on renal oncocytoma in November 2010, being the fourth in weight (1973 g, dimensions 27 × 16 × 13 cm), Anastasiadis et al. [6] reviewed the largest and heaviest cases published in literature: Demos et al. [7] (4652 g, 27 × 20 × 15cm), Banks et al. [8] (3090 g , 21 × 18 × 15 cm), and Kiliç et al. [9] (2680 g, 20 × 15 × 10 cm). In no single case renal oncocytoma could be differentiated from RCC preoperatively [10] neither clinically nor by using radiological imaging.

It is very important to carefully examine both kidneys because 13% of patients have multiple oncocytomas and up to 32 % have synchronous RCC [11]. The definitive diagnosis is made by histopathological examination of the tumor, applying histochemical and immunohistochemical methods and ultrastructural analysis in some cases [12]. Renal oncocytomas show similar immunoprofile as RCC, especially with eosinophilic variations. Some studies suggest the use of different markers such as Vimentin, S-100 protein and CD82, but the interpretation of these markers must be done with caution [13].

Conclusion

Renal oncocytomas may be asymptomatic for prolonged periods of time and can became very large in size. Inadequacy of specific diagnostic methods and the overlapping of radiological characteristics with RCC make their clinical differentiation hard. Definitive diagnosis is usually made after removing the tumor surgically, through histopathological examination using adequate (immuno) histochemical analysis.

This case confirms the difficulties in making a preoperative diagnosis even by the use of contrasting the enhanced graphic representation of such a large tumor lesion. This emphasizes the necessity to include renal oncocytomas in the differential diagnosis of such lesions, as a reliable preoperative diagnosis of oncocytomas provides for nephron sparing surgery.

REFERENCES

1. *Kuroda N, Toi M, Hiroi M, Shuin T, Enzan H.* Review of renal oncocytoma with focus on clinical and pathobiological aspects. Histol Histopathol 2003; 18(3): 935−42.
2. *Campbell CS, Novick CA, Bukowski MR.* Renal tumours. In: *Campbell CS*, editor. Campbell's urology. 9th ed. Philadelphia: WB Saunders; 2007. p. 1567−637.
3. *Zippel L.* Zur Kenntnis der oncocytem. Virchow Arch Pathol Anat 1942; 308: 360–82.
4. *Eble JN, Sauter G, Epstein JI, Sesterhenn IA.* World Health Organization classification of tumours. Pathology and Genetics of Tumours of the Urinary System and Male Genital Organs. Lyon: IARC Press; 2004.
5. *Licht MR.* Renal adenoma and onkocytoma. Semin Urol Oncol 1995; 13(4): 262−6.
6. *Anastasiadis A, Dimitriadis G, Radopoulos D.* Incidental giant renal oncocytoma: a case report. J Med Case Reports 2010; 4: 358.
7. *Demos TC, Malone AJ Jr.* Computed tomography of a giant renal oncocytoma. J Comp Assist Tomogr 1988; 12(5): 899−900.
8. *Banks KL, Cherullo EE, Novick AC.* Giant renal oncocytoma. Urology 2001; 57(2)**:** 365.
9. *Kiliç S, Altinok MT, Ipek D, Ergin H.* Case report of a giant renal oncocytoma. Int Urol Nephrol 2003; 35(1)**:** 83−4.
10. *Choudhary S, Rajesh A, Mayer NJ, Mulcahy KA, Haroon A.* Renal oncocytoma: CT features cannot reliably distinguish oncocytoma from other renal neoplasms. Clin Radiol 2009; 64(5): 517−22.
11. *Bostwick DG, Cheng L.* Urologic surgical pathology. 2nd ed. Philadelphia: Elsevier, Mosby; 2008.
12. *Kostov M, Cerovic S, Tatic V, Spasic P, Skaro-Milic A, Knezevic-Usaj S,* et al. Morphological, histochemical, immunohistochemical and ultrastructural features of renal oncocytoma. The 7th Congress of Pathology of Yugoslavia with International Participation; 1996 June 13–15; Podgorica-Budva, Monte Negro, Yugoslavia. (Serbian)
13. *Truong LD, Shen SS.* Immunohistochemical diagnosis of renal neoplasms. Arch Pathol Lab Med 2011; 135(1): 92−109.

Received on May 16, 2011.
Revised on May 25, 2011.
Accepted on June 7, 2011.

CASE REPORT

UDC: 616.98-022.9
DOI: 10.2298/VSP1301072K

Atypical form of cat scratch disease in immunocompetent patient

Atipična forma bolesti mačjeg ogreba kod imunokompetentne bolesnice

Miroslav Kojić*, Dragan Mikić*†, Darko Nožić*†, Lidija Zolotarevski†‡

***Clinic for Infectious and Tropical Diseases, ‡Institute for Pathology, Military Medical Academy, Belgrade, Serbia; Faculty of Medicine of the Military Medical Academy, University of Defence, Belgrade, Serbia**

Abstract

Introduction. Cat scratch disease (CSD) is an acute infectious disease with benign course caused by the bacteria *Bartonella henselae.* Clinically, it is usually manifested as regional lymphadenopathy and mild infective syndrome. Rare forms of the disease which usually occur in immunocompromised presons are: encephalitis, transverse myelitis, neuroretinitis, granulomatosus conjunctivitis, arthritis, hepatitis etc. **Case report.** We presented an atypical form of cat scratch disease in a young immunocompetent female person. The disease was manifested with prolonged fever, rash, purulent lymphadenitis and hepatitis. The diagnosis was based on characteristic patohystological finding and exclusion of the other causes of lymphadenopathy. The patient was treated by antibiotics for a few weeks, with surgical incision and drainage of the purulent lymphadenitis. **Conclusion.** Atypical forms of CSD could be an important differential-diagnostic problem, especially if there is no opportunity for serological confirmation of the disease.

Key words:
cat-scratch disease; lymphadenitis; diagnosis; drug therapy; antibacterial agents; treatment outcome.

Apstrakt

Uvod. Bolest mačjeg ogreba (BMO) je akutno infektivno oboljenje benignog toka čiji je izazivač bakterija *Bartonella henselae.* Klinički, najčešće se ispoljava kao regionalna limfadenopatija uz blag infektivni sindrom. U retke forme bolesti, koje se obično javljaju kod imunokompromitovanih, spadaju encefalitis, transverzalni mijelitis, neuroretinitis, granulomatozni konjunktivitis, artritis, hepatitis i druge. **Prikaz bolesnika.** U radu je prikazan atipičan oblik BMO kod mlade imunokompetentne osobe ženskog pola. Bolest se ispoljila produženom febrilnošću, ospom, gnojnim limfadenitisom i hepatitisom. Definitivna dijagnoza je postavljena na osnovu karakterističnog patohistološkog nalaza i isključivanjem drugih uzročnika gnojnog limfadenitisa. Lečenje je sprovedeno višenedeljnom primenom antibiotika uz hiruršku inciziju i drenažu gnojnog limfadenitisa. **Zaključak.** Atipične forme BMO mogu predstavljati značajan diferencijalno-dijagnostički i terapijski problem, posebno kada nema mogućnosti za serološku potvrdu bolesti.

Ključne reči:
bolest mačje ogrebotine; limfadenitis; dijagnoza; lečenje lekovima; antibiotici; lečenje ishod.

Introduction

Cat scratch disease (CSD) is an acute infectious disease with benign course and good prognosis caused by *Bartonella (B) henselae*. It is the most frequent cause of chronic lymphadenopathy in children and adolescents [1]. *B. henselae* is a gram negative slow-growing, intracelullar bacteria which causes granulomatosus inflammation of the skin and regional lymph nodes [2–6]. The first case of CSD was described by Robert Debre in Paris in 1931, in a ten-year old boy with limphadenitis who was in contact with a cat, but *B. henselae* as causative agent was identified in 1985 [2, 4, 7]. The illnes is classified in the group of zoonosis, since it is transmitted from cats or kittens, what is more frequent, during scratches, bites or licking[3]. CSD is registred all over the world, although there is no exact data about the prevalence, as is the case in our area. Seropositivity against *B. henselae* ranges from 3.1% to 61.6% in general population in some parts of the world, which shows that a small number of infected persons become sick [8, 9]. It is registred about 22,000 patients with CSD per year in the USA and the incidence is 9.3 patients per 100,000 inhabitants [1, 6, 9, 10]. The disease appears more frequently during automn and winter in temperate climate zones, most frequently in children and adolescents, so younger than 21 make 80% of all cases of CSD. Males suffer more than females [9].

Correspondence to: Miroslav Kojić, Clinic for Infectious and Tropical Diseases, Military Medical Academy, Crnotravska 17, 11 000 Belgrade, Serbia. Phone: +381 64 21 29 655. E-mail: miroslav_kojic@hotmail.com

Incubation period is 1–2 weeks in 90% of patients (3–12 days). It is followed by papulopustular lesion on the bitten or scratched place (primary lesion), which lasts about 1–3 weeks. After that, in 90% of patients, characteristic regional lymphadenopathy is developed and is followed by mild fever, anorexy, nausea, fatigue or headache [1, 2, 7, 11]. Truncal maculopapular rash apears rarely [12]. Lymph nodes are painful and suppurate in 25%–30% of cases. CSD is a self-limited disease with exellent prognosis even in a severe form of the disease. Recovery is spontaneous in 2–5 months, but immunocompromised persons can develop severe and potentialy life-threatening forms of the disease [13]. Atypical forms of the disease, without papula at the site of inoculation and visible regional lymphadenopathy are present in 10% of cases with CSD. They include: encephalitis with seizures, transverse myelitis, arthritis, neuroretinitis, granulomatosus conjunctivitis, aseptic meningitis, hepatitis, osteomyelitis, endocarditis, myocarditis, pneumonia, splenic abscess, hemolytic anemia, trombocytopenic purpura etc [2, 14–27]. These atypical forms of CSD could be misdiagnosed as other infectious process or neoplasma [7].

The aim of this study was to show rare form of CSD with prolonged fever, purulent regional lymphadenitis, maculopapular rash and hepatitis in a young immunocompetent female person.

Case report

A 29-year-old female person was bitten by a kitten the third finger of her left hand on 12 June 2010. Three weeks later papulopustular lesion appeared on the bitten place. After about 6 weeks, her axillar lymph nodes became swollen and painful on palpation and a tumor formation appeared in the region of the left elbow on 24 July 2010. By the end of July, the patient became febrile, about 39°C, followed by extensive night sweets. In that period her lymph nodes were grouped into packages, followed by skin redness behind them and extreme palpatory tendreness. Laboratory analyses from that period are shown in Table 1. As causative agents were excluded hepatitis A, B and C viruses, Epstain-Barr virus and *Cytomegalovirus*. The therapy with ciprofloxacin 1,000 mg per day was initiated in the regional hospital with suspition on CSD, but maculopapular rash on truncus, limbs and face appeared after 10 days. Ciprofloxacin was changed with doxycycline in a daily dose of 200 mg and antialergic therapy was initiated by a dermatologist. Since the treatment did not lead to a significant improvement, the patient was admitted to the Clinic for Infectious and Tropical Disease, Military Medical Academy, Belgrade, on 18 August 2010.

At admission, the patient was subfebrile, pale, in good general condition, with the present crust on the bitten place and with rash in regression at the medial side of both forearms, which absolutely dissapeared after 7 days. In the region of the left axilla, a lymph node package was registred (Figure 1). It was painful on palpation and without signs of supuration. In the region of the medial side of the left elbow, a tumor formation of firmer consistency about 2 cm in diameter was noticed. Physical examinations of pharynx, lung and heart were normal; there was no hepatosplenomegaly.

Pathological laboratory analyses at admission are shown in Table 1. Using serological analyses, as causative agents were excluded Human Immunodeficiency Virus (HIV), *Toxoplasma gondii*, *Francisella tularensis, Echinococcus granulosum* and *Toxocara canis.*

Table 1

Laboratory analyses in the patient with cat scratch disease (CSD)

Laboratory analyses	Before admission	On admission	After three months
ESR (mm/1h)	106	139	16
CRP (mg/L)	47.3	30.4	3.4
Fibrinogen (g/L)	/	6.4	/
Procalcitonin (ng/L)	/	0.05	/
AST (U/L)	162	101	104
ALT (U/L)	507	178	304
Gamma GT (U/L)	/	159	/

ESR – Erythrocyte Sedimentation Rate; CRP – C reactive protein; AST – aspatrate aminotransferase; ALT – alanine aminotransferase; gamma GT– gama glutamyl transpeptidase

Five distinct oval hypoechogenic formations which were partialy in block, localized deeply in the muscle medialy beside the chest wall were registrated by ultrasound. They seemed to be enlarged and altered lymph nodes. A heterogeneous formation 3 cm in diameter was noticed behind the left elbow, medialy in the muscle, which seemed to be "a parasitic change". Ultrasound examinations of the neck and abdomen were normal.

After admission, the therapy with doxycyclin was being conducted for 7 days when it was stopped. A biopsy of the change in the left elbow was performed (Figure 1). The cap-

Fig. 1 – Regional lymphadenitis in the patient with cat scratch disease

sule of the tumor was opened, pus drainaged, which culture was sterile. Histopathological analyses of the tumor showed star-shaped granulomas with caseous necrosis and palisade deployed hystiocytes which correspond to CSD. No microorganisams were isolated in the tissue specimen using special paintings, including silver painting by the Warthin Starry method. The antimicrobial therapy was reintroduced on September 3 with doxycycline in a daily dose of 200 mg, for 20 days and was continued with ciprofloxacin 500 mg per day, for 3 weeks.

In repeated ultrasound examination, 5 weeks after the first one, a lobular heterogeneous liquid collection was registred, 4 cm in diameter, without capsula. Beside it a reactive lymph node was noticed 12 mm large (Figure 2). There were no pathologic findings in the subcutaneous tissue and muscle of the left axilla. A spontaneous drainage of abscess collection happened at that time and the patient became afebrile, with elevated erythrocyte sedimentation rate (ESR), C reactive protein (CRP), fibrinogen and serum transaminases (Figure 3). On October 4 2010, surgical procedure, incision and drainage of abscess collection, was done and a necrotic lymph node in the left axilla was eliminated. Purulent content was obtained, with no growth on pathogenic bacteria or *Mycobacterium tuberculosis.*

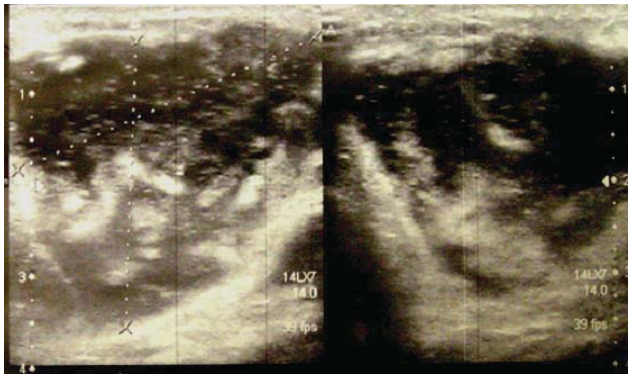


Fig. 2 – Ultrasound of axillar lymph nodes in the patient with cat scratch disease

Fig. 3 – Purulent lymphadenitis after spontaneous drainage in the patient with cat scratch disease

The antimicrobial therapy was definitely stopped on October 18 2010 when the patient was without symptoms and with absolutely normal physical examination. Laboratory findings performed at the end of October are shown in Table 1. Elevated serum transaminases and other normal biochemical findings were noticed in the middle of December, while total laboratory findings were normal only at the end of January 2011.

Discussion

CSD is an illness of children and youths under the age of 20 and is manifested as benign and self-limited lymphadenopathy caused by bacteria *B. henselae* [9, 12]. According to Erik [2] the disease appears in patients younger than 21 in 80% of cases. The reason for this could be in the fact that children are in contact with cats and kittens more freqently, so it is more probable to be scratched or bitten. The incidence of disease is not known in our country and only sporadic cases are reported.

After the incubation period of 3–12 days, on the bitten or scratched place, papulopustular lesions appear in about 90% of patients which lasts about 1–3 weeks [2]. In the presented patient the lesions were present but they appeared a little bit latter, 3 weeks after the bite. The most impressive clinical sign of infection is regional lymphadenopathy and it is present in 80% of patients. In about 10% of inflammed lymph nodes, the skin behind them becomes red. After that they fluctuate, what is the sign of suppuration and it is followed by spontaneous drainage [10]. Erik [2] cites in his research that lyphadenopathy is manifested primary in axillar lymph nodes and that they suppurate in about 25%–30% of cases. According to the same author, in 50% of cases only one lymph node is changed, in 30% more lymph nodes from different part of the body are infected and in 20% of cases a few lymph nodes from the same region are infected. In the presented patient one cubital and a few axillar lymph nodes were changed.

Mild fever is present in 30%–60% of patients and usually lasts about 1–2 weeks [6]. The presented patient was febrile about 20 days with some epizodes of high fever. In the literature are described cases of systemic illness in immunocompetent persons which arise hematogeneously. These forms are characterized with long-lasting fever, hepatosplenomegaly, granulomatous hepatitis, abdominal pain, weight loss, headache, weakness and malaise [28]. The presented patient had prolonged fever and signs of hepatitis with values of serum transaminases which were multiple as high, but granulomatous lesions in liver were not detected by ultrasound, and other diagnostic procedures (MSCT and liver biopsy) were not performed.

Dermal manifestations of CSD are quite infrequent and appear in about 5% of patients. Eryhema nodosum, erythema multiforme, erythema marginatum and non-specific maculopapular, morbiliform or petechial rash have been described till now [29]. Dzelalija et al. [12] described a similar case to the case we presented, with maculopapular rash, purulent lymphadenitis, slightly elevated serum transaminases and parameters of inflammation. The presented patient had macu-

lopapular rash considered allergic manifestation, but the same was exluded later, with the reintroduction of ciprofloxacin in the therapy, in hospital settings.

Diagnostic criteria for CSD are characteristic clinical picture, positive epidemiological data, exclusion of other causes of lymphadenitis, morphological and histopathological examination of the biopted lymph nodes, serological confirmation by detecting specific serum antibodies using immunofluorescency methods and detection of *B. henselae* genom using PCR method [3, 30–33].

Morphological examinations (US, CT, NMR) are of great importance for diagnosing CSD. Lymph nodes are visualised as round or ovoid masses in diameter of about 1–5 cm by ultrasound examination [34]. In about ⅔ of patients it is affected only one or more lymph nodes from the same region (hand, neck or axilla). Affecting more different regions is a sign of multiple inoculations or dissemination of the disease. Disseminated form of the disease can be registred by finding granulomas in the liver and the spleen by ultrasound [3]. The presented patient showed some signs for disseminated form of the disease, but on ultrasound no granuloma was detected.

The causative agent is difficult to be isolated from the human tissue, but isolation and identification of the agent is important in the detection of the disease in animals [3]. Cultivation of the microorganism from tissue specimens requires special cicumstances and is possible in well-equiped laboratories [35]. In modest labs, using special paintings (Gram, hematoxilin-eosin, Ziehl-Neelsen, Warthin Starry etc), characteristic histopathological findings can indicate CSD by the characteristic shape of granuloma what was used for the diagnosis in the presented patient. According to data from the literature, the presence of *B. henselae* in lymph node specimens is more freqent in patients with supurative lymphadenitis (67%) comparing to patients with non-supurative lymphadenitis (22%) [10, 33]. In our patient, although it was supurative lymphadenitis a causative agent is not proven using silver painting by the Warthin Starry method, probably because the patient had already started antimicrobial therapy. The most applied serologial method for detection of serum antibodies againest *B. henselae* is indirect immunofuorescency (IIF) [32]. Sensitivity of the method is 88% and specificity 97%, although they vary between labs, so sensitivity of the method ranges from less than 30% to 100% [10, 35]. In our country, unfortunately, no reference laboratory performs serological diagnosis of CSD.

The course of the disease in immunocompetent persons is favourable, yet complications appear in 5%–13% of patients as purulent lymphadenitis, maculopapular rash, bilateral recurent iridocyclitis, endocarditis, pericarditis, and/ or myocarditis [11, 12, 16, 17, 29, 25–27]. The presented patient had a prolonged-course fever that lasted about a month, with reverse damage of the liver which lasted almost 6 months.

B. henselae is sensitive on macrolids, fluoroquinolones, tetracyclines, rifampicin and sulfametoxazol-trimethoprim [3]. However, antimicrobial therapy very often has no efect on the course of the disease. There is no consensus about antimicrobial therapy of CSD in immunocompetent person, nor on duration of therapy, and the need for therapy. The presented patient was treated with ciprofloxacin and tetracyclines for 9 weeks, but according to the clinical and laboratory monitoring we could not conclude that antimicrobial therapy had good effect on the course of the disease.

Conclusion

Atypical forms of CSD could be an important differential-diagnostic problem, especially if there is no opportunity for serologial confirmation of disease.

R E F E R E N C E S

1. *Carithers HA*. Cat-scratch disease. An overview based on a study of 1,200 patients. Am J Dis Child 1985; 139(11): 1124−33.
2. *Erik S*. Catscratch Disease. eMedicine. Available from: http://emedicine.medscape.com/article/781320-overview [cited 2009 September 24].
3. *Chomel BB, Boulouis HJ, Breitschwerdt EB*. Cat scratch disease and other zoonotic Bartonella infections. J Am Vet Med Assoc 2004; 224(8): 1270−9.
4. *Gerber MA, Sedgwick AK, MacAlister TJ, Gustafson KB, Ballow M, Tilton RC*. The aetiological agent of cat scratch disease. Lancet 1985; 1(8440): 1236−9.
5. *Little S*. Cat Scratch Disease. In: *Little S,* editor. CFA Health Committee [online]. Available from: www.cfainc.org. [accessed 2002 November 10].
6. *Friedman AD*. Catscracth Disease.eMedicine. Available from: http://emedicine.medscape.com/article/962445-overview [accesed 2009 May 28].
7. *Lamps LW, Scott MA*. Cat-scratch disease: historic, clinical, and pathologic perspectives. Am J Clin Pathol 2004; 121 Suppl: S71−80.
8. *Sander A, Posselt M, Oberle K, Bredt W*. Seroprevalence of antibodies to Bartonella henselae in patients with cat scratch disease and in healthy controls: evaluation and comparison of two commercial serological tests. Clin Diagn Lab Immunol 1998; 5(4): 486−90.
9. *Zangwill KM, Hamilton DH, Perkins BA, Regnery RL, Plikaytis BD, Hadler JL,* et al. Cat scratch disease in Connecticut. Epidemiology, risk factors, and evaluation of a new diagnostic test. N Engl J Med 1993; 329(1): 8−13.
10. *Lina LC, Rosalind S, Chong AV, Toha A, Shaffie B*. Cat Scratch Disease: A Diagnostic Dilemma Med J Malaysta 2010; 65(2): 155−6
11. *Rombaux P, M'Bilo T, Badr-el-Din A, Theate I, Bigaignon G, Hamoir M*. Cervical lymphadenitis and cat scratch disease (CSD): an overlooked disease? Acta Otorhinolaryngol Belg 2000; 54(4): 491−6.
12. *Dzelalija B, Medić A, Rode OD, Mazzi A*. Rash and purulent lymphadenitis in cat scratch disease. Acta Med Croatica 2006; 60(5): 483−6. (Croatian)
13. *Birtles RJ, Harrison TG, Taylor AG*. Cat scratch disease and bacillary angiomatosis: aetiological agents and the link with AIDS. Commun Dis Rep CDR Rev 1993; 3(8): R107−10.
14. *Smith RA, Scott B, Beverley DW, Lyon F, Taylor R*. Encephalopathy with retinitis due to cat-scratch disease. Dev Med Child Neurol 2007; 49(12): 931−4.
15. *Durá-Travé T, Yoldi-Petri ME, Gallinas-Victoriano F, Lavilla-Oiz A, Bove-Guri M*. Neuroretinitis Caused by Bartonella

henselae (Cat-Scratch Disease) in a 13-Year-Old Girl. Int J Pediatr 2010; 2010: 763105.
16. *Skorin L Jr.* Perinaud`s oculoglandular syndrome An atypical manifestation of cat-scratch disease. Available from: http://www.optometry.co.uk/files/9429c5d753381c2ed7f7936e73f2ea0c [published 2005 March 11]
17. *Martínez-Osorio H, Calonge M, Torres J, González F.* Cat-scratch disease (ocular bartonellosis) presenting as bilateral recurrent iridocyclitis. Clin Infect Dis 2005; 40(5): e43−5.
18. *Pinto Jr VL, Curi AL, Pinto Ada S, Nunes EP, Teixeira Mde L, Rozental T,* et al. Cat scratch disease complicated with aseptic meningitis and neuroretinitis. Braz J Infect Dis 2008; 12(2): 158−60.
19. *Tasher D, Armarnik E, Mizrahi A, Liat BS, Constantini S, Grisaru-Soen G.* Cat scratch disease with cervical vertebral osteomyelitis and spinal epidural abscess. Pediatr Infect Dis J 2009; 28(9): 848−50.
20. *Vermeulen MJ, Rutten GJ, Verhagen I, Peeters MF, van Dijken PJ.* Transient paresis associated with cat-scratch disease: case report and literature review of vertebral osteomyelitis caused by Bartonella henselae. Pediatr Infect Dis J 2006; 25(12): 1177−81.
21. *Robson JM, Harte GJ, Osborne DR, McCormack JG.* Cat-scratch disease with paravertebral mass and osteomyelitis. Clin Infect Dis 1999; 28(2): 274−8.
22. *Kodama Y, Maeno N, Nishi J, Imuta N, Oda H, Tanaka S,* et al. Multifocal osteomyelitis due to Bartonella henselae in a child without focal pain. J Infect Chemother 2007; 13(5): 350−2.
23. *Roubaud-Baudron C, Fortineau N, Goujard C, Le Bras P, Lambotte O.* Cat scratch disease with bone involvement: a case report and literature review. Rev Med Interne 2009; 30(7): 602−8. (French)
24. *de Kort JG, Robben SG, Schrander JJ, van Rhijn LW.* Multifocal osteomyelitis in a child: a rare manifestation of cat scratch disease: a case report and systematic review of the literature. J Pediatr Orthop B 2006;15(4): 285−8.
25. *Mariani M, Pagani M, Inserra C, De Servi S.* Complete atrioventricular block associated with toxoplasma myocarditis. Europace 2006; 8(3): 221−3.
26. *Meininger GR, Nadasdy T, Hruban RH, Bollinger RC, Baughman KL, Hare JM.* Chronic active myocarditis following acute Bartonella henselae infection (cat scratch disease). Am J Surg Pathol 2001; 25(9): 1211−4.
27. *Pipili C, Katsogridakis K, Cholongitas E.* Myocarditis due to Bartonella henselae. South Med J 2008; 101(11): 1186.
28. *Liao H, Huang F, Wang N, Chen B.* Systemic Cat Scratch Disease. J Formos Med Assoc 2006; 105(8): 674−9.
29. *Daye S, McHenry JA, Roscelli JD.* Pruritic rash associated with cat scratch disease. Pediatrics 1988; 81(4): 559−61.
30. *Hansmann Y, DeMartino S, Piémont Y, Meyer N, Mariet P, Heller R,* et al. Diagnosis of cat scratch disease with detection of Bartonella henselae by PCR: a study of patients with lymph node enlargement. J Clin Microbiol 2005; 43(8): 3800−6.
31. *Eroğlu C, Candir N, Dervişoğlu A, Kefeli M.* A case of cat scratch disease. Mikrobiyol Bul 2007; 41(4): 603−6. (Turkish)
32. *Flexman JP, Chen SC, Dickeson DJ, Pearman JW, Gilbert GL.* Detection of antibodies to Bartonella henselae in clinically diagnosed cat scratch disease. Med J Aust 1997; 166(10): 532−5.
33. *Suzumija J, Ohshima K, Takada T, Kanda M, Tamura K, Kikuchi M.* Prevalance of Bartonella henselae in granulomatous lymphadenitis : A useful tool for the diagnosis of the cat scratch disease by polymerase chain reaction. Clin Exp Hematop 2001; 41(2): 101−6.
34. *Hopkins KL, Simoneaux SF, Patrick LE, Wyly JB, Dalton MJ, Snitzer JA.* Imaging manifestations of cat-scratch disease. AJR Am J Roentgenol 1996; 166(2): 435−8.
35. *Rolain JM, Lepidi H, Zanaret M, Triglia JM, Michel G, Thomas PA,* et al. Lymph node biopsy specimens and diagnosis of cat-scratch disease. Emerg Infect Dis 2006; 12(9): 1338−44.

