

UDK 615 (497.11)

ISSN 0004-1963 (Štampano izd.)  
ISSN 2217-8767 (Online)

# ARHIV ZA FARMACIJU

Godina 68

Broj 2

Beograd, 2018.

## ČASOPIS SAVEZA FARMACEUTSKIH UDRUŽENJA SRBIJE

SPECIJALNI BROJ/SPECIAL ISSUE

VII Kongres farmaceuta Srbije sa međunarodnim učešćem

*Zajedno stvaramo budućnost farmacije*

Beograd, 10-14. oktobar 2018.

VII Serbian Congress of Pharmacy with international participation

*Creating the future of pharmacy together*

Belgrade, October 10-14, 2018

2/2018

# ARHIV ZA FARMACIJU

**ČASOPIS SAVEZA FARMACEUTSKIH UDRUŽENJA SRBIJE**  
**ARCHIVES DE PHARMACIE - ARCHIVES OF PHARMACY**

IZLAZI OD 1951. GODINE

## IZDAVAČ

SAVEZ FARMACEUTSKIH UDRUŽENJA SRBIJE

11000 Beograd, Bulevar vojvode Mišića 25, pošt. fah 664

tel/fax: + 381 11 2648 385; +381 11 2648 386

e-mail: [fps@sbb.rs](mailto:fps@sbb.rs); [sfus@farmacija.org](mailto:sfus@farmacija.org)

[www.farmacija.org](http://www.farmacija.org)

## IZDAVAČKI SAVET

Milana Dučić - Apoteka „Beograd”,

Sonja Kuštrin-Đorđević - Udruženje farmaceuta Beograda,

Ivana Miletić - Savez farmaceutskih udruženja Srbije,

Dubravka Urošev - Savez farmaceutskih udruženja Srbije,

Nenad Vulović - Udruženje farmaceuta Beograda

## UREDNUĆA ARHIVA

Marija Primorac

Univerzitet u Beogradu - Farmaceutski fakultet, Katedra za farmaceutsku tehnologiju i kozmetologiju

## ZAMENIK GLAVNOG UREDNIKA

Radica Stepanović-Petrović

Univerzitet u Beogradu - Farmaceutski fakultet, Katedra za farmakologiju

Sažeci radova nisu lektorisani

---

Radove objavljene u časopisu Arhiv za farmaciju indeksiraju: EMBASE i SCOPUS

---

ARHIV ZA FARMACIJU izlazi šest puta godišnje  
na sajtu Saveza farmaceutskih udruženja Srbije  
[www.farmacija.org](http://www.farmacija.org)

## **Naučni odbor/Scientific Committee**

### **Predsednik/President**

Jelena Paroјčić

### **Članovi/Members**

Danica Agbaba

Milica Atanacković Krstonošić

Erem Bilensov

Branka Brzaković

Silva Dobrić

Svetlana Ibrić

Vladimir Jakovljević

Vesna Spasojević Kalimanovska

Nada Kovačević

Tereza Kowalska

Gordana Leposavić

Vesna Matović

Branislava Miljković

Miroslav Savić

Svetlana Stojkov

Slađana Šobajić

Ljiljana Tasić

Radmila Veličković Radovanović

Andreas Zimmer

## **Reč gostujućeg urednika**

*Specijalni brojevi Arhiva za farmaciju 2/2018 i 3/2018 obuhvataju sažetke radova VII Kongresa farmaceuta Srbije sa međunarodnim učešćem koji se održava u Beogradu od 10. do 14. oktobra 2018. godine. Naučni program Kongresa obuhvatačetiri plenarna predavanja, 86 predavanja po pozivu, 48 usmenih saopštenja i 214 poster prezentacija iz različitih oblasti farmaceutskih nauka.*

*Vođeni sloganom „Zajedno stvaramo budućnost farmacije”, članovi Naučnog odbora osmislili su program koji obuhvata raznolike teme, počev od najnovijih dostignuća u oblasti istraživanja i otkrića lekova, do njihove efikasne i bezbedne primene kod različitih populacija pacijenata, s ciljem da zajedno potražimo odgovore na pitanja kako vidimo budućnost farmacije i kako da se, kao profesija i kao pojedinci, pripremimo za izazove koji nam predstoje.*

*Imajući u vidu zdravstvene izazove koji se javljaju zbog produženja životnog veka, obuhvaćene su teme koje se odnose na zdravo starenje i napredne pristupe u terapiji hroničnih bolesti, uključujući psihijatrijska i neurološka, autoimunska i maligna oboljenja i metabolički sindrom; takođe su predstavljeni inovativni pristupi, materijali i tehnologije koji se koriste u razvoju, proizvodnji i kontroli kvaliteta lekova, analiza koristi i rizika koji se odnose na primenu dijetetskih suplemenata, uticaj zagađivača životne sredine na zdravlje i zabrinjavajući trend porasta broja trovanja psihoaktivnim supstancama; sagledane su potrebe i mogućnosti za razvoj i implementaciju novih kliničkih usluga farmaceuta kao osnov za unapređenje zdravstvenih ishoda, kao i novi pristupi u stručnom osposobljavanju i kontinuiranom profesionalnom usavršavanju farmaceuta.*

*U okviru poster sesija predstavljeni su rezultati istraživanja iz različitih oblasti farmacije sprovedenih u okviru apoteka/bolničkih apoteka, farmaceutske industrije, regulatornih tela i univerziteta, kako u Srbiji, tako i u inostranstvu.*

*Zahvaljujem se svim predavačima i autorima poster prezentacija na njihovom doprinosu, uz čvrsto verovanje da su ovi specijalni brojevi Arhiva za farmaciju odraz kreativnosti, profesionalnih kompetencija, snage i značajnog naučnoistraživačkog kapaciteta farmaceuta u Srbiji i regionu.*

*Jelena Parožić  
predsednik Naučnog odbora  
VII Kongresa farmaceuta Srbije*

## **Guest Editor's Preface**

*Special issues 2/2018 and 3/2018 of the Archives of Pharmacy include abstracts from the VII Serbian Congress of Pharmacy which is held in Belgrade from October 10 to 14, 2018. The scientific programme includes four plenary lectures, 86 invited lectures, 48 oral presentations and 213 poster presentations from various fields of pharmaceutical sciences.*

*Guided by the slogan „Creating the future of pharmacy together”, Scientific Committee members have built the versatile programme covering topics ranging from the latest findings in drug discovery and development, to effective and safe drug use in diverse patient populations, with the aim to explore together the vision for the future of pharmacy and how to best prepare, as profession, as well as the individuals, for the upcoming challenges.*

*Having in mind the healthcare challenges caused by longevity extension, topics related to healthy aging and advanced treatments of chronic diseases, including psychiatric and neurological disorders, autoimmune and malignant diseases, and metabolic syndrome are included; furthermore, novel approaches, materials and technologies used in drug development, manufacture and quality control are presented, as well as the benefit risk analysis of dietary supplements consumption, the health effects of environmental pollutants, and increased number of poisoning caused by psychoactive substances abuse; the needs and possibilities for development and implementation of new clinical pharmacy services are considered as a basis for health outcomes improvement, as well as the increasing requirements for qualification and continuing professional development of pharmacists.*

*Poster sessions included the research outcomes from different areas of pharmacy, conducted within the community and hospital pharmacies, pharmaceutical industry, regulatory authorities and academia.*

*I would like to acknowledge the contribution of all the authors with firm belief that these special issues of the Archives of Pharmacy reflect the creativity, professional competencies, strengths and considerable research capacity of pharmacist in Serbia and the region.*

*Jelena Parožić  
president of the Scientific Committee  
of the VII Serbian Congress of Pharmacy*

**Usmena izlaganja**

**Oral Presentations**

## SADRŽAJ – CONTENTS

US 1

**FARMAKOTERAPIJSKI PROBLEMI I INTERVENCIJE FARMACEUTA U  
BOLNICI ZA MEDICINSKU REHABILITACIJU**

**DRUG RELATED PROBLEMS AND INTERVENTIONS OF PHARMACISTS  
IN A PHYSICAL REHABILITATION HOSPITAL**

- **Gordana Ljubojević** 269

US 2

**POTENCIJALNE INTERACIJE IZMEĐU LEKOVA U TERAPIJI NA  
OTPUSTU KOD GERIJATRIJSKIH PACIJENATA**

**POTENTIAL DRUG-DRUG INTERACTION IN GERIATRIC PATIENTS AT  
DISCHARGE**

- **Ivana Baralić, Vesna Janković, Tatjana Savković,  
Mirjana Stanojlović, Višnja Glišić, Goran Marković, Sonja Sim** 271

US 3

**UNAPREĐENJE KOMPETENCIJA STUDENATA FARMACIJE**

**IMPROVING THE COMPETENCIES OF PHARMACY STUDENTS**

- **Dragana Jocić, Ljiljana Tasić, Andrijana Milošević Georgijev,  
Dušanka Krajanović, Valentina Marinković** 273

US 4

**B.Cell: INTERAKTIVNA EDUKACIJA I EDUKATIVNA INTERAKCIJA**

**B.Cell: INTERACTIVE EDUCATION & EDUCATIVE INTERACTION**

- **Neda Trivić, Milica Puđa, Tamara Kovačević** 275

US 5

**UTICAJ PRAVILNIKA KOJIM SE REGULIŠU MAKSIMALNE  
VELEPRODAJNE CIJENE LIJEKOVA NA CIJENE LIJEKOVA U BOSNI I  
HERCEGOVINI**

**INFLUENCE OF THE RULEBOOK FOR REGULATING MAXIMUM  
WHOLESALE PRICES ON MEDICINE COST IN BOSNIA AND  
HERCEGOVINA**

- **Biljana Tubić, Jelena Aničić, Tijana Spasojević,  
Ana Cvijanović, Aleksandar Zolak** 277

US 6

**OD TRADICIONALNE MEDICINE DO RACIONALNE FITOTERAPIJE – 50  
GODINA FARMACEUTSKE SLUŽBE U OKVIRU BILJNE APOTEKE  
INSTITUTA ZA PROUČAVANJE LEKOVITOG BILJA „DR JOSIF PANČIĆ“  
FROM TRADITIONAL MEDICINE TO RATIONAL PHYTOTHERAPY – 50  
YEARS OF PHARMACEUTICAL PRACTICE WITHIN THE HERBAL  
PHARMACY OF THE INSTITUTE FOR MEDICINAL PLANTS RESEARCH  
„DR JOSIF PANČIĆ“**

- [Nebojša Menković](#)

**279**

US 7

**APOTEKAR I JAVNO-ZDRAVSTVENI PROSVETITELJ MR PH MILIVOJE  
MOLJAC: PEČAT U VREMENU**

**PHARMACIST AND PUBLIC HEALTH EDUCATOR MILIVOJE MOLJAC:  
SEAL IN TIME**

- [Stevan Vukov](#)

**281**

US 8

**HOMEOPATHY 222 YEARS AFTER – THE HISTORICAL KNOWLEDGE  
AND VIEWS OF SAMUEL HAHNEMANN IN CONTEXT OF HIS WRITINGS**

- [Łukasz Komsta](#)

**283**

US 9

**KRATAK ISTORIJSKI PRIKAZ MAGISTRALNIH LEKOVA KOJI SE  
PRIMENJUJU KOD OPSTIPACIJE**

**SHORT HISTORICAL OVERVIEW OF EXTEMPORANEOUSLY  
COMPOUNDED MEDICINES FOR CONSTIPATION**

- [Ilinka Vuković, Mirjana Gajdaš, Dušanka Krajnović](#)

**284**

US 10

**FALSIFIKOVANI LEKOVI - IZAZOVI REGULATIVE U SPROVOĐENJU I  
PROMOCIJI BEZBEDNE UPOTREBE LEKOVA**

**FALSIFIED MEDICINES - REGULATORY CHALLENGES IN SAFE DRUG  
USE AND ITS PROMOTION**

- [Pavle Zelić, Saša Jačović](#)

**286**

US 11

**DEFEKT KVALITETA LEKA - REGULATORNI ZAHTEVI I SAVREMENI TRENDovi**

**MEDICINE QUALITY DEFECT - REGULATORY REQUIREMENTS AND CURRENT TRENDS**

- **Svetlana Mihaljica, Žarko Jović, Boris Bojić,  
Marija Malešević, Gordana Pejović**

**288**

US 12

**SPECIFIČNOSTI FARMAKOVIGILANCE BIOTEHNOLOŠKIH LEKOVA  
SPECIFICITIES OF PHARMACOVIGILANCE OF BIOTECHNOLOGICAL MEDICINES**

- **Milena Miljković, Cvijeta Bielen, Aleksandra Pajić**

**290**

US 13

**ULOГA FARMAKOGENETIKE U PERSONALIZOVANOJ TERAPIJI KOD PACIJENATA SA TRANSPLANTIRANIM BUBREGOM NA TAKROLIMUS-BAZIRANOJ IMUNOSUPRESIJI**

**THE ROLE OF PHARMACOGENETICS IN PERSONALIZED THERAPY OF RENAL TRANSPLANT RECIPIENTS ON TACROLIMUS-BASED IMMUNOSUPPRESSION**

- **Nikola Stefanović, Radmila Veličković-Radovanović,  
Katarina Dinić, Tatjana Cvetković**

**292**

US 14

**ULOГA I ZNAЧAJ MITOHONDRIJALNIH MARKERA APOTOZE U TERAPIJI KARCINOMA KOLONA**

**THE ROLE AND SIGNIFICANCE OF MITOCHONDRIAL MARKERS OF APOPTOSIS IN COLON CANCER TREATMENT**

- **Ivana Damnjanović, Gordana Kocić, Stevo Najman,  
Sanja Stojanović, Andrej Veljković, Srđan Pešić**

**294**

US 15

**INTERAKCIJE ERLOTINIBA U TERAPIJI ONKOLOŠKIH BOLESNIKA NA KLINICI ZA PLUĆNE BOLESTI**

**INTERACTIONS OF ERLOTINIB IN THE TREATMENT OF ONCOLOGIC PATIENTS AT THE LUNG DISEASE CLINIC**

- **Tijana Kovačević, Branislava Miljković,**

**Sandra Vezmar-Kovačević, Mirko Stanetić, Peđa Kovačević**

**296**

US 16

**POTENCIJALNE INTERAKCIJE LEKOVA KOD PACIJENATA SA HIPERTENZIJOM**

**POTENTIAL DRUG INTERACTIONS IN HYPERTENSIVE PATIENTS**

- **Zorica Cvetković, Aneta Perić, Ana Udilović**

**298**

US 17

**POVRATAK U BUDUĆNOST: KAKO RACIONALIZOVATI BOLNIČKU POTROŠNJU ANTIBIOTIKA?**

**BACK TO THE FUTURE: HOW TO OPTIMIZE HOSPITAL ANTIBIOTIC CONSUMPTION?**

- **Aneta Perić, Bojana Milenković, Jelena Brusić-Renaud,**

**Mirjana Antunović, Vesna Šuljagić**

**300**

US 18

**TROVANJA OLANZAPINOM U NACIONALNOM CENTRU ZA KONTROLU TROVANJA SRBIJE U 2017. GODINI**

**OLANZAPINE INTOXICATIONS IN NATIONAL POISON CONTROL CENTER OF SERBIA IN 2017**

- **Snežana Đorđević, Vesna Kilibarda, Gordana Brajković,**

**Nataša Perković Vukčević, Gordana Vuković Ercegović,**

**Jasmina Jović Stošić, Slavica Vučinić**

**302**

US 19

**EFIKASNOST OMEGA-3 MASNIH KISELINA U PREVENCIJI KARDIOVASKULARNIH BOLESTI: DOKAZI I PREPORUKE**

**EFFECTIVENESS OF OMEGA-3 FATTY ACIDS IN PREVENTION OF CARDIOVASCULAR DISEASES: EVIDENCE AND RECOMMENDATIONS**

- **Silva Dobrić**

**304**

US 20	KORISTI I RIZICI UPOTREBE DIJETETSKIH SUPLEMENATA BENEFITS AND RISKS OF DIETARY SUPPLEMENTS USE	
- Davor J. Korčok, Bogdan Mitić		306
US 21	DIJETETSKI SUPLEMENTI SA VITAMINIMA I MINERALIMA NA TRŽIŠTU SRBIJE VITAMIN AND MINERAL SUPPLEMENTS ON THE SERBIAN MARKET	
- Bojana Vidović, Mirko Lazović, Lana Kostić, Slađana Šobajić		308
US 22	ISPITIVANJE BRZINE RASTVARANJA RESVERATROLA IZ DIJETETSKIH SUPLEMENATA ANALYSIS OF RESVERATROL DISSOLUTION FROM DIETARY SUPPLEMENTS	
- Mira Mikulić, Ljilja Torović, Milica Atanacković Krstonošić, Veljko Ćućuz, Jelena Hogervorst		310
US 23	ISPITIVANJE ANTIMIKROBNOG POTENCIJALA ETARSKOG ULJA HERBE CRVENOG ZDRAVCA ( <i>GERANIUM ROBERTIANUM L.</i> ) EXPLORATION OF ANTIMICROBIAL POTENTIAL OF THE ESSENTIAL OIL FROM AERIAL PARTS OF <i>GERANIUM ROBERTIANUM L.</i>	
- Jelena Antić Stanković, Nikola Krstić, Nikola Jakovljević, Dubravka Bigović		312
US 24	EFFECT OF EXENATIDE LAR IN TYPE-2 DIABETIC PATIENTS WITH VS. WITHOUT ELEVATED ADIPO-INFLAMMATORY RISK SCORE AT BASELINE: AN 8-MONTH PROSPECTIVE INTERVENTION STUDY	
- Dragana Nikolić, Roberta Chianetta, Giuseppe Castellino, Jelena Vekić, Aleksandra Zeljković, Vesna Spasojević-Kalimanovska, Manfredi Rizzo		314

US 25

**POVEZANOST IZMEĐU OBRAZACA HOMEOSTAZE HOLESTEROLA I KONCENTRACIJA NE-HDL HOLESTEROLA KOD ZDRAVIH OSOBA I PACIJENATA SA ISHEMIJSKOM BOLEŠĆU SRCA KOJI NISU NA TERAPIJI STATINIMA**

**ASSOCIATION BETWEEN CHOLESTEROL HOMEOSTASIS PATTERNS AND NON-HDL CHOLESTEROL IN HEALTHY PEOPLE AND NON-STATIN TREATED CORONARY ARTERY DISEASE PATIENTS**

- **Tamara Gojković, Sandra Vladimirov,  
Vesna Spasojević-Kalimanovska, Aleksandra Zeljković,  
Jelena Vekić, Zorana Jelić-Ivanović**

**315**

US 26

**LONG TERM EFFECTS OF LIRAGLUTIDE ON GLYCO-METABOLIC PARAMETERS AND CIMT IN PATIENTS WITH TYPE-2 DIABETES: 5 YEARS PROSPECTIVE REAL-WORLD STUDY**

- **Giuseppa Castellino, Dragana Nikolic,  
Roberta Chianetta, Jelena Vekic, Aleksandra Zeljkovic,  
Vesna Spasojevic-Kalimanovska, Manfredi Rizzo**

**317**

US 27

**INTERAKCIJA REZISTINA I CAP-1 RECEPTORA SA HDL-HOLESTEROLOM KOD PACIJENATA SA KOLOREKTALNIM KANCEROM**

**INTERACTION OF RESISTIN AND CAP1 RECEPTOR WITH HDL-CHOLESTEROL IN COLORECTAL CANCER PATIENTS**

- **Marija Mihajlović, Ana Ninić, Miron Sopić,  
Milica Miljković, Aleksandra Stefanović,  
Vesna Spasojević-Kalimanovska, Aleksandra Zeljković**

**318**

US 28

**POTENCIJALNI RAZLOZI ZA SMANJENJE ANTIOKSIDATIVNE AKTIVNOSTI HDL ČESTICA KOD PACIJENATA NA HEMODIJALIZI**

**POTENTIAL REASONS FOR DECREASED ANTIOXIDATIVE ACTIVITY OF HDL PARTICLES IN HEMODIALYSIS PATIENTS**

- **Milica Miljković, Aleksandra Stefanović, Jelena Vekić,  
Sanja Simić-Ogrizović, Vesna Spasojević-Kalimanovska,  
Jelena Kotur-Stevuljević**

**320**

US 29

**METABOLIČKI ZDRAVA GOJAZNOST I RIZIK ZA RAZVOJ  
KARDIOVASKULARNIH BOLESTI**

**METABOLICALLY HEALTHY OBESITY AND CARDIOVASCULAR  
DISEASE RISK**

- [Sanja Vujčić, Jelena Vekić, Aleksandra Zeljković,  
Lidija Memon, Nataša Bogavac-Stanojević,  
Vesna Spasojević-Kalimanovska](#)

322

US 30

**ISPITIVANJA NA PACOVSKOM VALPROATNOM MODELU AUTIZMA  
OTKRIVAJU POZITIVNU MODULACIJU ALFA5GABAA RECEPTORA  
KAO MOGUĆI NOVI TERAPIJSKI PRISTUP**

**THE VALPROATE RAT MODEL REVEALS POSITIVE MODULATION OF  
ALPHA5GABAA RECEPTORS AS A NOVEL TARGET FOR TREATMENT  
OF AUTISM SPECTRUM DISORDER**

- [Anja Santrač, Marija Banićević, Jovana Aranđelović,  
Bojan Marković, Guanguan Li, James Cook, Miroslav Savić](#)

324

US 31

**DISULFIRAM – POTENCIJALNE TERAPIJSKE PRIMENE STAROG LEKA  
DISULFIRAM – POTENTIAL THERAPEUTIC APPLICATIONS  
OF AN OLD DRUG**

- [Ana Đurić, Mirjana M. Đukić](#)

326

US 32

**UTICAJ VINIFIKACIJE NA SADRŽAJ FENOLNIH KISELINA I  
ANTIOKSIDATIVNE OSOBINE VINA OD ARONIJE**

**VINIFICATION INFLUENCE ON PHENOLIC ACID CONTENT AND  
ANTIOXIDANT PROPERTIES OF BLACK CHOKEBERRY WINE**

- [Uroš Čakar, Aleksandar Petrović, Vlatka Vajs, Brižita Đorđević](#) 328

US 33

**ISPITIVANJE RETENCIONOG PONAŠANJA ODABRANIH LIGANADA IMIDAZOLINSKIH RECEPTORA U REVERZNO-FAZNOJ I TEĆNOJ HROMATOGRAFIJI HIDROFILNIH INTERAKCIJA**

**INVESTIGATION OF RETENTION BEHAVIOUR OF SELECTED IMIDAZOLINE RECEPTOR LIGANDS IN REVERSED-PHASE AND HYDROPHILIC INTERACTION LIQUID CHROMATOGRAPHY**

- **Darija Obradović, Slavica Oljačić, Danica Agbaba**

**330**

US 34

**PRAĆENJE NIVOA OLOPATADINA U HUMANIM SUZAMA HILIC-ESI/MS/MS METODOM**

**MONITORING OF OLOPATADINE LEVEL IN HUMAN TEARS BY HILIC-ESI/MS/MS METHOD**

- **Jelena Maksić, Ana Stajić, Miroslav Knežević,  
Bojana Dačić Krnjaja, Biljana Jančić-Stojanović,  
Mirjana Medenica**

**332**

US 35

**PRIMENA HPLC METODE U ODREĐIVANJU KONSTANTI STABILNOSTI KOMPLEKSA  $\beta$ -CIKLODEKSTRINA SA ODABRANIM ANTIPSIHOTICIMA**

**APPLICATION OF HPLC METHOD IN DETERMINING THE COMPLEX STABILITY CONSTANTS BETWEEN  $\beta$ -CYCLODEXTRIN AND SELECTED ANTIPSYCHOTICS**

- **Nevena Maljurić, Biljana Otašević,  
Jovana Krmar, Mira Zečević, Ana Protić**

**334**

US 36

**KVANTIFIKOVANJE VEZE STRUKTURE ARIPIPRAZOLA I SRODNIH NEČISTOĆA SA GENERISANIM ESI ODGOVOROM PRIMENOM METODA MAŠINSKOG UČENJA**

**QUANTITATIVE STRUCTURE – PROPERTY RELATIONSHIP MODELING OF ESI RESPONSE OF ARIPIPRAZOLE AND ITS IMPURITIES USING MACHINE LEARNING METHODS**

- **Jovana Krmar, Ljiljana Tolić, Tatjana Đurkić, Ana Protić,  
Nevena Maljurić, Mira Zečević, Biljana Otašević**

**336**

US 37

**SIMULACIJE MOLEKULSKE DINAMIKE I VIRTUAL SCREENING  
STUDIJA INHIBITORA SIRTUINA 2**

**MOLECULAR DYNAMICS-BASED VIRTUAL SCREENING OF SIRTUIN 2  
INHIBITORS**

- **Nemanja Đoković, Katarina Nikolić,  
Danica Agbaba, Maija Lahtela-Kakkonen**

**338**

US 38

**ISPITIVANJE FIZIČKO-HEMIJSKIH SVOJSTAVA SMEŠA POLIMERA I  
POVRŠINSKI AKTIVNIH MATERIJA KAO POTENCIJALNIH NOSAČA  
LEKOVA**

**PHYSICO-CHEMICAL EVALUATION OF POLYMER-SURFACTANT  
MIXTURES AS POTENTIAL DRUG DELIVERY SYSTEMS**

- **Maja Milanović, Veljko Krstonošić,  
Milica Atanacković Krstonošić**

**340**

US 39

**ACE I  $\alpha$ -GLUKOZIDAZNA INHIBITORNA AKTIVNOST METANOLNOG  
EKSTRAKTA *ALCHEMILLA VIRIDIFLORA* ROTHM. (ROSACEAE)**

**ACE AND  $\alpha$ -GLUCOSIDASE INHIBITORY ACTIVITY OF METHANOL  
EXTRACT OF *ALCHEMILLA VIRIDIFLORA* ROTHM. (ROSACEAE)**

- **Jelena Radović, Nada Grozdanić, Tatjana Stanojković,  
Relja Suručić, Tatjana Kundaković-Vasović**

**342**

US 40

**PROCENA BEZBEDNOSNOG PROFILA ETARSKIH ULJA TAKSONA  
RODA *HERACLEUM* L. (APIACEAE) U ODNOSU NA UTVRĐENI SADRŽAJ  
FURANOKUMARINA**

