YU ISSN 0042-8450 vol. 69, br. 6, 2012.

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: 4 000 dinara za građane Srbije, 8 000 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. 69 No. 6, 2012

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 4,000.00 Din, institutions 8,000.00 Din in Serbia, and foreign subscribers 150 €.

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

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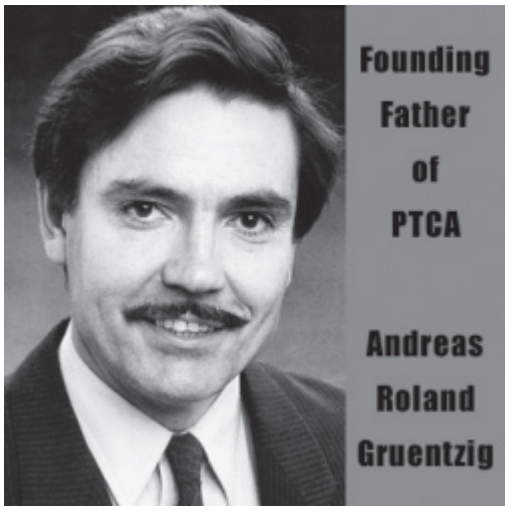

Dr Andreas Roland Greuntzig (25. jun 1939 – 27. oktobar 1985), nemački kardiolog, je utemeljivač perkutane transluminalne koronarne angioplastike (*percutaneous transluminal coronary angioplasty* – PTCA) koja je označila početak nove ere u lečenju koronarne bolesti, ere interventne kardiologije.
Ove godine navršava se 35 godina od prve uspešno izvedene koronarne angioplastike na čoveku koju je dr Greuntzig obavio na bolesniku sa stenozom > 80% u medijalnom delu leve prednje descedentne grane koronarne arterije (vidi str. 541–4).

Dr. Andreas Roland Greuntzig (June 25, 1939 – October 27, 1985), a German cardiologist, is a founder of percutaneous transluminal coronary angioplasty (PTCA) that marked the beginning of a new era in cardiology – interventional cardiology era.
This year marks the 35 years of a successfully performed PTCA an the man. This intervention Dr. Greuntzing performed on the patient with more than 80% stenosis of the medial part of the left anterior descending artery (see pages 541–4).

U V O D N I K

Plagijarizam ili "*copy-paste*" manipulacije

Plagiarism or "copy-paste" manipulation

Silva Dobrić

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Nedavna ostavka mađarskog predsednika Pala Šmita (Pál Schmitt) zbog skandala koji je nastao otkrivanjem da je njegova doktorska teza, odbranjena 1992. godine, kompilacija tekstova drugih autora, ponovo je aktuelizovala problem plagijarizma, jedne od najvećih pošasti savremene naučne publicistike. Preuzimanje rezultata, ideja, delova ili celog teksta nekog rada, bez navođenja originalnog izvora, uz falsifikovanje i fabrikovanje (izmišljanje) rezultata, predstavlja, svakako, najteži oblik nepoštenja u nauci i zahteva najstrože sankcije. Neretko se pribegava i autoplagijarizmu, tj. višekratnom publikovanju istog sopstvenog rada u različitim časopisima sa malim izmenama u naslovu ili sadržaju. Autori koji posežu za ovom vrstom manipulacije, najčešće to čine da bi napredovali u karijeri i obezbedili finansijsku podršku za svoja istraživanja, budući da se u većini zemalja, pa tako i kod nas, broj objavljenih radova, pogotovo u visokorangiranim časopisima, uzima kao merilo vrednosti jednog naučnika. Stoga, pred uredništva naučnih časopisa postavlja se ozbiljan zadatak da pojačaju budnost pri prijemu svakog rukopisa kako bi na vreme otkrili i sprečili eventualne slučajeve plagijarizma ili autoplagijarizma. To zahteva i vreme i energiju, pogotovo danas, kada se svakodnevno objavljuje na hiljade članaka sa rezultatima novih istraživanja, metaanalizama, opštim prikazima ili aktuelnim temama iz neke naučne oblasti, zbog čega je izuzetno teško utvrditi koji je od njih originalan, odnosno da li i u kojoj meri sadrži tuđe rezultate ili sadržaje iz tuđih ili sopstvenih, već objavljenih radova.

Uredništvo i redakcija *Vojnosanitetskog pregleda* (VSP) ovom problemu poklanjaju veliku pažnju već duži niz godina. Zahvaljujući tome, ali i našim recenzentima koji dobro prate novine u svojoj naučnoj oblasti, mnogi slučajevi plagijarizma i/ili autoplagijarizma na vreme su prepoznati i sprečeni. U jednom slučaju smo, čak, izvršili i retrakciju (povlačenje) članka iz časopisa posle saznanja da je isti rad, sa nešto izmenjenim naslovom, već objavljen u drugom časopisu [1]. Pre pet godina u jednom od uvodnika dat je detaljniji osvrt na problem plagijarizma i mere koje će uredništvo VSP preduzimati protiv počinaoca takvih prekršaja [2, 3]. Prva, najblaža

A recent resignation of the Hungarian President Pál Schmitt due to a scandal caused by revealing that his doctoral dissertation, defended in 1992, is a compilation of texts by other authors, has again made plagiarism burning problem, one of the greatest scourges of modern scientific publishing. Downloading results, ideas, parts or entire texts of articles, without referring to original sources, falsification and fabrication of results, are certainly the most serious form of misconduct (dishonesty) in science and require the most stringent sanctions. Repeated publication of one's own article, so-called self-plagiarism, in various journals changing a little the title or content are also very often. Authors who resort to this kind of manipulation, usually do that to make progress in their careers and ensure financial support for their research, since in most countries, including Serbia, the number of published articles, especially in high-ranking journals, is taken as a measure for scientist value. Therefore, editorail boards of scientific journals have a serious task to be more vigilant to receiving any manuscript in order to timely detect and prevent possible cases of plagiarism or self-plagiarism. It requires time and energy, especially today, when thousands of articles are published daily with the results of new research, meta-analyses, a general review or some current topics from the scientific fields, making it extremely difficult to determine which of them is original, or whether and to what extent contains results or contents of their own or others, already published articles.

The editorial Board and Editorial Staff of the *Vojnosanitetski Pregled* (VSP) have devoted great attention to this problem for many years. Thanks to this and our reviewers who regularly follow new scientific research in their field, many cases of plagiarism and / or self-plagiarism have been timely identified and prevented. An article was even retracted by the VSP after finding that the same article under a slightly changed title, had been already published in other journal [1]. Five years ago in one of the editorials a detailed overview of plagiarism and measures which the VSP Editorial Board would take against such misconducts was given [2, 3]. The first, mildest, measure implies sending warning letters to the

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mera podrazumeva slanje pisma upozorenja autorima spornog rada, dok najstroža podrazumeva javno iznošenje svih podataka o učinjenom prekršaju (obaveštavanje institucija i strukovnih udruženja u kojima autori rade, odnosno čiji su članovi, kao i uredništava drugih časopisa), povlačenje spornog rada iz časopisa i zabranu publikovanja autorima u VSP u određenom vremenskom periodu čija dužina zavisi od vrste i učestalosti kršenja etičkih principa publikovanja od strane tih autora [3].

Od početka ove godine svaki rukopis koji pristigne u redakciju VSP proverava se na plagijarizam pomoću softvera *iThenticate* koji, u slučaju postojanja istih delova teksta, identifikuje preklapanje pristiglog rukopisa sa već objavljenim radovima, uz navođenje procenta preklapanja. Posle dobijanja izveštaja sa provere *iThenticate*-om stručni redaktori VSP vrlo lako mogu da ocene da li se radi o slučaju "grubog" ili "blagog" plagijarizma, tj. da li je preklapanje teksta preobimno ili ne. U zavisnosti od stepena preklapanja i mesta u rukopisu koja su istovetna sa tekstom u već objavljenim radovima, zaključuje se o postojanju ili nepostojanju plagijarizma. U slučaju blagog preklapanja, autorima se sugeriše kako da poprave rukopis da se ne bi svrstao u neku od etički spornih kategorija. Rukopisi sa visokim procentom preklapanja odmah se odbijaju, a ubuduće će i prema autorma takvih radova, takođe, biti preduzete određene kaznene mere, zbog očite namere da na nečastan način dođu do publikacije.

Uredništvo VSP odlučno je u nameri da istraje na održanju najviših etičkih standarda u publikovanju. To će, verujemo, unaprediti uspostavljanje pravih vrednosti u naučnoistraživačkom radu i doprineti ugledu koji naš časopis uživa danas u medicinskoj naučnoj javnosti.

authors of a ncontroversial article, while a severe one means the public disclosure of all information on the committed offense (notifying institutions and professional associations in which the authors work, or whose members are, as well as the editorial boards of other journals), article retraction, and prohibition of publishing in the VSP for the authors over a period depend on the type and frequency of violations of ethical principles in publishing by those authors.

Since the beginning of this year, each manuscript received by the VSP Editorial Staff has been checked by the text-matching software iThenticate that, in the case of the same parts of the text, identifies the overlap extent (in percentage) of manuscript with the other, already published articles. After receiving reports from the iThenticate, a editors of the journal can easily estimate whether it is a case of "severe" or "mild" plagiarism, *ie* whether the overlapping text is too large or not. Depending on the overlap degree and places in the manuscript which are identical with the text in already published papers, it can be concluded on the presence or absence of plagiarism. In case of a slight overlap, the authors suggested how to improve the manuscript not to be classified in any of the contentious ethical categories. Manuscripts with a high percentage of overlap are immediately rejected and further on their authors are punished because of the obvious intention to get publication in an unapplicable (dishonorable) way.

The VSP Editorial Board is resolute in the intention to maintain the highest ethical standards in publishing. This, we believe, will promote the establishment of real value in scientific research and publishing and contribute to the reputation our journal enjoys today in the medical scientific community.

L I T E R A T U R A

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ORIGINAL ARTICLES

UDC: 364.4-053.2/.6
DOI: 10.2298/VSP1206469D

Self-assessment of the quality of life of children and adolescents in the child welfare system of Serbia

Samoprocena kvaliteta života dece i adolescenata u sistemu socijalne zaštite Srbije

Maja Damnjanović*, Aneta Lakić*, Dejan Stevanović†, Ana Jovanović*, Jasna Jančić*, Mirjana Jovanović*, Ljubica Leposavić‡

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Abstract

Background/Aim. Children and adolescents who enter a child welfare system are at higher risk of suffering from mental disorders, physical health, and/or social and educational problems than the general population of the same age is. This study was organized with the aim to evaluate the general characteristics of quality of life (QOL) in children and adolescents living in residential and foster care in Serbia. **Methods.** Two hundred and sixteen children and adolescents, aged 8–18 years, from residential and foster care and 238 children and adolescents from the general population participated in the study. QOL was assessed using the Pediatric Quality of Life Inventory (PedsQL) – Serbian version. Three groups were created: residential care group (RCG), foster care group (FCG), and control group (children and adolescents from biological families – CG). Descriptive data were calculated for all questionnaires' scores, while *t*-test and ANOVA were used to compare them. **Results.** The mean value of the total PedsQL was lower in the RCG, 67.47 ± 17.75, than in the FCG and the CG, 88.33 ± 11.27 and 80.74 ± 11.23, respectively. Additionally, the RCG reported lower all PedsQL Scale scores, but the lowest value was for the psychosocial domain. These differences were statistically significant (F value ranged from 17.3 to 49.89, $p < 0.000$). However, only the scores of the RCG were statistically different from the FCG and the CG, while the differences between the FCG and the CG were statistically insignificant ($p > 0.05$). **Conclusion.** Children and adolescents living in residential care have significantly poorer QOL than those living in foster care or in biological families. On the other side, QOL in children and adolescents from foster care is similar to the one of those living in biological families.

Key words:
quality of life; child; adolescent; questionnaires; child, abandoned.

Apstrakt

Uvod/Cilj. Deca i adolescenti koji se nalaze u sistemu socijalne zaštite pod većim rizikom su od mentalnih poremećaja, problema sa fizičkim zdravljem i/ili od socijalnih i obrazovnih probleme, nego deca u opštoj populaciji. Ova studija sprovedena je sa ciljem da se procene opšte karakteristike kvaliteta života kod dece i adolescenata koji žive u domovima i hraniteljskim porodicama u Srbiji. **Metode.** U studiji je učestvovalo 216 dece i adolescenata, uzrasta od 8 do 18 godina, iz domova i hraniteljstva, i 238 dece i adolescenata iz opšte populacije. Kvalitet života procenjivan je pomoću Pedijatrijskog upitnika o kvalitetu života (PedsQL) – srpska verzija. Formirane su tri grupe ispitanika: grupa iz doma (RCG), hraniteljstvo (FCG) i kontrolna grupa (deca i adolescenti iz bioloških porodica – KG). Deskriptivni podaci izračunati su za rezultate svih upitnika, dok su *t*-test i ANOVA korišćeni da ih uporede. **Rezultati.** Srednja vrednost ukupnog skora PedsQL bila je niža u grupi RCG, 67,47 ± 17,75, nego u grupama FCG i KG, 88,33 ± 11,27 i 80,74 ± 11,23. Pored toga, RCG imala je niže skorove PedsQL na svim skalama, ali je najniža vrednost bila za psihosocijalni domen. Ove razlike bile su statistički značajne (F vrednost bila je od 17,3 do 49,89; $p < 0,000$). Međutim, samo su između RCG i FCG i KG postojale statistički značajne razlike, dok su te razlike između FCG i KG bile statistički beznačajne ($p > 0,05$). **Zaključak.** Deca i adolesceati koji žive u domovima imaju znatno niži kvalitet života od onih koji žive u hraniteljstvu ili u biološkim porodicama. S druge strane, kvalitet života dece i adolescenata iz hraniteljstva sličan je onome koji imaju deca iz bioloških porodica.

Ključne reči:
kvalitet života; deca; adolescenti; upitnici; deca, napuštena.

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Introduction

Children and adolescents who enter a child welfare system, whether through foster care, kinship care (placement with relatives), or residential institution care, are a vulnerable population. Over the past decades, it was shown that these children and adolescents are at higher risk of suffering from mental disorders, physical health problems, and/or social and educational problems [1]. The data reported that more than a half of children and adolescents who are in the public care have some kind of psychopathology [1–3], while the prevalence rates of mental health problems could reach up to 70% [4, 5]. Additionally, these children were more likely to have different pediatric illnesses in a higher degree than those from the general population, especially related to physical growth and development [6, 7]. Finally, they also have significantly poorer educational outcomes and they are frequently marginalized, socially withdrawn or isolated [8].

Although the data about health and social problems in this population accumulated over the years, well-being and quality of life (QOL), as more complex health parameters, were insufficiently studied [9–11]. Only two studies were organized about QOL in this population and they demonstrated that children and adolescents who are in the child welfare system have significantly poorer well-being and QOL as compared to the general population of the same age [10, 11]. Additionally, these studies showed some risk factors for QOL, like younger age and residing in an institution, and that mental health problems were associated with impaired daily functioning in this population.

During 2009, we initiated a project with the aims to evaluate mental health status in children and adolescents residing living in residential and foster care in Serbia. As a part of that project, a QOL research was organized in this population. Here, we reported on the general characteristics of QOL in children and adolescents residing in institutions and foster care as compared to those living in biological families.

Methods

The total number of children, adolescents, and young adults up to 26 years of age in the Serbian child welfare system at the time of the study initiation was 5,831 according to the Serbian Ministry of Labour and Social Policy. The study sample was selected from a pool of children and adolescents aged 8–18 residing in residential or foster care. The only inclusion criterion was literacy. Based on the criteria, it was estimated that about 700 of these individuals resided in designated institutions and about 2,000 in foster care.

Three major regional childcare centers in Serbia participated in the study (Belgrade, Niš, Kruševac). All children and adolescents who accepted to participate and who completed the informed consent were included. The informed consent was also obtained from caregivers of the children and adolescents residing in foster care.

School psychologists contacted 300 children and adolescents living with biological parents (aged 8–18 years, equally boys and girls) from four elementary schools in Serbia (Belgrade, Niš, Surdulica) to participate in the study as a control group. They informed all children and adolescents about the purpose of the study, as well as their parents.

Quality of life was assessed with the Pediatric Quality of Life Inventory™ Version 4.0 (PedsQLTM) [12]. This is a self-report questionnaire with 23.5-point-scaled items assessing QOL in the following dimensions: Physical Functioning, Emotional Functioning, Social Functioning, and School Functioning. All the items were reverse-scored and linearly transformed to a 0–100 scale, where higher scores indicated better QOL. The Physical Health score (eight items) was the same as the Physical Functioning Scale and the Psychosocial Health score (15 items) and was the sum of the items divided by the number of items answered in the Emotional, Social and School Functioning scales. The total score was the sum of all the items over the number of items answered on all the scales.

The Serbian PedsQLTM 4.0 Generic Core Scales version was provided by Mapi Institute after the permission had been obtained from its developer, Dr. James W. Varni. The validation study reported the Serbian version was an equivalent to the original with appropriate reliability and validity.

Three study groups were formed: residential care group (RCG), foster care group (FCG) and control group (CG). Descriptive data were calculated for all questionnaires' scores for each group [mean standard deviation (SD) and 95% confidence intervals (CI)]. Independent samples *t*-tests and one-way between-groups analysis of variance (ANOVA) test, with Bonferroni *post-hock* analysis were used to compare PedsQL scores for various groups. The value of $p = 0.05$ was considered as statistically significant.

The Ethics Committee of the Clinic for Neurology and Psychiatry for Children and Youth Belgrade approved the study. The proper authorities of the participating childcare centres approved the study as well.

Results

The overall response rate was 72%. Twentyfive percent of all the contacted children and adolescents from residential and foster care refused to participate, while the rest returned inappropriately completed questionnaires. The residential care group (RCG) included 111, the foster care group (FCG) 105, and the control group (CG) 238 subjects (Table 1). The total amount of missing data was 0.2%.

The mean values with SD and 95% CI for all the groups are given in Table 2. The mean value of the PedsQL was lower in the RCG – 67.47 ± 17.75 than in FCG and CG, 88.33 ± 11.27 and 80.74 ± 11.23, respectively. Additionally, RCG reported lower all PedsQL Scale scores. ANOVA corrected for age (whereas there were significant differences in age between the groups) demonstrated that these differences were statistically significant (F value ranged from 17.3 to 49.89, $p < 0.000$). However, only the scores of the RCG were statistically significantly different from the FCG and the CG, while the differences between the FCG and the CG were not significant ($p > 0.05$).

Table 1

General demographic characteristics of the groups

Parameters	Residential Care (RCG) (n = 111)	Foster Care (FCG) (n = 105)	Control Group (CG) (n = 238)
Age (years), mean (SD)*	14.43 (2.57)	13.15 (2.87)	12.76 (2.17)
Age group, n (%)			
children, 8–12 years	25 (22.5)	49 (46.7)	118 (49.6)
adolescents, 13–18 years	86 (77.5)	56 (53.3)	120 (50.4)
Gender, n (%)			
male	61 (55.0)	41 (39.0)	107 (45.0)
female	50 (45.0)	64 (61.0)	131 (55.0)

*RCG *vs* FCG, $p < 0.000$; RCG *vs* CG, $p < 0.000$; FCG *vs* CG, $p = 0.51$

Table 2

Pediatric Quality of Life Inventory (PedsQL) General Scale Scores' mean (M), standard deviation (SD) and 95% confidence interval (CI) in the studied groups

PedsQL	Groups	M	SD	CI 95% lower bound	CI 95% upper bound	F* *p*
Emotional functioning	Residential Care (n = 111)	56.26	21.46	52.74	59.77	20.76** (0.000)
	Foster Care (n = 104)	72.45	19.14	68.82	76.08	
	Control (n = 238)	70.65	17.34	68.25	73.05	
Social functioning	Residential Care (n = 111)	70.2	26.03	66.82	73.58	49.89** (0.000)
	Foster Care (n = 104)	88.4	14.81	84.91	91.9	
	Control (n = 238)	88.34	14.61	86.03	90.64	
School functioning	Residential Care (n = 111)	64.28	21.94	61.04	67.51	27.66** (0.000)
	Foster Care (n = 104)	79.55	15.75	76.21	82.89	
	Control (n = 238)	78.49	15.49	76.28	80.7	
Physical health	Residential Care (n = 111)	71.37	21.07	68.47	74.26	17.03** (0.000)
	Foster Care (n = 104)	80.53	14.33	77.54	83.53	
	Control (n = 238)	82.32	12.75	80.34	84.3	
Psychosocial health	Residential Care (n = 111)	63.58	18.81	60.9	66.26	49.05** (0.000)
	Foster Care (n = 104)	80.13	12.76	77.37	82.9	
	Control (n = 238)	79.16	12.5	77.33	80.99	
Total	Residential Care (n = 111)	67.47	17.75	65.02	69.92	38.8** (0.000)
	Foster Care (n = 104)	80.33	11.27	77.80	82.87	
	Control (n = 238)	80.74	11.23	79.06	82.41	

*One-way ANOVA corrected for age; **Bonferroni: Residential Care *vs* Foster Care – $p < 0.000$; Residential Care *vs* Control, $p < 0.000$; Foster Care *vs* Control – $p \geq 0.98$

Table 3 shows the mean ± SD PedsQL scores according to gender. Females tended to rate the PedsQL scores lower in the studied groups than males, but there was a statistically significant difference in the FGC in the Emotional and School Functioning scale and the Psychosocial Health. However, males, as well as females, in the RCG had significantly lowered all PedsQL scores than those in the FCG or the CG ($p < 0.000$). Between the FCG and the CG, there were no statistically significant difference ($p > 0.05$).

The mean (SD) of the PedsQL scores according to age, children (8–12 years) and adolescents (13–18 years), were given in Table 4. Only children in the RCG reported lower PedsQL scores than adolescents, but this was not statistically significant ($p > 0.05$). However, children, as well as adolescents, in the RCG had significantly lower all PedsQL scores than those in the FCG or the CG ($p < 0.000$), but between the FCG and the CG there were no statistically significant differences ($p > 0.05$).

Discussion

This is the first study to our knowledge on QOL in children and adolescents from the child welfare system in Serbia and the results indicate that children and adolescents residing in institutions report significantly poorer QOL than those living in foster care or with biological parents.

Analyzing general characteristics of quality of life in these children and adolescents the following was observed.

In general, children and adolescents living in residential care institutions valued their QOL significantly lower across all PedsQL scales than those living in foster care or in biological families. Children and adolescents from residential care especially valued lower emotional functioning and much lower psychosocial than physical health. This difference was negligible in the other two groups. Nevertheless, children and adolescents living in foster care reported similar QOL scores to those living in biological families. These findings partially agree with the previous findings. One study reported that children and adolescents from residential care tended to value their QOL significantly lower, especially in psychological domains, than those from the general population, what agrees with our results [11]. However, the children and adolescents from foster care in this study had similar QOL as the general population, what disagrees with the previously reported lower QOL in this group as compared to the general population [10].

Table 3

Pediatric Quality of Life Inventory (PedsQL) General Scale Scores' mean (M) and standard deviation (SD) in the groups according to gender

PedsQL	Group	Gender	M	SD	*T** (*p*)	F_{Male}** (*p*)	F_{Female}** (*p*)
Emotional functioning	Residential care	Male (n = 61)	58.31	18.44	1.12 (0.27)		
		Female (n = 50)	53.75	24.62			
	Foster care	Male (n = 40)	78.81	17.11	2.7 (0.008)	19.64 (0.000)	11.72 (0.000)
		Female (n = 64)	68.47	19.38			
	Control	Male (n = 107)	72.89	17.29	1.81 (0.07)		
		Female (n = 131)	68.81	17.22			
Social functioning	Residential care	Male (n = 61)	70.98	24.32	0.35 (0.73)		
		Female (n = 50)	69.25	28.19			
	Foster care	Male (n = 40)	87.37	18.32	-0.56 (0.58)	16.64 (0.000)	24.96 (0.000)
		Female (n = 64)	89.05	12.23			
	Control	Male (n = 107)	87.33	14.6	-0.96 (0.34)		
		Female (n = 131)	89.16	14.62			
School functioning	Residential care	Male (n = 61)	64.91	20.23	0.34 (0.74)		
		Female (n = 50)	63.5	24.05			
	Foster care	Male (n = 40)	84	14.36	2.38 (0.02)	18.83 (0.000)	14.14 (0.000)
		Female (n = 64)	76.77	16.05			
	Control	Male (n = 107)	77.73	15.15	-0.68 (0.49)		
		Female (n = 131)	79.1	15.8			
Physical health	Residential care	Male (n = 61)	71.77	21.78	0.22 (0.82)		
		Female (n = 50)	70.87	20.37			
	Foster care	Male (n = 40)	82.27	12.89	1.11 (0.27)	14.42 (0.000)	6.91 (0.001)
		Female (n = 64)	79.45	15.16			
	Control	Male (n = 107)	84.97	11.72	2.95 (0.004)		
		Female (n = 131)	80.15	13.18			
Psychosocial health	Residential care	Male (n = 61)	64.74	17.3	0.72 (0.47)		
		Female (n = 50)	62.16	20.6			
	Foster care	Male (n = 40)	83.39	13.16	2.06 (0.04)	27.47 (0.000)	26.25 (0.000)
		Female (n = 64)	78.1	12.17			
	Control	Male (n = 107)	79.32	12.56	0.97 (0.86)		
		Female (n = 131)	79.03	12.5			
Total	Residential care	Male (n = 61)	68.26	17.31	0.51 (0.61)		
		Female (n = 50)	66.52	18.4			
	Foster care	Male (n = 40)	82.83	11.09	1.87 (0.06)	25.41 (0.000)	18.91 (0.000)
		Female (n = 64)	78.77	11.19			
	Control	Male (n = 107)	82.15	10.69	0.25 (0.08)		
		Female (n = 131)	79.59	11.57			

*** *t*-test; **One-way ANOVA with Bonferroni *post-hoc* analysis:**
male – residential care *vs* foster care, $p \leq 0.002$; residential care *vs* control, $p < 0.000$; foster care *vs* control, $p \geq 0.11$
female – residential care *vs* foster care, $p \leq 0.000$; residential care *vs* control, $p < 0.000$; roster care *vs* control, $p = 1.0$

Girls and boys, separately, from residential care reported significantly lower QOL than those living in foster care or with biological parents. Although girls, generally, valued their QOL lower than boys across all the groups, and there was statistically significant difference in the Emotional and School Functioning scale and the Psychosocial Health only among those from foster care.

The QOL analysis according to age demonstrated that children from residential care valued QOL lower than adolescents, although this difference was not statistically significant. Between the two from foster care, there were similar scores. On the contrary, adolescents from biological families had lower QOL than children, what agreed with the findings from the literature [13, 14]. Nevertheless, children, as well as adolescents, living in residential care institutions had significantly lower all PedsQL scores than those in foster care or biological families. As according to gender, both children and adolescents from foster care had similar QOL sores as those from biological families. The above findings could indicate that children from residential care tend to be more vulnerable group or that adolescents are much more resilient.

The strengths of the present study are the following. First, its good response rate of 72%. Second, the study included both children and adolescents from residential and foster care, which is about 8.5% of this population. Another strength of the study was the availability of a comparison of the group assessed using the same measure, which made it possible to compare the QOL of children in the child welfare system with that of a comparable group of children in the community. Finally, QOL was assessed with a psychometrically sound and referent measure – PedsQL.

Table 4

Pediatric quality of life inventory (PedsQL) General Scale Scores' mean (M) and standard deviation (SD) in the groups according to age

PedsQL	Groups	Age	M	SD	T (*p*)	$F_{Children}$ *	$F_{Adolescents}$ **
Emotional functioning	Residential care	Children (n = 25)	53.20	13.6	-0.81 (0.42)	20.25 (0.000)	14.53 (0.000)
		Adolescents (n = 86)	57.15	23.24			
	Foster care	Children (n = 49)	73.64	16.84	0.63 (0.53)		
		Adolescents (n = 55)	71.38	21.07			
	Control	Children (n = 118)	75.74	16.39	4.68 (0.000)		
		Adolescents (n = 120)	65.64	16.84			
Social functioning	Residential care	Children (n = 25)	55.8	23	-3.28 (0.01)	46.59(0.000)	21.41 (0.000)
		Adolescents (n = 86)	74.38	25.47			
	Foster care	Children (n = 49)	86.22	15.92	-1.43 (0.15)		
		Adolescents (n = 55)	90.35	13.59			
	Control	Children (n = 118)	89.44	14.03	1.16 (0.25)		
		Adolescents (n = 120)	87.25	15.14			
School functioning	Residential care	Children (n = 25)	55	19.47	-2.47 (0.16)	26.78 (0.000)	14.74 (0.000)
		Adolescents (n = 86)	66.97	22			
	Foster care	Children (n = 49)	79.15	17.12	-0.3 (0.76)		
		Adolescents (n = 55)	79.91	14.57			
	Control	Children (n = 118)	81.10	15.27	2.61 (0.01)		
		Adolescents (n = 120)	75.92	15.33			
Physical health	Residential care	Children (n = 25)	64.26	16.13	-1.94 (0.05)	24.65 (0.000)	7.69 (0.001)
		Adolescents (n = 86)	73.43	21.95			
	Foster care	Children (n = 49)	82.59	10.79	1.28 (0.2)		
		Adolescents (n = 55)	78.7	16.76			
	Control	Children (n = 118)	84.11	13.02	2.16 (0.03)		
		Adolescents (n = 120)	80.56	12.28			
Psychosocial health	Residential care	Children (n = 25)	54.66	13.92	-2.77 (0.07)	51.77 (0.000)	25.89 (0.000)
		Adolescents (n = 86)	66.17	19.32			
	Foster care	Children (n = 49)	79.67	11.8	-0.33 (0.74)		
		Adolescents (n = 55)	80.55	13.66			
	Control	Children (n = 118)	82.09	12.18	3.69 (0.000)		
		Adolescents (n = 120)	76.27	12.18			
Total	Residential care	Children (n = 25)	59.46	11.03	-2.63 (0.01)	49. 86 (0.000)	18.57 (0.000)
		Adolescents (n = 86)	69.8	18.68			
	Foster care	Children (n = 49)	81.13	9.96	0.62 (0.53)		
		Adolescents (n = 55)	79.62	12.37			
	Control	Children (n = 118)	83.1	11.12	3.28 (0.000)		
		Adolescents (n = 120)	78.41	10.98			

*** *t*-test; **One-way ANOVA with Bonferroni *post-hoc* analysis:**
male – residential care *vs* foster care, $p \leq 0.000$; residential care *vs* control, $p < 0.000$; foster care *vs* control, $p \geq 0.1$
female – residential care *vs* foster care, $p \leq 0.009$; residential care *vs* control, $p < 0.009$; foster care *vs*. control, $p \geq 0.25$

The main limitation of the present study was the availability of only two studies from the literature that evaluated QOL in this population, so it was not possible to compare the results properly. Then, we do not have any results from systemic studies on the characteristics of our child welfare system and its implications in the lives of these children. Therefore, the findings of the present study were not possible to comment in respect of it. Additionally, there was a selection bias, whereas only children and adolescents from four child welfare system centers in Serbia participated. Finally, using only self-assessment questionnaire and not considering QOL assessments from proxies could be also limiting.

Conclusion

In general, as well as according to gender and age, children and adolescents living in residential care institutions reported significantly lower QOL than those living in foster care or in biological families, especially in the psychosocial domains. However, the quality of life of children and adolescents from foster care is similar to the ones of those living with biological parents. With the results of the incoming study, where different risk and protective factors for QOL and mental health would be reported, it will be possible to suggest interventions to improve QOL of these children and adolescents, as well as to evaluate different models of QOL.

Acknowledgements

We would like to thank to all the children and adolescents who accepted to participate in the study and to their caregivers. Many thanks to Dobrila Grujić (Foster Care Centre of Serbia, Belgrade, Serbia), Zoran Milačić (Residential Care for Infants, Children and Youth Belgrade), Violeta Blagojevic (Foster and Residential Care for Children Nis), coordinators, and psychologists from the participating care centers.

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Received on July 05, 2010.
Revised on July 19, 2010.
Accepted on July 27, 2010.

ORIGINALNI ČLANAK

UDC: 340.64:616-091.1/.6
DOI: 10.2298/VSP1206475M

Premortalni podaci u procesu identifikacije skeletnih ostataka

Premortal data in the process of skeletal remains identification

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Apstrakt

Uvod/Cilj. Prilikom ekshumacije masovnih grobnica ili kod masovnih nesreća, osnovni zadatak forenzičara je utvrđivanje identiteta osobe. Utvrđeni rezultati zavise od stepena izraženosti postmortalnih promena i upoređuju se sa premortalnim podacima koji se dobijaju od članova porodica nestalih i nastradalih. Iskustvo prilikom ekshumacije pokazalo je velike razlike između rezultata dobijenih ekshumacijom i premortalnih podataka. Cilj rada bio je da se ukaže na postojanje razlika između premortalnih podataka i rezultata dobijenih ekshumacijom za iste parametre i usmeri uzimanje premortalnih podataka na specifične odlike skeleta. **Metode.** Izvršili smo uporednu analizu rezultata ekshumacije skeletnih ostataka iz jedne masovne grobnice i premortalnih podataka za identifikovane osobe. Najmanji broj individua u ovoj masovnoj grobnici izračunat je na osnovu najvećeg broja gornjih okrajaka desnih butnih kostiju, na osnovu čega je izračunato da je najmanji broj individua u masovnoj grobnici 48. Od toga je identifikovano 27 osoba. Određivanje pola vršeno je prema metričkim i morfološkim odlikama karličnih kostiju. Individualna starost u momentu smrti određivana je na osnovu morfoloških odlika preponske simfize, morfologije sternalnog okrajka rebara i zapažanja na ostalim delovima skeleta. Visinu smo izračunavali na osnovu srednje vrednosti proizvoda dužine dugih kostiju i koeficijenata po Rollet-u **Rezultati.** Potpuno poklapanje postajalo je u pogledu pola osoba. Godine starosti su se poklapale u određenom intervalu koji može da se odredi na osnovu skeletnih ostataka. Svi ostali parametri su se razlikvali, što je znatno otežalo identifikaciju. **Zaključak.** Premortalni podaci su važan elemenat identifikacije. Trebalo bi da ih uzima lekar forenzičar i da se usmeravaju na detaljnije opisivanje skeletnog sistema.

Ključne reči:
medicina, sudska; grobnice, masovne; kostur; antropometrija; antropologija, forenzička; porodica; upitnici.

Abstract

Background/Aim. The basic task of a forensic examiner during the exhumation of mass graves or in mass accidents is to establish identity of a person. The results obtained through these procedures depend on the level of perceptibility of post mortal changes and they are compared with premortal data obtained from family members of those missing or killed. Experience with exhumations has shown significant differences between the results obtained through exhumation and the premortal data. The aim of the study was to suggest the existance of the difference between premortal data and the results obtained by exhumation regarding the some parameters, as well as to direct premortal data colection to the specific skeletal forms. **Methods.** We performed comparative analysis of the results of exhumation of skeletal remains in a mass grave and the premortal data concerning the identified persons. The least number of individuals in this mass grave was calculated according to the upper parts of the right femur and it helped in calculating the smallest number of individuals in mass graves to be 48. A total of 27 persons were identified. Sex was determined by metrics and morphology of the pelvis. Personal age in the moment of death was determined by morphology features of groin symphisis and morphology of sternal edge of ribs and other parts of scelets observations. The hight was calculated as average results of length of long bones and Rollet coefficients. **Results.** There was a complete match in terms of sex and age matched within an interval that could be established based on the skeletal remains. All the other parameters were different, however, which made identification significantly more difficult. **Conclusion.** The premortal data is an important element of identification process and it should be obtained by the forensic doctor and directed towards more detailed examination of the skeletal system.

Key words:
forensic medicine; mass casualty incidents; skeleton; anthropometry; forensic anthropology; family; questionnaires.

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Uvod

Prilikom ekshumacija masovnih grobnica ili kod masovnih nesreća, osnovni zadatak forenzičkog antropologa je utvrđivanje identiteta osoba. Identifikacija se vrši na leševima, delovima tela i skeletnim ostacima. Obuhvata čitav niz postupaka: daktiloskopiju, opisivanje, fotografisanje, utvrđivanja pola, starosti, visine tela, zubnog statusa, uzimanje uzoraka za DNK analizu i dr [1]. Utvrđeni rezultati upoređuju se sa premortalnim podacima koji se dobijaju od članova potencijalnih porodica nestalih i nastradalih. Kada se ekshumacija vrši nekoliko godina ili nekoliko desetina godina posle sahranjivanja, zbog postmortalnih promena ostaju samo skeletni ostaci. Međutim, postmortalne promene zavise od temperature okoline, vlažnosti, odevenosti leša i niza drugih faktora. Brzina truljenja izražena Casparovim pravilom je na vazduhu četiri puta veća nego u vodi, a u vodi dva puta veća nego u zemlji [1]. Zbog brojnih faktora koji utiču na postmortalne promene i stepen očuvanosti leševa je različit. Navodi se da meko tkivo u zemlju zakopanog leša iščezava nakon 3 do 4 godine [1]. Naše iskustvo demantuje podatke iz literature jer je ekshumacija, koju smo obavili nakon nešto više od godinu dana od nestanka osoba, pokazala da meka tkiva nedostaju u potpunosti. Međutim, u konkretnom slučaju leševi su određeni vremenski period boravili u vodi, a nakon toga prebačeni u zemlju. Boravak u vodi uslovio je ubrzani proces postmortalnih promena.

Cilj rada bio je da se ukaže na postojanje razlika između premortalnih podataka i rezultata dobijenih ekshumacijom za iste parametre i usmeri uzimanje premortalnih podataka na specifične odlike skeleta.

Metode

Izvršili smo uporednu analizu premortalnih podataka na osnovu standardnih upitnika koje su uzimali predstavnici Organizacije za nestala lica i rezultata ekshumacije skeletnih ostataka iz masovne grobnice. Najmanji broj individua u ovoj masovnoj grobnici izračunat je na osnovu najvećeg broja istih kostiju koje su se nalazile u skeletnom materijalu. To su bili gornji okrajci desnih butnih kostiju, na osnovu čega je izračunato da je najmanji broj individua u masovnoj grobnici 48. Do sada je od tog broja identifikovano 27 osoba. Određivanje pola vršeno je prema metričkim i morfološkim odlikama karličnih kostiju (kotiloishiadični indeks, zadnji ugao). Od 27 identifikovanih, 26 bile su osobe muškog pola, a jedna osoba bila je ženskog pola. Individualna starost u momentu smrti određivana je na osnovu morfoloških odlika preponske simfize, morfologije sternalnog okrajka rebara i zapažanja na ostalim delovima skeleta. Telesnu visinu izračunavali smo na osnovu srednje vrednosti proizvoda dužine dugih kostiju i koeficijenata prema Rollet-u. Prema ovoj metodi kod osoba muškog pola dužina žbice množi se koeficijentom 6,86, lakatne kosti koeficijentom 6,41, golenjače koeficijentom 4,53, a lišnjače koeficijentom 4,58. Kod osoba ženskog pola koeficijent kojim se množi dužina žbice je 7,16, lakatne kosti 6,66, golenjače 4,61, a lišnjače 4,66.

Za statističku obradu korišćen je *Microsoft Office Excel*. Prikazane su srednja vrednost ($\bar{x}$) i standardna devijacija (SD) merenih parametara.

Rezultati

Premortalni podaci uzimani su na standardnim upitnicima od strane Međunarodne organizacije za nestala lica, na prosečno 19 strana, od kojih su odgovori na većinu pitanja nedostaljali i uzimani su pet i šest godina nakon nestanka osoba.

Ekshumacijom su pronađeni samo skeletni ostaci, bez mekog tkiva, neki predmeti i delovi odeće.

Zaživotne godine starosti poklopile su se kod 16 (59,25%) slučajeva, kod četiri slučaja nije bilo dovoljno elemenata da se odredi starost, a kod 7 slučajeva starost se nije uklapala u postmortalno određenu granicu. U slučajevima gde je to bilo moguće starost je određivana sa tačnošću u rasponu dve godine, ali tamo gde nije bilo dovoljno elemenata za preciznije određivanje mogući raspon godina bio je 15 (tabela 1).

Tabela 1
Godine starosti u premortalnim podacima i procenjene godine starosti nakon ekshumacije

Premortalni podaci (godine)	Procena nakon ekshumacije (godine)
33	24–28
19	17–19
44	35–45
38	35–45
55	/
48	35–45
62	/
42	35–50
33	28–35
17	17–19
32	28–35
23	30–35
33	40–50
38	35–45
22	20–30
42	/
20	19–20
52	/
32	35–45
25	30–45
21	19–21
33	30–45
33	25–35
16	16–18
25	25– 35
36	40–45
36	30–40
$\bar{x}$ ± SD: 33,70 ± 11,85	32,83 ± 8,73

/ – nije bilo elemenata za određivanje godina starosti

Telesna visina koja se navodila u premortalnim podacima samo u dva slučaja bila je identična sa visinom izračunatom nakon ekshumacije (tabela 2).