Received on May 17, 2011.
Revised on August 3, 2011.
Accepted on August 18, 2011.

CASE REPORT

UDC: 616.89-008.441:613.25]:616.34
DOI: 10.2298/VSP1301077M

Celiac disease diagnosed after uncomplicated pregnancy in a patient with history of bulimia nervosa

Celijačna bolest dijagnostikovana posle nekomplikovane trudnoće kod bolesnice sa anamnezom bulimije nervoze

Nemanja Milisavljević, Mirjana Cvetković, Goran Nikolić, Branka Filipović, Nikola Milinić

Department of Gastroenterohepatology, Clinical and Hospital Center "Bežanijska kosa", Belgrade, Serbia

Abstract

Introduction. The association between celiac disease and eating disorders has been very rarely reported. This is the first report on celiac disease associated with bulimia in this part of Europe. **Case report.** An adult female patient with history of bulimia and one uncomplicated pregnancy was admitted to the Gastroenterology Department, due to long lasting dyspeptic symptoms, constipation, major weight loss and fatigue. After positive serological screening, the diagnosis of celiac disease was confirmed with histopathology examination of duodenal biopsy specimen. **Conclusion.** Complicated interactions between celiac disease and bulimia can make them difficult to diagnose and treat. It is important to consider the presence of celiac disease in patients with bulimia and gastrointestinal symptoms.

Key words:
celiac disease; eating disorders; pregnancy; diagnosis.

Apstrakt

Uvod. Udruženost celijačne bolesti i poremećaja ishrane je retko opisivana u literaturi. Ovo je prvi prikaz bulimije udružene sa celijačnom bolešću u ovom delu Evrope. **Prikaz bolesnika.** Odrasla bolesnica sa bulimijom i jednom nekomplikovanom trudnoćom primljena je u Odeljenje za gastroenterologiju zbog tegoba kao što su dugotrajna dispepsija, konstipacija, gubitak telesne mase i malaksalost. Nakon pozitivnog serološkog skrininga, dijagnoza celijačne bolesti potvrđena je histopatološkim pregledom bioptata mukoze duodenuma. **Zaključak.** Komplikovani odnosi između celijakije i bulimije mogu ih učiniti teškim za dijagnostikovanje i lečenje. Važno je razmotriti postojanje celijačne bolesti kod bolesnika sa bulimijom i gastrointestinalnim simptomima.

Ključne reči:
celijakija; apetit, poremećaji; tudnoća; dijagnoza.

Introduction

Celiac disease is an inflammatory condition of the small intestinal mucosa induced by gluten consumption in genetically susceptible individuals, leading to the spectrum of gastrointestinal manifestation. Previously, celiac disease was thought to be a disease of infancy, manifesting during the first years of life as the malabsorption syndrome with chronic diarrhea, abdominal distension, and failure to thrive. In the recent years it became increasingly evident that celiac disease can affect individuals of any age. Because of the variety of ways celiac disease can manifest itself in adults, it is often not still diagnosed. Psychiatric symptoms and disorders are commonly found in association with celiac disease [1, 2]. Most reports concern the association between celiac disease and anxiety, irritability and depression [3]. The association between celiac disease and eating disorders has been rarely reported in the literature [4]. Most of the case reports describe onset of anorexia nervosa in patients with already diagnosed celiac disease after the introduction of gluten free diet [5].

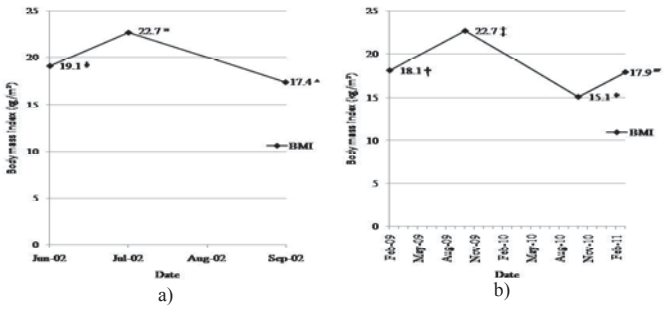
We presented an adult patient with celiac disease and history of bulimia nervosa, in who celiac disease had been diagnosed after pregnancy.

Case report

A 27-year-old woman with a history of bulimia and one uncomplicated pregnancy was admitted to the Gastroenterology Department in 2010 due to long lasting dyspeptic symptoms, constipation, weight loss and fatigue. During

Correspondence to: Nemanja Milisavljević, Department of Gastroenterohepatology, Clinical and Hospital Center Bežanijska kosa, 11 080, Belgrade, Serbia. E-mail: nemanja_vesa@yahoo.com

childhood the patient suffered from mild underweight, nocturnal enuresis, and occult spina bifida. The patient had a late onset of menarche at the age of 15, followed by irregular, painful periods. Also, the patient had never been treated for infertility. Family history was negative for celiac disease or eating disorders. The patient's eating difficulties started in 2002, when she started binge eating and gained 10 kg in body weight (Figure 1 a). Then the patient started to use diet products, trained excessively for a few months and vomited and binge eating 3 to 4 times a week for the next 7 years in order to lose weight (Figure 1 a), fulfilling all the criteria of the Diagnostic and Statistical Manual (DSM-IV) for bulimia. Also, in 2002 the patient complained on dyspeptic symptoms and received *Helicobacter pylori* eradication therapy. As her dyspeptic symptoms had persisted she underwent upper gastrointestinal endoscopy which was normal findings. Celiac disease was not suspected as her duodenal mucosa had normal macroscopic appearance. Because of a long-lasting constipation in 2006 underwent colonoscopy and small bowel barium follow through test, which were both normal. In 2009, the patient got pregnant for the first time in her life and in the first trimester of pregnancy stopped her bulimic behavior. Routine pregnancy laboratory tests were within normal range. Pregnancy and childbirth were uncomplicated. During the next 12 months, prior to admission to hospital, the patient lost nearly 21 kg in weight (Figure 1 b) and experienced severe dyspeptic symptoms. Both weight loss and dyspeptic symptoms had begun soon after childbirth. The patient had not bulimic symptoms after pregnancy. Both psychiatrist and psychologist suspected a hidden bulimia relapse. Family history was negative for celiac disease or eating disorder.



a) b)

Fig. 1 – Body mass index (BMI) variations: a) before onset of bulimia$^{\Phi}$, during binge phase $^{\Omega}$, during purge phase^; b) at the beginning† and at the end of pregnancy‡, at hospital admission*, and six months after introduction of gluten free diet$^{///}$

Physical examination at admission revealed cachexia.

Routine laboratory tests were within normal range. According to the standardized rating scale for eating disorders – the Bulimic Investigatory Test Edinburgh (BITE) [6], the patient was in remission from bulimia. Bioimpedance analysis revealed a low level of body mass index (BMI) (15.1 kg/m^2) and total body fat (TBF) (8.6%). The screening serology test for celiac disease – IgA anti-transglutaminase antibodies was performed which were highly elevated – 425.61 RU/mL (< 20 RU/mL). Upper gastrointestinal endoscopy revealed atrophic macroscopic appearance in duodenum (Figure 2). Histopathology examination of duodenal biopsy specimen revealed type IIIb lesions according to the Marsh-Oberhuber classification. Osteodensitometry displayed a normal bone mineral density.

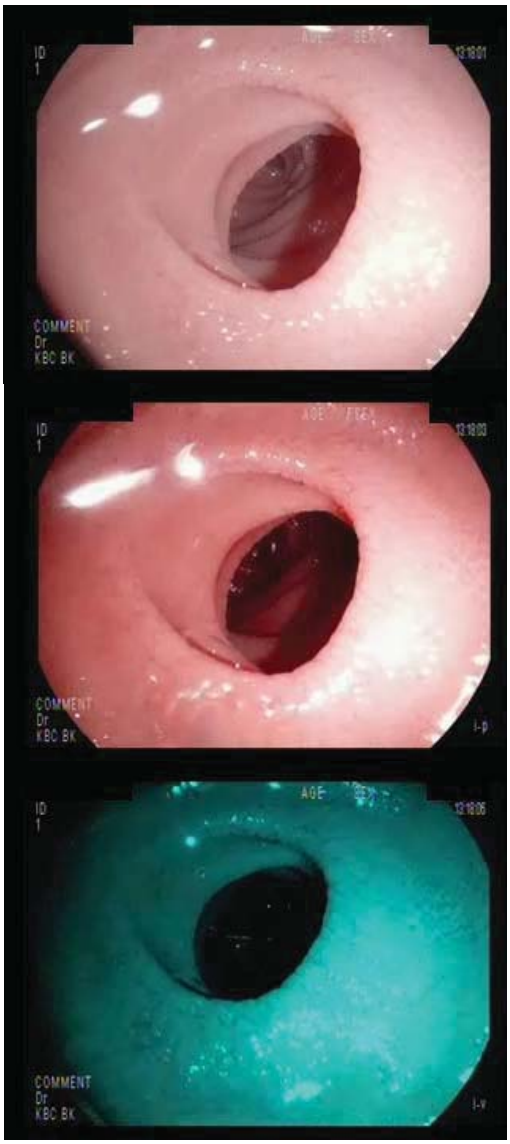


Fig. 2 – Endoscopic appearance of atrophic duodenal mucosa (with and without i-scan surface enhancement)

After diagnosing celiac disease, the patient was recommended to undergo strict life-long gluten free diet, and underwent cognitive behavioural psychotherapy performed by the psychologist for 20 weeks in order to prevent possible bulimia relapse. She was recommended to use paroxetine in combination with psychotherapy.

During 6-month follow-up the BMI of the patient improved (Figure 1 b) and IgA anti-transglutaminase antibodies were only slightly elevated (32.4 RU/mL). According to BITE, the patient had not relapsed into bulimia during the follow-up period.

Discussion

This paper presented an adult patient with celiac disease and previous history of bulimia nervosa and one uncomplicated pregnancy. To the best of our knowledge, this is the first case of celiac disease associated with an eating disorder in this part of Europe. According to the available sources, only 2 similar case reports, [1, 2] and one case series [4] have been reported so far. There is a strong clinical evidence for suspecting celiac disease early in our patient.

Celiac disease is an inflammatory condition of the small intestinal mucosa that is induced by ingestion of gluten in genetically susceptible individuals, leading to spectrum of gastrointestinal manifestations. It is one of the most common genetically conferred disorders reaching the prevalence of 1% in overall population. Many patients remained undiagnosed, either because of misdiagnosed, asymptomatic, latent, silent or potential celiac disease. It is known that a high proportion of adults with celiac disease has a prior history of psychiatric disorder even for years [3, 7]. Also, it is known that undiagnosed celiac disease can lead to infertility in women. Bulimia nervosa is characterized by episodes of binge eating (uncontrolled consumption of a large amount of food in a relatively short period of time) followed by an inappropriate "compensation" such as forced vomiting, laxative or diuretic abuse, a subsequent fast or period of food restriction, or excessive exercising [8].

The mechanisms involved in the pathogenesis of psychiatric disorders related to celiac disease are not well understood. Malabsorption of tryptophan, folic acid, and vitamin B6, known to occur in celiac disease, may lead to disturbances in brain serotonin function [9]. Decreased serotonin activity in the brain is associated with enhanced appetite, and thus may be related to bulimia [10]. Adult celiac patients taking a gluten-free diet showed an increase in the concentrations of serotonin metabolites in cerebrospinal fluid [11]. On the other hand, treatment-induced weight gain can act as a risk factor for altered eating behavior [12]. Studies evaluating the course of bulimia during pregnancy have reported an improvement in bulimic symptoms and a return to prepregnancy symptom levels or even a worsening of symptoms in the postpartum period [13, 14]. A reason for improvement of bulimic symptoms during pregnancy may be found in the significant increase of serotonin concentration in the brain through pregnancy [15]. Pregnancy and delivery can trigger celiac disease in susceptible people for reasons that are not well-understood.

A long history of nonspecific gastrointestinal symptoms and low body weight, described in our patient, were neglected by physicians in the presence of bulimia and uncomplicated pregnancy. Furthermore, worsening of symptoms after pregnancy was mistakenly attributed to a hidden bulimia relapse.

Conclusion

Complicated interaction between celiac disease and bulimia can make the patient difficult both to diagnose and treat. It is important to consider the presence of celiac disease in patients with bulimia and gastrointestinal symptoms. Serological testing for celiac disease in patients with eating disorders may be useful. Further studies are needed to determine the true nature of the relationship between celiac disease and eating disorders, particularly bulimia.

REFERENCES

1. *Yucel B, Ozbey N, Demir K, Polat A, Yager J.* Eating disorders and celiac disease: a case report. Int J Eat Disord 2006; 39(6):530-2.
2. *Jost PJ, Stengel SM, Huber W, Sarbia M, Peschel C, Duyster J.* Very severe iron-deficiency anemia in a patient with celiac disease and bulimia nervosa: a case report. Int J Hematol 2005; 82(4): 310−1.
3. *Addolorato G, Leggio L, D'Angelo C, Mirijello A, Ferrulli A, Cardone S,* et al. Affective and psychiatric disorders in celiac disease. Dig Dis 2008; 26(2): 140−8.
4. *Leffler DA, Dennis M, Edwards George JB, Kelly CP.* The interaction between eating disorders and celiac disease: an exploration of 10 cases. Eur J Gastroenterol Hepatol 2007; 19(3): 251−5.
5. *Ricca V, Mannucci E, Calabro A, Bernardo MD, Cabras PL, Rotella CM.* Anorexia nervosa and celiac disease: two case reports. Int J Eat Disord 2000; 27(1):119–22
6. *Henderson M, Freeman C*P. A self-rating scale for bulimia. The 'BITE'. Br J Psychiatry 1987; 150: 18−24.
7. *Pynnönen PA, Isometsä ET, Aronen ET, Verkasalo MA, Savilahti E, Aalberg VA.* Mental disorders in adolescents with celiac disease. Psychosomatics 2004; 45(4): 325−35.
8. American Psychiatric Association. Diagnostic and Statistical Manual of Mental Disorders. DSM-IV. 4th ed. Washington, DC: Amazon; 1994.
9. *Pynnönen PA, Isometsä ET, Verkasalo MA, Savilahti E, Aalberg VA.* Untreated celiac disease and development of mental disorders in children and adolescents. Psychosomatics 2002; 43(4): 331−4.
10. *Jimerson DC, Wolfe BE, Metzger ED, Finkelstein DM, Cooper TB, Levine JM.* Decreased serotonin function in bulimia nervosa. Arch Gen Psychiatry 1997; 54(6): 529−34.
11. *Hallert C, Sedvall G.* Improvement in central monoamine metabolism in adult coeliac patients starting a gluten-free diet. Psychol Med 1983; 13(2): 267−71.
12. *Karwautz A, Wagner G, Berger G, Sinnreich U, Grylli V, Huber WD.* Eating pathology in adolescents with celiac disease. Psychosomatics 2008; 49(5): 399−406.
13. *Crow SJ, Keel PK, Thuras P, Mitchell JE.* Bulimia symptoms and other risk behaviors during pregnancy in women with bulimia nervosa. Int J Eat Disord 2004; 36(2): 220−3.
14. *Morgan JF, Lacey JH, Sedgwick PM.* Impact of pregnancy on bulimia nervosa. Br J Psychiatry 1999; 174: 135−40.
15. *Uluitu M, Duşleag L, Constantinescu D, Petcu G, Catrinescu G, Pană S.* Serotonin through pregnancy. Comparative researches in different species and in mankind. Physiologie 1975; 12(4): 275−80.

Received on June 6, 2011.
Revised on July 28, 2011.
Accepted on Avgust 5, 2011.

CASE REPORT

UDC: 616.716.4-089.87-089.844:616.314-76/-77-089.843
DOI: 10.2298/VSP1301080K

Possibilities of reconstruction and implant-prosthetic rehabilitation following mandible resection

Mogućnosti rekonstrukcije i implantološko-protetičke rehabilitacije nakon resekcije mandibule

Vitomir S. Konstantinović*, Vladimir S. Todorović*, Vojkan M. Lazić†

***Clinic of Maxillofacial Surgery, †Clinic of Dental Prosthetics, Faculty of Dental Medicine, University of Belgrade, Belgrade, Serbia**

Abstract

Introduction. Mandible reconstruction is still very challenging for surgeons. Mandible defects could be the consequence of ablative surgery for malignancies, huge jaw cysts, infection and trauma. Segmental resection of the mandible may compromise orofacial function and often lead to patients psychological disorders. Despite very frequent use of microvascular flaps, autogenous bone grafts are still very reliable technique for mandible reconstruction. Comprehensive therapy means not only mandible reconstruction, but prosthodontic rehabilitation supported by dental implants, which can significantly improve patients quality of life. The aim of this paper was to evaluate possible techniques of mandible reconstruction and to present a patient who had been submitted to mandible resection and reconstruction with autogenous iliac bone graft and prosthodontic rehabilitation with fixed denture anchoraged by disc-shaped implants in early loading protocol. **Case report.** Mandible reconstruction was performed simultaneously with resection. Autogenous iliac bone graft was taken, reshaped and placed in two parts, to the required optimal contour of the mandible. After graft consolidation, decision was made for prosthodontics rehabilitation with fixed dentures supported by implants. In addition to the standard preoperative procedures, planning was done based on a biomodel gained by rapid prototyping after CT scan. It offered a real 3D planning to obtain a proper shape, dimension and the position of implants. **Conclusion.** If bone dimensions of a reconstructed mandible are insufficient, like in the presented case, the use of basal osseointegrated implants may be a method of choice. Avoiding bone augmentation procedures, as well as early loading protocol for this type of implants, shorten the total rehabilitation time, which is very convenient for patients. Fixed denture supported by dental implants is the best solution for comprehensive rehabilitation after mandible resection.

Key words:
oral surgical procedures; mandibular injuries; reconstructive surgical procedures; dental prothesis, implant-supported; rehabilitation, treatment outcome

Apstrakt

Uvod. Rekonstrukcija mandibule još uvek predstavlja izazov za hirurga. Defekti mandibule mogu biti posledica radikalnog hirurškog tretmana malignih tumora, velikih viličnih cista, infekcija i povreda. Nedostatak dela vilične kosti kompromituje sve orofacijalne funkcije, a česti su i psihološki poremećaji. Pored sve učestalije primene mikrovaskularnih režnjeva, slobodni koštani graftovi su još uvek veoma pouzdan metod rekonstrukcije mandibule. Sveobuhvatna terapija pored rekonstrukcije mandibule podrazumeva i implantološko-protetičku rehabilitaciju, kojom se znatno poboljšava kvalitet života bolesnika. Cilj ovog rada bio je da se kroz pregled literature ocene metode rekonstrukcije mandibule i da se prikaže bolesnica kod koje je nakon segmentalne resekcije mandibule izvršena rehabilitacija fiksnom zubnom nadokanadom nošenom implantatima oblika diska. **Prikaz bolesnika.** U istom aktu sa resekcijom izvršena je i rekonstrukcija slobodnim koštanim ilijačnim graftom, koji je preoblikovan i postavljen iz dva dela, kako bi se uspostavio optimalan kontinuitet i forma mandibule. Po konsolidaciji grafta, postavljena je indikacija za izradu fiksne zubne nadoknade nošene implantatima. Pored standardnih preoperativnih procedura, izvršeno je realno trodimenzionalno planiranje na biomodelu dobijenim softverskom analizom CT podataka. Na taj način određen je oblik, veličina i najpovoljniji položaj implantata. **Zaključak.** Ukoliko su koštane dimenzije rekonstruisane mandibule nedovoljne, kao kod prikazane bolesnice, primena bazalnih oseointegrišućih implantata oblika diska može biti metoda izbora. Njihovom upotrebom izbegava se dodatna nadoknada kosti što uz rano opterećenje predstavlja pogodnost za bolesnike, jer se znatno skraćuje vreme rehabilitacije. Izrada fiksne zubne nadoknade nošene implantatima je najbolji način definitivne rehabilitacije bolesnika sa rekonstruisanom mandibulom.

Ključne reči:
hirurgija, oralna, procedure; mandibula, povrede; hirurgija, rekonstruktivna, procedure; zubna proteza, implantatom podržana; rehabilitacija; lečenje, ishod.

Correspondence to: Konstantinović S. Vitomir, Clinic of Maxillofacial Surgery. Dr Subotića 4, 11 000 Belgrade, Serbia.
Phone. +381 11 2685 342. E-mail: vskvita@sbb.rs

Introduction

Mandible reconstruction has been a challenge for surgeons for more than a century. Mandible defects resulting in face deformity of various stages are mostly the consequence for ablative surgery for malignancies, huge jaw cysts, infections (osteomyelitis) and trauma, that may compromise orofacial functions and cause subsequent psychological disorders.

Adequate anatomic reconstructing assumes the outcome which should provide satisfactory mandible dimensions, form and shape. Also, muscle attachments which enable normal functioning should be established again. It is necessary to consider a definitive prosthetic rehabilitation and to think about the space for the placement of oral implants. In spite of a significant progress achieved, particularly in the last 40 years, none of the existing reconstruction techniques is completely satisfactory [1].

According to the algorithm developed by Takushima et al. [2], mandible defects are classified as either „lateral" or „anterior". Soft-tissue defects are classified into three categories: "none" (no or minimal defect on both sides of facial skin and oral mucosa); "skin or mucosal" (only skin or mucosal defect); and "through-and-through" (defect is through-and-through from the oral mucosa to the facial skin). To select a suitable reconstruction method, bone-defect should be considered first, followed by the soft tissue condition. In accordance with this, autologous bone grafts, alloplastic materials and tissue engineered matrix origin cell grafts, are utilized for the mandible reconstruction [1]. Most frequently used are autologous bone grafts which can be applied in three principally different ways, such as: free bone grafts, pedicled bone grafts and microvascular bone grafts (flaps) [3].

The introduction of microvascular surgery has led to a significant progress in mandible defects treatment [1]. Microvascular bone grafts can be „osteomuscular" which, apart from the bone, contain the periosteum and the attached muscle, or „osteomusculocutaneous", which also contain the skin on their surface. These, so-called composite grafts can be taken from the different donor regions: fibula, iliac and scapula. They are indicated for the reconstruction of large bone defects, defects in recipient sites of poor quality (scarred tissue, irradiated tissue, etc.), and when a simultaneous bone and soft tissue reconstruction is preferred [3]. According to the literature, the most frequently applied is fibula flap [1, 4]. The basic advantage of microvascular composite flaps is the possibility of one-stage treatment of both bone and soft tissue defects by using a single donor site, with over 90% efficacy, even in irradiated patients [1, 5, 6]. Foster et al. [5] state that the success of implants osseointegration in microvascular bone flaps was recorded in 99% cases. Compared with free bone grafts, there is a less risk to develop postoperative complications such as resorption or infection. However, complications of various degrees can be developed in the donor region, such as pain, difficulties in walking (limping), pathological fractures, herniation, etc. [7]. On the other hand, duration of surgery may impose a serious problem with patients with a compromised general condition [1]. It should be also mentioned that such procedures require qualified staff and well-equipped institutions, yet not always possible to provide.

Initial efforts to use free bone grafts date back to the beginning of the 19th century. Owing to the experience gained during the First and Second World War, this technique became widely accepted as standard in treatment of mandible defects. Until 1970s, fixation of these grafts was done with a wire, taking a longer period of intermaxillary fixation with the level of success ranging from 20% to 90%. It is known that the success of free bone grafts depends on the fixation and revascularization of the recipient site. Revascularization is very important for the process of resorption and deposition of a new bone, which is referred to as creeping substitution [1]. Also, it is known that even micromovements, if fixation is not enough strong, could jeopardize the viability of a graft or lead to graft failure. There is surprisingly little literature about the success rate of free bone grafts fixed with plates and screws as compared to wire fixation. However, fixation is nowadays routinely done with reconstructive plates and screws. The usual donor sites are: the iliac crest, rib or tibia. With regard to the bone quantity and quality, the best characteristics are provided by the iliac crest. The anterior iliac crest is the donor region of choice in most of cases. Iliac grafts could be taken as „cancellous", „thin cortical", „corticocancellous", and „bicorticocancellous" (full thickness) bone grafts [3]. The technique of raising free iliac bone grafts is simple one, their shape matches mandible contours and dimensions and they provide enough amount of bone that is very significant for implant placement. Generally, patients experience a postoperative course without difficulties, and donor site complications are rare (12%).

A definitive functional reconstruction implies prosthetic rehabilitation, which may be done with mobile restorations which are retained by means of the existing teeth. However, apart from the limited function and discomfort of patient, mobile prosthesis also causes an additional bone resorption. An ideal reconstruction is achieved with fixed dentures anchoraged by dental implants. If bone dimensions are not sufficient, bone augmentation or adequate implantation systems can be used.

Disk-shaped dental implants placed in jaw bones by lateral approach were described even in 1972. Significantly improved in the sense of their design and surface, they have been recently applied as the so-called basal osseointegrated implants [8]. Owing to their design which enables bicortical osseointegration in the basal, the most resoption-resistant part of the jaw bone, they can be also placed even when vertical and horizontal dimensions of the residual alveolar ridge are insufficient, that is a huge advantage over other implantation systems. Moreover, the possibility of early loading, conditioned by the achievement of a balanced occlusion, provides a patient with great comfort.

The aim of this paper was to evaluate possible techniques of mandible reconstruction and the contemporary approach to comprehensive functional rehabilitation of patient after a segmental mandible resection.

A female patient with mandible resection followed by immediate reconstruction with an autogenous iliac bone graft was presented. After a complete graft integration, in the second phase, the basal osseointegrated implants were inserted and early loaded with the fixed denture.

Case report

A 55-years-old female patient was admitted to the Clinic of Maxillofacial Surgery, Faculty of Dental Medicine, University of Belgrade, with pathological lesion in mandible which was identified during the routine radiographic examination. The patient was not complaining to any discomfort which might indicate the presence of lesion. A radiological finding was unspecific, showing confined multiloccular bone radiolucency of the corpus, angulus and ramus of the mandible at the right side (Figure 1). After biopsy and histopathological finding the diagnosis of odontogenic keratocyst was made. After preoperative planning, a segmental mandible resection from the canine to the subcondylar region including a coronoid process was performed. A total length of the resected part was about 8 cm. Simultaneously, a primary reconstruction with a free bone graft from the iliac bone was done. Due to the size of defect, the graft was reshaped and placed in two parts in order to achieve the most optimal continuity and mandible form. Titanium reconstructive plate and screws (Synthes GmbH, Switzerland) were used for graft fixation (Figure 2).