**EVALUATION OF SAFETY PROFILE OF THE ESSENTIAL OILS OF  
*HERACLEUM* L. TAXA (APIACEAE) RELATED TO DETERMINED  
FURANOCOUMARIN CONTENT**

- **Ljuboš Ušjak, Silvana Petrović**

**344**

US 41

**ISPITIVANJE POGODNOSTI GRANULATA OBLOŽENOG TOPLJENJEM  
ZA KOMPRIMOVANJE U TABLETE DEFINISANE DEBLJINE**

**ASSESSING THE ABILITY OF HOT MELT COATED GRANULES TO  
PRODUCE TABLETS OF CONTROLLED THICKNESS**

- **Ana Milanović, Marija Bujišić, Katarina Drezgić,  
Jovana Drobnjak, Ivana Aleksić, Sandra Cvijić**

**346**

US 42

**FORMULACIJA I OPTIMIZACIJA ORALNO-DISPERZIBILNIH TABLETA  
IZRAĐENIH DIREKTNOM KOMPRESIJOM SA VISOKIM UDELOM  
AKTIVNE SUPSTANCE**

**FORMULATION AND OPTIMIZATION OF ORODISPERSIBLE TABLETS  
CONTAINING HIGH DRUG LOAD PREPARED BY DIRECT COMPRESSION**

- **Milica Drašković, Erna Turković, Jelena Đuriš, Jelena Parojević** **348**

US 43

**SUPERKRITIČNA IMPREGNACIJA TABLETA MIKROKRISTALNE  
CELULOZE IBUPROFENOM**

**SUPERCritical IMPREGNATION OF MICROCRYSTALLINE  
CELLULOSE TABLETS WITH IBUPROFEN**

- **Jovana Potpara, Jasna Ivanović, Svetlana Ibrić**

**350**

US 44

**UTICAJ SADRŽAJA LEKA NA DINAMIKU MEĐUPOVRŠINSKOG SLOJA  
NISKOENERGETSKIH NANOEMULZIJA – STUDIJA SA KURKUMINOM**

**DRUG LOADING INFLUENCE ON THE INTERFACIAL MEMBRANE  
DYNAMICS OF THE LOW-ENERGY NANOEMULSIONS -A CURCUMIN  
CASE STUDY**

- **Ines Nikolić, Evgenia Mitsou, Dominique Jasmin Lunter,  
Vassiliki Papadimitriou, Aristotelis Xenakis,  
Rolf Daniels, Snežana Savić**

**352**

US 45

**BIOKOMPATIBILNE NANOEMULZIJE ZA ISPORUKU ACEKLOFENAKA U/KROŽ KOŽU PRIMENOM HEMIJSKIH POJAČIVAČA PENETRACIJE I ČVRSTIH MIKROIGALA**

**BIOCOMPATIBLE NANOEMULSIONS FOR ACECLOFENAC DELIVERY INTO/THROUGH THE SKIN USING CHEMICAL PENETRATION ENHANCERS AND SOLID MICRONEEDLES**

- **Tanja Ilić, Sanela Savić, Bojan Batinić, Jelena Đoković**

**354**

US 46

**ISPITIVANJE VARIJABILNOSTI U KONCENTRACIJAMA METOTREKSATA IZMEĐU CIKLUSA TERAPIJE**

**INVESTIGATION OF VARIABILITY IN METHOTREXATE CONCENTRATIONS BETWEEN THERAPY CYCLES**

- **Biljana Škorić, Marija Jovanović, Branislava Miljković, Miloš Kuzmanović, Dragan Mićić, Katarina Vučićević**

**356**

US 47

**OPTIMIZACIJA PROTOKOLA ZA SAKUPLJANJE UZORAKA KRVI ZA ISPITIVANJE FARMAKOKINETIKE ZONISAMIDA KOD PEDIJATRIJSKIH PACIJENATA**

**OPTIMIZATION OF THE BLOOD SAMPLING PROTOCOL FOR THE ZONISAMIDE PHARMACOKINETIC STUDY IN PEDIATRIC PATIENTS**

- **Maša Roganović, Branislava Miljković, Marija Jovanović, Andelija Malenović, Nebojša Jović, Katarina Vučićević**

**358**

US 48

**ISPITIVANJE UTICAJA FUNKCIONALNOG VOLUMENA ŠTITASTE ŽLIJEZDE NA VJEROVATNOĆU ISHODA TERAPIJE  $^{131}I$  KOD PACIJENATA SA BENIGNIM OBOLJENJIMA ŠTITASTE ŽLIJEZDE**

**INVESTIGATION OF THE INFLUENCE OF FUNCTIONAL THYROID VOLUME ON THE PROBABILITY OF  $^{131}I$  THERAPY OUTCOME IN PATIENTS WITH BENIGN THYROID DISEASE**

- **Valentina Topić Vučenović, Dijana Jelić, Zvezdana Rajkovača, Branislava Miljković, Katarina Vučićević**

**360**

## FARMAKOTERAPIJSKI PROBLEMI I INTERVENCIJE FARMACEUTA U BOLNICI ZA MEDICINSKU REHABILITACIJU

**Gordana Ljubojević**

Zavod za fizikalnu medicinu i rehabilitaciju „Dr Miroslav Zotović”, Banja Luka  
(Bosna i Hercegovina)

Aktivnosti bolničkih farmaceuta značajno su se promijenile u poslednjih 30 godina, sa većim naglaskom na klinički rad i farmaceutsku zdravstvenu zaštitu. Cilj ovog istraživanja bio je ustanoviti farmakoterapijske i logističke probleme, farmaceutske intervencije i ishode tih intervencija tokom svakodnevnog rutinskog rada farmaceuta u bolnici za medicinsku rehabilitaciju.

Sistematično, prospektivno, opservaciono istraživanje provedeno je tokom 2,5 godine u Zavodu za fizikalnu medicinu i rehabilitaciju, Banjaluka, Bosna i Hercegovina, sa 594 bolnička kreveta. Tri bolnička farmaceuta pružala su konsultacije u vezi sa farmakoterapijskim i/ili logističkim problemima, a na lični ili telefonski upit doktora, medicinskih sestara/tehničara, fizioterapeuta i pacijenata. Glavne mjere ishoda bile su vrsta farmakoterapijskog ili logističkog problema, pokretač konsultacije, ishod i vrsta farmaceutske intervencije, te vrijeme utrošeno za rješavanje problema. Farmaceuti su imali pristup medicinskoj dokumentaciji, premda nisu bili rutinskih prisutni na odjeljenjima. Od 1515 farmaceutskih intervencija, 48,8% odnosilo se na rješavanje farmakoterapijskih problema, od kojih su najzastupljeniji bili preporuka za izbor ili doziranje lijeka i potreba za dodatnim informacijama o lijekovima. Cijena i mogućnost nabavke lijeka bili su najčešći logistički problemi. Više su vremena iziskivale farmakoterapijske konsultacije, koje su najčešće inicirali doktori (Mann-Whitney U test,  $p \leq 0,001$ ), nego rješavanje logističkih problema. Intervencije na rješavanju farmakoterapijskih problema bile su slabije prihvaćene (83,7%) u odnosu na logističke intervencije (95,2%;  $p \leq 0,00$ ). Prosječan broj identifikovanih farmakoterapijskih problema po pacijentu (1,37) niži je u poređenju sa rezultatima sličnih istraživanja (2,6-6,5), a kako farmaceuti (0,51/100 bolničkih kreveta) nisu rutinski radili reviziju farmakoterapije to može biti obrazloženje za samo 19% farmakoterapijskih problema identifikovanih od strane farmaceuta.

Bolnički farmaceuti su se tokom rutinskog rada bavili približno istim brojem farmakoterapijskih i logističkih problema. Ukupna stopa prihvaćenih farmaceutskih intervencija bila je visoka, te naši rezultati ukazuju na potrebu za većim angažmanom farmaceuta u kliničkim aktivnostima, ali i za većim brojem bolničkih farmaceuta u Bosni i Hercegovini.

## **DRUG RELATED PROBLEMS AND INTERVENTIONS OF PHARMACISTS IN A PHYSICAL REHABILITATION HOSPITAL**

**Gordana Ljubojević**

Institute for the Physical Medicine and Rehabilitation „Dr Miroslav Zotovic”,  
Banja Luka (Bosnia and Herzegovina)

In the last 30 years, activities of hospital pharmacists have gone through significant changes and pharmacists are increasingly involved in patient care. The aim of this study was to explore drug-related and logistic problems, interventions, and outcomes during routine work of pharmacists in a physical rehabilitation hospital. During the 2.5 years a systematic, prospective observational study was performed in the 594-bed Institute for physical medicine and rehabilitation, Banjaluka, Bosnia and Herzegovina. Medical doctors, nurses, physiotherapists, and patients addressed three pharmacists, face-to-face or by telephone, with drug-related problems (DRPs) and/or logistic issues. Type of DRP or logistic issue, intervention, outcome, initiator and time spent for solving the problem were documented for each consultation as the main outcome measures. Pharmacists had access to electronic medical records of patients but they were not ward based. Out of 1515 interventions, 48.8% were aimed at solving DRPs. The most common DRPs were the recommendation of a drug or dose and need for additional information about drugs. Drug price and supply were the most prevalent logistic issues. DRPs were more frequently initiated by doctors and required more time to solve the problem compared to logistic issues (Mann-Whitney U test,  $p \leq 0.001$ ). The acceptance rate of interventions to solve DRPs (83.7%) was lower compared to logistic issues (95.2%;  $p \leq 0.001$ ). The average number of DRPs/patient (1.37) was lower compared to other studies in hospital settings (2.6-6.5). Pharmacists (0.51/100 beds) did not perform medication reviews routinely which could explain the low level of clinical DRPs identified by pharmacists (19%).

Hospital pharmacists were faced with an approximately equal number of DRPs and logistic issues during routine work. The overall acceptance rate of pharmacists' interventions was high, and our results indicate that hospital pharmacists in Bosnia and Herzegovina should be more involved in clinical activities and more pharmacists are clearly needed.

## POTENCIJALNE INTERACIJE IZMEĐU LEKOVA U TERAPIJI NA OTPUSTU KOD GERIJATRIJSKIH PACIJENATA

**Ivana Baralić, Vesna Janković, Tatjana Savković, Mirjana Stanojlović,  
Višnja Glišić, Goran Marković, Sonja Simić**

Kliničko-bolnički centar „Zvezdara”, Beograd (Srbija)

Interakcije između lekova mogu dovesti do neželjenih događaja i zdravstvenih problema koji se mogu sprečiti. Interakcije su važan faktor, posebno kod gerijatrijske populacije, gde je primena većeg broja lekova uobičajena. Cilj ovog istraživanja je bio da se identifikuju potencijalne interakcije između lekova u terapiji koja je propisana gerijatrijskim pacijentima na otpustu iz Kliničko-bolničkog centra „Zvezdara”. Ova retrospektivna studija je sprovedena pregledom otpusnih lista pacijenata koji su bili hospitalizovani na Kliničkom odeljenju za gerijatriju u Kliničko-bolničkom centru „Zvezdara” u periodu od januara do aprila 2018. godine. Epocrates aplikacija je korišćena za utvrđivanje potencijalnih interakcija između lekova. Identifikovane interakcije su prema nivou opasnosti podjeljene u 4 grupe: kontraindikovane, izbegavati/koristiti alternative, modifikovati/pratiti i savetuje se oprez. Studija je obuhvatila 70 pacijenata. Srednja starost ispitanika je bila  $79,8 \pm 6,87$  godina. Ukupan broj propisanih lekova je bio 525, dok je presečan broj lekova po pacijentu bio  $7,5 \pm 2,83$ . Šezdeset četiri (91,43%) pacijenta uzima 5 i više lekova. Ukupno 430 interakcija je identifikovano u terapiji 65 pacijenata. Kontraindikovane kombinacije lekova su uočene kod samo 2 pacijenta (2,86%). Trideset devet (9,07%) interakcija izbegavati/koristiti alternative je pronađeno u terapiji 22 pacijenta. Trista trideset četiri (77,67%) interakcije tipa modifikovati/pratiti je uočeno u terapiji 59 pacijenata. Pedeset i pet (12,79%) interakcija kod kojih se savetuje oprez detektovano je u terapiji 32 pacijenta.

Polifarmacija i starija populacija su dokazani faktori rizika za veću učestalost interakcija između lekova. Rezultati ove studije su pokazale visoku učestalost interakcija između lekova u terapiji koja je propisana gerijatrijskim pacijentima nakon hospitalizacije. Ipak, pokazana je niska učestalost kontraindikovanih kombinacija, dok je najčešće uočena interakcija bila tipa „modifikovati/pratiti”. Na osnovu dobijenih rezultata, bilo bi od koristi definisati sistem praćenja i optimizacije upotrebe lekova od strane farmaceuta, da bi se negativne posledice interakcija između lekova izbegle i svele na najmanju moguću meru.

## POTENTIAL DRUG-DRUG INTERACTION IN GERIATRIC PATIENTS AT DISCHARGE

**Ivana Baralić, Vesna Janković, Tatjana Savković, Mirjana Stanojlović,  
Višnja Glišić, Goran Marković, Sonja Simić**

Zvezdara University Medical Center, Belgrade (Serbia)

Drug-drug interactions (DDIs) may often lead to preventable adverse drug events and health damage. This may be an important factor, particularly in geriatric population, as multiple drug therapy is common. The aim of the present study was to identify potential DDIs between drugs prescribed to the patients at discharge from the Zvezdara University Medical Center. A retrospective study was conducted by reviewing discharge lists of hospitalized patients in geriatric clinical ward in Zvezdara University Medical Center, Belgrade, Serbia, from January to April 2018. Epocrates drug interaction checker was used for screening potential DDIs. The identified DDIs were categorized by level of severity into 4 groups: contraindicated, avoid/use alternative, modify/monitor and caution advised. The study included 70 patients. The mean age of patients was  $79.8 \pm 6.87$  years. Total number of drugs prescribed was 525 and average number of drugs per patient was  $7.5 \pm 2.83$ . Sixty four patients (91.43%) received 5 and more drugs. Total 430 DDIs were identified in therapy prescribed to 65 out of 70 patients. Contraindicated DDIs were recorded only in two patients (2.86%). Thirty nine (9.07%) avoid/use alternative interactions were found in 22 patients. Three hundred thirty four (77.67%) modify/monitor interactions were identified in 59 patients. Fifty five (12.79 %) interactions where caution is advised were detected in 32 patients.

Polypharmacy and older age are proven risk factors to potential drug interactions. The findings of the present study showed high prevalence of DDIs in therapy prescribed to the geriatric patients. However, we found low rate of contraindicated interactions, while the most prevalent type of interaction was type modify/monitor. Based on these findings, it could be helpful to established clinical pharmacy system to monitor and optimize medication use, in order to avoid and minimize negative outcomes of drug interactions.

## UNAPREĐENJE KOMPETENCIJA STUDENATA FARMACIJE

**Dragana Jocić<sup>1</sup>, Ljiljana Tasić<sup>2</sup>, Andrijana Milošević Georgijev<sup>2</sup>,  
Dušanka Krajnović<sup>2</sup>, Valentina Marinković<sup>2</sup>**

<sup>1</sup>ZU Apoteka BENU, <sup>2</sup>Katedra za socijalnu farmaciju i farmaceutsko zakonodavstvo, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Profesionalne i lične kompetencije farmaceuta predstavljaju podršku stručnim i organizacionim kompetencijama i neophodne su u uslovima savremene farmaceutske prakse. Potrebno je ove kompetencije razvijati tokom studija u akademskom okruženju, kao i u nastavnim bazama, kako bi studenti bili pripremljeni za realno radno okruženje. Cilj ovog rada je unapređenje profesionalnih i ličnih kompetencija studenata farmacije neophodnih u uslovima savremene farmaceutske prakse.

Projekat „Profesionalni razvoj studenata farmacije“ realizovan je u saradnji ZU Apoteke BENU i Centra za razvoj farmaceutske i biohemijske prakse Univerziteta u Beogradu. Projekat je organizovan kroz tri radionice i odbranu projektnog zadatka i trajao je od 15.2.2018. do 15.6.2018. godine. Teme projektnih zadataka bile su: izgled apoteke; kartica poverenja; facebook/instagram u cilju unapređenja eksterne komunikacije ka pacijentima i kupcima; unapređenje komunikacije farmaceut - lekar u cilju dobrobiti pacijenta; procesi koji će povećati prodaju OTC proizvoda i savetovanje pacijenata; e-karton kao platforma za unapređenje kvaliteta života i optimizaciju nege pacijenta.

U projektu je učestvovalo 70 studenata farmacije. Projekat je realizovan kroz tri modula sa fokusom na profesionalni razvoj i unapređenje kompetencija studenata upoznavanjem sa operativnim procesima u apoteci, kvalitetu radnih procesa, upravljanje i marketing različitih kategorija proizvoda u uslovima savremene farmaceutske prakse. Nakon završenih modula studenti su kroz 6 mini projekata implementirali stečene veštine. Projekat je uspešno odbranilo 48 studenata farmacije. Evaluacija postignuća je sprovedena postavljanjem pitanja od strane mentora tokom odbrane projekata. Sistematski pristup unapređenju kompetencija studenata farmacije organizacijom projekata sa fokusom na profesionalni razvoj studenata i sticanje dodatnih znanja i veština potrebnih za rad u realnom radnom okruženju može doprineti unapređenju profesionalnih i ličnih kompetencija studenata farmacije kao značajnoj potpori stručnih kompetencija koje se razvijaju u akademskom okruženju, kao i njihovoj boljoj pripremljenosti za realno radno okruženje.

Povezivanje procesa u akademskom okruženju sa procesima u realnom radnom okruženju može doprineti unapređenju profesionalnih i ličnih kompetencija farmaceuta i unapređenju farmaceutske prakse.

## **IMPROVING THE COMPETENCIES OF PHARMACY STUDENTS**

**Dragana Jocić<sup>1</sup>, Ljiljana Tasić<sup>2</sup>, Andrijana Milošević Georgijev<sup>2</sup>,**  
**Dušanka Krajnović<sup>2</sup>, Valentina Marinković<sup>2</sup>**

<sup>1</sup>BENU Pharmacy, <sup>2</sup>Department of Social Pharmacy and Pharmaceutical Legislation, University of Belgrade - Faculty of Pharmacy (Serbia)

Professional and personal competences of pharmacists represent support for professional and organizational competences and are necessary in the conditions of modern pharmaceutical practice. These competencies need to be developed during studies in the academic environment in order to prepare students for a real working environment. The aim is to improve the professional and personal competencies of pharmacy students that are necessary in the conditions of modern pharmaceutical practice.

Project „Professional development of students of pharmacy” was realized in cooperation with BENU Pharmacy and Center for Development of Pharmaceutical and Biochemical Practice of the University of Belgrade. The project was organized through three workshops and project presentation task. Project lasted from 15.2.2018. to 15.6.2018. The themes of the project tasks were: pharmacy layout, loyalty card, facebook/instagram in order to improve external communication to patients and customers, improve communication between pharmacist and physician in order to benefit the patient, processes that will increase the sale of OTC products and patient counseling, e-card as a platform for improving the quality of life and optimizing patient care.

The project was attended by 70 students of pharmacy. The project was realized through three modules focusing on professional development and improvement of students' competence by familiarizing with the operational processes in pharmacy, quality, management and marketing in the conditions of modern pharmacy practice. After the completed modules, students implemented the acquired skills through 6 mini projects. The project was successfully presented by 48 students of pharmacy. Evaluation of the project was done by asking questions from a mentor during project presentation. A systematic approach to improving the competence of pharmacy students by organizing projects with a focus on professional development of students and acquiring additional knowledge and skills are necessary for working in a real working environment. That can contribute to the improvement of the professional and personal competences of pharmacy students as a significant support of professional competences that are developed in the academic environment, as well as their better preparedness for a real working environment.

Linking processes in the academic environment with processes in a real work environment can contribute to the improvement of professional and personal competencies of pharmacists and the advancement of pharmacy practice.

## B.CELL: INTERAKTIVNA EDUKACIJA I EDUKATIVNA INTERAKCIJA

**Neda Trivić, Milica Puđa, Tamara Kovačević**

Nacionalna asocijacija studenata farmacije – NAPSer (Srbija)

Onkologija i biotehnologija, sfere izuzetnih dostignuća i primeri najdinamičnijih oblasti medicine, sa konstantnim povećanjem incidence malignih oboljenja zahtevaju proporcionalan porast u broju stručnjaka i inovativnosti u prevenciji i terapiji. Tim povodom susretom farmacije, medicine i informacionih tehnologija na navedenim poljima formirana je B.Cell platforma, u vidu aplikacije i veb sajta. Namenjena je studentima biomedicinskih usmerenja i stažerima, upravo jer predstavlja brz, jednostavan, savremen i besplatan način preuzimanja informacija, podizanja svesti i upotpunjavanja stečenog znanja kako studenata tako i mladih stručnjaka u navedenim sferama.

Platforma nudi mogućnost elektronskog učenja i pruža relevantne, praktično primenljive i najnovije naučno zasnovane vesti sa fokusom na onkologiju. Korisnici imaju pristup edukativnim materijalima, kao što su naučne publikacije, online časopisi, predavanja i kursevi. Specifičnost ponuđene edukacije se oslikava u tome što pored dinamičnog informisanja, B.Cell motiviše korisnike da za svoje angažovanje na samoj platformi budu nagrađeni i viđeni od strane fakulteta, kompanija, budućih poslodavaca i poslovnih partnera. Korisnici rešavanjem kvizova znanja i informisanosti, mini kliničkih studija, diskusijama i volontiranjem sakupljaju „Super celije”, koje mogu da iskoriste na različite načine, kao što su posete farmaceutskim kompanijama, prisustva stručnim predavanjima, seminarima, treninzima, radionicama ili pak ostvarivanje prava na stručne prakse. Za uspešnu realizaciju ovih programa oslonac su partneri B.Cell-a, društveno odgovorne kompanije, kao i nastavnici Medicinskog i Farmaceutskog fakulteta Univerziteta u Beogradu koji svojom stručnošću podržavaju kredibilitet platforme.

## **B.CELL: INTERACTIVE EDUCATION & EDUCATIVE INTERACTION**

**Neda Trivić, Milica Puđa, Tamara Kovačević**

National Association of Pharmacy Students – NAPSer (Serbia)

Oncology and biotechnology, the spheres of exceptional achievements and examples of the most dynamic areas of medicine, with a constant increase in the incidence of malignancies, require proportional increase in the number of experts and innovations in prevention and therapy. On that basis, combining pharmacy, medicine and information technologies, B.Cell platform was formed, as an application and a web site. It is intended for students and interns of biomedical professions because it represents a fast, simple, modern and cost effective way of acquiring information, raising awareness and knowledge improvement of students and young professionals.

The platform offers the possibility of e-learning and provides relevant, practically applicable and latest scientifically based news with a focus on oncology. Users have access to educational materials, such as scientific publications, online journals, lectures and courses. Apart from dynamic information that platform provides, specificity is reflected in the fact that B.Cell motivates users to be rewarded and seen by faculties, companies, future employers and partners for their engagement on the platform itself. The users collect „Super Cells” by solving knowledge quizzes, mini-business and clinical studies, by discussing on various cases or volunteering, which then can be used in different ways, such as visits to pharmaceutical companies, attendance at expert lectures, seminars, trainings, workshops or the opportunity for professional practice placements.

B.Cell is connected with its partners - socially responsible companies, as well as academic staff from the University of Belgrade - Faculty of Medicine and Faculty of Pharmacy which, by their expertise, support the credibility of the platform.

## **UTICAJ PRAVILNIKA KOJIM SE REGULIŠU MAKSIMALNE VELEPRODAJNE CIJENE LIJEKOVA NA CIJENE LIJEKOVA U BOSNI I HERCEGOVINI**

**Biljana Tubić, Jelena Aničić, Tijana Spasojević,  
Ana Cvijanović, Aleksandar Zolak**

Agencija za lijekove i medicinska sredstva Bosne i Hercegovine  
(Bosna i Hercegovina)

Bosna i Hercegovina je u 2017. godini prvi put implementirala podzakonski akt kojim se reguluše nivo maksimalnih veleprodajnih cijena lijekova. Cilj rada je ocjeniti prve rezultate uticaja Pravilnika na nivo veleprodajnih cijena lijekova na tržištu Bosne i Hercegovine.

Upoređene su veleprodajne cijene lijekova za 2016. godinu kada Pravilnik nije postojao u odnosu na 2017. godinu kada je Pravilnik prvi put implementiran. Izvori veleprodajnih cijena lijekova bili su podaci dobijeni od velprometnika za godišnje izvještaje o potrošnji lijekova koje naša agencija publikuje svake godine na svojoj internet prezentaciji [www.almbih.gov.ba](http://www.almbih.gov.ba).

Ukupna sredstva izdvojena za lijekove u 2017. godini niže su u odnosu na 2016. godinu. Takođe, veleprodajne cijene lijekova koji se izdaju na recept ljekara niže su u 2017. godini u odnosu na 2016. godinu. S druge strane, može se vidjeti da su veleprodajne cijene lijekova koji se izdaju bez ljekarskog recepta (OTC lijekovi) više u 2017. u odnosu na iste u 2016. Međutim, ovi bezreceptni lijekovi čine samo 15 % tržišta lijekova, tako da to navedeno nije imalo uticaj na ukupna finansijska sredstva izdvojena na lijekove u 2017. godini u Bosni i Hercegovini. Implementacija Pravilnika i uvođenje sistema maksimalnih veleprodajnih cijena lijekova ima pozitivan uticaj na budžet fondova zdravstvenog osiguranja u Bosni i Hercegovini.

# **INFLUENCE OF THE RULEBOOK FOR REGULATING MAXIMUM WHOLESALE PRICES ON MEDICINE COST IN BOSNIA AND HERCEGOVINA**

**Biljana Tubić, Jelena Aničić, Tijana Spasojević,  
Ana Cvijanović, Aleksandar Zolak**

Agency for Medicinal Products and Medical Devices of Bosnia and Herzegovina  
(Bosnia and Herzegovina)

Bosnia and Herzegovina have implemented rulebook for regulating maximum wholesale prices at the first time in 2017. The aim of this study was to assess the first results of the influence of the rulebook on the level of wholesale prices of medicines on the market of Bosnia and Herzegovina.