Tabela 2
Telesna visina u premortalnim podacima i izračunata visina nakon ekshumacije

Premortalni podaci (cm)	Izračunata visina (cm)
183	168
170	162
170	178
167	178
170	163
168	168
160	170
180	166
178	164
165	170
178	182
170	155
180	173
175	181
175	167
183	177
184	192
165	163
175	168
175	179
175	171
169	177
176	176
178	174
165	169
170	177
175	173
$\bar{x}$ ± SD: 173,29 ± 6,22	171,88 ± 7,69

Telesna konstitucija, u upitnicima je bila označena kao srednja kod 6 slučajeva, a kod jednog kao jače razvijena. Telesna težina opisana je kod šest slučajeva. Zubni status na osnovu zubnog kartona stomatologa nije opisan ni u jednom slučaju, a u tri slučaja bilo je navedeno da je izvađen zub, ali se ne zna lokacija zuba. Ekshumacijom su ustanovljeni skeletni ostaci jedne osobe koji su prema dužini i poprečnim promerima mogli odgovarati jače razvijenoj osobi, a telesna masa nije bila od značaja, jer su bili pronađeni samo skeletni ostaci. Za svih 27 identifikovanih postojali su delovi ili cela donja i gornja vilica i zubni kartoni bili bi od neprocenjive važnosti.

Kosa je bila detaljno opisana u svim slučajevima. Delovi lica (čelo, obrve, oči, nos, uši, usne, kosmatost lica) opisani su delimično u pet slučajeva, a delovi tela (vrat, ruke, noge) u sedam slučajeva (tabela 3). Svi ovi opisi imali bi smisla da je ekshumacija rađena u kraćem vremenskom intervalu nakon smrti, a u konkretnom slučaju nisu pomogli u postupku identifikacije.

Od bolesti u upitnicima navedeni su čir na dvanaestopalačnom crevu i stomačne tegobe uz korišćenje terapije (ranitidin), u jednom slučaju perforacija bubne opne, ožiljci kože kod šest osoba i to u predelu leve ruke (dužine 2 cm), leve strane vrata (dužine 4 cm), prednje strane trupa, desne noge, leve noge i ožiljak od opekotina na obe noge, a u dva slučaja navedene su bubrežne bolesti gde su u jednom slučaju bili navedeni problemi sa bubrezima i lečenje 1996, a u drugom da je 1992. godine osoba bila u bolnici u Švajcarskoj.

Ekshumacijom je pronađena desna golenjača za koju je u donjoj polovini pričvršćena pravougaona uzdužna „šina“ od belog metala sa sedam šrafova koji prolaze kroz celu debljinu kosti, koja je ugrađena od strane lekara u cilju lečenja najverovatnije preloma tog dela kosti (slika 1). U premortalnim podacima za tu osobu naveden je samo ožiljak u predelu desne noge. U jednom slučaju nađeno je nepravilno zadebljanje desne ramenjače, najverovatnije kao posledica sraslog preloma, a u premortalnim podacima za tu osobu ne navodi se nikakva povreda.

Sl. 1 – Golenjača sa metalnom „šinom“

Kao identifikaciona dokumenta koja su nestali imali kod sebe bili su navođeni: studentski indeks, zdravstvena knjižica, crni kožni novčanik, novac i fotografije porodice i prijatelja, vozačka dozvola, pasoš i lična karta. Ekshumacijom je pronađena jedna lična karta i vozačka dozvola. U rubrici fotografije bilo je navedeno da fotografije postoje za sve nestale osobe. U devet slučajeva bilo je navedeno da postoji otisak prsta i u kom Ministarstvu unutrašnjih poslova se nalazi, u jednom slučaju nije bilo otiska prsta, a u ostalih 17 nije bilo podataka. Otisci prsta, takođe, nisu mogli poslužiti u identifikaciji zbog postmortalnih promena na mekom tkivu prstiju. Za sve nestale u premortalnim podacima bilo je navedeno da su bele rase, što se i pokazalo ekshumacijom na osnovu antropoloških odlika lobanje.

U rubrikama za nakit koji su nestali nosili bilo je navedeno: lanac od žutog metala oko vrata, sat od belog metala sa kućištem zelene boje, marke „seiko“, plastični crni sat marke „swatch“, sat od belog metala sa kućištem bele boje, burma od žutog metala, sat od belog metala sa kućištem zelene boje marke „anker“, prsten od žutog metala sa svetlooranž kamenom, sat od belog metala sa braon kućištem i sat od belog metala sa zelenim kućištem. Ekshumacijom je pronađena samo narukvica na crnom koncu sa okruglim perlicama zelene, crne i crvene boje.

Odeća je bila detaljno opisana u smislu materijala, boje i veličine, ali je poklapanje po jednog odevnog predmeta ustanovljeno samo u šest slučajeva ekshumiranih i identifikovanih, a u jednom slučaju su se poklopila dva odevna predmeta. Od ukupno 138 odevnih predmeta poklopilo se samo osam (5,79%) (tabela 4).

Tabela 3

Opisi građe tela i pojedinih delova tela u premortalnim podacima

Skeletni ostaci (br.)	Kostitucija	Telesna masa (kg)	Kosa	Delovi lica	Delovi tela
1.	srednja	72	prirodna, kratka, srednje braon, prava, tanka, razdeljak levo	čelo srednje, obrve lučne, oči srednje braon, nos srednji prav,uši srednje veličine, srednje priljubljene, nema pirsinga, usne srednje	vrat srednji, veličina okovratnika 42, ruke srednje
2.	/	/	prirodna, kratka, tamnobraon, kovrdžava	/	/
3.	/	/	prirodna, kratka, tamnobraon, prava,	/	/
4.	/	/	prirodna, srednje dužine, crna, prava	/	/
5.	/	/	prirodna, kratka, crna, prava	/	/
6.	srednja	65	prirodna, kratka, tamno braon, srednje dužine, talasasta		dužina cipele 44
7.	/	/	prirodna, kratka, tamnobraon, prava	/	/
8.	/	/	prirodna, kratka,svetlobraon, kovrdžava	/	/
9.	/	/	prirodna, kratka, crna, kovrdžava	/	dužina cipela 40
10.	/	/	prirodna, srednja, tamnobraon, prava	/	/
11.	srednja	82	prirodna, kratka, svetlobraon, prava, razdeljak desno	čelo srednje,obrve prave, oči plave, nos srednji, konveksan, uši srednje, osrednje priljubljene, usne srednje, zubi prirodni	vrat srednji, okovratnik 42, ruke srednje, noge duge
12.	/	/	prirodna, kratka, bela, prava	/	/
13.	srednja	/	prirodna, srednje dužine, srednje braon, srednje debljine, kovrdžava	/	/
14.	/	/	prirodna, srednje dužine, proseda, kovrdžava	/	/
15.	srednja	75	prirodna, kratka, crna, debela, prava	čelo srednje, obrve lučne, oči plave, nos srednji, prav, uši srednje, priljubljene, usne srednje,	vrat srednji, okovratnik 42, ruke srednje, noge srednje
16.	/	/	prirodna, srednje dužine, svetlo braon, kovrdžava	/	/
17.	/	/	prirodna, kratka, braon, prava	oči braon	/
18.	/	/	prirodna, kratka, crna, prava	/	/
19.	/	/	prirodna, kratka, svetlobraon, prava	/	/
20.	/	/	kratka, tamnobraon, prava	/	/
21.	/	/	prirodna, srednje dužine, braon, talasasta	oči braon	/
22.	/	/	kratka, crna, prava	/	/
23.	/	/	prirodna, kratka, tamnobraon, prava	/	/
24.	/	/	prirodna, srednje dužine, tamnobraon, prava	/	/
25.	krupna	104	prirodna,srednje dužine,proseda, tanka, prava	/	okovratnik 42, cipele 39
26.	/	/	prirodna, srednje dužine, crna, prava	/	/
27.	srednja	65	prirodna, kratka, crna, srednje debljine, prava	/	cipele 42

/ – nedostaju podaci

Tabela 4

Odevni predmeti navedeni u premortalnim podacima nakon ekshumacije i njihovo poklapanje

Odevni predmeti		
Premortalni podaci (n*)	Podaci sa ekshumacije (n**)	Poklapanje
11	5	/
4	6	/
3	5	/
10	5	1 – majica bela dug rukav
2	3	/
3	3	/
4	2	/
9	4	2 – crni vuneni džemper, pantalone
5	2	/
6	6	1 – farmerke
3	7	1 – trenerka donji deo zelena
6	2	/
3	7	1 – vunene helanke crne
6	8	/
3	6	/
4	/	/
4	1	/
6	2	/
8	6	/
6	2	/
1	7	/
11	6	/
3	6	/
5	7	1 – donji deo trenerke
2	0	/
4	2	1 – donji deo trenerke
4	5	/
Ukupno 138	117	8

n* – broj opisanih odevnih predmeta; n– broj nađenih odevnih predmeta; / – nema poklapanja**

Diskusija

Osnovne informacije do kojih se može doći na osnovu pregleda pronađenih skeletnih ostataka su: da li su u pitanju ljudske kosti ili ne, vreme iz koga datira grobnica, godine starosti, visina, pol, rasa i najmanji broj individua koji se nalazi u određenoj grobnici. Moguće informacije koje se mogu dobiti na osnovu skeletnih ostataka su postojanje povreda, anomalija, oboljenja i neke druge individualno specifične odlike osobe, što sve zajedno pomaže pri identifikaciji [2].

Određivanje pola vršeno je na osnovu odlika karličnih kostiju i tu je postojalo poklapanje sa postmortalnim podacima u svim slučajevima.

Određivanje životnog doba poklopilo se kod 59,25% slučajeva. Najpreciznije su određene godine starosti kod osoba mlađih od 20 godina zbog uočavanja lobanjskih šavova koji nisu srasli i nesrastanja epifize dugih kostiju. Kod osoba starijeg životnog doba manje je precizno određivanje godina starosti zbog individualno različite brzine starenja kostiju.

Mali procenat poklapanja telesne visine posledica je delom neprecizno uzetih premortalnih podataka, a delom i Rollet-ovih formula na osnovu kojih je izvršeno izračunavanje [3]. Naime, prilikom ove, a i drugih ekshumacija, ustanovljeno je da postojeće formule za izračunavanje visine koje datiraju iz XIX i prve polovine XX veka, ne daju pouzdane vrednosti visine za pripadnike naše populacije. Zato su i učestala istraživanja koja daju specifične antropometrijske odlike za svoje populacije [4–6]. Zbog toga smo u okviru našeg forenzičko-antropološkog istraživanja koje je u toku, a na osnovu rendgenskog merenja dužine dugih kostiju na živim osobama i telesne visine, lineranom regresijom dobili formule specifične za našu populaciju. Nadamo se da će pri budućim ekshumacijama korišćenje ovih formula davati preciznije vrednosti visine u okviru naše populacije.

Telesna konstitucija je u upitnicima označena u pet slučajeva kao srednja, a u jednom krupna. Pojam srednja konstitucija uglavnom ne može mnogo pomoći prilikom identifikacije, dok jače razvijena konstitucija može da se poveže sa nalazom kostiju veće dužine, većih poprečnih promera i masivnijih okrajaka. U obrascima premortalnih podataka postoje i opcije za opis oblika glave frontalno i profilno, ali za analizirane slučajeve nijedna nije popunjena, kao ni opcije za oblik čela i brade. Kako je u svim sličajevima navedeno da postoje fotografije osoba, neophodno je da se te fotografije prilože ili odštampaju u obrascu. Na osnovu fotografija mogao bi se odrediti oblik glave, čela i brade, što bi se uporedilo sa pronađenim kostima lobanje i eventualnom superpozicijom došlo do identifikacije [7, 8]. Kosa je opisana detaljno u svim slučajevima, a ekshumacijom zbog ubrzanih postmortalnih promena nije pronađena ni u jednom slučaju.

Noge su posebno opisane u upitnicima u samo dva slučaja i to jednom kao duge, a u jednom kao srednje. Dužina cipela opisana je kod četiri slučaja. Međutim, kod skeletnih

ostataka, imajući u vidu da stopalo čini 26 kostiju poređanih u tri grupe, ne može se precizno odrediti dužina stopala. Obuća se u masovnim grobnicama, kada su u pitanju skeletni ostaci, uglavnom nalazi udaljeno od delova tela. Dužinu obuće bi trebalo opisati sa odećom, ali uz veličinu navesti i boju, materijal, način pertlanja ili drugog zakopčavanja, što bi eventualno pomoglo u identifikaciji.

Ruke su posebno opisane u tri slučaja kao srednje duge. Trebalo bi detaljnije opisati odnos dužina nadlaktice i podlaktice, kao i natkolenice i potkolenice, „X" ili „O" noge. Kako se navodi u udžbeniku Sudske medicine prof. Tasića i sar. [9], čuveni anatom Vesalius je rekao: „Oblik bilo kog stvorenja određen je oblikom skeleta. Ono što su štapovi za šatore, to su kosti za kičmenjake." Oblik grudne kosti u smislu deformiteta, ugnutog sternuma kao familijarne nasledne deformacije ili pak ispupčenog kao posledica rahitisa, treba takođe uvrstiti u uzimanje premortalnih podataka.

U opisima delova tela u upitnicima su navedene veličine okovratnika u četiri slučaja, sve 42, što se kod pronalaženja skeletnih ostataka ne može primeniti, a na pronađenim delovima odeće zbog boravka u vodi oznake veličine nisu bile vidljive. Kod opisa vrata trebalo bi insistirati na dužini, što bi eventualnom rekonstrukcijom pršljenova moglo pomoći rezultatima ekshumacije.

U ukupno 11 slučajeva navedene su neke zdravstvene tegobe nestalih, ali nijedna od njih na skeletnim ostacima nije mogla biti ustanovljena. U slučaju gde je pronađena „šina" koja spaja delove golenjače u premortalnim podacima naveden je samo ožiljak kože desne noge.

U identifikaciji bi bilo od značaja i eventulano postojanje zaživotnog radiografskog snimka lobanje, koji bi se uporedio sa postmortalnim nalazom na kostima lobanje i na osnovu oblika i veličine frontalnih sinusa mogla bi se izvršiti identifikacija.[10] Zaživotni radiografski snimci pomogli bi i kod postojanja akcersornih kostiju stopala, zaraslih preloma kostiju, ortopedskih hirurških intervencija, deformacija pršljenova i sl [11].

Kod opisa odeće i ličnih predmeta, nakita, satova i sl, premortalni podaci su najpotpuniji. Međutim, poklapanje pronađenih odevnih predmeta sa opisanim utvrđeno je kod svega 5,79% slučajeva. Mali procenat poklapanja odevnih predmeta moguće nastaje zbog protoka vremena u toku kojeg su članovi porodice zaboravili šta su nestali imali na sebi od odeće, a delimično zbog toga što su nestali u periodu proteklom od vremena nestanka do smrti mogli promeniti određene delove odeće. Nakit i satovi ne poklapaju se ni u jednom slučaju.

Premortalni podaci predstavljaju važnu kariku u procesu identifikacije. Ukoliko su kvalitetno uzeti i usmereni na prava pitanja, mogu pomoći u procesu identifikacije. Nedovoljno precizni podaci i usmeravanje na pogrešne detalje mogu biti samo problem u procesu identifikacije. Bez obzira na superiornost DNK identifikacije, postoje slučajevi kada ni ta metoda ne može da pomogne. Prilikom nekih ekshumacija zbog mesta na kojem su sahranjeni skeletni ostaci i protoka vremena, DNK identifikacija nije se mogla izvršiti i tada se možemo osloniti samo na premortalne podatke.

U konkretnom slučaju za 27 identifikovanih, dobili smo kod tri slučaja rezultate DNK analize koji su odgovarali jednom od dva nestala brata. Trebalo je na osnovu postojećih premortalnih podataka utvrditi o kojoj osobi se radi. Zbog nedovoljno premortalnih podataka mogli smo u identifikaciji koristiti samo telesnu visinu i životno doba.

Zaključak

Postoji neslaganje između premortalnih podataka i nalaza sa ekshumacije masovnih grobnica. Prilikom uzimanja premortalnih podataka treba imati u vidu postmortalne promene i pitanja usmeriti na skeletnu građu i oboljenja, deformitete i povrede kostiju. Premortalne podatke trebalo bi uvek da uzima lekar forenzičar, ali kako je broj lekara specijalista sudske medicine mali potrebno je izvršiti adekvatnu obuku osoba koje uzimaju premortalne podatke.

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Primljen 7. VII 2010.
Revidiran 7. IX 2010.
Prihvaćen 5. X 2010.

ORIGINAL ARTICLE

UDC: 616.8-009.24-085.213-092.9:661.98
DOI: 10.2298/VSP1206481J

Influence of NG-nitro-L-arginine methyl ester on clinical and biochemical effects of methylene blue in pentylenetetrazole-evoked convulsions

Uticaj NG-nitro-L-arginin metil estra na kliničke i biohemijske efekte metilen plavog kod konvulzija izazvanih pentilentetrazolom

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Abstract

Background/Aim. Despite years of research in a number of experimental models the question whether nitric oxide (NO) and methylene blue (MB) have pro- or anticonvulsant effects remains to be fully resolved. **Methods.** In adult Wistar rats the influence of a nonselective inhibitor of nitric oxide synthase NG-nitro-L-arginine methyl ester (L-NAME, 10 μg) on clinical and biochemical effects of MB (10 μg) given before the intraperitoneally administered chemical convulsant pentylenetetrazole (PTZ, 80 mg/kg) was examined. MB and L-NAME were applied intracerebroventricularly. PTZ application was followed by a 4-minute observation time, after which rats were sacrificed and elements of oxido-reductive balance were measured in a crude mitochondrial fraction of forebrain cortex, hippocampus and striatum. **Results.** Convulsive responses (forelimb dystonia – FLD, generalised clonic- and clonic-tonic convulsions – GCC and GCTC respectively) were observed in all rats received PTZ, together with significantly decreased lipid peroxidation in the forebrain cortex and striatum and increased superoxide dismutase activity in the hippocampus, in comparison to controls (saline treated). It was registered anticonvulsant effects of L-NAME pretreatment. However, these effects were insignificant. In the hippocampus of these animals there was decreased lipid peroxidation ($p < 0.01$, $p < 0.05$ *vs* saline-treated and PTZ-treated rats, respectively) and reverted PTZ-induced increase of superoxide dismutase activity. But MB individually pretreatment significantly decreased the incidence of CTCs and GCCs (FLD: $p = 0.0513$), prolonged the convulsive latent time for FLD, GCTCs and GCCs, in all the examined brain regions increased lipid peroxidation and decreased the level of superoxide anion. Administration of L-NAME 10 minutes before MB reverted all MB-evoked clinical and biochemical effects. **Conclusion.** Methylene blue applied individually before PTZ has strong anticonvulsant effects that were eliminated by L-NAME pretreatment. These effects and changed biochemical parameters in the brains of animals treated by L-NAME before MB in comparison to MB-treated group suggest involvement of NO in MB's effects in the animal model of PTZ-evoked convulsions.

Key words:
seizures; nitric oxide; methylene blue; pentylenetetrazole; rats; oxidoreductases.

Apstrakt

Uvod/Cilj. I pored višegodišnjeg istraživanja na različitim eksperimentalnim modelima, nije potpuno odgovoreno na pitanje da li azot-oksid (NO) i metilen plavo (MP) deluju konvulzivno ili antikonvulzivno. **Metode.** Na odraslim pacovima Vistar soja ispitivan je uticaj NG-nitro-L-arginin metil estra (L-NAME, 10 μg), neselektivnog inhibitora azot oksid sintaze, na kliničke i biohemijske efekte metilen plavog (MP, 10 μg) datog intracerebroventrikularno pre hemijskog konvulziva pentilentetrazola (PTZ, 80 mg/kg), primenjenog intraperitonealno. Pacovi su posmatrani četiri minuta posle davanja PTZ-a, posle čega su žrtvovani i u neprečišćenoj mitohondrijskoj frakciji prednjeg mozga, hipokampusa i strijatuma određivani su parametri oksidoreduktivne ravnoteže. **Rezultati.** Posle primene PTZ-a, konvulzivni odgovor (distonija prednjih nogu – DPN, generalizovane klonične – GKK i generalizovane kloničnotonične konvulzije – GKTK) bio je ispoljen kod svih životinja, kao i statistički značajno sniženje lipidne peroksidacije u kori prednjeg mozga i strijatuma, i povećanje aktivnosti superoksid dizmutaze (SOD) u hipokampusu, u poređenju sa kontrolnom grupom (dobila fiziološki rastvor NaCl). Registrovani su antikonvulzivni efekti L-NAME

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koji nisu bili statistički značajni. U hipokampusu ovih životinja bila je snižena lipidna peroksidacija ($p < 0,01$ u poređenju sa kontrolnom grupom, $p < 0,05$ u poređenju sa životinjama koje su dobile PTZ), kao i aktivnost SOD u poređenju sa životinjama koje su dobile PTZ. Samo metilen plavo dovelo je do statistički značajnog smanjenja incidencije GKK I GKTK (DPN: $p = 0,0513$), produžilo je latentni period DPN, GKK i GKTK, a u svim ispitivanim strukturama mozga bila je povećana lipidna peroksidacija i smanjen nivo superoksidnog anjona. Svi klinički i biohemijski efekti izazvani primenom MP u potpunosti su odstranjeni primenom L-NAME 10 minuta pre davanja MP. **Zaključak.** Metilen plavo, dat samostalno pre PTZ, ispoljio je snažne antikonvulzivne efekte. Nestanak ovih efekata i izmenjeni biohemijski parametri u mozgovima pacova koji su pre MP dobili L-NAME, sugerišu da je NO uključen u efekte MP ispoljene na životinjskom modelu konvulzija izazvanih primenom PTZ-a.

Ključne reči:
konvulzije; azot, oksid; metilensko plavilo; pentilentetrazol; pacovi; oksidoredukcija.

Introduction

An accumulated body of evidence supports multiple physiological as well as pathological roles for nitric oxide (NO) in its free radical form. Acting both presynaptically and postsynaptically NO accomplishes its complex participation in a wide range of physiological and pathophysiological phenomena including regulation of vascular tone, inflammation and signalling in the central nervous system (CNS) *via* polysynaptic interacting circuits [1, 2].

Nitric oxide is synthesised from L-arginine by nitric oxide synthases (NOS) in response to N-methyl-D-aspartate (NMDA) and non-NMDA receptor activation, and it is inhibited by a number of NOS inhibitors, among which is NG-nitro-L-arginine methyl ester (L-NAME). Alternatively, it can be released from NO donors. Besides hemoglobin, NOS and other metaloenzymes which are targets for NO binding, the interaction between NO and heme moiety within the soluble (cytosolic) form of guanylate cyclase (sCG) has particular physiological significance. The consequence of NO binding to sGC is the activation of the latter resulting in an elevation of cyclic guanosine 3',5'-monophosphate (cGMP) and the initiation of a cascade of target cell-specific events [3]. Although much effort has been made to elucidate how NO interacts and stimulates sGC the precise mechanism is still unclear to date.

Despite years of research, the role of NO in the pathogenesis of epilepsy and convulsions still remains controversial. NO can provoke convulsions but also it can exhibit anticonvulsant effects [4–6]. In addition, some convulsive patterns are NO-independent [7, 8]. Such unbalanced reports maybe reflected by the use of different experimental conditions, involving not only different animal species and strains, but also other factors including age, sex, convulsive models, the form, the route and doses of the applied substances.

During convulsions the concentration of cGMP increases in specific brain regions. Within the first four minutes of pentylenetetrazole (PTZ) application, which evokes generalised clonic convulsions (GCCs) and generalised clonic-tonic convulsions (GCTCs) in almost all mice, increases in cGMP (3- to 5-fold) were found primarily in the hippocampus and cerebral cortex but also (to a lesser extent) in the cerebellum [9]. Within the first minute of PTZ application a rapid and very high level of cGMP was found in ventilated and non-ventilated guinea pigs in all the above-mentioned brain structures and also in the striatum, suggesting that increased cGMP is a pathogenic component of PTZ-evoked convulsions. The above mentioned brain structures are most important in convulsions initiation and propagation.

Methylene blue (MB), a highly active redox compound that readily cycles between oxidised (methylene blue) and reduced (leukomethylene blue) states, is a blue-coloured organic dye. Besides in experimental conditions, MB has importance in human clinical practice too, although data about its effects in humans are not uniform [10, 11].

The effects of MB on cGMP and on NO have already been documented in several studies. Since MB is a nonselective and a weak incomplete inhibitor of sGC, this effect of MB on sGC has been widely studied [12, 13]. MB can attenuate cGMP accumulation by inhibiting NO-stimulated cGC without an effect on the basal cGMP level [14]. Masaki and Kondo [15] intracerebrovetricularly administered MB to rats 30 minutes prior to sacrificing them. In whole brain homogenate the level of cGMP decreased in a MB dose-dependent manner. MB administration can also decrease the NOS activity and NO content in certain brain regions and also prevents experimental seizures [16, 17].

The NO-sGC-cGMP signalling pathway is present in virtually all cells but sGC is expressed at particularly high levels within neurons. This pathway is largely influenced by glutamatergic neurotransmission that is modulated in response to PTZ treatment [18].

According to a number of data, as well as to our own results, it is obvious that elements of oxidative/antioxidative balance are changed in the brains of rats with convulsive response to PTZ and upon L-NAME treatment in such kind of experimental convulsions [19, 20]. Because of these facts but also due to MB's ability as well as L-NAME and PTZ to interfere with synthesis/actions of NO, and due to their convulsive/anticonvulsive properties, we sought to determine their influence on both clinical and brain biochemical changes (parameters of oxidative defence and stress) in a PTZ seizure model in rats [5–7, 12, 16, 17].

Methods

Animals and surgery

Experiments were performed on 13-week old male Wistar rats housed in a temperature-controlled room (23 ± 2ºC) with 11-hour light/13-hour dark cycles with free access

to food and water. Experiments were conducted within a period of two weeks always between the hours of 9:00 am and 2:00 pm. Animals were randomly assigned to different drug treatment regimes. All protocols for handling the rats were approved by the local ethical committee for the use of experimental animals (Military Medical Academy, Belgrade, Serbia).

For intracerebroventricular (*icv*) application of drugs a polyethylene cannula was stereotaxically implanted into the left lateral ventricle (coordinates: 1.3 mm behind the bregma, 1.8 mm left from the midline suture, 3.7 mm ventral from the durra [21]) under sodium pentobarbital intraperitoneally (*ip*) anaesthesia (45 mg/kg body weight, Vetanarcol®, Werfft-Chemie, Vienna). The cannula was fixed to the skull with dental cement and two jeweller screws. The postoperative recovery period lasted six days before experiments were continued.

Treatments

Rats were divided into five groups (Figure 1; in each group there were 7–8 rats). PTZ (Sigma Chemical Co., St Louis) was used as a chemoconvulsant. It was applied *ip* in a single dose of 80 mg/kg body weight. The control group received sterile isotonic saline *icv* twice: 30 minutes and 20 minutes before *ip* application of the same solution. Another group of rats also received sterile isotonic saline *icv* twice: 30 and 20 minutes before *ip* application of PTZ (PTZ-treated group). The third group received 10 μg NG-nitro-L-arginine methyl ester (L-NAME, Sigma Chemical Co.) *icv* 30 minutes before PTZ. Ten minutes after the L-NAME injection the rats also received sterile isotonic saline *icv*.

MB (10 μg) was administered *icv* to another two groups of rats 10 minutes after sterile isotonic saline (*icv*) or L-NAME (10 μg, *icv*) [15]. Twenty minutes after MB (30 minutes after sterile isotonic saline/L-NAME treatment), both groups of rats received PTZ. All drugs were dissolved in sterile isotonic saline. Volume of *ip* and *icv* applied solutions were 1 mL/kg body weight and 10 μL, respectively.

Behavioural evaluation

Immediately after PTZ administration the rats were placed individually in transparent perspex cages. The time of observation following PTZ injection was limited to 4-min. During that time convulsive responses were monitored and were graded according to the following scale: fore limb dystonia (FLD), generalised clonic seizures (clonus of the whole body with a loss of righting reflex, GCCs) and generalised clonic-tonic convulsions with tonic hind limb extension (GCTCs). The appearance (incidence) and the time to onset of every convulsive pattern were registered. If the rats did not exhibit convulsive responses, a latent time of 240 sec was assigned.

Biochemical parameters

All the rats were sacrificed by decapitation 4 min after PTZ administration. Their heads were immediately frozen in liquid nitrogen and stored at -70ºC.

For biochemical analysis the forebrain cortex, striatum and hippocampus from the contralateral hemisphere relative to cannula insertion were dissected on ice, homogenized in ice-cold buffer, centrifuged and sonicated. Biochemical parameters were determined spectrophotometrically in the crude mitochondrial fraction according to the method of Gurd et al. [22].

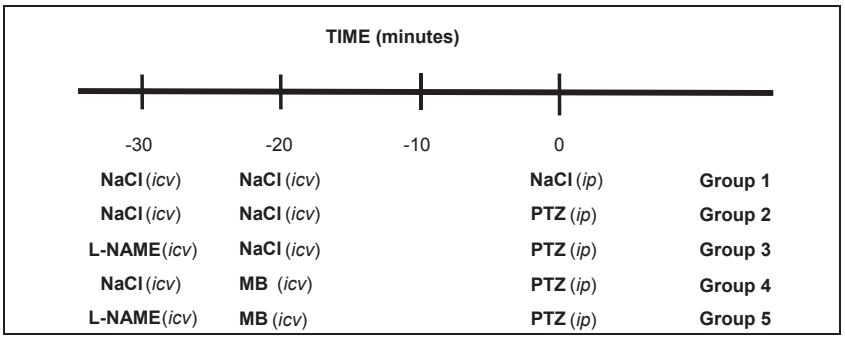

Fig. 1 – Protocol of treatments (n = 7–8 Wistar rats in each group)
NaCl – 0.9% saline; PTZ – pentylenetetrazole; L-NAME – NG-nitro-L-arginine methyl ester; MB – methylene blue; *ip* – intraperitoneally; *icv* – intracerebroventricularly

The assay for the superoxide radical is based on the reduction of nitrobluetetrazolium (Merck) in an alkaline, nitrogen-saturated buffer with absorbance monitoring at 515 nm [23].

Total superoxide dismutase (SOD) activity was measured as the inhibition of epinephrine auto-oxidation (Sigma Chemical Co.) in sodium carbonate buffer (Serva) containing 0.1 mM EDTA (Sigma Chemical Co.) at 480 nm [24].

Lipid peroxidation was measured as a function of malondialdehyde (MDA) production at 533 nm [25].

The incidence of convulsive patterns was expressed as the percentage of convulsing rats of the total number of rats in a group and was analysed using Kolmogorov-Smirnov test. The latent time of convulsions was calculated in seconds and expressed as the mean ± standard deviation (SD) and was analysed using the Kruskal-Wallis test. Biochemical parameters were expressed as the mean ± SD and were analysed using the ANOVA, followed by Tuckey test. Differences between experimental groups were considered significant when $p < 0.05$.

Results

Behavioural effects

In the control group of rats (sterile isotonic saline treatment, *icv* and *ip*) no convulsions were observed. In contrast, convulsive responses were observed in all PTZ-treated rats (Figure 2).

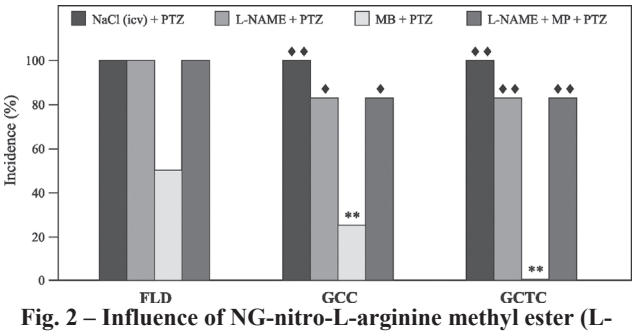

Fig. 2 – Influence of NG-nitro-L-arginine methyl ester (L-NAME, 10 μg) and methylene blue (MB, 10 μg) on pentylenetetrazole (PTZ, 80 mg/kg) – evoked convulsions in Wistar rats (n = 7–8)

***p* < 0.05, *p* < 0.01 is the level of significance when compared with the corresponding PTZ (*, **) and MB + PTZ-treated rats (♦, ♦♦; Kolmogorov-Smirnov test). PTZ was applied intraperitoneally, 0.9% NaCl, L-NAME and MB intracerebroventricularly; FLD – forelimb dystonia; GCC – generalised clonic convulsions; GCTC – generalised clonic-tonic convulsions**

Pretreatment with L-NAME led to some anticonvulsant, however insignificant effects on PTZ-induced convulsions.

MB pretreatment was very effective against PTZ-evoked convulsions. GCTCs were completely prevented ($p < 0.01$, compared to the PTZ-treated group). GCCs appeared only in 25% of animals ($p < 0.01$). FLD was apparent in 50% of the rats ($p = 0.0513$).

When L-NAME was administered before MB, the anticonvulsant effects of MB were lost, in particular GCTCs and GCCs; their incidence increased from 0 to 83%; ($p < 0.01$), and from 25 to 83% ($p < 0.05$), respectively. In addition the incidence of FLD was doubled, from 50 to 100% ($p = 0.0513$).

The latent time of PTZ-evoked convulsions was not influenced by L-NAME pretreatment (Table 1). MB very strongly influenced the latent time of all three convulsive patterns ($p < 0.01$ for FLD and GCTC, $p < 0.05$ for GCC, compared with the PTZ-treated group and groups pretreated with L-NAME). But MB's ability to extend the latent time of PTZ-induced convolutions was completely blocked by L-NAME.

Biochemical effects

In the PTZ-treated group of rats decreased lipid peroxidation in the forebrain cortex and striatum compared with the control group was found ($p < 0.05$ and $p < 0.01$, respectively) (Figure 3). In addition, increased SOD activity in the hippocampus compared with the control group was noted ($p < 0.05$) (Figure 4).

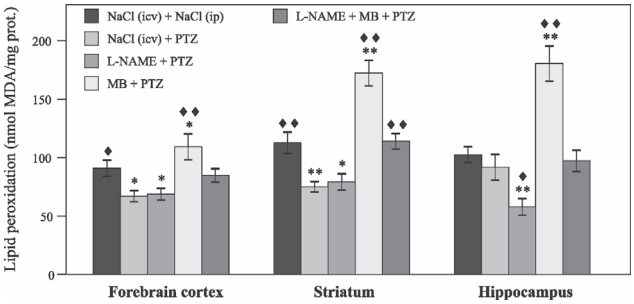

Fig. 3 – Influence of NG-nitro-L-arginine methyl ester (L-NAME 10 μg) and methylene blue (MB, 10 μg) on lipid peroxidation in the brain of Wistar rats (n = 7–8) treated with pentylenetetrazole (PTZ, 80 mg/kg)

***p* < 0.05, 0.01 is the level of significance when compared with the corresponding values (mean ± SD) of 0.9% NaCl (*, **) and PTZ-treated rats (♦, ♦♦; ANOVA test). PTZ was applied intraperitoneally, NaCl intraperitoneally and intracerebroventricularly, L-NAME and MB intracerebroventricularly**

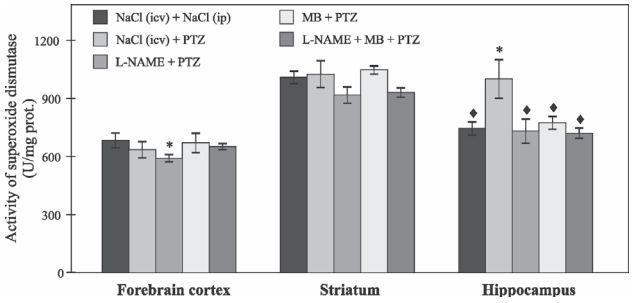

Fig. 4 – Influence of NG-nitro-L-arginine methyl ester (L-NAME 10 μg) and methylene blue (MB, 10 μg) on the superoxide dismutase (SOD) activity in the brain of Wistar rats (n = 7–8) treated with pentylenetetrazole (PTZ (80 mg/kg)

***p* < 0.05, 0.01 is the level of significance when compared with the corresponding values (mean ± SD) of 0.9% NaCl (*, **) and PTZ-treated rats (♦, ♦♦; ANOVA). PTZ was applied intraperitoneally, NaCl intraperitoneally and intracerebroventricularly, L-NAME and MB intracerebroventricularly**

Table 1

Time to onset of pentylenetetrazole (PTZ, 80 mg/kg) – evoked convulsions in Wistar rats (n = 7–8)

Treatment	Latent time (seconds)		
L-NAME and MB (μg)	FLD	GCC	GCTC
NaCl + PTZ	49.5 ± 11.3♦♦	118.6 ± 25.8♦	131.7 ± 35.6♦♦
L-NAME (10) + PTZ	57.7 ± 11.9♦♦	125 ± 68.3♦	128 ± 67♦♦
MB (10) + PTZ	159.2 ± 88.7**	213.3 ± 65.3*	240 ± 0**
L-NAME (10) + MB (10) + PTZ	53.3 ± 8.8♦♦	123.5 ± 70.4♦	126.7 ± 70♦♦

***p* < 0.05, 0.01 is the level of significance when compared with the corresponding PTZ (*, **) and MB+PTZ treatments (♦, ♦♦; Kruskal-Wallis test). Values are expressed as means ± SD. In the case without convulsive response, the latent time was assigned as 240 seconds. PTZ was applied intraperitoneally, 0.9% NaCl, L-NAME and MB intracerebroventricularly**

FLD – forelimb dystonia; GCC – generalised clonic convulsions; GCTC – generalised clonic-tonic convulsions; L-NAME – NG-nitro-Larginine methyl ester; MB – methylen blue

Treatment with L-NAME prior to PTZ administration decreased lipid peroxidation in the hippocampus ($p < 0.01$, compared with the control group; $p < 0.05$, compared with the PTZ-treated group) (Figure 3). SOD activity was decreased in the forebrain cortex ($p < 0.05$, compared with the control group) (Figure 4). Superoxide anion content of the hippocampus was also decreased ($p < 0.05$, compared with the control group) (Figure 5).

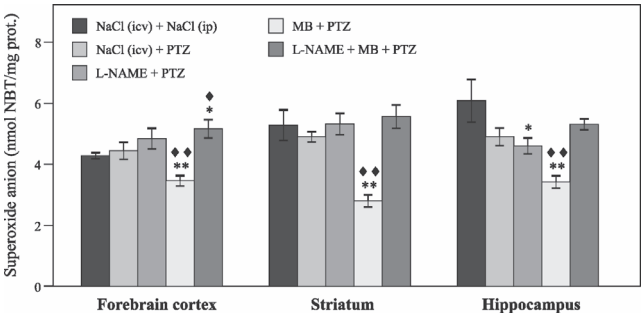

Fig. 5 – Influence of NG-nitro-L-arginine methyl ester (L-NAME 10 μg) and methylene blue (MB, 10 μg) on the superoxide anion content of the brain of Wistar rats (n = 7–8) treated with pentylenetetrazole (PTZ, 80 mg/kg).
p < 0.05, 0.01 is the level of significance when compared with the corresponding values (mean ± SD) of 0.9% NaCl (*, **)) and PTZ-treated rats (♦, ♦♦; ANOVA). PTZ was applied intraperitoneally, NaCl intraperitoneally and intracerebroventricularly, L-NAME and MB intracerebroventricularly

MB administered before PTZ resulted in increased lipid peroxidation compared with the control group ($p < 0.05$ in the forebrain cortex, $p < 0.01$ both in the striatum and hippocampus) and compared with the PTZ-treated group of rats ($p < 0.01$ for all brain regions) (Figure 3).

The level of superoxide anion was decreased in the MB+PTZ treated group of rats compared with both the control and PTZ-treated groups ($p < 0.01$ in all brain regions) (Figure 5).

Pretreatment with L-NAME reverted the MB+PTZ-induced increase in lipid peroxidation (Figure 3) and decrease in superoxide anion (Figure 5) in all brain regions back to the control values, except in the forebrain cortex where the superoxide content was increased, while SOD activity was decreased ($p < 0.05$ in comparison with the control and PTZ-treated groups of rats, and the control group, respectively).

Discussion

We found that both L-NAME and MB affected PTZ-evoked convulsions (insignificant and almost completely/completely prevented, respectively). However, co-administration of L-NAME and MB did not result in an additive/synergistic effects. Instead the effects (clinical and biochemical) of MB were severely abrogated by L-NAME.

The convulsant effects of PTZ are quite complex and are not yet completely understood. Reduced γ-aminobutyric (GABA) activity (GABA suppresses NOS), and enhanced excitatory amino acids release/transmission is believed to be the underlying mechanism [26, 27]. Thus, PTZ's effects on both the GABA-ergic and glutamatergic system lead to overproduction of NO and potentiation of sGC activity.