A postoperative course was uneventful. A control orthopantomogram, which was made immediately after the operation, showed a good position of the graft. Three months after the operation, a complete graft integration with a certain degree of resorption was radiologically confirmed.

On the basis of the control radiography after 10 months, a definite resorption of graft was estimated to be about 20%.

A final decision about prosthetic rehabilitation with fixed dentures supported by implants in the early loading protocol was made in agreement with the patient after having been informed about all the eventual possibilities.

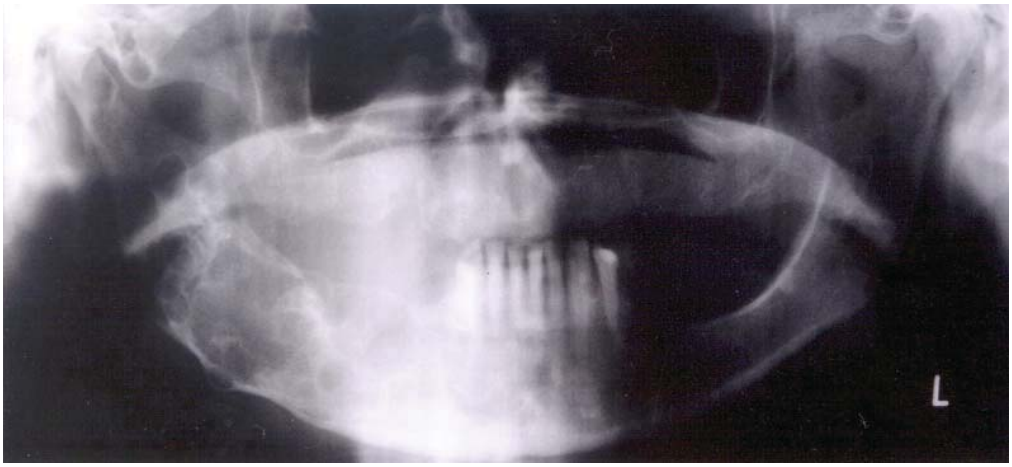


Fig. 1 – Orthopantomograph with odontogenic keratocyst

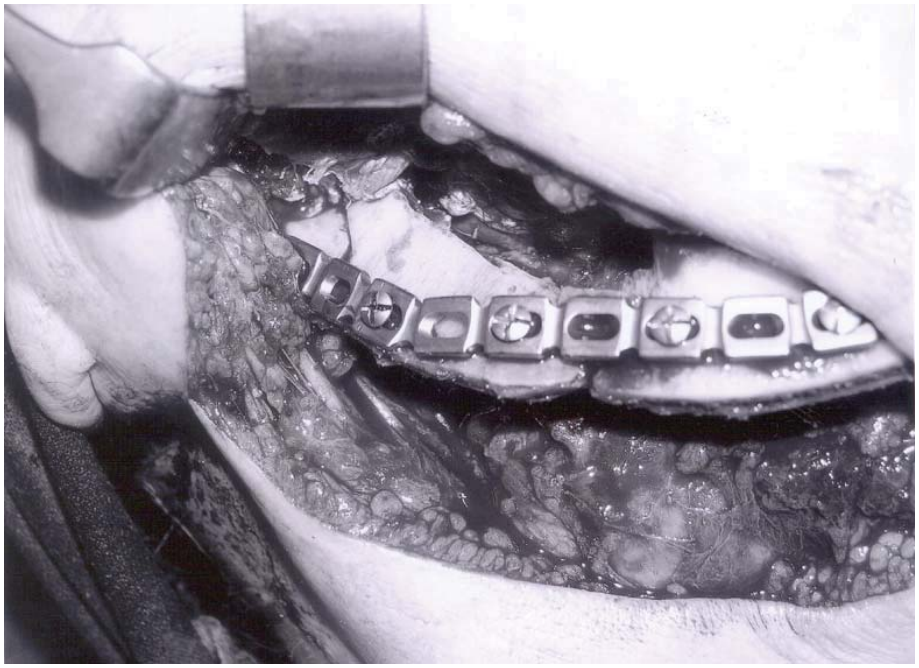


Fig. 2 – Fixation of the graft with reconstructive plate and screws

Based on the orthopantogram, vertical and horizontal bone dimensions were insufficient for placement of conventional screw implants. To have an insight into the exact situation, the procedure continued with the 3D planning on the basis of CT data using the software (Mimics, Materialise, Belgium). This software allowed manufacturing of a mandible biomodel by means of 3D copying (rapid prototyping). In the mean time, the impressions of both jaws were taken and plaster study models were obtained. The biomodel served to carefully analyse the available bone, in order to determine the exact shape, size and the position of implants (Figure 3). In addition, a surgical stent which helped in inserting implants in the pre-determined specific positions was made.

The implantation procedure was performed under general anesthesia. Intraoperatively, following rising mucoperiosteal flap, the stability of bone graft was confirmed. After surgical stent adaptation, implantation was done according to the protocol for disk-shaped implants (lateral insertion). In the residual alveolar ridge in the molar region at the left side, an implant was placed, and in the graft at the right side, two disk-shaped implants were inserted (Diskos-ID Brand, Dr. Ihde Dental AG, Switzerland) (Figure 4). Six days after the operation, the impressions were taken, and the implants were loaded with a temporary composite bridge on the day 10.

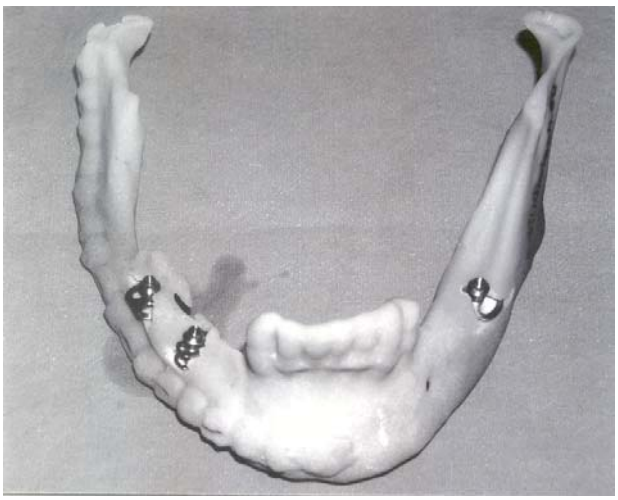


Fig. 3 – Biomodel with implants

The control CT and orthopantomographs showed an excellent position of implants. Six months later, a complete integration of implants was determined clinically (radiologically), and a definite metalceramic circular bridge was produced. There were no signs of marginal bone loss around the loaded implants after 2 years, which was confirmed by control orthopantomograph (Figure 5).

Fig. 4 – Disk implants inserted in reconstructed mandible

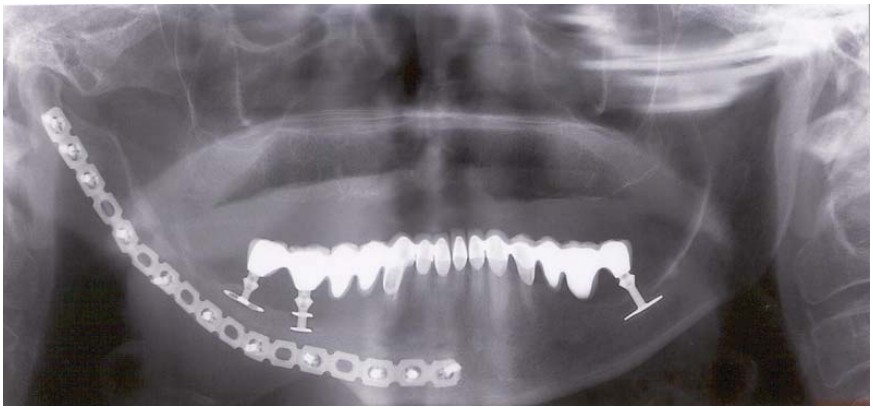


Fig. 5 – Orthopantomograph two years after implantation

Discussion

There are no many papers on mandible reconstruction with free bone grafts and oral implants in the literature. However, there are several hesitations concerning this topic which need further consideration.

First of all, considering the currently available techniques for mandible reconstruction, the question arises about which cases are suitable for the reconstruction with free bone grafts as a method of choice. There are several crucial criteria for a definite decision making. As concerns the size of the defect, Goh et al. [1] believe that free bone grafts are still a good option for defects smaller than 5 cm, if the surrounding soft tissues are in good condition. Hotz [9] gives priority to the delayed mandible reconstruction with a free iliac graft for defects smaller than 8 cm. Foster et al. [5] in their comparative study concluded that the use of avascular bone grafts is limited to smaller bone defects (< 5–6 cm), in patients who do not undergo irradiation therapy and/or do not have a compromised general condition. Pogrel et al. [10] share similar opinion indicating the dimension of 6 cm as an upper limit for use of avascular grafts. Contrary to them, Chiapasco et al. [11] in their retrospective study showed that the limiting factor for the use of avascular grafts is not the size of defect but surely the insufficient quality and quantity of surrounding soft tissues, in the sense of compromised revascularization. The mentioned study presents successful reconstructions also for defects which spread from the symphyseal part to the condylar region of the mandible.

The majority of authors agree that the iliac crest is the best donor site, because of easy approach and possibility for taking a large amount of bone [3, 9, 10, 12].

One of the dilemmas is whether the reconstruction should be done simultaneously with mandible resection or subsequently. Hotz [9] indicates an important problem of simultaneous reconstruction in cases with malignancies, because it is not possible to perform a histopathological verification of the tumour free margins. Also, compared with the postponed reconstruction, the duration of intervention is significantly prolonged and therefore the risk of postoperative complications is increased.

Since the patient presented in this paper was involved with a benign lesion (the patient was not irradiated), the size of the bone defect was estimated to be about 8 cm, and soft tissues were of a satisfactory quality, the decision was made for a primary reconstruction with a free bone iliac graft. According to the exact dimensions of the resected part of the mandible, the graft was reshaped and fixed in two parts in order to adequately reconstruct the mandible contours.

A contemporary approach to a patient definitive rehabilitation after mandible resection does not imply an anatomic reconstruction only, but also a prosthetic rehabilitation. In the past, patients were mostly rehabilitated with mobile dentures of limited functional and aesthetic values. The introduction of endosseal implants provided rehabilitation with fixed dentures showing to be more comfortable, and significantly improving both function and aesthetic. There are numerous studies describing successful application of conventional screw implants in reconstructed mandibles [6, 9, 12, 13].

When considering the right timing for implants placement, there are two reasons in favour of the delayed implantation. The first is in the fact that the successful osseointegration depends on osteoblasts capable for osteogenesis, and bone grafts are, so to say “a dead bone” as long as the process of the so-called “acceptance of the grafts” does not start. Another reason is that the simultaneous implantation is proportionally more demanding and rarely meets prosthetic requirements. Lekholm et al. [14] have concluded that implantation success is higher with the delayed approach. Lundgren et al. [13] in their research revealed that the delayed procedure not only results in bigger amount of bone on the implant’s surface but also stimulates further remodelling and formation of a new bone. Foster et al. [5] mentioned that it is necessary to wait for 5 to 6 months after reconstruction, so that the implantation procedure may be successful. Considering the size of the graft and the time necessary for remodelling and formation of osteogenetic potential in the presented case, the decision was made to place the implants subsequently, at least after 6 months.

The basic prerequisite of successful implantation procedure is a sufficient quantity of bone. When free bone grafts are concerned, the expected resorption is 25% in relation to the initial graft volume [15]. One study tested the average vertical resorption of the graft and the value obtained was 3.53 cm [14]. Stošić [16] indicates an average resorption of graft to be 15%–30%. Also, it is important to know whether the patient underwent a postoperative irradiation therapy [17].

A problem might occur with the lack of bone for placement of screw implants. If so, it is possible to apply various augmentation procedures or to use particular implant systems [18, 19]. The use of basal osseointegrated (disk) implants in immediate or early loading protocols is a useful solution if a bone dimensions are insufficient for placing conventional screw type implants. By means of that, total rehabilitation time could be significantly shortened, which is very convenient to patients.

It is very important to point out that the multidisciplinary approach and good planning together with the use of adequate measuring is of crucial importance for an overall favourable outcome [20]. A real 3D determination of future implant positions on a biomodel obtained on the basis of CT data appears very useful if a limiting anatomic factor is present. Also, it was possible for the patient to become familiar with the planned procedure.

Conclusion

It is the fact that the introduction of microvascular flaps has reduced indications for the use of free bone grafts. However, in favourable condition of soft tissues of the accepting region, free bone grafts are still very reliable method for mandible reconstruction. The best way to achieve definitive patient rehabilitation with a reconstructed mandible is to make a fixed denture supported by implants, because it is

very comfortable for a patient both in the functional and aesthetic sense.

In case of insufficient bone dimensions of the reconstructed mandible, the use of disk implants in immediate or early loading protocols is an useful optional method of rehabilitation.

Acknowledgement

A part of this research was financed with the Grant No 175075 of the Ministry of Education, Science and Technological Development of the Republic of Serbia.

REFERENCES

1. *Goh BT, Lee S, Tideman H, Stoelinga PJ.* Mandibular reconstruction in adults: a review. Int J Oral Maxillofac Surg 2008; 37(7): 597−605.
2. *Takushima A, Harii K, Asato H, Momosawa A, Okazaki M, Nakatsuka T.* Choice of osseous and osteocutaneous flaps for mandibular reconstruction. Int J Clin Oncol 2005; 10(4): 234−42.
3. *Ehrenfeld M, Hagenmaier C.* Autogenous Bone Grafts in Maxillofacial Reconstruction. In: *Greenberg AM, Prain J*, editors. Craniomaxillofacial Reconstructive and Corrective Bone Surgery: Principles of Internal Fixation Using the AO/ASIF Technique. New York: Springer; 2002. p. 295−309.
4. *Wedler V, Farshad M, Sen M, Koehler C, Hanschin A, Graetz K, et al.* Retrospective analysis and clinical evaluation of mandible reconstruction with a free fibula flap. Eur J Plast Surg 2007; 29(6): 285–91.
5. *Foster RD, Anthony JP, Sharma A, Pogrel MA.* Vascularized bone flaps versus nonvascularized bone grafts for mandibular reconstruction: an outcome analysis of primary bony union and endosseous implant success. Head Neck 1999; 21(1): 66−71.
6. *Hidalgo DA, Pusic AL.* Free-flap mandibular reconstruction: a 10-year follow-up study. Plast Reconstr Surg 2002; 110(2): 438−49; discussion 450−1.
7. *Ghassemi A, Ghassemi M, Riediger D, Hilgers RD, Gerressen M.* Comparison of donor-site engraftment after harvesting vascularized and nonvascularized iliac bone grafts. J Oral Maxillofac Surg 2009; 67(8): 1589−94.
8. *Ihde S.* Principles of BOI, Clinical Scientific, and Practical Guidelines to 4-D Dental Implantology. Berlin, Heidelberg, New York: Springer- Verlag; 2005.
9. *Hotz G.* Reconstruction of mandibular discontinuity defects with delayed nonvascularized free iliac crest bone grafts and endosseous implants: a clinical report. J Prosthet Dent 1996; 76(4): 350−5.
10. *Pogrel MA, Podlesh S, Anthony JP, Alexander J.* A comparison of vascularized and nonvascularized bone grafts for reconstruction of mandibular continuity defects. J Oral Maxillofac Surg 1997; 55(11): 1200−6.
11. *Chiapasco M, Colletti G, Romeo E, Zaniboni M, Brusati R.* Long-term results of mandibular reconstruction with autogenous bone grafts and oral implants after tumor resection. Clin Oral Implants Res 2008; 19(10): 1074−80.
12. *Chiapasco M, Abati S, Ramundo G, Rossi A, Romeo E, Vogel G.* Behavior of implants in bone grafts or free flaps after tumor resection. Clin Oral Implants Res 2000; 11(1): 66−75.
13. *Lundgren S, Rasmusson L, Sjöström M, Sennerby L.* Simultaneous or delayed placement of titanium implants in free autogenous iliac bone grafts. Histological analysis of the bone graft-titanium interface in 10 consecutive patients. Int J Oral Maxillofac Surg 1999; 28(1): 31−7.
14. *Lekholm U, Wannfors K, Isaksson S, Adielsson B.* Oral implants in combination with bone grafts. A 3-year retrospective multicenter study using the Brånemark implant system. Int J Oral Maxillofac Surg 1999; 28(3): 181−7.
15. *Stoll P, Prein J, Bähr W, Wächter R.* Considerations in the fixation of bone grafts for the reconstruction of mandibular continuity defects. In: *Greenberg AM, Prain J*, editors. Craniomaxillofacial Reconstructive and Corrective Bone Surgery: Principles of Internal Fixation Using the AO/ASIF Technique. New York: Springer; 2002. p. 317−26.
16. *Stošić S.* Mandibular reconstruction. Belgrade: Rubikon; 2005. (Serbian)
17. *Ihde S, Kopp S, Gundlach K, Konstantinović VS.* Effects of radiation therapy on craniofacial and dental implants: a review of the literature. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2009; 107(1): 56−65.
18. *Konstantinović V.* Aspekte der implantologischen Versorgung mit BOI im Bereich des Sinus maxillaris. ZMK 2003; 19: 568−75.
19. *Konstantinović VS, Lazić VM, Stefan I.* Nasal epithesis retained by basal (disk) implants. J Craniofac Surg 2010; 21(1): 33−6.
20. *Arsić S, Perić P, Stojković M, Ilić D, Stojanović M, Ajduković Z, Vučić S.* Comparative analysis of linear morphometric parameters of the humane mandibula obtained by direct and indirect measurement. Vojnosanit Pregl; 2010; 67(10): 839–46. (Serbian)

Received on June 13, 2011.
Revised on August 25, 2011.
Accepted on September 5, 2011.

IN FOCUS

UDC: 618.146-006
DOI: 10.2298/VSP1301086P

Cervical cancer screening in Serbia

Skrining karcinoma grlića materice u Srbiji

Živko Perišić*†, Vesna Plešinac-Karapandžić‡, Milica Džinić†, Milena Zamurović†, Nataša Perišić†

***Faculty of Medicine, University of Belgrade, Serbia; †University Clinic of Gynecology and Obstetrics "Narodni front", Belgrade, Serbia; ‡Institute of Oncology and Radiology of Serbia, Belgrade, Serbia**

Key words:
uterine cervical neoplasms; mass screening; diagnosis; cytological techniques; incidence.

Ključne reči:
grlić materice, neoplazme; masovno ispitivanje; dijagnoza; citološke tehnike; incidenca.

Introduction

The article focuses on the beginning of cervical cancer screening in Serbia. The opportunistic screening was introduced in a regular gynecological practice in the early sixties and preventive gynecological examination has been performed since then on a yearly basis. The proportion of population screened was unknown, and the standards for quality assurance and control were not available. The national program for such screening was established in 2011 encouraging women between 25 and 69 years to undergo a preventive gynecological examination together with Pap smear once in a 3-year period [1].

Based on the experience of countries with effectively organized screening programs, a decision was made in 2006 by the Minister of Health to nominate a group of experts to prepare a proposal for organized cervical cancer screening program after testing the methodology in a pilot study in the District of Braničevo.

The specific objectives were to evaluate the reduction of the incidence and mortality from cervical cancer in the Province by means of an organized low-intensity cervical cytology program, as well as to demonstrate the different aspects of program implementation as a potential model for nationwide implementation.

Screening activities were integrated in the existing health care system. Organized screening for women in the target population (aged 25–69 years) were planned to be free of charge. Sample taking was done by the gynecologists and primary health care personnel in the local health care centers. Sample quality was under continuous control by the cytology laboratories. Confirmation and treatment were integrated into the normal health care routines. The screening results of the program, including the histologically confirmed diagnosis, were registered at the National Cancer Institute [1].

The impact of the screening program was assessed indirectly by comparing trends in invasive cervical cancer, changes in coverage, and changes in the interval between Pap smears.

Overview of the cervical cancer screening in Serbia during the years before

According to the data of the Register of Central Serbia for the Malignant Tumors 1,400 new cases of cervical cancer are discovered on the territory of Serbia every year. Considering its frequency, this is the second frequent cancer in women in Serbia, after breast cancer. The standardized incidence rate of cervical cancer in the Central Serbia in 2002 was 27.2 in 100,000 women, which was the highest incidence rate in Europe. Similar, high rates were also recorded in Romania, Albania and Bosnia and Herzegovina. According to the Institute of Statistics of the Republic of Serbia for the year 2002, a total of 452 women died because of cervical cancer. The standardized mortality rate was 7.2 in 100,000 which was lower than in the mentioned countries of the region [2].

The incidence rate of cervical cancer was higher in Central Serbia than in the Province of Vojvodina. Apart from this, there were some significant differences in incidences between the districts of Central Serbia. In 2002 the lowest incidences (16.6 per 100,000 were recorded in the District of Mačva) and the highest (more than two times as high) were recorded in the Eastern Serbia, in the border area with Romania and in Belgrade [3].

Correspondence to: Živko Perišić, University Clinic for Obstetrics and Gynecology „Narodni front", Kraljice Natalije 62, 11000 Belgrade, Serbia. Phones: +381 63 21 66 77; +381 65 221 66 77. E-mail: zivkoperisic@gmail.com

Age distribution of patients with cervical cancer in Serbia

The risk for occurrence of cervical cancer increases with the age. The highest number of patients is between 45 and 54 years of age. However, the illness may, although very rare, occur even much earlier, e.g. even before 20 years of age. Age distribution of cervical cancer patients in Central Serbia in 2002 [3] is shown in Table 1.

Table 1

Age distribution of cervical cancer patients in Central Serbia in 2002 [3]

Patient's age (years)	Patients		Age specific rate of cervical cancer per 100,000 patients
	n	%	
0–14	0	0.0	0.0
15–19	1	0.1	0.6
20–24	1	0.1	0.6
25–29	23	2.3	12.4
30–34	40	4.0	22.7
35–39	75	7.5	42.9
40–44	108	10.8	56.7
45–49	162	16.1	75.3
50–54	173	17.2	77.9
55–59	105	10.5	66.3
60–64	89	8.9	55.4
65–69	93	9.3	52.2
70–74	77	7.7	46.5
75+	57	5.7	29.5

According to the available data, less than one third of the cases of cervical cancer are discovered in an early invasive phase in which only operative treatment can be successfully applied. Most of patients are in later stages, when it is only possible to conduct radiotherapy, which results in longer treatment, different complications and significantly increased treatment expenses.

New cervical cancer screening program in Serbia

On July 3 2006 the Ministry of Health of the Republic of Serbia appointed the Expert Commission for the prevention of cervical cancer primarily aimed at developing a National Program for Prevention of Cervical Cancer.

Regarding the importance of the problem as well as the fact that in Serbia one woman dies per day due to cervical cancer, respecting the recommendations of WHO, and analyzing screening programs of other countries and using the experiences from the Pilot Program from the District of Braničevo, the Commission made the Program to enable the beginning of screening of cervical cancer in our country. This Program was adopted by the Government of the Republic of Serbia issued in Official Gazette No 54 from 23 May 2008 [1].

Serbia has enough gynecologists and other medical personnel to conduct screening. A partial change in organization was expected to be achieved by introducing organized screening. This primarily refers to education of cytoscreeners and to organization of laboratory service for cytology.

On the territory of Serbia, screening of cervical cancer is conducted through an organized decentralized program.

The target group for cervical cancer screening

The decision on target group for cervical cancer screening, as well as on interval between check-ups is most usually made on the national level considering the presence of cervical cancer, frequency of human papilloma virus (HPV) infections and available infrastructure means. The World Health Organization (WHO) recommends that the new screening programs should include women starting from 30 years of age. Women between 25 and 29 should be included only in the case when women of 30 + have been screened. Screening should not include women under 25 years of age. In addition, screening in women older than 65 can be stopped, if they have two consecutive negative findings [4].

According to data from the Central Serbia the frequency rate of cervical cancer is higher in all age groups in women between 35 and 74 years of age (Table 1). Because of this, establishing the upper age limit, even over 65 years, would enable the revealing of prevalent cases of cervical cancer. Moving the lower limit of target group toward younger age groups, even despite the small risk for cervical cancer occurence, would enable revealing precancerous lesions.

The screening program included women between 25 and 69 years of age. The target group was identified with the help of election lists or data base of personal identification numbers and list of insured persons of the Republic of Serbia Bureau for Health Insurance. The call for testing was sent to all women from the target group.

Testing included cytological examination of cervical smear (Papanicolau test) observing the professional-methodological instructions. Screening was performed in every third year. Taking cervical smears was done by the Service for the Protection of Women's Health in the Primary Health Care Center.

Cytological examination of cervical smear (Papanicolau test) underwent in cytological laboratory in the Primary Health Care Center and only for that Primary Health Care

Center, or in Primary Health Care Center which is responsible for a whole district [1].

Reading the results of tests as well as determining the dynamic and content of further monitoring and referring to the other diagnostics was done by the Service for the Protection of Women's Health in the Primary Health Care Center. The results were read by the gynecologists who initially took smears for cytological diagnostics or the selected gynecologists in the Primary Health Care Center.

In case of negative result, a patient was given a time recommendation for the next control examination and this was evidenced in the suitable Data Base of organized screening of cervical cancer. A patient stayed in a regular screening program and was called for the next check-up [3].

A positive result was reported by the doctor so that a patient understood the significance of further diagnostics. In case of inflammatory reversible changes, the patients were treated in the facilities of primary level and cytological test control was scheduled not earlie then 6 weeks after the treatment.

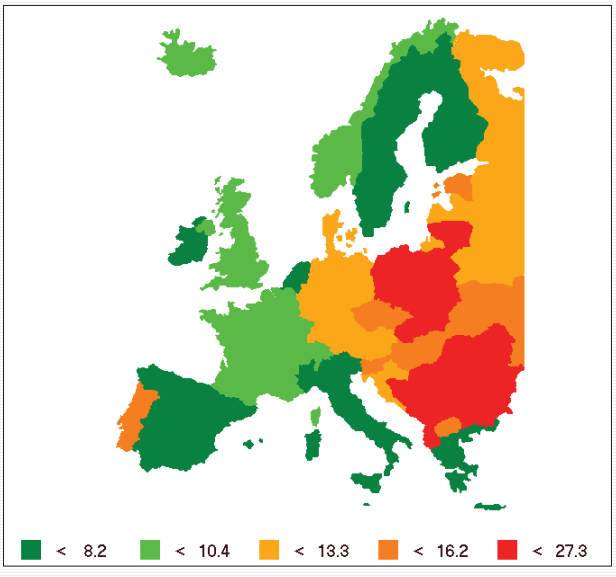
All the other histopathological findings required further procedure in accordance with the protocol for monitoring after receiving results from cytological examination. If a smear was unsatisfactory for cytological analysis, a patient was called again for the control examination in 6–8 weeks.

levels of health care, as well as communication channels taking special care that the procedure be easy, acceptable and understandable for patients with the minimum of stress. It is extremely important to ensure adequate communication with patients at every level. This means explaining all possibilities of the treatment and its results, and getting an acceptance of further treatment, giving a patient an opportunity to freely ask questions in every phase of the treatment.

Women with low-grade lesions are submitted to routine follow-up smears. High-grade preinvasive disease was further evaluated by repeating Pap smear, conization or biopsy and subsequent treatment through surgical removal or ablation. This organized low-intensity cervical cytology programme showed a considerable increase in cervical intraepithelial neoplasia (CIN) II–III cases and should reduce incidence of and mortality from cervical cancer in the future. Screening with the Papanicolaou smear plus adequate follow-up diagnosis and therapy can achieve major reductions in both incidence and mortality rates.