Medicine prices in the years 2017 and 2016 were analyzed. The level of wholesale prices in 2016, when rulebook has not existed, were compared with wholesale prices of medicines in 2017 when rulebook was implemented for the first time. The wholesale prices of medicines were data obtained from wholesalers which we collect every year for Annual reports about the distribution of medicines in Bosnia and Herzegovina and which are published on website [www.almbih.gov.ba](http://www.almbih.gov.ba) by our agency.

The total financial expenses for medicines were reduced in 2017 compared to 2016. Also, it was shown that the wholesale prices of Rx medicines are decreased compared with 2016. On the other side, it could be seen that OTC medicines have increased prices in 2017 as compared with the year 2016. But, OTC medicines make 15% of the whole market of medicines and this increase did not have the influence on financial cost in all. Implementation of the Rulebook and system of maximal wholesale prices of Rx medicines has positive influence on budget of funds for healthcare insurance in Bosnia and Herzegovina.

**OD TRADICIONALNE MEDICINE DO RACIONALNE FITOTERAPIJE – 50  
GODINA FARMACEUTSKE SLUŽBE U OKVIRU BILJNE APOTEKE  
INSTITUTA ZA PROUČAVANJE LEKOVITOG BILJA „DR JOSIF PANČIĆ”**

**Nebojša Menković**

Institut za proučavanje lekovitog bilja „Dr Josif Pančić”, Beograd (Srbija)

Značaj lekovitih biljaka i pripravaka od biljnih droga, iz godine u godinu ima tendenciju porasta. Tako je i u našoj zemlji i u svetu, a posebno u razvijenim zemljama. Potražnju lekovitih i aromatičnih biljaka stimulišu i potkrepljuju pozitivna iskustva tradicionalne medicine koja su i dalje osnov za moderna naučna istraživanja i, paralelno sa sveobuhvatnim istraživanjima savremenim laboratorijskim metodama, doprinose razvoju novih biljnih lekova. Ovakav pristup razvoja je i ideja vodilja Instituta za proučavanje lekovitog bilja „Dr Josif Pančić“. Ove godine Institut slavi 70 godina postojanja i rada, kao i 50 godina rada Biljne Apoteke, u okviru Instituta.

Biljna apoteka Instituta je svojevrsni centar fitoterapije gde su prethodne i današnje generacije pronalazile rešenje za mnoge zdravstvene tegobe. Savetovalište za fitoterapiju i lekovito bilje osnovano je u okviru Biljne Apoteke. Njime prvenstveno želimo da unapredimo kontakt sa pacijentima i ponudimo dodatni podsticaj za korišćenje proizvoda na bazi lekovitog bilja na racionalni način (promocija racionalnih fitofarmaka standardizovanih na aktivne ili preovlađujuće materije, proizvedenih u definisanim uslovima u institutskim farmaceutskim pogonima i laboratorijama kao i „ex tempore“). Sa druge strane, razgovor sa pacijentima i njihova iskustva u korišćenu lekovitog bilja su značajan izvor informacija za istraživače Instituta.

Od velikog značaja je i sakupljanje terenskih informacija o upotrebi poznatih i manje poznatih lekovitih biljaka koje se u narodu koriste za različite bolesti i simptome. Čitav taj sistem prikupljanja informacija, omogućava da se na kvalitetan način razvije formulacija i postpak izrade racionalnog fitofarmaka.

U ovom radu predstavljamo naša iskustva kroz rad: Savetovališta, Botaničke baštne lekovitog bilja „Akademik Jovan Tucakov“ u Valjevu, kao i terenskih i drugih istraživanja u okviru Instituta.

# **FROM TRADITIONAL MEDICINE TO RATIONAL PHYTOTHERAPY -50 YEARS OF PHARMACEUTICAL PRACTICE WITHIN THE HERBAL PHARMACY OF THE INSTITUTE FOR MEDICINAL PLANTS RESEARCH „DR JOSIF PANČIĆ”**

**Nebojša Menković**

Institute for Medicinal Plants Research „Dr Josif Pančić“ (Serbia)

The importance of medicinal plants and herbal remedies, from year to year, has a tendency to increase. This is the situation in our country as well as in the world, especially in developed countries.

The demand for medicinal and aromatic plants is stimulated and supported by positive experiences of traditional medicine which are still the basis for modern scientific research and, along with comprehensive research with modern laboratory methods, contribute to the development of new herbal remedies. This type of approach is also the leading idea for the development of the Institute for Medicinal Plants Research „Dr Josif Pančić“. This year, the Institute celebrates 70 years of existence and work, as well as 50 years of work of the Herbal Pharmacy, within the Institute.

Herbal Pharmacy of the Institute is a kind of phytotherapy center where the previous and present generations who worked in the Institute have found a solution for many health problems. Counseling center for phytotherapy and medicinal herbs was established within the Herbal Pharmacy. We primarily want to improve contact with patients and offer additional incentives for the use of medicinal products in a rational manner (promotion of rational phytopharmaceuticals standardized on active or predominant compounds produced in defined conditions in Institutes production sector, laboratories as well as „ex tempore“). On the other hand, talking with patients and their experiences in the use of medicinal herbs are of significant importance for the researchers of the Institute.

Collection of terrain information on the use of well-known or less well-known medicinal plants that are used by the people for various diseases and symptoms are of great importance as well. All this information gathering enables us to make the formulation and production of rational phytopharmaceuticals in a quality way.

In this paper we present our experiences through the work of Counseling center, Botanical Garden of medicinal herbs „Akademik Jovan Tucakov“ in Valjevo, as well as terrain and other research within the Institute.

## **APOTEKAR I JAVNO-ZDRAVSTVENI PROSVETITELJ MR PH MILIVOJE MOLJAC: PEČAT U VREMENU**

**Stevan Vukov**

Apoteka „Sent Andreja”, Zrenjanin (Srbija)

Ovaj rad ima za cilj da prikaže sveukupni doprinos apotekara Milivoja Moljca i na taj način otrgne od zaborava njegov lik i delo. Korišćeni su primarni i sekundarni izvori za istorijsku analizu, kako biografskih podataka o životu Moljca, tako i hronoloških podataka o njegovom radu.

Milivoje Moljac (1890 - 1979) bio je farmaceut po obrazovanju, i apotekarski poziv obavljaо je tokom celog profesionalnog života. Od kako je 1919. godine dobio koncesiju za otvaranje šeste po redu apoteke „Kod Svetog Jovana” u ondašnjem Velikom Bečkereku (današnji Zrenjanin), pa sve do prisilnog otkupa apoteke 1949. godine aktivno je učestvovao na različitim poljima, ne samo stručne već i kulturno-prosvetne delatnosti. Bio je vlasnik i urednik apotekarskih časopisa, učestvovao u izradi Jugoslovenske farmakopeje (1933), bio u upravnom odboru veledrogerije „Slavija”, učestvovao u organizaciji Sveslovenskog kongresa apotekara (1939). Istovremeno, obavljaо je značajne dužnosti u Crvenom krstu, Sokolskom društvu, Narodnoj odbrani, kao i Jadranskoj straži. Kao istaknuti član Demokratske stranke aktivno je učestvovao u političkom životu. Tokom Drugog svetskog rata pripadaо je neformalnoj organizaciji građanskih intelektualaca koja je prikupljala hranu, lekove i novac, ne samo za hiljade izbeglica, već i za članove pokreta otpora. Posle rata nastavlja sa radom u sopstvenoj apoteci, pod budnim okom novih vlasti. Tokom 1949. godine, njegova apoteka je prešla u državnu svojinu. Sve do odlaska u penziju ostao je na mestu upravnika nekad svoje, a potom Treće narodne apoteke. Njegovi potomci, čerka Vukosava i sin Slobodan, nastavili su očevim stopama i ostali verni apotekarskom pozivu. U zaključku, možemo naglasiti da se Moljčev doprinos kao receptariusa nesumnjivo osećao i izvan okvira apoteke, te da je on ostavio pečat u kulturnom i političkom razvoju grada. S druge strane, njegov doprinos farmaceutskoj profesiji je daleko prevazišao lokalni karakter i može se reći da je rad apotekara Moljca bio od velike važnosti za razvoj farmacije u Jugoslaviji između dva svetska rata.

# **PHARMACIST AND PUBLIC HEALTH EDUCATOR MILIVOJE MOLJAC: SEAL IN TIME**

**Stevan Vukov**

Pharmacy „Sent Andreja” Zrenjanin (Serbia)

The aim of this work is to show overall contribution of pharmacist Milivoje Moljac, and turn away from forgetting his character and working. Primary and secondary sources, biographical data about life and activities of Moljac have been used for historical analysis.

Milivoje Moljac (1890-1979) was a pharmacist, and he was engaged in pharmacy practice through entire his professional life. Since he received a concession to open the sixth consecutive pharmacy „St. John”, in 1919, in Veliki Beckerek, until the forcible acquisition in 1949, he actively participated in various fields, not only professional, but also cultural and educational activities. He was the owner and editor of pharmaceutical journals, also he participated in the development of the Yugoslav Pharmacopoeia (1933), he was member of the board of the wholesaler „Slavija” and participated in the organization of the all Slovenian Congress of Pharmacists (1939). At the same time, he performed important duties in the Red Cross, the Sokol Society, the National Defense, and the Adriatic Guard. During the Second World War he belonged to the non-formal organization of civil intellectuals who collected food, medicine and money, not only for thousands of refugees, but also for members of the resistance movement. After the war, he continues to work in his own pharmacy, under the watchful eye of new authorities. During 1949, his pharmacy has passed into the State ownership. Until his retirement he remained the manager of what was formerly his own, and later the Third People's Pharmacy. In conclusion, we can emphasize that Moljac's contribution as a receptarius was extended beyond the pharmaceutical area, and he left a mark on the cultural and political development of the city. On the other hand, his contribution to the pharmaceutical profession has overcome the local character and it can be said that the work of Moljac was of great importance for the development of pharmacy in Yugoslavia between the two World Wars.

## **HOMEOPATHY 222 YEARS AFTER – THE HISTORICAL KNOWLEDGE AND VIEWS OF SAMUEL HAHNEMANN IN CONTEXT OF HIS WRITINGS**

**Łukasz Komsta**

Medical University of Lublin (Poland)

Homeopathy is one of the few trends in alternative medicine that have survived to our times and have not gone away since its beginning. Despite the lack of convincing scientific evidence, it still has many followers. As almost all literature sources are subjective and present the point of view of an enthusiast or a hater. Therefore, there is a general need to look with a distance and see the original motivations that led Hahnemann to think in the homeopathic way.

The point-by-point reading of original Organon is the best way to see what Hahnemann thought and how he perceived the nature in his times. It allows to understand the neglected and forgotten details of medical knowledge and to verify many views in today's knowledge of the etiology of diseases and classical pharmacology. It is also beneficial to see Organon in context of Hahnemann's contemporary philosophical views, to sort in a chronological manner scientific milestones and to understand the placement of Hahneman's life around the most important nineteenth century chemical discoveries.

In current scientific methodology, the „similia similibus curantur” paradigm cannot be even called a theory and it is a great example of a priori deduction. The contemporary philosophical arguments an example of another paradigm and a scientific troublesome. The only paradigm of treatment are the laws of nature, which do not depend on the adopted philosophical view and one has to understand the lack of the knowledge in Hahnemann's times.

It is very hard to perceive the homeopathy in any scientific way. It should be perceived only as a historical curiosity without any unneeded emotional background. If there is any undiscovered medical mechanism beyond any homeopathic drug, it surely cannot be derived from „similia...” principle.

## KRATAK ISTORIJSKI PRIKAZ MAGISTRALNIH LEKOVA KOJI SE PRIMENJUJU KOD OPSTIPACIJE

**Ilinka Vuković<sup>1</sup>, Mirjana Gajdaš<sup>2</sup>, Dušanka Krajnović<sup>3</sup>**

<sup>1</sup>Srpsko lekarsko društvo, <sup>2</sup>Apoteka Beograd, <sup>3</sup>Katedra za socijalnu farmaciju i farmaceutsko zakonodavstvo, Univerzitet u Beogradu–Farmaceutski fakultet (Srbija)

Izrada magistralnih lekova pominje se još u prvim recepturnim priručnicima lekar-apotekara, a i značajni nacionalni istorijski izvori prvog reda, poput Hilandarskog medicinskog kodeksa, na više mesta se bave sredstvima za čišćenje na bazi šljive, aloje, belog duda i drugih biljnih droga. U savremenoj terapiji magistralni lekovi zauzimaju značajno mesto, a njihova izrada podržana je i Rezolucijom Evropske komisije (Resolution CM/ResAP, 2011), odobrenom od Saveta Evrope. U Evropskoj farmakopeji postoji monografija o preparatima ex tempore, što odgovara magistralnim lekovima. Među onima koji se koriste u terapiji bolesti organa za varenje nalazimo veći broj lekova koji se preporučuju kod opstipacije, čiji uzroci mogu biti brojni, funkcionalni ili organski kao posledica neke bolesti, a lečenje je kompleksno i zavisi od uzroka. Iako je opstipacija češća kod starih osoba, mogu je imati i deca, trudnice, ili može biti posledica uzimanja lekova (analgetici, antacidi, opioidi, antiparkinsonici, antibiotici...).

U radu je analizirano 38 preskripcija za magistralne lekove koji se primenjuju kod opstipacije, preuzetih iz nekoliko izvora: 8 magistralnih lekova iz Sveske magistralnih lekova dr Miloša Todorova (1894-1969), privatna ordinacija somborskog lekara, 4 iz Formulae magistrales et reagentia FM II (1966), 6 lekova iz FM III (1979), 12 preskripcija iz Farmakoterapije Dragutina Tomića (1989) i 8 monografija iz važećih propisa Magistralne formule (2008). Prisutno je više farmaceutskih oblika lekova, a najviše preskripcija se odnosi na podeljene praškove (12), pilule (6), supozitorije (6), oralne emulzije (6) i čajne mešavine (3). U pogledu sastava prisutno je preko 60 aktivnih komponenti i ekscipijenasa, a dominiraju droge biljnog porekla. U zaključku možemo reći da su analizirane preskripcije magistralnih lekova za opstipaciju, prisutne u više farmaceutskih oblika, primenjivane peroralno i rektalno, pružale mogućnost, uz druge mere lečenja, za prilagođavanje terapije prema individualnim potrebama pacijenta.

## **SHORT HISTORICAL OVERVIEW OF EXTEMPORANEOUSLY COMPOUNDED MEDICINES FOR CONSTIPATION**

**Ilinka Vuković<sup>1</sup>, Mirjana Gajdaš<sup>2</sup>, Dušanka Krajnović<sup>3</sup>**

<sup>1</sup>Serbian Medical Society, <sup>2</sup>Pharmacy Beograd, <sup>3</sup>Department of Social Pharmacy and Pharmaceutical Legislation, University of Belgrade - Faculty of Pharmacy (Serbia)

Pharmaceutical compounding can be traced back to early practitioner-pharmacists' prescription manuals, including prominent national historical sources such as The Hilandar Medical Codex, which contains several references to treatments for defecation, based on plum, aloe, white mulberry and other plants. Extemporaneously compounded medicines are relevant in contemporary treatment and their preparation is supported by European Committee Resolution (Resolution CM/ResAP, 2011), approved by The European Council, while The European Pharmacopoeia includes a monograph on ex tempore products which correspond to extemporaneously compounded medicines. Amongst medications used in treating digestive disorders, many are recommended for constipation. Its causes are either functional or organic, or a result of another illness, the treatment is complex, and it depends on the exact cause. Even though it often affects the elderly, it might occur in children, during pregnancy or as a consequence of taking other prescription drugs (analgesics, antacids, opioids, antiparkinsonics, antibiotics...).

Our analysis includes a total of 38 prescriptions for extemporaneously compounded medicines used for treating constipation, from several sources: 8 extemporaneously compounded medicines from the Extemporaneously Compounded Medicines Notebook by a physician Miloš Todorov (1894-1969), private physician's practice in Sombor, Serbia, 4 from Formulae magistralis et reagentia FM II (1966), 6 extemporaneously compounded medicines from FM III (1979), 12 prescriptions from Pharmacotherapy by Dragutin Tomić (1989), and 8 monographs from contemporary regulations Formulae Magistralis (2008). We found several pharmaceutical dosage forms, and the majority prescriptions are for divided powders (12), pills (6), suppositories (6), oral emulsions (6), and tea concoctions (3). In terms of ingredients there are over 60 active substances and excipients, with plant-based drugs being dominant. We can conclude that analysed prescriptions for extemporaneously compounded medicines for constipation, which existed in several pharmaceutical dosage forms, and were administered either orally or rectally, facilitated the adjustment of therapy to individual needs of the patient, in addition to other ways of treatment.

## **FALSIFIKOVANI LEKOVI - IZAZOVI REGULATIVE U SPROVOĐENJU I PROMOCIJI BEZBEDNE UPOTREBE LEKOVA**

**Pavle Zelić, Saša Jaćović**

Agencija za lekove i medicinska sredstva Srbije (Srbija)

Rad ima za cilj da predstavi razvoj legislative u oblasti falsifikovanih lekova i trenutno stanje u pogledu terminologije, pravnih akata, smernica i ostalih relevantnih dokumenata na nacionalnom i međunarodnom planu koji se tiču lažnih lekova i medicinskih sredstava. Na ovaj način bi se pokazala određena disjunktnost i neusaglašenost između organa zaduženih za regulativu i sprovođenje mera u svrhu minimizacije javno-zdravstvenih rizika od ovih ilegalnih medicinskih proizvoda. Uporedjivana su pravna akta na nivou Saveta Evrope, Evropske Unije, Svetske zdravstvene organizacije, Internacionale konferencije o harmonizaciji i drugih međunarodnih aktera, kao i nevladinih organizacija koje se bave borbom i podizanjem svesti o problematici falsifikovanih medicinskih proizvoda. Ova saznanja su komparirana i na nacionalnom nivou, kako u pogledu zakona i podzakonskih akata u Republici Srbiji, ali i regionu i ključnim državama Evrope i sveta kao što su SAD, Ruska federacija, Japan, Australija, Kina, Indija, Švajcarska i Kanada. Takođe je pravljeno poređenje kako farmaceutske, ali i legislative u pogledu zaštite prava intelektualne svojine i konačno, praktične implikacije svih ovih dokumenata u stvarnom životu.

Utvrđena su značajna odstupanja počev od samog pojma lažnog odnosno falsifikovanog leka, preko njegove definicije u užem i širem smislu, do daljih elaborata o pristupu ovom problemu i njegovom rešavanju kroz operativne i kaznene odredbe. Nedovoljan broj radova na ovu temu ukazuje da se ova važna oblast nije na adekvatan način obrađivala u naučne svrhe, niti postoji uzajamno razumevanje između ključnih činilaca koji bi, uz određeno usmeravanje, morali da obezbede objedinjen i sveobuhvatan pristup problematici, bez dupliranja posla, a naročito nejasnoća u pogledu pravne utemeljenosti koja podržava akcije na terenu. Za regulatorne autoritete u Srbiji i drugim zemljama, neophodno je da se pronađu pravi modaliteti i primeri koji bi olakšali kako donošenje propisa, ali i njihovu primenu uz određeni naučni osnovi svih aktivnosti.

## **FALSIFIED MEDICINES – REGULATORY CHALLENGES IN SAFE DRUG USE AND ITS PROMOTION**

**Pavle Zelić, Saša Jaćović**

Medicines and Medical Devices Agency of Serbia (Serbia)

The paper aims to present the development of legislation for falsified medicines and medical devices and current situation regarding terminology, legal acts, guidelines and other relevant documents on the national and international level. Certain disagreement between the regulatory authorities and the implementation of measures will be demonstrated in order to minimize the public health risks of illegal medicinal products. Legal acts from the Council of Europe, the European Union, the World Health Organization, the International Conference on Harmonization and other international actors, and non-governmental organizations fighting and raising awareness about the problem of falsified medical products were compared. This is comparable on national level, in terms of laws and by-laws in the Republic of Serbia, but also in the region and key countries of Europe and the world - United States, the Russian Federation, Japan, Australia, China, India, Switzerland and Canada. Further comparison was made of both pharmaceutical and legislative with regard to the protection of intellectual property rights and, practical implications of all these documents in real life.

Significant deviations were identified, starting from the notion of a false or falsified drug, through its definition in broader sense, to further studies on access to this problem and its resolution through operational and penal provisions. Insufficient number of papers on this topic indicate this important area has not been adequately addressed for scientific purposes, nor there is a mutual understanding between key factors that, with a certain direction, should provide a unified and comprehensive approach to the problem, without duplication of work, and especially uncertainty in terms of legal basis that supports action on the ground. For regulatory authorities in Serbia and other countries, it's necessary to find real modalities and examples that would facilitate both the adoption of regulations and their application, along with a certain scientific basis for all activities.

## **DEFEKT KVALITETA LEKA - REGULATORNI ZAHTEVI I SAVREMENI TRENDJOVI**

**Svetlana Mihaljica, Žarko Jović, Boris Bojić,  
Marija Malešević, Gordana Pejović**

Agencija za lekove i medicinska sredstva Srbije (Srbija)

Defekt kvaliteta lekova ima veliki značaj u post-marketinškom praćenju kvaliteta lekova. Defekt kvaliteta lekova može da dovede do povlačenja serija koje odstupaju od standarda kvaliteta i do suspenzije ili zabrane njihovog prometa. Regulatorna očekivanja za rukovanje reklamacijama i povlačenje proizvoda se zasnivaju na GMP/EU-EudraLex Vol.4 Part I (Medicinal Product), Chapter 8 and Part II (Active Substance used as Starting Materials), Section 15. U radu će biti dat pregled svih propisa koji su u vezi sa defektom kvaliteta/procesom povlačenja u zemljama EU i Republici Srbiji i odgovornostima svih uključenih učesnika u ovom postupku.

Sve reklamacije i druge informacije koje su u vezi sa mogućim proizvodima koji odstupaju od standarda kvaliteta moraju biti pažljivo pregledane u skladu sa pisanom procedurom. Treba da postoji sistem koji omogućava brzo i efikasno povlačenje poznatih proizvoda ili onih za koje postoji sumnja da odstupaju od standarda kvaliteta iz prometa, ako je potrebno. Postoji spisak kategorija odstupanja za klasifikaciju defekta proizvoda (kroskontaminacija, neusaglašenost sa dozvolom za lek, FDA Warning Letter, OOS, test stabilnosti...). Inicijalne akcije trebaju da budu preduzete kao rezultat preliminarne istrage za defekt kvaliteta (Rapid Alert, povlačenje). Povlačenja serije za defekt kvaliteta su klasifikovana zavisno od moguće ugroženosti po život i rizika po zdravlje.

Koji su sledeći koraci istrage: Glavni uzrok defekta je određen? CAPA su analizirane i implementirane? U skladu sa Aneksom 16 GMP, QP je odgovoran da obezbedi da je svaka pojedinačna serija proizvedena u skladu sa zahtevima dozvole za lek i sa GMP. Dodatno, QP je odgovoran da obezbedi da sve reklamacije koje su u toku, istrage ili povlačenja budu prepoznate za sertifikaciju predmetne serije. Postoji veliki značaj za implementaciju regulative za defekt kvaliteta, kao i potrebe za jačanje sistema za efikasno povlačenje lekova koji odstupaju od kvaliteta u cilju da se obezbedi kvalitet, efikasnost i bezbednost lekova u prometu.

## **MEDICINE QUALITY DEFECT - REGULATORY REQUIREMENTS AND CURRENT TRENDS**

**Svetlana Mihaljica, Žarko Jović, Boris Bojić,**  
**Marija Malešević, Gordana Pejović**

Medicines and Medical Devices Agency of Serbia (Serbia)

Medicines quality defect is of increasing importance in post-marketing medicines quality monitoring, as it can lead to the recall of a defected batch and potential suspension or prohibition on the market. Regulatory expectations for complaint handling and product recall are based on GMP/ EU-EudraLex Vol.4 Part I (Medicinal Product), Chapter 8 and Part II (Active Substance used as Starting Materials), Section 15. Review of regulation related to the quality defect/recall process in EU countries and Republic of Serbia and responsibilities of all participants involved in this procedure has been performed.

All complaints and other information concerning potentially defective products must be reviewed carefully according to written procedures. A system should be designed to recall, if necessary, promptly and effectively products known or suspected to be defective from the market. There is a list of categories of deficiencies to classify product defects (cross-contamination, deviation from MA, FDA Warning Letter, OOS, stability testing...). Initial actions should be taken as a result of preliminary investigation for quality defect (rapid alert, recall). Batch recalls for quality defects are classified depending on potentially life-threatening and risk to health.

What are the next steps of investigation: Root Cause is determined? CAPA are analysed and implemented? In line with Annex 16 GMP, the QP is responsible for ensuring that each individual batch has been manufactured in accordance with the MA and GMP requirements. Additionally, QP has responsibility to ensure that any on-going complaint, investigation or recall does not negate the conditions for certification of the batch in question. There is a great importance for the implementation of quality defect regulations, as well as for strengthening the system for effective recall of the defective medicines in order to achieve quality, efficacy and safety of medicines on the market.

## SPECIFIČNOSTI FARMAKOVIGILANCE BIOTEHNOLOŠKIH LEKOVA

**Milena Miljković<sup>1</sup>, Cvijeta Bielen<sup>2</sup>, Aleksandra Pajić<sup>2</sup>**

<sup>1</sup>PrimeVigilance, Beograd (Srbija), <sup>2</sup>PrimeVigilance, Zagreb (Hrvatska)

Tokom poslednje dve decenije upotreba biotehnoloških lekova u medicini stalno se povećava. Otkrivanje monoklonskih antitela i njihovo uvođenje posebno u terapiju autoimunskih/inflamatornih i malignih bolesti, smatra se jednim od najvećih napredaka biotehnologije do sada. Cilj ovog rada je da rezimira specifičnosti farmakovigilance visoko sofisticiranih biotehnoloških lekova.

Proučeni su relevantni pregledi, smernice i preporuke, objavljene u medicinskoj literaturi i preko regulatornih agencija tokom poslednjih pet godina, uključujući i odobrene proizvode.