NO preferentially reacts with other radicals despite being a free radical itself. Its reactivity with biological molecules is low but it readily reacts with the hydroxyl radical [28]. In this way it can act as an antioxidant by scavenging free radicals thereby inhibiting lipid peroxidation. Chiueh [29] proposed at least four mechanisms by which NO acts as an antioxidant, including inhibition of lipid peroxidation. Thus, within the first few minutes of PTZ administration when cGMP production is rapidly increased [9], which is an indirect measure of NO production, a defence reaction against PTZ's toxic effects (via the antioxidative activity of NO) may take place. Under very strong production of NO and at later times this antioxidative defence is overcome and lipid peroxidation ensues, as described by a number of studies. For example, Patsoukis et al. [30, 31] found that the intensity of lipid peroxidation was increased in the mouse hippocampus and the striatum 15 min after PTZ was applied ip at a dose of 60 mg/kg. Furthermore, increased lipid peroxidation was within control values 30 min and 24 h post PTZ administration. However, lipid peroxidation was not increased in the mouse cerebral cortex in each monitoring time [32]. A lower dose of PTZ (40 mg/kg) did not in any way influence lipid peroxidation, meaning that lipid peroxidatrion is PTZ dose- and time- dependent.

In the research of Bashkatova et al. [33] PTZ (120 mg/kg administered subcutaneously) caused convulsions in all animals within 2–3 minutes. Moreover, one hour later the level of NO was increased 5-fold and lipid peroxidation 2-fold in the frontal cortex. Despite the fact that PTZ was used as a chemoconvulsant in both our and the above-mentioned studies, to some degree different results concerning lipid peroxidation are most likely explainable due to different experimental procedures.

The anticonvulsant effects of MB described in our studies are in accordance with those of Furian et al. [17]. In addition to MB's ability to increase the latency of methylmalonate-induced convulsions in adult male Wistar rats, abrogation of the methylmalonate-induced striatal NO level elevation was also observed. Furthermore, in the case of catalepsy induction Echeverry et al. [34] demonstrated similar effects of L-NAME and MB, both applied *icv*. Also, the level of NO products was decreased in the striatum.

However, discrepancies between our results and the results obtained by Deutsch et al. [35], who did not record the anticonvulsant effects of MB in electrically precipitated tonic hind limb extension in mice, could be explained by the use of different experimental models of convulsions, routes of drug administration, time points and dose of MB application, since pharmacokinetics and organ distribution of MB depends on the way of its application [36].

Apart from inhibition of sGC, MB has several other nonspecific effects on NO including inhibition of NOS. In cultured endothelial cells Shimizu et al. [37] found that MB, which is known to inactivate iron-containing enzymes, reacts with the heme moiety of NOS. Vallo et al. [16] found similar effects on hippocampal brain NOS activity *in vivo*. Further-

more, MB-mediated inhibition of hippocampal NOS was dose-dependent and the degree of inhibition was similar to that obtained with an unselective NOS inhibitor N-G-nitro-L-arginine (L-NNA). In comparison with sGC inhibition, MB-mediated NOS inhibition appeared more potent [12]. The susceptibility of different NOS isoforms to MB remains unclear. Because of NOS inhibition by MB, in our research it was expected that L-NAME, a competitive non-selective NOS inhibitor, enhances MB's anticonvulsant effects. However, the opposite was apparent: MB's anti-convulsant effects were reverted back to control values. In other words, combined pretreatment with L-NAME and MB exerted deleterious effects on MB-preventable PTZ-evoked convulsions. It is difficult to propose the reason for the obtained events. In addition to nitrergic system a number of other effects of both substances have to be taken into consideration before any conclusion, such as their influence on cholinergic, dopaminergic, serotoninergic and other transmissions that are important in the pathogenesis of convulsions [38].

Despite much more evidence pointing towards its antioxidant effects, MB can also be prooxidative causing singlet oxygen and superoxide formation, weak peroxide generation and direct glutathione oxidation [39]. Inactivation of NO by superoxide is ranked as one of the most rapid nonenzymatic reactions in biology. In other words one of MB effect could be related to the removal of NO already present. The chemical reaction between NO and superoxide, which could be generated in the presence of MB, may lead to the formation of peroxynitrite anion. The latter is a source of the hydroxyl radical which can intensify lipid peroxidation [40]. In our previous study it was found increased NO leveles in the hippocampus, forebrain cortex, striatum and some other brains structures within 30 minutes of PTZ application in the PTZ-evoked convulsions in rats [41]. Therefore, abovementioned effects of PTZ are in agreement with our observation regarding increased lipid peroxidation and a decreased level of superoxide anion in all the examined brain structures isolated from rats pre-treated with MB. Reestablishing level of lipid peroxidation in all brain regions and superoxide anion (except in the forebrain cortex) without increased SOD activity when L-NAME was coadministered with MB coincide with the suggested explanation of increased lipid peroxidation upon MB treatment *ie* it could indicate decreased interaction between NO and superoxide anion. However, increased lipid peroxidation that was associated with anticonvulsant effects of MB is undesired for any antiepileptic drug and substance and needs scientific evidence reliable of benefit/risk ratio with the aim to study whether benefits of such treatments (anticonvulsant effects) overwhelm risks of their potential harmful effects (increased lipid peroxidation).

A cascade of prooxidative events associated with MB pretreatment was not at all disrupt by coadministration of L-NAME. That was demonstrated in increased level of superoxide anion in the forebrain cortex. It could be the result of L-NAME effects on SOD activity, which was decreased in this structure when L-NAME was individually applied before PTZ. Thus, L-NAME did not only prevent anticonvulsant and lipid peroxidation effects of MB, but also it made conditions for forebrain cortex oxidative damage. In other words, all three structures in which most of the complex events of epileptic seizures are generated were very sensitive and prone to PTZ, MB and L-NAME influence.

In our PTZ-mediated convulsing rat model it was registered very early and rapid events (within the first 4 minutes after PTZ application): MB, when applied individually, prevented convulsive responses, especially GCTCs and GCCs; modulation of the NO system by MB in the settings of PTZ-induced convulsions may elicit anticonvulsant effects, but only if MB was applied on its own, not in combination with L-NAME, indicating that NO plays important role into anticonvulsant effects of MB under applied conditions; convulsive responses to PTZ was associated with a decreased and/or unchanged level of lipid peroxidation in the forebrain cortex, striatum and hippocampus. In contrast, anticonvulsant effects of MB coincide with increased lipid peroxidation. These clinical and biochemical effects of MB were abandon by L-NAME treatment.

Conclusion

MB's strong anticonvulsant effects in PTZ-evoked convulsions, especially against generalised clonic and clonic-tonic convulsions, and changes of the examined biochemical parameters in brain structures, as well as prevention of all these effects by L-NAME applied before MB could be partly the result of the nitrergic system modulation. Our results have far reaching therapeutic implications, *ie* translation into clinical practice benefits, particularly the anticonvulsant effects of MB. Future investigations will hopefully shed some light on the intricacies of MB's effects.

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Received on July 13, 2010.
Revised on November 5, 2011.
Accepted on November 11, 2011.

ORIGINAL ARTICLE

UDC: 616.831-009.1:616.313-008-07
DOI: 10.2298/VSP1206488Z

Tongue mobility in patients with cerebral palsy

Pokretljivost jezika kod bolesnika sa cerebralnom paralizom

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Abstract

Background/Aim. In children with cerebral palsy speech is a big problem. Speech of these children is more or less understandable, depending on the degree of reduced mobility of articulatory organs. Reduced mobility is affected by inability to control facial grimacing and poor muscle strength when performing targeted movements. The aim of this study was to determine the mobility of tongue in patients with cerebral palsy. **Methods**. The study included a sample of 34 children – patients with cerebral palsy who had been treated in the Special Hospital for the Cerebral Palsy and Developmental Neurology in Belgrade. The patients were divided according to the determined diagnosis into two groups: *Quadriparesis spastica* (n = 11) and *Morbus* Little (n = 16). The children, aged 8–12 years, had preserved intelectual abilities, and all of them had preserved hearing. The study was conducted during the period from January to September 2009. The functional state of articulatory organs in both groups was tested by the C-test that examines the anatomic structure and mobility of the articulatory organs. **Results**. Our research showed that both groups of the patients had impaired functional state of the tongue – the most mobile articulatory organ. Also, the research showed that the functional state of the tongue was worse in children diagnosed with *Quadriparesis spastica.* A statistically significant correlation between the diagnosis and the functional state of the tongue, the tongue test performance and the retention of the tongue in a given position was found ($r = 0.594$, $p < 0.005$; $r = 0.816$, $p < 0.01$ and $r = 0.738$, $p < 0.001$, respectively). **Conclusion**. A large percentage of children with cerebral palsy were not able to establish control over the position of articulatory organs, especially the tongue, and its retention in a given position, all of which affect the quality of speech.

Key words:
speech disorders; cerebral palsy; tongue; speech articulation tests; child.

Apstrakt

Uvod/Cilj. Govor predstavlja veliki problem kod dece sa cerebralnom paralizom. Govor te dece je manje ili više razumljiv u zavisnosti od stepena redukovane pokretljivosti artikulacijskih organa. Na smanjenu pokretljivost jezika utiču i nemogućnost kontrole facijalnih grimasa i slaba mišićna snaga prilikom izvođenja ciljanih pokreta. Cilj ovog istraživanja bio je da se utvrdi pokretljivost jezika kod bolesnika sa cerebralnom paralizom. **Metode**. Ispitivanjem je bilo obuhvaćeno 34 dece sa cerebralnom paralizom koja su bila na lečenju u Specijalnoj bolnici za cerebralnu paralizu i razvojnu neurologiju u Beogradu. Deca, uzrasta 8–12 godina, imala su očuvane intelektualne sposobnosti i sluh. Deca su bila podeljena na dve grupe prema dijagnozi: *Quadriparesis spastica* bila je zastupljena kod 18 dece, a *Morbus* Little kod 16 dece. Ispitivanje je obavljeno u periodu od januara do septembra 2009. godine. Funkcionalno stanje artikulacijskih organa kod obe grupe bolesnika ispitano je C-testom, koji utvrđuje pokretljivost jezika**. Rezultati.** Naše istraživanje pokazalo je narušeno funkcionalno stanje jezika kao najpokretljivijeg artikulatora kod obe grupe bolesnika. Znatno više bila su ugrožena deca sa dijagnozom *Quadriparesis spastica.* Nađena je značajna korelacija između funkcijskog stanja jezika i dijagnoze ($r = 0{,}594$; $p < 0{,}005$), kao i između izvođenja proba jezikom i dijagnoze ($r = 0{,}816$; $p < 0{,}001$). Takođe, nađena je značajna korelacija između dijagnoze i zadržavanja jezika u zadatom položaju ($r = 0{,}738$; $p < 0{,}001$). **Zaključak**. Utvrđeno je da visok procenat dece sa cerebralnom paralizom nije u mogućnosti da uspostavi kontrolu položaja artikulacijskih organa, pre svega jezika, kao i njegovo zadržavanje u zadatom položaju, što sve skupa utiče na kvalitet govora.

Ključne reči:
govor, poremećaji; paraliza, cerebralna; jezik; govor, testovi artikulacije; deca.

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Introduction

"Cerebral palsy describes a group of permanent disorders of the development of movement and posture, causing activity limitation, that are attributed to non-progressive disturbances that occurred in the developing fetal or infant brain. The motor disorders of cerebral palsy are often accompanied by disturbances of sensation, perception, cognition, communication, and behaviour, by epilepsy, and by secondary musculoskeletal problems" [1].

Optimistic expectations that the constant progress of science and neonatal diagnosis would decrease the incidence of cerebral palsy, have not come true. The incidence of cerebral palsy is 1.5–2.5 per 1 000 live births [2]. The cure has not yet been found, so we need to observe current state of each patient individually in a way to help him/her in overcoming the difficulties caused by this desease [3]. Bax [4] suggests that in these patients skeletal muscles, as well as chewing, swallowing and speech muscles are affected by basic pathological processes that cause the difficulties in articulation. In children with cerebral palsy, there is insufficient mobility of the muscles of individual speech organs involved in the process of articulation. It is generally known that there is no good quality speech without good functional state of the articulation apparatus. The tongue is an organ that is most important articulator. It is located in the oral cavity and plays an important role in chewing, swallowing, sucking and speech. The tongue represents the muscle structure, fan-like spread, and is one of the most mobile organs. Anatomically, the tongue is divided in two parts, the front or the horizontal and the back or the vertical part (tongue basis). The front of the tongue is attached to the center line of the mouth (*frenulum linguae*), and sometimes can be so short that prevents normal movement of tip of the tongue, making articulation more difficult [5]. The tongue has the motor, sensitive, gustatory and tactile innervation. It receives motor innervation over the twelfth cerebral nerve (*nervus hypoglossus*). Field of innervation is strictly divided, right *hypoglossus* innervates the right side, and left *hypoglosssus* the left side of tongue. This is clinically important because of the unilateral nerve lesions that lead to muscle atrophy, and thus a mobility reduction of the appropriate side of the tongue [6]. For the voice articulation both motor and sensitive innervation are very significant. The tongue is a three-dimensional muscle which can move to the three main directions owing to the action of the external muscles. The external muscles enable motions upward – forward, upward – backward and downward – backward while the internal muscles enable shapes change of the tongue at any position [7]. By the contraction of these muscles the tongue can be made shorter, narrower, can bend at any directions, the gutters can be made and alike.

The aim of this study was to determine the condition of articulatory organs of certain groups of patients with cerebral palsy, ie. the mobility of tongue as one of the most important articulators.

Methods

The study included a sample of 34 children with cerebral palsy who had been treated in the Special Hospital for Cerebral Palsy and Developmental Neurology in Belgrade. The children, aged of 8–12 years, had preserved intellectual abilities, and hearing. The children were divided into two groups according to the diagnosis: *Quadriparesis spastica* was diagnosed in 18 patients, and *Morbus* Little in 16. This age was chosen because in eight-year-old children the automation of the articulation basis was finished. The study was conducted in a period from January to September 2009. The functional state of articulation organs in both groups was tested by the C-test that examines anatomic structure and mobility of the articulation organs [8]. The software package SPSS-16 was used for making a database. For processing the obtained data appropriate statistical methods were used.

Results

Examination of functional state of the tongue showed that in the group diagnosed with *Morbus* Little 50% of the children had normal tongue condition, 37.5% of them had hypertonic tongue, 6.25% spastic and 6.25% hypotonic (Table 1). In the children with *Quadriparesis spastica*, 33.33% of them had hypertonic tongue, 38.89% spastic, 27.78% atrofic, while normal and hypotonic state of tongue were not found.

Table 1
The functional state of the tongue in both groups of children

Functional state of the tongue	Children diagnosed with			
	Morbus Little		*Quadriparesis spastica*	
	n	%	n	%
Normal	8	50.00	0	0.00
Hypertonic	6	37.50	6	33.33
Spastic	1	6.25	7	38.89
Hypotonic	1	6.25	0	0.00
Atrofic	0	0.00	5	27.78
Total	16	100.00	18	100.00

r = 0.594 (Pearson's correlation coefficient); $p < 0.005$

Some tongue tests were performed by using C-tests. Accuracy and time required for performing certain movements were measured in both groups of the patients. In *Morbus* Little group testing was normally performed by 43.75% of the patients, 43.75% of patients have delayed test performance and 12.5% incorrect (Table 2). In the children with *Quadriparesis spastica* 38.89% of them incorrectly performed the test, with delay 5.56%, and 55.56% of them were not able at all to adequately perform fine and precise articulation movements.

We tested retention of the tongue in a given position, examining that way its muscle strength, and we did not come to optimistic results. In the group diagnosed with *Morbus* Little only 31.25% of the patients normally retained the tongue in a given position, 56.25% did it with difficulty, while 12.5% were not able to retain the tongue at all (Table 3). In the group

Table 2

The tongue test performance in both groups of children

Tongue test performance	Children diagnosed with			
	Morbus Little		*Quadriparesis spastica*	
	n	%	n	%
Normally	7	43.75	0	0.00
Slowly	7	43.75	1	5.56
Incorrectly	2	12.50	7	38.89
Unable to perform	0	0.00	10	55.56
Total	16	100.00	18	100.00

r = 0.816 (Pearson's correlation coefficient); $p < 0.001$

Table 3

The abilitly to retain the tongue in both groups of children

Tongue retention in determined position	Children diagnosed with			
	Morbus Little		*Quadriparesis spastica*	
	n	%	n	%
Normally	5	31.25	0	0.00
With difficulty	9	56.25	2	11.11
Unable to perform	2	12.50	16	88.89
Total	16	100.00	18	100.00

r = 0.738 (Pearson's correlation coefficient); $p < 0.001$

of patients diagnosed with *Quadriparesis spastica* the results were much worse, even 88.89% of patients were not able to retain the tongue in a given position, and only 11.11% of them performed retention, but with great effort.

Discussion

Scientific community has focused attention on dysarthria as a global problem of this population and the consequences deriving from it, especially unintelligible speech. Strauss et al. [9] have find that estimations based on a simple, easily measurable functions, such as lead pose, ability for taking food, quality of articulation and speech intelligibility, can lead to valuable information about this desease forecasting. Our research showed functional state of the tongue as the most mobile articulator is impaired in both groups of the patients. But generally, functional state of tongue was significantly worse in patient diagnosed with *Quadriparesis spastica*. The correlation between functional state of the tongue and the diagnosis was r = 0.594, $p < 0.005$.

When performing the tongue test, 43.75% of the patients diagnosed with *Morbus* Little managed to accomplish the test. In the group with *Quadriparesis spastica* no patient was able to fully carry out the task. The limitation of tongue mobility may be associated with spasm of tongue and chin muscles [10]. Active movements were more limited in children with *Quadriparesis spastica*, because in these patients both the lower and the upper limbs muscles were affected by spasms including orofacial muscles, while in patients with *Morbus* Little motor deficit was mostly associated with the lower limbs. The correlation between tongue test performance and the diagnosis was significant (r = 0.816, $p < 0.001$).

A motor deficit of orofacial muscles is often accompanied by poor muscle strength of the tongue. Retaining the tongue in a certain position somewhat showed better results in the patients diagnosed with *Morbus* Little, indicating that this articulatory organ in children with cerebral palsy in adition to impaired inervation is accompanied by extremely weak muscle strength, all of which results in poor sounds, articulation that requires active participation of the tongue along with other structures. There was a significant correlation found between the diagnosis and retention of tongue in a given position (r = 0.738, $p < 0.001$).

This study is strictly related to motor deficits of the tongue as articulation organ in children with cerebral palsy and preserved intelligence and it is the first research of this type conducted in Serbia. Although the sample is small, we hope that the research will contribute to better understanding of articulation problems of this population and help in taking appropriate and timely habilitation measures for their reducing. Platt et al. [11] conducted a similar research, but it included adults of reduced intelligence quotient with cerebral palsy.

Conclusion

After analyzing the results obtained by this study it could be concluded that both groups of the patients had impaired functional condition of the tongue, decreased ability to perform certain movements and weaker tongue muscles strength. The worse results were obtained in the children diagnosed with *Quadriparesis spastica*. Therefore, they had less control of articulation apparatus, increased salivation and less intelligible speech. The facts indicate that immediately after making the diagnosis, it is necessary to include these children in an early habilitation treatment, well before the automation of articulation, so that the mechanisms of neuroplasticity could help them build basic levels of speech functions.

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Received on October 4, 2010.
Revised on July 13, 2011.
Acepted on July 28, 2011.

ORIGINAL ARTICLE

UDC: 616.831-073-71:616.8-053.31-036
DOI: 10.2298/VSP1206492V

The prognostic value of amplitude-integrated electroencephalography in neonates with hypoxic-ischemic encephalopathy

Prognostička vrednost elektroencefalografije integrisanih amplituda kod novorođenčadi sa hipoksičko-ishemijskom encefalopatijom

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Abstract

Background/Aim. Diagnosis of perinatal hypoxic-ischemic encephalopathy (HIE) and early prediction neurological outcome is important and difficult. The aim of this study was to determine the prognostic value of amplitude-integrated electroencephalography (aEEG) for abnormal neurodevelopment outcome in a neonate with HIE. **Methods.** A total of 90 neonates > 32 gestational age (GA) with HIE were enrolled prospectively. All neonates with HIE were categorized into three grades according to the Sarnat and Sarnat clinical scoring system (mild HIE, moderate HIE and severe HIE). aEEG traces were recorded with a cerebral function monitor (CFM) during the first 72 h of life. The neurodevelopment outcome was assessed at 12 months of age of corrected gestational age. **Results.** The pattern of aEEG correlated with the severity of HIE ($p < 0.0001$) and subsequent neurodevelopment outcome ($p < 0.001$). We found that aEEG background patterns exhibited superior prediction of abnormal outcomes at 12 months of age (sensitivity of 91.7% and specificity of 94.3%, positive predictive value of 78.6% and negative predictive value of 98.1%) when compared to aEEG seizure (sensitivity of 94% and specificity of 48%, positive predictive value of 57% and a negative predictive value of 92%). Electroclinical dissociation seizure was detected in 28% of the neonates with HIE. **Conclusions.** Our results confirm that aEEG is simple and accurate bedside diagnostic method for assessing extension of hypoxic-ischemic brain damage and early identification of neonates with perinatal HIE who are at high risk of neurodevelopmental impairment.

Key words:
elektroencephalography; intensive care units, neonatal; hypoxia-ishemia, brain; infant, premature; infant, newborn; prognosis.

Apstrakt

Uvod/Cilj. Dijagnoza perinatalne hipoksično-ishemičke encefalopatije (HIE) i prognoza kasnijeg neurološkog ishoda je važna i, istovremeno, vema teška. Cilj ove studije bio je da se utvrdi prognostički značaj elektroencefalograma integrisanih amplituda (aEEG) za utvrđivanje neurološke prognoze kod novorođenčeta sa HIE. **Metode.** Ovom prospektivnom studijom bilo je obuhvaćeno 90 novorođenčadi > 32 gestacijske nedelje (GN) sa HIE. Sva ispitivana novorođenčad bila su neurološki procenjivana prema Sarnat i Sarnat kliničkom skoru i podeljena u 3 grupe (blaga HIE, srednje teška HIE i teška HIE). Snimanje aEEG vršeno je cerebralnim funkcionim monitorom (CFM) tokom prva 72 sata života. Neurološka procena ispitivane dece sprovedena je u uzrastu od 12 meseci i korigovana je prema gestacijskoj zrelosti na rođenju. **Rezultati.** Registrovana aEEG aktivnost bila je u korelaciji sa težinom HIE ($p < 0,0001$) i kasnijim razvojem neuroloških sekvela ($p < 0,001$). Naši rezultati ukazuju da registrovana osnovna aEEG aktivnost ima višu prediktivnu vrednost za loš neurološki ishod u izrastu od 12 meseci (senzitivnost 91,7%, specifičnost 84,3%, pozitivna prediktivna vrednost 78,6% i negativna prediktivna vrednost 98,1%) od registrovanih aEEG konvulzija (senzitivnost 94%, specifičnost 48%, pozitivna prediktivna vrednost 57% i negativna prediktivna vrednost 92%). Elektroklinička disocijacija konvulzija je registrovana kod 28% novorođenčadi sa HIE. **Zaključak.** Naši rezultati ukazuju da je aEEG jednostavna i precizna dijagnostička metoda za procenu ekstenzivnosti hipoksično-ishemičkog moždanog oštećenja i ranu identifikaciju novorođenčadi sa HIE kod kojih je prisutan visok rizik od kasnijeg nastanka neuroloških sekvela.

Ključne reči:
elektroencefalografija; intenzivna nega, neonatalna; mozak, hipoksija-ishemija; nedonošče, novorođenče; prognoza.

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Introduction

Perinatal hypoxic-ischemic encephalopathy (HIE) is a common cause of neonatal morbidity and mortality and neurological disabilities among survivors [1]. Each year 1.2 million neonates die and about 1 million infants have permanent neurological disability caused by HIE [2]. Determination of severity of perinatal HIE by the clinical criteria and appropriate neuroimaging or neurophysiologic techniques remains the main prognostic tool.

Neonates with mild HIE have a uniformly good prognosis and those with severe HIE a very high risk for early death or severe disabilities. Thus, especially for the neonates with moderate HIE, neuroimaging and neurophysiological examinations have a great value. Unfortunately, the predictive values of neuroimaging techniques (i.e. magnetic resonance imaging, computerized tomography and ultrasound) are limited during the first days of life [3]. In contrast, neurophysiological examinations of functional integrity of the brain have been shown to be useful also during the first days of life. High predictive values have been reported for evoked potentials (somatosensory, visual and auditory), conventional encephalography (EEG) and amplitude-integrated encephalography (aEEG) [4]. Both serial registration of conventional EEG and measurement of evoked potentials in a neonatal intensive care unit (NICU) require considerable technical skill, time and expertise in interpretation and may not be rapidly available in most hospitals. An alternative technique is aEEG recorded with a cerebral function monitor (CFM), designed for long-term monitoring brain activity at bedside [5]. CFM is a simplified single- or two-channel electroencephalogram monitor [6]. A signal is obtained from a single pair of electrodes placed at the P3 and P4 position of the 10–20 International System, i.e. in the left and right parietal region. A guard or reference electrode positioned anterior to the vertex was also used to reduce the effects of electrical interference. The use of two channels (two pair of bilateral frontoparietal electrodes) has the advantage of defining laterality in unilateral lesions, not available in the single channel devices. The signal is amplified and passed through an asymmetrical band filter which attenuates activity below 2 Hz and above 15 Hz in order to minimize artefacts from muscle activity and electrical interference. Additional processing includes semi logarithmic amplitude compression, rectification and time compression. A signal is presented electronically on the device monitor and can be recorded on paper with a semi-logarithmic scale at slow speed (6 cm/hr). A second trace continuously records the electrode impedance. EEG waveform can also be displayed on the monitor. The bandwidth in aEEG traces reflects variations in upper and lower margins of activity or patterns of aEEG, both of which depend on the maturity and severity of the illness of a newborn infant [7].

In this study we prospectively evaluated the prognostic value of aEEG for assessing extension of hypoxic-ischemic brain damage and early identification of neonates with perinatal HIE who are at high risk of neurodevelopmental impairment.

Methods

Our study was performed from January 2007 to January 2009 and was approved by the Ethical Committee for Medical Research of the Medical Faculty at the University of Belgrade. Our institute serves as a referral center for high-risk pregnancies, with delivery numbers of 7,000–7,500 per year. We studied 90 neonates under 32 weeks gestational age (GA) with perinatal HIE admitted to neonatal intensive care units (NICU) at the Institute of Gynecology and Obstetrics, Clinical Center of Serbia, Belgrade. Written consent was obtained from all parents. Perinatal HIE was diagnosed if fetal distress (meconium staining of liquor or abnormal fetal heart rate), metabolic acidosis [pH < 7.20, base excess (BE) ≥ 10 mmol/L and lactate > 3mmol/L in arterial cord blood within 60 min of birth], immediate neonatal depression Apgar score (AS) ≤ 6 at 5 min and/or delayed spontaneous respiration, necessitating artificial ventilation at 5 min, and early neonatal encephalopathy (within the first 24 of life) were presented. All the neonates were resuscitated according to the guidelines of the Newborn Resuscitation Program of the American Academy of Pediatrics and American Heart Association [8, 9].

A complete obstetrical history and physical examinations were obtained on admission. Perinatal HIE was categorised into three stages according to the Sarnat and Sarnat clinical scoring system [10]. Head sonograms were performed on all the neonates before enrolment.

aEEG recordings started after initial stabilization in all 90 neonates with HIE. aEEG was recorded during the first 72 h of life using the Cerebral Function Monitor Olympic 6000 (Olympic Biomedical, USA) from biparietal adhesive electrodes and displayed on the integral printer at 6 cm/h. Handling of the neonates, observed clinical seizures, and administration of anticonvulsants or sedatives were recorded by the nursing staff. We excluded any aEEG records within 30 minutes of anticonvulsant administration. The CFM also recorded the impedance across the electrodes which was always below 10 kΩ.

Evaluation of aEEG recording should begin with the background pattern and then proceed to the presence or absence of seizures. aEEGs were described with either voltage criteria, using the upper and lower margins of activity or patterns of aEEG.

Different aEEG background patterns (Figure 1) in a postasphyctic period were classified as [11]: 1) normal aEEG background patterns (the upper margin of the trace is above 10 μV and the lower margin is greater than 5 μV. In healthy full term neonates the trace alters in width according to the awake and sleep state of the neonate); 2) moderately abnormal aEEG background patterns (the upper margin of the trace is greater than 10 μV and the lower margin is less than 5 μV). This pattern can be seen in neonates with moderately severe encephalopathy or immediately after administration of drugs such as anticonvulsants and sedatives. Excessive background discontinuity pattern ("dysmature pattern") may also be seen in very preterm neonates); 3) severely abnormal aEEG background patterns (the upper margin of the trace is

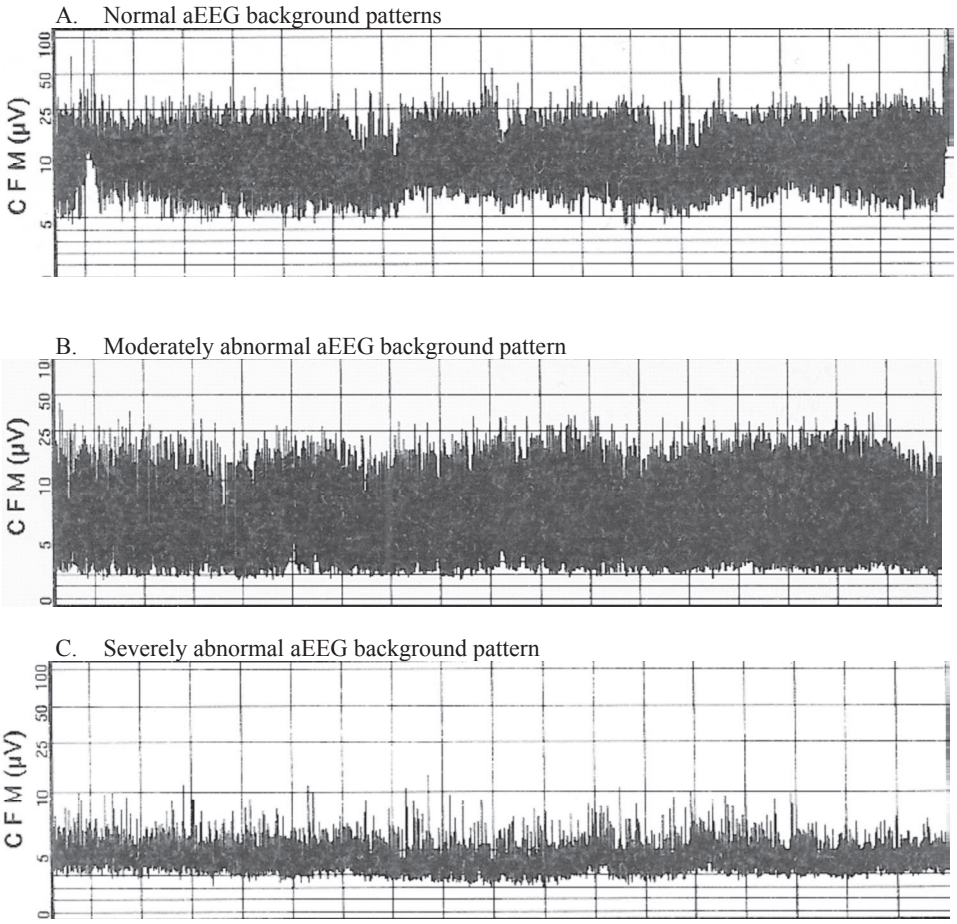

Fig. 1 – The different amplitude-integrated encephalography (aEEG) background patterns

less than 10 μV and the lower margin is usually less than 5 μV. A severely abnormal trace is characterized by a general suppression of amplitude and this pattern may be accompanied by brief bursts of higher voltage spikes which appear as single spikes above the background activity – "burst suppression". A severely abnormal trace is usually seen with severe encephalopathy and is often accompanied by seizure activity).

In addition, any of these three groups could be accompanied by seizures. Seizures were manifested as periods of sudden increase in voltage accompanied by narrowing of the band of aEEG activity and followed by a brief period of suppression (Figure 2). Arousal during care procedures may be misinterpreted as seizures. It is therefore important that all the procedures are documented to facilitate correct interpretation of aEEG. Administration of anticonvulsants or sedatives or handling of the neonates was recorded by the nursing staff. Status epilepticus often looks like a 'saw tooth' pattern but a continuously raised background may also be seen. Correct interpretation is only possible when simultaneous raw EEG is also available.

Neurodevelopment outcome was assessed at 12 months of corrected gestational age by the neonatologist and pediatric neurologist using the Denver Developmental Screening Test (DDST). Neurodevelopment outcomes were classified as normal, mild motor abnormality (slight abnormality in muscular tone or mild delayed of motor development) and severe adverse outcome [cerebral palsy (CP), epilepsy or if died in follow-up period].

We excluded neonates with congenital malformations, chromosomal abnormalities, inherited metabolic disorders, congenital or acquired neonatal infections and maternal drug addiction.

Statistical Package for Social Science software 11.5.1 for Windows (SPSS Inc., Chicago, IL, U.S.A.) was used for

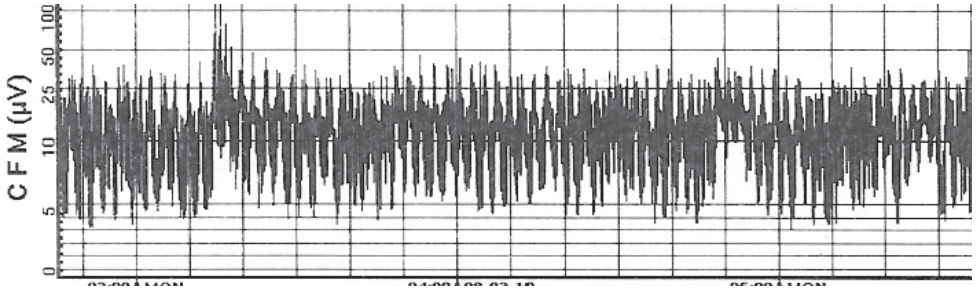

Fig. 2 – Seizures amplitude-integrated encephalography (aEEG) activity

data management and statistical analysis. Values of $p < 0.05$ were considered statistically significant. All values were expressed as mean value with standard deviation (SD) or as percentages (%) for descriptive purposes unless otherwise stated. Group comparisons were performed with the Fisher exact test or Kruskall-Wallis test and Wilcoxon rank sum test. The predictive value of aEEG for determining neurodevelopmental prognosis was assessed by calculation of sensitivity and specificity and positive and negative predictive values (PPV and NPV, respectively). Primary analysis was made by assessing the predictive value of abnormal aEEG background activity alone and/or seizure aEEG activity.

Results

We studied 90 neonates (> 32 GA) with perinatal HIE. The majority of neonates in our study developed mild HIE (Table 1). Birth weight (BW) and gestational age were similar, while gender distribution was different among the neonates with mild and advanced clinical stage of HIE ($p < 0.05$) (Table 1). At term, the incidence of HIE ranged from 3 to 4 per 1,000 live births during our study period. Predictive capacities AS at 1 and 5 min and arterial blood cord values of pH, BE and lactate for severity of HIE and abnormal outcome are shown in Table 1. Apgar scores at 1 min (W = 166; $p < 0.05$) and AS at 5 min (W = 181; $p < 0.01$) and pH values of arterial blood cord (W = 184; $p < 0.001$), BE (W = 167; $p < 0.05$) and lactate (W = 51; $p < 0.05$) correlated well with severity of HIE. Arterial cord blood pH had best predictive capacities for the severity of HIE and subsequent abnormal neurological outcome (W = 916; $p < 0.001$). We detected no significant difference among arterial blood cord pH values, BE and lactate or AS at 1min and 5 min in preterm and term neonates.

A relation between aEEG patterns and severity of HIE and neurologic outcome is summarized in Figure 3. There was a close relationship between the findings on aEEG patterns and severity of HIE ($p < 0.0001$) (Figure 3). The aEEG patterns in the first 72 h of life also had high predictive value for abnormal outcome ($p < 0.001$) (Figure 3). All neonates with normal aEEG patterns had good outcome while neonates with severely abnormal aEEG patterns (low voltage or inactive isoelectric background activity) either died or subsequently had an abnormal neurologic outcome. All the three HIE groups could be accompanied by seizures (Figure 3). Electrographic seizures without clinical correlates were detected on the CFM in 28% of the neonates with HIE (Figure 4). In the neonates with seizures, we found that the interictal aEEG activity correlated with the subsequent outcome. Severely abnormal aEEG patterns accompanied with seizures had a poor outcome. We detected no significant difference among aEEG patterns in preterm and term neonates.

Neurodevelopment outcome at 12 months of corrected gestational age corresponded well with severity of HIE ($p < 0.01$) and gestation age ($p < 0.05$) (Table 2). All the infants with severe HIE developed neurological sequels (cerebral palsy or epilepsy). Two preterm neonates with severe HIE died, one within the early neonatal period because of multiorgan failure, the other later because of respiratory dysfunction. In determining the prognostic value of aEEG, we included death and neurodevelopmental impairment as a single outcome group. Incidences of neurological sequels were significantly higher in preterm infants ($p < 0.05$). We found that aEEG background pat-

Table 1

Clinical and biochemical characteristics at birth in neonates with different stage of hypoxic-ischemic encephalopathy

Parameters	HIE	HIE I	HIE II	HIE III	NS
GA	36.6 ± 2.6	36.4 ± 2.5	37.4 ± 2.6	36.2 ± 3.1	35.8 ± 2.7
BW (g)	2711 ± 810	2699 ± 775	2889 ± 796	2311 ± 1000	2450 ± 93
M /F(%)	62/38	54/46	67/33*	100/0*	67/33*
AS (1min)	3.4 ± 1.4	4.1 ± 1.1	2.6 ± 1.3	1.3 ± 0.7*	2.5 ± 1.6*
AS (5min)	5.5 ± 1.7	5.6 ± 0.5	4.3 ± 1.2	2.7 ± 1.0*	3.8 ± 1.8**
pH	7.09 ± 0.11	7.15 ± 0.05	7.03 ± 0.11**	6.88 ± 0.09***	6.97 ± 0.14***
BE (mmol/L)	-13.1 ± 4.2	-11.0 ± 1.7	-15.5 ± 4.5*	-19.7 ± 3.9**	-16.5 ± 5.1*
Lactate(mmol/L)	7.4 ± 4.4	5.4 ± 2.9	9.6 ± 4.6*	13.9 ± 3. 9*	11.0 ± 5.1*
∑N	90	57	24	9	15

Values are expressed as mean± standard deviation (SD) or percentages (%). Kruskal-Wallis χ^2 test and Wilcoxon rank sum test with continuity correction. Significance *$p < 0.05$ or **$p < 0.01$ or ***$p < 0.001$. BW = birth weight: AGA = appropriate BW for gestation age: SGA = small BW for gestation age and LGA = large BW for gestation age; GA = gestational age; M = male and F = female; AS = Apgar score; BE = base excess; ∑N = numbers of neonates; HIE = hypoxic-ischemic encephalopathy (HIE stage I, HIE stage II and HIE stage III); NS = neurological sequels.

Table 2

Neurodevelopment outcome (NDO) in neonates with different stages of hypoxic-ischemic encephalopathy and different gestation age at birth

NDO	HIE	HIE I	HIE II	HIE III	PRETERM	TERM
Normal NDO (%)	73.9	91.3**	54.2**	0**	62.7	84.4*
M (%)	12.5	7	29.2**	0*	16.3 *	8.9
CP (%)	10.2	1.7**	12.5	71.4**	14*	6.7
EPI (%)	3.5	0	4.1	28.6**	13.3 *	0

Values are expressed as percentages (%); Fisher exact test significance; *$p < 0.05$ or ** $p < 0.01$; M = mild motor abnormality; CP = cerebral palsy and EPI = epilepsy; HIE = hypoxic-ischemic encephalopathy (HIE stage I, HIE stage II and HIE stage III);

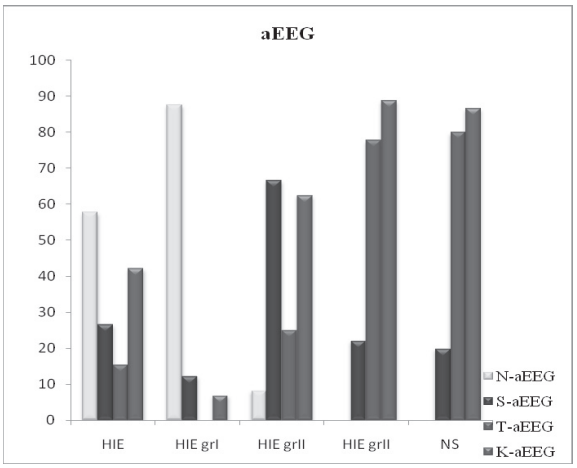

Fig. 3 – The different amplitude-integrated encephalography (aEEG) characteristics in neonates with different stages of hypoxic-ischemic encephalopathy and subsequent neurologic outcome

N-aEEG = normal aEEG background patterns; S-aEEG- = moderately abnormal aEEG background patterns; T-aEEG- severely abnormal aEEG background patternsM; K-aEEG = seizures aEEG activity; HIE = hypoxic-ischemic encephalopathy (HIE stage I, HIE stage II and HIE stage III); NS = neurological sequels.