Uterine cervical cancer was one of the leading cancer among women in Serbia with age-standardized incidence rates of 23–27 per 100,000 in 2002 [2]. Cervical cancer incidence rate in Europe is shown in Figure 1.

Uterine cancer cervix is the second most common cancer in females in the world with about half a million new pa-



		MT
C. Serbia	27.3	12.6
Romania	23.9	13.0
Bosnia & Herz.	21.3	8.0
Bulgaria	18.7	8.0
Slovakia	18.5	6.1
Poland	18.4	7.8
Czech Republick	16.2	5.5
Slovenia	16.1	4.7
Hungary	15.7	6.7
FYR Macedonia	13.9	7.6
Portugal	13.5	4.5
Croatia	13.3	5.0
Netherlands	7.3	2.3
Ireland	7.2	3.5
Finland	4.3	1.8

Fig. 1 – Cervical Cancer Incidence Rate in Europe [2]
(standardised rate per 100,000 women)

Referring the patients to the further diagnostics

A patient was referred to further diagnostics in a regional health facility of secondary level. With histopathological finding the patient returned to the chosen doctor who, depending on the kind of a diagnosed change acted in accordance with the recommendations of the protocol for monitoring women after receiving results from histopathological finding. In order to fulfill the whole value of the procedure it necessary to clearly define all the competences of various

tients per year [4]. Cervical cancer is an important public health problem among adult women in developing countries [5]. Since the introduction by Papanicolaou of cervical smear screening, the incidence of cervical cancer has declined in many developed countries [2, 6]. A decrease in the incidence of and the mortality from cervical cancer is mainly due to the organized mass screening using Pap smear programs. To organize an effective screening program in developing countries it should find adequate financial resources, develop the infrastructure, train the needed manpower, and

elaborate surveillance mechanisms for screening, investigating, treating, and following up the targeted women. The findings from the large body of research on various screening approaches carried out in developing countries and from the available managerial guidelines should be taken into account when reorganizing existing programmes and when considering new screening initiatives [7].

Cytological screening has reduced the incidence of cervical cancer in countries with organized screening, but in Europe in 1995 there were still an estimated 68,000 incident cases [5, 7, 8]. Cytology has limited reproducibility, and both meta-analyses and pooled analyses of cross sectional studies have established that tests for HPV have higher sensitivity than cytology in detecting high grade CIN and that combined HPV and cytology testing have high negative predictive values for CIN [9–11].

Cost-effectiveness modeling of screening strategies, however, depends greatly on reliable and generalisable estimates of the longitudinal, long term predictive values of testing. A long-term negative predictive value is the main determinant of a safe screening interval to use, a key factor for the cost efficiency of a screening program. The long-term positive predictive value is an important measure of the extent of unnecessary procedures induced by screening, another major factor in evaluations of cost-efficiency. Several randomized controlled trials are currently being conducted to compare primary screening based on HPV detection with conventional cytology screening [12–17]. Data from these trials indicate that HPV based screening results in detection of more high grade CIN lesions (a higher sensitivity) but a reduced specificity compared with cytology based screening [18–21].

Conclusion

The coordinated screening programme provides a low-cost, increases the coverage of the female population, and consequently reduces the rate of invasive cervical cancer.

REFERENCES

1. Official Gazette No 64/2008. The National Program for the prevention of cervical cancer. Available from: http://www.slglasnik.com, www.e-glasnik.rs
2. *Ferlay J, Bray F, Pisani P, Parkin DM.* GLOBOCAN 2002: Cancer Incidence. Mortality and Prevalence Worldwide. Lyon, France: IARC Press; 2004.
3. *Miljuš D, Živković S.* Incidence and mortality of cervical cancer in central Serbia. Belgrade: Institute of Public Health of Serbia "Dr Milan Jovanović - Batut"; 2004.
4. *Sankaranarayanan R, Budukh AM, Rajkumar R.* Effective screening programmes for cervical cancer in low- and middle-income developing countries. Bull World Health Organ 2001; 79(10): 954−62.
5. *Bray F, Loos AH, McCarron P, Weiderpass E, Arbyn M, Møller H*, et al. Trends in cervical squamous cell carcinoma incidence in 13 European countries: changing risk and the effects of screening. Cancer Epidemiol Biomarkers Prev 2005; 14(3): 677−86.
6. *Boyle P, Levin B.* World Cancer Report 2008. Lyon: International Agency for Research on Cancer (IARC); 2008.
7. *Dillner J, Rebolj M, Birembaut P, Petry KU, Szarewski A, Munk C*, et al. Long term predictive values of cytology and human papillomavirus testing in cervical cancer screening: joint European cohort study. BMJ 2008; 337: a1754.
8. *Bray F, Sankila R, Ferlay J, Parkin DM.* Estimates of cancer incidence and mortality in Europe in 1995. Eur J Cancer 2002; 38(1): 99−166.
9. *Stoler MH, Schiffman M.* Atypical Squamous Cells of Undetermined Significance-Low-grade Squamous Intraepithelial Lesion Triage Study (ALTS) Group. Interobserver reproducibility of cervical cytologic and histologic interpretations: realistic estimates from the ASCUS-LSIL Triage Study. JAMA 2001; 285(11): 1500−5.
10. *Cuzick J, Clavel C, Petry KU, Meijer CJ, Hoyer H, Ratnam S*, et al. Overview of the European and North American studies on HPV testing in primary cervical cancer screening. Int J Cancer 2006; 119(5): 1095−101.
11. *Arbyn M, Sasieni P, Meijer CJ, Clavel C, Koliopoulos G, Dillner J.* Chapter 9: Clinical applications of HPV testing: a summary of meta-analyses. Vaccine 2006; 24 Suppl 3: S3/78−89.
12. *Sherman ME, Lorincz AT, Scott DR, Wacholder S, Castle PE, Glass AG*, et al. Baseline cytology, human papillomavirus testing, and risk for cervical neoplasia: a 10-year cohort analysis. J Natl Cancer Inst 2003; 95(1): 46−52.
13. *Hoyer H, Scheungraber C, Kuehne-Heid R, Teller K, Greinke C, Leistritz S,* et al. Cumulative 5-year diagnoses of CIN2, CIN3 or cervical cancer after concurrent high-risk HPV and cytology testing in a primary screening setting. Int J Cancer 2005; 116(1): 136−43.
14. *Mayrand MH, Duarte-Franco E, Rodrigues I, Walter SD, Hanley J, Ferenczy A*, et al. Human papillomavirus DNA versus Papanicolaou screening tests for cervical cancer. N Engl J Med 2007; 357(16): 1579−88.
15. *Bulkmans NW, Berkhof J, Rozendaal L, van Kemenade FJ, Boeke AJ, Bulk S,* et al. Human papillomavirus DNA testing for the detection of cervical intraepithelial neoplasia grade 3 and cancer: 5-year follow-up of a randomised controlled implementation trial. Lancet 2007; 370(9601): 1764−72.
16. *Kitchener HC, Almonte M, Wheeler P, Desai M, Gilham C, Bailey A*, et al. HPV testing in routine cervical screening: cross sectional data from the ARTISTIC trial. Br J Cancer 2006; 95(1): 56−61.
17. *Kotaniemi-Talonen L, Nieminen P, Anttila A, Hakama M.* Routine cervical screening with primary HPV testing and cytology triage protocol in a randomised setting. Br J Cancer 2005; 93(8): 862−7.
18. *Naucler P, Ryd W, Törnberg S, Strand A, Wadell G, Elfgren K,* et al. Human papillomavirus and Papanicolaou tests to screen for cervical cancer. N Engl J Med 2007; 357(16): 1589−97.
19. *Ronco G, Giorgi-Rossi P, Carozzi F, Dalla Palma P, Del Mistro A, De Marco L*, et al. Human papillomavirus testing and liquid-based cytology in primary screening of women younger than 35 years: results at recruitment for a randomised controlled trial. Lancet Oncol 2006; 7(7): 547−55.
20. *Wahlström C, Iftner T, Dillner J, Dillner L.* Population-based study of screening test performance indices of three human papillomavirus DNA tests. J Med Virol 2007; 79(8): 1169−75.
21. *Cuzick J, Szarewski A, Mesher D, Cadman L, Austin J, Perryman K,* et al. Long-term follow-up of cervical abnormalities among women screened by HPV testing and cytology – Results from the Hammersmith study. Int J Cancer 2008; 122(10): 2294−300.

Received on March 23, 2011.
Revised on August 29, 2011.
Accepted on September 5, 2011.

I S T O R I J A M E D I C I N E

UDC: 355.415.6(091)

Sanitet dobrovoljačkog pokreta Južnih Slovena u Rusiji (1914–1919) – srpski dobrovoljački pokret

South Slav Volunteer Movement Medical Service in Russia (1914–1919) – Serbian Volunteer Movement

Aleksandar S. Nedok

Akademija medicinskih nauka Srpskog lekarskog društva, Beograd, Srbija

Ključne reči:
vojni sanitet, dobrovoljci, srpski dobrovoljački korpus, rusija, prvi svetski rat.

Key words:
military medical service, south slav volunteers, serbian volunteer corps, russia, first world war.

Uvod

Već od samog početka Prvog svetskog rata u Rusiji, među austrougarskim vojnicima slovenskih naroda koji su bili prebegli ili zarobljeni, pojavio se pokret za učešće u borbama na savezničkoj strani kako bi i lično doprineli oslobođenju svojih naroda od viševekovne tuđinske vlasti. Pokret je bio posebno snažan među mlađim, školovanim zarobljenicima koji su bili zadojeni slobodoumnim idejama nastalim tokom XIX veka unutar Austrougarske carevine. Nižeobrazovani su za njih bili manje zainteresovani, što je bila prirodna posledica denacionalizujuće politike polufeudalne države u kojoj su živeli [1–3].

Pošto ruska Vlada skoro do kraja 1915, poštujući međunarodne konvencije, nije bila spremna da na svojoj teritoriji od zarobljenika stvara vojne jedinice, ona je sve te dobrovoljce upućivala u Srbiju. Tek u kasnu jesen 1915, kada je neprijateljskom ofanzivom prekinuta komunikacija sa Srbijom, Rusija se, odlukom cara Nikole II, saglasila da se srpske dobrovoljačke jedinice počnu stvarati na njenoj teritoriji [4]. Sa svoje strane, srpska Vlada i njena vojska još od početka rata i kasnije svesrdno su prihvatale ovaj pokret, svesne ne samo njegovog vojničkog, već i političkog značaja. Politički, jer se u potpunosti uklapao u stavove „Niške deklaracije“ iz decembra 1914. godine o ratnim ciljevima Srbije i njenoj borbi za oslobođenje i ujedinjenje svih Južnih Slovena u zajedničku državu, i vojnički, imajući u vidu ratne potrebe: popune postojećih ili obrazovanja novih jedinica, makar bilo to privremeno daleko od, za Srbiju glavnog, Solunskog fronta.

Dobrovoljci u Srbiji (1914–1915)

Rusija je odmah po otpočinjanju neprijateljstava uspostavila putem Dunava vezu sa Srbijom. Svojom Dunavskom flotilom i svim raspoloživim srpskim brodovima, ona je neprekidno slala izdašnu pomoć u sanitetskom materijalu i stručnom osoblju, namirnicama, odeći, naoružanju i drugim potrepštinama, a povratno su u Rusiju plovile strane delegacije i ratna tehnika sa Zapada (Marsej–Solun–Niš–Prahovo–Odesa). Bez ovog konstantnog pomaganja iz Rusije Srbija ne bi izdržala ratne napore i njen bi slom bio neminovan već i iz najosnovnijeg razloga – propale letine 1914. i nemogućnosti da se prehrane vojska i narod, ne uzimajući u obzir druge vidove pomoći u naoružanju, municiji, vojnoj odeći i obući koji su bili naročito intenzivni posle Cerske bitke i tokom bitke na Drini [4, 5].

U prahovsko pristanište, gde je stvorena velika baza, stizale su dovoljne količine brašna, pa ga je uvek „bilo u potpuno dovoljnim količinama za ishranu naše vojske“, kako svedoči đeneral Živko Pavlović, pomoćnik vojvode Putnika, dodajući da je „u bazi bilo (i) stovarište oružja i municije, pa i nekoliko baterija“ za isporuku srpskoj vojsci [6]. Dnevno je stizalo u proseku po 40 tona ljudske i stočne hrane, od čega se deo slao i crnogorskoj vojsci.

Tim putem krajem 1914. (manje) i tokom devet meseci 1915. (većinom) u Srbiju prebačeno je oko 3 500 dobrovoljaca koji su u jesen 1915. učestvovali u borbama na bugarskoj granici u sastavu „Južnomoravskih trupa“. Najveći deo tih dobrovoljaca je izginuo.

Među dobrovoljcima još 1914. bilo je nekoliko studenata medicine koji su radili u ruskim bolnicama u Nišu (Đorđe Milić, Krsta Grabovački, Milan K. Jovanović, Kosta Živanović i Vuko Bošković) [4].Takođe, sa ruskim misijama došlo je i nekoliko lekara i milosrdnih sestara srpskog porekla koji su se zadesili u Rusiji na školovanju ili radu (lekari dr Radul Backović, dr Arsenije Džuverović i dr Risto Žerajić i studentkinje medicine Jelisaveta Sundečić i Ruskinja Tamara udata Petrović-Njegoš) [4].

Correspondence to: Aleksandar S. Nedok, Akademija medicinskih nauka Srpskog lekarskog društva, Džordža Vašingtona 17, Beograd, Srbija. E-mail: anedok@sbb.rs

Međutim, ne treba zaboraviti da je pored dobrovoljaca koji su u manjim grupama počeli da pristižu iz Rusije Dunavom bilo i onih koji su prebegli na razne načine iz BiH, Vojvodine i drugih pokrajina Austrougarske države, ili iz austrougarskih jedinica na srpskom frontu, ili su zarobljeni tokom bitaka na Ceru, Drini i Kolubari i potom svojevoljno pristupali Srpskoj vojsci.

Politikom srpskog vojnog rukovodstva svi dobrovoljci, bilo kojim putem ili načinom da su se našli u Srbiji, da bi se izbeglo njihovo zarobljavanje, a samim tim i smrtna osuda za njih i progon njihovih porodica, najčešće su slati u jedinice Trupa novih oblasti, na granice prema Bugarskoj i Albaniji (među njima medicinari Josif Goldberg, Lovro Klemenčič-Dušan) [7], ili u druge pozadinske sanitetske ustanove (dr Nikola Blagojević, dr Ivo Petković, dr Milan Simonović, dr Karel Freja-Dragutin, dr Toma Živanović, dr Miloš Pučelka, dr Petar Zec, dr Roman Fedina, dr Jovan Popović, studenti medicine Franjo Vuletić, Ludvik Horvatin-Lujo, Bogdan Vorkapić, Miloš Ćirić, Viktor Sosič, Viktor Stibl, Ivan Kalčič, Radoslav Miletić, Jovan Milošević, Dinko Cvitanović, Savatije Mitrović, Franja Navratil, Antun Saso, Kornel Radulović i zubar Ludvik Zadnjak-Đorđe [7], svi pripadnici slovenskih naroda sa teritorije Austrogarske: Srbi, Hrvati, Slovenci, Česi, Slovaci i Ukrajinci.

Među dobrovoljcima bilo je i onih koji su u Srbiji ostali još iz vremena balkanskih ratova, ili su se u njoj našli za vreme Vidovdanske proslave 1914. godine, odlučivši da se ne vrate; oni su od samoga početka rata učestvovali u borbama, pa je među njima bilo poginulih i ranjenih. U Cerskoj bici, prvom velikom sukobu dve vojske i prvoj srpskoj i, uopšte, prvoj savezničkoj pobedi u ratu, poginula su dva jugoslovenska rodoljuba, mladi Augustin Jenko, osnivač i duhovni predvodnik ljubljanske đačkostudentske organizacije „Preporod“ iz koje su potekli mnogi članovi dobrovoljačkih jedinica u Rusiji, i kapetan 2 klase Srpske vojske, komandir čete bivši austrougarski aktivni oficir Slovenac Martin Javornik, posmrtno odlikovan „Karađorđevom zvezdom sa mačevima“ [8], a ranjen još jedan preporodovac, Vladislav Fabjančič. Oni su se borili u sastavu 2. prekobrojnog puka Kombinovane divizije. Za vreme borbi za Adu Ciganliju poginuo je dobrovoljac četničkog odreda, Slovenac Viktor Dajsinger [9].

Slično je i sa Slovencem, poručnikom Srpske vojske iz vremena balkanskih ratova, inž. Ivanom Gosarom, koga su, teško ranjenog u jesen 1915, zarobili Bugari i predali austrougarskim vlastima. Ubijen je u Beogradu 1917. odbivši da se pokaje i izjavi vernost caru („Ja sam srpski oficir“ [8]). Kasnije, na Solunskom frontu, naročito u Odredu vojvode Vuka, tokom osvajanja Kajmakčalana, izginulo je dosta dobrovoljaca Južnih Slovena, mahom Slovenaca.

I u bitkama na Drini i Kolubari, takođe, bilo je poginulih među dobrovoljcima slovenskih nacija (npr. bolničarka, Ruskinja Darija Aleksandrovna Korovkina iz Petrograda [4].

Slovenka Antonija Javornik –„Natalija Bjelajac“ kao bolničarka učestvovala je u oba balkanska rata, u toku novoga rata je u činu narednika nastavila svoju samarićansku službu i bila ranjena (12 rana!), dobivši „Karađorđevu zvezdu sa mačevima“. Posle ratova, do smrti je živela u Beogradu [9].

Od boraca, najveći broj dobrovoljaca iz početnog ratnog perioda, poreklom pretežno Srba iz susedne BiH, bio je raspoređen u tzv. komitske (četničke) odrede koji su bili namenjeni vršenju gerilskih akcija u neprijateljskoj pozadini. U julu mesecu 1914. od njih su bila obrazovana dva takva odreda, nešto kasnije još dva (Zlatiborski, Jadarski, Rudnički i Gornjački), u njima su se našli zajedno dobrovoljci iz Austrougarske i Srbi dobrovoljci iz Srbije i Crne Gore [9]. Bilo je među njima i pripadnika drugih slovenskih naroda. Veliki broj ih je poginuo ili je stradao mučeničkom smrću posle zarobljavanja.

Stvaranje dobrovoljačkih jedinica u Rusiji, njihovo borbeno angažovanje i sanitet – Srpski dobrovoljački odred i Prva srpska dobrovoljačka divizija (1915–1916)

U trenutku bugarskog napada na Srbiju, kada je prebacivanje dobrovoljaca Dunavom prekinuto, u Odesi prema pisanju konzula Marka Cemovića, zateklo se njih 36. Kako su dobovoljci nastavili da dolaze, konzul i tamo prisutna srpska vojna lica krajem 1915. godine, kada je taj broj porastao na 11 oficira i 560 podoficira i vojnika, obrazovali su Srpski dobrovoljački odred i njegov mali sanitet od jednog lekara (dr Stevan Poljak) i jednog medicinara (Milan Mirković) [2].

U međuvremenu je i stav ruske Vlade izmenjen, čemu je odlučujuće doprinela podrška ruskog Cara Nikole II. Time je omogućeno sistematsko upućivanje dobrovoljaca u Odesu, te je početkom 1916. godine, uz dogovor sa srpskom vladom, započeto sa obrazovanjem pukova iz kojih će do kraja proleća nastati Prva srpska dobrovoljačka divizija sa četiri pešadijska puka, artiljerijom, konjicom i sanitetom. Njeno ratno ustrojstvo bilo je po ruskim propisima i sa ruskom opremom. Divizija je bila podeljena na dve brigade, svaka sa dva puka i jednom poljskom bolnicom. Na nivou divizije obrazovana je bolničarska četa koja je u vreme borbi formirala 2–3 zavojišta, svaki puk je dobio pukovskog lekara i pukovsko previjalište, a svaki od tri bataljona po jednog mlađeg lekara ili medicinara i felčera. Pitanje finansiranja rešeno je posebnim sporazumom između ruske i srpske vlade [1, 3, 5].

Ovako formirana divizija brojala je oko 450 oficira i 18 500 podoficira i vojnika. Konjica i artiljerija su bile ruske, dok je sanitet bio mešanog sastava (nedostajući broj popunili su ruski kadrovi). Kako su se ti kadrovi mobilisali uglavnom sa teritorije Odeskog vojnog okruga gde je jevrejsko stanovništvo bilo brojno, to je i među ruskim sanitetskim kadrovima dodeljenim diviziji bilo dosta Jevreja. Sa Krfa srpska vlada u prvoj partiji poslala je 170 vojnih lica, najvećim delom oficira za rukovodeći kadar divizije, ostali oficiri su bili dobrovoljci, a u sanitetu, artiljeriji i konjici preovlađivali su Rusi. U jesen 1916. poslati su oficiri za štab Korpusa i novu, 2. diviziju čije je formiranje bilo u toku [1]. Kasnije je bilo još pojedinačnih slanja sve do 1918. godine radi podrške zarobljenicima sa teritorija jugoslovenskih zemalja.

I divizija u Dobrudži borila se u sastavu 47 posebnog korpusa, u kome su, pored nje, bile ruske 61. pešadijska i 3. konjička divizija. Dve rumunske divizije su bile samostalne, u taktičkoj vezi sa rusko-srpskim korpusom [1, 3, 5, 10]. Tokom skoro dvomesečnih borbi, u jesen 1916. poginulo je ili umrlo od rana 42 oficira i 718 podoficira i vojnika, ranjeno je 203 oficira i 6 047 podoficira i vojnika, što ilustruje intenzitet borbi, dok je „nestalih“ (ranjenici evakuisani ruskim ili ru-

munskim sanitetom, ostali na bojnom polju iza neprijateljskih linija, zarobljeni ili pobegli) bilo 1 321 podoficir i vojnik. Ukupni gubici divizije iznosili su 8 086 vojnika, podoficira i oficira [1, 2, 5].

Srpski dobrovoljački korpus (kasnije Dobrovoljački korpus Srba, Hrvata i Slovenaca), 1916–1917

Junaštvo i borbeni uspesi divizije ispoljeni tokom krvavih okršaja ostali su nenagrađeni pobedom zbog lošeg vođenja ruskog korpusa kome je divizija pripadala i zbog katastrofalno lošeg učinka rumunskih trupa koje su samo predajom tvrđave Tutrakan na Dunavu izgubile skoro 30 000 vojnika i 100 topova, u potpunosti kompromitujući stabilnost fronta, što se i u kasnijim borbama ponovilo, vodeći gubitku skoro cele teritorije Dobrudže.

Hrabro držanje Srpske 1. divizije doprinelo je odluci, donetoj krajem meseca avgusta 1916. u ruskoj Stavci da se obrazuje nova, 2. dobrovoljačka divizija iz ljudstva Dopunskog bataljona i novih kontingenata vojnika i oficira iz logora u kojima su se nalazili slovenski ratni zarobljenici. Najveći takav logor nalazio se u mestu Darnice kod Kijeva [2, 3].

Tom prilikom, međutim, učinjena je greška koja će u mnogome iskomplikovati odnose ne samo u toj diviziji koja je bila u fazi formiranja, već i među ljudstvom Prve, izmožđenoj kako teškim gubicima u borbama kroz koje su prošli, tako i novim političkim i personalnim problemima koji su se u njoj pojavili (disidenti).

Pojava disidentstva imala je više uzroka: snažna austrougarska propaganda i pretnje upućene dobrovoljcima i njihovim porodicama, uvođenje revolucionarnih organa u jedinice divizije u vidu radničko-soldatskih sovjeta i njihovih deputata i, uopšte, nesrećni događaji koji su od februara 1917. godine revolucionisali ruski narod. Tome treba dodati i nastale unutrašnje probleme: razlike u shvatanju namene dobrovoljačkog pokreta i njegovih ciljeva, razlike u pogledu organizovanja buduće države (integralni Jugosloveni, federalisti, unitaristi), razlike u platama oficira-dobrovoljaca i oficira poslatih sa Krfa (prve finansirala ruska, a druge srpska vlada), pitanje primanja srpskog državljanstva, pitanje imena Korpusa, sprovođenje stroge vojničke discipline bez rasprave o političkim pitanjima zagovarane od srpskih oficira pridošlih sa Krfa za razliku od oficira-dobrovoljaca, pitanje fizičkog kažnjavanja vojnika (za bivše austrougarske vojnike nepoznate pojave), uzbuna među banatskim Srbima i istarskim Slovencima posle saznanja da su saveznici obećali Banat Rumuniji, a Istru Italiji. Većina ovih problema su rešeni, ali je „zao duh“ bio posejan, bilo je potrebno vreme da se među mladim i temperamentnim ljudima duhovi smire, a naneta šteta delimično sanira [2, 3].

Iako je disidentstvom naneta šteta Korpusu, ti su mladi ljudi u najvećem broju čista srca pristupili dobrovoljaštvu, mnogi od njih su prošli sa Prvom divizijom teška borbena iskušenja u Dobrudži, u njima se istakli, bili ranjavani i odlikovani, ali su se u kasnijim raspravama razočarali, napuštajući Korpus i prelazeći u redove ruske vojske da nastave borbu sa neprijateljem. Kasnije će se deo njih vratiti u Korpus, a dobar deo se naći u sibirskim jugoslovenskim pukovima, dok će neki stupiti u jedinice „crvenih“ ili „belih“, prolivajući svoju krv i dajući živote u košmaru ruskog građanskog rata. Ne može se, međutim, negirati da se među njima nisu nalazili i ubačeni zlonamerni elementi, čak i špijuni, oni su najčešće vrlo brzo identifikovani i udaljeni. Sa druge strane, i neki od poslatih srpskih oficira nisu umeli da se snađu u za njih neuobičajenoj situaciji što je, takođe, doprinosilo razvoju zategnutih odnosa. Novopridošli komandant korpusa, đeneral Mihailo Živković svojim taktičkim ponašanjem, naročito u vreme uvođenja radničko-soldatskih odbora u jedinicama i pojačanog uticaja revolucionarnih previranja u Rusiji, uticao je na smirivanje i stabilizovanje međuljudskih odnosa unutar Korpusa.

Najveća greška, ipak, učinjena je slanjem u Korpus ne samo pravih dobrovoljaca, već i svih ostalih zarobljenika iz redova Južnih Slovena, koji su sami sebe nazvali „silovoljci“. To je učinjeno na zahtev predstavnika „Jugoslovenskog odbora“ u Rusiji, koji je želeo što veći broj svojih sunarodnika u Korpusu i pogrešnom procenom posledica ovoga čina u ruskoj Stavci. Svojim ponašanjem i odbijanjem vršenja vojničkih zadataka, često i nasilnim radnjama, pa i pobunom koja je koštala 13 izgubljenih života, oni su umnogome zatrovali odnose, od čega će se Korpus izlečiti tek kada svi nezadovoljnici po ličnom opredeljenju budu otpušteni i vraćeni u zarobljeničke logore. Odlaskom disidenata i „silovoljaca“, Korpus je ponovo postao po duhu jedinstvena borbena jedinica vođena zajedničkom željom da svojim učešćem u borbenim operacijama na Solunskom frontu doprinese oslobađanju Srbije i ostalih južnoslovenskih pokrajina i njihovom ujedinjenju u zajedničku državu.

U toku svih tih događanja izvršena je promena imena u „Dobrovoljački korpus Srba Hrvata i Slovenaca“, kako bi se izašlo u susret zahtevima dobrovoljaca.