Biotehnološki lekovi su biološki proizvodi dobijeni korišćenjem biotehnoloških metoda i bioloških sistema za njihovo stvaranje ili modifikaciju. Biološki slični lekovi su skoro identične kopije originalnih biotehnoloških proizvoda, sa kojima su veoma slični, ali ne i identični. Postoje određeni aspekti farmakovigilance biotehnoloških lekova koji zahtevaju posebnu pažnju. Značajna karakteristika bezbednosti biotehnoloških lekova je njihov kapacitet da indukuju imunogenost. Svi biološki lekovi proteinske strukture pokreću imunski odgovor, a kao rezultat nastaju anti-lek antitela. Brojni faktori vezani za sam lek, pacijenta ili način davanja leka mogu da utiču na imunogenost. U pojedinim slučajevima, razvoj imunskog odgovora na biotehnološki lek može smanjiti njegovu efikasnost, uz blage neželjene efekte, dok u drugim slučajevima imunski odgovori mogu dovesti do ozbiljnih, a ponekad i fatalnih neželjenih efekata. Dodatno, pri primeni biotehnoloških lekova mogu se pojaviti i infuzijske reakcije. One mogu biti anafilaktičke (IgE-posredovane) i anafilaktoidne (nisu IgE-posredovane). Ove reakcije javljaju se već u toku davanja infuzije ili u okviru jednog sata od njenog završetka i mogu imati širok spektar neželjenih efekata, ponekad i nespecifičnih. Kod biotehnoloških proizvoda, predmet većine regulatornih bezbednosnih mera bile su reakcije na mestu primene leka, infekcije, neoplazme i poremećaji imunološkog sistema.

U postmarketinškom periodu, biotehnološki lekovi zahtevaju pažljivo praćenje imunološki posredovanih neželjenih efekata i infuzijskih reakcija, dok za imunomodulatorne lekove treba pažljivo pratiti razvoj oportunističkih infekcija i sekundarnih neoplazmi.

## SPECIFICITIES OF PHARMACOVIGILANCE OF BIOTECHNOLOGICAL MEDICINES

**Milena Milković<sup>1</sup>, Cvijeta Bielen<sup>2</sup>, Aleksandra Pajić<sup>2</sup>**

<sup>1</sup>PrimeVigilance, Belgrade (Serbia), <sup>2</sup>PrimeVigilance, Zagreb (Croatia)

Over the past two decades, the use of biopharmaceuticals in medicine was constantly increasing. The discovery of monoclonal antibodies and particularly their introduction in the therapy of autoimmune/inflammatory and malignant diseases, are considered as one of the greatest advances in the field of biotechnology so far. The aim of this work is to summarize specificities of pharmacovigilance of highly sophisticated biopharmaceuticals.

Relevant reviews, guidelines and recommendations published in the medical literature and through regulatory agencies during the last five years, including approved products, have been examined.

Biotechnology-derived medicines are biologicals manufactured by biotechnological methods and biological systems to create or modify products. Biosimilars are almost identical copies of original biotechnological medicines, with which they are highly similar but not identical. There are certain aspects of pharmacovigilance of biopharmaceuticals that require special attention. An important feature of biopharmaceuticals safety is their capacity to induce immunogenicity. All biopharmaceuticals trigger an immune response, and as a result, anti-drug antibodies are produced. Numerous factors associated with the medicine itself, the patient, or the method of administration may affect immunogenicity. In some cases, development of immune response to biopharmaceuticals can decrease their efficacy with only mild adverse effects, while in other cases, immune responses can lead to serious, and sometimes fatal, adverse events. Additionally, infusion related reactions can occur with biopharmaceuticals. These reactions can be anaphylactic (IgE-mediated) or anaphylactoid (non-IgE-mediated). They appear already during infusion or within one hour of its completion, can be broad, and at times non-specific. Administration site reactions, infections, neoplasms, and immune system disorders were the subject of most safety-related regulatory actions for biopharmaceuticals.

In the post-marketing period, biopharmaceuticals require careful monitoring of immune-mediated adverse events and infusion related reactions, while for immunomodulatory agents careful surveillance for development of opportunistic infections and secondary malignancies is needed.

## ULOGA FARMAKOGENETIKE U PERSONALIZOVANOJ TERAPIJI KOD PACIJENATA SA TRANSPLANTIRANIM BUBREGOM NA TAKROLIMUS-BAZIRANOJ IMUNOSUPRESIJI

**Nikola Stefanović<sup>1</sup>, Radmila Veličković-Radovanović<sup>1</sup>,  
Katarina Dinić<sup>2</sup>, Tatjana Cvetković<sup>3</sup>**

<sup>1</sup>Katedra za farmaciju, Univerzitet u Nišu-Medicinski fakultet, <sup>2</sup>Univerzitet u Nišu-Medicinski fakultet, <sup>3</sup>Katedra za Biohemiju, Univerzitet u Nišu-Medicinski fakultet (Srbija)

Kliničku primenu takrolimusa komplikuje izražena interindividualna varijabilnost u farmakokineticu i hronična nefrotoksičnost leka. Glavni cilj ovog istraživanja bila je procena potencijalnog uticaja citohrom P450 3A5 (CYP3A5) 6986A>G i ABCB1 3435C>T genskih polimorfizama na dozom-prilagođenu koncentraciju takrolimusa (C0/D) u toku 36 meseci nakon transplantacije bubrega (RTx). Dodatno, istražili smo da li ispitivani polimorfizmi mogu ispoljiti negativan uticaj na bubrežnu funkciju u posmatranom post-transplantacionom periodu.

Studija je uključila 93 pacijenta sa transplantiranim bubregom na takrolimus-baziranoj imunosupresiji. Njima je određen CYP3A5 i ABCB1 genotip (kodira P-glikoprotein) korišćenjem alel-specifične PCR metodologije. C0/D (ng/mL/mg) je izračunat kao količnik predozne koncentracije i odgovarajuće doze takrolimusa. Procenjena brzina glomerularne filtracije (mL/min/1.73m<sup>2</sup>, eGFR) je izračunata korišćenjem MDRD formule.

Pacijenti su genotipizirani na CYP3A5 (12,9% CYP3A5\*1/\*3; 87,1% CYP3A5\*3/\*3) i ABCB1 (25,8% CC; 47,3% CT; 26,9% TT) genski polimorfizam. Nosioci CYP3A5\*1/\*3 imali su niže vrednosti C0/D takrolimusa u poređenju sa nosiocima CYP3A5\*3/\*3 nakon 1, 6, 12, 24 i 36 meseci post-transplantacionog perioda ( $0,91 \pm 0,38$  vs.  $1,35 \pm 0,54$ ,  $p=0,008$ ;  $1,17 \pm 0,69$  vs.  $2,02 \pm 1,14$ ,  $p=0,011$ ;  $1,35 \pm 0,56$  vs.  $2,41 \pm 1,57$ ;  $p=0,015$ ;  $1,50 \pm 0,71$  vs.  $2,28 \pm 1,45$ ,  $p=0,046$ ;  $1,86 \pm 1,22$  vs.  $2,44 \pm 1,14$ ,  $p=0,041$ , respektivno). Nije bilo razlike u C0/D takrolimusa u odnosu na ABCB1 genotip. Multivariatna regresiona analiza potvrdila je da CYP3A5 predstavlja nezavisan prediktor C0/D u toku posmatranog perioda. Nosioci CYP3A5\*1/\*3 imali su nižu eGFR u poređenju sa pacijantima sa CYP3A5\*3/\*3 genotipom nakon 24 i 36 meseci post-transplantacionog perioda ( $37,62 \pm 12,70$  vs.  $51,89 \pm 15,97$ ,  $p=0,022$ ;  $37,25 \pm 16,48$  vs.  $50,99 \pm 16,74$ ,  $p=0,023$ ; respektivno).

CYP3A5 genotip doprinosi interindividualnoj varijabilnosti u dozi takrolimusa neophodnoj da održi optimalnu imunosupresiju, ne samo u ranom, već i kasnjem periodu nakon RTx. Smanjenje bubrežne funkcije može biti izraženje kod pacijenata sa CYP3A5\*1/\*3 genotipom u dugoročnom periodu nakon RTx. Uvođenje CYP3A5 genotipizacije zajedno sa terapijskim monitoringom takrolimusa može obezbediti personalizovanu terapiju kod pacijenata sa transplantiranim bubregom.

# **THE ROLE OF PHARMACOGENETICS IN PERSONALIZED THERAPY OF RENAL TRANSPLANT RECIPIENTS ON TACROLIMUS-BASED IMMUNOSUPPRESSION**

**Nikola Stefanović<sup>1</sup>, Radmila Veličković-Radovanović<sup>1</sup>,  
Katarina Dinić<sup>2</sup>, Tatjana Cvetković<sup>3</sup>**

<sup>1</sup>Department of Pharmacy, University of Niš-Faculty of Medicine, <sup>2</sup>University of Niš - Faculty of Medicine, <sup>3</sup>Department of Biochemistry, University of Niš - Faculty of Medicine (Serbia)

The clinical use of tacrolimus is complicated by marked inter-individual variability in its pharmacokinetics and chronic nephrotoxicity. The main goal of this study was to evaluate potential influence of cytochrome P450 3A5 (CYP3A5) 6986A>G and ABCB1 3435C>T gene polymorphisms on tacrolimus dose-adjusted trough concentrations (C0/D) up to 36 months after renal transplantation (RTx). Additionally, we aimed to investigate whether tested polymorphisms might have negatively affected renal function or not, in the observed post-transplant period.

The study enrolled 93 renal transplant recipients on tacrolimus-based immunosuppression, who were genotyped for CYP3A5 and ABCB1 (encoding P-glycoprotein) using allele-specific PCR method. We calculated C0/D (ng/mL/mg) as trough concentration divided by corresponding tacrolimus dose. Estimated glomerular filtration rate (mL/min/1.73m<sup>2</sup>, eGFR) was calculated by MDRD formula.

We genotyped patients for CYP3A5 (12.9% CYP3A5\*1/\*3; 87.1% CYP3A5\*3/\*3) and ABCB1 (25.8% CC; 47.3% CT; 26.9% TT) gene polymorphism. CYP3A5\*1/\*3 carriers had lower tacrolimus C0/D than CYP3A5\*3/\*3 carriers after 1, 6, 12, 24 and 36 months of post-transplant period (0.91±0.38 vs. 1.35±0.54, p=0.008; 1.17±0.69 vs. 2.02±1.14, p=0.011; 1.35±0.56 vs. 2.41±1.57, p=0.015; 1.50±0.71 vs. 2.28±1.45, p=0.046; 1.86±1.22 vs. 2.44±1.14, p=0.041; respectively). There was no difference in tacrolimus C0/D with respect to ABCB1 genotype. Multivariate regression analysis confirmed that CYP3A5 gene polymorphism was an independent predictor of C0/D within observed period. The carriers of CYP3A5\*1/\*3 genotype had lower eGFR compared to patients with CYP3A5\*3/\*3 genotype after 24 and 36 months of post-transplant period (37.62±12.70 vs. 51.89±15.97, p=0.022; 37.25±16.48 vs. 50.99±16.74, p=0.023; respectively).

CYP3A5 genotype contributes to the inter-individual variability in tacrolimus dose requirements in order to maintain optimal immunosuppression, not only in the early, but as well in the late period after RTx. Renal function decline may be more pronounced in patients with CYP3A5\*1/\*3 genotype in long-term periods after RTx. The introduction of CYP3A5 genotyping alongside therapeutic monitoring of tacrolimus may provide personalized therapy in renal transplant recipients.

## ULOGA I ZNAČAJ MITOHONDRIJALNIH MARKERA APOPTOZE U TERAPIJI KARCINOMA KOLONA

**Ivana Damnjanović<sup>1</sup>, Gordana Kocić<sup>2</sup>, Stevo Najman<sup>3</sup>,**  
**Sanja Stojanović<sup>3</sup>, Andrej Veljković<sup>2</sup>, Srđan Pešić<sup>4</sup>**

<sup>1</sup>Katedra za farmaciju, Univerzitet u Nišu - Medicinski fakultet, <sup>2</sup>Institut za biohemiju, Univerzitet u Nišu - Medicinski fakultet, <sup>3</sup>Institut za biologiju i humanu genetiku, Univerzitet u Nišu - Medicinski fakultet, <sup>4</sup>Katedra za farmakologiju, Univerzitet u Nišu - Medicinski fakultet (Srbija)

Mnoge komponente mitohondrijalnog puta apoptoze su deregulisane u ćelijama karcinoma. Članovi Bcl-2 familije proteina predstavljaju važne markere ćelijske apoptotične funkcije, igrajući važnu ulogu u unutrašnjoj, mitohondrijalnoj kaskadi apoptoze. Cilj ovog istraživanja bio je ispitati efekat alfa-liponske kiseline (ALA) same ili u kombinaciji sa cisplatinom (CP) i 5-fluorouracilom (FU) na proliferaciju i Bcl-2/Bax ekspresiju u kulturi Caco-2 ćelija, humanih ćelija karcinoma kolona.

Ispitivan je efekat različitih koncentracija ALA, same ili u kombinaciji sa CP i FU na proliferaciju Caco-2 ćelija MTT testom. Kvantitativna ekspresija Bcl-2 i Bax proteina u kulturi Caco-2 ćelija takođe je ispitivana. Podaci su analizirani SPSS programom, verzija 17.0, Čikago, USA. Rezultati su prikazani kao srednja vrednost absorbance ± SD.

ALA pokazuje citotoksični i antiproliferativni efekat u kulturi Caco-2 ćelija. Ispitivane supstance pokazuju tendenciju smanjenja Bcl-2 i povećanja nivoa Bax ekspresije u poređenju sa kontrolnim uzorcima. ALA dovodi do signifikantne inhibicije ekspresije Bcl-2 proteina u koncentraciji od 1000 µM. Odnos Bax/Bcl-2 proteina može uticati na osetljivost ćelije na apoptozu, kao i progresiju i agresivnost karcinoma, dok povećana ekspresija antiapoptotičkih proteina, ne samo da doprinosi progresiji karcinoma, već potpomaže razvoju rezistence na primjenjeni terapijski protokol.

Prema našim rezultatima, alfa-lipoinska kiselina se može smatrati obećavajućim agensom u borbi protiv karcinoma kolona uzimajući u obzir njenu efiksanost i značajni uticaj na mitohondrijalni put apoptoze. ALA dovodi do smanjenja ekspresije Bcl-2 i povećanja ekspresije Bax proteina kao važnih regulatornih činioca apoptoze. Fokusiranje na mitohondrijalne proteine kao dela apoptotičnog puta može biti atraktivn koncept za pronalaženje novih antikancerskih lekova, međutim još uvek postoje veliki izazovi koje treba prevazići.

*Istraživanje je podržano od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije (projekat br. TR 31060 i projekat br. III41017).*

## **THE ROLE AND SIGNIFICANCE OF MITOCHONDRIAL MARKERS OF APOPTOSIS IN COLON CANCER TREATMENT**

**Ivana Damnjanović<sup>1</sup>, Gordana Kocić<sup>2</sup>, Stevo Najman<sup>3</sup>,**  
**Sanja Stojanović<sup>3</sup>, Andrej Veljković<sup>2</sup>, Srđan Pešić<sup>4</sup>**

<sup>1</sup>Department of Pharmacy, University of Niš - Faculty of Medicine, <sup>2</sup>Institute of Biochemistry, University of Niš - Faculty of Medicine, <sup>3</sup>Institute of Biology and Human Genetics, University of Niš - Faculty of Medicine, Serbia, <sup>4</sup>Department of Pharmacology, University of Niš - Faculty of Medicine (Serbia)

Many components of the mitochondrial apoptosis pathway are deregulated in cancer cells. Members of Bcl-2 family proteins are important markers of cell apoptotic function and they play a major role in the intrinsic mitochondrial apoptotic cascade. The aim of this study was to examine the effects of both pure alpha-lipoic acid (ALA)/ALA combined with cisplatin (CP) and 5-fluorouracil (FU) on the proliferation and Bcl-2/Bax quantitative expression in human colon cancer Caco-2 cell line.

We examined the effect of different concentrations of both pure ALA or combined with CP and FU on proliferation of Caco-2 by MTT test. Bcl-2 and Bax quantitative expression were also performed. The data were analyzed by SPSS (v. 17.0). The results were in the range of the average value of absorbance  $\pm$  SD.

The research results show that ALA exerts cytotoxic and antiproliferative effects on Caco-2 cells. The tested compounds tended to decrease Bcl-2 and increase Bax expression levels, compared with control samples. It was found that ALA exerts a significant inhibitory effect on Bcl-2 expression at the concentration of 1000  $\mu$ M. Bax/Bcl-2 ratio can act as a rheostat which determines cell susceptibility to apoptosis and affects tumor progression and aggressiveness as well. However, an over expression of prosurvival proteins not only contributes to the progression of cancer, but also confers resistance to the therapeutic treatments.

According to our results, alpha-lipoic acid may be considered a promising agent in the battle against colon cancer due to its efficiency and significant impact on mitochondrial apoptosis pathway. This way ALA inhibits Bcl-2 or activates Bax apoptotic checkpoints and regulators. Targeting of Bcl-2 family proteins as a part of apoptotic pathway may be an attractive concept to finding new anticancer therapies, although there are still huge challenges to meet.

*The study was supported by the Ministry of Science and Technological Development of the Republic of Serbia (project no. TR 31060 and project no. III 41017).*

## INTERAKCIJE ERLOTINIBA U TERAPIJI ONKOLOŠKIH BOLESNIKA NA KLINICI ZA PLUĆNE BOLESTI

**Tijana Kovačević<sup>1</sup>, Branislava Miljković<sup>2</sup>, Sandra Vezmar-Kovačević<sup>2</sup>,  
Mirko Stanetić<sup>3</sup>, Peđa Kovačević<sup>4</sup>**

<sup>1</sup>Klinička apoteka, Univerzitetski Klinički Centar Republike Srpske, <sup>2</sup>Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija), <sup>3</sup>Klinika za plućne bolesti, Univerzitetski Klinički Centar Republike Srpske, <sup>4</sup>Klinika intenzivne medicine za nehirurške grane, Univerzitetski Klinički Centar Republike Srpske (Bosna i Hercegovina)

Karcinom pluća predstavlja najčešći uzrok smrti od malignog oboljenja, odmah nakon karcinoma prostate kod muškaraca i karcinoma dojke kod žena. Erlotinib je inhibitor tirozin kinaze indikovan za liječenje nemikrocelularnog karcinoma pluća sa aktiviranom mutacijom receptora epidermalnog faktora rasta. Erlotinib ulazi u klinički značajne interakcije sa drugim lijekovima što za posljedicu ima promjenu serumske koncentracije erlotiniba.

Cilj rada je da se pokaže da učešće kliničkog farmaceuta u onkološkom konzilijumu na klinici za plućne bolesti značajno smanjuje broj interakcija u liječenju oboljelih od nemikrocelularnog karcinoma pluća ciljanom terapijom erlotinibom.

Grupa od 44 bolesnika označena je kao intervencijska grupa i podaci o njima su prikupljeni prospektivno u vremenskom periodu od 01.01.2017.-01.05.2018. godine za vrijeme učešća na redovnom onkološkom konzilijumu. Kontrolnu grupu sačinjavalo je 44 od ukupno 110 bolesnika koji su liječeni lijekom erlotinib od kada je dostupan u Univerzitetskom Kliničkom Centru republike Srpske (UKC RS) izabranih uparivanjem sa ispitanicima iz intervencijske grupe (*matched pair analiza*), a prije uključivanja kliničkog farmaceuta u rad onkološkog konzilijuma na Klinici za plućne bolesti UKC RS.

Klinički značajne interakcije identifikovane su kod čak dvije trećine ispitanika u studiji (57 od 88). U najvećem broju interakcija, čak 38%, dolazi do smanjenja serumske koncentracije erlotiniba. Klinički farmaceut je dao sugestije za 32 od 44 (72,72%) ispitanika od kojih je većina prihvaćena od strane ljekara. U intervencijskoj grupi bilo je statistički značajno manje klinički značajnih interakcija u odnosu na kontrolnu grupu (10 vs 24,  $p = 0,044$ ).

Značajno manji broj klinički značajnih interakcija sa erlotinibom u intervencijskoj grupi pacijenata ukazuje na doprinos kliničkog farmaceuta u sprovođenju racionalne terapije pacijenata sa karcinomom pluća.

## **INTERACTIONS OF ERLOTINIB IN THE TREATMENT OF ONCOLOGIC PATIENTS AT THE LUNG DISEASE CLINIC**

**Tijana Kovačević<sup>1</sup>, Branislava Miljković<sup>2</sup>, Sandra Vezmar-Kovačević<sup>2</sup>, Mirko Stanetić<sup>3</sup>, Peđa Kovačević<sup>4</sup>**

<sup>1</sup>Pharmacy Department, University Clinical Centre of the Republic of Srpska,

<sup>2</sup>Department of Pharmakocinetics and Clinical Pharmacy, University of

Belgrade - Faculty of Pharmacy (Serbia), <sup>3</sup>Lung Disease Clinic, University Clinical Centre of the Republic of Srpska, <sup>4</sup>Clinic for nonsurgical intensive care medicine, University Clinical Centre of the Republic of Srpska (Bosnia and Herzegovina)

Lung cancer is the most common cause of death from malignancy, immediately after prostate cancer in men and breast cancer in women. Erlotinib is a tyrosine kinase inhibitor indicated for treating non-small cell lung cancer with an activated epidermal growth factor receptor mutation. Erlotinib interacts with other medicinal products, which results in a change of erlotinib serum concentration.

The aim of the study was to demonstrate that clinical pharmacist, as a member of oncology consilium at the lung disease clinic, significantly reduces the number of erlotinib interactions in the treatment of non-small cell lung cancer patients.

A group of 44 patients was labeled as intervention group and they were analysed prospectively in the period from 01.01.2017. - 01.05.2018. during clinical pharmacist's participation in the regular oncology consilium. The control group consisted of 44 out of 110 patients treated with erlotinib since it was available in University Clinical Centre of the Republic of Srpska, match paired with patients in intervention group, and before the involvement of a clinical pharmacist in the oncology consilium at the University Clinical Centre of the Republic of Srpska lung diseases clinic. Clinically significant interactions were identified in two-thirds of studied patients (57 out of 88). Most drug interactions, 38%, reduce serum concentration of erlotinib. Clinical pharmacist gave suggestions for 32 out of 44 (72.72%) patients, most of which were accepted by doctors. In the intervention group there were statistically significantly less clinically significant interactions compared to the control group (10 vs 24, p = 0.044). A significantly lower number of clinically significant erlotinib interactions in the intervention group of patients advocates the contribution of a clinical pharmacist in the implementation of rational therapy for lung cancer patients.

## POTENCIJALNE INTERAKCIJE LEKOVA KOD PACIJENATA SA HIPERTENZIJOM

**Zorica Cvetković<sup>1</sup>, Aneta Perić<sup>2</sup>, Ana Udilović<sup>3</sup>**

<sup>1</sup>Vojna apoteka „Slavija”, Centralna apoteka, <sup>2</sup>Sektor za farmaciju, Vojnomedicinska akademija, Medicinski fakultet Vojnomedicinske akademije, Univerzitet odbrane, <sup>3</sup>Medicinski fakultet Vojnomedicinske akademije, Univerzitet odbrane (Srbija)

Interakcije lek-lek (DDI) predstavljaju ozbiljan problem među hipertenzivnim pacijentima koji u terapiji koriste veći broj lekova (polifarmacija). Poznavanje potencijalnih DDI-a (pDDI) može pomoći lekarima da minimiziraju štetne efekte pažljivim kombinovanjem upotrebljenih lekova. Cilj naše studije je bio identifikovanje najčešćih pDDI među hipertenzivnim pacijentima i procena mehanizama nastanka i ozbiljnosti interakcija.

Prospektivna, opservaciona studija je sprovedena među hipertenzivnim ambulantnim pacijentima lečenim na Vojnomedicinskoj akademiji u periodu od mesec dana. Studijom su obuhvaćeni pacijenti oba pola stariji od 18 godina koji su u terapiji koristili dva ili više leka. Softver za kontrolu interakcije lekova *Medscape* korišćen je za identifikaciju i analizu pDDI. Deskriptivna statistika i višestruka analiza linearne regresije sprovedena je upotrebom PASW 18.0 (SPSS Inc, Chicago, Illinois).

Od 350 pacijenata u starosnoj grupi od 36 do 98 godina (prosečno  $75 \pm 11$ ), bilo je neznatno više žena (51,4%). Kod 72,6% pacijenata identifikovane su pDDI od kojih je 18,29% imalo ozbiljne, 65,71% značajne i 28,86% beznačajne pDDI. Utvrđena je pozitivna korelacija između broja propisanih lekova i pDDI ( $r = 0,709$ ;  $p < 0,001$ ). Upotreba digoksina, nesteroidnih antiinflamatornih lekova, antikoagulantnih lekova i statina pozitivan je prediktor za ozbiljne pDDI. Broj značajnih interakcija je niži u poređenju sa drugim studijama u kojima se broj kretao u rasponu od 71,29% do 95,42%. Utvrđena je pozitivna korelacija između pDDI i broja propisanih recepata ( $r = 0,788$ ,  $p < 0,001$ ).

Studija je istakla podložnost hipertenzivnih pacijenata na pDDI i posledično pojavu neželjenih reakcija na lekove. Pacijenti na terapiji sa više lekova su skloniji lek-lek interakcijama.

## POTENTIAL DRUG INTERACTIONS IN HYPERTENSIVE PATIENTS

**Zorica Cvetković<sup>1</sup>, Aneta Perić<sup>2</sup>, Ana Udilović<sup>3</sup>**

<sup>1</sup>Military Pharmacy „Slavija”, Central Department for Pharmacy Service, <sup>2</sup>Sector for Pharmacy, Military Medical Academy, Medical Faculty of the Military Medical Academy, University of Defense, <sup>3</sup>Medical Faculty of the Military Medical Academy, University of Defense (Serbia)

Drug-drug interactions (DDIs) are a serious concern among hypertensive patients receiving multidrug therapy. Knowing the potential DDIs (pDDIs) may help physicians minimize adverse effects by careful combining the drugs that are used. Purpose of our study was to identify the most common pDDIs among hypertensive patients and evaluate the mechanism and severity of such interactions.

A prospective, observational study was conducted among the hypertensive outpatients treated at the Military Medical Academy over the period of one month. Patients of both genders over the age of 18 taking two or more drugs were included in the study. Medscape drug interaction checker software was used to identify and analyze the pattern of pDDIs. Descriptive statistics and multiple linear regression analysis were performed using PASW 18.0 (SPSS Inc, Chicago, Illinois).