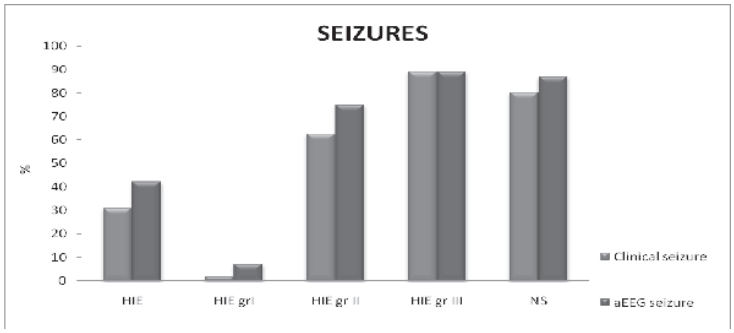

Fig. 4 – Electroclinical dissociation seizure in neonates with different stage of hypoxic-ischemic encephalopathy and subsequent neurologic outcome

HIE = hypoxic-ischemic encephalopathy (HIE stage I, HIE stage II and HIE stage III); NS = neurological sequels

terns exhibited superior prediction of abnormal outcomes at 12 months of age (sensitivity of 91.7% and specificity of 94.3%, positive predictive value of 78.6% and negative predictive value of 98.1%) when compared to aEEG seizure (sensitivity of 94% and specificity of 48%, positive predictive value of 57% and negative predictive value of 92%) (Table 3).

Table 3

The predictive value of amplitude-integrated electroencephalography (aEEG) background activity and seizures aEEG activity for determining neurodevelopmental prognosis in neonates with hypoxic-ischemic encephalopathy

aEEG activity	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Background	91.7	94.3	78.6	98.1
Seizures	94	48	57	92

PPV = positive predictive value; NPV = negative predictive value.

Discussion

Outcome prediction after perinatal asphyxia is important and difficult. Despite vast advances in neonatal intensive care the incidence of HIE continues to have a significant impact on the perinatal morbidity and mortality. The incidence of perinatal HIE varies at different gestational ages. At term, the incidence ranges from 3 to 5 per 1,000 live births and an incidence approaching 60% in premature newborns [12]. In our study the incidence of perinatal HIE was similar. The age-dependent vulnerability to hypoxic-ischemic insults seen in the immature brain can be explained by the high density of N-methyl-d-aspartate (NMDA) receptors and neuronal nitrie oxide synthase (nNOS)-positive cells [13, 14]. The immature brain is especially sensitive to oxidative damage relative to the mature brain are poor antioxidant capabilities and a high concentration of free iron and lipids.

In our study males had higher incidence of long term developmental disabilities than females. A recent analysis of a European database of 4,500 children with CP found that the incidence of CP was 30% higher in males than females.

Basic research of the causes of CP has revealed that gender may influence the pathogenesis of developmental brain injury. Sex differences in the immature brain appear to be strongly influenced by intrinsic differences between male and female cells and this is influenced by sex chromosomes and sex hormones [15, 16].

The Apgar scoring system, first devised in 1952, has been used to assess a newborn's condition and to reflect the need for resuscitation [17]. Over time, the Apgar score has also been used to define asphyxia, which is inappropriate, as many other conditions (e.g., congenital anomalies, prematurity, maternal drug administration) can result in low scores that are not reflective of asphyxia [18]. On the other hand, another study stated that low Apgar scores at 5 min are associated with death or cerebral palsy, and this association increased if both Apgar scores at 1 and 5 minute were low [19]. Our results also showed that the Apgar score at 5 minute remained a valid predictor of neonatal mortality, but using it alone to predict long-term outcome was inappropriate.

The incidence of long-term complications depends on the severity of hypoxic-ischemic encephalopathy which is in concordance with our results. In adults, neuronal necrosis and apoptosis after global ischemia are slow, and last for several hours to several days. Studies in neonatal experimental model suggest a quicker cellular destruction and energy substrates in the neonatal brain continue to run down for 12 h to 48 h after perinatal hypoxia [20]. Therefore, neuroprotective intervention might be effective 6 h to 8 h after perinatal hypoxic-ischemic insult [18]. Intervention and treatment following perinatal asphyxia may not be free of risk.

A CFM could be useful for selecting those neonates who might benefit from early intervention after perinatal asphyxia, avoid unnecessary risks related to treatment for those neonates that do not need intervention [21, 22]. A CFM provides a continuous real-time display of cerebral electrical activity at bedside to assist the clinician in making immediate treatment decisions and in identifying high-risk neonates, which requires closer attention. The benefit of CFM in a NeuroCare Unit (NCU) setting is its simplicity and ease of interpretation. It can be applied rapidly by nursing staff at any time of the day. We showed that aEEG correlates closely with neurologic outcome in neonates with HIE. The ability of aEEG background abnormalities to predict abnormal outcome has been previously studied in asphyxiated neonates [23]. Our study showed that neonates with normal aEEG patterns within the first 72 h of life were likely to survive without sequelaes, by contrast, neonates with severely abnormal aEEG patterns or worse pattern were at a risk of death or neurological sequela. These findings are in accordance with previous studies [24]. Our data also show that normalization of abnormal background patterns is associated with normal neurologic outcomes, whereas severely abnormal aEEG patterns that persisted beyond the age of 72 h are associated with adverse outcomes. These findings are in accordance with previous studies [25].

Diagnosis of seizures in a neonate has been based on clinical recognition of repetitive, stereotypic motor activity or behavioural phenomena. All neonates with either clinical or silent seizures were treated with antiepileptic drugs (phenobarbital as drug of first choice). Treatment with antiepileptic drugs has never changed a normal pattern into a severely abnormal one, although in some infants, the pattern became transiently more discontinuous than before, for 30 to 60 min. By excluding from analysis the parts of the records that were associated with drug administration or handling of the neonates we minimized possible confounding effects. A CFM aids in the detection of seizures and displays their severity, duration and frequency in real time [26]. In neonates with seizures, we found that the interictal aEEG activity correlated with a subsequent outcome. The neonates with seizures and a normal-amplitude aEEG had a normal outcome, whereas the neonates with seizures and moderately abnormal or suppressed amplitude aEEG had a poor outcome. A CFM also helps to evaluate response to anticonvulsive therapies and to identify subclinical seizures. Electrographic seizures without clinical correlates are common in neonates with HIE. From previous studies, it is estimated that > 50% of seizures identified on EEG or aEEG in neonates may be silent [27]. However, there are studies with the opposite results, reporting a rate of electroclinical dissociation seizure of approximately < 30%–50% in neonates with HIE, which is in accordance with our findings. Moreover, once anticonvulsant therapy is initiated, electrographic seizures may persist well after cessation of clinical seizure activity [28]. The importance of electrographic seizures is underscored by recent data showing that electrographic seizures with or without clinical correlates may have detrimental effects on the neonatal brain. Animal and human data indicate that seizures in the developing brain may be harmful, at least in the short term, considering disturbances in cerebral blood flow, energy metabolism, and excitotoxic amino acids. This suggests that anticonvulsant therapy that suppresses clinical but not electrographic seizures may not be fully effective in preventing brain injury and that an appropriate goal of anticonvulsant therapy is to suppress both clinical and electrographic seizures. Brief seizure activity may be missed, and neonatologists with limited experience in reading aEEGs may misinterpret the presence or absence of seizures [29]. These studies stress the fact that experience is required to be able to interpret aEEG traces, and one should also be aware of the limitations of the technology. The long duration of aEEG recording outweighs the limitations of obtaining detailed information during much shorter, 30–40 min of full montage EEG recording. Newer systems provide access to raw EEG, and may offer 2 to 4 channels of recording. The use of these two modalities in conjunction is likely to provide the best information at the current state of the technology [30].

A CFM is a useful tool in deciding when to initiate neuroprotective interventions and it can aid in evaluating the progress or recover from hypoxic-ischemic brain injury [31]. The CFM identifies neonates that require further neurological assessment by MRI or multi-lead EEG [32]. We did not investigate very preterm infants. Interpretation of aEEG is more difficult in preterm neonates because of EEG changes related to gestational age: a burst suppression pattern observed in extremely preterm neonates changes to a discontinuous pattern in more mature neonates [33]. Therefore, it is

very unlikely that immaturity influenced our results. Our study shows that when CFM is used in combination with standard neurological examination, it enhances the clinician's ability to identify neonates at risk for poor long-term neurodevelopment outcome. Just like monitoring of respiration, heart rate, and saturation is routine in neonatal intensive care settings for high risk neonates, continuous aEEG recording to monitor brain function may be appropriate for neonates with HIE and may be considered a standard of care.

Conclusion

In conclusion, our findings confirm that continuous aEEG is a simple and accurate bedside diagnostic method for assessing extension of hypoxic-ischemic brain damage and early identification of neonates with perinatal HIE who are at high risk of developmental delay. aEEG improves our ability to detect neonates at risk of hypoxic-ischemic brain injury at an earlier stage, when the window for therapeutic action is still open, optimize timing and assessment of neuroprotective treatment at the same time.

Acknowledgements

The authors are thankful to Dusica Gavrilovic, Institute for Radiology and Oncology, Department of Medical Education for her statistical help.

We declare that none of the authors has any competing interests with regard to the manuscript.

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Received on December 8. 2010.
Revised on February 1, 2011.
Accepted on February 24, 2011.

ORIGINAL ARTICLE

UDC: 616.322/.323-089.87/06:616-005.1]:616-053.2
DOI: 10.2298/VSP101231002M

Intensity of hemorrhage following tonsillectomy

Intenzitet krvarenja posle tonzilektomije

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Abstract

Background/Aim. Although post-tonsillectomy hemorrhage is one of the most frequent and potentially life-threatening complications, there is no generally accepted classification of post-operative bleeding intensity. The aim of this study was to evaluate the intensity of post-tonsillectomy hemorrhage according to the five-grade classification. **Methods.** A total of 408 consecutive patients, aged 2 to 54 years, undergoing elective tonsillectomy, with (n = 261) or without (n = 147) adenoidectomy, were included in this prospective study. Tonsillectomy was performed under general anesthesia using standard technique of cold dissection with a snare. Any bleeding event was recorded. The severity of post-operative hemorrhage was classified in five grades. **Results.** In 11 (2.70%) of the patients grade 1 hemorrhage following tonsillectomy occurred, 4 (0.98%) had grade 2 and 2 (0.49%) of the patients had grade 3 post-operative bleeding. Grades 4 and 5 were not recorded, and no patient received a blood transfusion. **Conclusion.** Post-tonsillectomy hemorrhage can be expected in a small number of patients undergoing tonsillectomy. Hemorrhage is mostly primary and rarely requires treatment under endotracheal anesthesia and blood transfusion.

Key words:
tonsillectomy; postoperative complications; hemorrhage; child, adult.

Apstrakt

Uvod/Cilj. Iako je krvarenje posle tonzilektomije jedna od najčešćih i potencijalno po život opasnih komplikacija, do danas ne postoji opšte prihvaćena klasifikacija intenziteta krvarenja. Cilj ovog rada bio je da se utvrdi intenzitet krvarenja posle tonzilektomije. **Metode.** Ova prospektivna studija obuhvatila je 408 bolesnika, starosti od 2 do 54 godine, kojima je načinjena tonzilektomija sa (n = 261) ili bez (n = 147) adenoidektomije. Tonzilektomija je izvođena u opštoj endotrahejskoj anesteziji standardnom tehnikom oštre i tupe disekcije sa omčom. Beleženo je svako krvarenje, a intenzitet krvarenja klasifikovan je u pet različitih stepena. **Rezultati.** Krvarenje stepena 1 zabeleženo je kod 11 (2,70%) bolesnika. Četiri (0,98%) bolesnika imala su krvarenje stepena 2 i dva (0,49%) bolesnika krvarenje stepena 3. Nije zabeleženo krvarenje stepena 4 i 5, kao ni primena transfuzija krvi. **Zaključak.** Krvarenje kao komplikaciju tonzilektomije možemo da očekujemo kod malog broja bolesnika. Krvarenje je uglavnom primarno i retko postoji potreba za ponovnom intervencijom u endotrahejskoj anesteziji i transfuzijom krvi.

Ključne reči:
tonzilektomija; postoperativne komplikacije; krvarenje; deca; odrasle osobe.

Introduction

In the literature, the data on the incidence of post-tonsillectomy hemorrhage are often conflicting. Over the years, the incidence of post-tonsillectomy hemorrhage has been reported from less than 1% to more than 10%. Some authors record only bleeding requiring hemostasis under general anesthesia, others include each bleeding event and recognise two [1] or three categories of bleeding [2] depending on the demand for treatment to achieve hemostasis. Up to the present day, there has been no universally accepted classification of post-tonsillectomy hemorrhage intensity. The five-grade classification of bleeding following tonsillectomy proposed by Windfuhr and Seehafer [3] records each bleeding event depending on the applied technique to accomplish hemostasis and blood transfusion. The aim of the study was to assess intensity and timing of post-tonsillectomy hemorrhage.

Methods

This prospective study was conducted in the Eaer, Nose and Throat (ENT) Department, University Hospital "Zvezdara" in Belgrade, during one year. The study included 408 consecutive patients (children and adults) undergoing elective bilateral tonsillectomy. Inclusion criteria were history of recurrent acute tonsillitis, chronic tonsillitis, quinsy,

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and hypertrophy of the tonsils (airway obstruction with sleep-disordered breathing and/or dysphagia). Exclusion criteria were personal or family history of bleeding disorder, unilateral tonsillectomy for suspected malignancy, tonsillectomy as a part of uvulopalatopharyngoplastica, and acute quinsy and infective mononucleosis. Tonsillectomy was performed by the five ENT consultants and three residents. All of them used the same technique for tonsil removal and hemostasis.

Tonsillectomy was performed under endotracheal anesthesia using cold steel dissection with snare, and bipolar diathermy for hemostasis. Pre-operative assesment of the patients included a complete blood count and bleeding time. Post-operative antibiotics were given for one week. All the patients had been admitted one day before and discharged on the third postoperative day. Traditionally, patients have to drink liquid two hours after procedure, and if it is possible, they are allowed to eat soft food. At the time of discharge, all the patients were scheduled to return for a routine follow-up appointment, one, two, four and six weeks following surgery, and all were placed on similar dietary and activity restrictions and given instructions to return immediately in the hospital if any bleeding occured at home. With this close supervision of the cases, both in the hospital and after discharge, the statistics given in this paper appear reliable.

The main outcome measure was the occurrence of any post-operative bleeding noted by the patients, parents or the staff. Primary and secondary bleeding events were recorded regardless of the severity or measures needed for their treatment and blood transfusion. In addition, patients` age and gender were recorded.

According to the criteria proposed by Windfuhr and Seehafer [3], the intensity of post-tonsillectomy hemorrhage was classified in grade 1 (stopped spontaneously or after clot removal), grade 2 (infiltration of local anesthetic necessary), grade 3 (treatment in general endotracheal anesthesia), grade 4 (ligature of the external carotid artery) and grade 5 (in case of lethal outcome). Primary hemorrhage following tonsillectomy was defined as any bleeding event occurring within the first 24 h after the operation. Secondary bleeding was defined as the reporting of any bleeding between 24 h and six weeks after the procedure. Primary and secondary post-operative bleeding events were grouped into five categories based on medical treatment used for achieving hemostasis.

Medical records data were entered into a computer spreadsheet program for statistical analysis. The χ^2-test, Mann-Whitney, and Fisher test were used to compare the groups to determine the presence of a statistically significant difference, taken as $p < 0.05$. Statistical analysis was performed by the IMSL routines for statistical analysis (IMSL Inc, 1989).

Results

During a one-year study period, 408 patients, aged from 2 to 54 years ($\overline{X}$ ± SD; 13 ± 10 years), underwent tonsillectomy, with (n = 261) or without (n = 147) adenoidectomy, in the ENT Department of the University Hospital"Zvezdara". There were significantly more patients who underwent tonsillectomy with adenoidectomy ($\chi 2$ test, $p \approx 0.00$). This study included significantly ($\chi 2$ test, $p \approx 0.00$) more children (n = 266) than adults (n = 142). The difference between the number of females (n = 196) and males (n = 212) was not significant ($\chi 2$ test, $p = 0.428$). In this study, males ($\overline{X}$ ± SD; 11 ± 9 years) were significantly younger (Mann-Whitney test, $p = 1.2 \times 10^{-5}$) than females ($\overline{X}$ ± SD; 16 ± 11 years).

The frequency of post-tonsillectomy hemorrhage in 408 patients according to the grade and timing of bleeding is shown in Table 1. Eleven patients had grade 1, four patients had grade 2 and two patients had grade 3 of post-operative bleeding. No ligature of the external carotid artery (grade 4) was recorded. In addition, there was no case with lethal outcome (grade 5) following tonsillectomy and no patient received blood transfusion.

Table 1

Frequency of post-tonsillectomy hemorrhage in 408 patients

Bleeding grade	Primary bleeding [n (%)]	Secondary bleeding [n (%)]	Total [n (%)]
1 º	6 (1.47)	5 (1.23)	11 (2.70)
2 º	3 (0.74)	1 (0.25)	4 (0.98)
3 º	2 (0.49)	0	2 (0.49)
4 º	0	0	0
5 º	0	0	0
Total	11 (2.70)	6 (1.47)	17 (4.17)

Post-tonsillectomy hemorrhage was primary in 2.70% of the patients occurring between 3 and 13 hours ($\overline{X}$ ± SD; 6 ± 3 h) after the surgery. Sixty four per cent of the patients bled during the first 6 after surgery and 82% of the patients had post-operative bleeding during 8 h after the surgery. Secondary hemorrhage was recorded in 1.47% of the patients and it occurred between the first and seventh postoperative day ($\overline{X}$ ± SD; 5 ± 2 days). The difference in the frequency between primary (65%) and secondary bleeding (35%) did not reach a statistical significance (χ^2 test, $p = 0.220$).

The overall hemorrhage rate was 4.17% (17/408). The patients who experienced post-tonsillectomy bleeding were aged between 4 and 40 years ($\overline{X}$ ± SD; 23 ± 2 years). Bleeding was recorded in 3.06% (6/196) of females and 5.19% (11/212) of the males with no significant difference ($\chi 2$ test, $p = 0.283$). The overall frequency of bleeding in the adult patients (14/142; 9.86%) was statistically significantly higher (χ^2 test, $p = 2.7 \times 10^{-5}$) than the frequency of bleeding in children (3/266; 1.13%). In addition, the overall frequency of bleeding in the group of patients who undergone tonsil-

lectomy without adenoidectomy (14/147; 9.52%) was statistically significantly higher ($\chi 2$ test, $p = 1.4 \times 10^{-4}$) than the frequency of bleeding in the group of patients who undergone tonsillectomy with adenoidectomy (3/261; 1.15%).

The comparison of post-tonsillectomy bleeding rates obtained in the this study and the study performed by Windfuhr and Seehafer (Table 2) showed no significant difference for grade 1, grade 2 and grade 4 bleeding. The difference between the two studies in hemorrhage rate for grade 3 and overall incidence of post-tonsillectomy bleeding were significant.

Table 2

Comparision of the results obtained in the present study and the study of Windfuhr/Seehafer [3]

Bleeding grade	Patients [n/total number (%)]		Test, *p*-value
	Present study	Windfuhr/Seehafer study	
1 º	11/408 (2.70)	21/602 (3.49)	χ^2, $p = 0.481$
2 º	4/408 (0.98)	1/602 (0.17)	χ^2, $p = 0.176$
3 º	2/408 (0.39)	14/602 (2.33)	χ^2, $p = 0.042$
4 º	0	2/602 (0.33)	Fisher, $p = 0.355$
5 º	0	0	0
Blood transfusion	0	0	0
Total	17/408 (4.17)	38/602 (6.31)	χ^2, $p = 0.140$

Discussion

Low overall incidence of post-operative bleeding (4.2%) in the present study confirms that tonsillectomy continues to be a very secure surgical procedure. Although post-tonsillectomy hemorrhage could be serious and life-threatening, most often it does not require any medical treatment. Usually the removal of a coagulum and infiltration of the tonsillar fossa with local anesthetic are sufficient to stop bleeding. The procedure under endotracheal anesthesia (0.4%) is rarely necessary for achieveing hemostasis and blood transfusion.

The overall incidence of postoperative hemorrhage of 4.2% reported in this study appears similar to the rates described in recently published papers on cold dissection tonsillectomy in non-selected patients [3–9]. In the literature, there is no agreement on the definition of significant or major bleeding, and consequently no uniform method for quantifying hemorrhage following tonsillectomy exists. As a result, some authors count all bleeding events and therefore report higher haemorrhage rates than do authors who include only hemorrhages requiring surgical intervention. Some authors classify posttonsillectomy bleedings in two [1] or three grades [2] depending on the need for surgical treatment in general anesthesia to achieve hemostasis. The advantage of generally accepted classification would include a direct comparison between two studies or different techniques and reliable evaluation of a vary rare case of severe post-tonsillectomy bleeding with lethal outcome.

In case of post-operative bleeding, effort has to be made to avoid treatment under second general anesthesia. Bleeding requiring no procedure under general anesthesia can be minimal and stop spontaneously without any medical treatment or after cloth removal. In addition, infiltration of tonsil fossa with local anesthetics is often sufficient for bleeding control. The management of more severe bleeding requires a procedure in general anesthesia including bipolar diathermy or suture ligation with or without blood transfusion. Rarely severe bleeding could not be sufficiently controlled by these procedures and multiply blood transfusion, arteriography, embolisation and/or ligature of external carotid artery could be necessary. Bleeding events with lethal outcome still exist although very rarely, or maybe their incidence could be underestimated.

Classification of postoperative hemorrhage includes five types of bleeding events according to medical treatment requiring to achieve hemostasis and blood transfusion. It allows us a detailed analysis of this unpredictable and potentially life-treathening complication and a direct comparision of different studies. In a one-year retrospective study of cold disection tonsillectomy with suture ligation hemostasis, Windfuhr and Seehafer [3] recorded bleeding grade 1, grade 2, grade 3 and grade 4 in 3.49%, 0.17%, 0.33% and 0.33% of 602 non-selected patients, respectively. One patient received blood transfusion and no bleeding with lethal outcome was recorded. The comparision between the study Windfuhr and Seehafer and our study points out a very similar post-tonsillectomy bleeding rate except for grade 3 bleeding, which was slightly lower in our study. In our experience, the five-grade classification of post-tonsillectomy hemorrhage is clear-cut and very helpful. It allows us a reliable comparison of different studies or techniques and helps us to improve preoperative and postoperative management as well as technique of tonsillectomy and hemostasis.

The cause of primary bleeding is generally acknowledged to be inadequate hemostasis during the procedure. Although the cause of secondary bleeding is less certain, the sloughing of the superficial eschar from the tonsillar fossa is believed to be the inciting event. No agreement on primary and secondary bleeding incidence exists. Some studies reported a higher rate of primary hemorrhage [3], but some articles cite a lower rate of primary bleeding and describe the rate of secondary hemorrhage as low and stabile [4–9]. The differences seem to be a consequence of no universally accepted definition of bleeding and classification scheme. In the present study, the overall rate of primary bleeding was higher than the rate of secondary bleeding. This is consistent with the findings of a recent study [3].

According to recently published studies, the technique of hemostasis [4], quinsy tonsillectomy [5, 6] and the surgeon experience [7] have no significant influence on bleeding inci-

dence. The bleeding rate seems to be somewhat higher among older patients and males. In present study, no significant difference in post-tonsilelectomy bleeding rate between females and males was found. Older patients and patients with tonsillectomy without adenoidectomy bled significantly more often than younger patients and patients with the tonsillectomy with adenoidectomy, which is consistent with results of previous studies [8, 9].

Usually, authors do not report data on post-operative diet and the length of refrain from drinking and eating [5, 7, 8, 10–12]. Early return to normal diet with liquid and soft food is important to avoid dehydratation and, probably, to improve healing of tonsillar bed. In our Department, traditional post-operative diet includes cold liquid intake 2 hours post-tonsillectomy while Windfuhr and Sehafer's study protocol delays liquid intake for 6 hours [3].The post-tonsillectomy healing process is characterized by a reactive inflammation with healing by second intention and the role of post-operative diet in healing tonsillar bed remains unclear.

Conclusion

Post-tonsillectomy hemorrhage can be expected in a small number of patients undergoing tonsillectomy. Hemorrhage is mostly primary and rarely requires treatment under endotracheal anesthesia. Any tonsillar bleeding is significant, but those requiring operative intervention to arrest hemorrhage are by their nature more serious.

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Received on December 31, 2010.
Revised on April 13, 2011.
Accepted on April 21, 2011.

ORIGINAL ARTICLE

UDC: 617.583-06-022.1
DOI: 10.2298/VSP1206504R

Infected total knee arthroplasty treatment outcome analysis

Analiza ishoda lečenja infekcije totalne artroplastike kolena

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Abstract

Background/Aim. Infected total knee arthroplasty (TKA) is a topic of great importance, because its diagnosing and treatment requires a lot of resources, and often has an unsatisfactory outcome. The aim of this study was to analyze the outcome of the treatment of infection developed following TKA. **Methods.** This retrospective study of infected TKAs was performed in the period from 1998 to 2008 in the Orthopedics & Traumatology Clinic of the Military Medical Academy (MMA) in Belgrade. A total of 654 primary and revised TKAs were performed in the said period. We registered and surgically treated 28 infected TKAs (primary TKAs: MMA – 22, other institutions – 6). The incidence of TKA infection in the MMA was 3.36%. The most common pathogens were: S*taphylococcus aureus* – 14 (50%) cases, and *Staph. epidermidis* – 3 (10.7%) cases. Other isolated pathogens were: *Enterococcus faecalis*, *Klebsiella pneum*., *Klebsiella spp*., *Streptoccocus viridans*, *Seratia spp*, *Micrococcus luteus* and *Peptostreptococcus spp.* In one case we had mixed anaerobic flora, and in 3 cases cultures were negative. We analyzed diagnostic challenges, risk factors (such as age and previous viscosupplementation) and treatment outcomes in our series of infected TKAs. **Results.** In our series 2 infections healed after *iv* antibiotics and debridement, 1 patient responded to open debridement with component retention, 4 patients responded fully to one-stage reimplantation, 10 cases responded fully to two-stage reimplantation, 11 patients ended with arthrodesis and we had 1 patient withabove knee amputation. **Conclusion.** Two-stage reimplantation remains gold standard for treatment of infected TKA, and we recommend it as treatment of choice for eradication of infection. The antibiotic loaded spacer prothesis concept in most cases allows infection eradication, good function and high patient satisfaction.

Key words:
arthroplasty, replacement, knee; bacterial infections; diagnosis; drug therapy; anti-bacterial agents; orthopedic procedures; treatment outcome.

Apstrakt

Uvod/Cilj. Infekcija totalne artroplastike kolena (TKA) je tema od velikog značaja, budući da dijagnoza i tretman zahtevaju značajne resurse sa često nezadovoljavajućim ishodom. Cilj ove studije bio je analiza ishoda lečenja infekcije nastale nakon totalne artroplastike kolena. **Metode.** Ova retrospektivne studija infekcije TKA obuhvatala je period od 1998. do 2008. godine u Klinici za ortopediju i traumatologiju Vojnomedicinske Akademije (VMA) u Beogradu. U navedenom periodu izvedene su 654 primarne i revizione TKA. Registrovali smo i hirurški lečili 28 infekcija TKA (22 TKA primarno izvedene u VMA i 6 TKA primarno izvedenih u drugim ustanovama). Incidencija infekcije u VMA bila je 3,36%. Najčešći uzročnici bili su: *Staphylococcus aureus* – 14 (50%) slučajeva i *Staph. epidermidis* – 3 (10,7%) slučaja. Ostali izolovani patogeni bili su: *Enterococcus faecalis*, *Klebsiela pneum*., *Klebsiela spp*., *Streptoccocus viridans*, *Seratia spp*, *Micrococcus luteus* i *Peptostreptococcus spp.* Kod jednog bolesnika ustanovljena je mešovita anaerobna flora, a u tri slučaja kulture su bile negativne. Analizirali smo dijagnostičke izazove, faktore rizika (između ostalih godine života i prethodne viskosuplementacije) i ishod lečenja u našoj seriji TKA. **Rezultati.** U našoj seriji dve infekcije sanirane su nakon *iv* primene antibiotika i debridmana, jedan bolesnik izlečen je debridmanom sa zadržavanjem komponenata, četiri bolesnika izlečena su *one-stage* reimplantacijom, 10 bolesnika primenom *two-stage* reimplantacije, 11 bolesnika je rešeno artrodezom i urađena je jedna natkolena amputacija. **Zaključak.** *Two-stage* reimplantacija predstavlja zlatni standard u lečenju infekcije TKA, i preporučujemo je kao metodu izbora za eradikaciju infekcije. Koncept primene antibiotskih *spacer* proteza u najvećem broju slučajeva omogućuje eradikaciju infekcije, dobru funkciju i visok stepen zadovoljstva bolesnika.

Ključne reči:
artroplastika kolena; infekcija, bakterijska; dijagnoza; lečenje lekovima; antibiotici; ortopedske procedure; lečenje, ishod.

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Introduction

Average population in many countries is getting older, and total knee arthroplasty (TKA) is becoming more often performed. Infected TKA is a topic of great importance, because it is a diagnostic challenge, requires a lot of resources and often has an unsatisfactory outcome. Infection after TKA is catastrophic both to patients and surgeons. It can cause persistent pain, and loss of function. It can damage the periarticular bone, causing prosthesis loosening and compromising further revision surgery, and may end up in septicaemia, even life-threatening conditions. Reported rates of infected TKA in the literature are about 2% [1, 2] and about 0.4% for those ensuing within 3 months after operation [1]. Risk factors have been identifed [1, 2] and various methods have been devised to decrease the chance of this complication [1–4]. In this study, we tried to determine the infection rate of TKA, the most common bacteria, and the succes rate of various surgical treatment options in the Military medical Academy (MMA) in Belgrade. We also attempted to determine the risk factors for infection and ways of minimising the risk.

Numerous studies have shown the TKA infection incidence of less than 1% to 2%. The average annual cost of managing infected TKAs in the United States is $150 to $200 million [5]. Infected TKA requires 3 to 4 times the hospital resources when compared with primary TKA, and double the resources when compared with aseptic revision TKA [5]. An exact estimate of the costs in Serbia is not yet known since the data we have are only from our institution's artrhoplasty register. We consider that primary uncomplicated TKA peformed in MMA requires an average of 6.32 days of hospitalization, while infected TKA requires an average of 4.92 hospitalizations with a total of 39.2 days of hospital treatment.

The aim of this study was to analyze the outcome of the treatment of infection developed following TKA.

Methods

This is a retrospective study of all surgically treated infected TKAs in the MMA. Our team of orthopaedic surgeons reviewed all relevant medical records in the period from january 1998 to december 2008. A total of 654 TKAs were performed on 533 patients. There were 121 bilateral TKAs in single procedure, and others were unilateral TKAs or bilateral as two-stage procedure. Out of a total number, 386 patients were female and 147 were male. The mean patient age was 70.1 years ranging from 36 to 94 years. The mean patient age in the infected TKA group was 74.8 years while in the noninfected TKA cases the mean age was 69.7 years. We used different total knee prosthesis (Zimmer, Depuy-Johnson and Johnson, and Stryker models) and all TKAs were cemented. The mean follow-up period for infected TKAs was 39 months.

We define TKA as infected when there are: 2 or more cultures positive, or cultures negative (in less than 7%) but we have clinical and laboratory manifest infection, acute inflammation or purulence at the time of the surgery, sinus tract. General signs of infection – fever, chills, malaise, wound erythema, are uncommon in the most infected TKAs. Sometimes the only presenting symptoms such as pain, sweelling, warmth and synovitis are hard to distinguish from aseptic failure. TKA infections are primarily bacterial, although fungal ones were reported [6]. They are hematogenous or they occur after intraoperative contamination.

Diagnosis of infected TKA consists of: anamnesis – local status; laboratory findings (CBC, sedimentation, CRP, fibrinogen; knee aspirate – analysis of synovial fluid and culture); and imaging – radiography, scintigraphy.

Laboratory findings for infected TKA – sedimentation and CRP are highly sensitive but lack specificity. We noted increased sedimentation in 85% of cases CRP was elevated in 96%, fibrinogen was increased in 75% and leukocytosis was presented in 36% of cases. Silva et al. [7] and Rand and Brown [8] noted that only 28% of cases had leucocytosis greater than 11,000 in the presence of deep knee infectio.

Knee aspirate serves for assesement: the color, clarity and, viscosity of the fluid – string test, cell count – Le > 20,000 with more than 65% polymorphonuclear leukocytes (PMN) (values are significantly lower than in septic arthritis of the knee that was not subjected to previous arthroplasty), culture and antibiogram (antibiotics should be discontinued two weeks before taking swab or aspirate). Histological examination of intra-operative tissue could also be taken as suggestive of infection, as could intraoperative findings of pus, turbid fluid, unhealthy granulation tissue, synovitis, bone destruction, and prosthesis loosening [9].

Radiography is usually more useful in chronic infected TKAs. It can show the presence of radiolucent zones. In the presence of long lasting infections we can also see periosteal bone formation (indicating poor prognosis these knees have bad response to two-stage reimplantation), and loosening of components. Fistulography is rarely indicated for infected TKA.

Scintigraphy is used in the evaluation of infected knee arthroplasty with variable results. Indium scans have good sensitivity but poor specificity. Rand and Brown [8] found that indium scans had a sensitivity of 83% and accuracy of 83%. We used scintigraphy in 2 cases (two cases with negative cultures). We had focal hot uptake around the prosthesis – positive results in all the cases, but we could not know for sure, in one case of failed TKA, if it was mechanical or septic loosening. We do not recommend routine use of radioisotope scanning since other diagnostic methods are cheaper, safer, faster and more reliable.

In the last two decades numerous studies have identified risk factors, and the immeasurable importance of adequate recognition. Immunocompromised host factors (leukopenia and malnutrition) and existing infections are of the utmost importance. Care should be taken to minimize risks for possible sources of contamination operating room (OR) environment, the skin of the patient or remote sources – dental infections, chronic ulcers, urinary tract infections, gastrointestinal, gynecological infections, even endocarditis. In order to minimize the risks of contamination, the necessary measures are: prophylactic antibiotics, sterile technique, drapes,

self contained exhaust suits, ORs with less traffic, laminar flow, carefull closing.

Viscosupplementation as a risk factor – in the study on effect of intraarticular hyaluronic acid agents on subsequent rate of infection following TKA Petrella and Mahadeva [10] recorded 18 infections after TKA in a group of 415 TKA, who had previously received intraarticular viscosupplementation injections, and 21 infected TKA in the group of patients who had not recieved viscosupplementation previous to knee arthroplasty. In our series we identified patients who had underwent previous knee viscosupplementation, there were 654 TKAs and 126 of them had underwent knee viscosupplementation previous to TKA. 28 patients had infected TKA only 2 of them were given viscosupplemetantion knee injections prior to TKA. Since there was no statistically significant relationship between viscosupplementation and infection after TKA we could not say that viscosupplemenation injections promote infections.

Classification of TKA infection – Tsukayama et al. [11] presented a classification of infection based on clinical presentation (they differentiate between early, acute onset < 4 weeks post surgery, subdivided in superficial and deep, and late, chronic onset > 4 weeks post surgery). The two remaining categories are: acute hematogenous infection and infection based on positive intraoperative culture. We used this classification and we have registered 15 acute infections (< 4 weeks post surgery) two superficial and all other were deep infections. We noticed 13 late chronic infections (> 4 weeks post surgery).

Prophylactic measures in the MMA – one operating room is assigned for total joint replacements. We have not routinely used body exhaust suits, but water repellant paper gowns, drapes, and double gloves have been mandatory. We prepare the lower limb distal to tourniquet with kodan tincture twice: once by an assistant surgeon before gowning, and once more by surgeon, and routinely change the outer pair of gloves after draping. Prophylactic antibiotic has always been given (in most cases 1g of ceftriakson or 1g of cefazolin) 1 hour preoperatively, antibiotic therapy continued in the same dose the next three days. In all cases we administered low molecular weight heparin (LWMH) once or twice daily (according to recommended dosage of specific product).

Results

In a period 1998–2008 in the MMA – Orthopaedics & Traumatology Clinic, 654 primary and revision TKAs were performed. During this period we registered and surgicaly treated 28 infected TKAs (MMAs primary TKAs – 22, other institutions primary TKAs – 6). The incidence of TKA infection in MMA was 3.36%. In our series of infected TKAs the most common pathogenes were *Staphylococcus aureus* – 14 (50%) cases, and *Staph. epidermidis* – 3 (10.7%) cases. Other isolated pathogens were: *Enterococcus faecalis, Klebsiela pneum., Klebsiela spp., Streptoccocus viridans, Seratia spp, Micrococcus luteus* and *Peptostreptococcus spp.* In one case we had *mixed anaerobic flora*, and in 3 cases cultures were negative. The most common bacteria in the study of Silva et al. [7] on 590 infected TKA were: *Staphylococcus aureus* 48.7%, *S. Epidermidis* 16.3%, *Pseudomonas* 5.1%, *Streptococcus* 4.8%, *Enterococcus* 4.5 %, *polimikrobe flora* 5.4% *i others* 15.2%).

We registered 2 superficial and 26 deep infections. Superficial infections were treated by *iv* antibiotics and surgical debridement. Both healed well after the treatment, both patients enjoyed a painless knee without any signs of infection at the follow-up. A total of 26 deep TKA infections required a more serious approach. In the MMA we adopted the algorithm of treatment of TKA infections that all the MMA surgeons have to follow. Our strategy after clinical and microbiological confirmation of infected TKA was: debridement and, if found as viable option, retention of components or one-stage reimplantation, two-stage reimplantation, then if two-stage reimplanatation fails the next option was arthrodesis, and finally if everything else fails above knee amputation.

In our series, 2 infections healed after *iv* antibiotics and debridement, 1 patient responded to open debridement with component retention, 4 patients responded fully to one-stage reimplantation, 10 cases were successfully treated with two-stage reimplantation, 11 patients ended with arthrodesis (9 external fixator, 2 plating technique) and we had 1 case of above knee amputation.

We performed two stage reimplantation in 21 cases. The rate of infection eradication in TKA with two-stage reimplantation was 76.19%.

Discussion

Infection eradication, alleviation of pain and restoration of function are goals of treatment of infected TKA. In effort to achieve the first two goals, we often have to sacrifice the function.

In general, treatment options for infected TKA are: treatment that retains prostheses – antibiotic suppression and open debridement with component retention; exchange arthroplasty (one- and two-stage reimplantation); salvage procedures – arthrodesis, resection arthroplasty and amputation.

Antibiotic suppression therapy if chosen is usually lifelong 12. It is only adequate in the earliest and most benign infections and it can be considered if a patient is not suitable for surgery, if there is low bacterial virulence, no sepsis, no severe antibiotic side effects and if prothesis is stable. In our series we did not have patients on lifelong antibiotic suppression.

Some authors recommend arthroscopy in management of infected TKA: and/or continuous irrigation [13–16]. Arthroscopy theoretically has several advantages over open debridement: it is less invasive, has faster rehabilitation, but has also disadvantages to open debridement: poly change is not possible, debridement is less radical, possibility for eradication is worse than in open debridement.

We perfomed arthroscopic debridements and irrigations in five cases of infected TKA, but with poor results, which required further surgical treatment, so we cannot speak in favor of this kind of treatment.

Open debridement with component retention as a treatment option has few advantages, including reduced stress for a patient, less bone loss, better function of the knee, and less cost to the health care system [17]. Following criteria necessary for this treatment approach are: low-virulence organism (sensitive to antibiotics), acute infection (< 4 weeks), no signs of component loosening, no sinus tracts. Deirmengian et al. [18] used open irrigation and debridement to treat 33 patients with acute gram-positive infected TKA with poor success. Of the 31 patients who underwent debridement with component retention, 20 (65%) experienced recurrent infection and eventual removal of components. We used open debridement with component retention in 8 cases, yet only one of them responded. It was the patient whose culture was positive to *Seratia species*, all the other infected TKA underwent further one- or two-stage reimplantation.