Ipak, time je Korpus izgubio veliki broj vojnika i oficira, te je sa 45 000, koliko je brojao na svome vrhuncu, spao na oko 18 000 [2], pri čemu je njegov sastav postao dominantno srpski u svim segmentima, što je vidljivo iz statističkog pregleda ljudstva na dan 30. maja 1917. koji je dao dr Ante Mandić delegat „Jugoslavenskog odbora“ u Rusiji, u svome pismu centrali odbora (tabela 1) [2].

Reorganizacija Korpusa (povratak u Solun)

U leto 1917. godine, kada se očekivalo da proslavljena 1. divizija ponovo krene na front izvršeno je reorganizovanje Korpusa ukidanjem 2. divizije iz čijeg je sastava popunjena Prva divizija do punog borbenog brojnog stanja, dok je od ostatka obrazovan Rezervni puk i pojačan Dopunski bataljon do 12 četa [11, 12]. Za popunu 1. divizije upućeno je 108 oficira i 3 510 podoficira i vojnika, u Rezervni puk 145 oficira i 2 558 podoficira i vojnika, dok je ostatak pripojen Dopunskom bataljonu. Zvanično je Druga divizija prestala da postoji na dan 17. jula 1917. godine [13].

Ruska revolucionarna vlada želela je da Prvu diviziju, kao snažnu i proverenu jedinicu, upotrebi na svome jugozapadnom frontu u sastavu Šeste ruske armije u njenoj strateškoj rezervi, u stvari kao potporu poljuljanim redovima ruskih vojnika. Iako je srpska vlada u početku pristala, snažni otpor rukovodstva i ljudstva u Korpusu učinio je da se takvo reše-

Tabela 1

Sastav Dobrovoljačkog korpusa Srba, Hrvata i Slovenaca

Narodnost	Oficiri	Podoficiri i vojnici	Svega
Srbi	411	16 562	16 973
Hrvati	98	668	766
Slovenci	39	243	282
Česi†	98	90	188
Ostali	8	62	70
Ukupno*	654	17 625	18 179

*** Od ukupnog broja na Dopunski bataljon otpadalo je 2 165 (81 oficir i 2 084 podoficira i vojnika); Rusa u Korpusu bilo je ukupno 1 972, od toga 125 oficira i 1 847 podoficira i vojnika); †Česi oficiri, za razliku od vojnika, najvećim delom nisu hteli da napuste Korpus i pređu u jedinice češke vojske koja se stvarala u Rusiji**

nje povuče i da se dobrovoljci spasu iz haosa koji je već počeo da se ocrtava u Rusiji. Uprkos otporima koji su postojali, postignut je dogovor o evakuaciji dobrovoljačkih jedinica na Solunski front.

U tri velike grupe sa po nekoliko ešalona izvršeno je prebacivanje oficira, podoficira i vojnika, koji su u Solun pristizali sledećim redom: 7.decembra 1917. godine štab Korpusa i Rezervni puk, 145 oficira i 2 518 vojnika, koji su evakuisani preko Arhangelska; tokom januara 1918. iskrcani su 1. i 2. puk, evakuisani, takođe, preko Arhangelska, sa njima je evakuisana druga poljska bolnica, bez njenog komandira kapetana dr Dragutina Kostića, koji je ostao u Rusiji radi lečenja (kasnije se pridružio „Puku SHS majora Blagotića"), a bolnicu je primio češki dobrovoljac kapetan 2. klase dr Karel Freja – „Dragutin". Sa njima je evakuisana i bolnica „Škotskih žena" zajedno sa svojom već smrtno bolesnom dr Elzi Inglis i gospođom Evelinom Haverfild; 29. marta 1918. prispeo je 3. puk, a 1. maja 1918, 4. puk. Njihovo putovanje bila je prava avantura, jer su, zapavši u revolucionarne bitke između crvenih i belih, skrenuti preko Sibira do Dalekog istoka, odakle su preko Singapura i Sueckog kanala prispeli u Solun da se pridruže Srpskoj vojsci već opasno proređenih redova. Sa njima je putovala Prva poljska bolnica 1. divizije sa svojim komandirom kapetanom 1. klase dr Vladimirom Stanojevićem i ostalim osobljem.

Sa ovim trupama pošao je i izvestan broj ruskih oficira, među njima i neki lekari koji su služili u Korpusu i sa njime prošli borbeni put na frontu (dr Lev Polivec, dr Aleksej Šepelj, dr Konstantin Kržiškovski, dr Roman Strutinski). Oni su na Solunskom frontu raspoređeni u srpske pukove [4], osim dr Šepelja koji je u međuvremenu umro [3]. Sa njima je stigla i grupa čeških oficira i medicinara koji će nešto kasnije većinom preći u novoformirane češke jedinice u Francuskoj.

Ukupno, u Solun je pristiglo 553 oficira i vojnih činovnika i 11 385 podoficira, kaplara i vojnika (usput je u Engleskoj i Francuskoj ostalo nekoliko stotina obolelih oficira i vojnika). Pristigli su raspoređeni u srpske pukove, popunivši njihove proređene redove za veliku ofanzivu koja se uveliko spremala.

Zaostali dobrovoljci i njihov sanitet u Rusiji, 1917-1919 – borba za opstanak i povratak u zemlju

Jače delove Dopunskog bataljona (oko 1 000 vojnika) zadržale su u Rusiji komande savezničkih trupa, u Arhangelsku, Kandalakši i Murmansku. Te su jedinice, pod savezničkom komandom, bile zadržane radi čuvanja pristanišnih postrojenja, ali su, nažalost, bile upletene i u ruske međusobne oružane sukobe. Mala sanitetska ekipa u Kandalakši (tek diplomirani u Moskvi dr Nikola Kešeljević i narednik-medicinar Valentin Meršol), pored toga, morala je da se bori i sa epidemijama (grip) koje su vladale i ratnim povredama [3]. U novu državu SHS vratili su se tek posle završetka rata.

U beskrajima Rusije ostali su, rasuti po bolnicama i oporavilištima, ranjeni pripadnici. I. divizije, bivši pripadnici Korpusa (disidenti) i mnogo zarobljenih pripadnika jugoslovenskih naroda po logorima koji su raspušteni pod vihorima nadolazeće revolucije. O njima je srpska vlada preko svoga poslanstva i konzularnog delegata pokušavala da vodi aktivnu brigu i da ih organizuje uz saradnju delegacije Jugoslavenskog odbora, u kojoj su se, pored drugih, od medicinara nalazili kapetan dr Milivoje Jambrišak i narednik, junak sa Kajmakčalana u Odredu vojvode Vuka, Lovro Klemenčič-„Dušan", prvi Hrvat, a drugi Slovenac.

Od te amorfne mase Jugoslovena stvorena su dva jaka puka: u Čeljabinsku od prezdravelih vojnika iz „Slabosilne komande" Korpusa, zaostalih delova Dopunskog bataljona i naknadno prikupljenih vojnika „Puk Srba, Hrvata i Slovenaca majora (Mateje) Blagotića". Njihovi lekari bili su kapetan 2. klase dr Dragutin Kostić, zaostao od Korpusa zbog lečenja i Crnogorac dr Nikola Petrović, diplomac Vojnomedicinske akademije u Petrogradu [14]; u Tomsku „Prvi jugoslovenski puk Matija Gubec", od bivših pripadnika Dobrovoljačkog korpusa (disidenti) i ratnih zarobljenika iz raspuštenih logora. Pukovski sanitet činili su Slovenci „major" (raniji poručnik DK) dr Božidar Fajdiga i studenti medicine Tone Lovšin i Josip Erat [3].

Oba ova puka, prvi kasnije, a drugi od početka, bili su u sastavu čehoslovačkih trupa i pod francuskom komandom i bili su silom prilika upleteni u građanski rat boreći se protiv boljševika. Posle velikih peripetija, preko Dalekog istoka vratili su se savezničkim konvojima zajedno sa Čehoslovacima u domovinu tek polovinom 1920. godine [3, 14].

Personalni sastav dobrovoljačkih jedinica, 1915–1917 – medicinski kadar

Već u periodu reorganizacije ostataka Srpske vojske na Krfu, u februaru 1916. godine, na vesti iz Rusije o prilivu dobrovoljaca iz redova slovenskih zarobljenika i obrazovanju već brojno snažnog odreda u Odesi pod komandom tamo prisutnih srpskih oficira, srpska Vlada donela je odluku o slanju potrebnog broja vojnih lica radi obrazovanja dobrovoljačke

divizije. Tako je aktima Ministarstva vojnog i Vrhovne komande tokom februara određena i grupa sanitetskih oficira, medicinara i veterinara, koja je upućena u Rusiju [15–18]. Za tom grupom sledile su u jesen 1916. druge, u vreme obrazovanja Druge divizije i korpusnog štaba, a kasnije je bilo i pojedinačnih slanja sve dok su u Rusiji boravili ratni zarobljenici iz redova jugoslovenskih naroda [3, 4].

Pored srpskih vojnih lica prispelih sa Krfa ili Soluna, sanitet divizije, organizovan po propisima Ruske vojske, sačinjavali su dobrovoljci iz redova austrijskih Srba, Hrvata i Slovenaca, Česi, Slovaci, po koji Poljak i Ukrajinac (Rusin), kao i dodeljeno rusko sanitetsko osoblje radi dopune nedostajućih kadrova.

Za Prvom divizijom ubrzo je iz Velike Britanije stigla poljska bolnica humanitarne organizacije „Škotskih žena" pod rukovodstvom velike dobrotvorke srpskog naroda, dr Elzi Inglis, uključivši se u sastav saniteta usred borbi u Dobrudži u okolini Medžidije, kada je počela da prima srpske ranjenike. Dr Elsi Inglis će kasnije, 1917. godine, iako već ophrvana smrtonosnom bolešću, odbiti da napusti Rusiju „sve dok se to ne dozvoli i hrabrim srpskim jedinicama", izvršivši snažan pritisak na britansku vladu i, indirektno, na ruske demokratsko-revolucionarne vlasti. Uz njenu bolnicu stigla je i sanitetska kolona mis Eveline Haverfild sa 16 malih „fordova". Pored ove bolnice, u Medžidiji se nalazila i humanitarna misija britanskog Crvenog Krsta sa bolnicom na čijem se čelu nalazio, Srbima iz 1914. dobro poznati, dr Džejms Beri. Svi oni će u završnim bitkama u Dobrudži posle evakuacije ruskih bolnica iz Medžidije, zajedno sa srpskim lazaretima pružati medicinsku pomoć ne samo srpskim, već i ruskim i rumunskim ranjenicima. *

Rasporedi sanitetskog osoblja (Ordre de bataille) od prvih dana do evakuacije na Solunski front (1915–1917)

Period prikupljanja dobrovoljaca u Odesi (novembar 1915 – april 1916)

Prvi dokument u kome se navode imena medicinskih lica iz grupe dobrovoljaca koji su u Odesi zaostali posle prekida veze Dunavom između Rusije i Srbije usled neprijateljske ofanzive, nosi datum 11. novembra 1915. godine. To je naredba br. 5 komandanta Srpskog dobrovoljačkog odreda u Odesi [13], kojom se za medicinsko osoblje odreda imenuju: lekar dr Stepan Poljak i medicinar Milan Mirković. Obojica će, prvi kao poručnik, drugi kao potporučnik Srpske vojske biti do kraja u sastavu divizije (korpusa) i posle evakuacije pridružiće se njenim jedinicama na Solunskom frontu.

Kako je uskoro usledio priliv većeg broja dobrovoljaca, a u međuvremenu su pristigli i srpski kadrovi poslati sa Krfa, pristupilo se obrazovanju većih jedinica (pukova), od kojih će nastati divizija. Srpska vlada i Vrhovna komanda su se trudile da u Rusiju pošalju prvenstveno kadrove koji su školovani u Rusiji. Pukovi su stvarani sukcesivno, kako su dobrovoljci pristizali

Period formiranja pukova i 1. divizije (april – juli 1916)

Naredbama o upućivanju sanitetskih kadrova u Rusiju [15–18] lekari i medicinari su pristizali u nekoliko grupa, zaobilaznim putem, morem i kopnom, iz Soluna preko Engleske i Skandinavije. Tako su u periodu proleće–jesen 1916. godine pristigli pukovnik dr Milan Žerajić, potpukovnici dr Dragoslav Popović i dr Mihailo Veličković, major dr Momčilo Ivković, kapetani 1. klase dr Vladimir Stanojević i dr Božidar Janković, kapetani 2. klase dr Dragutin Kostić, dr Stojan

*Dokumenti obe divizije i korpusa u celini su sačuvani u Vojnom Arhivu Vojske Srbije u Beogradu, popisnik 10, izabrani deo građe svojevremeno je objavio dr Nikola Popović [2].

Sanitet I divizije prošao je celu kampanju u Dobrudži o čemu je ostalo, pored sačuvanog arhiva, i dosta pisanih svedočanstava u vidu dnevnika, uspomena i stručnih dela na tu temu. Navešćemo neka koja zbog se svoje autentičnosti mogu svrstati po svojoj vrednosti odmah uz primarnu građu:

Slovenci koji su učestvovali u svim ovim zbivanjima u Rusiji dali su neobično iscrpan zbornik „Kladivarji (kovači) Jugoslavije", u okviru koga je, pored mnoštva priloga posthumno objavljen dobrudžanski dnevnik rano preminulog medicinara potporučnika Milka Gnezde o tim herojskim danima dobrovoljaca [3]. Sličan ovome je i dnevnik medicinara Milutina Velimirovića [19, 20] koji se čuva u SANU. U „Spomenici I. SDD" [1], kao i u zborniku „Golgota i Vaskrs Srbije" [21] dati su realistični opisi ratnih dejstava tokom kampanje u Dobrudži. Tu su i ratne beleške dr Vlade Stanojevića [4] i njima slične uspomene dr Rudolfa Trušnoviča [10].

Kada govorimo o sanitetu kao organizacionom delu jedne borbene trupe u vreme ratnih i revolucionarnih zbivanja u Rusiji, sasvim je prirodno da se tokom 1916. i 1917. godine personalni sastav sanitetskih jedinica stalno menjao, prvo zbog borbenih gubitaka /obolevanja usled teških uslova, potom zbog službenih premeštaja ili napuštanja službe usled političkih neslaganja (disidentski pokret), a bilo je i prelaska u rusku revolucionarnu „Crvenu Armiju" ili u jedinice „Belih", ili odlazaka u češke jedinice koje su se uveliko obrazovale širom Rusije (ipak najveći broj oficira Čeha i Slovaka, koji u se borili sa 1. divizijom u Dobrudži, njih 98, ostali su sa svojim ratnim drugovima).

Sačuvani službeni izveštaji koji se odnose na personalne sastave su nažalost često manjkavi, jer su u njima navođene samo osobe sa oficirskim činovima, što kod medicinara nije uvek bio slučaj. Zapravo, medicinari poslati sa Krfa nisu ni imali oficirske, već u najboljem slučaju podoficirske činove, dok je većina austrijskih medicinara, prošavši po maturi kroz kadetske ili škole jednogodišnjih dobrovoljaca taj čin stekla, što im je pri stupanju u dobrovoljačke jedinice priznato. Svi ostali medicinari bez čina, bilo da su došli sa Krfa ili (retko) dobrovoljci, nisu u ratnim rasporedima (*Ordre de bataille*) najčešće pojedinačno, uopšte, navođeni [4].

Kod ruskih članova saniteta u izveštajima vrlo često nije navođen ni čin niti lično i očevo ime, već samo porodično. Oni koji su bili na oficirskoj dužnosti (lekari) redovno su navođeni, što sa ostalima („feldšeri") nije bio slučaj, pa je njihov identitet u najvećem broju slučajeva ostao nepoznat, iako je i iz sistematizacije i iz memoarskih dela bilo jasno da ih je bilo mnogo više.

Na rasporedima najčešće nema datuma kada su pisani, a neki su i bez označenog broja iz delovodnog protokola, pa je tek upoređivanjem bilo moguće njihovo vremensko lociranje.

Svi ti nedostaci su dosta otežali rad na rekonstrukciji sastava saniteta divizija i korpusa u periodu 1916–1917. godine. Neka eventualna kasnija istraživanja, uvidom u drugu građu koja je u Popisniku 10 pohranjena: rešenja o postavljenjima, premeštajima, unapređenjima, odlikovanjima i ostalim aktima koji se odnose na sanitet (periodični izveštaji i operativni dnevnici) omogućila bi da se do detalja izuči kretanje sanitetskog osoblja i njihov rad izrazi u statističkim brojevima, što smo mi samo delimično i u glavnim crtama mogli da učinimo.

Pored prevashodno vojničke građe iz Popisnika 10, 3 i 3a (građa Ministarstva vojnog i Vrhovne komande), postoji i druga, uglavnom politička, građa koja se nalazi u Arhivu Srbije (Arhiv Ministarstva spoljnih poslova) i u Arhivu Hrvatske akademije znanosti i umjetnosti u Zagrebu (Trumbićev arhiv), čija bi dokumenta koja se odnose na našu osnovnu temu (sanitet), takođe, bilo interesantno proučiti. Što se tiče saniteta, našu studiju smatramo nedovršenom, ali i u ovome obliku interesantnom za objavljivanje imajući u vidu činjenicu da je rad ratnih saniteta kod nas do sada istoriografski bio poprilično zanemaren. Nažalost, događaji u toku poslednje dekade XX veka ovakva istraživanja su veoma otežali uspostavljanjem novih granica među bivšim jugoslovenskim republikama, ali i doveli do gubitka interesovanja za ovu temu, pa je pitanje da li će se do nekih novih saznanja moći doći.

Stevanović, dr Karel-Dragutin Freja (Čeh), dr Fedor-Teodor Serebrenikov (Rus), poručnici dr Aleksej-Aleksa Šepelj i dr Apostol Hadži-Gligorije, medicinari Aleksandar Petrović, Svetozar Nikolajević, Milutin Velimirović, Milutin Đorić, Milan Matić, Savatije Mitrović, Mihailo Babović, Franjo Vuletić, Bogoljub Kurandić, i veterinar Mihailo Petrović.

Do proleća 1916. u Odesi se prikupilo preko 12 000 dobrovoljaca, pa je 16. aprila izdata naredba o obrazovanju I divizije, u okviru nje i saniteta [22].

Sanitet divizije obrazovao se postepeno, puk po puk. U Štabu divizije (došli sa Krfa) bili su referent saniteta, potpukovnik dr Dragoslav Popović; medicinar-ađutant referenta, Svetozar Nikolajević; higijeničar divizije, major dr Momčilo Ivković.

U Prvom puku*, koji je obrazovan 20. januara 1916, nalazili su se: v.d. trupnog lekara kapetan 1.klase dr Božidar Janković i medicinari, potporučnik Juraj–Đura Dekanić, potporučnik Dušan Krstić i Milan Matić.

Drugi puk, obrazovan 6. marta 1916, imao je sledeći sastav: v.d. trupnog lekara kapetan 2. klase dr Karel–Dragutin Freja i medicinare, potporučnika Milana Mirkovića, potporučnika Stojana Grgurova i Mihaila Babovića.

Treći puk, obrazovan 17. marta 1916. bio je bez određenog sanitetskog osoblja, dok je 4. puk (obrazovan 9. aprila1916) imao v.d. trupnog lekara dr Ivana Smuljskog (ruski lekar). U sastavu Dopunskog bataljona (obrazovan 14. juna 1916) nalazio se v.d. trupnog lekara potporučnik dr Stevan Poljak [23].

Priliv dobrovoljaca bio je veoma brz, tako da je popunjavanje redova divizije završeno do letnjih meseci. U svome punome sastavu divizija je brojala oko 18 500 ljudi, uključujući i pridodate ruske artiljerijske i konjičke jedinice i delove saniteta, a sastav njenog saniteta bio je sledeći:

Sanitet I divizije na dan 16. 7. 1916. (uoči odlaska na front u Dobrudžu u svome punom borbenom sastavu) imao je referenta saniteta (potpukovnik dr Dragoslav Popović); higijeničara divizije i člana regrutne komisije (major dr Momčilo Ivković) i medicinara u štabu (Svetozar Nikolajević) [24]. U sastavu Dopunskog bataljona bio je poručnik dr Božidar Fajdiga.

U 1. puku bio je kapetan 2. klase. dr Stojan Stevanović i medicinari potporučnik Đura Dekanić i potporučnik Dušan Krstić i Milan Matić; u 2. puku: lekar kapetan dr Fedor–Teodor Serebrenikov; medicinari potporučnik Milan Mirković, potporučnik Stojan Grgurov, potporučnik Josip Gazarek, potporučnik Franja Štrodel; u 3. puku: lekar poručnik dr Aleksej Šepelj; medicinari, potporučnik Milan Jovanović, potporučnik Josip Hebajn, potporučnik Milko Gnezda, potporučnik Bedžih Opletal i u 4. puku: potporučnik dr Bogoslav Bouček, potporučnik Sima Ilić i potporučnik Jože Ermenc.

Sastav 1. lazareta činili su: komandir, kapetan dr Vladimir Stanojević, vojni lekari Gelman i Godlevski, medicinari Aleksandar Petrović, Milan Matić i Milutin Đorić i apotekar, potporučnik Vladimir Šubčik, dok je 2. lazaret imao sledeći sastav: komandir kapetan dr Dragutin Kostić, vojni lekari dr Gomberg, dr Mutermilh, dr Lev Ivanovič Polivec; medicinari, potporučnik Josip Erat i Milutin Velimirović, te apotekar potporučnik Bedžih Koržinek.

Koliko nedostaju navođenja svih sanitetskih lica vidi se u primeru koji je dao u svojim uspomenama medicinar Milutin Velimirović [19], kada je naveo da su se u sastavu ovoga lazareta nalazili, pored gorenavedenih i 4 starija i 6 mlađih ruskih felčera, tj. po današnjoj terminologiji lekarskih pomoćnika) koji, kao podoficiri, nisu posebno navođeni.†

U sastavu Sanitetske čete bili su: komandir 1 klase dr Božidar Janković; hirurg, vojni lekar, dr Feliks Bergman i medicinar Mihailo Babović. U Dezinfekcionom odredu bio je vojni lekar dr Ivan Kolesničenko. Komandir osoblja bio je češki dobrovoljac kapetan Rudolf–Radovan Gajda, kasniji general Gajda, komandant čeških jedinica u Sibiru. Medicinar Velimirović u svojim uspomenama ga navodi kao komandira odreda za vreme borbi u Dobrudži [19].

Sanitet Ruskog artiljerijskog puka i Ruskog konjičkog odreda bio je sastavljen od ruskog medicinskog osoblja.

Rad saniteta Prve divizije tokom i neposredno posle borbenih dejstava u Dobrudži

Divizija je ukrcana u brodove 15–28. avgusta i transportovana do Černavode, gde je ušla u sastav Ruskog 47. korpusa pod komandom generala Zajončkovskog, zajedno sa Ruskim 61. pešadijskom i 3. konjičkom divizijom. Dve rumunske divizije su bile samostalne, u taktičkoj saradnji sa korpusom.

U toku septembra i oktobra divizija je učestvovala u pet teških bojeva, prvo nastupnim ka Dobriču kod Kara sinana, koji je divizija uspešno rešila, ali je zbog neuspeha krilnih

---

* U ovome puku za komandira 1. čete 1.bataljona bio je postavljen potporučnik Rudolf Trušnovič, istarski Slovenac iz okoline Gorice, inače student medicine IV godine, koji je izričito zahtevao da bude u borbenim redovima, a ne u sanitetu, koristeći činjenicu da je uoči studija završio kadetsku školu pešadijskog smera. U borbama u Dobrudži teško je ranjen i dugo bolovao u ruskim bolnicama. Kasnije je stupio u ruske nacionalne jedinice („bele“) zajedno sa pobratimom Hrvatom potporučnikom Ignjatom Frankom iz 3. puka. Frank je u borbama izgubio ruku, kasnije je u Donskoj armiji komandovao Kozačkim pukom u kome je Trušnovič bio jesaul. Frank je poginuo, a Trušnović je, ostavši u Rusiji, završio medicinu u Harkovu i u Jugoslaviju se vratio polovinom tridesetih godina [10], službujući u Sremu (Morović) i u Ministarstvu narodnog zdravlja u Beogradu. Emigrirao je 1944. u Nemačku i ubijen šezdesetih godina u Berlinu od sovjetskih agenata.

Slično je bilo i u 3. puku, gde je za vodnog oficira postavljen medicinar Nikola Šajatović, rodom iz Jezernice (Hrvatska), koji je 1. oktobra hrabro poginuo kod Hardalija na čelu svoga voda [19].

† Sastav jednog ruskog lazareta (poljske bolnice) bio je, prema dr V. Stanojeviću daleko bogatiji od Srpske poljske bolnice. Bio je sastavljen od medicinskog i nemedicinskog osoblja [25].

Medicinsko osoblje činili su: komandir (stariji lekar) – 1; ordinatori (lekari) stariji – 1; ordinatori (lekari) mlađi – 3; feldšeri stariji – 4; feldšeri mlađi – 6; apotekari stariji – 1; apotekari mlađi – 1; veterinari – 1; bolničari (nadzornik bolnice – 1; bolničari stariji – 5; bolničari mlađi – 20; služitelji – 24).

U sastavu nemedicinskog osoblja bili su: šef ekonomata – 1; njegov pomoćnik – 1; knjigovođa – 1; sveštenik – 1; crkvenjak – 1; pisar viši – 1; pisar niži – 4; majstori – 6; posilni – 9; komordžijski podoficir – 1; komordžije – 29.

Pored toga, bolnica je imala: 26 apotekarskih sanduka sa fiokama i pregradama, 8 dvokolica za apoteku i gazdinstvo, kola za konjsku vuču, pokretnu kuhinju, 49 konja za vuču i 4 jahaća konja. Od intendantskog materijala tu su bili posteljni, sobni, kuhinjski i ostali bolnički nameštaj, stolarski, obućarski i kovački alat, razni kancelariski materijal, uputstva, propisi i, na kraju, kasa sa dvostrukim zaključavanjem.

ruskih i rumunskih trupa morala da se povlači, i sledeća četiri odbranbena, kod Hardalija, Kokardže, Amzače i Endže-Mahale – Isakče. U ovim borbama koje su obilovale ranjenicima, najveći problem saniteta bio je nedostatak transportnih sredstava za evakuaciju ranjenika pod sopstvenom komandom, jer su sanitetske kolone bile koncentrisane na nivou korpusa, od koga su morale da budu tražene u slučaju potrebe (ovo iskustvo je korišćeno pri obrazovanju II divizije, pa je u njoj od početka obrazovana sanitetska kolona).

Naročito su bile teške poslednje borbe pošto su nemačko-bugarsko-turske snage probile rumunski deo fronta, zauzele Medžidiju, Černavodu i Konstancu, zapretivši opkoljavanjem celoj rusko-srpsko-rumunskoj grupaciji. Tada se evakuisalo svima dostupnim sredstvima u neopisivoj žurbi i haosu što je plastično opisano u dnevnicima medicinara Gnezde [3] i Velimirovića [19], a prema nemačkim izvorima [26] zarobljeno je 37 600 rusko-rumunsko-srpskih vojnika i podoficira i 400 oficira i zaplenjeno 400 topova i obilje drugog ratnog materijala.