Among 350 patients in the age group of 36 to 98 years (average  $75 \pm 11$ ), most were female (51.4%). There were 72.6% patients with pDDIs of which 18.29% patients had serious, 65.71% significant and 28.86% minor pDDIs. A positive correlation was observed between the number of drugs prescribed and pDDIs ( $r=0.709$ ;  $p<0.001$ ). Use of digoxin, nonsteroidal anti-inflammatory drugs, anticoagulant drugs and statins was found to be a positive predictor for serious pDDIs. The number of significant interactions was lower compared to other studies in which the number ranged from 71.29% to 95.42%. A positive correlation between pDDI and the number of prescribed recipes was determined ( $r = 0.788$ ,  $p < 0.001$ ).

The study highlighted the susceptibility of hypertensive patients to DDIs and therefore adverse drug reactions. Patients on multidrug therapy are more prone to these interactions.

## POVRATAK U BUDUĆNOST: KAKO RACIONALIZOVATI BOLNIČKU POTROŠNJU ANTIBIOTIKA?

**Aneta Perić<sup>1,2</sup>, Bojana Milenković<sup>1</sup>, Jelena Brusić-Renaud<sup>1</sup>,  
Mirjana Antunović<sup>1,2</sup>, Vesna Šuljagić<sup>2,3</sup>**

<sup>1</sup>Služba za farmaceutsku delatnost, Vojnomedicinska akademija, <sup>2</sup>Medicinski fakultet Vojnomedicinske akademije, Univerzitet odbrane, <sup>3</sup>Odsek za prevenciju i kontrolu bolničkih infekcija, Vojnomedicinska akademija (Srbija)

Antibiotici su najčešće korišćeni lekovi u bolničkim uslovima. Praćenje i predviđanje potrošnje antibiotika u bolnicama je značajno za optimizaciju terapije. Prognoza vremenskih serija je metoda koja se koristi za predviđanje buduće potrošnje, a bazira se na analizi trendova potrošnje u proteklom, dužem vremenskom periodu.

Cilj ove studije je da se proceni potrošnja antibiotika u Vojnomedicinskoj akademiji (VMA), posebno cefalosporina i karbapenema, i da se predvidi potrošnja u narednom petogodišnjem periodu.

Retrospektivna studija je sprovedena u periodu od 2001-2017. godine u VMA, tercijernoj, univerzitetskoj bolnici, sa 1200 kreveta. Analizirani su antibiotici za sistemsku primenu (Anatomsko-terapijsko-hemijska (ATC) klasifikacija - J01-antibiotici za sistemsku primenu). Korišćeni su podaci o potrošnji antibiotika iz bolničkog informacionog sistema, a potrošnja je izražena kao Definisana dnevna doza na 100 bolničkih dana (DDD/100 BD), korišćenjem ATC/DDD indeksa za 2017. godinu. Podaci o potrošnji su prikazani kao srednja vrednost  $\pm$  standardna devijacija. Prognoza vremenskih serija je sprovedena korišćenjem srednjih vrednosti podataka za svaku grupu antibiotika, za svaku analiziranu godinu. Na osnovu trenda srednjih vrednosti predvideli smo buduću potrošnju u odabranom petogodišnjem vremenskom periodu (2018-2022. godine) pomoću ARIMA predikcionog modela.

Prosečna potrošnja antibiotika je bila  $51,3 \pm 7,9$  DDD/100 BD. Najčešće korišćeni su bili cefalosporini ( $15,1 \pm 2,8$  DDD/100 BD), a zatim karbapenemi ( $2,9 \pm 1,6$  DDD/100 BD). Trendovi potrošnje pokazali su da bi očekivano povećanje bilo  $0,2$  DDD/100 BD godišnje za cefalosporine i  $0,3$  DDD/100 BD za karbapeneme. Njihova potencijalna prekomerna upotreba će dovesti do visoke stope rezistencije Gram-negativnih bakterija i porasta broja gljivičnih infekcija. Procenjeno je da je antibiotska hirurška profilaksa bila razlog velike potrošnje cefalosporina. Pokazano očekivano povećanje potrošnje antibiotika u bolničkim uslovima lečenja naglašava potrebu za pažljivim praćenjem korišćenja ovih lekova.

## **BACK TO THE FUTURE: HOW TO OPTIMIZE HOSPITAL ANTIBIOTIC CONSUMPTION?**

**Aneta Perić<sup>1,2</sup>, Bojana Milenković<sup>1</sup>, Jelena Brusić-Renaud<sup>1</sup>,  
Mirjana Antunović<sup>1,2</sup>, Vesna Šuljagić<sup>2,3</sup>**

<sup>1</sup>Department of Pharmacy, Military Medical Academy, <sup>2</sup>Faculty of Medicine of the Military Medical Academy, University of Defence, <sup>3</sup>Department of Infection Control, Military Medical Academy (Serbia)

Antibiotics are the most frequently used drugs in hospitalized patients. Investigating, monitoring and predicting the consumption of antibiotics in hospital is necessary in order to encourage prudent use of these drugs. Forecasting analysis is the process of making predictors of the future based on past and present data and analysis of trends. The aim of this study was to evaluate the antibiotic consumption in the Military Medical Academy (MMA), especially cephalosporins and carbapenems, and to predict future consumption in a five-year period.

The retrospective study was conducted from 2001 to 2017 in MMA, Belgrade, a tertiary, university hospital, with 1200 beds. Antibacterials for systemic use (Anatomical Therapeutic Chemical (ATC) code J01) were included in this study. Data regarding the use of antibiotics were extracted from hospital computer system and expressed as Defined Daily Dose per 100 bed days (DDD/100 BD), using the 2017 version of the ATC/DDD index. Results are expressed as mean value  $\pm$  standard deviation. Forecasting analysis was performed on mean values of all data for each group of antibiotics in a single year. Based on mean value trend we predicted how this variable is likely to behave in the future, using ARIMA prediction model in a selected time horizon (2018-2022).

The average antibiotic consumption was  $51.3 \pm 7.9$  DDD/100 BD. The most frequently used were cephalosporins ( $15.1 \pm 2.8$  DDD/100 BD), followed by carbapenems ( $2.9 \pm 1.6$  DDD/100 BD). Trends of consumption showed that the expected increase would be 0.2 and 0.3 DDD/100 BD per year for cephalosporins and carbapenems, respectively. Potential overuse will lead to the high rate of both Gram-negative bacteria resistance and fungal infections. It is estimated that surgical antibiotic prophylaxis is the reason for high levels of cephalosporin consumption. Demonstrated expected increase in antibiotic consumption highlights the need for careful drug use monitoring.

## TROVANJA OLANZAPINOM U NACIONALNOM CENTRU ZA KONTROLU TROVANJA SRBIJE U 2017. GODINI

**Snežana Đorđević<sup>1</sup>, Vesna Kilibarda<sup>1</sup>, Gordana Brajković<sup>2</sup>,  
Nataša Perković Vukčević<sup>1</sup>, Gordana Vuković Ercegović<sup>1</sup>,  
Jasmina Jović Stošić<sup>1</sup>, Slavica Vučinić<sup>1</sup>**

<sup>1</sup>Vojnomedicinska akademija - Medicinski fakultet, Univerzitet Odbrane,

Beograd, <sup>2</sup>Nacionalni centar za kontrolu trovanja, Beograd (Srbija)

Olanzapin je lek koji pripada novoj generaciji antipsihotika. Koristi se za lečenje šizofrenije i bipolarnog poremećaja ličnosti. Efekti nastaju verovatno kao rezultat blokiranja ili antagonizovanja dopaminskih D<sub>2</sub> receptora. Kao i drugi netipični antipsihotici olanzapin je jak antagonist 5-HT<sub>2A</sub> serotoninskih receptora. Akutna trovanja olanzapinom su retka. Simptomi trovanja podrazumevaju dublji ili fluktuirajući poremećaj stanja svesti sa hipersalivacijom i miozom. Visoke doze mogu da uzokuju komu i smrt. Terapijske koncentracije olanzapina u krvi su u opsegu od 0,01-0,05 mg/L. Letalan ishod može nastati pri koncentracijama olanzapina u plazmi većim od 1 mg/L.

Cilj ovog rada je da prikaže slučajeve akutnih trovanja olanzapinom u Nacionalnom centru za kontrolu trovanja (NCKT) Srbije u 2017. godini.

Prema podacima NCKT registrovan je 31 pacijent (26 žena i 6 muškaraca) zbog sumnje na trovanje olanzapinom. Svi pacijenti su imali olanzapin u svojoj redovnoj terapiji. Određivanje koncentracije olanzapina vršeno je validiranim metodom tečne hromatografije sa masenom spektrometrijom (LC-MS).

Rezultati pokazuju da je 15 pacijenata (10 žena i 5 muškaraca) imalo terapijske koncentracije olanzapina u krvi, a kod 16 pacijenata (15 žena i 1 muškarac) su registrovane toksične i čak letalne koncentracije olanzapina. Jedna pacijentkinja je zbog pokušaja suicida bila hospitalizovana 2 puta u toku tri meseca. Koncentracije olanzapina na prijemu od 1,75 mg/L i 2,44 mg/L su u oba slučaja bile letalne. Ona je imala karakterističnu kliničku sliku trovanja olanzapinom. Hospitalizacija je trajala više od jedne nedelje, ali je bila sa povoljnijim ishodom.

Trovanja olanzapinom su retka i uglavnom lakšeg stepena. Međutim, u izvesnim slučajevima visoke koncentracije mogu da izazovu ozbiljna trovanja. S obzirom da ne postoji antidot kod trovanja olanzapinom, primena adekvatne simptomatske i suportivne terapije dovodi do povoljnog terapijskog ishoda čak i u slučajevima trovanja sa visokim koncentracijama olanzapina.

## **OLANZAPINE INTOXICATIONS IN NATIONAL POISON CONTROL CENTER SERBIA IN 2017**

**Snežana Đorđević<sup>1</sup>, Vesna Kilibarda<sup>1</sup>, Gordana Brajković<sup>2</sup>,  
Nataša Perković Vukčević<sup>1</sup>, Gordana Vuković Ercegović<sup>1</sup>,  
Jasmina Jović Stošić<sup>1</sup>, Slavica Vučinić<sup>1</sup>**

<sup>1</sup>Medical Faculty Military Medical Academy, University of Defense, Belgrade,

<sup>2</sup>National Poison Control Centre, Belgrade (Serbia)

Olanzapine is the drug that belongs to the group of new generation of antipsychotics. It is used for treating of schizophrenia and bipolar disorder. The effects are probably result of blocking or antagonizing of dopamine D2 receptors. Like other atypical antipsychotics, olanzapine also strongly antagonizes the 5-HT2A serotonin receptor. Acute poisonings with olanzapine are rare. Symptoms of an overdose include deeper or fluctuating disorder of consciousness with hypersalivation and miosis. High doses can cause coma and death. Therapeutic blood concentration of olanzapine is in the range of 0.01-0.05 mg/L. Fatalities generally have occurred with olanzapine plasma concentrations greater than 1 mg/L.

The aim of this work is to present cases of acute poisonings with olanzapine in National Poison Control Centre (NPCC) Serbia in 2017.

According to NPCC data, there were 31 patients (26 female and 6 male) under suspicion of olanzapine intoxication. All of the patients had olanzapine in their regular therapy. Determination of olanzapine was done by validated liquid chromatographic mass spectrometric (LC-MS) method.

Results showed that 15 patients (10 female and 5 male) had olanzapine concentration in the therapeutic range, and 16 in toxic and even lethal range (15 female and 1 male). One of the patients with a suicide attempt has been hospitalized two times during the three months period. Olanzapine concentrations of 1.75 mg/L and 2.44 mg/L after reception in both cases were in the fatal range. She had characteristic clinical picture for acute olanzapine poisoning. Hospitalization lasted more than one week, but with favorable outcome.

Olanzapine intoxications are rare and mostly with mild degree. But in some cases, high concentration can cause severe intoxication. Since there is not antidote for olanzapine poisoning, applying of adequate symptomatic and supportive therapy lead to favorable outcome even in cases with high olanzapine concentration.

## EFIKASNOST OMEGA-3 MASNIH KISELINA U PREVENCICI KARDIOVASKULARNIH BOLESTI: DOKAZI I PREPORUKE

Silva Dobrić

Univerzitet odbrane - Medicinski fakultet Vojnomedicinske akademije, Institut za naučne informacije, Beograd (Srbija)

Veći broj opservacionih studija ukazao je na povezanost redovnog konzumiranja ribe bogate omega-3 masnim kiselinama (omega-3 MK) sa smanjenim rizikom od smrti zbog kardiovaskularnih bolesti (KVB). Ova zapažanja rezultirala su povećanim interesom za primenu dijetetskih suplemenata sa omega-3 MK u prevenciji KVB. Međutim, kliničke studije, u kojima je ispitivana njihova protektivna efikasnost kod osoba sa povećanim rizikom od KVB, dale su kontradiktorne rezultate. Stoga je cilj ovog rada bio da se kritički analiziraju rezultati studija objavljeni u poslednjih 10 godina kako bi se sagledala uloga i mesto suplemenata sa omega-3 MK u smanjenju fatalnih i nefatalnih kardiovaskularnih događaja kod osoba sa povišenim rizikom od KVB.

Analizom su bili obuhvaćeni rezultati kontrolisanih kliničkih studija i meta-analiza o upotrebi suplemenata sa omega-3 MK u prevenciji KVB objavljeni u časopisma indeksiranim u bazi *PubMed/MEDLINE* u periodu 2008-2018.

Rezultati analiziranih kliničkih studija, većinom dvostruko-slepih i kontrolisanih placeboom, pokazali su da redovno uzimanje suplemenata sa omega-3 MK u dozi 0,5-2 g/dan nije dovelo do značajnog smanjenja neželjenih kardiovaskularnih događaja kod osoba sa povišenim rizikom od KVB. Ovi nalazi ne podupiru preporuke koje sugerisu upotrebu otprilike 1 g/dan omega-3 MK kod osoba sa istorijom ishemiske bolesti srca. Trenutno je u toku nekoliko velikih studija, u koje je uključeno blizu 55000 osoba sa rizikom od razvoja velikih kardiovaskularnih događaja, u kojima se ispituje protektivnu efikasnost omega-3 MK, primenjenih u dozama od 3-4 g/dan. Rezultati ovih studija treba da pokažu da li će više doze omega-3 MK, od trenutno preporučenih, imati značajan efekat na smanjenje rizika od neželjenih kardiovaskularnih događaja.

Rezultati novijih kliničkih studija pokazuju da upotreba suplemenata sa omega-3 MK nema značajan efekat u prevenciji fatalnih i nefatalnih vaskularnih događaja kod osoba sa povišenim rizikom od KVB.

# **EFFECTIVENESS OF OMEGA-3 FATTY ACIDS IN PREVENTION OF CARDIOVASCULAR DISEASES: EVIDENCE AND RECOMMENDATIONS**

**Silva Dobrić**

University of Defense - Faculty of Medicine of the Military Medical Academy,  
Institute for Scientific Information, Belgrade (Serbia)

A number of observational studies have highlighted the association of regular consumption of fish rich in omega-3 fatty acid (omega-3 FA), with a reduced risk of death from cardiovascular disease (CVD). These observations have resulted in increased interest in the use of omega-3 FA supplements in the prevention of CVD. However, clinical studies, in which their protective effectiveness was investigated in people with an increased risk of CVD, gave contradictory results. Therefore, the aim of this paper was to critically analyze the results of clinical studies published over the past 10 years to look at the role and place of omega-3 FA supplements in reducing unwanted cardiovascular events in people at high risk of CVD.

The analysis included results of controlled clinical studies and meta-analyses on the use of omega-3 supplements in the prevention of CVD, published in journals indexed at the PubMed/MEDLINE database in the period 2008-2018.

Results of analyzed studies showed that regular use of omega-3 FA (0.5-2 g / day) failed to significantly reduce fatal and non-fatal cardiovascular events in people with high risk of CVD. These findings do not support recommendations suggesting the use of approximately 1 g/day omega-3 FA in people with ischemic heart disease. Several ongoing large trials, involving almost 55000 people at risk of developing major cardiovascular events, in which the protective effectiveness of 3-4 g/day omega-3 FA is tested, will provide evidence whether higher doses of omega-3 FA than currently recommended may have a significant effect on reducing the risk of unwanted cardiovascular events.

The results of recent clinical studies show that the use of omega-3 FA supplements has no significant effect in the prevention of fatal and non-fatal vascular events in people at high risk of CVD.

## KORISTI I RIZICI UPOTREBE DIJETETSKIH SUPLEMENATA

**Davor J. Korčok, Bogdan Mitić**

Abela Pharm d.o.o., Beograd (Srbija)

Dijetetski suplementi kao farmaceutski dozirani oblici se mogu naći u slobodnoj prodaji u apotekama na internetu, drogerijama i drugim objektima i važno je pravilno proceniti njihove koristi kao i rizike. Cilj ovog rada jeste procena koristi i bezbednosnih rizika upotrebe dijetetskih suplemenata u zdravstvene svrhe kod ljudi pregledom kliničkih studija i naučnih radova.

Urađena je pretraga baza podataka na PUBMED-u i Google Scholar website-u za radove koji su objavljeni periodu od 2011 do 2018. godine. Procena studija je urađena od strane autora rada. Da bi bili uključeni u rad, studije su morale da sadrže bezbednosne aspekte i korist upotrebe dijetetskih suplemenata kod ljudi.

Pregledom velikog broja radova koji su dobijeni unosom sledećih pojmova, uz njihovu kombinaciju u pretragu: „safety”, „efficiency”, „dietary supplements” i „food supplements”, odabранo je 10 radova u kojima su praćeni pozitivni ishodi suplementacije dijetetskim suplementima kao što su: smanjenje rizika od nastanka određene bolesti, poboljšanje opšteg zdravstvenog stanja i drugi. Podaci o dijetetskim suplementima koji su sadržali sledeće supstance su razmatrani: vitamin D, kalcijum, probiotici, glukozamin, hondroitin, S-adenozilmletonin, folati, karnitin, cimet, vitamin C i druge. Rezultati su pokazali da su se dijetetski suplementi koristili u cilju poboljšanja opšteg zdravlja pojedinca ili u cilju poboljšanja određenog stanja bez dodatnih rizika po njihovo zdravlje.

Dokazi pokazuju da korist upotrebe dijetetskih suplemenata prevazilazi njihove bezbednosne rizike po zdravlje ljudi, i da se njihova upotreba generalno može preporučivati, međutim, potrebno je uraditi još istraživanja i proceniti u kojim situacijama primena suplementa neće izazvati neželjene događaje i iskazati efikasnost.

## BENEFITS AND RISKS OF DIETARY SUPPLEMENTS USE

**Davor J. Korčok, Bogdan Mitić**

Abela Pharm d.o.o., Belgrade (Serbia)

Dietary supplements as pharmaceutical dosage forms can be marketed and sold in pharmacies, on the Internet, in drugstores and other facilities, so it is important to properly assess their benefits and risks. The aim of this paper is to evaluate the benefits and safety risks of dietary supplements use for health purposes in humans by review of clinical studies and scientific papers.

Database search on PubMed and Google Scholar has been completed for the research papers published in the time period from 2011 until 2018. The evaluation of the studies was performed by the authors of this paper. In order to be included, the studies had to include the safety aspects and the benefits of dietary supplements use in humans.

After the performed review of large number of scientific papers that were found using the key words: „safety”, „efficiency”, „dietary supplements” and „food supplements”, 10 of them were included in this overview, and the positive outcomes of a supplementation with dietary supplements were analyzed such as: risk reduction for the development of certain disease, improvement of general health of the individuals, and others. Data on dietary supplements containing the following supplements were assessed: vitamin D, calcium, probiotics, glucosamine, chondroitin, S-adenosyl methionine, folate, carnitine, cinnamon, vitamin C and others. The results showed that dietary supplements were generally used to improve general health of an individual or to improve a certain health condition without additional risks to their health.

Evidence suggests that the benefits of dietary supplements use outweigh their safety risks for human health, and that their use can generally be recommended. However, further research is needed to assess the situations when the supplement use would not cause side effects and demonstrate complete efficacy.

## DIJETETSKI SUPLEMENTI SA VITAMINIMA I MINERALIMA NA TRŽIŠTU SRBIJE

**Bojana Vidović, Mirko Lazović, Lana Kostić, Slađana Šobajić**

Katedra za bromatologiju, Univerzitet u Beogradu - Farmaceutski fakultet  
(Srbija)

Vitamini i mineralne materije, kao esencijalni nutrijenti, ubrajaju se u najčešće aktivne sastojke dijetetskih suplemenata. Cilj rada bio je da se dobije uvid u zastupljenost, karakteristike sastava i hemijske oblike vitamina i minerala u dijetetskim suplementima na tržištu Srbije.

Istraživanje je sprovedeno prikupljanjem i analizom deklaracija dijetetskih suplemenata sa vitaminima i mineralima koji se nalaze na našem tržištu.

Od ukupno 705 dijetetskih suplemenata koji sadrže vitamine i/ili minerale, 19,43% je proizvedeno u Srbiji, a ostali su iz uvoza. Monokomponentnih dijetetskih suplemenata je bilo 36,4%. Od ukupnog broja polikomponentnih proizvoda, 35,8% sadrži samo vitamine, 4,1% samo minerale, dok ostatak čine kombinacije vitamina i minerala, sa i bez dodatka drugih biološki aktivnih sastojaka. Vitamin C je najzastupljeniji vitamin (u 59,4% proizvoda), slede vitamini B grupe, dok je vitamin K prisutan u svega 6,4% dijetetskih suplemenata. Najzastupljeniji makroelementi su magnezijum (41,7%) i kalcijum (35,6%), a od mikroelemenata cink (36,9%) i selen (26,7%). Najčešće korištene organske soli kao izvori minerala su citrati i glukonati, a od neorganskih izvora: karbonati, oksidi i sulfati. Prirodni izvori vitamina zastupljeni su u 6,2% dijetetskih suplemenata. Upoređivanjem dnevnih doza vitamina i minerala (na osnovu predviđenog načina upotrebe navedenog na deklaraciji) sa njihovim nutritivnim referentnim vrednostima (NRV), utvrđeno je da veliki broj dijetetskih suplemenata sadrži vitamine i minerale u količinama većim od 150% NRV.

Na tržištu se nalazi veliki broj dijetetskih suplemenata sa vitaminima i/ili mineralima, uključujući i one čijom se primenom u organizam unose količine vitamina i minerala koje su višestruko veće od 100% NRV. U cilju racionalne i bezbedne upotrebe ovih proizvoda veoma je značajna savetodavna uloga zdravstvenih radnika, prvenstveno farmaceuta.

## VITAMIN AND MINERAL SUPPLEMENTS ON THE SERBIAN MARKET

**Bojana Vidović, Mirko Lazović, Lana Kostić, Slađana Šobajić**

Department of Bromatology, University of Belgrade - Faculty of Pharmacy  
(Serbia)

Vitamins and minerals, as essentials nutrients, are the most common active compounds in dietary supplements. The aims of this work were to investigate the presence, sources, and amounts of vitamins and minerals in the dietary supplements on Serbian market.

The research was conducted by analyzing the declarations of dietary supplements from Serbian market.

During the survey, it was established that 705 dietary supplements contained vitamins and/or minerals. In Serbia 137 dietary supplements (19.43%) have been produced, and the rest was imported. There were 36.4% mono-component products. Among the multicomponent, 35.8% contain only vitamins, 4.1% contain only minerals, and the rest was the combination of vitamins and minerals with or without the addition of other bioactive components. The most common vitamin was vitamin C (59.5%), followed by B-vitamins group while the least frequent vitamin K was present in only 6.4% of the products. The most common macroelements were magnesium (41.7%) and calcium (35.6%). Zinc and selenium were the most common microelements present in 36.9% and 26.7% of dietary supplements, respectively. The most common organic salts of minerals were citrates and gluconates; among inorganic sources, these were carbonates, oxides, and sulfates. Natural vitamin sources were present in 6.2% of the dietary supplements. The recommended daily dosage vitamins and minerals have been compared with the reference doses for nutrition labeling (NRV) and a large number of dietary supplements have exceeded the value of 150% NRV.

On the market, there is a large number of dietary supplements with the content of vitamins and minerals higher than NRV values. In order to improve rational and safe use of dietary supplements, the consumers before use need to consult with pharmacists or other health professionals.

## ISPITIVANJE BRZINE RASTVARANJA RESVERATROLA IZ DIJETETSKIH SUPLEMENATA

**Mira Mikulić, Ljilja Torović, Milica Atanacković Krstonošić,  
Veljko Ćućuz, Jelena Hogervorst**

Katedra za farmaciju, Univerzitet u Novom Sadu - Medicinski fakultet (Srbija)

Resveratrol (*trans*-3,5,4'-trihidroksistilben) je fitoaleksin koji pripada grupi stilbena - fenolnih jedinjenja prisutnih u različitim biljnim vrstama. Njegovi najpoznatiji prirodni izvori su crveno vino, grožđe, kikiriki i borovnice. Na tržištu raste broj dijetetskih suplemenata koji sadrže ovo jedinjenje, jer se smatra da može imati značajnu ulogu u prevenciji kardiovaskularnih i malignih oboljenja, pre svega zbog svoje antioksidativne i antiinflamatorne aktivnosti. Cilj ovog rada je određivanje brzine rastvaranja resveratrola iz komercijalno dostupnih dijetetskih suplemenata koji sadrže resveratrol ili ekstrakt grožđa.

Ispitivano je 12 različitih dijetetskih suplemenata koji sadrže resveratrol (7 uzoraka, F6-F12) ili neku vrstu ekstrakta grožđa (5 uzoraka, F1-F5). Test brzine rastvaranja rađen je na ERWEKA DT800 aparatu u acetatnom puferu pH 4,5, a uzorkovanje je vršeno nakon 45 minuta. Identifikacija i kvantifikacija resveratrola je izvedena na Agilent 1100 Series tečnom hromatografu uz upotrebu Poroshell 120 EC-C18 kolone, UV detekciju na 305 nm i gradijentnu eluciju.