One-stage reimplantation as treatment option offers possibility to only one surgery, but there is no second chance, no second debridement, no local antibiotics, and it is not recommended if bacterium is not identified. One-stage reimplantation may be a viable alternative to the two-stage revision procedure. In one-stage surgery, incision and arthrotomy are performed in the usual way; and thorough irrigation and debridement are done, all infected total knee components are removed, and new total knee components are placed during the same session. In the study of Buechel et al. [19–22] infected total knees were revised using single-stage resection and reimplantation. Out of 21 patients nineteen showed no signs of recurrent infection at an average of 10.2 years of follow-up.

In our series of infected TKA we had 50% rate of success with this approach, there were 4 successful one-stage reimplatations out of 8 perfomed. All the patients were then treated with 4 weeks of *iv* antibiotics, followed by 2 months of oral antibiotics, in all cases we consulted the infectious disease specialist.

Two-stage reimplantation consists of two stages**.** In stage one after confirming the diagnosis of infected arthroplasty, first steps are irrigation, debridement, and resection of components. Polyethylene, tibial, femoral, patellar components and bone cement are removed. Care must be taken to preserve as much bone stock as possible to allow a foundation for the reimplantation. The soft tissues are thoroughly debrided, the joint is then irrigated using antibiotic solution. The second part of stage one of the procedure is to place an antibiotic spacer in the joint to maintain the space for reimplantation of the components in the future and possibly to allow the patient to ambulate until component reimplantation can be completed. The spacers are made with bone cement impregnated with antibiotics. Cement allows antibiotics to elute into the joint and surrounding tissues over time, usually several weeks, to help eradicate infection. The goal of the spacer is to provide patient comfort and mobility, prevent the loss of joint space, enhance bone quality, and allow for treatment of the infection locally with time-released high local concentration of antibiotics [20]. Spacers can be simple self-made block-shaped or articulating devices. Recent studies suggest that using an articulating antibiotic spacer allows for better functioning for patients between the stage I and stage II procedures [21–23]. Articulating spacers may be custom made – the components removed at debridement can be reused to construct an articulating spacer. The femoral component is debrided, cleared of adherent bone and cement, autoclaved for 20 minutes, and coated with antibiotic-impregnated cement on its nonarticulating surface. A new polyethylene insert is open, and is coated, too, on its nonarticulating surface, and both are implanted with cement in a doughy stage, so that there is limited interdigitation with bone [22]. Or articulating spacers may be constructed from prefabricated cement molds. After an antibiotic spacer is implanted, a patient may be allowed to have at least partial weight bearing and range of motion of the joint. The patient is also treated with *iv* antibiotic therapy, with the choice of antibiotic dependent on microorganism culture and sensitivity results. Courses of antibiotics range from 14 days to 12 weeks [24–26].

The stage two occurs after the infection is eradicated. Indications for reimplantation are good bone stock, adequate soft tissues, immunocompetent patient and sensitive microorganism. Contraindications for reimplantation are persistent infection, immunocompromised host, extremely poor bone stock, poor soft tissues.

The lapse of time between the first-stage and second-stage procedures ranges from 4 to 58 weeks and depends on a patient's medical condition, a physical condition of the joint itself, and results of repeated aspiration/culture and tests for inflammatory markers [22, 24–26]. Stage two consists of removal of antibiotic spacers, debridement, and reimplantation of total knee components, in some cases revision knee implants are needed. The two-stage revision procedure is generally considered to be the gold standard treatment for infected TKA. Cuckler [26] had no recurrence of infection with an average follow-up of 5.4 years for 44 infected TKA treated with two-stage revision using articulating spacers. In a study by Hirakawa et al. [24], a success rate of 66.7% was found when high-virulence organisms (*Staphylococcus aureus*, *Enterococcus* species, methicillin-resistant *S. aureus*) were involved. The success rate was 80% when infection was with low-virulence organisms (*Staphylococcus epidermidis*, *Streptococci*, *Proteus* species) and 71.4% with polymicrobial organisms [24].

We performed two stage reimplantation in 21 cases. When we closed folow-up in june 2010 and headed to the processing of data, we found 10 patients responding fully to two stage reimplantation and had good functional outcome, while the other 10 ended with arthrodesis and one patient had above knee amputation. The rate of good functional outcome with two-stage treatment was 47.6%. But we noticed that the mean follow-up period in the patients who had good functional outcome was 35 months, much shorter than in the group who ended with arthrodesis or amputation where the mean follow-up was 47 months. We must say that 6 of 10 patients who ended with arthrodesis did not undergo this procedure because of infection, they had mechanical problems due to instability and loosening of prosthesis components. We are under the impression that many patients after

two-stage reimplantation after a longer period of follow-up are in need for revision arthroplasty or arthrodesis due to loss of bone stock, or soft tissue problems. In our series only in 5 cases out of 21 of infected TKA, two-stage reimplantation failed to eradicate infection. So, the rate of eradication of infection in TKA with the two-stage reimplantation approach was 76.19%. In our series we have used self-made articularting spacers in all cases, and *iv* antibiotics from 2 to 4 weeks. In some cases we added orally rifampin 2 × 600 mg with *iv* antibiotics, and rifampin 600 mg 1× for 4 weeks orally after we discontinued *iv* antibiotics, based on infectious diseases consultants recommendations.

Salvage procedures for infected TKA are: resection arthroplasty arthrodesis and amputation.

Resection arthroplasty as a definitive treatment for infected TKAs is reserved for patients who are medically ill and sedentary. It results in a significant loss of function, instability, and potentially persistent pain. In most cases, arthrodesis require is eventually. In our series of infected TKA we did not perform resection artrhoplasties. But in the MMA we found that it could be a viable option for some cases of infected total hip arthoplasty.

Arthrodesis is the treatment of choice when it is thought that reimplantation will have a high rate of failure, due to inadequate joint mechanics, soft tissue envelope, or immune system deficiency.

The relative contraindications to arthrodesis of infected TKA are: significant contralateral limb dysfunction, coexistent ipsilateral ankle or hip disease, inadequate bone stock for fusion. The available arthrodesis techniques are: intramedullary nailing, external fixation and plates and screws. We had 11 arthrodesis after infected TKA, in 9 cases we applied an external fixator (7 – type Mitković and 2 – Ilizarov), and in two cases we used the double plating technique.

Amputation is indicated when other attempts to salvage the knee have failed and when further salvage procedures would likely be ineffective. One patient in a 42-months-period from primary TKA, underwent multiple debridements, two-stage reimplantation and arthrodesis. Refractory infection (negative *Staph. aureus*) and severe bone defects compromised all reconstructive and salvage procedures. Above knee amputation was a definitive treatment.

Conclusion

Two-stage revision arthroplasty is the most commonly used treatment for infected TKA and had the best treatment results reported in the literature. However it is not almighty procedure, though numerous studies percentages of success are individually different.

Since success is not always guaranteed, a patient should be always aware of the back-up options and in best case scenario surgeons should expect some loss of function after two stage reimplantation.

Two-stage reimplantation remains gold standard for treatment of infected TKA, and we recommend it as treatment of choice for eradication of infection. The antibiotic loaded spacer prothesis concept in most cases allows infection eradication, good function and high patient satisfaction.

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Received on January 4, 2011.
Revised on February 3, 2011.
Accepted on February 11, 2011.

A K T U E L N A T E M A

UDC: 616.89-07/-08:[159.95::616.895.8
DOI: 10.2298/VSP1206510T

Neuropsihološka procena i mogućnosti lečenja kognitivnog deficita kod shizofrenih bolesnika

Neuropsychological assessment and treatment possibilities of cognitive deficit in schizophrenic patients

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Ključne reči:
shizofrenija; kognicija; antipsihotici; rehabilitacija.

Key words:
schizophrenia; cognition; antipsychotic agents; rehabilitation.

Uvod

Shizofrenija je danas prihvaćena kao neurobiološki poremećaj sa snažnim neurokognitivnim komponentama [1, 2]. Tokom poslednjih nekoliko decenija značajna su neuropsihološka istraživanja zasnovana na utvrđivanju specifičnosti neurokognitivnih obrazaca povezanih sa shizofrenijom što doprinosi razjašnjavanju neuroanatomskih i neuropsiholoških sistema koji leže u osnovi bolesti dajući klinički doprinos razvoju efikasnijih rehabilitacionih mera [3]. Rasprava o „funkcionalnoj" u odnosu na „organsku" prirodu shizofrenije bazirana na neurobiološkoj konceptualizaciji ovog poremećaja još od vremena Emila Krepelina, aktuelna je i danas [4]. Neuropsihološki deficiti utvrđeni u shizofreniji u okviru brojnih kognitivnih sposobnosti, uobičajeno se opisuju kao „generalizovani deficit" [5]. Prilikom analize uočenih deficita, suština neuropsihološkog pristupa ogleda se u integraciji strukturne i funkcionalne organizacije moždanih sistema (na primer, analiza funkcija vizuelne integracije kroz strukturnu organizaciju veza između neurona okcipitalnog korteksa), za razliku od dosadašnjeg pristupa koji se bazirao na razlikama između strukturne i funkcionalne organizacije ovih sistema. Neuropsihološki poremećaji čiji se intenzitet kreće od umerenih do izraženih prisutni su kako tokom prve epizode bolesti, tako i tokom niza godina sa minimalnim promenama uprkos postignutoj kliničkoj stabilizaciji primenom farmakoterapije, ne menjajući se značajno čak ni tokom ponovnih epizoda bolesti [5, 6]. Opšti zaključak iz dosadašnje literature ukazuje da su neurokognitivni deficiti u okviru različitih kognitivnih domena sastavni deo kliničke slike shizofrenije, pa se, stoga, fokus novijih istraživanja usmerava ka uočavanju kauzalne veze između kognitivne disfunkcije i određenih psihopatoloških fenomena (na primer, povezanost deficita u radnoj memoriji sa poremećajima mišljenja) [7]. Učestalost kognitivnog poremećaja varira u zavisnosti od podgrupa shizofrenih bolesnika sa primarno pozitivnim i primarno negativnim simptomima ili između paranoidne i neparanoidne podgrupe, a rezultati neuropsiholoških istraživanja govore u prilog izraženijeg neuropsihološkog deficita u podgrupi sa primarno negativnim simptomima, odnosno neparanoidnoj podgrupi [8]. Razmatranjem uloge farmakoterapije kod shizofrenih bolesnika može se zaključiti da primena antipsihotika ne utiče značajno na prisustvo kognitivnog poremećaja, zbog čega se može prihvatiti stav da je kognitivni poremećaj u shizofreniji relativno stabilna varijabla koja ima karakteristike crta, bez obzira što intenzitet kognitivne disfunkcije tokom bolesti može da varira [9, 10].

Neurobiološki korelati kognitivne disfunkcije u shizofreniji

Precizna neuropatološka osnova shizofrenije i povezanost sa neurokognitivnim deficitima još uvek je nedovoljno jasna. Utvrđeni su neki od bazičnih obrazaca neuropatoloških korelata potvrđenih studijama magnetne rezonance (MR) kao što su proširenje moždanih komora, redukcija volumena sive mase (posebno u predelu gornje temporalne vijuge, medijalnog dela temporalne regije i limbičkih struktura kao što su amigdala, hipokampus, parahipokampalna vijuga), redukcija volumena frontalnih i parijetalnih režnjeva, izostanak nor-

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malne asimetrije, promene u veličini i obliku korpusa kalozuma, limbičkom sistemu, talamusu i bazalnim ganglijama) [11–14].

Proučavanje prefrontalnih i temporalnih kortikalnih disfunkcija u shizofreniji ukazuje da ove promene mogu biti osnov kognitivnih abnormalnosti prisutnih kod obolelih osoba, što je potvrđeno u studijama koje koriste metode vizuelizacije mozga, kao što su pozitron emisiona tomografija (PET) i pozitron emisiona kompjuterizovana tomografija (SPECT), kojima je utvrđena abnormalna aktivacija ovih regiona na vizuelnim i verbalnim zadacima [2, 15].

Među „kognitivnim" aspektima funkcija bazalnih ganglija najznačajnija je njihova uloga u kontroli pažnje, funkcionisanju radne memorije i proceduralnom učenju [16].

Novije studije fokusiraju pažnju na talamus, kao centralno moždano senzorno raskršće, implikujući značaj ove strukture u patofiziologiji shizofrenije [17]. Činjenica da je medijalni talamus ključna struktura u komunikaciji između različitih asocijativnih kortikalnih oblasti ukazuje na to da bi promene u ovom talamičkom jedru mogle dovesti do disfunkcije u kortikosupkortikalnim i kortikokortikalnim vezama i podržava diskonekcionu hipotezu shizofrenije [17]. Pri proceni kratkoročne memorije, zapaženo je povećanje metabolizma glukoze u medijalnom talamusu, što ukazuje da ova struktura može biti uključena u prve faze obrade informacija za pamćenje i učenje, dok pulvinar i talamička retikularna jedra imaju ulogu u različitim aspektima regulacije pažnje [18] .

Mali mozak ima kompleksnu strukturu i zahvaljujući bogatoj unutrašnjoj beloj masi svojih projekcija direktno je povezan sa korom velikog mozga i talamusom, a preko moždanog stabla sa bazalnim ganglijama, retikularnim i perifernim sistemom. Funkcionalne studije i studije magnetne rezonance mozga pokazale su cerebelarno oštećenje kod shizofrenije, a dobijeni podaci upućuju da mali mozak ima značajnu ulogu u različitim moždanim funkcijama. Pored motorne kontrole u obezbeđivanju preciznosti, povezivanju pokreta u koordinisane složene obrasce ili proceduralnom učenju, učestvuje u emocionalnoj regulaciji, senzornoj obradi (na primer, u vremenskoj organizaciji i percepciji), jezičkim kao i drugim kognitivnim funkcijama [19, 20] .

Postoje dokazi da bolesnici sa shizofrenijom imaju promenjene kortiko-cerebelarne konekcije [21]. Andreasen i Pierson [22] predložili su da na poremećaju kortikalno-talamičko-cerebelarno-kortikalnih kola (KTCKK) može da se zasniva bar deo simptomatologije koja se sreće u shizofreniji. Analogno cerebelarnoj ulozi u omogućavanju brzog i nesmetanog izvođenja motornih zadataka, autori su predložili da KTCKK mogu imati sličnu ulogu u praćenju i koordinaciji izvršenja mentalnih aktivnosti što rezultuje normalnim kognitivnim funkcionisanjem [21, 23]. Globalno smanjenje cerebeluma kod bolesnika sa shizofrenijom izgleda da je u nekim slučajevima udruženo sa perinatalnim povredama mozga [24], sa muškim polom [25], početkom u detinjstvu [26], ali i kod bolesnika sa kasnim početkom [27], hroničnim tokom [28] i pozitivnim psihotičnim simptomima [29]. Drugi autori su primetili da je atrofija ograničena na predeo vermisa [25]. Abnormalnosti vermisa su češće u zadacima koji aktiviraju limbički region (npr. studije emocija), dok su bočni neocerebelarni regioni disfunkcionalni u zadacima koji aktiviraju neokortikalne regione (npr. kodiranje memorije i pronalaženje) [22, 30]. Protok krvi u cerebelumu bolesnika sa shizofrenijom je smanjen u širokom spektru zadataka koji uključuju različite funkcionalne sisteme mozga, pamćenje, pažnju, socijalnu kogniciju, i emocije [30].

Neuropsihološki profil kognitivnog deficita u shizofreniji

Utvrđivanje specifičnosti neurokognitivnog profila u shizofrenijii vrši se, najčešće, upoređivanjem shizofrenih bolesnika sa bolesnicima sa unipolarnim i bipolarnim afektivnim poremećajem [31, 32] ukazujući da je neuropsihološki poremećaj u shizofrenijii stabilniji tokom vremena i dovodi do izraženijeg onesposobljavanja nego što je to slučaj kod bipolarnog afektivnog poremećaja kod kog su evidentirani deficiti epizodični i značajno manje onesposobljavajući [33].

Uprkos postojanju značajnih metodoloških problema koji prate utvrđivanje fokalnih deficita, ipak je moguće utvrditi kognitivnu geografiju mozga i to zahvaljujući kliničkoj interpretaciji podataka studija. Rezultati većine neuropsiholoških studija shizofrenije ukazuju na postojanje jednog zajedničkog profila kognitivnog poremećaja, koji obuhvata poremećaje pažnje, pamćenja i egzekutivnih funkcija, jasno ga izdvajajući od difuznog poremećaja. Međutim, istraživanja su pokazala da uočeni deficiti pokazuju varijabilnost u stepenu izraženosti, prisutnom obrascu poremećaja, kao i na postojanje grupe bolesnika (čak i do 25 procenata) kod kojih neuropsihološki deficiti nisu uočeni [34]. Heterogenost prisustva profila i izraženosti neuropsihološkog deficita dovela je do kontradiktornih stavova u odnosu na postojanje podgrupa bolesnika sa ili bez neuropsihološke disfunkcije, opisujući kategoriju "neuropsihološki normalnih" shizofrenih bolesnika [35, 36] i predlažući kognitivne podtipove na osnovu neuropsihološke klaster analize [37].

Neuropsihološka procena kognitivnih deficita u shizofreniji

Neuropsihološko ispitivanje se sastoji od skrining tehnika i posebnih testova koji ispituju pojedinačno sve neuropsihološke funkcije standardizovanim instrumentima koji imaju odgovarajuće norme za datu populaciju (prema polu, obrazovanju i godinama) kao i kvalitativnim zapažanjima [38, 39].

Pažnja

Poremećaj pažnje jedan je od najčešće uočenih deficita u shizofreniji, ali do danas nisu potvrđeni specifični regioni mozga koji bi bili uključeni u ove procese. Neke studije sugerišu pozitivnu povezanost pažnje (*vigilance*) sa volumenom kaudatusa i putamena [40]. Neuropsihološka istraživanja ukazuju da pažnja nije jedinstven konstrukt i da verovatno uključuje nekoliko komponenti koji su pod kontrolom različitih regiona mozga. Literatura koja ukazuje na poremećaje pažnje je obimna, ali je kognitivno-anatomska interpretacija tih rezultata veoma nejasna. Bolesnici sa shizofrenijom imaju

oštećenje procesa pažnje, koji može da odražava srž poremećaja [41] i obezbedi pouzdane endofenotipove za genetsku vulnerabilnost [42]. Smatra se da je poremećaj pažnje u smislu hiperaktivnosti najčešći premorbidni poremećaj koji se može uočiti kod osoba kod kojih će se razviti klinička slika shizofrenije [2].

Pamćenje

Pamćenje je složen sistem prikupljanja, skladištenja i prisećanja informacija [43]. Osobe sa shizofrenijom ne koriste efikasne strategije, kao što je semantičko grupisanje, čak ni kada im je rečeno da je u pitanju klaster informacija. Međutim, tendencija ka intaktnoj rekogniciji [44] sugeriše postojanje značajnijeg oštećenja u eksplicitnoj u poređenju sa implicitnom memorijom [45]. Neuropsihološkom procenom pokazano je da kod shizofrenih bolesnika postoji usporeno učenje, relativno normalna ili blago povećana brzina zaboravljanja i nedostatak prajming efekta, što ukazuje na teškoće u konsolidovanju materijala u depoima dugotrajnog pamćenja [46].

Pokazano je da bolesnici sa shizofrenijom imaju očuvano implicitno pamćenje na zadacima koji uključuju identifikaciju materijala bez percepcije i produkciju semantičkih kategorija, dok je eksplicitno pamćenje poremećeno [45, 46].

Deficit u vizuelnom delu memorije je u nekim studijama okarakterisan kao nasledni, s obzirom da su slične promene nađene i kod neobolelih rođaka. Smatra se da je poremećaj vizuelne radne memorije povezan sa promenama na hromozomu 2q, a poremećaj verbalne memorije sa promenama na hromozomu 4q [46].

Govor

Najinteresantija protivrečnost u neuropsihologiji shizofrenije je u domenu govora, s obzirom na razlike između kliničkih opservacija i formalne neuropsihološke procene. Dok konverzaciju shizofrenih bolesnika karakteriše nedostatak zamenica, nelogičnosti i disocijacije, na testovima koji procenjuju govor bolesnici sa shizofrenijom pokazuju neočekivano dobre rezultate. Rezultati pokazuju da se adekvatno koriste lingvistička pravila, a postignuća na verbalnim testovima iz Vekslerove skale inteligencije (*Wexler Adult Inteligence Scale revised* – WAIS-R), koji uključuju ekspresivni rečnik, uočavanje sličnosti i razumevanje socijalnih situacija su blizu normalnih [47]. Smatra se da skraćenje reakcionog vremena, tj. povišena semantička aktivacija može da bude kritičan faktor u stvaranju dezorganizovanog govora, tj. disocijacija. Studije primenom PET-a ukazuju na abnormalnu aktivaciju levog temporalnog lobusa i prednjeg cinguluma kod zadataka verbalne fluentnosti [48].

Međutim, novije studije su pokazale da su oštećenja govora kod shizofrenih bolesnika veća nego što se ranije mislilo. Metaanaliza koju su sproveli Dikinson i sar. [49] ukazuje na teška oštećenja u vokabularu shizofrenih bolesnika. Za razliku od ranijih tvrdnji da bolesnici sa shizofrenijom obično ne pokazuju oštećenje na testovima za afaziju [50], metaanaliza koju su sproveli Henri i Kraford [51] pokazala je umereno visoko oštećenje na Bostonskom testu imenovanja (*Boston Naming Test* – BNT) što znači da je konfrontaciono imenovanje više oštećeno. Ovakvi oprečni rezultati istraživanja i kontradiktorni stavovi otvaraju novo poglavlje u neuropsihologiji govora shizofrenih bolesnika, ukazujući na značaj preciznijeg utvrđivanja neuroanatomske i neurofiziološke osnove ovih poremećaja u cilju boljeg razumevanja klinički opserviranog poremećaja govora kod shizofrenih bolesnika.

Vizuelno opažanje

Dva različita kognitivna sistema imaju ulogu u vizuelnoj obradi informacija. Sistem lociranja objekta (okcipitoparijetalni region) određuje orijentaciju prema objektima u prostoru i odnose među njima i daje odgovor na pitanje gde se nešto nalazi. Sistem prepoznavanja objekta (okcipitotemporalni region) govori o identitetu objekta, baziran na kritičnoj grupi perceptivnih obeležja objekta i daje odgovor na pitanje „šta je to nešto". Postignuća osoba sa shizofrenijom na testovima lociranja objekta koji obuhvataju prostorne analize i testovima prepoznavanja objekta, bila su adekvatna, što je doprinelo stavu da je vizuelna obrada informacija u shizofrenijii intaktna a funkcionalnost posteriornih režnjeva očuvana [52]. Međutim, studija koju su sproveli Dickinson i sar. [49] ukazuje na postojanje ozbiljnijih deficita u vizuospacijalnom domenu nego što se ranije mislilo. Neke studije ukazuju na redukciju volumena bilateralnog okcipitalnog režnja i sugerišu da to može biti neurobiološki supstrat za neke od deficita uočenih u ranom vizuelnom procesiranju kod shizofrenih bolesnika [53].

Egzekutivne funkcije

Sa stanovišta neuropsihologije, uočeni su poremećaji koji su u osnovi izvršni (egzekutivni), odnosno uključuju upotrebu informacije, a ne osnovnu obradu informacije [43]. Neuropsihološka procena pokazuje da bolesnici sa shizofrenijom često nisu uspešni kada je potrebno da održe voljnu kontrolu nad obradom informacija. Oni imaju teškoće sa formulisanjem planova, sa njihovim započinjanjem, ne koriste greške da bi se ispravili (neefikasna povratna sprega), zaboravljaju šta su prethodno započeli, ne koriste prethodno znanje za nove kombinacije, imaju teškoće sa rešavanjem zadataka čija rešenja nisu odmah vidljiva.

Smatra se da bolesnici sa shizofrenijom nisu u mogućnosti da zadrže informaciju nakon kraćeg odlaganja, u okviru kojeg su imali interferirajući zadatak, sugerišući teškoće u zadržavanju i transformaciji informacije u toku kratkih odlaganja, čak i nakon davanja instrukcija što delimično rasvetljava komponente uočenih egzekutivnih disfunkcija [46]. Pretpostavlja se da disfunkcionalnost ovog sistema ima za posledicu dva naizgled kontradiktorna ponašanja: rasejanost, pošto su svi stimulusi iz spoljne sredine podjednako značajni i, stoga, dolazi do grešaka, i perseveraciju, kada ne koriste nove strategije nakon odgovora koji su jednom bili tačni, ali u aktuelnoj situaciji više nisu [54].

Motorne funkcije

Poznato je da poremećaji motorike predstavljaju sastavni deo kliničke slike shizofrenije. Bolesnici sa shizofrenijom su spori u započinjanju pokreta, što se pokazuje kroz produženo reakciono vreme, ponavljani pokreti se obavljaju sporo i ta usporenost se povećava sa kompleksnošću motor-

nog akta, kompleksni oblici motornih aktivnosti su, takođe, poremećeni. Priprema obrade informacije pre započinjanja motornog akta kod shizofrenih bolesnika se odvija uz odlaganje. Postoje teškoće sa kontrolom sopstvenih akcija, tako da često nisu u mogućnosti da isprave sopstvene greške [55]. Mnogi stavovi koji su utvrđeni za složenije kognitivne funkcije u shizofrenijii, mogu se odnositi i na motorne funkcije [56].

Uticaj antipsihotika na kognitivnu disfunkciju

Mogućnost antipsihotika u poboljšanju i redukciji kognitivnih poremećaja predstavlja novi izazov moderne psihofarmakologije [57]. Metaanaliza podataka iz 34 studije objavljene između 1957. i 2002. godine pokazala je da konvencionalni antipsihotici imaju blage pozitivne efekte na postignuća u neuropsihološkim zadacima [57]. Značajni efekti se odnose na pažnju, automatsku obradu, jezik, perceptivnu obradu, sa negativnim efektima na motorne funkcije (u vezi su sa ekstrapiramidnim neželjenim efektima – EPS) i pamćenje (odnose se na antiholinergička svojstva ili korišćenje antiholinergika sa ciljem redukcije EPS). Konvencionalni antipsihotici indukuju promene kognitivnih funkcija, sa pretpostavkom da je korelacija obrnuto dozno zavisna [58]. Kognitivni deficiti se pojavljuju čak i kada se lečenje sprovodi nižim dozama [59].

Poređenja postignuća bolesnika lečenih konvencionalnim i atipičnim antipsihoticima pokazala su nedosledan nalaz kroz studije i ukazuju da značajna pitanja ostaju bez odgovora [57]. Rezultati studije CATIE pokazuju da su konvencionalni antipsihotici, kao perfenazin, imali najveći uticaj na kogniciju posle 18 meseci lečenja, ali da je stepen poboljšanja neznatan [60]. Pozitivan efekat perfenazina u odnosu na antipsihotike druge generacije (olanzapin, kvetiapin, risperidon, ziprasidon) na kognitivno poboljšanje dovodi se u pitanje s obzirom da je učinak antipsihotika druge generacije u većini slučajeva kompromitovan prethodnom terapijom [61]. Nekoliko metaanaliza pokazalo je da su noviji antipsihotici efikasniji od konvencionalnih u efektima na kognitivno poboljšanje, ali ostaje nejasno da li su pretpostavljena kognitivna poboljšanja direktni efekti farmakoterapije ili pak indirektni, posredovani smanjenjem EPS-a pomoćnom antiholinergičkom terapijom [61, 62]. Rezultati studije Dejvidsona i sar. [63] pokazali su da nema značajne razlike u poboljšanju kognitivnog funkcionisanja između bolesnika koji su lečeni konvencionalnim (haloperidol) i antipsihoticima druge generacije (amisulprid, olanzapin, kvetiapin i ziprasidon).

U svetlu dosadašnjih saznanja u vezi sa kognitivnim poboljšanjem tokom lečenja antipsihoticima kako prve, tako i druge generacije može se zaključiti da je kognitivno poboljšanje neznatno i shvata se primarno kao efekat povlačenja psihotičnih simptoma i posledično efikasnijeg korišćenja kognitivnih resursa i u vezi je sa unapređenjem funkcionalnog ishoda. Farmakoterapija kognitivnih deficita u shizofrenijii ostaje terapijski izazov. Kroz istraživačke projekte koji se bave istraživanjima i merenjima efekata lekova na poboljšanje kognicije u shizofreniji čine se napori da se farmakološki ciljano deluje na kognitivne deficite kao ključnu komponentu bolesti u efikasnom individualnom, socijalnom i profesionalnom funkcionisanju [64].

Studije muskarinskih receptora ističu značaj promene ovih receptora u shizofreniji i mogućnost terapijske modulacije transmisije u pravcu poboljšanja kognitivnog funkcionisanja. Složenost muskarinskog sistema ukazuje na jedinstvenu distribuciju svih podtipova muskarinskih receptora (M1 do M5). Receptori M1 su najviše zastupljeni u rostralnim oblastima mozga: neokorteksu, hipokampusu (posebno CA1 region), strijatumu, nukleusu akumbensu; M2 receptori se nalaze u bazalnim moždanim regionima, talamusu i moždanom stablu, a M3 receptori su lokalizovani u hipokampusu, talamusu, neokorteksu; M4 receptori se nalaze pored M1, u strijatumu, hipokampusu i neokorteksu; M5 receptori su po gustini najniži od svih, a lokalizovani su sa dopaminergičnim neuronima u supstancija nigra i hipokampusu [65]. Postoji nekoliko obdukcionih studija gustine muskarinskih receptora u ljudskom mozgu. Jedna studija je otkrila 28% smanjenje M1 receptora u gornjem frontalnom girusu kod shizofrenih bolesnika u poređenju sa zdravim osobama i pokazala da M1 podtip muskarinskih receptora može biti podtip najčešće promenjen u shizofrenijii [66]. Studije koje prate dejstva lekova na muskarinske receptore i korelaciju sa poboljšanjem kognitivnih performansi sugerišu da bi definisanje selektivnih muskarinskih agonista M1 receptora značajno popravilo kognitivno funkcionisanje shizofrenih bolesnika, ali danas takav profil leka nije poznat, a ispitivanja lekova kao što su AF-150(s), WAY-132983, Lu25-109, 1,2,5-tiadiazolni analozi, ksanomelin i NDMC su u toku [67].

Rehabilitacija (nefarmakološke intervencije za poboljšanje kognitivnih deficita u shizofreniji)

Kognitivni trening koji koristi kompjuterska vežbanja, individualne instrukcije ili grupne tehnike je nefarmakološki pristup za poboljšanje kognitivnih oštećenja u shizofreniji utičući na poboljšanje održavanja pažnje, jezičko procesiranje, egzekutivne funkcije i rekogniciju, radnu memoriju, brzinu procesiranja i rešavanje socijalnih problema [68–73]. Program neuralnog poboljšanja (*Neural Enhancement Therapy* –NET) je program celovite kognitivne remedijacije koji se sastoji od kompjuterskog kognitivnog treninga. Praćenjem grupe bolesnika kod koje je primenjen NET uočeno je da su posle jednogodišnjeg tretmana značajno poboljšali svoje rezultate na merenjima egzekutivnih funkcija i radne memorije [74].

Poboljšanje kognitivnih deficita koji utiču na kvalitet života, predstavljaju cilj terapije kognitivne remedijacije (*Cognitive Remediation Therapy* – CRT). Ova terapija predstavlja pokušaj da se poboljšaju kognitivni deficiti davanjem instrukcija za učenje strategija obrade informacija kroz mentalne vežbe. Jedna od studija je imala za cilj vrednovanje efikasnosti ublažavanja kognitivnih deficita u poređenju sa uobičajenim tretmanima. Glavni ishod je bio meren kroz kogniciju (memorija, kognitivna fleksibilnost i planiranje) i sekundarne ishode (simptomi, socijalni kontakti, samopoštovanje). Primena CRT je proizvela značajna poboljšanja u kognitivnoj fleksibilnosti mereno Viskonsin testom sortiranja karata (*Wisconsin Card Sorting Test* – WCST) [75]. U smislu poboljšanja neurokognitivnog i sociokognitivnog funkcionisanja procenjivan je i efekat terapije kognitivnog poboljšanja

(*Cognitive Enhancement Therapy* – CET). Ova terapija je multidimenzionalni pristup remedijaciji socijalno kognitivnih i neurokognitivnih deficita. Fokus primene CET je povećanje mentalne izdržljivosti i procesiranja najpre aktivnih, a zatim i pasivnih informacija. Rezultati pokazuju da postoji značajno poboljšanje, naročito u domenu pažnje, koje se održavalo i godinu dana posle tretmana [76].

Zaključak

Pregled literature koja se bavi neuropsihološkim aspektima shizofrenije ukazuje na heterogenost u kognitivnom funkcionisanju među shizofrenim bolesnicima, značajnu stabilnost kognitivne disfunkcije tokom bolesti, i da je stepen izraženosti kognitivnih deficita, a ne psihopatološka fenomenologija, primarna odrednica poremećaja funkcionalnog kapaciteta i funkcionalnog statusa bolesnika. Uočeni kognitivni poremećaji u shizofreniji su primarno na nivou pripreme odgovora, izvršenja i kontrole, pa se mogu sresti u bilo kom modalitetu i na bilo kom nivou obrade informacija. Neuropsihološki profil shizofrenih bolesnika obuhvata kao najprominentnije deficite pažnje, pamćenja i egzekutivnih funkcija, sa blažim poremećajima funkcije govora i vizuoprostorne obrade informacija, što upućuje na mogućnost da kognitivni poremećaji imaju neurobiološki supstrat u specifičnim regionima mozga. Takav profil ukazuje na frontalno-medijalno temporalnu disfunkciju, uz relativnu očuvanost funkcije posteriornih regiona. Ipak, neurohemijski, neurološki ili neuropsihološki korelati ključnih simptoma shizofrenije još uvek su nepoznati. Prepoznavanje funkcionalnih efekata kognitivnih deficita ima značajnu ulogu u razvoju terapijskih smernica u smislu pomeranja fokusa farmakoterapije sa lečenja uglavnom pozitivnih psihotičnih simptoma, na lečenje kognitivnih deficita i negativnih simptoma, koji značajnije determinišu kako samu bolest, tako i stepen funkcionalnog onesposobljavanja. Otkrivanje specifičnih markera shizofrenije kojima teže savremena istraživanja će dati bar neka od rešenja za mnoge dijagnostičke i terapijske probleme vezane za ovaj poremećaj.

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Primljen: 14. VII 2010.
Revidiran: 1. IX 2010.
Prihvaćen 6. IX 2010.

CASE REPORT

UDC: 616.132.2-005.6-08
DOI: 10.2298/VSP1206517S

Life-saving percutaneous coronary interventions on the unprotected left main coronary artery in patients with acute coronary syndrome in the catheterization laboratory without cardiosurgical back-up

Spasonosne perkutane koronarne intervencije na nezaštićenom glavnom stablu leve koronarne arterije kod bolesnika sa akutnim koronarnim sindromom u kateterizacionoj laboratoriji bez podrške kardiohirurgije

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Abstract

Introduction. The optimal revascularization strategy for unprotected left main coronary disease (ULMCD) is the subject of ongoing debate and patients with ULMCD still represent a challenge for interventionalist, especially in the setting of an acute coronary syndome (ACS). **Case report.** We presented two cases of percutaneous treatment of ULMCD in the settings of ACS (ST Segment Myocardial Infarction and Non ST Segment Myocardial Infarction – STEMI and NSTEMI) in a catheterization laboratory without back-up of cardiosurgical department. Both patients were hemodynamically unstable with clinical signs of cardiogenic shock. Coronary angiography revealed left main thromobosis and using intra-aortic balloon pump as hemodynamic support primary angioplasty procedures were performed. Immediately after the procedures the patients hemodynamically improved and remained stable till discharge from hospital. **Conclusion.** Percutaneous coronary intervention (PCI) has become the most common strategy of revascularization in ACS patients with ULMCD and is generally preferred in patients with multiple comorbidities and/or in very unstable patients. In cases with no cardiosurgical departments PCI is an inevitable, bail-out, life saving procedure.

Key words:
coronary disease; myocardial infarction; angioplasty, balloon; stents.

Apstrakt

Uvod. Optimalna revaskularizaciona strategija u slučaju suženja glavnog stabla leve koronarne arterije, nezaštićenog graftom (ULMCD), predstavlja veliki izazov za interventne kardiologe, a naročito u situaciji akutnog koronarnog sindroma. (AKS). **Prikaz bolesnika.** Prikazana su dva slučaja perkutanog lečenja tromboze glavnog stabla leve koronarne arterije u akutnom koronarnom sindromu sa i bez elevacije ST segmenta (STEMI i NSTEMI) u centru sa kateterizacionom salom, ali bez kardiohirurške podrške. Oba bolesnika bila su hemodinamski nestabilna, sa kliničkim znacima kardiogenog šoka. Nakon koronarne angiografije kada je registrovana tromboza glavnog stabla, plasirana je intraaortna balon pumpa i urađena primarna angioplastika. Odmah posle procedure registrovano je subjektivno i objektivno poboljšanje hemodinamskog statusa bolesnika i oni su otpušteni sa klinike u stabilnom stanju, bez komplikacija. **Zaključak.** Bolesnici sa AKS usled tromboze glavnog stabla leve koronarne arterije sve češće se podvrgavaju perkutanim koronarnim intervencijama, posebno u slučaju postojanja komorbiditeta ili značajne hemodinamske nestabilnosti. U centrima bez kardiohirurgije, perkutane procedure su neizbežne u cilju spašavanja života jako ugroženih bolesnika.

Ključne reči:
koronarna bolest; infarkt miokarda; angioplastika, balonska; stentovi.

Introduction

The optimal revascularization strategy for unprotected left main coronary disease (ULMCD) is the subject of ongoing debate [1]. Although it is noteworthy that 2 of the first 5 coronary balloon dilations ever performed were in the left main coronary artery, however several complications such as acute vessel closure precluded the widespread use of multivessel percutaneous transluminal coronary angioplasty (PTCA) in patients with severe coronary disease and left main involve-

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ment [2]. On the other hand, a recently performed SYNTAX study has demonstrated good results after ULMCD with similar mortality but higher revascularisation rates in percutaneous coronary interventions (PCI) patients and higher stroke rate in coronary artery bypass graft (CABG) patients [3].

Left main stenting has been carried out with increasing frequency during the last years. Nevertheles, patients with ULMCD still represent a challenge for interventionalist, especially in the setting of an acute coronary syndome (ACS) [4]. Angiographic and technical details in ULMCD stenting, such as localization of the lesion within the left main (LM), the need for intravascular ultrasound (IVUS) and the technique used to approach the bifurcation, may be extremely important in the acute and long-term results and have already been studied [5, 6]. The number of patients who need a CABG in the acute phase is limited, but CABG may be indicated after failed PCI, coronary occlusion not amenable for PCI, the presence of refractory symptoms after PCI, cardiogenic shock or mechanical complications such as ventricular rupture, acute mitral regurgitation or ventricular septal defect [7].

Case report

We presented two cases of percutaneous treatment of ULMCD in the settings of ACS (ST Segment Myocardial Infarction and Non ST Segment Myocardial Infarction – STEMI and NSTEMI) in a catheterization laboratory without back-up of cardiosurgical department.