Prvi lazaret je za vreme borbi primio i evakuisao 2 461 ranjenika i bolesnika. Za 2. lazaret, koji se diviziji nešto kasnije pridružio, nedostaju tačni podaci. O njemu u svojim beleškama Velimirović [19] navodi da su samo za tri dana borbi u reonu Medžidije imali 1 600 ranjenika.

Sanitet je tokom svih bitaka obradio 6 250 ranjenika sve tri vojske, a mnoge je ranjenike primio i susedni ruski sanitet iz 61. divizije, kojoj su tokom borbi privremeno dodeljivani bataljoni ili čak i ceo puk iz sastava srpskih jedinica. Tokom tih borbi „Bolnica škotskih žena“ dr Elzi Inglis tesno je sarađivala sa 1. poljskom bolnicom srpske divizije, u čijem sastavu je organizaciono i bila. U njenom susedstvu radila je i bolnica britanskog Crvenog krsta pod vođstvom, Srbima dobro poznatog, dr Džejmsa Berija. Britanske bolnice, izvanredno opremljene, pružale su pomoć, pretežno hiruršku, velikom broju (preko 1 000) srpskih ranjenika. Kada se među vojnicima pojavila kolera, bolnica dr Inglis je organizovala posebno „II odeljenje“ radi lečenja zaraženih bolesnika.

Gubici divizije u borbama, prema zvaničnom izveštaju koji je u Ismailiji podneo po povratku sa fronta komandant divizije pukovnik Hadžić bili su (tabela 2) [27, 28]:

Prvi izveštaj o brojnom stanju divizije, posle povratka u pozadinu, poslao je 14. novembra telegramom Vrhovnoj komandi srpski vojni ataše pri rumunskoj Vrhovnoj komandi, pukovnik Andonović [2, 29]. Prema toj depeši brojno stanje divizije iznosilo je (posle gubitka 53% njenog sastava u mrtvim, ranjenim i nestalim) 373 oficira i 9 047 vojnika. Poređenja radi, 61. ruska i 10. i 19. rumunska divizija izgubile su 70%, a 114. ruska divizija 90% svog sastava.

Pošto su prikupljeni oficiri i vojnici, koji su se tokom povlačenja odvojili od svojih jedinica, definitivno brojno stanje je na dan 18. novembra iznosilo 418 oficira i 10 735 podoficira i vojnika [30].

Divizija je bila smeštena u kantonmane sa sanitetom čiji je sastav posle svih događaja kroz koje je prošla i bez popune nedostajućih kadrova koji su se našli u „rasturu“ zbog bolesti ili nekih drugih razloga (službena ili privatna odsustvovanja, kursevi, nastavak studija koji je nekim studentima odobren) bio donekle izmenjen [31–33]. U štabu su se nalazili: referent saniteta, potpukovnik dr Dragoslav Popović; higijeničar i član regrutne komisije major dr Momčilo Ivković (od 16. 04. 1917. biće premešten u poslanstvo Srbije u Petrogradu) i ađutant med. Svetozar Nikolajević; u dopunskom bataljonu: kapetan 2 klase dr Stojan Stevanović; u 1. puku: puk. lekar poručnik dr Apostol Hadži-Gligorije i bataljonski lekar, pporučnik med. Dušan Krstić; u 2. puku bili su: puk. lekar kapetan 2.klase dr Teodor Serebrenikov i bataljonski lekari, pporučnik med. Milan Mirković, pporučnik med. Stojan Grgurov, pporučnik med. Franjo Štrodel, pporučnik med. Josip Gazarek (pri puku su se vodili kao odsutni dr Karel Freja i dr Danilo Vasiljenko); u 3. puku bili su: puk. lekar poručnik dr Aleksej Šepelj i bataljonski lekari, pporučnik med. Josip Hebajn, pporučnik med. Milko Gnezda, pporučnik med. Milan Jovanović; u 4. puku su se nalazili: vojni lekar dr Ivan Kolesničenko; bataljonski lekari, pporučnik med. Bogosav Bouček, pporučnik med. Karl Sadilek, pporučnik med. Rudolf Lehki, pporučnik med. Sima Ilić. U Bolničarskoj četi komandir je bio kapetan 1. klase dr Božidar Janković, a hirurg: dr Feliks Bergman. Sastav 1 lazareta činili su: komandir kapetan 2 klase dr Vladimir Stanojević, mlađi lekari, med. Aleksandar Petrović, med. Ludvik Godlevski, med. Gelman i apotekar, pporučnik Vladimir Šubčik, a sastav 2. lazareta: komandir kapetan 2 klase dr Dragutin Kostić i prikomandovan vojni lekar dr Leo Braškin (verovatno kao zamena dr Kostića koji će uskoro otići na bolovanje), te mlađi lekari, dr Gomberg, dr Luterman, dr Lev Ivanovič Polivec. Dezinfekcioni odred činili su: vojni lekar dr Ivan Kolesničenko i rusko osoblje, dok je u sastavu artiljerije i konjice bilo rusko osoblje.

Iz dnevnika ili beležaka neposrednih sanitetskih učesnika tokom ove kampanje (Stanojević, Velimirović, Gnezda)

Tabela 2

Gubici 1. divizije

Pripadnici divizije	Karasinan	Hardali	Kokardža	Amzača	Endže Mahale-Isakča	Ukupno
Oficiri	33	11	116	32	39	231
poginuli	7	2	19	5	2	35
ranjeni	26	9	97	27	36	195
nestali	–	–	–	–	1	1
Vojnici	1 360	694	4 234	1 180	1 528	8 996
poginuli	161	46	226	149	102	684
ranjeni	1 002	547	2 912	992	595	6 048
nestali	197	101	1 096	39	831	2 264

Prema prvom, nepotpunom izveštaju, među poginulima bilo je: 687 Srba, 13 Hrvata, 3 Slovenca i 3 Čeha, a među nestalima: 1 123 Srba, 19 Hrvata, 38 Slovenaca i 23 Čeha.

vidi se da je zbog njene specifičnosti rad saniteta bio tome prilagođen. Naime, sam teren (ravnica, delom bezvodna, naseljena većim delom primitivnim stanovništvom bugarskog ili turskog porekla) nametnuo je manevarski tip kampanje sa dosta brzih pokreta u oba pravca, a njena oštrina, koja se ogledala u vrlo krvavim i energičnim sukobima, povećala je dodatno značaj brzine reagovanja saniteta, tim pre što je nad velikom većinom vojnika i oficira lebdela stalna opasnost od zarobljavanja sa nesagledivim posledicama po njih i njihove porodice. Stoga, kao i u svim sličnim kampanjama, uloga saniteta najčešće se svodila na hitrost u prikupljanju ranjenika, ukazivanju najnužnije pomoći (imobilizacija povređenih udova, zaustavljanje životno ugrožavajućih krvavljenja i što brža evakuacija u pozadinu). Ni u jednom od tih zapisa nema opisa ozbiljnijih hirurških intervencija neposredno iza borbene linije, što se u borbama i u Srbiji i na Solunskom frontu i te kako često događalo.

Poseban problem je postojao u brzini evakuacije ranjenika, jer za razliku od uobičajenog u srpskoj vojsci, u kojoj je sanitetska kolona bila u sastavu divizijskog saniteta, ovde su po naređenju komandujućeg 47. korpusa, generala Zajončkovskog, sva transportna sredstva bila pod komandom Korpusa. I na raspolaganje podređenim jedinicama stavljana su na njihov zahtev, što je neminovno dovodilo do velikih problema. Iz navedenih svedočenja vide se napori da se ta organizaciona besmisao prevaziđe improvizacijama, najčešće stavljanjem ranjenika na sredstva koja su služila za prevoz opreme same sanitetske jedinice, pukovske ili bolničke. Naročito se taj problem ispoljavao prilikom brzih odstupanja kada je pretilo zarobljavanje, pa se u pojedinim trenucima sve pretvaralo u haotično stanje i paniku, posebno u završnom periodu kampanje.

U principu srpski sanitet je svoje ranjenike predavao ruskom i rumunskom sanitetu radi dalje evakuacije i smeštaja u bolnicu, on nije posedovao sopstvene bolnice u pozadini. To je otežalo kako tačno evidentiranje svih ranjenika i bolesnika, tako i njihovo dalje praćenje. Za otpuštene ranjenike kasnije je stvorena tzv. „Slabosilna komanda“ (komanda lakih ranjenika i rekonvalescenata) u kojoj su prikupljani svi otpušteni iz bolnica kako bi bili pod kontrolom srpskih vojnih vlasti do osposobljavanja za vojnu službu.

Kasnije se pojavio i problem vojnonesposobnih invalida otpuštenih iz bolnica koji su ostali bez obezbeđenog smeštaja i ishrane, sa ružnim slikama po ulicama Odese kojima su lutali.

Sanitet Dobrovoljačkog korpusa

Po odluci ruske Stavke i naređenju srpskog Ministarstva vojnog započeto je sa formiranjem 2. divizije iz Dopunskog bataljona i iz novih grupa zarobljenika koji su stizali iz zarobljeničkih logora (Rusija je već vrlo brzo počela da odvaja slovenske zarobljenike od ostalih, koristeći ih za poverljive radove iza fronta, ili koncentrišući ih na nekoliko mesta, najviše u logoru Darnice kod Kijeva) još u vreme dok se 1. divizija borila u Dobrudži. Obrazovanje novih jedinica iz Dopunskog bataljona omelo je popunu borbenih gubitaka 1. divizije do uspostavljanja njenog punog brojnog stanja i dovelo do problema u odnosima između komandi Korpusa i 1. divizije. Tako je ona, posle gubitka preko 8 000 ljudi, samo delimično dopunjena sa 45 oficira i oko 2 400 podoficira i vojnika.

Kada je završeno formiranje, sastav saniteta korpusnog štaba i 2. divizije bio je sledeći: štab Dobrovoljačkog korpusa (sanitetski deo) [34]: načelnik saniteta, pukovnik dr Milan Žerajić; ađutant načelnika, potporučnik medicinar Zdravko Jeftanović; na radu u odeljenju kapetan 2. klase dr Milivoj Jambrišak (do početka 1917. kada je prešao u Jugoslavenski odbor u Rusiji, gde je dalje bio vrlo aktivan u agitaciji po zarobljeničkim logorima); lekar intendantske službe korpusa, dr Justin Smuljski; komandir sanitarno higijenskog i dezinfekcionog odreda, dr Ivan Vološinov; mlađi lekar u odredu, bakteriolog Josif Braclavski; hemičar i bakteriolog dr Gornik (iz ruskog Crvenog krsta); 2. divizija, prva formacija [35]: referent saniteta, potpukovnik dr Mihailo Veličković; komandir divizijskog zavojišta, koleški savetnik dr Julij Zeland; zamenik komandira divizijskog zavojišta, titularni savetnik dr Lev Genjaš; lekar poručnik dr Stevan Poljak; vojni lekar Emanuel Guberman; divizijska sanitetska kolona – stariji lekar Leonid I. Rubanovič; sastav lazareta – komandir kapetan 2. klase dr Stojan Stevanović; lekar ordinator dr Aleksej Boldirev; medicinar (dr?) Isak Kenski; medicinar (vrač?) Fedor Savčenko; medicinar (vrač?) Avram Čerkaski; medicinar (vrač) Parižer; Dezinfekcioni odred – komandir pukovnik stariji vrač dr Dimitrije Uvarov; 5. pešadijski puk – komandir sanit. odeljenja kapetan vrač Leonid Maksimovič; lekar poručnik dr Roman Strutinski; medicinar potporučnik Konstantin Jacenko; medicinar potporučnik Jaroslav Kohoušek; 6. pešadijski puk – komandir sanit. odeljenja stariji vrač Josif V.Citiljevski; medicinar potporučnik Franja Vuletić (prebegao 1914. u Srbiju, sa Krfa poslat u Rusiju); medicinar potporučnik Semen Ž. Judelevič; medicinar potporučnik Mihail Davidov; 7. pešadijski puk – komandir vojni lekar dr Stevan Voisovski; medicinar potporučnik Vojislav Veselinović; medicinar potporučnik Branko Kešanski; medicinar Nikolaj Skvorcev; 8. pešadijski puk – komandir dr Nikolaj Zernov; medicinar Sergej Grabovski; medicinar Emanuel Solovjev; medicinar potporučnik Adolf Šebesta.

Iz prikazanog sastava zapaža se da je u ovoj diviziji bilo mnogo više sanitetskih oficira iz redova ruske vojske, novopridošlih dobrovoljaca je bilo manje nego ranije, pa je popuna nedostajućih kadrova rešavana na taj način.

Dopunski bataljon Korpusa (12 četa) činili su [36]: komandir saniteta – kapetan 2. klase dr Karel–Dragutin Freja; medicinar potporučnik Karel Sadilek; prikomandovani lekari – medicinar potporučnik Jaroslav Kohoušek; medicinar potporučnik Đura Dekanić; medicinar potporučnik Isa Nedeljkov; medicinar potporučnik Sima Ilić; medicinar potporučnik Tone Eržen; apotekar Nikola Gajić.

Sastav komande lakih ranjenika Korpusa („Slabosilna komanda“) činili su [37]:

1. komanda – komandir poručnik dr Božidar Fajdiga; medicinar narednik Valentin Meršol; medicinar Mihailo Babović; apotekar potporučnik Andra Posavac.

2. komanda – lekar med. poručnik dr Bogosav Bouček (ostalo osoblje nije navedeno).

Tokom svog postojanja 2. divizija nije imala sreće jer je još od početka trpela unutrašnje potrese usled pogrešne odluke ruskih vlasti da se u nju i silom upućuju zarobljenici južnoslovenskog porekla. Neprekidno odlaženje i dolaženje novih regruta dovodilo je i do promena u sastavu saniteta, koji ni sam nije bio imun na ova kretanja, pa je i među sanitetskim oficirima postojao disidentski pokret o čemu je već bilo reči. U revolucionarnoj agitaciji među sanitetskim osobljem posebno su se isticali, od lekara dr Bergman i dr Kolesničenko, a od ostalih najvećim delom feldšeri.

U daljem tekstu biće navedene samo one promene u sanitetu koje su se tokom tih događaja desile (prema pronađenim dokumentima u korpusnom arhivu): 2. divizija, nova formacija saniteta [38] – referent saniteta ppukovnik dr Mihailo Veličković; 5. puk – pukovski lekar kapetan 2. kl. dr Karel-Dragutin Freja; bataljonski lekari: poručnik dr Stevan Poljak, potporučnik med. Franjo Vuletić i potporučnik Konstantin Nikolajevič Jacenko; 6. puk – pukovski lekar poručnik dr Br. Košnickoj, bataljonski lekari: potporučnik med. Vojislav Veselinović, potporučnik med. Adolf Šebesta, vojni lekar Judelevič, vojni lekar Mih. Davidov; 7. puk – pukovski lekar, potporučnik vrač Ivan Dubnovski; bataljonski lekari: potporučnik med. Branko Kešanski, potporučnik med. Franc Tobijaš i potporučnik (med.?) Nikolaj Skvorcev; 8. puk – pukovski lekar dr Sergej Grabovski (u spisku označen kao bataljonski); bataljonski lekari: (med.?vrač?) Emanuil Solovjev, potporučnik med. Isa Nedeljkov, potporučnik med. Golovjev; Bolničarska četa – komandir vojni lekar dr Julije Zeland; lekari: vojni lekar dr Fedor Savčenko i vojni lekar Emanuel Guberman; 1. lazaret – komandir kapetan 2 klase dr Karel-Dragutin Freja i mlađi lekari: Grešovič, Čerkaski, Kenski, Parižer; 2. lazaret – komandir vojni lekar vrač Eršov i mlađi lekari: Arhisovič, Pres, Zabjelina; Dezinfekcioni odred – komandir pukovnik dr Dimitrije Uvarov .

Poslednje izmene desile su se u 2. brigadi 2. divizije 9. juna 1917, mesec dana uoči rasformiranja divizije [39]: 5. puk – pukovski lekar kapetan dr Leonid Maksimovič; bataljonski lekari: vojni lekar dr Vuljf Zelcer, vojni lekar Konstantin Nikolajevič Jacenko i poručnik dr Roman Strutinski; 6. puk – komandir vojni lekar dr Josif Vartolomejevič Cipilevski; 1. bataljon potporučnik med. Franjo Vuletić; 2. bataljon vojni lekar Semen Avramovič Judelevič; 3.bataljon vojni lekar Mihail Davidov.

Polovinom 1917. stanje u ruskoj vojsci bilo vrlo loše, te je Stavka pokušavala da privoli srpsku Vrhovnu komandu na pristanak da se 1. divizija uputi na front kao pojačanje posustalim i nesigurnim ruskim jedinicama. Pošto je pristanak za to dobijen od srpske Vrhovne komande, pristupilo se njenoj popuni do punog sastava iz redova 2. divizije, dok je za nju odlučeno da se rasformira.

Za popunu 1. divizije 10. jula 1917. poslato je 108 oficira i 3 510 podoficira i vojnika, dok je od ostatka 2. divizije 12. jula 1917. obrazovan Rezervni puk [11].

Brojno stanje Rezervnog puka iznosilo je 145 oficira i 2.588 podoficira i vojnika, a njegov nacionalni sastav bio je: Srbi (2 395), Hrvati (198), Slovenci (77), Česi (59), Slovaci (6) i Poljaci (8) [12].

Druga divizija prestala je da postoji 17. jula 1917.

U 1. diviziji, sanitet posle popune, 26. 7. 1917, činili su [40]: referent saniteta, ppukovnik dr Dragoslav Popović; u 1. puku (novi) komandir vojni lekar dr Gelman (poručnik dr Apostol Hadži Gligorije prešao je u ruski vojni sanitet, učestvovao je u građanskom ratu na strani „belih“ i u Srbiju se vratio tek posle rata); u 1. bataljonu potporučnik med. Dušan Krstić; u 2. bataljonu potporučnik med. Đura Dekanić; u 3. bataljonu potporučnik med. Isa Nedeljkov; u 2. puku komandir kapetan 2 klase dr Teodor Serebrenikov i bataljonski lekari, potporučnik med. Milan Mirković i potporučnik med. Josif Gazarek; u 3. puku, komandir poručnik dr Aleksej Šepelj i bataljonski lekari, potporučnik med. Milan Jovanović, potporučnik med. Josip Hebajn i potporučnik med. Milko Gnezda; u 4. puku, komandir vojni lekar dr Ivan Kolesničenko (ovaj će lekar kasnije prići Crvenoj armiji) i bataljonski lekari: potporučnik med. Sima Ilić, potporučnik med. Karel Sadilek, potporučnik med. Rudolf Lehki i poručnik (dr?) Bogoslav Bouček; u Bolničarskoj četi, komandir kapetan 1 klase dr Božidar Janković, hirurg vojni lekar dr Feliks Bergman, komandir nosilaca ranjenika kapetan 2 klase Josip Klančić; u 1. lazaretu, komandir kapetan 1 klase dr Vladimir Stanojević; mlađi lekari – vojni lekari Ludvik Godlevski i Gelman, te medicinar Aleksandar Petrović; u 2. lazaretu, komandir kapetan 2 klase dr Karel–Dragutin Freja; mlađi lekari – vojni lekari dr Gomberg i dr Mutermilh i medicinar Svetozar Nikolajević; Profijant kolona, v.d. lekara potporučnik med. Stojan Grgurov i veterinar Mihailo Petrović.

Prema naredbi od 18. 3. 1917. u Komandi lako ranjenih bili su [41]: 1. komanda (rekonvalescenti) – trupni lekar i upravnik ambulante, poručnik dr Božidar Fajdiga; 2. komanda (stalno i privremeno nesposobni) – trupni lekar i upravnik ambulante poručnik dr Bogoslav Bouček; i apotekar obe komande, Andrija Posavac

U međuvremenu, zajedničkim otporom oficira i vojnika divizije i naporima plemenite dr Elzi Inglis, uspelo se da se divizija ne šalje na front, već da se celokupno ljudstvo Korpusa brodovima uputi u Solun za pojačanje Srpske vojske čiji su redovi bili već opasno proređeni.

Načelnik saniteta Korpusa, pukovnik dr Milan Žerajić, ostao je u Odesi (u dokumentu se ne navodi razlog) [42].

Time je završen rad Dobrovoljačkog korpusa i njegovih divizija u Rusiji.

Ostali medicinari koji su bili na službi u Rusiji [43-45]

Za sledeće ličnosti prema „Knjizi lekara i medicinara“ [44], vođenoj u Sanitetskom odeljenju Vrhovne komande, i nekih drugih dokumenata iz Arhiva srpske vojske, kao i ostalih izvora u vezi sa temom, zna se da su se nalazili u Korpusu. To su bili: poručnik dr Jovan Popović (prebegao u Srbiju 10. 8. 1914, iz Soluna poslat u Rusiju u jesen 1916, vratio se aprila 1918), nema ga ni u jednom *Ordre de bataille*, moguće je da je obavljao dužnosti van Korpusa (obilasci zarobljeničkih logora, oficir za vezu sa ruskim vlastima i sl). Ni u njegovom personalnom dosijeu K-1399/52 koji se nalazi u Vojnom arhivu nema detalja, osim da je „poslat u Rusiju sa drugom partijom oficira radi popune 2. dobrovoljačke divizije“; medicinar potporučnik Milivoje Malušev; medicinar

Savatije Mitrović, poslat sa Krfa (u nekoj kasnijoj grupi); medicinar Milutin Đorić, poslat sa Krfa 16. 9. 1916, ubrzo pušten da u Moskvi nastavi studije, diplomirao 1917, u Srbiju se vratio jula 1919. (kao državljanin Srbije nije bio dobrovoljac, već vojni obveznik); dr Milan Simonović, dobrovoljac, 19. 9. 1917. unapređen u čin poručnika, u Rusiji bio 1916–1918, vratio se 27. 8.1918. u Solun; kapetan 1 klase dr Vladimir Ivanov (Rus, u srpskoj vojsci od 1912, poslat iz Soluna u Dobrovoljački korpus 12. 6.1917. godine, 1919. je bio „još u Rusiji", kasnije se ne pominje vojnoj dokumentaciji, verovatno ostao u Rusiji); kapetan 1 klase dr Jakov Rjabin (Rus, u Srpskoj vojsci od 1913, poslat u Rusiju iz Soluna 14. 1. 1918. u srpsko poslanstvo u Moskvi 1918, posle se ne pominje u vojnoj dokumentaciji, pa se pretpostavlja da je ostao u Rusiji.

Posleratne sudbine medicinara i lekara

Smatrajući da je posle 90 godina od ovih događaja, koji su zbog svega što se naknadno na jugoslovenskim prostorima dešavalo od 1941. do danas, uveliko predati zaboravu, teško vratiti u sećanje ove mlade plemenite ličnosti, nastojali smo se da saznamo njihovu posleratnu sudbinu. To je posle raspada Jugoslavije 1991. postao vrlo težak zadatak. Za veći broj onih koji su živeli i radili van Srbije nismo uspeli, pored svih uloženih napora, da od arhiva ili lekarskih društava novostvorenih država dobijemo željene podatke, a mnoga naša pitanja putem Interneta ostala su bez odgovora. Za većinu, ipak, uspeli smo da rekonstruišemo bar delimične biografske podatke.

Srbi [43-47]

Dr Aleksandar Petrović (1893–?), dugogodišnji šef ATD službe u Kikindi;

Dr Svetozar D. Nikolajević (1892–?), hirurg, pukovnik, šef Urološkog odeljenja Glavne vojne bolnice u Beogradu, odlikovan ordenima beli orao 5. reda, jugoslovenska kruna 5. reda, čehoslovačkim ordenom ratni krst;

Dr Milan Matić (1888–1955), ostao u Rusiji da diplomira, potom radio kao lekar, po povratku u Kragujevac bio opštinski lekar i sanitarni inspektor;

Dr Milutin Velimirović (1893–1973), sreski lekar (Knjaževac, Ćuprija, Jagodina), primarijus Gradske bolnice u Beogradu, književnik i putopisac;

Dr Mihailo Babović (1890–1961), pukovnik, referent saniteta Bosanske divizijske oblasti, odlikovan ordenima zlatna medalja za hrabrost, zlatna medalja za vojničke vrline, Sveti Sava 5. reda;

Dr Milutin Đorić (1888–1930), završio medicinu u Rusiji 1917. (imao 10 semestara do rata), sreski lekar u Petrovcu na Mlavi;

Dr Vojislav Veselinović (1889–1961), šef saniteta banske uprave u Novom Sadu, bio u zarobljeništvu 1941–1945;

Dr Stojan Grgurov (1892–1944), lekar Okružnog ureda u Somboru, 1941. prebegao u Srbiju, gde su ga 1944. ubili Nemci;

Dr Đura Dekanić (1891–1977), specijalista ginekolog, primarijus, šef odeljenja u Velikom Bečkereku, Pokrajinskoj bolnici u Novom Sadu i Centralnom onkološkom dispanzeru SR Srbije u Beogradu, profesor i upravnik Babičke škole, ranjen i odlikovan u Dobrudži zlatnom medaljom za hrabrost;

Dr Sima Ilić (1892–1980), specijalista dermatovenerolog, profesor Medicinskog fakulteta u Beogradu i upravnik Dermatovenerološke klinike;

Dr Zdravko Jeftanović (1893–?), specijalista rendgenolog, lekar državnih bolnica u Sarajevu, Skoplju i Beogradu;

Dr Milan Jovanović (1884–?), dugogodišnji lekar i upravnik Doma narodnog zdravlja u Velikom Bečkereku (Zrenjanin);

Dr Branko Kešanski (1889–?), lekar u svome rodnom selu (Šajkaški Sv. Ivan kod Titela),

Dr Jovan Popović (1881–1972), lekar u Novom Sadu (sreski, šef odeljenja u Higijenskom zavodu, šef odseka u Povereništvu za narodno zdravlje);

Dr Nikola Kešeljević (1890–?), specijalista dermatovenerolog, Beograd;

Dr Milan Mirković, bio rezervni oficir bivše Jugoslovenske vojske;

Student medicine Isa Nedeljkov, bio na Solunskom frontu u 1. jugoslovenskom puku, (nema ga u rang-listama rezervnih oficira bivše Jugoslovenske vojske);

Student medicine Dušan Krstić, nepoznato (nema podatka u rang-listama rezervnih oficira bivše Jugoslovenske vojske);

Student medicine Bogoljub Kurandić, nepoznato (nema ga u rang-listama rezervnih oficira bivše Jugoslovenske vojske);

Student medicine Milivoje Malušev 1919. bio v.d. lekara u Drinskoj diviziji (6. puk, 1. bataljon), dalje nepoznato (nema ga u rang-listama rezervnih oficira bivše Jugoslovenske vojske);

Neki od dobrovoljaca iz 1914. godine, posle rata nastavili su aktivnu vojnu službu i u njoj napredovali do čina pukovnika. To su bili: dr Roman Fedina, dr Dinko Cvitanović i dr Apostol Hadži Gligorije.