Od 12 testiranih suplemenata, resveratrol je kvantifikovan u samo 4 suplementa (F6, F7, F8, F12) koji su svi imali deklarisan sadržaj ove komponente. Među njima, jedino je kod dve kapsule suplementa F7 utvrđeno rastvaranje više od 75% u odnosu na deklarisani sadržaj. U ostalim uzorcima količina rastvorenog resveratrola nije prelazila 33%. U uzorcima na bazi ekstrakta grožđa, resveratrol nije detektovan. Dobijeni rezultati mogu biti posledica slabe rastvoljivosti resveratrola, neadekvatne formulacije, ali i niskog sadržaja u samim suplementima.

Budući da nijedan od testiranih suplemenata nije pokazao zadovoljavajuću brzinu oslobađanja resveratrola, potrebno je posebno obratiti pažnju na ovu vrstu komercijalno dostupnih preparata. Svakako, ovakvi rezultati dovode u pitanje i potencijalnu efikasnost ovih preparata u ljudskom organizmu.

## **ANALYSIS OF RESVERATROL DISSOLUTION FROM DIETARY SUPPLEMENTS**

**Mira Mikulić, Ljilja Torović, Milica Atanacković Krstonošić,  
Veljko Ćućuz, Jelena Hogervorst**

Department of Pharmacy, University of Novi Sad - Faculty of Medicine (Serbia)

Resveratrol (*trans*-3,5,4'-trihydroxystilbene) is a phytoalexin from group of stilbenes – phenolic compounds present in various plant species. Natural sources rich in resveratrol include red wine, grapes, peanuts and blueberries. Number of dietary supplements containing resveratrol on the market is rising, mainly because of its potential role in the prevention of cardiovascular and malignant diseases as well as antiaging properties. It has been shown that this component possesses antioxidant and anti-inflammatory activity. Therefore, the aim of this study was assessment of dissolution of dietary supplements containing resveratrol or grape extract.

Twelve different dietary supplements with declared resveratrol (7 samples, F6-F12) or grape extract content (5 samples F1-F5) were analyzed. Dissolution test was performed on ERWEKA DT800 apparatus in acetate buffer pH 4.5, and sampling was done after 45 minutes. Identification and quantification of resveratrol was performed on Agilent 1100 Series liquid chromatograph with Poroshell 120 EC-C18 column, UV detection on 305 nm and gradient elution.

From total of 12 analyzed supplements, resveratrol was quantified only in 4 (F6, F7, F8, F12) which all had declared resveratrol content. Among them, more than 75% of the declared content was dissolved only from two capsules of supplement F7. In the rest of the samples, amount of dissolved resveratrol did not exceed 33% of the declared content. In 5 dietary supplements with grape extracts, resveratrol was not detected. Obtained results can be consequence of different factors, including low solubility of resveratrol, inadequate formulation as well as its low content in analyzed samples.

Since none of the tested supplements has shown optimal dissolution properties, there is a need for more detailed analysis of resveratrol supplements present on the market. Additionally, these results indicate that potential efficacy of this type of preparations in humans should be further studied.

## ISPITIVANJE ANTIMIKROBNOG POTENCIJALA ETARSKOG ULJA HERBE CRVENOG ZDRAVCA (*GERANIUM ROBERTIANUM L.*)

**Jelena Antić Stanković<sup>1</sup>, Nikola Krstić<sup>1</sup>,**  
**Nikola Jakovljević<sup>1</sup>, Dubravka Bigović<sup>2</sup>**

<sup>1</sup>Katedra za mikrobiologiju i imunologiju, Univerzitet u Beogradu - Farmaceutski fakultet, <sup>2</sup>Institut za proučavanje lekovitog bilja „Dr Josif Pančić“ (Srbija)

Imajući u vidu sve češću pojavu rezistencije mikroorganizama prema konvencionalnim antibioticima, jedan od ciljeva istraživačkih timova širom sveta je ispitivanje antimikrobnog potencijala jedinjenja biljnog porekla i definisanje hemijskih sastojaka koji ispoljavaju antimikrobnu aktivnost. Cilj ovog rada je bio određivanje antimikrobne aktivnosti etarskog ulja crvenog zdravca, *Geranium robertianum L.*, dobijenog destilacijom nadzemnog dela biljke u cvetu, prema standardnim sojevima *Staphylococcus aureus* i *Escherichia coli*, kao i efekat kombinovane primene etarskog ulja sa amoksicilinom.

Za ispitivanje antimikrobne aktivnosti, korišćena je agar mikrodilucionna metoda. Inokulum je pripremljen u skladu sa smernicama Instituta za kliničke i laboratorijske standarde (CLSI). Sveže pripremljene kolonije vrsta *S. aureus* (ATCC 25923), *E. coli* (ATCC25922) (18-24h) su suspendovane u određenoj zapremini fiziološkog rastvora da bi se postigao turbiditet vrednosti 0,5 koja prema McFarland standardu odgovara 1-2x10<sup>8</sup> CFU/ml. U hranljivom bujonu je od pripremljene suspenzije napravljeno razblaženje 1:10 (10<sup>7</sup> CFU/ml bakterija). Za inokulaciju na površinu agara u bazenu mikrotitracione ploče korišćeno je 5 µl ovako pripremljene bakterijske suspenzije. Temperatura Mueller-Hinton bujona je bila 55±5°C, kako bi medijum bio tečan pre inkorporiranja odgovarajućih koncentracija etarskog ulja i/ili amoksicilina.

Agar mikrodilucionom metodom utvrđeno je da minimalna inhibitorna koncentracija (MIK) etarskog ulja *G. robertianum* iznosi 2,39 mg/ml prema testiranim mikroorganizmima. Poređenja radi, disk difuzionom metodom, efekat testiranog etarskog ulja uočen je pri koncentraciji koja je nekoliko puta veća. Na osnovu vrednosti indeksa frakcione inhibitorne koncentracije (FICI), zaključeno je da testirano etarsko ulje pokazuje sinergistički efekat sa amoksicilinom prema *S. aureus*, a indiferentan u kombinaciji sa amoksicilinom prema *E.coli*.

Agar mikrodilucionna metoda se pokazala kao pogodna za *in vitro* ispitivanje antimikrobne aktivnosti etarskog ulja, a izdvaja se od ostalih prema ekonomičnosti, jednostavnosti i smanjenju pojave razdvajanja hidrofobnog etarskog ulja i hidrofilnog medijuma.

## **EXPLORATION OF ANTIMICROBIAL POTENTIAL OF THE ESSENTIAL OIL FROM AERIAL PARTS OF *GERANIUM ROBERTIANUM* L.**

**Jelena Antić Stanković<sup>1</sup>, Nikola Krstić<sup>1</sup>,  
Nikola Jakovljević<sup>1</sup>, Dubravka Bigović<sup>2</sup>**

<sup>1</sup>Department of Microbiology and Immunology, University of Belgrade - Faculty of Pharmacy, <sup>2</sup>Institute for Medicinal Plant Research „Dr Josif Pančić“ (Serbia)

Bearing in mind the increasingly frequent occurrence of microbial resistance to conventional antibiotics, one of the goals of research teams around the world is investigation of the antimicrobial potential of compounds of plant origin and identification of chemical ingredients that exhibit antimicrobial activity. The aim of this work was investigation of antibacterial activity of essential oil obtained by distillation of the aerial parts of *Geranium robertianum* L. during the flowering period, against standard bacterial strains *Staphylococcus aureus* and *Escherichia coli*. Also, effect of essential oil in combination with amoxycillin was investigated.

An inoculum was prepared according to the Clinical and Laboratory Standards Institute methods (CLSI). Briefly, fresh (18–24 h) bacterial colonies of *S. aureus* (ATCC 25923) and *E. coli* (ATCC 25922) were suspended in Mueller-Hinton broth to achieve a turbidity of 0,5 McFarland standard corresponding to approx.  $1\text{--}2 \times 10^8$  colony-forming units (CFU). Next, 0,5 McFarland suspensions were diluted 1:10 in fresh Mueller-Hinton broth to obtain a concentration of  $10^7$  CFU/ml and then 5 ml of prepared bacterial suspension was applied to the surface of agar in each well of the microplate.

During the experiment, the Miller-Hinton agar temperature of  $55\pm 5^\circ\text{C}$  was maintained, so that the agar was liquid prior to mixing with essential oil or/and antibiotic solution.

Agar microdilution method showed that the minimal inhibitory concentration (MIC) of *G. robertianum* essential oil against the tested microorganisms is 2.39 mg/ml. For comparison, using the disc diffusion method, the effect of the tested essential oil was observed at a several times higher concentration. Based on the values of fractional inhibitory concentration index (FICI), it was concluded that the tested essential oil exhibits a synergistic effect with amoxicillin against *S. aureus*, and is indifferent in combination with amoxicillin according to *E. coli*.

The agar microdilution method proved to be suitable for *in vitro* testing of antimicrobial activity of essential oil, and it distinguishes itself from others in terms of economy, simplicity and reduction in the occurrence of separation of hydrophobic essential oil and hydrophilic medium.

## **EFFECT OF EXENATIDE LAR IN TYPE-2 DIABETIC PATIENTS WITH VS. WITHOUT ELEVATED ADIPO-INFLAMMATORY RISK SCORE AT BASELINE: AN 8-MONTH PROSPECTIVE INTERVENTION STUDY**

**Dragana Nikolic<sup>1</sup>, Roberta Chianetta<sup>1</sup>, Giuseppa Castellino<sup>1</sup>, Jelena Vekic<sup>2</sup>, Aleksandra Zeljkovic<sup>2</sup>, Vesna Spasojevic-Kalimanovska<sup>2</sup>, Manfredi Rizzo<sup>1</sup>**

<sup>1</sup>University of Palermo (Italy), <sup>2</sup>University of Belgrade – Faculty of Pharmacy (Serbia)

The effect of exenatide once-weekly (long-acting release, LAR) on cardiovascular risk may be mediated by the modulation of several cytokines involved in inflammatory process and endothelial function. We evaluated different cytokines associated with inflammation at the endothelial level and glycemic decompensation in subjects with type 2 diabetes (T2DM), in order to make a new adipo-inflammatory risk score. The effect of exenatide LAR was then assessed in such T2DM patients in relation to the presence of elevated adipo -inflammatory risk score at baseline.

Sixty subjects with T2DM (41 men and 19 women) naïve to incretin-based therapies were treated with exenatide LAR as add-on to metformin (from 1500 up to 3000 mg/day) for 8 months. Elevated cardiometabolic risk score at baseline was defined by the combination of the following 5 cytokines: adiponectin, leptin, resistin, monocyte chemotactic protein 1, plasminogen activator inhibitor-1, E-Selectin, and soluble intercellular adhesion molecule. The median value of each cytokine was used as cut-off for defining the „abnormal value” of each cytokine, and the cohort of patients was then subdivided in 2 groups: with elevated adipo-inflammatoty risk score at baseline (n=28) and without elevated adipo-inflammatoty risk score at baseline (n=32). Carotid intima media thickness (cIMT) was assessed by B-mode real-time ultrasound, while endothelial function by flow mediated dilation (FMD) of the brachial artery.

We found improvements in most of the investigated cardio-metabolic parameters among subjects with and without elevated adipo-inflammatoty risk score at baseline, and did not find any significant difference among the two subgroups of patients.

This study showed that exenatide LAR similarly improved cardio-metabolic parameters in T2DM subjects with vs. without elevated adipo-inflammatoty risk score at baseline. Our data somewhat extend previous findings of anti-inflammatoty effects of exenatide in animal models.

## POVEZANOST IZMEĐU OBRAZACA HOMEOSTAZE HOLESTEROLA I KONCENTRACIJA NE-HDL HOLESTEROLA KOD ZDRAVIH OSOBA I PACIJENATA SA ISHEMIJSKOM BOLEŠĆU SRCA KOJI NISU NA TERAPIJI STATINIMA

**Tamara Gojković, Sandra Vladimirov, Vesna Spasojević-Kalimanovska, Aleksandra Zeljković, Jelena Vekić, Zorana Jelić-Ivanović**

Katedra za medicinsku biohemiju, Univerzitet u Beogradu – Farmaceutski fakultet (Srbija)

Homeostaza holesterola predstavlja ravnotežu između sinteze i apsorpcije holesterola. Određivanjene holesterolskih sterola (NHS) kao markera sinteze i apsorpcije holesterola, može ukazivati na rani razvoj dislipidemije i predvideti odgovor na terapiju statinima. Ne-HDL holesterol (ne-HDL-h) je bolji prediktor rizika za ishemijsku bolest srca (IBS) od LDL holesterola.

Studija je obuhvatala 47 IBS pacijenata bez terapije statinima i 31 zdravog ispitanika (KG). Koncentracije NHS-a su kvantifikovane metodom gasne hromatografije sa plamenojonizacijonom detekcijom (GC-FID). Koncentracije ukupnog holesterola i HDL-holesterola, merene su rutinskim metodama na Ilab 300+ analizatoru.

Obrasci homeostaze holesterola su dobijeni na osnovu medijalnih vrednosti latosterola i  $\beta$ -sitosterola ( $L/\beta$ ). U KG, učesnici sa većim nivoima sinteze imali su veće vrednosti ne-HDL-h u odnosu na podgrupu sa manjom sintezom ( $p<0,05$ ). Nivoi apsorpcije u obe podgrupe su bili isti. Pacijenti bez terapije statinima sa povećanom sintezom holesterola, bez obzira na apsorpciju, imali su povećane nivoje ne-HDL-C u poređenju sa podgrupama sa slabom sintezom i slabom apsorpcijom ( $p<0,01$ , za oba poređenja).

Odnos  $L/\beta$  može biti korisna alatka za procenu individualnih obrazaca homeostaze holesterola, a samim tim i rano otkrivanje dislipidemije i predviđanje odgovora na terapiju statinima.

## **ASSOCIATION BETWEEN CHOLESTEROL HOMEOSTASIS PATTERNS AND NON-HDL CHOLESTEROL IN HEALTHY PEOPLE AND NON- STATIN TREATED CORONARY ARTERY DISEASE PATIENTS**

**Tamara Gojković, Sandra Vladimirov, Vesna Spasojević-Kalimanovska,  
Aleksandra Zeljković, Jelena Vekić, Zorana Jelić-Ivanović**

Department of Medical Biochemistry, University of Belgrade - Faculty of  
Pharmacy (Serbia)

Cholesterol homeostasis represents the balance between cholesterol synthesis and absorption. Evaluation of non-cholesterol sterols (NCSs) as synthesis and absorption markers may indicate the dyslipidemia early development and predict statin response. Non-HDL cholesterol (non-HDL-C) is a better predictor of coronary artery disease (CAD) risk than LDL cholesterol. This study investigates associations of different cholesterol homeostasis patterns with non-HDL-C concentration.

We enrolled 47 statin-untreated CAD patients and 31 controls (CG). NCSs concentrations were quantified using gas chromatography-flame ionization detection (GC-FID). Concentrations of total cholesterol (TC) and HDL-cholesterol (HDL-C) were measured by routine methods on an iLab 300+ analyzer.

Cholesterol homeostasis patterns were obtained according to lathosterol and  $\beta$ -sitosterol median values ( $L/\beta$ ). CG participants with same absorption levels, but elevated cholesterol synthesis had higher non-HDL-C concentration compared to those with reduced synthesis ( $p<0.05$ ). Statin-untreated patients with increased cholesterol synthesis, regardless of absorption, had increased levels of non-HDL-C compared to subgroups with poor synthesis and poor absorption ( $p<0.01$ , for both).

$L/\beta$  ratio could be a useful tool for estimating individual cholesterol homeostasis patterns, and consequently dyslipidemia early development and statin therapy response.

## LONG TERM EFFECTS OF LIRAGLUTIDE ON GLYCO-METABOLIC PARAMETERS AND cIMT IN PATIENTS WITH TYPE-2 DIABETES: 5 YEARS PROSPECTIVE REAL-WORLD STUDY

**Giuseppa Castellino<sup>1</sup>, Dragana Nikolic<sup>1</sup>, Roberta Chianetta<sup>1</sup>, Jelena Vekic<sup>2</sup>, Aleksandra Zeljkovic<sup>2</sup>, Vesna Spasojevic-Kalimanovska<sup>2</sup>, Manfredi Rizzo<sup>1</sup>**

<sup>1</sup>University of Palermo, (Italy), <sup>2</sup>University of Belgrade – Faculty of Pharmacy (Serbia)

Non-glycemic effects of liraglutide in subjects with type-2 diabetes (T2DM) including carotid intima-media thickness (cIMT), a recognized marker of subclinical atherosclerosis, are well established. Furthermore, the LEADER study determined its beneficial cardiovascular (CV) effect. However, the long-term effects of liraglutide on CV risk markers, including cIMT, in real-world setting are still limited. Here we investigated whether the reduction in glycemic and metabolic parameters, with particular focus on cIMT, could be maintained in T2DM subjects under routine clinical practice.

Thirty one T2DM subjects (19 men and 12 women; mean age:  $60\pm17$  years) without prior history of a major CV event were included in this prospective 5 years real-world study. All of them were naïve to incretin-based therapies and treated with metformin only. Liraglutide (1.2 mg/day) was added to stable dose of metformin (1500-3000 mg/day). cIMT was measured by B-mode real-time ultrasound.

As expected, there was a significant reduction in fasting glycemia, HbA1c, total- and LDL- cholesterol over the time. Of interest, also cIMT significantly reduced during the 5 years follow-up. Yet, changes in cIMT did not correlate with changes in any other variable studied.

Long-term liraglutide treatment in real world settings effectively maintained the reduction of several glyco-metabolic parameters in T2DM subjects. The main finding of the present study, a reduction of cIMT over the time with the use of liraglutide is in accordance with its CV actions. Our data together with the results from the LEADER study, support the fact that liraglutide's effect could be translated into an effective CV prevention independently of its effect on plasma glucose and/or lipids.

## INTERAKCIJA REZISTINA I CAP-1 RECEPTORA SA HDL-HOLESTEROLOM KOD PACIJENATA SA KOLOREKTALNIM KANCEROM

**Marija Mihajlović, Ana Ninić, Miron Sopić, Milica Miljković,**  
**Aleksandra Stefanović, Vesna Spasojević-Kalimanovska,**  
**Aleksandra Zeljković**

Katedra za medicinsku biohemiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Rezistin se može smatrati adipocitokinom od interesa u kolorektalnom karcinomu (CRC), s obzirom da su njegove koncentracije povećane kod ovih pacijenata u odnosu na zdrave, asimptomatske osobe. Razvoj CRC je takođe povezan sa poremećajima u lipidnom statusu; stoga je cilj naše studije bio da se ispita veza koja postoji između rezistina i njegovog receptora, proteina udruženog sa adenilat ciklazom (CAP1) kao inflamatorne komponente i koncentracije HDL-holesterol (HDL-H) kao lipidnog markera i jednog od nosilaca antiinflamatornog statusa.

Naša studija je uključila 100 pacijenata sa CRC i 109 zdravih osoba. Koncentracija rezistina u plazmi je određena ELISA metodom. Rutinsko laboratorijsko određivanje je primenjeno za HDL-H. Određivanje nivoa iRNK rezistina i CAP1 iz mononuklearnih ćelija periferne krvi (MČPK) je sprovedeno qRT-PCR metodom.

Naša studija je pokazala povišen nivo iRNK CAP1 ( $p<0,05$ ) i snižen nivo iRNK rezistina ( $p<0,001$ ), više vrednosti koncentracije rezistina ( $p<0,001$ ) i sniženu koncentraciju HDL-H ( $p<0,001$ ), kod pacijenata u odnosu na kontrolu. Značajna negativna korelacija je uočena između HDL-H i koncentracije rezistina u CRC ( $r=-0,250$ ;  $p<0,05$ ). Koncentracija rezistina ( $OR=1,074$ ;  $p<0,001$ ) i HDL-H ( $OR=0,210$ ;  $p<0,001$ ) su izdvojene kao značajne determinante povećanog rizika za razvoj CRC.

Naši rezultati su ukazali na značajnu povezanost između rezistina i HDL-H, kao i značajan prediktivni potencijal oba markera za razvoj CRC. S obzirom da se ovi markeri nalaze na suprotstavljenim stranama na inflamatornom raskršću, njihove međusobne interakcije treba dalje istraživati u CRC.

## **INTERACTION OF RESISTIN AND CAP1 RECEPTOR WITH HDL-CHOLESTEROL IN COLORECTAL CANCER PATIENTS**

**Marija Mihajlović, Ana Ninić, Miron Sopić, Milica Miljković,  
Aleksandra Stefanović, Vesna Spasojević-Kalimanovska,  
Aleksandra Zeljković**

Department of Medical Biochemistry, University of Belgrade - Faculty of Pharmacy (Serbia)

Different types of meta analyses revealed resistin as an adipocytokine of interest in colorectal cancer (CRC), since its concentrations were generally higher when compared with healthy asymptomatic individuals. CRC is also associated with lipid status disorders; therefore the aim of our study was to investigate the link between resistin and its receptor adenylate cyclase-associated protein 1 (CAP1), as inflammatory components and HDL cholesterol (HDL-C), as a lipid marker and one of the anti-inflammatory status carriers.

Our study included 100 colorectal cancer patients and 109 healthy controls. Plasma resistin concentration was measured by ELISA method. Routine laboratory method was applied for HDL-C determination. Resistin and CAP1 mRNA levels from peripheral blood mononuclear cells (PBMC) were measured using qRT-PCR method.

Our study showed increased CAP1 mRNA levels ( $p<0.05$ ) and lower resistin mRNA levels ( $p<0.001$ ), higher values of resistin protein concentration ( $p<0.001$ ) and lower HDL-C concentration ( $p<0.001$ ) in CRC patients relative to control group. Significant negative correlation was observed between HDL-C and resistin in CRC ( $\rho=-0.250$ ;  $p<0.05$ ). Univariate logistic analysis showed that both resistin ( $OR=1.074$ ;  $p<0.001$ ) and HDL-C ( $OR=0.210$ ;  $p<0.001$ ) are significant determinants of increased risks for CRC development.

Our results indicated significant associations between resistin and HDL-C, as well as notable predictive potential of both individual markers for CRC development. Since these markers represent opponents at the inflammatory crossroads, their mutual relationship in CRC should be further explored.

## POTENCIJALNI RAZLOZI ZA SMANJENJE ANTIOKSIDATIVNE AKTIVNOSTI HDL ČESTICA KOD PACIJENATA NA HEMODIJALIZI

**Milica Miljković<sup>1</sup>, Aleksandra Stefanović<sup>1</sup>, Jelena Vekić<sup>1</sup>,**  
**Sanja Simić-Ogrizović<sup>2</sup>, Vesna Spasojević-Kalimanovska<sup>1</sup>,**  
**Jelena Kotur-Stevuljević<sup>1</sup>**

<sup>1</sup>Katedra za medicinsku biohemiju, Univerzitet u Beogradu – Farmaceutski fakultet, <sup>2</sup>Klinika za nefrologiju, Klinički Centar Srbije; Univerzitet u Beogradu - Medicinski fakultet (Srbija)

Paraoksonaza1 (PON1) predstavlja glavni antioksidativni enzim na HDL česticama. Dislipidemija, oksidativni stres i inflamacija kod pacijenata na hemodijalizi mogu dovesti do promena u strukturi apoA-I, što može uticati na aktivnost PON1. Oksidativna modifikacija apoA-I u HDL česticama, kao posledica smanjene aktivnosti PON1 može dovesti do formiranja proaterogenih HDL čestica. Cilj ove studije je bio da se ispita povezanost između koncentracije apoA-I i aktivnosti PON1 kod pacijenata na hemodijalizi.

U studiju je uključeno 57 pacijenata na hemodijalizi i 20 zdravih kontrola, Koncentracija apoA-I je izmerena imunoturbidimetrijski, dok je arilesterazna aktivnost PON1 određena korišćenjem fenilacetata kao supstrata.

Dobijene vrednosti koncentracija apoA-I i aktivnosti PON1 su bile značajno niže kod pacijenata na hemodijalizi u odnosu na zdrave ispitanike ( $p<0,01$ ). Pronađena je i jaka pozitivna korelacija između koncentracije apoA-I i aktivnosti PON1 ( $\rho=0,649$ ,  $p<0,01$ ) što potencijalno ukazuje da strukturne promene u apoA-I koje nastaju kao posledica povišenog oksidativnog stresa i inflamacije, mogu uticati na aktivnost PON1.

Ispitivanje strukture i funkcije HDL čestica merenjem koncentracije apoA-I i aktivnosti PON1 omogućava bolju procenu antiaterogenog potencijala ovih čestica.

## POTENTIAL REASONS FOR DECREASED ANTIOXIDATIVE ACTIVITY OF HDL PARTICLES IN HEMODIALYSIS PATIENTS

**Milica Miljković<sup>1</sup>, Aleksandra Stefanović<sup>1</sup>, Jelena Vekić<sup>1</sup>,  
Sanja Simić-Ogrizović<sup>2</sup>, Vesna Spasojević-Kalimanovska<sup>1</sup>,  
Jelena Kotur-Stevuljević<sup>1</sup>**

<sup>1</sup>Department of Medical Biochemistry, University of Belgrade – Faculty of Pharmacy, <sup>2</sup>Clinic for Nephrology, Clinical Center of Serbia; University of Belgrade - Faculty of Medicine (Serbia)

Paraoxonase1 (PON1) presents the most important antioxidative enzyme at HDL particles. Dyslipidemia, oxidative stress and inflammation in hemodialysis patients can change apoA-I structure and influence PON1 activity. Oxidative modification of apoA-I in HDL particles as a consequence of decreased PON1 activity could lead to formation of proatherogenic HDL particles. The aim of the present study was to examine association between apoA-I concentration and PON1 activity in hemodialysis patients.

This study included 57 hemodialysis patients and 20 healthy controls. apoA-I concentration was measured by immunoturbidimetric method, while arylesterase activity of PON1 was determined kinetically using phenylacetate as substrate.

The concentration of apoA-I and PON1 activity showed significantly lower values in hemodialysis patients compared to healthy subjects ( $p < 0.01$ ). A strong positive correlation was found between apoA-I concentration and PON1 activity ( $\rho = 0.649$ ,  $p < 0.01$ ), which potentially indicates that structural changes in apoA-I as a result of increased oxidative stress and inflammation can affect PON1 activity.

Examination of the structure and function of HDL particles by measuring concentration of apoA-I and PON1 activity enables a better estimation of antiatherogenic potential of these particles.