The first patient, a hemodynamically unstable 72-year-old male patient, was admitted to our hospital with an acute coronary syndrome with ST segment elevation in precordial leads. He suffered from arterial hypertension and dyslipidemia, and was a former smoker. Because of intensive chest pain lasting three hours, the patients had called emergency medical service and was transfered to the center with catheterization laboratory. He was treated with unfractionated heparin (UFH, 70 IU /kg), aspirin 300 mg, clopidogrel 600 mg, oxygen, dopamin and analgetics in the emergency means of transportation. At admission the patient was hypotensive with systolic blood pressure of 95 mmHg and with pulmonary basal rales. Emergent coronary angiography revealed spontaneous coronary dissection and thrombosis of the left main with TIMI 1 flow (penetration of the contast material without perfusion) (Figure 1a). The right coronary artery was without significant stenosis (Figure 1b). After implantation of an intraaortic baloon pump, a guidewire was placed in left anterior descending (LAD) artery, intermediate branch and in right circumflex artery (RCX). Predilatation was performed using Dura-Star compliant balloon 3.0 × 10 mm at 14 atm (Figure 2). A bare metal stent Tsunami Gold 4.0 × 13 was deployed into main stream and inflated at 19 atm (Figure 3), and after that another Tsunami Gold 4.0 × 13 at 14 atm to-

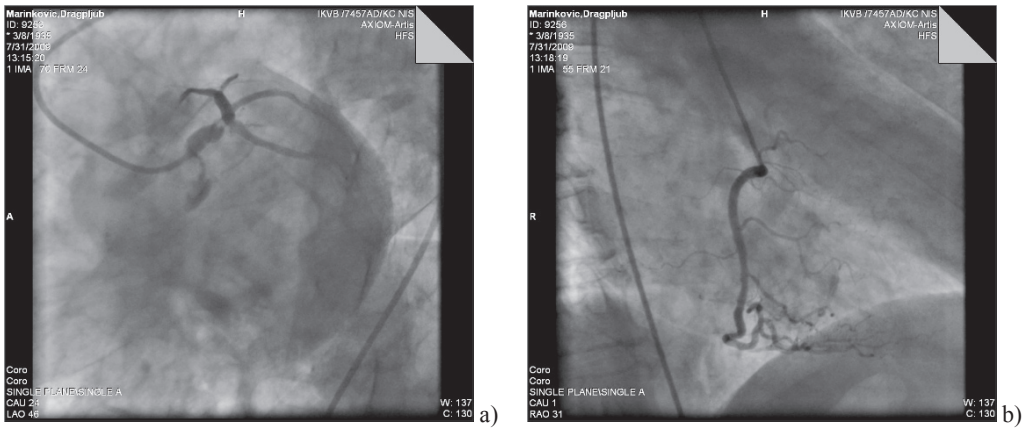

Fig. 1 – Emergent coronary angiography in ST segment myocardial infarction (STEMI)
a) spontaneous coronary dissection and thrombosis of the left main artery
b) the right coronary artery without significant stenosis

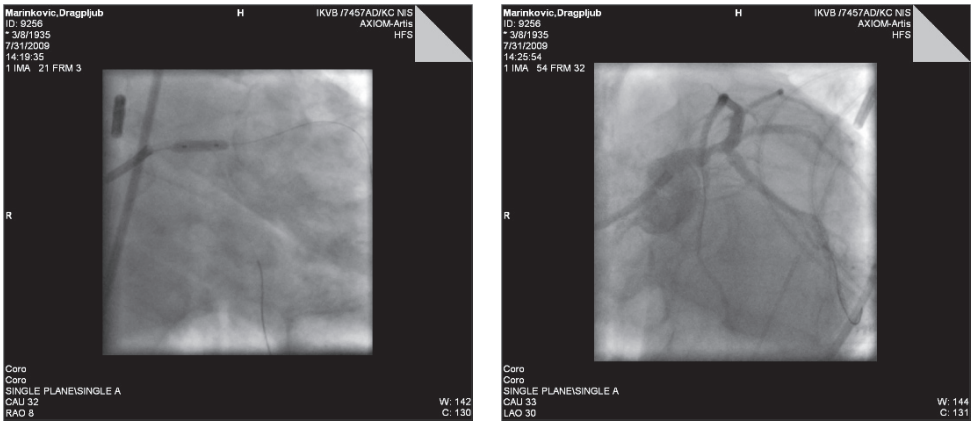

Fig. 2 – Predilatation with Dura-Star compliant balloon at 14 atm

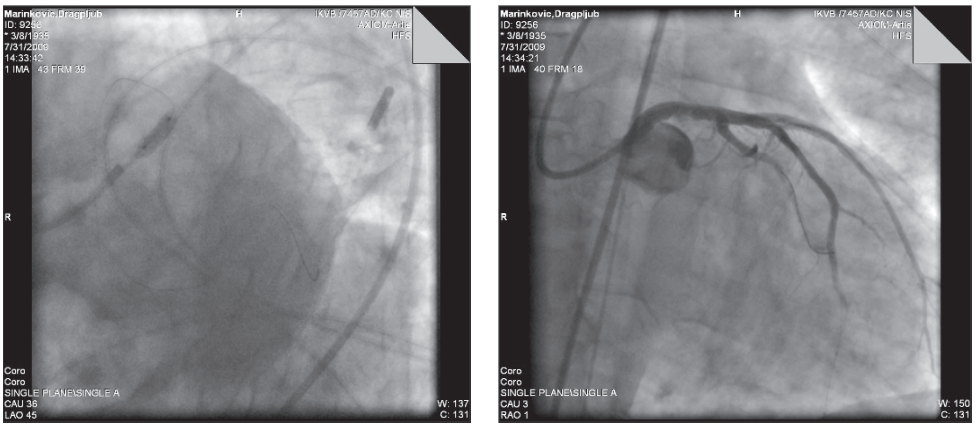

Fig. 3 – Implantation of a bare-metal stent Tsunami Gold that was inflated at 19 atm

ward LAD because of visible thrombus nearby ostium (Figure 4). Neither RCX nor right intermediate (RI) were compromised and the procedure was concluded (Figure 5). After 5 days the patient was discharged hemodinamically stable and without any complication.

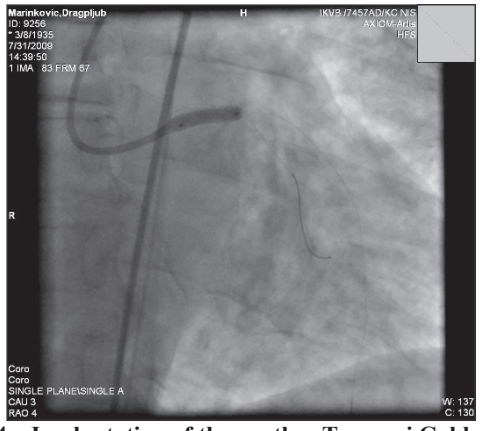

Fig. 4 – Implantation of the another Tsunami Gold metal stent toward left anterior descendent (LAD) artery

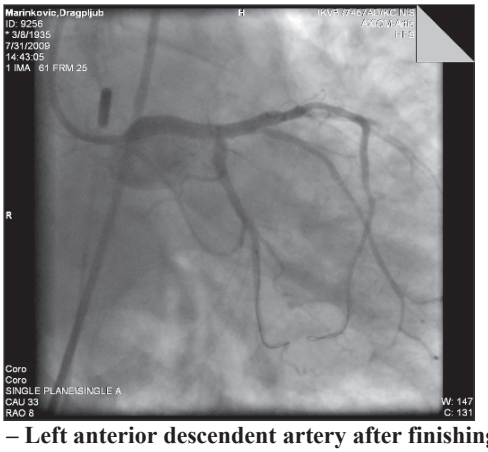

Fig. 5 – Left anterior descendent artery after finishing the procedure

The second patient was a hemodynamically unstable 78-year-old male, with no risk factor, admitted to our hospital due to chest pain at rest lasting more than 12 h before admission with concomitant ST segment depression of 4 mm. The patient was treated with aspirin 300 mg, clopidogrel 600 mg, analgetics and UFH (70 IU/kg), and underwent emergent coronary angiography. As culprit lesion ostial LM dissection with thrombus and TIMI 1 flow was found (Figure 6). The right coronary artery (RCA) was without significant stenosis. After implanting an intraortic balloon pump (IABP), a guide wire was introduced in LAD and RCX. Predilatation was performed using a Sprinter NC balloon at 14 atm (Figure 7). Thereafter a 4.0 × 13 baremetal stent Tsunami Gold was inflated at 18 atm (Figure 8a), and proximal part was postdilated with the stent balloon at 20 atm (Figure 8 b and c). During hospitalization of 5 days the patient remained free of symptoms and adverse symptoms.

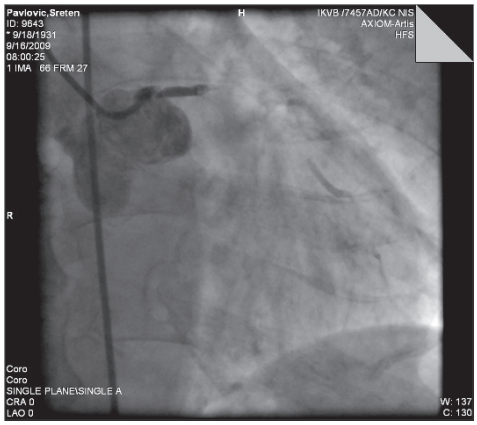

Fig. 6 – Ostial left main (LM) culprit lesion, dissection and thrombosis on coronary angiography

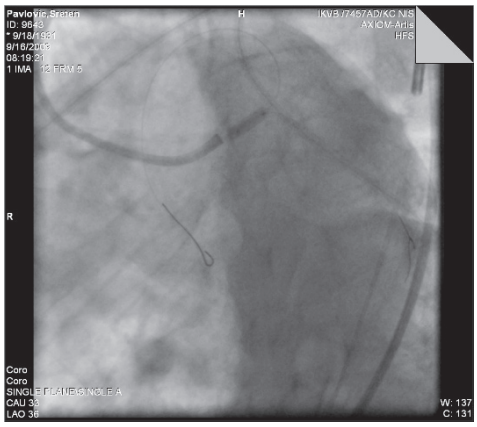

Fig. 7 – Predilatation with a sprinter NC balloon

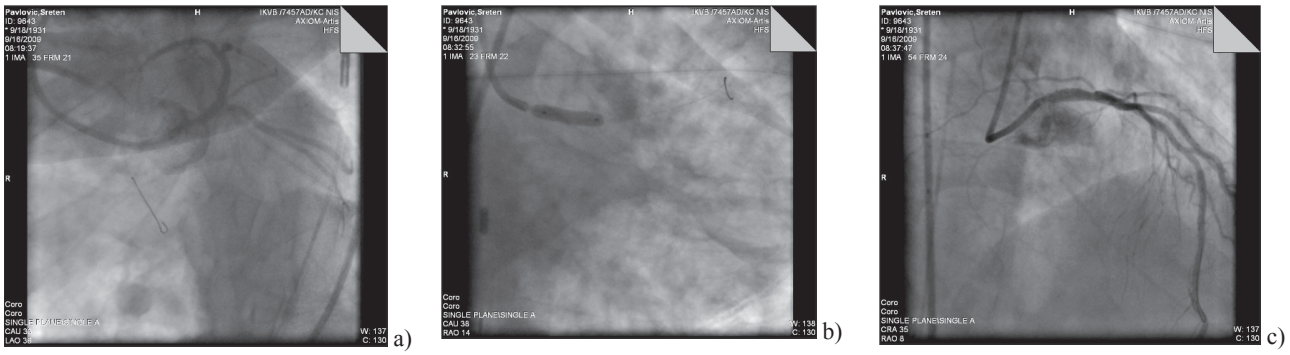

Fig. 8 – Implantation of a bare-metal stent Tsunami Gold
a – Stent baloon inflated at 18 atm
b and c – proximal part postdilated with stent baloon at 20 atm

Discussion

Unprotected left main coronary artery disease in patients with an ACS is a serious situation with high in-hospital mortality, especially in those presenting with STEMI and/or hemodynamic or arrhythmic instability Montalescot et al. [1] reported the results of patients with ACS and left main disease included in the Global Registry of Acute Coronary Events (GRACE) between 2000 and 2007 and a trend towards more PCI and less CABG was observed. Overall in-hospital mortality was 7.7% but reached 11% in patients who presented with STEMI or left bundle brunch block (LBBB) and was as high as 34% in patients with cardiogenic shock or cardiac arrest [4]. A complete occlusion of the left main is usually associated with cardiogenic shock and therefore represents a very high risk situation requiring immediate life support strategies and urgent revascularization often in conjuction with the use of left ventricular assist devices [8, 9].

In PRECOMBAT (Premier of Randomized Comparison of Bypass Surgery versus Angioplasty Using Sirolimus-Eluting Stent in Patients with Left Main Coronary Artery Disease) prospective, randomized trial involving patients with unprotected left main coronary artery stenosis, PCI with sirolimus-eluting stents was noninferior to CABG with respect to the primary composite end point of major adverse cardiac or cerebrovascular events at 1 year. In addition, the two groups had similar rates of individual components of death, myocardial infarction, and stroke. However, the rate of ischemia-driven target-vessel revascularization at 2 years was lower in the CABG group than in the PCI group [10].

In a recently on-line published metaanalysis of randomized patients with unprotected left main stenosis, the risk of death and myocardial infarction was comparable between CABG and PCI. However, patients undergoing CABG had a higher risk of stroke, whereas patients undergoing PCI were at a higher risk for repeated revascularization [11].

In Serbia as developing country, many unresolved clinical needs remain for revascularization strategies. Cardiosurgical departments in most cases are far away and are not able to provide sufficient capacity for surgical back up. Therefore, interventional strategies need to be implemented, even in scenarios in which clear cut recommendations are given for surgical treatment. The presented cases demonstrate that immediate PCI of an unprotected LMA stenosis together with implantation of a left ventricle (LV) assist system (IABP) can be a life saving procedures and should be performed without any delay if surgery is not immediately available.

Conclusion

Unprotected left main coronary artery desase in patients with an ACS is rare but serious situation with high in-hospital mortality, especially in those presenting with STEMI and/or hemodynamic or arrhythmic instability. Percutaneous coronary intervention has become the most common strategy of revascularization in ACS patients with ULMCD and is generally preferred in patients with multiple comorbidities and/or in very unstable patients. In cases with no cardiosurgical departments PCI is an inevitable, bail-out, life saving procedure.

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Received on July 2, 2010.
Revised on April 17, 2012.
Accepted on April 17, 2012.

P R I K A Z S L U Č A J A

UDC: 616.98:579.87] 616,34
DOI: 10.2298/VSP1206522P

Viplova bolest

Whipple's disease: a case report

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Apstrakt

Uvod. Viplova bolest (Morbus Whipple) je hronična, multisistemska, infektivna bolest koju izaziva bakterija *Tropheryma whipplei*. Najčešće se javlja kod muškaraca bele rase, srednjih godina. To je primarno gastrointestinalno oboljenje koje se manifestuje malapsorpcijskim sindromom, a dijagnoza se postavlja endoskopijom i intestinalnim biopsijama. Ekstraintestinalne manifestacije bolesti nisu retke. **Prikaz bolesnika.** Prikazan je bolesnik star 48 godina, koji je primljen zbog proliva, smanjenja telesne mase i slabosti u ekstremitetima. Fizikalnim nalazom je utvrđena difuzna hiperpigmentacija, pleuralna efuzija i edem potkolenica. Laboratorijski su otkriveni anemija, zapaljenski sindrom i znaci malapsorpcije. Dijagnoza je potvrđena biopsijama tankog creva. Bolesnik je lečen antibioticima i simptomatskom terapijom. Nakon 9 meseci, bolesnik je bio bez tegoba, sa urednim kliničkim i laboratorijskim nalazom. **Zaključak.** Viplova bolest je retka. Za dijagnozu je najbitnije da se posumnja na to oboljenje čije manifestacije su: malapsorpcija, artritisi, povišena temperatura, neurološka simptomatologija. Pravovremeno postavljanje dijagnoze i primena adekvatne terapije sprečavaju progresiju bolesti i fatalan ishod.

Ključne reči:
viplova bolest; dijagnoza; malapsorpcija, sindromi; anemija; biopsija; crevo, tanko; lečenje, ishod.

Abstract

Introduction. Whipple's disease is a chronic, multisystem, infectious disease caused by *Tropheryma whipplei*. It most commonly affects Caucasian males, middle-aged. Morbus Whipple is primarily gastrointestinal disease, manifested as malabsorption syndrome, and diagnosed by endoscopy and intestinal biopsy. Extraintestinal manifestations are not rare. **Case report**. A 48-year-old male was admitted due to diarrhea, weight loss and weakness in the extremities. Physical examination findings pointed out diffuse hyperpigmentation, pleural effusion and leg edema. Anemia, inflammatory syndrome and malabsorption signs were discovered through laboratory tests. The diagnosis was confirmed by intestinal biopsy. The patient was treated with antibiotic and symptomatic therapy. After 9 months, the patient had no symptoms, and clinical and laboratory findings were regular. **Conclusion.** Whipple's disease is a rare disease. A high degree of clinical suspicion for the disease (malabsorption, arthritis, fever, neurological symptoms) is the most important for diagnosis. Timely diagnosis and appropriate therapy prevent the disease progression and fatal outcome.

Key words:
whiplle disease; diagnosis; malabsorption syndromes; anemia; biopsy; intestine, small; treatment outcome.

Uvod

Viplova bolest (VB) je retka, hronična, infektivna bolest koja može zahvatiti bilo koji organ ili sistem organa, a izazivač je intracelularni, gram pozitivni bacil *Tropheryma whipplei*. Precizni podaci o prevalenciji i incidenciji te bolesti ne postoje. Podaci dobijeni prilikom autopsije ukazuju na to da je učestalost bolesti niža od 0,1% [1]. Bolest se može javiti u bilo kom životnom dobu, ali se najčešće javlja između 40. i 60. godine života, i to češće kod muškaraca bele rase [2]. Do sada je u literaturi opisano 1 000–1 500 bolesnika sa tim oboljenjem.

Prikaz bolesnika

Muškarac star 48 godina, primljen je u Kliniku za gastroenterologiju Kliničkog centra Srbije u Beogradu. Tegobe su počele pet meseci ranije pojavom kašastih stolica, 1–2 dnevno, a potom i prolivastih stolica do 10 dnevno. Pored toga, naveo je gubitak telesne mase 20 kg za pet meseci, mučninu, povraćanje, oticanje nogu i izrazitu slabost ruku i nogu. Hospitalizovan je u regionalnom zdravstvenom centru, ali je zbog pogoršanja opšteg stanja upućen u našu ustanovu. Alergičan je na penicilin i acetilsalicilnu kiselinu; porodična anamneza mu je bila pozitivna na ulkusnu bolest i hiperten-

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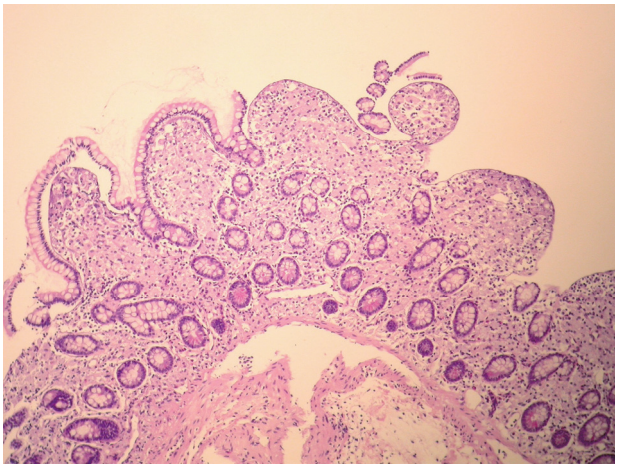
ziju; bio je višegodišnji pušač, ali nije pušio prethodnih pet meseci. Naveo je da živi u komfornim uslovima i da koristi vodovodsku vodu. Na prijemu bolesnik je imao asteničnu konstituciju, bio je pothranjen [telesna visina 182 cm, telesna masa 56 kg, indeks telesne mase (BMI) 16,9 kg/m^2], difuzno hiperpigmentisan, afebrilan, dehidriran, hipotenzivan (krvni pritisak 90/50 mmHg). Nije bilo znakova hemoragijskog sindroma ni periferne limfadenopatije. Na plućima je imao oslabljen disajni šum levo, bazalno. Izuzev življe peristaltike, abdomen je bio urednog nalaza. Prisutni su bili obostrani testasti edemi stopala i potkolenica. U laboratorijskim analizama otkriveni su normohromna normocitna anemija, pozitivan zapaljenski sindrom, znaci malapsorpcije i lako povišenje aspartat aminotransferaze (AST). Patološke vrednosti krvne slike i biohemijskih analiza na prijemu prikazani su u tabeli 1.

Tabela 1
Hematološki i biohemijski parametri kod bolesnika sa Viplovom bolesti na prijemu

Parametri	Nađene vrednosti	Referentne vrednosti
Hematološki		
eritrociti (x10^{12}/L)	2,7	4,3–5,7
hematokrit (L/L)	0,22	0,41–0,53
hemoglobin (g/L)	75,8	138–175
MCV (fL)	83,1	83–97
leukociti (x10^9/L)	10,4	3,4–9,7
neutrofili (x10^9/L)	7,49	2,1–6,5
Biohemijski		
proteini (g/L)	45	63–82
albumin (g/L)	18	39–50
holesterol (mmol/L)	1,52	0–5,2
gvožđe (µmol/L)	3,5	8,8–32,4
TIBC (µmol/L)	16,3	44,8–80,6
kalijum (mmol/L)	3,3	3,5–5,1
kalcijum (mmol/L)	1,96	2,10–2,55
AST (U/L)	57	14–50
sedimentacija eritrocita (mm/h)	40	2–10
fibrinogen (g/L)	5,74	2–4
protrombinsko vreme (%)	69	75–120

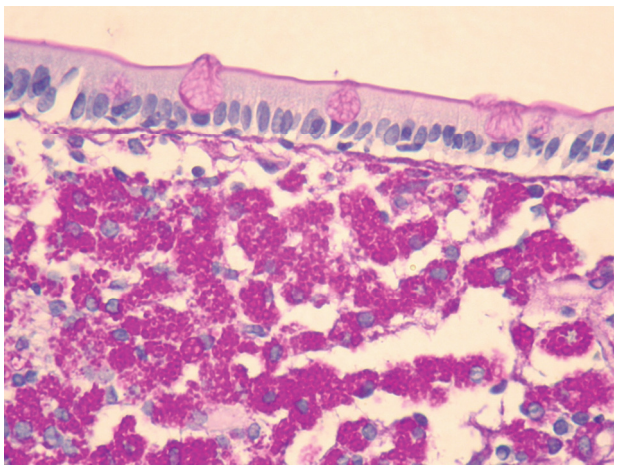
AST – aspartat aminotransferaza; MCV – prosečna veličina eritrocita
TIBC – *total iron binding capacity*

Površinski (*surface*) antigen virusa hepatitisa B (HBsAg), anti-hepatitis virus C (HCV), anti-virus humane imunodeficijencije (HIV) i imunološke analize bile su negativne. Tireostimulišući hormon (TSH), slobodni tiroksin (FT4) i kortizol bili su u referentnom opsegu. Stolica je bila pozitivna na svarena mišićna vlakna, skrob i masti; mikrobiološki, u stolici detektovan je umeren broj *Candida spp*. Nalaz urina ukazivao je na urinarnu infekciju prouzrokovanu *Acinetobacter spp*. Na radiografiji srca i pluća viđena je pleuralna efuzija levo. Ultrazvučni pregled abdomena ukazao je na manju količinu slobodne tečnosti u maloj karlici uz uredan nalaz na parenhimskim organima. Na ezofagogastroduodenoskopiji sluznica duodenuma bila je edematozna, sa erozijama, nareckanih ivica, mestimično "slaninastog" izgleda. Pri kolonoskopiji sa terminalnom ileoskopijom viđena je istanjena sluznica terminalnog ileuma sa erozijama, proširenih limfnih sudova. Patohistološkom analizom uzoraka tankog creva dobijeni nalaz bio je visokosugestivan za VB. Na svim uzorcima uočena je enormna akumulacija makrofagnih ćelija visoke uniformnosti, izgleda krupnih epiteloidnih ili delom okruglastih, nepravilnih ćelija čija je citoplazma eozinofilna i granulirana (slika 1). Infiltracija je prvenstveno bila u lamini



Sl. 1 – Enterobiopsija ukazuje na infiltraciju lamine proprije penastim makrofagama (H&E, 40×)

propriji. Nakon histohemijskog bojenja dobijena je intenzivna *periodic acid-Schiff* (PAS) pozitivnost citoplazme granuliranog tipa (slika 2). S obzirom na to da je bojenje po Ziehl-



Sl. 2 – Intenzivna *periodic acid-Schiff* (PAS) pozitivnost citoplazme penastih makrofaga (PAS, 240×)

Nielsen-u bilo negativno, isključena je mogućnost prisustva acidorezistentnih bacila. Radiografija gastroduodenuma i pasaža tankog creva ukazali su na diskretno izraženije Kerkingove nabore i naglašeniji reljef sluznice tankog creva. Ehokardiografski nalaz bio je uredan. Nalaz kompjuterizovane tomografije (KT) endokranijuma bio je uredan. Nakon postavljene dijagnoze započeta je antibiotska terapija tabletama kotrimoksazola (400 + 80 mg), 3 × 1 dnevno, i streptomicinom 1g/dan, intramuskularno, u trajanju od 2 nedelje, a potom je nastavljeno sa tabletama kotrimoksazola (400 + 80 mg), 2 × 1/dan. Zbog urinarne infekcije ordiniran je tigeciklin 50 mg/12 h, intravenski, u trajanju od 10 dana. Sve vre-

me hospitalizacije sprovođena je intenzivna rehidracijska i supstitucijska terapija. Na kontrolnom pregledu, posle 6 meseci, bolesnik se dobro osećao, imao je 1–2 normalno formirane stolice dnevno, dobio je 10 kg u telesnoj masi. Nije imao otoke, ni slabost u ekstremitetima. Ultrazvučni pregled abdomena i radiografija srca i pluća bili su urednog nalaza. Laboratorijske analize bile su u referentnom opsegu, izuzev makrocitne anemije sa niskim vrednostima vitamina B12 i folata u serumu, tako da je ordinirana nadoknada vitaminom B12 i folnom kiselinom. Na kontrolnom pregledu, nakon 9 meseci, bolesnik nije imao tegobe. Laboratorijske analize bile su u granicama normale. Nastavljeno je sa primenom kotrimoksazola (400 + 80 mg), 2 × 1/dan.

Diskusija

George H. Whipple je 1907 godine prvi opisao VP i to kod muškarca starog 36 godina, koji je imao gubitak telesne mase, prolive, bolove u trbuhu, poliartritis i poliserozitis [3]. U lamini propriji tankog creva i mezenterijalnim limfnim čvorovima otkrio je infiltraciju penastim makrofagama, koje su bile ispunjene velikim brojem nepoznatih štapićastih struktura, promera oko 2 μm. Otkrivenu bolest nazvao je intestinalnom lipodistrofijom, ali je nije povezao sa bakterijskom etiologijom. Zahvaljujući elektronskoj mikroskopiji, 1960. godine utvrđeno je da ove štapićaste strukture imaju karakteristike bakterija [4, 5]. Nakon 30 godina, tehnika lančane reakcije polimeraze (PCR) omogućila je amplifikaciju 16S rRNA uzročnika iz duodenalnih lezija bolesnika sa VB [6]. Prva uspešna kultivacija bakterije *Tropheryma whipplei* objavljena je 2000. godine iz valvule bolesnika sa endokarditisom u sklopu VB [7, 8]. Teoretski, VB može zahvatiti bilo koji organski sistem, ali su patološke promene smeštene predominantno u gastrointestinalnom i centralnom nervnom sistemu. Ova bolest prolazi kroz dva klinička stadijuma: prodromalni i manifestni. Prodromalni stadijum karakteriše se nespecifičnim simptomima, povišenom temperaturom, artralgijama i artritisima. Nakon prodromalnog stadijuma, u proseku za 6 godina, bolest ulazi u manifestnu fazu [1]. Manifestna faza se karakteriše simptomima koji zavise od zahvaćenog organskog sistema [9]. Gastrointestinalni trakt je zahvaćen kod jedne trećine bolesnika [10, 11]. Dominiraju simptomi malapsorpcije (proliv, nadimanje), kao i bolovi u trbuhu [12–14]. Stolice su obilne, 5–10 dnevno, neugodnog mirisa, kašaste ili tečne. Krvarenje može biti prisutno kao okultno ili manifestno [1]. Kao posledica izražene malapsorpcije mogu se javiti različiti nutritivni deficiti. Zbog opstrukcije protoka limfe i sledstvenog gubitka albumina, mogu se razviti ascites i periferni edemi [14]. Kod opisanog bolesnika bili su prisutni tipični gastrointestinalni simptomi VB, bez manifestnog prodromalnog stadijuma. Ascites i pleuralna efuzija objašnjeni su hipoalbuminemijom, i u potpunosti su se povukli na supstituciju albuminom. Najčešća ekstraintestinalna lokalizacija VB je nervni sistem. On je zahvaćen kod 6–63% obolelih [15, 16] sa poremećajem stanja svesti, kognitivnim poremećajima, paralizom, konvulzijama i dr. [14]. Opisani bolesnik imao je parezu gornjih i donjih ekstremiteta, što je shvaćeno kao posledica elektrolitnog disbalansa (hipokalijemija, hipokalcemija), jer su se nakon supstitucije u potpunosti povukle, dok sprovedeno neurološko ispitivanje nije ukazalo na zahvaćenost nervnog sistema. U fizikalnom nalazu obolelih mogu se videti limfadenopatija (45–60%), difuzna hiperpigmentacija (40%), znaci malnutricije sa gubitkom mišićnog i masnog tkiva, što može ići i do kaheksije [12]. Pored toga, prisutni su i znaci različitih nutritivnih deficita. Prilikom prijema, opisani bolesnik bio je pothranjen i dehidriran. S obzirom na difuznu hiperpigmentaciju kože, koja se javlja i u Adisonovoj bolesti, određen je i nivo kortizola koji je bio u referentnom opsegu. U laboratorijskim analizama mogu biti prisutni leukocitoza, leukopenija, eozinofilija, trombocitoza ili trombocitopenija, povišeni reaktanti akutne faze i ubrzana sedimentacija eritrocita. Prisutni su i biohemijski parametri karakteristični za malapsorpciju (anemija, hipoproteinemija, hipoalbuminemija, hipoholesterolemija hipokalijemija, hipokalcemija). Anemija može nastati usled nedostatka gvožđa, vitamina B12 ili folata, ali se, takođe, može javiti i anemija tipa hronične bolesti. U opisanom prikazu, laboratorijski su otkriveni pozitivan zapaljenski sindrom i malapsorpcija. Inicijalno, anemija je bila normocitna, normohromna, tipa anemije hronične bolesti, ali kasnije je došlo do pojave makrocitne anemije, kao posledice malapsorpcije folne kiseline i vitamina B12. Supstitucijskom terapijom anemija je izlečena. Najvažnija dijagnostička procedura je gornja endoskopija, sa biopsijama tankog creva. Makroskopski se vide promene na intestinalnoj mukozi, naročito u postbulbarnom regionu, u vidu zadebljanja i edema nabora sa eritemom i žućkastobeličastim plakovima i erozijama [12]. Dijagnoza se najčešće postavlja pregledom uzorka tkiva dobijenog duodenalnim biopsijama. Patohistološki se mogu videti promene u rasponu od gotovo normalnih resica do izražene atrofije sluznice tankog creva. Prisutna je infiltracija lamine proprije penastim makrofagama. Prilikom prebojavanja PAS tehnikom, vide se crveno prebojene inkluzije sa makrofagama u lamini propriji. S obzirom na to da se PAS pozitivne inkluzije sreću i kod infekcije sa *Mycobacterium avium-intracellulare*, diferencijalno dijagnostički je veoma bitno i bojenje po Ziehl-Nielsenu. Ono je kod VB negativno. Kod našeg bolesnika sumnja na VB postavljena je na osnovu endoskopije, a verifikovana patohistološki. Laboratorijske metode koje, takođe, omogućavaju dijagnozu VB su elektronska mikroskopija i PCR tehnika. Elektronska mikroskopija može detektovati jasan, trilaminarni ćelijski zid *Tropheryma whipplei* [17]. Detekcija 16s rRNA *Tropheryma whipplei* PCR tehnikom može se koristiti za inicijalnu evaluaciju, ali se dijagnoza mora postaviti duodenalnim biopsijama [12, 18]. Ova tehnika se može sprovoditi na bilo kom tkivu, krvi ili sekretu [19, 20]. Primenom drugih *imaging* tehnika za evaluaciju tankog creva (pasaža, enterokliza) vide se nespecifični znaci malapsorpcije, kao i zadebljanje mukoznih nabora, naročito u duodenumu i proksimalnom jejunumu [10]. Kod opisanog bolesnika, nalaz pasaže tankog creva ukazao je na diskretno izraženije Kerkingove nabore i reljef sluznice tankog creva, ali tipični radiografski znaci malapsorpcije nisu viđeni. Kompjuterizovanom tomografijom abdomena može se prikazati mezenterijalna i retroperitonealna limfadenopatija [21]. Nelečena VB vodi fatalnom ishodu. Terapija podrazumeva primenu antibiotika i supstitucijsku terapiju. Prvi

antibiotici koji su korišćeni za lečenje VB bili su tetraciklini. Međutim, nakon prekida terapije, relaps se javlja kod 28% bolesnika [1]. Antibiotska terapija za VB primenjuje se tokom dve nedelje – parenteralna primena ceftriaksona (2 g dnevno) ili streptomicina (2 g dnevno) sa penicilinom G (1,2 miliona internacionalnih jedinica dnevno). Posle toga, preporučuje se primena 160 mg trimetoprima i 800 mg sulfametoksazola dnevno, 1–2 godine [22, 23]. Ukoliko postoji alergija na neki od preporučenih lekova, tada se inicijalna terapija sprovodi hloramfenikolom, karbapenemima, penicilinom i cefalosporinima [12, 23]. Važeće preporuke za terapiju uglavnom su zasnovane na empirijskim podacima iz prikaza slučajeva ili serija slučajeva. Feurle i sar. [24] su 2010. godine objavili rezultate randomizovane kontrolisane studije u kojoj su dokazali da terapija ceftriaksonom ili meropenemom, praćena trimetoprim-sulfametoksazolom, dovodi do izlečenja VB [24]. I pored adekvatne antibiotske terapije, klinički relaps se može javiti kod 2–33% bolesnika u proseku posle 5 godina, naročito kod neurološke forme bolesti [25]. Uz kauzalnu terapiju sprovodi se i supstitucijska terapija. Primenjuju se vitamini, albumini, minerali, makro- i mikronutritijenti. Ova supstitucijska terapija ne mora biti dugotrajna jer, uvođenjem antibiotika, za 2–4 nedelje dolazi do dramatičnog povlačenja većine simptoma. Naš bolesnik je lečen indukcionom terapijom kotrimoksazolom i streptomicinom, a potom je nastavljeno kotrimoksazolom. U toku perioda praćenja nije registrovan relaps bolesti.

Zaključak

Za postavljanje dijagnoze VP najbitnije je da se posumnja na to retko oboljenje čije manifestacije su: malapsorpcija, povišena temperatura, artralgije i neurološka simptomatologija. Pravovremeno postavljanje dijagnoze i primena adekvatne terapije sprečavaju progresiju bolesti i fatalan ishod.

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Primljen 6. XII 2010.
Revidiran 13. XII 2011.
Prihvaćen 19. XII 2011.

CASE REPORT

UDC: 618.3-06:618.7]::616.12-008.46-055:26-08
DOI: 10.2298/VSP1206526I

Could it have been better? A patient with peripartum cardiomyopathy treated with conventional therapy

Da li je moglo biti bolje? Prikaz bolesnice sa peripartalnom kardiomiopatijom koja je lečena konvencionalnom terapijom

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Abstract

Introduction. Peripartum cardiomyopathy is a life threatening condition of unknown cause that occurs in previously healthy women. It is characterized by symptoms of heart failure due to left ventricular dysfunction that occurs in the last month of pregnancy or the first five months after delivery. **Case report.** We presented woman who underwent caesarean section due to preeclampsia. Two weeks after delivery first signs of heart failure appeared and only after six weeks following the onset of symptoms peripartal cardiomyopathy was recognized. A conventional treatment with diuretics, ACE inhibitor and beta blocker along with anticoagulant therapy was applied, which resulted in a complete recovery of the left ventricular function four months after. **Conclusion.** Timely detection and initiation of treatment are an important precondition for the complete or partial recovery.

Key words:
pregnancy complications, cardiovascular; cardiomypathies; diagnosis; drug therapy; treatment outcome.

Apstrakt

Uvod. Peripartalna kardiomiopatija je životno ugrožavajuće stanje nepoznatog uzroka koje se javlja kod prethodno zdravih žena. Karakterišu je simptomi srčane insuficijencije zbog disfunkcija leve komore koji se javljaju u poslednjem mesecu trudnoće ili u prvih pet meseci od porođaja. **Prikaz bolesnika.** Prikazana je bolesnica kod koje je zbog preeklampsije načinjen carski rez i kod koje su se, dve nedelje nakon porođaja, javili prvi znaci srčanog popuštanja, ali je peripartalna kardiomiopatija prepoznata tek šest nedelja od pojave simptoma. Sprovedeno je konvencionalno lečenje – diuretici, inhibitori, agiotenzin-konvertujućeg enzima (ACE) i beta blokatori, uz antikoagulantnu terapiju, što je nakon četiri meseca dovelo do potpunog oporavka funkcije leve komore. **Zaključak.** Pravovremeno otkrivanje i započinjanje lečenja je važan preduslov za potpuni ili delimični oporavak bolesnika sa peripartalnom kardiomiopatijom.

Ključne reči:
trudnoća, komplikacije, kardiovaskularne; kardiomiopatije; dijagnoza; lečenje lekovima; lečenje ishod.

Introduction

Peripartum cardiomyopathy (PPCM) is clinically defined as the occurrence of heart failure due to left ventricular dysfunction of unknown reasons, in late pregnancy or within the first five months after delivery [1].

Despite the low prevalence (approximately 1 out of 2,000 to 4,000 deliveries), this disease associated with pregnancy arises attention due to unexplained etiology and concerns about treatment [2].

We reported a patient with unrecognized PPCM who recovered entirely with conventional drug therapy including complete recovery of left ventricular systolic function. In this context we have considered a place of new modalities in the treatment of this disease.

Case report

Arterial hypertension was diagnosed in the third trimester of the first gravidity in a 33-year-old woman. The treatment with alpha-methyldopa resulted in a reduction but not normalization of blood pressure. Because of the development of preeclampsia – proteinuria and grade 3 arterial hypertension, pregnancy was terminated by cesarean section two weeks before the term. Two weeks after the delivery dyspnea appeared. Six weeks later the patient was admitted

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to the regional hospital because of dyspnea, paroxysmal nocturnal dyspnea and leg edema. Differential diagnosis was postpartum cardiomyopathy and the patient was sent to a tertiary institution.

On admission the patient was pale, respiratory rate was 24/min, heart rate > 120/min and blood pressure 120/80 mmHg. Her Body Mass Index (BMI) was 21 kg/m^2. Jugular veins were discretly distended. Auscultation of the lugs revealed normal breathing sound with late inspiratory crackles basally and bilaterally. Cardiac apex was palpable in the fifth intercostal space, 2 cm to the left of the medioclavicular line. Auscultation of the heart revealed systolic gallop rhythm with regurgitation murmur on the apex. The liver was palpable 3 cm below the rib. Bilateral pretibial edema was also detected.

The patient was free of previous cardiovascular disease or infection during pregnancy. During pregnancy the patient did not receive tocolytics. She did not use alcohol, cocaine and cigarettes.

Laboratory analyses at admission found sideropenic anemia (Hb 109 g/L, Ht 0.33, Fe 4 umol/L and transferrin saturation 5%), hypoproteinemia (54 g/L) with hipoalbuminemia (30 g/L), increased levels of uric acid (488 umol/L), ALT (110 U/L), LDH (727 U/L), and CRP (17 mg/dL). The brain natriuretic peptide (BNP) level was 3556 pg/mL. Other findings were within normal range, including prolactin levels and cardiac enzymes (CK-MB, troponin). The patient reported to have bronchopulmonary infection with bilateral pleural effusion and small pericardial effusion five years before pregnancy, which had resolved after three months. Because of this information we made basic immunological analyses (ANA, ANCA, immunoglobulin and complement components) which were all in referent ranges.

The first chest X-ray revealed enlarged heart and signs of lungs congestion. Sinus tachycardia was detected in ECG. Initial echocardiographic examination showed the dilated left ventricle with thin walls and septum and reduced systolic contractility (Figure 1), the dilated left atrium and the right ventricle, and Doppler ultrasound found mitral 2+ and tricuspid 3+ regurgitation. The right ventricular systolic pressure was estimated at 80 mmHg. Small pericardial effusion was also detected.

Table 1 shows the echocardiographic parameters of four consecutive examinations (on admission, after four, eight and 16 weeks of the start of the treatment).

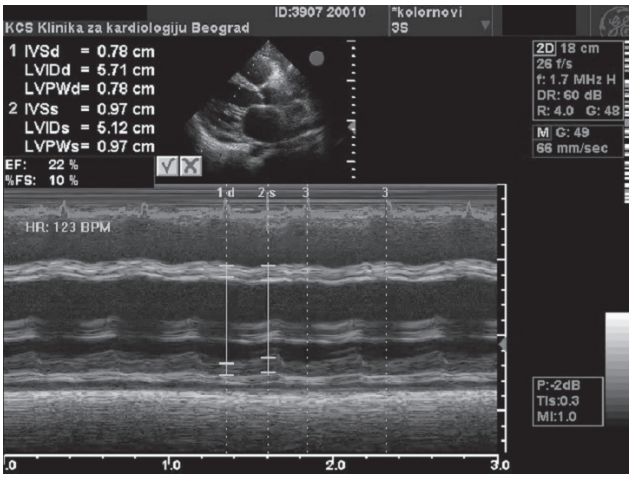

Fig. 1 – The echocardiographic finding of the left ventricle in M-mode (long parasternal view) at admission to our hospital

Magnetic resonance (MR) imaging with gadolinium showed the enlarged left ventricle echocardiographic end-diastolic diameter (EDD) 64 mm, end-systolic diameter (ESD) 53 mm) with ejection fraction (EF) 23%, end-diastolic volume (EDV) 145 mL, end-systolic volume (ESV) 114 mL, CO 2.3 L/min). Mitral regurgitation 2+ was also found. The right ventricular EF was estimated at 24% EDV 109 mL, ESV 83 mL, CO 1.81 L/min. There was a severe tricuspid regurgitation 3+. MR confirmed the existence of pericardial effusion. At the early stage after contrast application there were no signs to indicate the presence of intraluminal mass. In the late phase, 10 and 15 min after the application of contrast, the areas of delayed myocardial enhancement on MR images were not seen.