Slovenci [43-47]

Dr Božidar Fajdiga (1887–1969), dugogodišnji sreski lekar u Kranju i mesna legenda. U Drugom svetskom ratu pomagao partizane, od Nemaca interniran u okolinu Berlina, učestvovao u lečenju ranjenika. Posle rata radio u Kranju do smrti;

Dr Josip Hebajn (1891–1973), specijalista radiolog, profesor Medicinskog fakulteta u Ljubljani, nosilac ruskog ordena Svetog Stanislava sa mačevima 3. reda, ordena rumunske krune – oficirski red, jugoslovenskog belog orla 5 reda i drugih priznanja;

Dr Valentin Meršol (1894–1981), specijalista infektolog, primarijus, šef Infektivnog odeljenja bolnice Šiška u Ljubljani, nosilac ordena beli orao 5. reda, jugoslovenske krune 5. reda, češkoslovenskog valečnog križa, Zlatne medalje za revnosnu službu i drugih. Godine 1945, emigrirao u SAD gde mu žive potomci (Klivlend);

Dr Rudolf Trušnovič, student medicine 4. godine, komandir 1. čete u 1. bataljonu 1. puka 1. divizije, teško ranjen kod Kokardže, odlikovan: beli orao 5. reda, ruski orden

Sveta Ana 3. reda. Učesnik građanskog rata u Rusiji, komandir u kozačkom konjičkom puku „belih“, medicinu završio u Harkovu, u Jugoslaviju se vratio oko 1936, lekar u Moroviću (Srem), poslanički kandidat na izborima 1938. na listi „Zbor“, za vreme nemačke okupacije Srbije radio u Ministarstvu narodnog zdravlja, emigrirao 1945. u SAD, gde mu žive potomci (?), ubijen u Zapadnom Berlinu 1957. od strane sovjetskih agenata pod narezjašnjenim okolnostima (podatak usmeno dobijen od dr Jovana Kačakija);

Dr Lovro Klemenčič – Dušan (1891–1928), narednik-junak sa Kajmakčalana, član Jugoslavenskog odbora u Rusiji, predsednik Jugoslovenskog kluba studenata-marksista u Beču i narodni poslanik Konstituante posle rata na listi Komunističke partije (KPJ), poginuo 1926. u saobraćajnoj nesreći;

Dr Tone Lovšin, sreski lekar u Laškom (Slovenija);

Dr Josip Erat, lekar u Dravogradu-Prevalja (Slovenija);

Dr Jože Ermenc, specijalista ginekolog u Somboru, emigrirao 1945. u Argentinu,

Dr (?) Tone Eržen (nema podataka);

Student medicine Milko Gnezda (1894–1922), nastavio studije u Pragu, umro u Nišu kao aktivni oficir Jugoslovenske vojske ne dovršivši studije;

Dr Viktor Sosič (istarski Slovenac), sreski lekar u Makedoniji (Kratovo, Štip) i Srbiji, istovemeno vojni honorarni lekar.

Hrvati [43-47]

Dr Milivoj Jambrišak (1878–1943), specijalista stomatolog, učesnik NOB i većnik AVNOJ-a, prvi ministar zdravlja u ratnoj partizanskoj vladi umro 1943. godine po povratku sa Drugog zasedanja u Jajcu;

Dr Stjepan Poljak (1889–1955), asistent, Neuropsihijatrijska klinika, Zagreb (1923–1928), profesor neuroanatomije, Berkeley, Chicago, USA;

Dr Franjo Vuletić, specijalista internista, učestvovao po povratku iz Rusije i oslobođenja u „ratu za severne granice“ (bitke oko Štajerske i Koruške) 1919. kao lekar 1. srpskog konjičkog puka, kasnije lekar u Pućištu na otoku Braču;

Dr Ljudevit Horvatin – Lujo, sreski lekar u Ogulinu,

Dr Ivo Petković, sreski lekar u Splitu;

Dr Antun Saso, sreski lekar u Velesu;

Student medicine Nikola Šajatović, komandir voda u 3. puku 1. srpske dobrovoljačke divizije, poginuo kod Hardalija 1. 10. 1916.

Česi [43-45]

Česi su se po povratku pridružili češkim trupama u Francuskoj, ili su se posle završetka rata vratili u svoju domovinu: Josif Gazarek, Bedžih Opletal, Bogoslav Bouček, Karel Sadilek, Rudolf Lehki, Jaroslav Kohoušek i dr Karel Freja – Dragutin (nastavio vojnu službu u ČSR do čina pukovnika, Radovan Gajda (kasnije general ČSR).

Rusi [43-45]

Od ruskih lekara, koji su bili srpski sanitetski oficiri, dr Serebrenikov, dr Rjabin i dr Ivanov ostali su u Rusiji, dr Šepelj je umro krajem 1917, o dr Strutinskom i Kržiškovskom nema daljih podataka. Dr Lav Ivanovič Polivec – Ljoška kraće vreme bio je građanski lekar, potom se vratio u Jugoslovensku vojsku i napredovao do čina majora. Bio je lekar 6. vazduhoplovnog puka u Beogradu i 13. puka „Hajduk Veljko“ u Negotinu, gde je penzionisan.

Zaključna razmatranja

Od ovih događaja prošlo je više od 90 godina, skoro celo jedno stoleće, ispunjeno novim svetskim ratom i, poslednjih godina 20. veka, tragičnim zbivanjima na teritoriji bivše zajedničke države, a patina istorijskog zaborava je sve deblja: i poslednji, neposredni potomci ovih boraca već su u dubokoj starosti. Novim naraštajima, ukoliko budu zainteresovani, ostaje da od zaborava sačuvaju uspomenu na ove mlade ljude koji su, prevazišavši uske nacionalne međe, bili vesnici nečega što, igrom sudbine, ljudskom nesavršenošću i uticajem stranih faktora, nije trajno opstalo.

Ako se pogleda spisak 42 poginula oficira 1. divizije, uočava se da su među njima 30 potporučnika i pet poručnika, najmlađe intelektualne snage svojih naroda, čija bi prirodna misija bila da stasaju vremenom u njihove predvodnike. Posebno je tužno kada se, bez ikakve želje za nacionalnim prebrojavanjem, vidi da su jedan pored drugoga hrabro izginuli: Srbi (23), Česi (8), Hrvati (6), Slovenci (4) i Rusi (1), verujući da se bore i žrtvuju za neko bolje sutra njihovih naroda [45, 46, 48].

Za razliku od oficira, među poginulim vojnicima u ogromnoj većini bili su Srbi iz Bosne i Hercegovine, Srema, Bačke, Banata i Baranje, Dalmacije, Banije, Korduna i Slavonije, što je odgovaralo nacionalnom sastavu divizije [48].

Posle rata posmrtni ostaci poginulih većinom su sahranjeni ispod spomen-piramide podignute u Medžidiji.

L I T E R A T U R A

1. *Maksimović V*. Memorial of the 1. Serbian Volunteer Division in Russia 1916-1926. anonym. Belgrade: Prosveta; 1926. (Serbian)
2. *Popović N*. Yugoslav volunteers in Russia 1914−1918. Belgrade: Udruženje dobrovoljaca 1912−1918; 1977. (Serbian)
3. *Turk E, Jeras J, Pavlin R*. Volunteers, blacksmiths of Yugoslavia. Ljubljana, Maribor: Zveza ratnih dobrovoljcev; 1936. (Slovenian)
4. *Nedok AS*. Russian medical help during liberation and defence wars of Serbia in XIX and early XX century (1804-1917). Vojnosanit Pregl 2009; 66(7): 587−96. (Serbian)
5. *Stanojević V*. My war scripts and pictures, anonym. Ljubljana: Autorsko izdanje; 1934. (Serbian)
6. H.Q. New Territories Troops. Med. No. 24738, 6.02.1916. Belgrade: Vojnoistor Arh; pop.3a, kut. 116, fasc. 5, dok .1, list 2. (Serbian)
7. *Nedok A*. The Retreat of Serbian Army to the Albanian Coast, and their Evacuation to Corfu, Work of Military Medical Service. Beograd: AMD System; 2006. p. 92 and 99−101. (Serbian)
8. *Kranjc M*. Slovenian military inteligentsia. Grosuplje: Grafis Trade; 2005. p. 333. (Slovenian)

9. *Mitrović A*. Serbia in the Great War. Belgrade: Stubovi kulture; 2004. p. 106−14.
10. *Trušnović R.* The old and the new Russia, anonym. Skoplje: Južna Srbija; 1937. (Serbian)
11. H.Q. 2nd Serbian voluntary division, Confid. Adj. No. 1626, 1.07.1917. Belgrade: Vojnoistor Arh; pop. 10, kut. 6, fasc. 2, doc.17. (Serbian)
12. H.Q. 2nd Serbian volunteer division. A.No. 8764 12.07.1917. Belgrade: Vojnoistor Arh; pop. 10, kut. 6, fasc.2, dok. 18. (Serbian)
13. H.Q. Serbian volunteer detachment. Order No. 5, 11.11.1916. Belgrade: Vojnoistor Arh; pop. 10, kut. 48a, fasc. 1, dok. 23, list 8. (Serbian)
14. *Milanković J*. Memoires from Siberia 1918−1919. Belgrade: Autorsko izdanje; 1926. (Serbian)
15. Supreme Commanders Prince-Regent Alexander Order. 11.02.1916. List of Commanding officers. Belgrade: Vojnoistor Arhiv; pop.10, kut. 46, fasc. 1, dok. 2. (Serbian)
16. H.Q. Supreme Command: Adj. No. 62065, 2.02.1916. Belgrade: Vojnoistor Arh; pop.3a, kut. 116, fasc. 5, dok. 2, list 1. (Serbian)
17. H.Q. Supreme Command, Medic. Dept. Confid. Med. No. 33566, 23.02.1916. Belgrade: Vojnoistor Arh; pop. 3a, kut. 116, fasc. 5, dok. 52. list 1. (Serbian)
18. H.Q., Supreme Command: Adj. No. 62033, 23.02. 1916. Belgrade: Vojnoistor Arh; pop. 3a, kut. 116, fasc. 5, dok. 43, list 1. (Serbian)
19. *Velimirović M*. Papers (Personal documents). Belgrade: Srpska akademija nauka i umetnosti; rukopisni arhiv, No. 14325; 1971. (Serbian)
20. *Dimitrijevic V*. In the pleins of illusions. Belgrade: Filip Višnjić; 2008. (Serbian)
21. Albanian Medal Society: Golgotte and Resurrection of Serbia 1915−1916. Belgrade: Grafički zavod; 1971. (Serbian)
22. H.Q. 1th Serbian volunteer division: Order No. 19, 16.04.1916. Belgrade: Vojnoistor Arh; pop.10, kut. 1, fasc.1, dok.1, list 19. (Serbian)
23. H.Q. 1th Serbian volunteer division: Order No. 219 14.06.1916. Belgrade: Vojnoistor Arh; pop. 10, kut. 1a, fasc. 2,dok.11, list 89. (Serbian)
24. H.Q. 1th Serbian volunteer division: „Ordre de bataille" (no Nr. or Date). Belgrade: Vojnoistor Arh; pop.10, kut.3, fasc.1, dok.6 (Serbian)
25. *Stanojević V*. Our Medical Service in Dobrudja. In: *Stanojevic V*. History of Serbian Military Medical Service and Our War Medical Experience. Belgrade: Vojnoizdavački i novinski centar; 1992. p. 423−32. (Serbian)
26. Great German H.Q. The Dobrudja Campain, Meyer's Grosses Konversations-Lexicon, War Suppl. II. Leipzig,Wien:, Bibliographisches Institut; 1917; Suppl 2: 221−5. (German)
27. H.Q. 1th Serbian volunteer division: Survey of Losses during Operations in Dobrudja. No. offic. Nov.1916. Belgrade: Vojnoistor Arh; pop. 10, kut. 13, fasc. 2, dok. 15/4, list 2. (Serbian)
28. *Maksimović V*. 1th Serbian Volunteer Division. Presentation Held 19.09.1926. Belgrade: Vojnoistor Arh; pop.10, kut.10, fasc. 1, dok.2, list 8. (Serbian)
29. Serbian Military Atache Col. Andonovic, Rumanian H.Q. Telegramm 4.11.1916. Belgrade: Vojnoistor Arh; pop. 3, kut. 111, fasc. 9, dok. 7/35. (Serbian)
30. H.Q. 1th Serbian volunteer division. No. offic. 18.11.1916. Belgrade: Vojnoistor Arh; pop. 10, kut. 17, fasc.1, dok. 1/5, list 2. (Serbian)
31. H,Q. 1th Serbian volunteer division: „Ordre de bataille" No. offic. Nov. 1916. Belgrade: Vojnoistor Arh; pop. 10, kut .3, fasc. 1, dok. 6/14. (Serbian)
32. 2nd Regiment, 1th Serbian volunteer division: „Ordre de bataille". No.offic./ 10.11.1916. Belgrade: Vojnoistor Arh; pop. 10, kut. 20, fasc.1, dok. 25/2, list 3. (Serbian)
33. 3d Regiment, th Serbian volunteer division: „Ordre de bataille" . No.offic./ 12.12.1916. Belgrade: Vojnoistor Arh; pop.10, kut. 55, fasc. 1, dok.1/ 2, list 1. (Serbian)
34. H.Q. Serbian Volunteer Corps, „Ordre de bataille", No. offic./1916. Belgrade: Vojnoistor Arh; pop. 10, kut 3, fasc. 1, dok. 6/14. (Serbian)
35. H.Q. 2nd volunteer division: „Ordre de bataille". No. offic./1916. Belgrade: Vojnoistor Arh; pop. 10, kut. 3, fasc. 1, dok. 6/14. (Serbian)
36. H.Q. Serrbian Volunteer Corps: „Ordre de bataille, Complementary Batalion". Belgrade: Vojnoistor Arh; pop.10, kut. 3, fasc. 1, dok. 6/14. (Serbian)
37. H.Q. Volunteer Corps of Serbs, Croats and Slovenes: Order No.141/1917. Belgrade: Vojnoistor Arh; pop. 10, kut.1a, fasc. 2, dok.12, list 83. (Serbian)
38. H.Q. 2nd Serbian volunteer division: „Ordre de bataille", No.offic./1917. Belgrade: Vojnoistor Arh; pop.10, kut. 4a, fasc. 4, dok. 50. (Serbian)
39. H.Q. 2nd Serbian volunteer division „Ordre de bataille",O.No.1566, 9.06.1917. Belgrade: Vojnoistor Arh; pop. 10, kut. 35, fasc. 1, dok . 4. (Serbian)
40. H.Q. 1th Serbian volunteer division. Confid. Adj. No. 2627, 26.07.1917. Belgrade: Vojnoistor Arh; pop. 10. kut. 42, fasc.1, dok. 6. (Serbian)
41. H.Q. Volunteer Corps of Serbs, Croats and Slovenes: Order No.141, 18.03.1917. Belgrade: Vojnoistor Arh; pop. 10, kut. 1a, fasc. 2, dok.12, list 85. (Serbian)
42. H.Q. Volunteer Corps of Serbs, Croats and Slovenes. No. offic./1917. Belgrade: Vojnoistor Arh; pop.10, kut.4a, fasc.4, dok.43. (Serbian)
43. Kingdom of Yugoslavia Armed Forces: Official documents of active and reserve officers and military officials. Belgrade: Vojnoistor Arh; 1962. (Serbian)
44. Supreme Command, Medic. Dept. „The Book of doctors and medical students 1916-1919". Belgrade: Vojnoistor Arh; pop. 3a, kut. 95, fasc 1, dok. 4. (Serbian)
45. *Nedok A*. History of the Serbian Medical Military Service 1836−1918. Neobjavljeni podaci: rukopis kod autora. (Serbian)
46. *Nedok A*. Military Medical Service of the Serbian Volunteer Corps in Russia 1916-1917. Belgrade: Akademija medicinskih nauka Srpskog lekarskog društva; 2007. Book I(2), p. 163−80. (Serbian)
47. Ministry of social wellfare and of peoples's health. Belgrade: Medical Anual; 1933. (Serbian)
48. *Ristić RT*. The Dobrudja Epopee. Belgrade: Udruženje ratnih dobrovoljaca 1912−1918, njihovih potomaka i poštovalaca; 2006. p. 151−7. (Serbian)

Primljen: 24. X 2011.
Prihvaćen: 16. IV 2012.

INDEKS RADOVA ČASOPISA VOJNOSANITETSKI PREGLED, GODINA 2012

ANATOMIJA

MIŠIĆNO-SKELETNI SISTEM

KARDIOVASKULARNI SISTEM

NERVNI SISTEM

BOLESTI

BAKTERIJSKE I GLJIVIČNE INFEKCIJE

VIRUSNE BOLESTI

NEOPLAZME

BOLESTI MIŠIĆNO-SKELETNOG SISTEMA

BOLESTI DIGESTIVNOG SISTEMA

BOLESTI STOMATOGNATNOG SISTEMA

BOLESTI RESPIRATORNOG TRAKTA

BOLESTI UVA, GRLA I NOSA

BOLESTI NERVNOG SISTEMA

OČNE BOLESTI

UROLOŠKE I MUŠKE GENITALNE BOLESTI

GINEKOLOŠKE BOLESTI I POREMEĆAJI TRUDNOĆE

KARDIOVASKULARNE BOLESTI

NEONATALNE BOLESTI I ANOMALIJE

BOLESTI KOŽE I VEZIVNOG TKIVA

METABOLIČKE I NUTRICIONE BOLESTI

BOLESTI ENDOKRINOG SISTEMA

IMUNOLOŠKE BOLESTI

POREMEĆAJI IZAZVANI ŽIVOTNOM SREDINOM

HEMIJSKE MATERIJE I LEKOVI

PESTICIDI, NOKSE I DRUGI ZAGAĐIVAČI ŽIVOTNE SREDINE

SREDSTVA KOJA DELUJU NA CENTRALNI NERVNI SISTEM

SREDSTVA ZA KRVNI, GASTROINTEST. SISTEM I BUBREGE

IMUNOLOŠKI I BIOLOŠKI FAKTORI

FARMACEUTSKI PREPARATI

ANALITIČKE, DIJAGNOST. I TERAP. TEHNIKE I OPREMA

DIJAGNOSTIKA

TERAPIJSKE PROCEDURE

ANESTEZIJA I ANALGEZIJA

OPERATIVNE HIRURŠKE PROCEDURE

ISTRAŽIVAČKE METODE

STOMATOLOGIJA

PSIHIJATRIJA I PSIHOLOGIJA

PONAŠANJE I MEHANIZMI PONAŠANJA

MENTALNI POREMEĆAJI

BIHEVIORALNE DISCIPLINE I AKTIVNOSTI

BIOLOŠKE NAUKE

PROFESIJE U ZDRAVSTVU

FIZIOLOGIJA OKA, MIŠIĆNO-SKEL. I NERVNOG SISTEMA

Goran Ranković, Nataša Djindjić, Gorana Ranković-Nedin, Saša Marković, Dragan Nejić, Branislava Miličić, Boris Djindjić
The effects of physical training on cardiovascular parameters, lipid disorders and endothelial function
2012; 69(11): 956–960.

INFORMATIKA

Silva Dobrić
Nova godina – novi izazovi
2012; 69(1): 5–8.

Slobodan Obradović, Dragana Obradović
The Big Anniversary – 200 Years of The New England Journal of Medicine
2012; 69(2): 139–140.

Silva Dobrić
Autor godine Vojnosanitetskog pregleda za 2011.
2012; 69(3): 223–226.

Silva Dobrić
Plagijarizam ili "*copy-paste*" manipulacije
2012; 69(6): 467–468.

Snežana Janković, Silva Dobrić, Maja Marković, Sonja Andrić Krivokuća, Aleksandra Gogić
Plagiarism detection – how we do that
2012; 69(9): 743–746.

Silva Dobrić
Authorship misusing in scientific publications
2012; 69(12): 1028–1030.

ZDRAVSTVENA ZAŠTITA

EKONOMIJA I ORGANIZACIJE ZDRAVSTVENE ZAŠTITE

Sandra Stefan-Mikić, Siniša Sević, Radoslava Doder, Dejan Cvjetković, Nataša Jovanović, Maja Ružić
Troškovi lečenja infekcija urinarnog trakta kod primene pojedinih farmakoterapijskih smernica u Klinici za infektivne bolesti Kliničkog centra Vojvodine
2012; 69(8): 647–655.

Dubravka Jaganjacova, Petr Hava, Eva Kalvodova
Organization of healthcare about patients with cerebrovascular disease in the Czech Republic
2012; 69(9): 794–798.

KVALITET, PRISTUP I PROCENE ZDRAVSTVENE ZAŠTITE

Maja Damnjanović, Aneta Lakić, Dejan Stevanović, Ana Jovanović, Jasna Jančić, Mirjana Jovanović, Ljubica Leposavić
Self-assessment of the quality of life of children and adolescents in the child welfare system of Serbia
2012; 69(6): 469–474.

ISTORIJA MEDICINE

INDEKS AUTORA

INDEKS DESKRIPTORA

VOJNOSANITETSKI PREGLED
VOJNOMEDICINSKA AKADEMIJA
Crnotravska 17, 11040 **Beograd, Srbija**
Tel/faks: +381 11 2669689
vsp@vma.mod.gov.rs
vmavsp@hotmail.com

Poziv za reklamiranje u 2013. godini

U prilici smo da vam ponudimo mogućnost oglašavanja i reklamiranja proizvoda i usluga u časopisu „**Vojnosanitetski pregled**“ (VSP). To je sigurno najbolji vid i najzastupljeniji način upoznavanja eventualnih korisnika sa vašim uslugama i proizvodima.

Časopis „**Vojnosanitetski pregled**“, zvanični organ lekara i farmaceuta Vojske Srbije, naučno-stručnog je karaktera i objavljuje radove iz svih oblasti medicine, stomatologije i farmacije. Radove ravnopravno objavljuju stručnjaci iz vojnih i civilnih ustanova i iz inostranstva. Štampa se na srpskom i engleskom jeziku. Časopis izlazi neprekidno od 1944. godine do sada. Jedini je časopis u zemlji koji izlazi mesečno (12 brojeva), na oko 100 strana A4 formata, a povremeno se objavljuju i tematski dodaci (suplementi). Putem razmene ili pretplate VSP se šalje u 23 zemlje sveta. Radove objavljene u VSP-u indeksiraju: *Science Citation Index Expanded (SCIE)*, *Journal Citation Reports/Science Edition*, *Index Medicus (Medline*), *Excerpta Medica (EMBASE), EBSCO* (preko ove baze VSP je *on line* dostupan od 2002. godine u *pdf* formatu) i *Biomedicina Serbica*.

Cene reklama i oglasa u časopisu „Vojnosanitetski pregled“ u 2012. godini su:

1.	Oglas u crno-beloj tehnici A4 formata za jedan broj	20 000,00 dinara
2.	Oglas u c/b tehnici A4 formata za celu godinu (11-12 brojeva)	200 000,00 dinara
3.	Oglas u boji A4 formata za jedan broj	35 000,00 dinara
4.	Oglas u boji A4 formata za celu godinu (11-12 brojeva)	330 000,00 dinara
5.	Oglas u boji na koricama K3 za jedan broj	50 000,00 dinara
6.	Oglas u boji na koricama K3 za celu godinu (11-12 brojeva)	455 000,00 dinara
7.	Oglas u boji na koricama K2 i K4 za jedan broj	55 000,00 dinara
8.	Oglas u boji na koricama K2 i K4 za celu godinu (11-12 brojeva)	530 000,00 dinara

Za sva obaveštenja, uputstva i ponude obratiti se redakciji časopisa „Vojnosanitetski pregled“. Sredstva se uplaćuju na žiro račun kod Uprave javnih plaćanja u Beogradu broj: 840-941621-02 **VMA (za Vojnosanitetski pregled ili za VSP)**, **PIB 102116082**. Uplatnicu (dokaz o uplati) dostaviti lično ili poštom (pismom, faksom, *e-mail*-om) na adresu: Vojnosanitetski pregled, Crnotravska 17, 11000 Beograd; tel/faks: 011 2669 689, e-mail: vsp@vma.mod.gov.rs ili vmavsp@hotmail.com

VOJNOMEDICINSKA AKADEMIJA
INSTITUT ZA NAUČNE INFORMACIJE
Redakcija časopisa „Vojnosanitetski pregled“
Crnotravska 17, 11 000 Beograd

Kontakt tel./fax: **+381 011 26 69 689**
Elektronska pošta: **vsp@vma.mod.gov.rs**

VOJNOMEDICINSKA AKADEMIJA

INSTITUT ZA NAUČNE INFORMACIJE

Uređivački odbor

UPUTSTVO

ZA PISANJE RADOVA ZA

Vojnosanitetski pregled

„Vojnosanitetski pregled“ (VSP) objavljuje samo one radove koji nisu ranije objavljeni. Mogu se prihvatiti i radovi *in extenso* koji su prethodno delimično izloženi na naučnom/stručnom skupu. **Svaki pokušaj plagijarizma ili autoplagijarizma kažnjava se** (zabrana objavljivanja radova svim autorima u VSP-u u vremenskom periodu zavisno od stepena plagijarizma i o tome obaveštava rukovodstvo institucija u kojima autori rade i njihova strukovna udruženja).

Primaju se samo radovi napisani na engleskom sa apstraktom i na srpskom i na engleskom jeziku.

Od 1. januara 2012. godine Vojnosanitetski pregled prešao je na e-Ur: elektronsko uređivanje časopisa radova poslatih na adresu: http://scindeks-eur.ceon.rs/index.php/vsp

Svi autori, recezenti i urednici moraju biti registrovani korisnici sistema sa jednoznačnom e-mail adresom. Registraciju je moguće izvršiti na:

http://scindeks-eur.ceon.rs/index.php/vsp

ili

aseestant.ceon.rs/index.php

Tehničko uputstvo za korišćenje sistema **e-Ur: elektronsko uređivanje radova** može se takođe preuzeti na:

http://www.vma.mod.gov.rs/vsp

Prilikom prijave rada u sistem elektronskog uređivanja „Vojnosanitetskog pregleda“ neophodno je priložiti izjavu da su ispunjeni svi postavljeni tehnički zahtevi uključujući i izjavu potpisanu od strane svih autora i koautora da rad nije ranije ni u celini niti delimično objavljen niti prihvaćen za štampanje u drugom časopisu. Izjava o pojedinačnom doprinosu autora mora biti potpisana od strane svakog autora rada, skenirana i poslata uz rad kao dopunska datoteka. Takođe, autori su obavezni da dostave i potpisanu izjavu o nepostojanju sukoba interesa. Tim postupkom svi autori postaju odgovorni za ispunjavanje svih postavljenih uslova, čemu sledi odluka o prihvatanju za dalji uređivački postupak.

Sistem **Aseestant: elektronskog uređivanja časopisa** podrazumeva korišćenje servisa *CrossCheck*, pa se svi prijavljeni radovi automatski, pre prvog koraka uređivačkog postupka, proveravaju na **plagijarizam ili autoplagijarizam.**

Prihvaćeni radovi objavljuju se po redosledu koji određuje Uređivački odbor na predlog glavnog i odgovornog urednika.

U Vojnosanitetskom pregledu objavljuju se: **uvodnici**, **originalni** članci, **prethodna** ili **kratka saopštenja**, revijski radovi tipa **opšteg pregleda**, **aktuelne teme, metaanalize i seminari praktičnog lekara**, **kazuistika (prikaz bolesnika)**, članci iz **istorije medicine**, **lični stavovi**, **komentari**, **pisma uredništvu,** izveštaji sa naučnih i stručnih skupova, prikazi knjiga.

O r i g i n a l n i č l a n c i po obimu ne smeju prelaziti 16 stranica teksta (bez priloga) (vidi poglavlje **„Priprema rada“).**

P r e t h o d n o ili k r a t k o s a o p š t e n j e ne sme biti duži od pet stranica (bez priloga). Ona predstavlja početnu ili kratku belešku o istraživanjima koja nisu završena, ali su dobijene informacije od interesa za naučnu i stručnu javnosti. Sadrži sva poglavlja kao originalni naučni ili stručni članak, ali u znatno skraćenom obimu. **Rezultati** i **diskusija** mogu biti spojeni u jednu celinu, **zaključak** može izostati, ali se na kraju članka mora dati kratki tekst u vidu preliminarnog zaključka ili najave daljih istraživanja.