## METABOLIČKI ZDRAVA GOJAZNOST I RIZIK ZA RAZVOJ KARDIOVASKULARNIH BOLESTI

**Sanja Vujčić<sup>1</sup>, Jelena Vekić<sup>1</sup>, Aleksandra Zeljković<sup>1</sup>, Lidija Memon<sup>2</sup>,**  
**Nataša Bogavac-Stanojević<sup>1</sup>, Vesna Spasojević-Kalimanovska<sup>1</sup>**

<sup>1</sup>Katedra za medicinsku biohemiju, Univerzitet u Beogradu - Farmaceutski fakultet, <sup>2</sup>Kliničko-bolnički centar „Bežanijska kosa”, Beograd (Srbija)

Gojaznost je epidemija modernog doba i faktor rizika za razvoj kardiovaskularnih bolesti (KVB). Međutim, neke gojazne osobe su metabolički zdrave, dok normalno uhranjene mogu imati metaboličke poremećaje. Cilj rada je bio ispitivanje uticaja metaboličkog statusa na rizik od razvoja KVB, kod normalno uhranjenih osoba i kod osoba prekomerne telesne mase.

U ispitivanje su uključene 164 zdrave osobe i 163 pacijenta sa KVB. Na osnovu indeksa telesne mase (ITM) svi ispitani su najpre razvrstani na normalno uhranjene ( $ITM < 27 \text{ kg/m}^2$ ) i one sa prekomernom telesnom masom ( $ITM \geq 27 \text{ kg/m}^2$ ). Metabolički status je procenjen na osnovu prisustva sledećih faktora rizika: hipertenzija, povišen obim struka, hipertrigliceridemija, snižen HDL-bolesterol, hiperglikemija. Ispitani su  $\leq 2$  faktora rizika su označeni kao metabolički zdrave osobe.

Ispitani sa prekomernom telesnom masom su imali značajno viši rizik za razvoj KVB od normalno uhranjenih osoba (OR: 2,36; 95% CI: 1,51-3,68;  $P < 0,001$ ). Rizik za razvoj KVB je bio 11 puta veći kod metabolički nezdravih nego kod metabolički zdravih osoba (OR: 10,93; 95% CI: 6,54-18,28;  $P < 0,001$ ). U grupi ispitnika sa povišenom telesnom masom, metabolički nezdrave osobe su imale značajno viši rizik od metabolički zdravih (OR: 9,37; 95% CI: 4,49-19,53;  $P < 0,001$ ). Slično, u grupi normalno uhranjenih osoba, rizik za KVB je bio značajno viši kod metabolički nezdravih nego kod metabolički zdravih (OR: 12,38; 95% CI: 5,40-28,40;  $P < 0,001$ ). Sa druge strane, u poređenju sa normalno uhranjenim metabolički zdravim osobama, metabolički zdravi ispitani sa prekomernom telesnom masom nisu imali povišen rizik za KVB (OR: 1,23; 95% CI: 0,59-2,57;  $P = 0,585$ ).

Naši rezultati su potvrdili da gojaznost značajno doprinosi razvoju KVB. Takođe, utvrdili smo da metabolički zdrave osobe prekomerne telesne mase nisu pod većim rizikom za razvoj KVB u odnosu na metabolički zdrave, normalno uhranjene osobe, što ukazuje na potencijalni značaj otkrivanja metabolički zdravog fenotipa gojaznosti za prevenciju nastanka KVB.

## METABOLICALLY HEALTHY OBESITY AND CARDIOVASCULAR DISEASE RISK

**Sanja Vujićić<sup>1</sup>, Jelena Vekić<sup>1</sup>, Aleksandra Zeljković<sup>1</sup>, Lidija Memon<sup>2</sup>,**  
**Nataša Bogavac-Stanojević<sup>1</sup>, Vesna Spasojević-Kalimanovska<sup>1</sup>**

<sup>1</sup>Department of Medicinal Biochemistry, University of Belgrade - Faculty of Pharmacy, <sup>2</sup>Clinical Centre Bezanijska Kosa, Belgrade (Serbia)

Obesity has epidemic proportion and it is an important risk factor for developing cardiovascular disease (CVD). However, some people are metabolically healthy despite being obese, while normal weight people may exhibit metabolic disorders. The aim of this study was to examine the influence of the metabolic status on the risk for developing CVD in normal weight and overweight individuals.

The study included 164 healthy individuals and 163 patients with CVD. According to body mass index (BMI), the subjects were classified as normal weight ( $BMI < 27 \text{ kg/m}^2$ ) and overweight ( $BMI \geq 27 \text{ kg/m}^2$ ). The metabolic status was assessed on the basis of the presence of the following risk factors: hypertension, large waist circumference, hypertriglyceridemia, decreased HDL-cholesterol, hyperglycemia. Subjects with  $\leq 2$  risk factors were categorized as metabolically healthy (MH).

Overweight individuals were at significantly higher risk of developing CVD compared to normal weight individuals (OR: 2.36; 95% CI: 1.51-3.68; P<0.001). Metabolically unhealthy (MU) individuals had 11 fold higher CVD risk compared to MH subjects (OR: 10.93; 95% CI: 6.54-18.28; P<0.001). Among overweight subjects MU individuals had significantly higher CVD risk than their MH counterparts (OR: 9.37; 95% CI: 4.49-19.53; P<0.001). Similarly, among normal weight subjects, MU had higher risk compared to MH individuals (OR: 12.38; 95% CI: 5.40-28.40; P<0.001). When compared to MH normal weight subjects, the risk for development of CVD in MH overweight subjects was not significant (OR: 1.23; 95% CI: 0.59-2.57; P=0.585).

Obesity significantly contributes to the development of cardiovascular disease. Also, we found that MH overweight status is not associated with increased CVD risk, as compared to MH normal weight individuals, indicating the potential significance of detecting a metabolically healthy obese phenotype in prevention of CVD.

## ISPITIVANJA NA PACOVSKOM VALPROATNOM MODELU AUTIZMA OTKRIVAJU POZITIVNU MODULACIJU ALFA5GABAA RECEPTORA KAO MOGUĆI NOVI TERAPIJSKI PRISTUP

**Anja Santrač<sup>1</sup>, Marija Banićević<sup>1</sup>, Jovana Arandelović<sup>1</sup>, Bojan Marković<sup>2</sup>, Guanguan Li<sup>3</sup>, James Cook<sup>3</sup>, Miroslav Savić<sup>1</sup>**

<sup>1</sup>Katedra za farmakologiju, Univerzitet u Beogradu - Farmaceutski fakultet,

<sup>2</sup>Katedra za farmaceutsku hemiju, Univerzitet u Beogradu - Farmaceutski

fakultet (Srbija), <sup>3</sup>Univerzitet u Viskonsin-Milvokiju, Milvoki (SAD)

Spektar autističnih poremećaja (ASD) je spektar stanja koje karakterišu problemi u socijalnoj komunikaciji i interakciji, i restriktivni, repetitivni obrasci ponašanja, interesovanja i aktivnosti.

Cilj je bio da ispitamo uticaj pozitivne modulacije alfa5GABAA receptora na repetitivno i restriktivno ponašanje u pacovskom valproatnom (VPA) modelu ASD.

Sprovedena je farmakokinetička studija da bi se ustanovile doze pozitivnog modulatora alfa5GABAA receptora, MP-III-022, koje izazivaju blag i umeren selektivan odgovor. Trudne Wistar ženke su primile VPA ili fiziološki rastvor (SAL). Od 21. do 27. postnatalnog dana njihovo potomstvo oba pola je dnevno primalo rastvarač (SOL) ili MP-III-022 u dozama 0,3 (MP0,3) ili 1 mg/kg (MP1). Nakon toga, sprovedeni su test spontane lokomotorne aktivnosti (SLA) i reverzni Morrisov vodeni labyrin (rMWM).

Tokom SLA, VPA-SOL su bili više vremena aktivni, i napravili više rotacija, posebno nadesno, u poređenju sa SAL-SOL pacovima. Primenjen VPA mužjacima, MP0,3 je smanjio hiperaktivnost i rotacije nadesno, i imao tendenciju da smanji ukupne rotacije. Dat SAL mužjacima, MP0,3 ih je učinio hiperaktivnim, sa sklonosću da poveća njihove rotacije. Slično, MP1 je pogoršao sva triparametra u SAL, sa tendencijom da smanji rotacije nadesno kod VPA mužjaka. Kod ženki, MP1 je pokazao trend da smanji hiperaktivnost kod VPA, ali poveća je kod SAL, i slično, smanjio rotacije nadesno kod VPA, a povećao rotacije kod kontrole.

U rMWM, VPA-SOL životinje su imale manju efikasnost puta sa tendencijom da ulaze više puta u prethodnu ciljnu zonu. MP0,3 je poboljšao oba parametra kod VPA mužjaka; MP1 je pokazao isti trend za drugi parametar kod oba pola.

MP-III-022 je pokazao tendenciju da normalizuje praćene bihevioralne parametre kod VPA, ali da ih pogorša kod SAL životinja. Efekat doze je polno zavisан: MP0,3 ima veći uticaj na mužjake, MP1 na ženke.

Pozitivna modulacija alfa5GABAA receptora poboljšava repetitivno i restriktivno ponašanje u pacovskom modelu ASD.

# THE VALPROATE RAT MODEL REVEALS POSITIVE MODULATION OF ALPHA5GABAA RECEPTORS AS A NOVEL TARGET FOR TREATMENT OF AUTISM SPECTRUM DISORDER

**Anja Santrač<sup>1</sup>, Marija Banićević<sup>1</sup>, Jovana Aranđelović<sup>1</sup>,  
Bojan Marković<sup>2</sup>, Guanguan Li<sup>3</sup>, James Cook<sup>3</sup>, Miroslav Savić<sup>1</sup>**

<sup>1</sup>Department of Pharmacology, University of Belgrade - Faculty of Pharmacy,

<sup>2</sup>Department of Pharmaceutical Chemistry, University of Belgrade - Faculty of Pharmacy (Serbia), <sup>3</sup>University of Wisconsin-Milwaukee, Milwaukee (USA)

Autism spectrum disorder (ASD) is a range of conditions characterized with problems in social communication and interaction, and restricted, repetitive patterns of behavior, interests or activities. We aimed to determine the influence of a positive modulation of alpha5GABAA receptors on repetitive and restricted behavior in the rat valproate (VPA) model of ASD.

Pharmacokinetic study was performed to determine doses of a positive modulator of alpha5GABAA receptors, MP-III-022, that elicit a mild and a moderate selective response. Pregnant Wistar females were given VPA or saline (SAL). On postnatal days 21-27, the respective offspring of both sexes received daily doses of solvent (SOL) or MP-III-022, at 0.3 (MP0.3) or 1 mg/kg (MP1). Afterwards, spontaneous locomotor activity test (SLA) and reverse Morris water maze (rMWM) were performed.

In SLA, VPA-SOL spent more time in activity, making more rotations, especially clockwise, compared to SAL-SOL rats. Administered to VPA males, MP0.3 lowered hyperactivity, decreased clockwise rotations and tended to decrease overall rotations. MP0.3 made SAL males hyperactive, with a propensity to increase their rotations. Similarly, MP1 exacerbated all three parameters in SAL, while tended to decrease clockwise rotations in VPA males. In females, MP1 revealed a trend to lower hyperactivity in VPA, but increase it in SAL, and, similarly, to decrease clockwise rotations in VPA, while increase rotations in controls.

In rMWM, VPA-SOL animals had lower path efficiency with a tendency to visit more times the previous target zone. MP0.3 improved both parameters in VPA males; MP1 exerted the same trend for the second parameter in both sexes.

MP-III-022 tended to normalize the performance of VPA, but worsen it in SAL animals. The effect of the dose is sex-dependent: MP0.3 has a greater influence on males, MP1 on females. Positive modulation of alpha5GABAA receptors ameliorates repetitive and restricted behavior in the rat model of ASD.

## DISULFIRAM – POTENCIJALNE TERAPIJSKE PRIMENE STAROG LEKA

**Ana Đurić, Mirjana M. Đukić**

Katedra za toksikologiju „Akademik Danilo Soldatović”, Univerzitet u Beogradu  
- Farmaceutski fakultet (Srbija)

Disulfiram (DSF) se koristi u averzivnoj terapiji alkoholizma više od 60 godina. Do danas su se mnoge studije bavile različitim terapijskim primenama DSF. Disulfiram i njegovi glavni metaboliti ditiokarbamati (DTCs) se predlažu za korišćenje: u tretmanima nekih gljivičnih, bakterijski i virusnih infekcija; kod inflamacije; u terapiji trovanja niklom i bakrom (Cu); u eksperimentalnim tretmanima AIDS-a; kao dodatak hemoterapiji; kod kokainske i udružene kokainske i alkoholne zavisnosti. Helirajući esencijalne metale (Cu i/ili druge esencijalne metale) DTCs inhibiraju aktivnosti nekoliko Cu-zavisnih enzima: dopamin- $\beta$ -hidroksilaze, karboksiesteraze i holinesteraze. Smatra se da poznato Cu-helatno dejstvo indukuje inhibiciju proteazoma i posledičnu apoptozu ćelija kancera, što kandiduje DSF za korišćenje u antikancerskoj terapiji. Kako bismo ispitali uticaj DSF na redoks, metalni i androgeni status u testisima nakon i tokom izlaganja kadmijumu (Cd), sproveli smo studiju na Wistar pacovima.

Naši rezultati ukazuju da primena DSF nakon i tokom izlaganja Cd pokazuje protektivno dejstvo na narušen oksidativni status u testisima tretiranih pacova. Ovaj fenomen se može objasniti jakim helirajućim kapacitetom DSF kao i antioksidativnim kapacitetom DTC/DSF redoks para koji deluje slično kao endogeni redoks par redukovani glutation/oksidovani glutation. Vezujući toksične jone Cd DSF onemogućava ovaj metal da ostvari svoje štetne prooksidativne efekte na tkivo testisa. Takođe smo pokazali da DSF ne može popraviti smanjenu proizvodnju testosterona ni morfološke promene testisa nastale usled izlaganja pacova Cd. Iako je DSF pokazao značajan antioksidativni potencijal, on nije uspeo da ukloni štetne efekte izazvane izlaganjem Cd koji se odnose na proizvodnju testosterona i morfološke promene testisa pacova.

## **DISULFIRAM – POTENTIAL THERAPEUTIC APPLICATIONS OF AN OLD DRUG**

**Ana Djurić, Mirjana M. Djukić**

Department of Toxicology „Akademik Danilo Soldatović”, University of Belgrade  
- Faculty of Pharmacy (Serbia)

Disulfiram (DSF) has been used in the aversive therapy of alcoholism for more than 60 years. To date, many studies have dealt with different therapeutic applications of DSF. Disulfiram and its main metabolites dithiocarbamates (DTCs) are suggested for the treatment of some fungal, bacterial and viral infections; inflammation; nickel and copper (Cu) poisoning therapy; experimental treatment of AIDS; as an adjuncts for chemotherapy; and cocaine dependence alone or co-morbid cocaine and alcohol dependence. By chelating essential metals (Cu and/or other essential metals) DTCs inhibit activities of a few Cu-dependent enzymes: dopamine- $\beta$ -hydroxylase, carboxylesterase and cholinesterase. This same Cu-chelating action is thought to induce proteasome inhibition and subsequent cancer cell apoptosis, leading to the proposal that DSF could serve as an anticancer therapy. We conducted an animal study on Wistar rats to examine the influence of DSF on red-ox, metal and androgen status in testes after and during the exposure to cadmium (Cd).

Our results have indicated that application of DSF after and during Cd exposure, has shown beneficial effect on impaired oxidative status in the testes of treated rats. This phenomenon can be explained by strong chelating capacity of DSF and antioxidant capacity of DTC/DSF red-ox couple similar to endogenous reduced glutathione/oxidized glutathione redox couple. Binding toxic Cd ions DSF disables this metal to achieve its harmful pro-oxidant effects on testicular tissue. Also, we showed that DSF cannot repair the decreased production of testosterone and morphological changes of testes caused by exposure of rats to Cd. Although DSF showed significant antioxidant potential, it failed to remove the deleterious effects of Cd exposure on testosterone production and morphological changes in rat testes.

## UTICAJ VINIFIKACIJE NA SADRŽAJ FENOLNIH KISELINA I ANTIOKSIDATIVNE OSOBINE VINA OD ARONIJE

**Uroš Čakar<sup>1</sup>, Aleksandar Petrović<sup>2</sup>, Vlatka Vajs<sup>3</sup>, Brižita Đorđević<sup>1</sup>**

<sup>1</sup>Katedra za bromatologiju, Univerzitet u Beogradu - Farmaceutski fakultet,

<sup>2</sup>Institut za prehrambenu tehnologiju i biohemiju, Univerzitet u Beogradu - Poljoprivredni fakultet, <sup>3</sup>Centar za hemiju, Institut za hemiju tehnologiju i metalurgiju, Univerzitet u Beogradu (Srbija)

Aronija (*Aronia melanocarpa* Heynh.) je biljka koja je autohtona za severne krajeve Evrope i Azije. Zbog njenog pozitivnog zdravstvenog efekta na ljudski organizam aronija se uspešno danas gaji širom sveta, pa i u Srbiji. Vino predstavlja jedan od proizvoda koji se može dobiti preradom ovog voća.

Vino od aronije je proizvedeno postupkom mikrovinifikacije. Kontrolisano vreme aronije je sprovedeno uz pomoć dve različite čiste kulture kvasca. Pored toga, primenjeni su šećer i enzim koji su dodati u neke od uzoraka pre fermentacije, da bi se povećao sadržaj fenolnih jedinjenja u krajnjem proizvodu. Sadržaj fenolnih jedinjenja je određen UPLC TQ-MS/MS. Takođe je primenjena i Folin-Ciocalteu metoda za određivanje ukupnog sadržaja polifenola (USP). Antiradikalska aktivnost je određena uz pomoć DPPH radikala, a takođe je primenjena i FRAP metoda.

Rezultati ukazuju da postupak vinifikacije značajno utiče na sadržaj izabranih polifenolnih jedinjenja i antioksidativne osobine. Vinifikacije u kojima je dodat šećer i enzim pre početka fermentacije dale su vino od aronije sa najvišim sadržajem izabranih fenolnih jedinjenja kao i najboljim antioksidativnim osobinama. Kontrola koja je proizvedena bez dodatka šećera i enzima pokazala je najniže antioksidativne osobine kao i sadržaj izabranih polifenolnih jedinjenja. Posebno se ističe sadržaj fenolnih kiselina i njihovih derivata hidroksicimetne, među kojima su se istakle kafeinska, p-kumarinska, sinapinska i hlorogena. Sadržaj ukupnih polifenola u analiziranim voćnim vinima je bio od 2271,4 do 2477,6 mg GAE/L, dok su vrednosti za FRAP bile od 67,2 do 83,5 mmol/L Fe<sup>2+</sup>. Antiradikalska aktivnost određena DPPH testom predstavljena je IC<sub>50</sub> vrednostima koje su bile od 1,23 do 1,50%. Rezultati ukazuju da vino od aronije predstavlja proizvod sa pozitivnim zdravstvenim efektom na ljudski organizam koji ima mnogočestnu „hvatanja“ slobodnih radikala.

Dobijeni rezultati ukazuju da je vino od aronije bogat izvor derivata hidroksicimetne kiselina koji predstavljaju samo mali deo ostalih jedinjenja koja su odgovorna za jake antioksidativne i antiradikalske osobine ovog vina.

## VINIFICATION INFLUENCE ON PHENOLIC ACID CONTENT AND ANTIOXIDANT PROPERTIES OF BLACK CHOKEBERRY WINE

**Uroš Čakar<sup>1</sup>, Aleksandar Petrović<sup>2</sup>, Vlatka Vajs<sup>3</sup>, Brižita Đorđević<sup>1</sup>**

<sup>1</sup>Department of Bromatology, University of Belgrade - Faculty of Pharmacy,

<sup>2</sup>Institute of Food Technology and Biochemistry, University of Belgrade -

Faculty of Agriculture, <sup>3</sup>Center for Chemistry, Institute for Chemistry,

Technology and Metallurgy, University of Belgrade (Serbia)

Black chokeberry (*Aronia melanocarpa* Heynh.) is plant autochthonous for the northern parts of Europe and Asia. Due to their beneficial health effects on human organism black chokeberry is successfully introduced in Serbia and all around the world. Fruit wine is one product which can be derived from this fruit.

Black chokeberry wine was produced in micro-vinification procedure. The fermentation of black chokeberry must was conducted by using two different pure selected yeast cultures. Beside, in some samples enzyme and sugar were added to increase content of phenolic compounds in the final product. Phenolic profile was analyzed by UPLC TQ-MS/MS. Also, Folin-Ciocalteu method was used for determination of total phenolic content (TPC). Antiradical activity was investigated by DPPH radical while FRAP method was conducted too.

The results indicate that vinification process have significant influence on selected phenolic profile and antioxidant properties. Vinification process in which sugar and enzyme were added before fermentation resulted in black chokeberry wine with the highest content of selected phenolic compounds and the best antioxidant properties. The control produced without addition of enzyme and sugar showed the lowest antioxidant properties and content of selected phenolic compounds. Phenolic acids, such as hydroxycinnamic acid derivatives (caffeic, *p*-coumaric, sinapinic and chlorogenic acids) were the most abundant in analyzed samples. The TPC for fruit wine samples was in the range from 2271.4 to 2477.6 mg GAE/L while FRAP values were from 67.2 to 83.5 mmol/L Fe<sup>2+</sup>. Antiradical DPPH activity was presented with IC<sub>50</sub> values which were in the range from 1.23 to 1.50%. The findings indicate that black chokeberry wine represents product with beneficial health effects and ability in free radical scavenging.

Obtained results indicate that black chokeberry wine is a rich source of hydroxycinnamic acid derivatives which are only small part of many other compounds responsible for potent antioxidant and antiradical properties of this wine.

## ISPITIVANJE RETENCIONOG PONAŠANJA ODABRANIH LIGANADA IMIDAZOLINSKIH RECEPTORA U REVERZNO-FAZNOJ I TEČNOJ HROMATOGRAFIJI HIDROFILNIH INTERAKCIJA

**Darija Obradović, Slavica Oljačić, Danica Agbaba**

Katedra za farmaceutsku hemiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Primena *mixed-mode* HILIC/RP kolona omogućava analizu velikog broja strukturno različitih jedinjenja pri RP (*reversed-phase*) i HILIC (*hydrophilic interaction liquid chromatography*) hromatografskim uslovima koji se postižu adekvatnim odabirom mobilne faze. Cilj rada je bio da se ispita primenljivost particionog modela u opisivanju retencionog ponašanja liganada imidazolinskih receptora, da se odrede volumenske frakcije vodene faze pri kojima dolazi do smene RP i HILIC hromatografskih sistema ( $\varphi_{\min}$ ) i izdvoje najznačajniji molekulski deskriptori koji utiču na njihovu interkonverziju.

Retenciono ponašanje 17 liganada imidazolinskih receptora ispitano je na *mixed-mode* HILIC stacionarnoj fazi korišćenjem smeše acetonitrila i vodenog rastvora 20 mM amonijumacetata (pH 6) u širokom opsegu zapreminskih frakcija vodene faze ( $\varphi$ ). Višestruka linearna regresija je korišćena za izdvajanje najznačajnijih Abrahamovih deskriptora koji utiču na  $\varphi_{\min}$  kao i korelaciju  $\varphi_{\min}$  sa lipofilnošću ispitivanih jedinjenja.

Volumenska frakcija pufera pri kojoj dolazi do smene između retencionih RP i HILIC uslova izračunata je korišćenjem nelinearne relacije  $\log k$  vs  $\varphi$ . U sledećem koraku za svako pojedinačno jedinjenje, definisani HILIC i RP regioni su opisani linearnom relacijom između logaritma retencionih faktora i zapremine modifikatora mobilne faze. Dobro slaganje retencionih podataka za korišćeni hromatografski model potvrđeno je dobijenim visokim koeficijentima korelacijske,  $r>0,86$ . Pronađeno je da lipofilnost ispitivanih jedinjenja (AClogP) značajno utiče na minimalnu vrednost vodene frakcije pufera pri kojoj dolazi do smene retencionih uslova ( $r=0,90$ ). Pored toga, značajna korelacija je ostvarena i u modelu u kojem su izdvojene sledeće molekulske osobine: stepen jonizacije (Di), kiselost jedinjenja (Ai), i McGowan-ova zapremina (Vi) ( $r=0,86$ ).

Utvrđena je primenljivost particionog modela u opisivanju retencije u HILIC/RP sistemima odabranih jedinjenja. Izračunavanjem odabranih molekulskih deskriptora moguće je na brz i jednostavan način predvideti prevojnju tačku pri kojoj dolazi do smene između HILIC i RP retencionog mehanizma.

# INVESTIGATION OF RETENTION BEHAVIOUR OF SELECTED IMIDAZOLINE RECEPTOR LIGANDS IN REVERSED-PHASE AND HYDROPHILIC INTERACTION LIQUID CHROMATOGRAPHY

**Darija Obradović, Slavica Oljačić, Danica Agbaba**

Department of Pharmaceutical Chemistry, University of Belgrade - Faculty of Pharmacy (Serbia)

The use of mixed-mode HILIC/RP stationary phases allows the analysis of structurally different pharmaceutical compounds in the RP (*reversed-phase*) and HILIC (*hydrophilic interaction liquid chromatography*) systems. The aim of this study was to investigate applicability of the partition model in description of the retention behavior of the imidazoline-related compounds within the HILIC and RP systems, determination of the aqueous phase volume fraction when the turning point between the RP and HILIC mode occurs ( $\varphi_{\min}$ ), and selection of the most important physico-chemical properties which influence this interconversion.

Retention behaviour of 17 imidazoline receptor ligands was investigated on the mixed-mode HILIC stationary phase by using acetonitrile and the 20 mM aqueous ammonium acetate (pH=6) as a mobile phase in a wide range of volume fractions of the buffered eluent ( $\varphi$ ). Stepwise multiple linear regression was used to select the Abraham descriptors which influence the  $\varphi_{\min}$  value. Correlation between  $\varphi_{\min}$  and lipophilicity of the investigated compounds was also examined.