The treatment with the loop diuretics, aldosterone antagonist, ACE inhibitor, beta blocker and digoxin (only the first seven days, with control of serum concentrations) was started. The anticoagulant therapy with low molecular weight heparin was added shortly and it was replaced with warfarin after two weeks. Subjective improvement with the withdrawal of heart failure signs occurred after two weeks. The BNP level was reduced by > 80% after four weeks of the treatment, and decreased by > 90% after two months. However, it remained insignificantly increased even after four months (Table 1).

Table 1
The echocardiographic findings and brain natriuretic peptide (BNP) level from admission to the hospital to the last control (16 weeks)

Parameters	On admission	After 4 weeks	After 8 weeks	After 16 weeks
LVEDD (cm)	5.7	5.7	5.4	4.9
LVESD (cm)	5.1	4.5	4.2	3.4
EF (%)	22	35	38	57
FS (%)	10	15	17	30
LA (cm)	4,2	3.8	3.4	3.2
RV (cm)	3.2	2.8	2.4	2.2
RVSP (mmHg)	80	38	29	25
BNP (pg/ml)	3556	650	290	145

LVEDD – left ventricular end-diastolic diameter; LVESD – left ventricular end-systolic diameter; EF – ejection fraction of the left ventricle; FS – left ventricular shortening fraction; LA – left atrial diameter; RV – right ventricular diameter; RVSP – right ventricular systolic pressure

Controlled chest X-ray was performed after four weeks and it showed the decreased heart and the absence of congestion signs in the lungs. Eight weeks after the beginning of conventional treatment, significant improvement of systolic left ventricular function was registered (EF increased from 22% to 38%) with its normalization after 16 weeks (Table 1 and Figure 2).

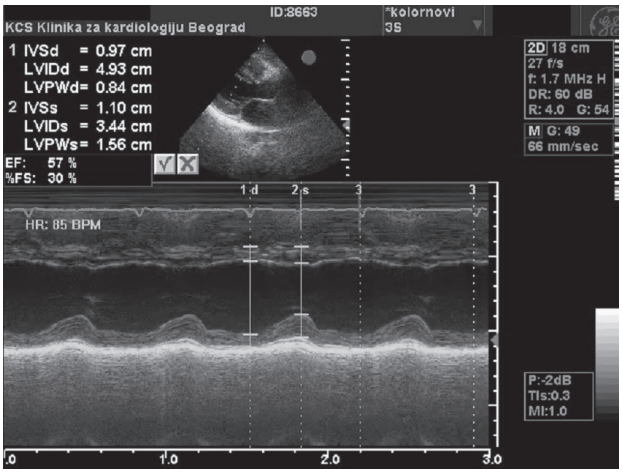

Fig. 2 – The echocardiographic finding of the left ventricle in M-mode (long parasternal view) 16 weeks after admission

Discussion

The etiology and pathogenesis of PPCM are unknown. There are many speculations about its etiopathogenesis.

During pregnancy intravascular volume and cardiac output increase while vascular resistance decreases. These changes could induce the left ventricular remodeling – eccentric hypertrophy with a mild reduction of systolic function. The theory that abnormal hemodynamic changes during pregnancy are responsible for PPCM is not convincing enough [3].

Based on the absence of differences in pathological findings between PPCM and idiopathic cardiomyopathy it was assumed that PPCM was a type of idiopathic dilated cardiomyopathy induced by hemodynamic stress in pregnancy. This hypothesis was called into question because, in this case, the incidence of PPCM would be higher, and it would be manifested in the second trimester when hemodynamic changes characteristic for pregnancy are most present and not in the third trimester or the postpartum period [3, 4]. Complete recovery in 30% and partial in about 50% of patients distinguish this form of cardiomyopathy from idiopathic dilated cardiomyopathy in which recovery is extremely rare.

The theory that myocarditis is the reason of PPCM was accepted for a long time [3]. It was based on the fact that during pregnancy there is a decrease in both cellular and humoral immunity, with an increased level of corticosteroids and "blocking antibodies". This hypothesis was challenged because myocarditis was confirmed in 0% to 100% of patients, according to different authors [3].

A possible reason for PPCM could be an abnormal immune response to pregnancy and within it the reduced possibility of "cleaning" of antigens which enter the circulation of the mother [5]. It was assumed, thus, that fetal cells, which remained unrecognized and unrejected from maternal circulation, could remain in cardiac tissue and could cause an abnormal autoimmune response in the postpartum period. Also, rapid degeneration of the uterus after delivery may result in fragmentation of tropocollagen and release of actin and myosin into the circulation which could result in cross-reactions with myocardial proteins. Detection of antibodies to β1 receptors in conjunction with increased adrenergic tone due to emotional and physical stress characteristic for pregnancy might represent another possible mechanism in the development of PPCM [3].

Apoptosis of cardiomyocytes is another possible reason for PPCM development [6]. Also, it was found that increased levels of CRP and tumor necrosis factor alpha, found in patients with PPCM, correlated with the increased left ventricular dimensions [6].

Prolonged tocolytic therapy also could be a possible cause of PPCM but it is not determined whether it directly affects its manifestation or unmasks preexisting subclinical disease [7]. Genetic predisposition was shown in the series of cases in families but there is still no confirmation of this hypothesis.

Interest in determination of the role of prolactin in the pathogenesis of PPCM has increased in the recent years. Prolactin exists in at least two biologically active forms with the opposite effects. A 23kDa form promotes angiogenesis and has a protective effect on endothelial cells in contrast to 16kDa that induces cellular apoptosis and disconnection of capillary structures, disturbs the metabolism of cardiomyocites and influences negatively contractility. It is assumed that the 16kDa form of prolactin which occurs under the influence of oxidative stress has a role in the development of PPCM [8, 9].

Numerous factors increase the risk of PPCM: higher maternal age, multiple gravity, twin gravity, poor socioeconomic status and arterial hypertension. The presented patient had a history of preeclampsia. Arterial hypertension (pregnancy-induced hypertension, preeclampsia and postpartum hypertension) was detected as a risk factor in 22% to 43% of patients with PPCM [9, 10]. The fact that in preeclampsia there are no significant changes in the left ventricular function classified hypertension as a risk factor and not as a cause [3, 11].

The diagnosis of PPCM is echocardiographic. Magnetic resonance imaging is a complementary method of diagnosing PPCM. In addition to determination of segmental and global left ventricular function, the finding of the delayed contrast enhancement (with gadolinium) can help in differentiating the inflammatory from noninflammatory form of PPCM. Subepicardial nonvascular nodular or linear distribution of contrast is characteristic of thr inflammatory form of PPCM [12]. The absence of delayed contrast enhancement, as in our patient, is typical of noninflammatory origin of PPCM. Based on this evidence we assumed that malnutrition, genetics, prolactin production excess, abnormal hor-

mone function and increased adrenergic tone caused PPCM in our patient. MR findings and the result of endomyocardial biopsy can influence the decision on the treatment modality.

The treatment of PPCM is empirically limited to a standard therapy of heart failure with ACE inhibitors/AT1 receptor blockers, beta blockers, diuretics and aldosterone antagonist. Because of the relationship between impaired left ventricular function and prothrombotic state, characteristic for pregnancy, and the increased risk of thromboembolic complications indicated a need for additional anticoagulant therapy.

There are attempts to use different modalities of treatment based on some of these possible mechanisms of PPCM. One of those was the idea to use intravenous immunoglobulin with immunomodulatory properties based on the hypothesis of abnormal maternal immunoglobulin response. The evidences of this modality of treatment are limited by a small number of included patients [13].

Based on the theory of inflammation, mediated by cytokines, in the development of PPCM, there was an attempt in pentoxifylline treatment that inhibits the production of tumor necrosis factor and prevents apoptosis. Sliwa et al. [14] showed that patients who receive conventional therapy and pentoxiphylline, six months after PPCM diagnosing, are significantly better in recovery of systolic left ventricular function [14]. But the absence of recent confirmation of this treatment did not make this treatment modality widely accepted.

The treatment of PPCM with bromocriptine was based on the concept that oxidative stress-mediated cleavage of prolactin in antiangiogenic and proapoptotic 16kDa form is responsible for the development of PPCM. Individual case reports about the application of bromocriptine in the treatment of PPCM showed faster recovery of the left ventricular function [15–18]. Last year Sliwa et al. [19] published the results of a pilot study in which they applied bromocriptine to 10 patients (2 × 2.5 mg two weeks and 2.5 mg for six more weeks). Significantly better recovery of left ventricular function and mortality reduction was reported in a group of patients who received bromocriptine in comparison to patients treated with empirical therapy [19]. The limitation of this study was the small number of patients and African origin of the patients which could imply phenotypic differences and may be the reason for inapplicability of the obtained results to patients from other regions.

Our patient was treated with conventional therapy. In adjusting intensity of the treatment, we were not guided primarily by subjective and objective improvement of the patients and by the recovery of heart systolic function (EF), but mostly by the BNP level (Table 1). Namely, the subjective and objective improvement occurred after two weeks. The BNP level in this period was significantly, but not satisfactorily reduced and we decided to continue intensive treatment in the hospital. A slow reduction of the BNP level corresponded to a slow recovery of left ventricular function that is characteristic for this form of cardiomyopathy.

During a prolonged recovery, there was a concern about whether conventional therapy would lead to a full recovery and if it was still necessary to apply a modern treatment modality. Among the previously mentioned modern therapy modalities, we preferred bromocriptine more than pentoxifylline primarily due to the results of gadolinium MR examination.

The reason for conventional therapy application was the lack of family consent for bromocriptine treatment. It is possible that the lack of agreement came after introduction of a patient and family members to numerous potential side effects of bromocriptine, which include increasing incidence of arterial hypertension, stroke, epilepsy, myocardial infarction, arterial thrombosis or intracardiac thrombus formation [20, 21]. We were extremely satisfied when detected that application of conventional therapy led to a complete recovery of left ventricular function. It remains controversial whether the use of bromocriptine would lead to earlier recovery. This doubt still remains in the shadow of the fact that all "modern" treatment modalities are used in addition to conventional therapy, and the question of actual profit from its application remains to be open.

Conclusion

Anyway, timely detection and initiation of treatment are an important precondition for complete or partial recovery of patients with PPCM.

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Received on December 8, 2010.
Revised on January 24, 2011.
Accepted on February 11, 2011.

CASE REPORT

UDC: 616-006-07:[616.233/.235-07:616-072.1
DOI: 10.2298/VSP1206531S

The role of autofluorescence bronchoscopy in monitoring a tumorous lesion in the bronchial mucosa: a case report

Uloga antifluorescentne bronhoskopije u praćenju tumorskih lezija bronhijalne mukoze

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Abstract

Introduction. Autofluorescence bronchoscopy (AFB) is a diagnostic procedure that is included in all diagnostic algorithms discovering precancerous lesions in the large airways. **Case report.** We presented a 71-year-old patient submitted to exploration due to prolonged cough. Both noninvasive and invasive pulmonary diagnostic management was carried out. On bronchoscopy, an endobronchial mass was detected in the apical bronchus. A positive endoscopy finding indicated AFB which disclosed a fluorescence alteration of the tumor mucosa and the former bronchoscopy site. Histopathological analysis of the catheter biopsy obtained samples from the right upper lobe confirmed fibrinous purulent pneumonia in organization. The applied treatment resulted in regression of both symptoms and the lesion in the right upper lobe. Due to a positive AFB finding, the patient was regularly observed over the following three years, having had three control AFB to monitor the initial finding. **Conclusion.** AFB may be utilized in the routine of everyday practice to assess the spread of the disease, as well as in the post-surgical and long-term follow-up of operated patients. The procedure may also be applied to enable an easier and more reliable observation of patients with suspicious endobronchial lesions, smokers with altered fluorescence of the bronchial mucosa, and chronic patients.

Key words:
lung neoplasms; diagnostic techniques and procedures; bronhoscopy; fluorescence angiography.

Apstrakt

Uvod. Autofluorescentna bronhoskopija (AFB) je dijagnostička metoda koja je zastupljena u svim dijagostičkim algoritmima za otkrivanje prekanceroznih lezija u velikim disajnim putevima. **Prikaz bolesnika.** U radu je prikazan bolesnik star 71 godinu, koji je ispitivan zbog prolongiranog kašlja. Sprovedena je neinvazivna i invazivna pulmološka dijagnostika. Bronhoskopski, u apikalnom segmentu desnog gornjeg bronha viđena je endobronhijalna masa. Zbog pozitivnog endoskopskog nalaza bila je indikovana AFB. Autofluorescentnom bronhoskopijom viđena je fluorescentno izmenjena sluznica tumora i mesto prethodne bronhoskopije. Patohistološkim pregledom materijala dobijenog kateter biopsijom iz desnog gornjeg režnja dokazana je fibrinozno gnojna pneumonija u toku organizacije. Nakon sprovedene terapije došlo je do povlačenja tegoba i regresije promene u području gornjeg lobusa sa desne strane. Zbog pozitivnog nalaza AFB bolesnik je bio na opservaciji tokom naredne tri godine. Ukupno su urađene tri kontrolne AFB na kojima je praćen stacionaran nalaz. **Zaključak.** Autofluorescentna bronhoskopija se može koristiti u svakodnevnoj praksi za proveru proširenosti bolesti, u postoperativnom praćenju nakon kurativne hirurgije i u dugoročnom praćenju hirurški lečenih bolesnika. Takođe, moguća je primena AFB za opservaciju bolesnika sa suspektnim endobronhijalnim promenama, kao i kod fluorescentno izmenjene sluznice pušača i hroničnih bolesnika.

Ključne reči:
pluća, neoplazme; dijagnostičke tehnike i procedure; bronhoskopija; angiografija, fluoresceinska.

Introduction

Lung cancer is the most common death cause in patients with malignant diseases. The treatment of choice in these patients includes early diagnosis and a radical tumor resection. There is a great interest to develop imaging techniques which might detect the slightest lesion in the mucosa of the airways. Autofluorescence bronchoscopy (AFB) is a relevant diagnostic procedure included in all diagnostic algorithms to discover precancerous lesions in the large airways, like metaplasia, dysplasia, carcinoma in situ and microinvasive cancer [1].

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Case Report

A 71-year-old male patient was examined in the Institute for Pulmonary Diseases of Vojvodina in Sremska Kamenica, in May 2008. The patient had gradually developed the symptoms of fatigue and productive cough without blood traces, reporting a 30 pack-year smoking history. The patient's chest X-ray finding revealed a non-homogeneous shadowing in the right suprahilar region, projecting in the upper lobe (Figure 1).

Computerized tomography of the chest was performed, disclosing an irregular, infiltrative lesion of the density of 40 Hu in the third segment of the right lobe, next to the incisure. The mediastinal lymph nodes were not enlarged (Figure 2).

Bronchoscopy was performed. The larynx, trachea, bifurcation and the left bronchial tree had a normal endoscopic finding, as well as the right main bronchus, intermediary bronchus and lower bronchi. However, a tumorous lesion was seen in the apical segment. Histopathology of the bronchial biopsy obtained from the right upper bronchus, as well as of the biopsy from endobronchial mass and the sample obtained by transbronchial needle biopsy *via* the right upper bronchus provided a purulent exudate and established the histological elements of chronic nonspecific inflammation, with no elements of tumor tissue. The positive endoscopic finding indicated hospitalization.

On admission to the Institute, the patient was eupnoic, afebrile, cordially compensated. His blood test findings were presented with leucocytosis (12.3×10^9/L), elevated erythrocyte sedimentation rate (70/-) and elevated levels of C-reactive protein (> 100 mg/L), fibrinogen (6.3 g/L), urea (8.4 mmol/L), and creatinine (135 μmol/L). Pulmonary gas exchange at rest suggested mild hypoxemia (SaO_2 = 94.2%). Bronchoscopy with autofluorescence imaging (AFI) was performed. Endo-

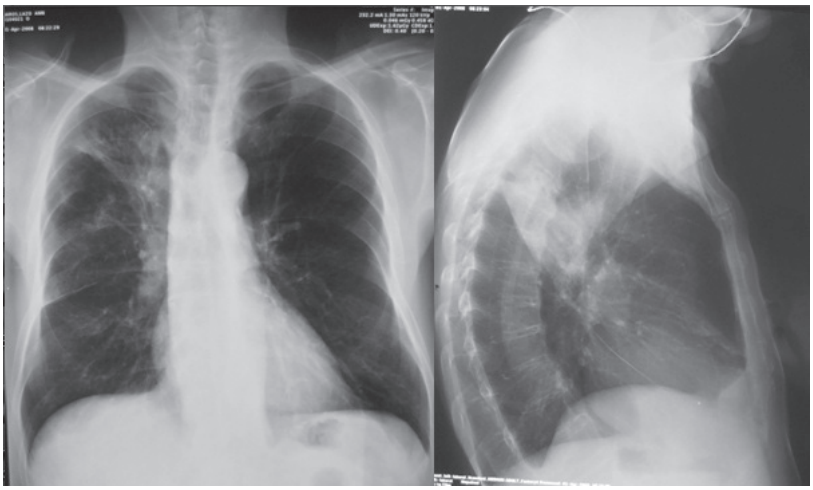


Fig. 1 – Posteroanterior and left lateral chest X-ray finding

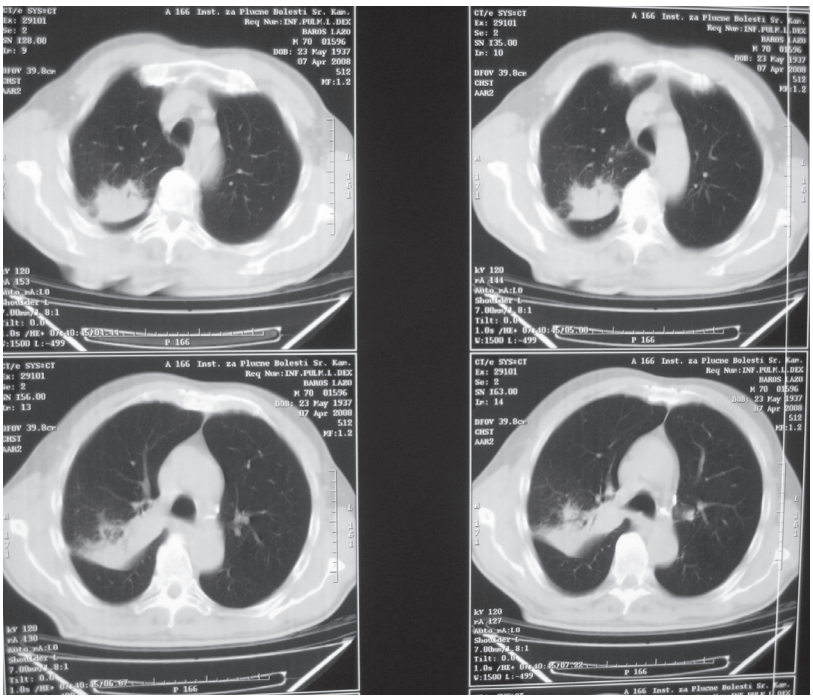


Fig. 2 – Computed tomography scan findings of the chest did not reveal enlarged mediastinal lymph nodes

scopically the superior bronchus was involved by a gelatinous tumorous formation which entirely obstructed the orifice for the apical segment. AFB revealed an altered fluorescence of tumor mucosa and the former bronchoscopy site (Figure 3).

The patient came for control six months later. On physical examination, the patient was entirely asymptomatic, but his chest X-ray finding was presented with more voluminous hilly and bilateral striped shadows pericardially (Figure 5).

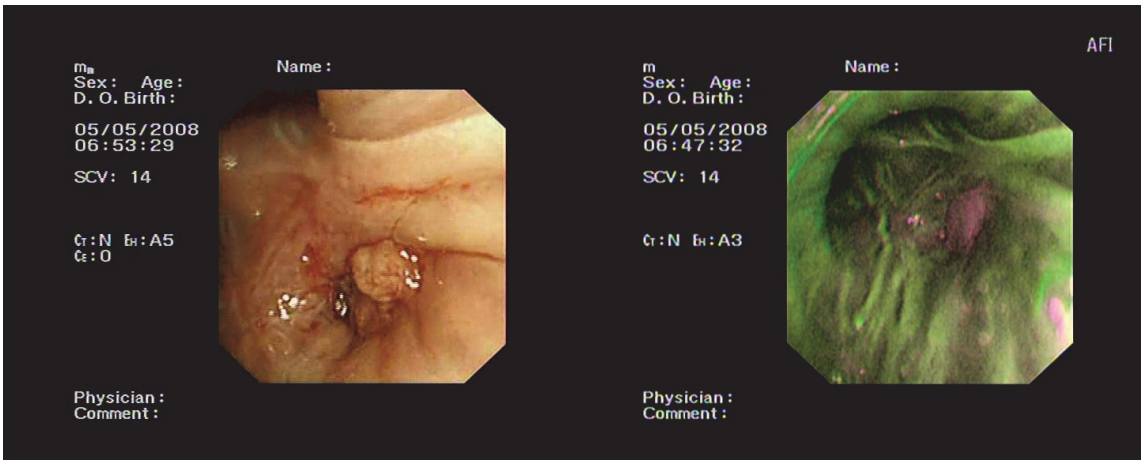


Fig. 3 – Endoscopy and autofluorescence findings of autofluorescence bronchoscopy

Histopathological analysis of the bronchial biopsy sample obtained from the right upper bronchus confirmed fibrinous purulent pneumonia in organization (Figure 4). No tumorous tissue was found in the examined sample.

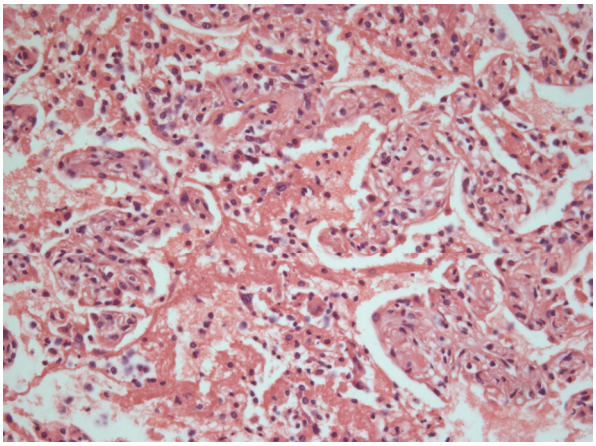


Fig. 4 – Histopathological finding: fibrinous purulent pneumonia in organization (hematoxylin-eozin, 400×)

The patient responded well to the applied antibiotic therapy and corticosteroids (prednisolone tablets in the dose of 30 mg) accompanied with H_2 receptor antagonists. Rehabilitation treatment with breathing exercises was also carried out. The control chest computed tomography (CT) finding showed considerable regression of the lesion in the right upper lobe. The control inflammation markers were within normal levels, including erythrocyte sedimentation rate (10/-), white blood count (7.1×10^9/L). C reactive protein (<6 mg/L), fibrinogen (3.5 g/L). The patient was discharged for further home treatment, with recommendations for control examination and bronchoscopy indicated by a positive autofluorescence finding.

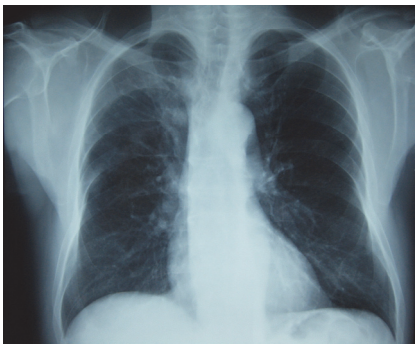


Fig. 5 – Posteroanterior chest X-ray finding six months after discharge revealed more voluminous hilly and bilateral striped shadows epricardially

Laboratory test findings, pulmonary gas exchange and lung function test findings were normal as well. The control chest CT finding delineated a discrete zone of hypodense lesions in the right upper lobe corresponding to post inflammatory lesions (Figure 6).

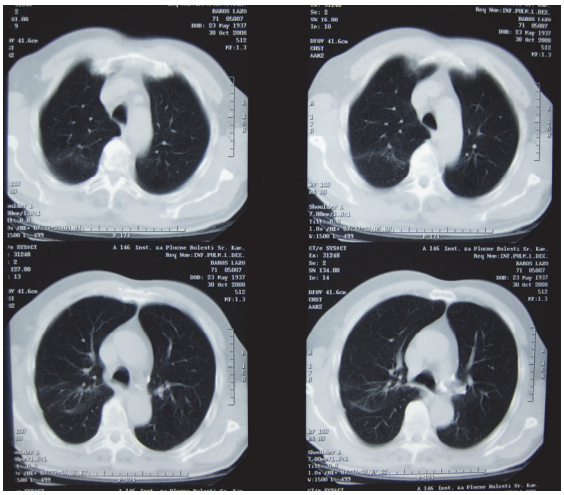


Fig. 6 – Control chest computed tomography finding: discrete zone of hypodense lesions in the right upper lobe

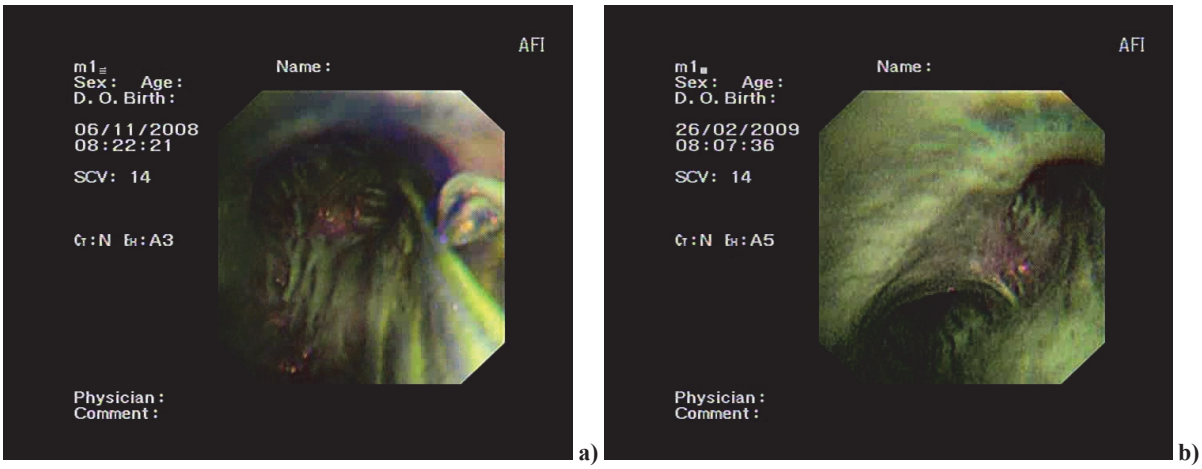
Bronchoscopy was performed. Two tiny granulations were observed in the upper arch of the right bronchus, as well as a wider intersegmental carina, with all orifices free. AFI detected no altered fluorescence of the granulations, unlike the altered fluorescence of the mucosa in the intersegmental carina, between the posterior and anterior orifices (Figure 7a).

The histopathological and cytological analysis of the samples showed no elements of tumorous tissue. The patient was suggested to have a control bronchological exploration in six months.

At the control six month later, the patient reported no subjective symptoms. The chest X-ray finding showed no deviations from the former one. A control bronchoscopy was performed, providing a normal endoscopic finding. In the bronchus for the right upper lobe, on its lateral wall towards the orifice for the posterior one, a tine granulation was detected. AFB revealed the altered mucosa on the intersegmental carina in the right upper lobe, at the possible site of the former biopsy. The granulation did not fluoresce (Figure 7b).

cancerous lesions than conventional bronchoscopy. Many multicentric studies have revealed that AFB performed in combination with a standard one, increases the percentage of early detected dysplasia, cancer *in situ* and microinvasive lung cancer [2–9]. It has been confirmed that the lesions in terms of metaplasia, dysplasia, dysplasia and carcinoma in situ may appear over large mucosa regions of the tracheobronchial tree, particularly in smokers and patients with invasive diseases [9, 10]. The studies having been carried out since 1990 using AFB to diagnose premalignant lesions of the central airways have confirmed that premalignant lesions' behavior is unpredictable. Certain advanced lesions, defined as “carcinoma in situ” have exhibited the capacity of spontaneous regression [11]. AFB has a higher potential than the conventional, white-light bronchoscopy. Recent studies have shown that it may be applied in the routine of everyday practice to assess the spread of the disease [12, 13].

Nowadays, there is a great debate about a large-scale application of AFB in the screening of lung cancer. Any new



a) b)

Fig. 7 – Endoscopy findings of autofluorescence bronchoscopy

a – no altered fluorescence of the granulations and altered fluorescence of the mucosa in the intersegmental carina (first control six month after disharge)
b – the altered mucosa at the possible site of the former biopsy (second control six month after the first post-disharge control)

The control bronchoscopy was performed one year later with normal endoscopic finding. The patient reported no symptoms. The radiological finding was unchanged. Laboratory test findings were within normal levels. Pulmonary gas exchange, spirometry and plethysmography findings were normal. The formerly detected granulations were not delineated. Autofluorescence showed fluorescence of sites of former biopsy at one point of the carina of the upper bronchus, as well as on the intersegmental carina. The histopathological analysis showed usual histological elements. The patient was dismissed with a recommendation to continue with regular observations in the future.

Discussion

The investigations performed so far have very well established that AFB achieves better results in detecting pre-

procedure requires a detailed analysis and assessment of possible negative effects on a patient.

Conclusion

When applied in combination with a standard bronchoscopy, AFB increases early detection of premalignant lesions, dysplasia, carcinoma *in situ* and micro invasive lung cancer. This method may be used in the routine of everyday practice to assess the spread of the disease, in the postoperative monitoring and a long-term follow-up of surgically treated patients. In addition, it may also be utilized to enable an easy and more reliable observation of patients with suspicious endobronchial lesions, as well as in smokers and chronic patients with altered mucosa on fluorescence.

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Received on December 21, 2010.
Accepted on February 21, 2011.

CASE REPORT

UDC: 615.065:612.017]:[616.24:616.36
DOI: 10.2298/VSP1206536M

Nitrofurantoin-induced immune-mediated lung and liver disease

Bolest pluća i jetre indukovana nitrofurantoinom i imunološki posredovana

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Abstract

Introduction. Nitrofurantoin, a furan derivative, introduced in the fifties has widely been used as an effective agent for the treatment and prevention of urinary tract infections (UTI). Spectrum of adverse reactions to nitrofurantoin is wide, ranging from eosinophilic interstitial lung disease, acute hepatitis and granulomatous reaction, to the chronic active hepatitis, a very rare adverse effect, that can lead to cirrhosis and death. **Case report.** We presented a 55-year- old female patient with eosinophilic interstitial lung disease, severe chronic active hepatitis and several other immune- mediated multisystemic manifestations of prolonged exposure to nitrofurantoin because of the recurrent UTI caused by *Escherichia coli*. We estimated typical radiographic and laboratory disturbances, also restrictive ventilatory changes, severe reduction of carbon monoxide diffusion capacity and abnormal liver function tests. Lymphocytic-eosinophylic alveolitis was consistent with drug-induced reaction. Hepatitis was confirmed by liver biopsy. After withdrawal of nitrofurantoin and application of high dose of glicocorticosteroids, prompt clinical and laboratory recovery was achieved. **Conclusion.** Adverse drug reactions should be considered in patients with concomitant lung and liver disease. The mainstay of treatment is drug withdrawal and the use of immunosuppressive drugs in severe cases. Consideration should be given to monitor lung and liver function tests during long term nitrofurantoin therapy.

Key words:
nitrofurantoin; urinary tract infections; drug toxicity; immunologic factors; hepatitis; pneumonia.

Apstrakt

Uvod. Nitrofurantoin, derivat furana, ušao je u upotrebu pedesetih godina i široko je upotrebljavan kao efikasan agens za terapiju i prevenciju infekcija urinarnog trakta. Spektar neželjenih reakcija na nitrofurantoin je veliki, od eozinofilne intersticijske bolesti pluća, akutnog hepatitisa, granulomatoznih reakcija, do veoma retkih efekata u vidu hroničnog aktivnog hepatitisa koji može dovesti do ciroze jetre i smrti. **Prikaz bolesnika**. Predstavili smo bolesnicu, staru 55 godina, sa eozinofilnim intersticijskim oboljenjem pluća, teškim hroničnim aktivnim hepatitisom i drugim imunološki posredovanim multisistemskim manifestacijama nakon produžene ekspozicije nitrofurantoinu zbog rekurentnih infekcija urinarnog trakta uzrokovanih *Escherichia coli*. Nađeni su tipični radiografski i laboratorijski poremećaji, takođe restrikcijski poremećaj ventilacije, teško oštećenje difuzijskog kapaciteta za ugljen monoksid i poremećeni testovi funkcije jetre. Limfocitno- eozinofilni alveolitis bio je konzistentan sa neželjenom reakcijom na lek. Hepatitis je potvrđen biopsijom jetre. Nakon ukidanja nitrofurantoina primenjene su visoke doze glikokortikosteroida, što je dovelo do brzog kliničkog i laboratorijskog oporavka. **Zaključak.** Neželjene reakcije na lekove trebalo bi razmatrati kod bolesnika sa istovremenim oboljenjem pluća i jetre. Osnova lečenja je ukidanje leka koji je doveo do reakcije i davanje imunosupresivnih lekova u težim slučajevima. Može se preporučiti praćenje funkcije pluća i jetre tokom dugotrajne primene nitrofurantoina.

Ključne reči:
nitrofurani; urinarni trakt, infekcije; lekovi, toksičnost; imunski faktori; hepatitis; pneumonija.

Introduction

Nitrofurantoin, a furan derivative, was introduced in the fifties and has widely been used as an effective agent for the treatment and prevention of urinary tract infections (UTI). Nitrofurantoin-induced hepatic injury was first reported in 1961 [1]. Since then a spectrum of adverse reactions to nitrofurantoin has been reported, ranging from eosinophilic interstitial lung disease, acute hepatitis, granulomatous reaction, to the very rare adverse effect of chronic active hepatitis that can lead to cirrhosis or death [2]. Autoimmune liver disease is not uncommon cause of chronic

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hepatitis in women. Although autoimmune destruction usually occurs without an identifiable trigger, some drugs such as methyldopa, minocycline and nitrofurantoin are associated with autoimmune liver disease [3]. Today, nitrofurantoin is well recognized as a cause of adverse drug reactions. Although the combination of lung and liver toxicity is rare, concomitant pulmonary and liver disease can occur together and it may well be that these share a common autoimmune mechanism [4–8]. Eighty five percent of patients having nitrofurantoin-associated pulmonary reactions are women. This observation may be related to the fact that women are more susceptible to recurrent UTI [9, 10].

We presented a middle age female patient with eosinophilic interstitial lung disease, severe chronic active hepatitis and several other immune–mediated multisystemic manifestations after prolonged exposure to nitrofurantoin.