O p š t i p r e g l e d, tematska studija (do 16 stranica), predstavlja sistematsko izlaganje o problemu na osnovu podataka iz literature, uključivši i najmanje pet radova autora članka iz uže oblasti iz koje je rad. On mora obuhvatiti svu dostupnu pripadajuću literaturu za određeni vremenski period. Autor članka mora dati i svoje viđenje problema u vidu zaključnog stava (podnaslov **Zaključak** nije obavezan) kojim se obično preporučuju pravci daljih istraživanja.

M e t a a n a l i z a, studija o studijama do 16 stranica, predstavlja analitičkosintetičku studiju većeg broja studija o nekoj značajnoj temi, uz analizu suprotstavljenih stavova i procenu praktične primenjivosti, dopušta preporuke i zaključivanje na osnovu tuđih podataka i mora da ima jasno formulisan zaključak i strukturisani apstrakt od 250 do 300 reči na srpskom i na engleskom jeziku.

A k t u e l n a t e m a, na 8–10 stranica, razmatra neko savremeno, nerešeno ili kontradiktorno pitanje od teorijskog i praktičnog značaja uz iznošenje sopstvenih rezultata istraživanja ili najnovijih važnih podataka iz literature. Konstrukcija članka je slobodna i nije obavezan zaključak, ali su poželjne kratke zaključne napomene sa jasnom porukom.

S e m i n a r p r a k t i č n o g l e k a r a, s t o m a t o l o g a ili f a r m a c e u t a, do 8 stranica teksta zajedno sa prilozima sa praktične tačke gledišta razmatra neko aktuelno pitanje iz prakse i preporučuje stavove koji doprinose poboljšanju profilakse, dijagnostike ili lečenja, odnosno rešavanja nekog problema od značaja za svakodnevni rad zdravstvenih stručnjaka.

K a z u i s t i k a, prikaz jednog ili nekoliko slučajeva oboljenja do 6 stranica i prilozi, obično didaktičkog karaktera (dijagnostički, terapijski ili iz domena preventivne medicine). Mogu se prikazivati i slučajevi vrlo retkih oboljenja ukoliko su od značaja za diferencijalnu dijagnozu. Uz rad se piše i strukturisani apstrakt na srpskom i engleskom jeziku (do 150 reči).

Za i s t o r i j u m e d i c i n e, tekst i prilozi do 16 stranica, objavljuju se materijali od interesa za rasvetljavanje pojedinih događaja iz istorije medicine, a posebno vojne medicine.

P r i k a z i k n j i g a, ne smeju biti duži od dve stranice i sadrže osnovne podatke o knjizi (autori, izvorni naslov, izdavač, mesto i godina izdanja), kratak sadržaj i pretpostavljeni domen interesovanja. Prikaz je osnovna informacija o publikaciji, ali može da sadrži i kritičke komentare.

L i č n i s t a v o v i, k o m e n t a r i i p i s m a u r e d n i š t v u mogu da se odnose na tekstove objavljene u „Vojnosanitetskom pregledu“, na teme od značaja za medicinsku praksu, uopšte, kao i na knjige (monografije) od posebnog medicinskog ili vojnosanitetskog značaja. Ne bi trebalo da su duži od 3 do 4 stranice, ali o tome odlučuje glavni i odgovorni urednik. Pišu se slobodno uz eventualno navođenje podataka iz literature. Objavljuju se prema odluci glavnog i odgovornog urednika.

Izveštaji sa naučnih i stručnih skupova predstavljaju kratak prikaz (do dve stranice) rada skupa uz isticanje najvažnijih referata ili zaključaka, odnosno preporuka od značaja za širi krug čitalaca „Vojnosanitetskog pregleda“.

Radovi se objavljuju na engleskom jeziku sa apstraktom na srpskom i engleskom za originalne članke, metaanalize i kazuistiku.

U celom radu obavezno je korišćenje međunarodnog sistema mera (SI). Izuzetak čine krvni pritisak (mm Hg) i temperatura (°C).

Prilikom pisanja radova koriste se standardne skraćenice, ali ne u naslovu i apstraktu. Pun naziv sa skraćenicom u zagradi navodi se u prvom pominjanju, a dalje u tekstu samo skraćenice.

Za pisanje rukopisa koristi se *word* tekst, sa proredom 1,5. na formatu A4 samo sa jedne strane, sa levom marginom od **4 cm.** Koristi se font veličine 12 (preporučuje se izvorni ***Times New Roman***), i izbegava **bold** i kurziv (*italic*) koji su rezervisani za podnaslove.

Prispeli radovi (bez imena autora) upućuju se na recenziju kod najmanje dva urednika/recenzenta. Primedbe i sugestije urednika i recenzenata (bez imena recenzenta) dostavljaju se autoru radi konačnog oblikovanja rada.

Prihvaćen rad, nakon stručne i redakcijske obrade upućuje se na **autorsko čitanje** pre publikovanja**,** korespondirajućem autoru putem Aseestant – sistema za uređivanje časopisa. Evantualne ispravke trebalo bi izvršiti u roku od dva dana. U ovoj fazi nije moguće izvršiti opsežnije izmene, već samo ispravke slovnih i drugih sitnih grešaka.

Ključne reči ne podležu autorskoj korekturi, pošto su one deskriptori iz Tezaurusa koje određuju stručni indekseri. Ukoliko ispravljeni tekst ne bude vraćen u tom roku, smatraće se da autor nema primedbi. *Rukopisi radova prihvaćenih za štampu ne vraćaju se autoru.*

Priprema rada

Radovi se pripremaju u skladu sa **Vankuverskim dogovorom** (V izdanje, revizija iz 1997) postignutim na inicijativu Međunarodnog komiteta urednika medicinskih časopisa (*International Committee of Medical Journal Editors)* ***Uniform Requirements for Manuscripts Submitted to Biomedical Journals. Ann Intern Med 1997; 126: 36–47. Updated October 2001.***

Delovi rada su: naslovna strana, apstrakt sa ključnim rečima, tekst rada, zahvalnost (po želji), literatura, prilozi (tabele, slike). Stranice treba numerisati redom (u gornjem ili donjem desnom uglu), počevši od naslovne strane.

1. Naslovna strana

a) Naslov rada trebalo bi da bude kratak, jasan i informativan, na srpskom i engleskom jeziku, bez skraćenica i da odgovara sadržaju rada. Podnaslove treba izbegavati.
b) Ispod naslova navode se puna imena i prezimena autora sa primerenim brojem koautora.
v) Navode se, takođe, puni nazivi organizacijske jedinice i ustanove u kojima je rad pripremljen kao i mesta i države u kojima se ustanove nalaze.
g) Znaci ***, †, ‡, ||, §, ¶, **, †† …** itd. pokazuju redom ustanove/organizacijske jedinice u kojima autori rade.

d) Ime, adresa i telefonski brojevi (fiksni, mobilni, faks) i *e-mail* adresa autora zaduženog za korespondenciju u vezi sa rukopisom.
đ) Ime i adresa autora kome se mogu slati zahtevi za separate.
e) Kratak naslov rada (do 40 znakova, uključujući i beline) na dnu naslovne strane.
ž) Neophodno je jasno navesti i potpisati pojedinačan doprinos koautora izradi studije i objavljivanje rada.

2. Apstrakt i ključne reči

Na drugoj stranici rada nalazi se **strukturisani apstrakt** na srpskom i engleskom jeziku, napisan kratkim i jasnim rečenicama koji obuhvata **Uvod/Cilj rada, Metode (**osnovni postupci, izbor ispitanika ili laboratorijskih životinja; metode posmatranja i analize), **Rezultate** (važni nalazi, konkretni podaci i njihova statistička značajnost) i **Zaključak**. Potrebno je da se naglase novi i značajni aspekti studije ili zapažanja. Strukturisani apstrakt **originalnih članaka i metaanaliza** ne bi trebalo da prelazi **300** reči, a kazuistike **150–200** reči, sa podnaslovima ***Uvod, Prikaz bolesnika* i *Zaključak*.** Apstrakt za članke iz ostalih rublika nije obavezan, a ukoliko autori žele da ga pripreme, ne treba da bude strukturisan (bez podnaslova), i piše se sa najviše **150** reči. Ispod apstrakta, u podnaslovu „Ključne reči“, dati 3–10 ključnih reči ili kratkih izraza koji ukazuju na sadržinu članka. Ključne reči predstavljaju pomoć u indeksiranju i ne moraju se u tom obliku naći u članku jer će se koristiti odgovarajući deskriptori, odnosno termini iz *Medical Subject Headings* (*MeSH*) liste *Index Medicus-a.*

3. Tekst članka

Neophodno je da originalni radovi sadrže poglavlja: **uvod, metode, rezultati** i **diskusija**. **Zaključak** može da bude posebno poglavlje ili se iznosi u poslednjem pasusu diskusije. Samo izuzetno, moguće je spajanje **rezultata** i **diskusije** u jedno poglavlje.

Uvod

U uvodu rada kratko se definiše predmet istraživanja, navode analize za istraživanje, hipoteza (ako postoji)

Metode

Jasno opisati izbor metoda posmatranja ili eksperimentalnih metoda (ispitanici ili eksperimentalne životinje). Identifikovati metode, aparaturu (ime i adresa proizvođača u zagradi) i proceduru dovoljno detaljno da bi se drugim autorima omogućilo ponavljanje rezultata. Za uhodane metode, uključujući i statističke, navesti samo podatke iz literature. Mogu se navesti podaci iz literature i kratak opis za metode koje su objavljene, ali nisu dovoljno poznate. Opisati nove ili značajno modifikovane metode, izneti razlog za njihovo korišćenje i proceniti njihova ograničenja. Tačno identifikovati sve primenjene lekove i hemikalije, uključujući generičko ime, doze i načine primene (*im, per os, iv, sc, ip,* itd). Ne koristiti komercijalna imena lekova i drugih preparata.

Etika

Kada se izveštava o eksperimentu na ljudima potrebno je naglasiti da li je procedura sprovedena u skladu sa etičkim standardima Komiteta za eksperimente na ljudima ili sa Helsinškom deklaracijom. Obavezna je i **saglasnost nadležnog etičkog komiteta**. Nije potrebno iznositi imena, inicijale niti bolničke brojeve ispitanika, naročito ukoliko je materijal ilustrovan. Kod eksperimenata na životinjama naznačiti da li su poštovani principi zaštite životinja iz propisa i zakona.

Statistika

Detaljno opisati statističke metode bi se dobro informisan čitalac mogao da proveri iznesene rezultate. Kada je moguće, kvantifikovati nalaze i prikazati ih uz odgovarajuće pokazatelje greške (npr. SD, SE ili granice poverenja). Izbegavati oslanjanje samo na statističko testiranje hipoteze, kao što je vrednost ***r***, što ne daje značajne kvantitativne informacije. Prodiskutovati prihvatljivost subjekata eksperimenta. Izneti detalje o randomizovanju (metodi slučajnog izbora). Opisati metode za slepo ispitivanje, izneti broj zapažanja. Izvestiti o gubicima kod zapažanja (npr. bolesnici isključeni iz kliničkog ispitivanja). Podaci iz literature za vrstu (tip) studije i statističke metode trebalo bi, ako i kada je moguće, da budu standardni radovi radije nego članci u kojima je to prvi put objavljeno. Naglasiti ako je primenjen neki kompjuterski program koji je u opštoj upotrebi. Opis statističkih metoda stavlja se u poglavlje o metodama.

Rezultati

Rezultate prikazati logičkim redosledom u tekstu, tabelama i ilustracijama navedenim, takođe, po redosledu. Nije potrebno ponavljati sve podatke iz tabela ili ilustracija unutar teksta, već samo naglasiti ili sumirati značajna zapažanja.

Kada se sumiraju rezultati, potrebno je naglasiti kojom statističkom metodom su analizirani. Tabele i slike ograničiti na one koje su neophodne da bi se objasnili i podržali stavovi u radu.

Slike su poželjnije umesto tabela sa mnogo podataka. Ne duplirati prikazivanje podataka slikom i tabelom. Definisati statističke termine, skraćenice i većinu simbola.

Diskusija

Naglasiti nove i značajne aspekte studije i zaključke koji iz njih slede. Ne ponavljati detaljno podatke ili drugi materijal koji je već prikazan u **uvodu** ili **rezultatima**. U diskusiju uključiti ono ošto proističe iz nalaza, kao i ograničenja i razloge za buduća istraživanja. Posmatranja dovesti u vezu sa drugim relevantnim studijama, u načelu iz poslednje tri godine, a samo izuzetno i starijim. Povezati zaključke sa ciljevima rada, ali izbegavati kategorične tvrdnje i zaključke koje podaci iz rada ne podržavaju u potpunosti. Izbegavati isticanje prednosti u nečemu i ukazivanje na rad koji nije dovršen. Izneti nove hipoteze kada je to opravdano i jasno ih naznačiti kao takve. Kada je to primereno, mogu se uključiti i preporuke.

Zaključak

U zaključku dati kratke zaključne napomene sa jasnom porukom koja su proističe iz rezultata istraživanja.

4. Zahvalnost

Posle zaključka, a pre navođenja literature, kada je to potrebno, izneti u jednoj ili više rečenica doprinos osobe kojoj treba odati priznanje, ali koja ne zaslužuje koautorstvo, kao što je podrška, zahvalnost za tehničku pomoć, zahvalnost za finansijsku i materijalnu pomoć, uz naznačavanje vrste pomoći itd.

5. Literatura

Potrebno je da se literatura numeriše onim redosledom kojim se na nju upućuje u tekstu, tabelama i legendama i to **arapskim brojevima**. Svi podaci o citiranoj literaturi moraju biti tačni. Preporučuje se citiranje samo radovi objavljenih u časopisima koje indeksiraju *Current Contents*, *Index Medicus* (*Medline*) ili *Excerpta Medica*. **Svi radovi, bez obzira na jezik izvora**, citiraju se na engleskom jeziku, a izvorni jezik navodi u zagradi, iza naslova.

Primeri citiranja koji slede u skladu su sa *Index*-om *Medicus*-om. Ne prihvata se citiranje apstrakata, sekundarnih publikacija, usmenih saopštenja, neobjavljenih radova, službenih i poverljivih dokumenata. Može se prihvatiti citiranje radova prihvaćenih za štampu, u toku postupka pripreme,tako što se navodi naziv i stavlja u zagradu časopisa *in press*. Informacije iz rukopisa koji su predati ali još nisu prihvaćeni za štampu u tekstu se citiraju kao neobjavljeni podaci i ne navode se u literature.

Primeri pravilnog navođenja literature:

R a d o v i u č a s o p i s i m a

(1) Standardni članak u časopisu (navesti sve autore do 6, posle i dodati et al.
Jurhar-Pavlova M, Petlichkovski A, TrajkovD, Efinska-Mladenovska O, Arsov T, Strezova A, et al. Influence of the elevated ambient temperature on immunoglobulin G and immunoglobulin G subclasses in sera of Wistar rats. Vojnosanit Pregl 2003; 60(6): 657–61.
Ako časopis ima kontinualno straničenje u celom volumenu, poželjno je navesti broj sveske.

(2) Organizacija kao autor
The Cardiac Society of Australia and New Zealand. Clinical exercise stress testing. Safety and performance guidelines. Med J Aust 1996; 164: 282–4.

(3) Bez autora
Cancer in South Africa [editorial]. S Afr Med J 1994; 84: 15.

(4) Volumen sa suplementom
Tadić V, Ćetković S, Knežević D. Endogenous opioids release: an alternative mechanism of cyanide toxicity? Iugoslav Physiol Pharmacol Acta 1989; 25 Suppl 7: 143–4.

(5) Sveska sa suplementom
Dimitrijević J, Đukanović Lj, Kovačević Z, Bogdanović R, Maksić Đ, Hrvačević R, et al. Lupis nephritis: histopathologic features, classification and histologic scoring in renal biopsy. Vojnosanit Pregl 2002; 59 (6 Suppl): 21–31.

6) Volumen sa dêlom (Pt)
Ozben T, Nacitarhan S, Tuncer N. Plasma and urine sialic acid in non-insulin dependent diabetes mellitus. Ann Clin Biochem 1995; 32 (Pt 3): 303–6.

(7) Sveska sa dêlom
Poole GH, Mills SM. One hundred consecutive cases of flap lacerations of the leg in ageing patients. N Z Med J 1994; 107 (986 Pt 1): 377–8.

(8) Sveska bez volumena
Turan I, Wredmark T, Fellander-Tsai L. Arthroscopic ankle arthrodesis in rheumatoid arthritis. Clin Orthop 1995; (320): 110–4.

(9) Bez volumena i sveske
Browell DA, Lennard TW. Immunologic status of the cancer patient and the effects of blood transfusion on antitumor responses. Curr Opin Gen Surg 1993: 325–33.

(10) Paginacija rimskim brojevima

Fisher GA, Sikic BI. Drug resistance in clinical oncology and hematology. Introduction. Hematol Oncol Clin North Am 1995; 9 (2): xi–xii.

K n j i g e i d r u g e m o n o g r a f i j e

(11) Pojedinac kao autor

Ringsven MK, Bond D. Gerontology and leadership skills for nurses. 2nd ed. Albany (NY): Delmar Publishers; 1996.

(12) Urednik (editor) kao autor

Balint B, editor. Transfusiology. Belgrade: Zavod za udžbenike i nastavna sredstva; 2004. (Serbian)

(13) Poglavlje u knjizi

Mladenović T, Kandolf L, Mijušković ŽP. Lasers in dermatology. In: *Karadaglić Đ*, editor. Dermatology. Belgrade: Vojnoizdavački zavod & Verzal Press; 2000. p. 1437–49. (Serbian)

(14) Zbornik radova sa kongresa

Kimura J, Shibasaki H, editors. Recent advances in clinical neurophysiology. Proceedings of the 10th International Congress of EMG and Clinical Neurophysiology; 1995 Oct 15–19; Kyoto, Japan. Amsterdam: Elsevier; 1996.

(15) Rad iz zbornika

Bengtsson S, Solheim BG. Enforcement of data protection, privacy and security in medical informatics. In: *Lun KC, Degoulet P, Piemme TE, Rienhoff O,* editors. MEDINFO 92. Proceedings of the 7th World Congress on Medical Informatics; 1992 Sep 6–10; Geneva, Switzerland. Amsterdam: North-Holland; 1992. p. 1561–5.

(16) Disertacija

Knežević D. The importance of decontamination as an element of complex therapy of poisoning with organophosphorous compounds [dissertation]. Belgrade: School of Veterinary Medicine; 1988 (Serbian).

O s t a l i o b j a v l j e n i m a t e r i j a l i

(17) Novinski članak

Vujadinović J. The inconsistency between federal and republican regulation about pharmacies. In between double standards. Borba 2002 February 28; p. 5. (Serbian)

(18) Sveto pismo

Serbian Bible. Belgrade: British and Foreign Biblical Society; 1981. Book of Isaiah 2: 19–22. (Serbian)

(19) Rečnici i slične reference

Kostić AĐ. Multilingual Medical Dictionary. 4th Ed. Belgrade: Nolit; 1976. Erythrophobia; p. 173–4.

N e o b j a v l j e n i m a t e r i j a l

(20) U štampi (in press)

Pantović V, Jarebinski M, Pekmezović T, Knežević A, Kisić D. Mortality caused by endometrial cancer in female population of Belgrade. Vojnosanit Pregl 2004; 61 (2): *in press.* (Serbian)

E l e k t r o n s k i m a t e r i j a l

(21) Članak u elektronskom formatu

Morse SS. Factors in the emergence of infectious disease. Emerg Infect Dis [5serial online] 1995 Jan–Mar. Dostupno na URL: http://www.cdc.gov/ncidod/EID/eid/htm

(22) Monografija u elektronskom formatu

CDI, clinical dermatology illustrated [monograph on CD-ROM]. Reeves JRT, Maibach H. CMEA Multimedia Group, producers. 2nd ed. Version 2.0. San Diego: CMEA; 1995.

(23) Kompjuterska datoteka

Hemodynamics III: the ups and downs of hemodynamics [computer program]. Version 2.2. Orlando (FL): Computerized Educational Systems; 1993.

Prilozi

Sistem **Aseestant: elektronsko uređivanje časopisa,** omogućuje individualno postavljanje priloga koji mogu biti u sastavu *word* datoteke, prema uputstvu "Vojnosanitetskog pregleda" iza liste literature.

Tabele

Svaka tabela kuca se sa dvostrukim proredom na posebnom listu hartije, ne u obliku fotografije, obeležena redosledom pojavljivanja arapskim brojem u desnom uglu (**Tabela 1**) sa kratakim naslovom. Svaka kolona treba da ima kratko ili skraćeno zaglavlje. Objašnjenja se daju u fusnoti, ne u zaglavlju. U fusnoti se objašnjavaju sve nestandardne skraćenice. U te svrhe mogu se koristiti simboli sledećim redosledom: ***, †, ‡, §, ||, ¶, **, ††,** itd.

Označiti statističke mere varijacije kao što su standardna devijacija (SD) i standardna greška (SE) srednje vrednosti ($\bar{x}$).

Ne koristiti horizontalne i vertikalne crte za razdvajanje redova i kolona u tabeli.

Svaka tabela obavezno se pominje u tekstu.

Ako se koriste tuđi podaci iz objavljenog ili neobjavljenog izvora, neophodna je saglasnost autora i navođenje kao i svakog drugog podatka iz literature.

Broj tabela trebalo bi uskladiti sa dužinom teksta.

Ilustracije (slike)

Svi grafički prilozi – fotografije, crteži, grafikoni, dijagrami, šeme – nazivaju se **slike** i predaju se u dva primerka (fotografije u jednom), oštre, crnobele na glatkom i sjajnom papiru, do formata dopisnice, a maksimalno 20 × 25 cm. Slova, brojevi i simboli jasni i ujednačeni, trebalo bi da budu dovoljne veličine da prilikom umanjivanja ostanu čitljivi. Naslovi i detaljna objašnjenja ne pišu se na samoj slici nego na legendama.

Svaku sliku na poleđini obeležiti brojem slike, imenom prvog autora (ne pisati direktno na fotografijama jer ih to oštećuje). Izbegavati upotrebu spajalica na fotografijama. Slike ne treba lepiti na karton.

Ako se koriste fotografije osoba (bolesnika), lik mora biti nejasan ili je potrebno dobiti pisanu dozvolu bolesnika sa fotografije za njeno korišćenje. Na prilozima (snimci rendgenom, skenerom, ultrazvukom itd) ukloniti sve što može da identifikuje bolesnika. Slike obeležiti brojevima onim redom kojim se navode u tekstu. Ukoliko je slika već negde objavljena potrebno je citirati izvor uz eventualno pisano odobrenje ako se radi o zaštićenom materijalu.

Legende za ilustracije

Legende za ilustracije pišu se na posebnom listu hartije, duplim proredom, koristeći arapske brojeve (**Sl. 1; Sl. 2** itd). Ukoliko se koriste simboli, strelice, brojevi ili slova za objašnjavanje pojedinih delova ilustracije, svaki pojedinačno treba objasniti u legendi. Za fotomikrografije treba navesti unutrašnju skalu i metod bojenja.

Merne jedinice

Koristiti mere za oblast hematologije i kliničke hemije iz Međunarodnog sistema mera (SI). Krvni pritisak izražavati u mm Hg, a temperatura u °C.

Skraćenice i simboli

Koristiti samo standardne skraćenice, ali ne u naslovu i apstraktu. Pun naziv sa skraćenicom u zagradi treba dati kod prvog pominjanja, u daljem tekstu dovoljna je samo skraćenica. Rečenice na srpskom jeziku nije poželjno počinjati skraćenicom, kao ni brojem, niti datumom.

VOJNOSANITETSKI PREGLED
VOJNOMEDICINSKA AKADEMIJA
Crnotravska 17, 11040 **Beograd**, **Srbija**
Tel/Fax: +381 11 2669689
vmaini1@EUnet.rs
vmavsp@hotmail.com

Časopis „Vojnosanitetski pregled“ izlazi godišnje u 12 brojeva.

Godišnja pretplata za 2013. godinu iznosi: 5 000 dinara za građane Srbije, 10 000 dinara za ustanove iz Srbije i 150 € za strane državljane i ustanove. Sredstva se uplaćuju na tekući račun Vojnomedicinske akademije Beograd kod Uprave za javna plaćanja u Beogradu broj: **840-941621-02 VMA (za Vojnosanitetski pregled ili za VSP)**. Uplatnicu (dokaz o uplati) dostaviti lično ili poštom (pismom, faksom, *e-mail*-om). Za zaposlene u MO i Vojsci Srbije moguća je i pretplata u 12 mesečnih rata putem trajnog naloga, tj. „odbijanjem od plate“. Popunjen obrazac poslati na adresu VSP-a.

PRIJAVA ZA PRETPLATU NA ČASOPIS „VOJNOSANITETSKI PREGLED“

Ime i prezime ili naziv ustanove	
Jedinstveni matični broj građana	
Poreski identifikacioni broj (PIB) za ustanove	
Mesto	
Ulica i broj	
Telefon / telefaks	

Pretplata na časopis „Vojnosanitetski pregled“ (zaokružiti):

1. Lično. Dokaz o pretplati dostavljam uz ovu prijavu.
2. Za pripadnike MO i Vojske Srbije: Dajem saglasnost da se prilikom isplate plata u Računovodstvenom centru MO iz mojih prinadležnosti obustavlja iznos mesečne rate (pretplate).
3. Virmanom po prijemu profakture.

Datum _____	Potpis _____

VOJNOSANITETSKI PREGLED
VOJNOMEDICINSKA AKADEMIJA
Crnotravska 17, 11040 **Beograd**, **Srbija**
Tel/Fax: +381 11 2669689
vmaini1@EUnet.rs
vmavsp@hotmail.com

Časopis „Vojnosanitetski pregled“ izlazi godišnje u 12 brojeva.

Godišnja pretplata za 2013. godinu iznosi: 5 000 dinara za građane Srbije, 10 000 dinara za ustanove iz Srbije i 150 € za strane državljane i ustanove. Sredstva se uplaćuju na tekući račun Vojnomedicinske akademije Beograd kod Uprave za javna plaćanja u Beogradu broj: **840-941621-02 VMA (za Vojnosanitetski pregled ili za VSP)**. Uplatnicu (dokaz o uplati) dostaviti lično ili poštom (pismom, faksom, *e-mail*-om). Za zaposlene u MO i Vojsci Srbije moguća je i pretplata u 12 mesečnih rata putem trajnog naloga, tj. „odbijanjem od plate“. Popunjen obrazac poslati na adresu VSP-a.

PRIJAVA ZA PRETPLATU NA ČASOPIS „VOJNOSANITETSKI PREGLED“

Ime i prezime ili naziv ustanove	
Jedinstveni matični broj građana	
Poreski identifikacioni broj (PIB) za ustanove	
Mesto	
Ulica i broj	
Telefon / telefaks	

Pretplata na časopis „Vojnosanitetski pregled“ (zaokružiti):

1. Lično. Dokaz o pretplati dostavljam uz ovu prijavu.
2. Za pripadnike MO i Vojske Srbije: Dajem saglasnost da se prilikom isplate plata u Računovodstvenom centru MO iz mojih prinadležnosti obustavlja iznos mesečne rate (pretplate).
3. Virmanom po prijemu profakture.

Datum _____	Potpis _____