Case report

A 55-year-old female was admited to hospital because of breathless, nonproductive cough and fever during six weeks. Few months prior admission the patient began to suffer from general weakness, nausea, weight lost and polyarthralgia without morning rigidity. Her past medical history included mesangioproliferative glomerulonephritis (diagnosed in 1985 and treated with systemic glicocorticosteroids), hypothyreosis (because of that she used levotiroxin substitution). The patient had been treated with nitrofurantoin 100 mg twice daily for the last six months because of the recurrent UTI caused by *Escherichia coli*. There was no history of liver disease; she denied consumption of any other medications, alcohol or tobacco. Physical examination on admission revealed profound jaundice, obesity, dark colour of skin with excoriated papulomatous rash on the face and arms (Figure 1). Auscultation of the lungs revealed normal breath sound with diffuse, bilateral, fine end-inspiratory crackles. Initial laboratory tests revealed an erythrocyte sedimentation rate (ESR) of 24 mm/h (normal range 0–12 mm/h), elevated C-reactive protein – 9 mg/L (normal 0–4 mg/L), a normal blood count with eosinophilia (880/mL, normal 100–250/mL), deranged liver function with total serum bilirubin of 139 μmol/L (normal range 2–21 μmol/L) with a direct fraction of 37 μmol/L (normal range 0–5 μmol/L), aspartate aminotransferase (AST) – 466 IU/L (normal range 0–34 IU/L), alanine aminotransferase (ALT) – 430 IU/L (normal range 7–49 IU/L), lactate dehydrogenase (LDH) – 535 IU/L (normal range 200–378 IU/L), alkaline phosphatase–1,111 IU/L (normal range 7–290 IU/L), gama-glutamyl-transpeptidase 1,590 IU/L (normal range 0–38 IU/L), normal total protein – 69 g/L, low albumin – 29 g/L (normal range 32–48 g/L). Other biochemical parameters and coagulation screen were normal. Antinuclear antibodies (ANA) were positive (++++ speckled pattern of fluorescence), also anti-smooth muscle antibodies – ASMA (++). Antibodies for extractable nuclear antigens, anticardiolipin, anti-mitochondrial, anti-CCP (cyclic citrullinated peptide), anti-neutrophil cytoplasmic (against myeloperoxidase and proteinase 3) were normal. Relative values of subpopulations of T lymphocytes in peripheral blood (CD4+ and CD8+) were normal, with normal CD4+/CD8+ ratio, so values of natural killer cells (CD16+, CD56+) were mildly elevated. There was an accompanying hyper-gammaglobulinemia with elevated IgG – 22 g/L (normal range 7–16 g/L), IgA 4.41 g/L (normal range 0.7–4 g/L), IgE 902 IU/mL (normal range 0–100 IU/mL) and normal IgM level. Serological tests for intestinal parasites, hepatitis A, B, C, human immunodeficiency, Epstein Barr and cytomegaloviruses were negative. No eggs of parasites were found in feces. Chest radiography (X-Ray) showed bilateral ground-glass and micronodular opacities, predominantly in lower lung fields (Figure 2). Computed

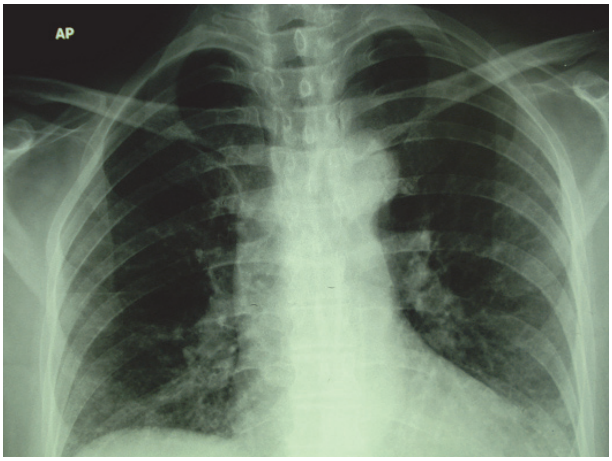
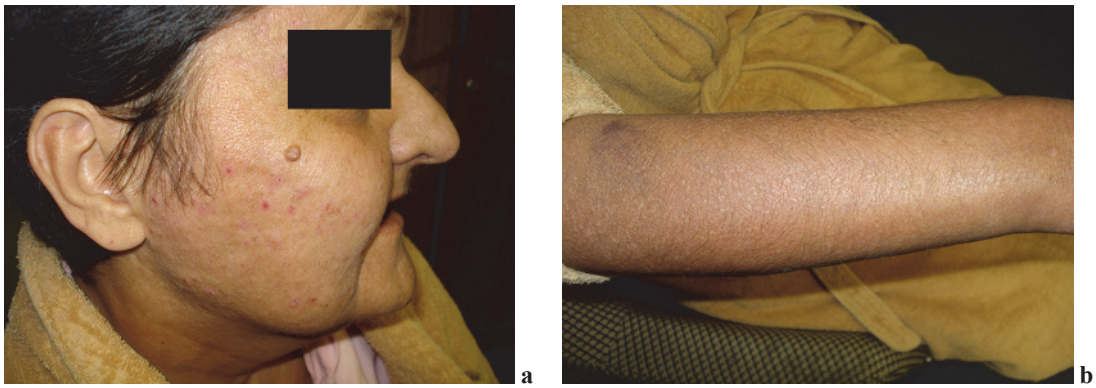


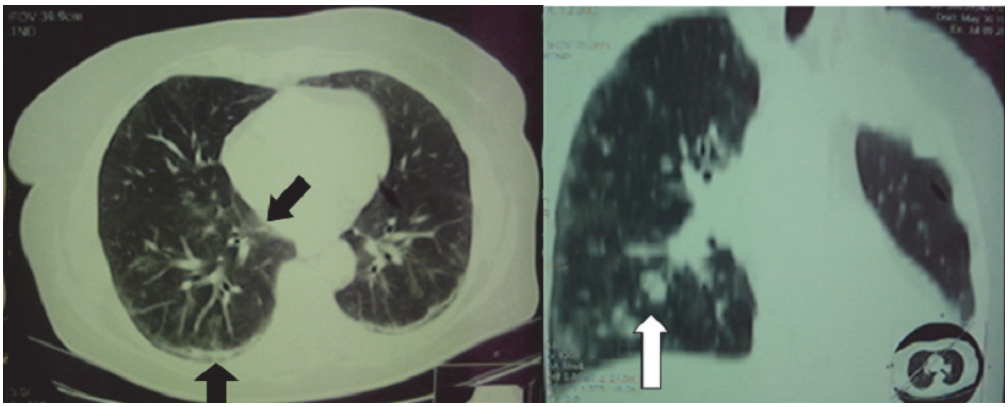
Fig. 2 – Chest radiography: ground glass and micronodular opacities, predominantly in lower lung fields



a **b**

Fig. 1 – A female patient, 55-year-old, with lichen simplex chronicus on the face (a) and the arm (b)

tomography (CT) revealed ground-glass opacities and consolidations without significant fibrotic changes (Figures 3). Pulmonary function tests showed moderate restrictive ventilatory changes (forced vital capacity was 55% predicted) and severe reduction carbon monoxide diffusion capacity (DLCO 47%, DLCO/VA 49% predicted). The respiratory arterial blood gases analysis at rest revealed mild hypoxemia with pO2 8 KPa (9.6 KPa normal for her age), oxygen saturation at 90% and severe hypocapnia with pCO2 2.8 KPa (normal range 4.6–6 KPa). Echocardiography and electrocardiography were normal. Abdominal ultrasonography found mild enlargement of spleen and liver with hyperechogenic structure with no evidence of gallstones or biliary dilatation. Doppler ultrasound showed no evidence of portal or hepatic vein occlusion. Bronchoscopic findings were normal. Histological finding of transbronchial biopsy was nonspecific. Bronchoalveolare lavage (BAL) fluid analysis did not show bacterial, fungal agents or acid fast bacilli. BAL cytology cell profile showed macrophages 14%, lymphocytes 75% and eosinophils 11% with decreased CD4/CD8 ratio. A liver biopsy was performed showing severe chronic active hepatitis, which was considered to be consistent with a drug induced hepatitis (Figure 4). Apperance of eyes and mouth dryness Shirmer`s test was performed which showed reduced secretion of tears (3 mm/5 min). Dermatological examination established Lichen simplex chronicus (Figure 1). After withdrawal of nitrofurantoin, high dose of glicocorticosteroids was applied – metylprednisolon in daily dose of 80 mg (60 mg in the morning and 20 mg in the evening). The result was a prompt clinical and laboratory recovery (symptoms and signs wanished, normalisation of acute phase reactants, liver and lung function parameters). The patient was discharged two weeks later and swiched to oral prednisone with taper to maintenance dose of 10 mg daily. Control examinations after three, six and twelve months showed normal physical findings, laboratory tests, chest X-ray and CT (Figure 5), abdominal ultrasonography, spirometry and carbon monoxide diffusion capacity. Follow-up was recommended.



a b

Fig. 3 – Chest computerized tomography: a – ground-glass opacities (black arrows); b – consolidation (white arrow) without significant fibrotic changes

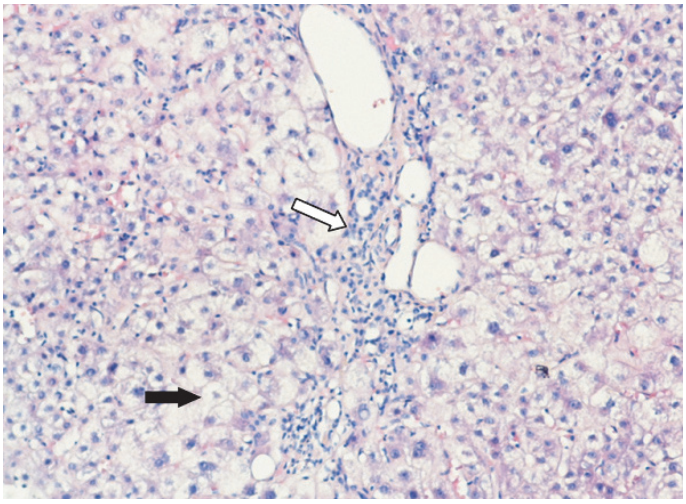
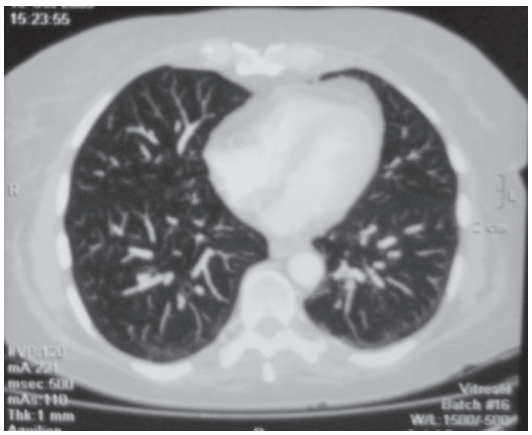


Fig. 4 – Histopathological finding of liver biopsy demonstrates chronic active hepatitis (H&E, 40×): a portal tract is expanded by a lymphoplasmacytic inflammatory infiltrate (white arrow). The interface between a portal tract and the parenchyma is disrupted by inflammation. The parenchymal sinusoids are suffused with lymphocytes and there is disruption of normal lobular hepatocyte architecture with ballooning of damaged hepatocytes (black arrow)


a)

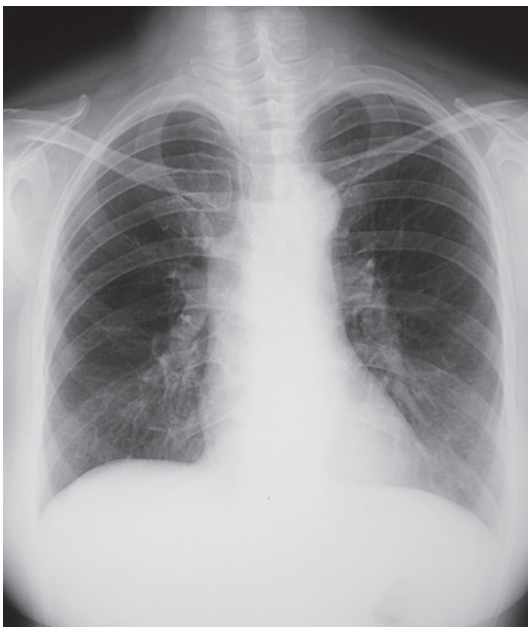

b)

Fig. 5 – Disappearance of pathological pulmonary changes six months after the treatment shown by multi-slice computerized tomography (a) and chest radiography (b)

Discussion

Nitrofurantoin is widely used for both acute and chronic management of UTI. It is cheap and effective, with a low incidence of resistance in common urinary pathogens; it is also safe in pregnancy [11]. Adverse drug reactions to nitrofurantoin include pulmonary reactions, hepatic toxicity, blood dyscrasias, peripheral neuropathy, etc [9]. Concomitant pulmonary and hepatic toxicity secondary to nitrofurantoin is rare with few reported cases [4–8]. The vast majority of pulmonary reactions to nitrofurantoin (90%) are acute and characterised by fever, cough, dyspnoea, and peripheral eosinophilia [9–10]. Nitrofurantoin also causes a range of subacute or chronic pulmonary disease, often presenting with insidious onset of increasing dyspnoea, dry cough and radiological evidence of fibrosis [10]. Because of that optimal duration of nitrofurantoin treatmen should not be over 14 days. Also, profilactic treatment of recurrent UTI should be discontinued, with switch by other effective antibacteriale medications. In patients who have some pulmonary, hepatic, alergic, neurologic disorder, anemia, diabetes or vitamin B deficiency special caution is necessery. Although severe adverse reactions caused by nitrofurantoin are rare, consideration should be given to monitoring lung and liver function tests during a long-term nitrofurantoin therapy. Pulmonary function tests (PFTs) may show a restrictive pattern with a reduced carbon monoxide diffusion capacity. Nitrofurantoin has been linked to autoimmune hepatitis, but in view of the rarity of the association, almost all reports of the association have been single case reports or small series [1–3]. Further information has been obtained from national adverse drug reaction monitoring agencies in the Netherlands [12] and Denmark [13] and it has been estimated that the incidence of nitrofurantoin-induced hepatic injury is low at about three cases in 1,000,000 [14].

The underlying mechanism behind nitrofurantoin toxicity remains uncertain; an immunological response is suggested by the presence of autoantibodies (ANA, ASMA). Direct cytotoxic mechanisms, for example by increased oxidative stress, have also been suggested [15]. Cytotoxic T-cells play a pivotal role in the pathogenesis of nitrofurantoin-induced liver injury. It has been hypothesized that a breakdown product of the drug or the drug itself, bound to an endogenous peptide, is presented by the class 1 HLA antigen on the hepatocyte cell membrane; this induces cytotoxic T-cell activation and subsequent hepatocyte death [16]. Ethnicity or genetic background may be a risk factor because of the variability in detoxification mechanisms (acetylator phenotype, human leukocyte antigen group) [17]. Our patient had a clear autoimmune disposition (mesangioproliferative glomerulonephritis and hypothireosis), and according to anamnestic, clinical, laboratory, imaging and other findings we estimated the existence of nitrofurantoin-induced, immune-mediated eosinophilic interstitial lung diseases, autoimmune hepatitis and several other multisystemic manifestations as lichen simplex chronicus and sicca syndrome, as well. There were no criterias for any diffuse connective tissue diseases, however it was possible that nitrofurantoin induced lupus-like syndrome associated with hepatitis [18]. Lung disease had subacute presentation with characteristic symptoms, clinical, X-ray, CT and PFTs findings. Lymphocytic-eosinophylic alveolitis was consistent with drug-induced reaction (DIR). Liver disease had chronic course. The positive ANA and ASMA results, hyper-gammaglobulinemia, histological features of liver biopsy and clinical response to immunosuppressive drugs were strongly suggestive of autoimmune hepatitis-type 1, triggered by nitrofurantoin. Definitive confirmation of DIR was positive rechallenge test according to WHO method [19]. Rechallenge, however, is not ethical due to severity of our patient`s clinical presentation. We applied the Naranjo algorithm for determination the likelihood of whether a DIR is actually due to the nitrofurantoin rather than the result of other factors and score was 6 – probable DIR. Initial treatment consists of drug withdrawal. In addition, we elected to use parenteral glucocorticosteroids because of the severe damage of lung and liver function. If glucocorticosteroid treatment fails, azathioprin may be introduced.

Conclusion

Adverse drug reactions should be considered in patients with concomitant lung and liver disease. The mainstay of treatment is drug withdrawal and the use of immunosuppressive drugs in severe cases. Consideration should be given to monitoring lung and liver function tests during a long term nitrofurantoin therapy.

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Received on December 21, 2010.
Revised on February 25, 2011.
Accepted on March, 2, 2011.

ISTORIJA MEDICINE

UDC: 61-05::616.12/091/

Dr Andreas Gruentzig – više od 30 godina blistave vizije lečenja koronarne bolesti

Dr Andreas Gruentzig – more than 30 years of the genius vision in therapy of coronary artery disease

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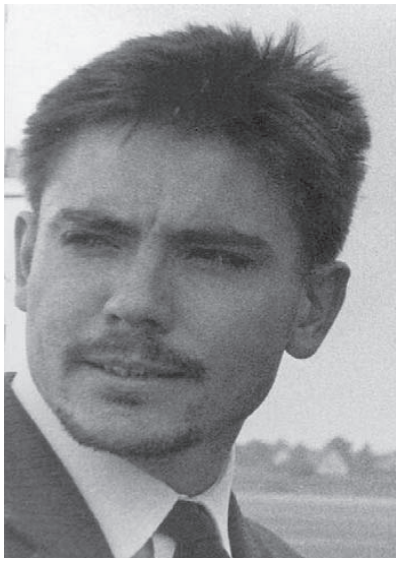
Ključne reči:
istorija medicine, XX vek; lekari; hirurgija, kardijalna, procedure; angioplastika, translumenska, perkutana, koronarna; lečenje, ishod.

Key words:
history, 20th century; physicians; cardiac surgical procedures; angioplasty, transluminal, percutaneous coronary; treatment outcome.

Uvod

Dr Andreas Gruentzig (slika 1) je genijalni lekar, koji je ostvario najveći uticaj na rađanje i ostvarenje ideje o perkutanoj translumenskoj koronarnoj angioplastici (*percutaneous trasluminal coronary angioplaty* – PTCA), kao začetka interventne koronarografije, i zlatnom standardu lečenja koronarne bolesti. Iako je od prve terapijske interventne procedure prošlo više od 30 godina, postulati koje je postavio dr Gruentzig tokom uvođenja i afirmacije te metode još uvek predstavljaju zlatna pravila interventne kardiologije.



Sl. 1 – Andreas Gruentzig

Put do ostvarenja cilja o kome je dr Gruentzig sanjao nije bio nimalo lak. Kao i većina ljudi koji su svojim izumima zadužili čovečanstvo, na početku svog rada osporavan je i kritikovan od strane tadašnjih vrhunskih kardiologa. Ipak, njegova genijalnost, upornost, predanost radu i vera u značaj sopstvenog istraživanja doveli su ga do najvećeg otkrića u kardiologiji u 20. veku, do PTCA koja je označila i početak nove ere u lečenju koronarne bolesti – ere interventne kardiologije.

Život u Drezdenu

Andreas Ronald Gruentzig rođen je 25. juna 1939. godine u Drezdenu, u istočnoj Nemačkoj. Roditelji, Wilmar i Charlotte, pripadali su malom krugu intelektualaca tog vremena – otac je bio doktor nauka u oblasti meteorologije, a majka učiteljica. Tokom rata otac je, kao vrsni meteorolog, regrutovan u stručni štab u Luftwaffa-e i poginuo je 1945. godine tokom završnih borbi za oslobođenje Berlina. Očeva ratna angažovanost u radu nemačke vazduhoplovne armije Luftwaffa-e je u posleratnom dobu bila velika prepreka u daljem školovanju Andreasa i njegovog starijeg brata Johana [1].

Charlotta Gruentzig je još od njihovih najranijih dana upućivala svoje sinove na to da vredno uče i radom se bore za ostvarivanje svog položaja u posleratnoj komunističkoj Nemačkoj. Teška vremena u kojima su odrastali u razorenoj Nemačkoj, koja je uz to nosila i teret glavnog krivca za Drugi svetski rat, naterali su porodicu Gruentzig da bude skromna, ali vrlo predana svojim obavezama, pa je, pored vrednog učenja, Andreas išao redovno i na časove klavira, što je u ono vreme bila retkost. Upravo godine najranijeg detinjstva provedene u siromaštvu,

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kao i odrastanje u Demokratskoj Republici Nemačkoj, sa komunističkom i prosovjetskom orijentacijom, uz teret oca poginulog na strani nemačke armije u Drugom svetskom ratu, ključno su uticale na formiranje karaktera Andreasa Gruentzig-a – postao je vredan, istrajan, posvećen svojim ciljevima i rešen da ih ostvari bez oslanjanja na pomoć, već isključivo na svoj rad.

Nekoliko godina posle završetka rata, Charlotte Gruentzig gonjena siromaštvom i lošim uspomenama, kao i veliki broj Nemaca, sa decom emigrira u Argentinu. Tamo se zadržala samo dve godine. Mučena nostalgijom i bez mogućnosti da se uklopi u način života u Novom svetu, porodica se vratila u Demokratsku Republiku Nemačku, u Lajpcig.

Univerzitet u Hajdelbergu

U školi se Andreas izdvajao po svom znanju i vrednom radu. Iako je završio Thomas gimnaziju u Lajpcigu kao najbolji u klasi, zbog političke pasivnosti u novom nemačkom poretku, nije dobio priliku da se upiše na fakultet, već mu je ponuđeno radno mesto u cementari. Osamnaestogodišnji Andreas nikako nije video sebe na mestu fabričkog radnika, te 1957. godine odlučuje da pređe u Zapadnu Nemačku. Odlazi u Istočni Berlin i seda u voz za Zapadni sektor, a zatim, mirno prešavši u Zapadnu berlinsku zonu, ulazi na teritoriju Zapadnog bloka koji od tog trena više nikada neće napustiti. Pridružuje se starijem bratu koji je već bio student Univerziteta u Hajdelbergu. Kako se veliki broj mladih ljudi koristio ovim trikom za miran prebeg iz Istočne Nemačke, Nemačka Demokratska Republika 1961. godine odlučuje da zaustavi odlazak svojih građana na zapad i gradi Berlinski zid [2].

U Hajdelbergu Andreas Gruentzig upisuje medicinu i dobija stipendiju. Upamćen je kao vrlo vredan student, posvećen predavanjima i još od studentskih dana vrlo zainteresovan za naučnoistraživački rad. Diplomirao je 1964. godine, u 25. godini. Nakon boravka u Hajdelbergu, 1969. godine, sa svojom verenicom Michela-om Seebrunner, koja je diplomirala psihologiju, bez mogućnosti za zaposlenje, zauvek napušta Nemačku i odlazi u Švajcarsku, u Cirih, gde se zapošljava na Odeljenju angiologije u Univerzitetskoj bolnici. U svojim memoarima, načelnik Odeljenja angiologije, dr Alfred Bollinger, opisuje predanost kliničkom i eksperimentalnom radu Andreasa Gruentzig-a, navodeći njegovu izjavu na početku rada: „Nameravam da svoj život posvetim vaskularnim bolestima“ [3].

Rađanje ideje o invazivnoj kardiologiji

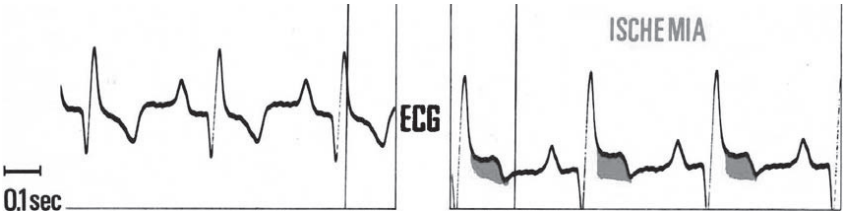
Uz podršku dr Bollinger-a, dr Gruentzig počinje sa eksperimentalnim radovima iz oblasti angiologije i periferne cirkulacije donjih ekstremiteta. Prvi rad objavljuje 1971. godine u časopisu *Angiology* pod nazivom „*Peripheral hemodynamics in patients with coarctation, normotensive and hypertensive arteriosclerosis obliterans of the lower limbs*“ [4, 5].

Prekretnica u radu dr Andreasa Gruentzig-a bila je 1971. godina, kada dobija priliku da poseti Aggertal kliniku u Engelskirchen-u, u Nemačkoj i upozna se sa dr Eberhart Zeitler-om, koji je prvi u Evropi počeo da koristi teleskopske dilatirajuće katetere za rešavanje tesnih stenoza u perifernim krvnim sudovima donjih ekstremiteta, po pionirskim istraživanjima dr Charles-a Dotter-a, iz Portlanda u Oregonu. Dr Gruentzig dolazi na ideju da istu tehniku primeni i na koronarnim arterijama, ali je ta zamisao u tom trenutku bila neostvarljiva jer tehničke karakteristike balona nisu odgovarale koronarnoj cirkulaciji. U Kantonalspital bolnici u Cirihu, otpočinje eksperimentali rad u kome mu je u eksperimentalnoj laboratoriji pomagala Maria Schlumpf, asistentkinja načelnika Angiologije dr Bollinger-a. Zabeleženo je da je skicu prototipa koronarnog katetera napravio prvi pijući kafu u restoranu na Gesnerallee, u blizini klinike, na jednoj od pauza.

Izrada adekvatnog katetera za prvu balon dilataciju koronarnih arterija trajala je nekoliko godina, zaokupivši potpuno dr Andreasa Gruntzig-a, koji je preneo celu eksperimentalnu laboratoriju u svoju kuhinju, da bi imao što više vremena da se posveti istraživanju. U rad je uključio i svoju suprugu, kao i Helmuth-a Schmidt-a, glavnog inženjera tada najpoznatije kompanije za radiološke igle „Hugo Schneider“. Najpre su uspeli da naprave prototip katetera sa jednim lumenom, a zatim i neophodan kateter sa dvostrukim lumenom.

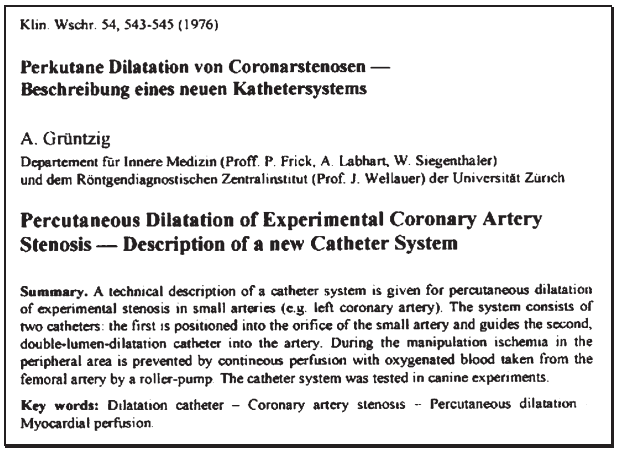
Novostvoreni jednolumenski polivinilski balon dr Gruentzig upotrebio je prvi put kod bolesnika sa suptotalnom stenozom arterije femoralis superficijalis 12. februara 1974. godine. Nakon toga, dr Andreas Gruentzig počeo je svoj eksperimentalni rad sa novim dvolumenskim kateterom, najpre na životinjama. Kateter sa dvostrukim lumenom, koji će kasnije biti upotrebljen za PTCA koronarnih arterija, prvi put je upotrebio za rešavanje stenoze ilijačne arterije, 23. janura 1975. godine. Novostvoreni kateter sa dvostrukim lumenom ubrzo je počeo komercijalno da se proizvodi u kompaniji „Cook and Schneider Companies“.

Svoje rezultate dr Gruentzig prikazao je na godišnjem skupu *American Heart Asociation* (AHA) 1976. godine, izazvavši veliko interesovanje izjavom da se sprema za lečenje stenoza u koronarnim arterijama (slika 2). Veliki broj renomiranih kardiologa skeptično je primio ta najnovija saznanja. Najveće interesovanje pokazao je dr Richard K. Myler iz Kalifornije (1980. godine osnovao je renomirani *San Francisco Heart Institute* u Kaliforniji), koji je odlučio da se odmah uputi u Cirih i na licu mesta uveri u opisane uspehe dr Gruentzig-a.



Sl. 2 – Deo originalnog EKG zapisa iz doktorske disertacije dr Andreasa Gruentzig-a, sa izazivanjem akutne okluzije cirkumfleksne koronarne arterije (Acx) podvezivanjem i posledičnom elevacijom ST segmenta (eksperiment na psu)

Svoja istraživanja iz oblasti dilatacije koronarnih arterija Dr Andreas Gruentzig prvi put je objavio u *Klinische Wochenschrift*, 1976. godine, u radu pod nazivom „*Percutaneous Dilatation of Experimental Coronary Artery Stenosis – Descriptio of a new Catheter System*“ (slika 3). Nakon toga, usledila je potraga za „idealnim bolesnikom“, koji će biti prikladan za novu tehniku i imati dovoljno entuzijazma da dâ pismeni informisani pristanak za novu proceduru.



Klin. Wschr. 54, 543-545 (1976)

Perkutane Dilatation von Coronarstenosen — Beschreibung eines neuen Kathetersystems

A. Grüntzig

Departement für Innere Medizin (Proff. P. Frick, A. Labhart, W. Siegenthaler) und dem Röntgendiagnostischen Zentralinstitut (Prof. J. Wellauer) der Universität Zürich

Percutaneous Dilatation of Experimental Coronary Artery Stenosis — Description of a new Catheter System

Summary. A technical description of a catheter system is given for percutaneous dilatation of experimental stenosis in small arteries (e.g. left coronary artery). The system consists of two catheters: the first is positioned into the orifice of the small artery and guides the second, double-lumen-dilatation catheter into the artery. During the manipulation ischemia in the peripheral area is prevented by contineous perfusion with oxygenated blood taken from the femoral artery by a roller-pump. The catheter system was tested in canine experiments.

Key words: Dilatation catheter – Coronary artery stenosis – Percutaneous dilatation Myocardial perfusion.

Sl. 3 – Naslovna strana rada dr Andreasa Gruentzig-a iz 1976. godine, u kome je prvi put opisao novu metodu lečenja koronarne bolesti – perkutanu translumensku koronarnu angioplastiku (PTCA)

Prva urađena perkutana translumenska koronarna angioplastika

Šesnaestog septembra 1977. godine, u Kantonspital bolnici u Cirihu, uz saglasnost i podršku dr Seninga, tadašnjeg šefa Kardiohirurgije, dr Andreas Gruentzig izveo je prvu koronarnu angioplastiku na živom bolesniku. Ta intervencija je označila istorijski pomak u lečenju koronarne bolesti i početak novog terapijskog pritstupa. I bolesnik, Adolf Bachman, i njegov lekar, dr Gruentzig, imali su po 38 godina. Bolesnik je imao simptome tipične za anginu na napor, a koronarografski nalaz je odgovarao jednosudovnoj bolesti sa stenozom od 85% u medijalnom delu leve prednje descendentne grane koronarne – *left anterior descending* (LAD) arterije. Sa kadriohirurške tačke gledišta aterosklerotična lezija bila je vrlo jednostavna i neko drugi na mestu dr Seninga mogao je insistirati da sam uradi relativno laku operaciju i na taj način spreči uvođenje nove terapijske metode. Tako je počela era saradnje kardiologa, kardiohirurga i vaskularnih hirurga za dobrobit bolesnika, koja i danas predstavlja ispravan multidisciplinarni put i omogućava napredak medicine na dobrobit bolesnika.

Nekoliko godina kasnije, 1984, dr Gruentzig je u jednom intervjuu opisao ovu proceduru: „Balon dilatacija stenoze je urađena bez ikakvih teškoća. Iako smo se strepeli, nije bilo ni ST elevacije, ventrikularne fibrilacije, pa čak ni jedne jedine ekstrasistole, a bolesnik je sve vreme bio bez bola u grudima. Uradio sam dve inflacije balona, a nakon toga se pritisak u distalnom delu koronarne arterije izjednačio sa pritiskom u aorti. Svi smo bili prijatno iznenađeni jednostavnošću intervencije“.

Prelazak u Sjedinjene Američke Države

Iako je otkriće dr Gruentzig-a, koje je promenilo sva dotadašnja shvatanja u lečenju koronarne bolesti, odjeknulo kao prasak u kardiološkoj naučnoj javnosti, uprava bolnice u kojoj je radio nije imala sluha za novu terapijsku metodu, dozvolivši mu da radi samo dve balon angioplastike nedeljno, što je on smatrao nedovoljnim brojem za afirmaciju i implementaciju nove metode. Nasuprot tome, naučni krugovi u SAD bili su vrlo zainteresovani za novu tehniku i ponudili su dr Gruentzig-u izvaredne uslove za prelazak u *Empory University Hospital*, u Atlantu, Džordžija. Najzad je dobio uslove za rad o kojima je maštao, uz obavezu da obučava interventne kardiologe sa teritorije SAD, za primenu nove metode.

Od svojih učenika zahtevao je da budu predani i posvećeni radu, sa pravilnim odabirom bolesnika. Kao neophodan uslov za uvođenje angioplastike u rad u nekom od centara postavio je kardiohirurišku *stand-by* podršku, u slučaju pojave proceduralnih komplikacija. Takođe, insistirao je na dugogodišnjem praćenju bolesnika i analizi uzroka komplikacija, posebno restenoza, želeći da spreči pogrešno tumačenje nove i tada još uvek neafirmisane tehnike, u koju je verovao celim svojim bićem.

U časopisu *Circulation*, 1983. godine, objavio je rad pod nazivom „*Should coronary arteries with less than 60% diameter stenosis be treated by angioplasty*?“ u kome navodi da: „PTCA u blagoj stenozi je vrlo uspešna, ali nosi povećan rizik od pojave komplikacija kao što su akutni infarkt miokarda i hitna operacija, a u nekim slučajevima čak može i ubrzati napredovanje koronarne bolesti”, zaključivši da PTCA ne treba raditi u stenozama manjim od 60% [6].

Godinu dana kasnije objavljuje svoja istraživanja na polju koronarne rezerve protoka (CFR) koju je merio tokom kateterizacije srca. Prvi put navodi značaj određivanja CFR kod stenoza manjih od 70%, navodeći da je „merenje CFR tokom kateterizacije srca vrlo važno ne samo za procenu značajnosti graničnih-intermedijalnih stenoza, već i za evaluaciju efikasnosti same intervencije“. Ovim ključnim razmatranjima procene značajnosti graničnih stenotičnih lezija postavio je osnovne kriterijume selekcije pacijenata za intervencije na koronarnim arterijama, koji se i danas poštuju [7].

Rad dr Gruentzig-a tragično je prekinut 27. oktobra 1985. godine, kada se avion kojim je pilotirao srušio u teško dostupnim planinskim predelima Srednjeg Zapada. Kao uzrok nesreće navedena je pilotska greška, iako njegov brat Johan, sumnjajući na sabotažu, nikada nije želeo da poveruje u to i zahtevao je dodatnu istragu, koja nije sprovedena.

Dr Gruentzig je za osam godina svog rada PTCA intervenciju uradio kod ukupno 169 bolesnika. Nakon njegove smrti, 1987. godine, u časopisu *New England Journal of Medicine* objavljen je rad Andresa Gruentzig-a i saradnika pod nazivom „*Long-term follow-up after percutaneous transluminal coronary angioplasty. The early Zurich experience*“ u kome su analizirane komplikacije i ishod intervencije kod

169 bolesnika kod kojih je dr Gruentzig uradio PTCA intervenciju. To su bili ujedno i rezultati prve analize uzroka restenoza kod do tada najveće grupe bolesnika. Autor je naveo: „Većina restenoza se javlja u prvih šest meseci posle angioplastike, mada se može javiti i znatno kasnije. Bolesnici sa jednosudovnom koronarnom bolešću imaju mnogo bolju prognozu u poređenju sa bolesnicima sa višesudovnom bolešću“ [8, 9].

Zaključak

Dr Andreas Gruentzig svojim radom postavio je temelje savremenom lečenju koronarne bolesti, osmislivši i realizujući ideju o PTCA. Njegova tragična smrt nije prekinula razvoj metode koju je izumeo. Sledbenici dr Gruentzig-a nastavili su da razvijaju PTCA, usavršavajući je uvođenjem stentova, danas široko prihvaćenih u terapiji koronarne bolesti. Dr Andreas Gruentzig je iza sebe ostavio potpuno novu viziju lečenja koronarne bolesti, koja je na početku bila nezamisliva za većinu vodećih kardiologa, a danas bi moderna kardiologija bila nezamisliva bez nje.

Zahvalnica

Autori se zahvaljuju dr Alexander-u Lembcke-u, načelniku Dijagnostičkog *imaging* centra Klinike „Charite“ u Berlinu, koji ih je tokom boravka u ovoj bolnici na obuci i u daljim kontaktima zainteresovao za život i delo dr Andreasa Gruentzig-a.

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Primljen: 22. II 2010.
Revidiran: 23. IV 2010.
Prihvaćen: 19. V 2010.

I N M E M O R I A M

dr vet. sci MILOMIR M. KUZMANOVIĆ pukovnik u penziji (1928–2012)

U Beogradu je 01. 03. 2012. godine preminuo puk. u penziji, dr sc vet. Milomir M. Kuzmanović, višegodišnji član kolektiva Vojnomedicinske akademije (VMA) u Beogradu, a zatim savetnik u Sektoru za odbrambene pripreme civilnih struktura društva Saveznog sekretarijata za narodnu odbranu (SSNO), sa koje dužnosti je i penzionisan.

Rođen je u selu Žabari, kraj Valjeva, u siromašnoj, ali nadasve časnoj i radnoj, seljačkoj porodici. Osnovnu školu i gimnaziju završio je u rodnom selu i Valjevu, a zatim vođen do kraja života neusahlom željom da stvara, nastavlja svoje obrazovanje kao vojni student na Veterinarskom fakultetu u Zagrebu. Studije na ovom fakultetu završava sa odličnim uspehom, te kao mlad sanitetski oficir, 1954. dobija prvi vojni i radni raspored u trupi, u Raškoj. Već na ovom prvom zadatku je pokazao svoje vanredne stručne, profesionalne i organizatorske sposobnosti, pa je nedugo zatim premešten u valjevski, zatim titogradski i najzad beogradski garnizon.

Nemirnog, ambicioznog i odlučnog duha, stalno radeći na svom stručnom usavršavanju, 1969. završava specijalističke studije na Veterinarskom fakultetu u Beogadu, na kome je 1975. odbranio doktorsku disertaciju pod naslovom „Uticaj gvanetidin sulfata na reprodukciju pacova“.

Radeći od 1964. u garnizonu grada Beograda, postavljen je na mesto upravnika Farme eksperimentalnih životinja Instituta za eksperimentalnu medicinu (sada Institut za medicinska istraživanja) VMA, kojom je rukovodio punih dvanaest godina. U ovoj ustanovi, njegov rad je, u svakom pogledu, više puta ocenjivan odličnim ocenama i istican je kao primer stručnosti, dobre organizacije i unapređenja rada kolektiva. Bez imalo preterivanja, može se reći da je bio dostojan reprezent svih progresivnih trendova kojima se VMA tih godina veoma brzo priključila svetski priznatim medicinskim istitucijama.

Objavio je veliki broj stručnih članaka, studija i prikaza u brojnim časopisima kao što su „Pozadina“, „Čovek i životna sredina“, „Privredni vjesnik“, a takođe je bio scenarista, koscenarista i autor teksta u filmskim ostvarenjima „Zaštita životinja u ratu i miru“, „RHB dekontaminacija“, u proizvodnji „Filmskih novosti“ iz Beograda. Bio je učesnik i organizator mnogih sručnih skupova širom bivše SFRJ sa tematikom zaštite čovekove okoline i civilne zaštite. Svoje znanje i iskustvo nesebično i strpljivo delio je sa kolegama i saradnicima, naročito mlađim, pa je 1987. imenovan za mentora u izradi jedne magistarske teze na Fakultetu narodne odbrane (sada Fakultet bezbednosti) u Beogradu.

Autor je poglavlja „Laboratorijske životinje“ u priručniku „Metodologija naučnoistraživačkog rada u medicinsko-biološkim naukama“, koje je zbog svog kvaliteta i praktične vrednosti, posle prvog, skromnog tiraža namenjenog potrebama VSU, štampana, između ostalog, i u izdanju „Medicinske knjige“, Beograd-Zagreb.

Autor je priručnika „Zaštita i spasavanje životinja, namirnica, hrane i vode“ koji obuhvata i sintetizuje ogromnu, na naučnim osnovama zasnovanu građu zaštite i spasavanja životinja, stočne hrane, hrane životinjskog porekla i vode. Priručnik namenjen civilnim i drugim društvenim struktura-

ma zaštite objavio je Vojnoizdavački i novinski centar iz Beograda.

Za doprinos struci i nauci, za stalno podizanje ugleda svog poziva, kao i ustanova u kojima je radio, odlikovan je sa četiri visoka mirnodopska ordena, među kojima je i Orden rada sa zlatnim vencem, a nosilac je i desetak medalja, pohvalnica i plaketa.

Bio je predan radnik, posvećen, istrajan i odlučan. Kao kolega i saradnik – širok i strpljiv, a kao čovek – nesebičan i plemenit, zbog čega je bio omiljen i poštovan među prijateljima i kolegama, a neizmerno voljen od strane svoje supruge i dvoje dece, kojima je posvetio svu svoju ljubav, svoj život i stvaralaštvo.

Neka mu je večna slava i hvala!

prof. dr Zvonko Magić,
načelnik Instituta za medicinska istraživanja,
Vojnomedicinska akademija

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Od 1. januara 2012. godine Vojnosanitetski pregled prešao je na e-Ur: Elektronsko uređivanje časopisa.

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Delovi rada su: **naslovna strana, apstrakt sa ključnim rečima, tekst** i **literatura.**

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Na drugoj stranici nalazi se strukturisani apstrakt sa naslovom rada. Kratkim rečenicama na srpskom i engleskom jeziku iznosi se **uvod i cilj** rada, osnovne procedure - **metode** (izbor ispitanika ili laboratorijskih životinja; metode posmatranja i analize), glavni nalazi - **rezultati** (konkretni podaci i njihova statistička značajnost) i glavni **zaključak**. Naglasiti nove i značajne aspekte studije ili zapažanja. Strukturisani apstrakt (**250** reči) ima podnaslove: *uvod/cilj, metode, rezultati* i *zaključak*. Za apstrakte na engleskom dozvoljeno je i do **450** reči. Strukturisani apstrakt je obavezan za metaanalize (istog obima kao i za originalne članke) i kazuistiku (do 150 reči, sa podnaslovima *uvod, prikaz slučaja i zaključak*). Ispod apstrakta, pod podnaslovom „Ključne reči" predložiti 3–10 ključnih reči ili kratkih izraza koji oslikavaju sadržinu članka.

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Tekst sadrži sledeća poglavlja: **uvod**, **metode**, **rezultate** i **diskusiju**. **Zaključak** može da bude posebno poglavlje ili se iznosi u poslednjem pasusu diskusije. U **uvodu** ponovo napisati naslov rada, bez navođenja autora. Navesti hipotezu (ukoliko je ima) i ciljeve rada. Ukratko izneti razloge za studiju ili posmatranje. Navesti samo strogo relevantne podatke iz literature i ne iznositi opširna razmatranja o predmetu rada, kao ni podatke ili zaključke iz rada o kome se izveštava.

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Rezultate prikazati logičkim redosledom u tekstu, tabelama i ilustracijama. U tekstu naglasiti ili sumirati samo značajna zapažanja.

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Primeri oblika referenci:

Đurović BM. Endothelial trauma in the surgery of cataract. Vojnosanit Pregl 2004; 61(5): 491–7. (Serbian)

Balint B. From the haemotherapy to the haemomodulation. Beograd: Zavod za udžbenike i nastavna sredstva; 2001. (Serbian)

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Christensen S, Oppacher F. An analysis of Koza's computational effort statistic for genetic programming. In: *Foster JA, Lutton E, Miller J, Ryan C, Tettamanzi AG,* editors. Genetic programming. EuroGP 2002: Proceedings of the 5th European Conference on Genetic Programming; 2002 Apr 3-5; Kinsdale, Ireland. Berlin: Springer; 2002. p. 182-91.

Abood S. Quality improvement initiative in nursing homes: the ANA acts in an advisory role. Am J Nurs [serial on the Internet]. 2002 Jun [cited 2002 Aug 12]; 102(6): [about 3 p.]. Available from: http://www.nursingworld.org/AJN/2002/june/Wawatch.htm

Tabele

Sve tabele štampaju se sa proredom 1,5 na posebnom listu hartije. Obeležavaju se arapskim brojevima, redosledom pojavljivanja, u desnom uglu (**Tabela 1**), a svakoj se daje kratak naslov. Objašnjenja se daju u fus-noti, ne u zaglavlju. Za fus-notu koristiti sledeće simbole ovim redosledom: ***, †, ‡, §, ||, ¶, **, ††, ...** Svaka tabela mora da se pomene u tekstu. Ako se koriste tuđi podaci, obavezno ih navesti kao i svaki drugi podatak iz literature.

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Parts of the manuscript are: **Title page; Abstract with key words; Text; References**.

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a) The title should be concise but informative. Subheadings should be avoided;

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The text of original articles is divided into sections with the headings: **Introduction**, **Methods**, **Results**, and **Discussion**. Long articles may need subheadings within some sections to clarify their content.

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Discussion is to emphasize the new and important aspects of the study and the conclusions that result from them. Relate the observations to other relevant studies. Link the conclusions with the goals of the study, but avoid unqualified statements and conclusions not completely supported by your data.

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Examples of references:

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): 657–612.

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Abood S. Quality improvement initiative in nursing homes: the ANA acts in an advisory role. Am J Nurs [serial on the Internet]. 2002 Jun [cited 2002 Aug 12]; 102(6): [about 3 p.]. Available from: http://www.nursingworld.org/AJN/2002/june/Wawatch.htm

Tables

Type each table double-spaced on a separate sheet. Number tables consecutively in the order of their first citation in the text in the upper right corner **(Table 1)** and supply a brief title for each. Place explanatory matter in footnotes, using the following symbols, in this sequence: ***, †, ‡, §, ||, ¶, **, ††, ...** Each table has to be mentioned in the text. If you use data from another source, acknowledge fully.

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Detailed Instructions are available at the web site:
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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 _____