Parameter  $\varphi_{\min}$  was calculated from the non-linear relation  $\log k$  vs  $\varphi$ . For each compound, the HILIC and RP regions were established from linear relation between  $\log k$  and the mobile phase modifier. A good fit of the retention data was obtained for the employed retention model ( $r>0.86$ ). It was found that lipophilicity (AClogP) ( $r=0.90$ ), as well as degree of ionization (Di), hydrogen bond acidity (Ai) and McGowan volume (Vi) ( $r=0.86$ ) reflect molecular properties of the investigated compounds which affect the turning point between the two retention mechanisms in a given chromatographic system.

Applicability of the assumed retention model in description of the RP and HILIC retention behaviour was successfully demonstrated. Calculation of selected molecular descriptors enables fast and easy prediction of the turning points between the RP and HILIC systems.

## PRAĆENJE NIVOA OLOPATADINA U HUMANIM SUZAMA HILIC- ESI/MS/MS METODOM

**Jelena Maksić<sup>1</sup>, Ana Stajić<sup>2</sup>, Miroslav Knežević<sup>3</sup>, Bojana Dačić Krnjaja<sup>3</sup>,  
Biljana Jančić-Stojanović<sup>2</sup>, Mirjana Medenica<sup>4</sup>**

<sup>1</sup>Odeljenje za ispitivanje i kontrolu lekova, Služba za farmaceutsku delatnost,  
Vojnomedicinska akademija, <sup>2</sup>Katedra za analitiku lekova, Univerzitet u  
Beogradu-Farmaceutski fakultet, <sup>3</sup>Klinika za očne bolesti, Klinički centar Srbije,

<sup>4</sup>Katedra za fizičku hemiju i instrumentalne metode, Univerzitet u Beogradu -  
Farmaceutski fakultet (Srbija)

Cilj rada bio je određivanje malih koncentracija olopatadina u humanim suzama i procena njegovog farmakokinetičkog ponašanja u oku. Za praćenje nivoa olopatadina upotrebljena je prethodno optimizirana i validirana metoda tečne hromatografije hidrofilnih interakcija (HILIC) u kombinaciji s tandem masenom spektrometrijom (MS/MS).

Hromatografska separacija izvršena je u UPLC Acquity BEH amidnoj koloni (2,1 mm x 100 mm, 1,7 µm veličina čestica) koristeći 0,1% mravlju kiselinu u vodi i acetonitril kao mobilnu fazu. Kvantifikacija je izvedena pozitivnom elektrosprejem ionizacijom (ESI) u MRM modu. Prekursor-proizvod jon tranzicije praćene za kvantitativnu analizu i strukturnu karakterizaciju bile su 338→165 i 338→247 m/z za olopatadin, odnosno 265→91 i 265→208 m/z za interni standard mianserin.

U prospektivnoj kliničkoj studiji ispitivani su uzorci suza dobijeni od 30 ambulantnih pacijenata nakon bilateralne primene 0,1% kapi za oči olopatadin-hidrohlorida. Suze su uzorkovane indirektnom tehnikom pomoću Širmerovih test traka. Precipitacija proteina s acetonitrilom kao agensom za denaturisanje definisana je kao pogodna procedura pripreme uzoraka. Jednoprostorni matematički model prvog reda primenjen je u cilju izračunavanja značajnih okularnih farmakokinetičkih parametara. Dobijeni rezultati omogućili su uvid u dužinu prisustva ispitivanog leka u očnoj vodici i u proces njegove eliminacije iz tkivnih struktura oka. Na taj način, opravdana je učestalost predloženog režima doziranja oftalmološkog rastvora olopatadina, kao i njegova efikasnost i bezbednost u terapiji alergijskog konjunktivitisa. Potvrđena je primenljivost novorazvijene, veoma osetljive i selektivne HILIC metode sa MS/MS detekcijom za pouzdanu i brzu kvantifikaciju olopatadina u ograničenim zapreminama humanih suza za vreme kliničke prakse. Takođe, postignuto je precizno predviđanje okularne farmakokinetike olopatadina.

## **MONITORING OF OLOPATADINE LEVEL IN HUMAN TEARS BY HILIC- ESI/MS/MS METHOD**

**Jelena Maksić<sup>1</sup>, Ana Stajić<sup>2</sup>, Miroslav Knežević<sup>3</sup>, Bojana Dačić Krnjaja<sup>3</sup>,  
Biljana Jančić-Stojanović<sup>2</sup>, Mirjana Medenica<sup>4</sup>**

<sup>1</sup>Department of Drug Control and Examination, Service for Pharmaceutical Activity, Military Medical Academy, <sup>2</sup>Department of Drug Analysis, University of Belgrade-Faculty of Pharmacy, <sup>3</sup>Clinic for Eye Diseases, Clinical Center of Serbia,

<sup>4</sup>Department of Physical Chemistry and Instrumental Methods, University of Belgrade-Faculty of Pharmacy (Serbia)

The objective of the paper was the determination of olopatadine small concentrations in human tears as well as the assessment of its pharmacokinetic behavior in the eye. For the monitoring of olopatadine level, previously optimized and validated Hydrophilic Interaction Liquid Chromatography (HILIC) method coupled with tandem mass spectrometry (MS/MS) was used.

The chromatographic separation was carried out on UPLC Acuity BEH amide column (2.1 mm x 100 mm, 1.7 µm particle size) using 0.1% formic acid in water and acetonitrile as the mobile phase. The quantification was performed by positive ion electrospray ionization (ESI) in the multiplexreaction monitoring (MRM) mode. The precursor-product ion transitions followed for the quantitative analysis and the structure characterization were 338→165 and 338→247 m/z for olopatadine as well as 265→91 and 265→208 m/z for the internal standard mianserin.

In the prospective clinical study, the tear samples obtained from 30 outpatients were investigated following bilateral administration of 0.1% olopatadine hydrochloride eye drops. The tears were sampled by an indirect technique using the Schirmer test strips. The protein precipitation with acetonitrile as a denaturation agent was defined as a suitable sample preparation procedure. The one compartment first-order mathematical model was applied for calculating the significant ocular pharmacokinetic parameters. The obtained results provided insight into the length of presence of the examined drug in aqueous humor and also in its elimination process from the eye tissue structures. In that manner, the frequency of the proposed dosage regimen of the olopatadine ophthalmic solution has been justified as well as its efficacy and safety in treatment of allergic conjunctivitis.

The applicability of a newly developed, highly sensitive and selective HILIC method with MS/MS detection for reliable and rapid quantification of olopatadine in limited volumes of human tears during clinical practice was confirmed. Additionally, a precise prediction of olopatadine ocular pharmacokinetics was achieved.

## PRIMENA HPLC METODE U ODREĐIVANJU KONSTANTI STABILNOSTI KOMPLEKSA $\beta$ -CIKLODEKSTRINA SA ODABRANIM ANTIPSIHOTICIMA

**Nevena Maljurić, Biljana Otašević, Jovana Krmar, Mira Zečević, Ana Protić**

Katedra za analitiku lekova, Univerzitet u Beogradu-Farmaceutski fakultet  
(Srbija)

Ciklodekstrini grade inkluzione komplekse sa velikim brojem različitih organskih jedinjenja. Na taj način menjaju fizičko-hemijske osobine kompleksiranih lekova, smanjujući njihova neželjena dejstva, povećavajući rastvorljivost i biološku raspoloživost. Kao aditivi mobilne faze, omogućavaju povećanje udela vodene, a smanjenje udela organske faze uz istovremeno smanjenje retencije analita. Zbog značajnosti primene, proces formiranja kompleksa, kao i njihova stabilnost su uvek aktuelne teme. Cilj rada je ispitivanje mogućnosti primene hromatografskog pristupa za izračunavanje konstanti stabilnosti kompleksa (K)  $\beta$ -ciklodekstrina sa odabranim antipsihoticima i njihovim nečistoćama.

K je izračunata na osnovu formule:  $1/k = 1/k_0 + K [\beta\text{-CD}]x/k_0$ , gde je k retencioni faktor,  $k_0$  retencioni faktor bez  $\beta$ -ciklodekstrina u mobilnoj fazi,  $[\beta\text{-CD}]$  koncentracija  $\beta$ -ciklodekstrina, a x stehiometrija kompleksa. Retencioni faktori su dobijeni HPLC metodom na Thermo Scientific, Dionex 3000 Ultra, dok je stehiometrija kompleksa određena pomoću ESI-MS na Thermo Scientific TSQ Quantum Access Max.

$\beta$ -ciklodekstrin modifikovani RP-HPLC sistemi su složeniji od regularnih zbog raspodele supstance između stacionarne, mobilne faze i rastvorenog  $\beta$ -ciklodekstrina. Ako je pH vrednost mobilne faze ispod 3, silanolne grupe su u neutralnom obliku, što za posledicu ima smanjenje interakcija sa stacionarnom fazom, pa su na pH 2 K za risperidon, nečistoću 1, nečistoću 2, nečistoću 3, olanzapin, nečistoću B i nečistoću C iznosile 185,52 M-1, 105,39 M-1, 423,89 M-1, 187,68 M-1, 20,24 M-1, 93,01 M-1 i 17,16 M-1, redom. Pri pH 3,5 i 5,0, sa porastom koncentracije  $\beta$ -ciklodekstrina u mobilnoj fazi retenciona vremena su se produžavala, zbog dominantnih interakcija sa stacionarnom fazom. Posledično, hromatografski pristup nije bio odgovarajući za određivanje K, izuzev u slučaju nečistoće B, koja se nalazi u neutralnom obliku pri ispitivanom pH opsegu i ceo molekul učestvuje u građenju inkluzionog kompleksa sa  $\beta$ -ciklodekstrinom.

Hromatografski pristup ima potencijal da bude upotrebljen za izračunavanje K u uslovima u kojima je retencija predvođena interakcijama analita sa  $\beta$ -ciklodekstrinom.

## **APPLICATION OF HPLC METHOD IN DETERMINING THE COMPLEX STABILITY CONSTANTS BETWEEN $\beta$ -CYCLODEXTRIN AND SELECTED ANTIPSYCHOTICS**

**Nevena Maljurić, Biljana Otašević, Jovana Krmar, Mira Zečević, Ana Protić**

Department of Drug Analysis, University of Belgrade-Faculty of Pharmacy  
(Serbia)

Cyclodextrins form inclusion complexes with variety of organic compounds. Therefore, physico-chemical characteristics of complexed drugs are affected, reducing their side effects and improving solubility and bioavailability. As mobile phase additives, they allow increase of water and decrease of organic solvent content with simultaneous reduction of analyte's retention. Due to inherent usefulness, the complex formation process and stability is always a challenging topic. The aim was to investigate the possibility to apply chromatographic approach for determination of complex stability constants ( $K$ ) between  $\beta$ -cyclodextrin and selected antipsychotics and their impurities.

$K$  was calculated according to formula:  $1/k = 1/k_0 + K [\beta\text{-CD}]x/k_0$ , where  $k$  is retention factor,  $k_0$  is retention factor without  $\beta$ -cyclodextrin in mobile phase,  $[\beta\text{-CD}]$   $\beta$ -cyclodextrin concentration and  $x$  is the complex stoichiometry. Retention factors were determined by HPLC method on Thermo Scientific, Dionex 3000, while the stoichiometry was determined by ESI-MS on Thermo Scientific TSQ Quantum Access Max.

$\beta$ -cyclodextrin modified RP-HPLC systems are more complicated than regular, since solute is distributed between stationary, mobile phase and dissolved  $\beta$ -cyclodextrin. If mobile phase pH is under 3, silanol groups are non-ionized, minimizing the solute's interactions with stationary phase, so at pH = 2  $K$  for risperidone, impurity 1, impurity 2, impurity 3, olanzapine, impurity B and impurity C was 185.52 M<sup>-1</sup>, 105.39 M<sup>-1</sup>, 423.89 M<sup>-1</sup>, 187.68 M<sup>-1</sup>, 20.24 M<sup>-1</sup>, 93.01 M<sup>-1</sup> i 17.16 M<sup>-1</sup>, respectively. If pH=3.5 or 5, retention times were prolonged with increasing  $\beta$ -cyclodextrin concentration due to stationary phase interactions. Consequently, the chromatographic approach appeared unsuitable for  $K$  determination, except in case of impurity B, which is non-ionized across the investigated pH range and whole molecule participates in inclusion complex formation with  $\beta$ -cyclodextrin.

Chromatographic approach could be used for  $K$  determination if the retention is governed by solutes interactions with dissolved  $\beta$ -cyclodextrin.

## KVANTIFIKOVANJE VEZE STRUKTURE ARIPIPRAZOLA I SRODNIH NEČISTOĆA SA GENERISANIM ESI ODGOVOROM PRIMENOM METODA MAŠINSKOG UČENJA

**Iovana Krmarić<sup>1</sup>, Ljiljana Tolić<sup>2</sup>, Tatjana Đurkić<sup>3</sup>, Ana Protić<sup>1</sup>, Nevena Maljurić<sup>1</sup>, Mira Zečević<sup>1</sup>, Biljana Otašević<sup>1</sup>**

<sup>1</sup>Katedra za analitiku lekova, Univerzitet u Beogradu - Farmaceutski fakultet,

<sup>2</sup>Inovacioni centar Tehnološko-Metalurškog fakulteta, <sup>3</sup>Katedra za inženjerstvo zaštite životne sredine, Univerzitet u Beogradu - Tehnološko-metalurški fakultet (Srbija)

Elektrosprej ionizacija (ESI) predstavlja najčešće korišćenu tehniku ionizacije u LC/MS analizi polarnih i umereno polarnih analita. Nedovoljno rasvetljeni mehanizmi generisanja ESI jona uslovljavaju dugotrajnu optimizaciju odgovora sistema, zasnovanu na primeni pristupa pokušaja-i-greške. Upotreba metodologije kvantifikovanja veze strukture analita sa osobinom od interesa (QSPR), odnosno, ESI signalom može da dâ doprinos razumevanju procesa ionizacije, utemeljen na fizičko-hemijskom značenju uvrštenih molekulskih deskriptora. Cilj rada bio je modelovanje ESI odgovora test supstanci – atipičnog antipsihotika aripiprazola i srodnih nečistoća primenom QSPR pristupa, radi sticanja uvida u faktore koji kontrolišu efikasnost ionizacije i sledstvene mogućnosti sistematičnog pospešivanja osetljivosti metode.

LC/ESI-MS analize izvedene su na hibridnom Dionex UltiMate 3000® LC-LTQ XL linearnom jon trap sistemu (Thermo Fisher Scientific), koristeći fenil-heksil kolonu (100 mm × 4,6 mm, 2,6 µm; Phenomenex). Promene površina pikova praćene su variranjem udela metanola (60–75%, v/v), pH vodene komponente mobilne faze (3,0–8,2), protoka mobilne faze (400–500 µl/min), napona raspršivanja (2,5–5,0 kV), temperature kapilare (200–400 °C), pritiska nebulizirajućeg gasa (12–52 AU) i pritiska desolvatacionog gasa (3–21 AU) prema Box-Behnken dizajnu eksperimenata. Molekulski deskriptori izračunati su za sve supstance u odgovarajućoj ionizovanoj/neionizovanoj formi primenom Dragon 6.0.7 softvera (Talete srl). QSPR modeli sagrađeni su tehnikom veštačkih neuronskih mreža i metodom potpornih vektora (RapidMiner Studio 6.5.002, RapidMiner, Inc.).

Prediktivna moć dobijenih modela procenjena je korišćenjem ukrštene validacije sa 10 odeljaka. Konstruisani modeli postigli su zadovoljavajuće performanse – niske vrednosti kvadratnog korena srednje vrednosti sume kvadrata greške i visoke vrednosti validacionog regresionog faktora, odnosno, koeficijenta determinacije.

Rezultati studije ukazali su na prikladnost korišćenog pristupa u proučavanju analitičkog problema. Računanje velikog broja deskriptora doprinelo je uspostavljanju sveobuhvatnijeg QSPR modela u odnosu na do sada razvijene. Ipak, shodno relativno malom broju ispitivanih struktura, predloženi mehanizmi ionizacije generalizovani su na nivo ispitivanog sistema.

# **QUANTITATIVE STRUCTURE – PROPERTY RELATIONSHIP MODELING OF ESI RESPONSE OF ARIPIPRAZOLE AND ITS IMPURITIES USING MACHINE LEARNING METHODS**

**Jovana Krmar<sup>1</sup>, Ljiljana Tolić<sup>2</sup>, Tatjana Đurkić<sup>3</sup>, Ana Protić<sup>1</sup>,  
Nevena Maljurić<sup>1</sup>, Mira Zečević<sup>1</sup>, Biljana Otašević<sup>1</sup>**

<sup>1</sup>Department of Drug Analysis, University of Belgrade - Faculty of Pharmacy,  
<sup>2</sup>Innovation Center of the Faculty of Technology and Metallurgy, <sup>3</sup>Department of Environmental Engineering, University of Belgrade - Faculty of Technology and Metallurgy (Serbia)

Electrospray ionization, ESI represents the most widespread ionization technique in LC-MS analysis of (moderately) polar analytes. Insufficiently elucidated mechanisms of ions' formation induce the time-consuming optimization of system's response. Quantitative Structure Property Relationship, QSPR study of ESI responsiveness may add to the understanding of the ionization process, based on physicochemical meaning of involved molecular descriptors. The aim was to model the ESI response of the aripiprazole and related impurities using QSPR approach, in order to optimize factors that control ionization efficiency.

LC/ESI-MS analysis were performed on the Dionex UltiMate 3000® LC-LTQ XL linear ion trap system (Thermo Fisher Scientific), using a Phenyl-Hexyl column (100 mm × 4.6 mm, 2.6 µm). The changes in peaks' areas were observed by varying the methanol content (60-75%, v/v), the pH of the aqueous component of mobile phase (3.0-8.2), the flow rate (400-500 µl/min), the spray voltage (2.5-5.0 kV), capillary temperature (200-400 °C), nebulizer gas pressure (12-52 AU) and desolvation gas pressure (3-21 AU) according to the Box-Behnken experimental design. Molecular descriptors were calculated for all substances in appropriate ionized/non-ionized form using the Dragon 6.0.7 (Talete srl). QSPR models were built using ANN and SVR (RapidMiner Studio 6.5.002, RapidMiner, Inc.).

The predictive power of the obtained models was estimated using a 10-fold cross-validation. Constructed models have achieved satisfactory performance in terms of low root mean square errors (RMSE) and the high values of the (cross-validated) coefficients of determination ( $R^2$  and  $Q^2$ ).

The results indicated the appropriateness of the utilized approach for studying the ESI ionization. Calculating a large number of descriptors has contributed to the establishment of more comprehensive QSPR models in comparison with the so far developed. According to the relatively small number of employed structures, generalization of proposed ionization mechanisms was precluded.

## SIMULACIJE MOLEKULSKE DINAMIKE I VIRTUAL SCREENING STUDIJA INHIBITORA SIRTUINA 2

**Nemanja Đoković<sup>1</sup>, Katarina Nikolić<sup>1</sup>, Danica Agbaba<sup>1</sup>,  
Maija Lahtela-Kakkonen<sup>2</sup>**

<sup>1</sup>Katedra za Farmaceutsku hemiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija), <sup>2</sup>School of Pharmacy (Pharmaceutical Chemistry) University of Eastern Finland (Finska)

Modulacija aktivnosti epigenetičkog brisača, NAD-zavisne protein deacetilaze sirtuina 2 (SIRT2), poslednjih godina se pokazala kao obećavajuća strategija u lečenju Parkinsonove bolesti, depresije, određenih tipova kancera, ishemisko-reperfuzionih povreda itd. Nasuprot terapijskom potencijalu, još uvek nijedan predstavnik ove grupe farmakološki aktivnih supstanci nije našao svoje mesto na tržištu. Najčešći problemi sa dosada opisanim SIRT2 inhibitorima predstavljaju niska potentnost, loša selektivnost, kao i loše farmakokinetičke osobine što dalje opravdava razvoj novih predstavnika.

Cilj ovog rada je bio ispitivanje konformacionih promena SIRT2 u prisustvu inhibitora i dalje poboljšanje dostupnih kristalografskih modela u cilju razvoja efikasnijeg protokola virtual screening-a.

Polazeći od 5 različitih kristalografskih struktura SIRT2-inhibitor kompleksa, ukupno 1,5 μs simulacija molekulske dinamike u eksplicitnom solventu je izvedeno. 3D deskriptori zasnovani na GRID-u i linearna diskriminantna analiza su korišćeni za virtual screening (VS) studiju.

Konformaciona fleksibilnost SIRT2-inhibitor kompleksa zabeležena tokom simulacija ukazuje na značajnu fleksibilnost aktivnog mesta i posledično na multiple vezivne modove inhibitora. Nakon procedure klasterovanja trajektorije, nekoliko relevantnih modela kompleksa je izdvojeno i uključeno u dalju VS studiju. VS modeli generisani pomoću tri relevantna kompleksa dobijena studijom molekulske dinamike su pokazali značajno bolje performanse u poređenju sa modelima dobijenim pomoću do danas opisanih kristalografskih struktura. Performanse generisanog VS protokola su značajno poboljšane i u odnosu na do danas publikovane protokole. Rezultati ove studije jasno ukazuju na značaj uračunavanja fleksibilnosti aktivnog mesta u racionalni dizajn SIRT2 inhibitora. Novi hemotipovi potenijalnih inhibitora SIRT2 su izdvojeni iz baza komercijalno dostupnih jedinjenja primenom generisanih VS modela.

U ovoj studiji formirani su realističniji modeli aktivnog mesta sirtuina 2 kojima su značajno poboljšane performanse virtual screening-a u odnosu na do danas publikovane studije. Rezultati ove studije, uključujući i opisane konformacione promene doprinose sveobuhvatnom razumevanju odnosa strukture i aktivnosti SIRT2 inhibitora i dodatno racionalizuju dizajn selektivnijih i potentnijih inhibitora.

# **MOLECULAR DYNAMICS-BASED VIRTUAL SCREENING OF SIRTUIN 2 INHIBITORS**

**Nemanja Đoković<sup>1</sup>, Katarina Nikolić<sup>1</sup>, Danica Agbaba<sup>1</sup>,  
Maija Lahtela-Kakkonen<sup>2</sup>**

<sup>1</sup>Department of Pharmaceutical Chemistry, University of Belgrade - Faculty of Pharmacy (Serbia), <sup>2</sup>School of Pharmacy (Pharmaceutical Chemistry) University of Eastern Finland (Finland)

Modulation of activity of epigenetic eraser, NAD-dependent protein deacetylase sirtuin 2 (SIRT2), recently emerged as promising therapeutic strategy for the treatment of many diseases, including Parkinson's disease, depression, some types of cancers, necrotic injuries (ischemic stroke, myocardial infarction) etc. Contrary to therapeutic potential, none of SIRT2 inhibitors reported to date has been approved for the market. Some of the most common problems with current SIRT2 inhibitors include poor potency, selectivity and pharmacokinetic properties which justify further development of novel inhibitors.

The Aim of this study was to explore conformational space of sirtuin2-inhibitor complexes and further refinement of available crystallographic structures in order to develop more efficient virtual screening (VS) protocol.

Starting from five different crystallographic structures of SIRT2 co-crystallized with inhibitors, total of 1.5 μs of molecular dynamics (MD) simulations in explicit solvent has been performed. GRID-based 3D descriptors and linear discriminant analysis were used for virtual screening.

Significant conformational flexibility of SIRT2-inhibitor complexes was observed during simulations indicating overall binding site flexibility and multiple binding modes of inhibitors. Several atomistic models of SIRT2-inhibitor complexes were extracted and used for structure-based VS study. VS models generated from three extracted SIRT2-inhibitor complexes were significantly better compared to VS models generated from available crystallographic structures. Generated VS protocol was also better in performance compared to published virtual screening studies. These results clearly indicate importance of considering flexibility of binding site in rational design of SIRT2 inhibitors. Obtained models were used for screening of commercial databases of compounds. Several chemotypes of potential novel SIRT2 inhibitors have been identified.

Refined atomistic models of SIRT2-inhibitor complexes have been generated and significant improvement of virtual screening performance has been achieved. These results further rationalize design of SIRT2 inhibitors with improved selectivity and potency.

## ISPITIVANJE FIZIČKO-HEMIJSKIH SVOJSTAVA SMEŠA POLIMERA I POVRŠINSKI AKTIVNIH MATERIJA KAO POTENCIJALNIH NOSAČA LEKOVA

**Maja Milanović, Veljko Krstonošić, Milica Atanacković Krstonošić**

Katedra za farmaciju, Univerzitet u Novom Sadu - Medicinski fakultet (Srbija)

Polimeri i površinski aktivne materije (PAM) ulaze u sastav brojnih farmaceutskih formulacija u cilju povećanja stabilnosti ili predstavljaju nosioce formulacije. Prisustvo interakcija između polimera i PAM, može značajno uticati na osobine ovih sistema u zavisnosti od njihove prirode, koncentracije i nanelektrisanja. Poznavanje fizičko-hemijskih svojstava smeša polimer-PAM neophodno je u cilju razvoja farmaceutskih formulacija unapređenih osobina. Cilj istraživanja je određivanje prisustva i mehanizma interakcije između anjonskog polimera-ksantan gume i nejonske PAM-Tween 80 primenom infracrvene spektroskopije sa Fourierovom transformacijom (FT-IR) i tenziometrije.

Nakon FT-IR analize strukture ksantan gume i Tween 80, u opsegu talasnih brojeva od 4000 do 600 cm<sup>-1</sup> (pri rezoluciji od 4 cm<sup>-1</sup>), površinski napon smeša ksantan guma-Tween 80 određen je metodom prstena po *du Noüy-u* na 25°C ± 0,1. Analizom FT-IR spektara može se pretpostaviti da osim hidrofobnih interakcija, ksantan guma međudejstvo sa Tween 80 ostvaruje i formiranjem vodoničnih mostova.

Krine zavisnosti površinskog napona od koncentracije Tween 80, pre i posle dodatka konstantne koncentracije ksantan gume, se razlikuju usled prisustva polimer-PAM interakcija. Formiranjem kompleksa ksantan guma-Tween 80 u unutrašnjosti rastvora smanjuje se količina monomera PAM na granici faza voda-vazduh što rezultuje višim vrednostima površinskog napona smeše u odnosu na rastvor čiste PAM, iste koncentracije. Nakon zasićenja lanaca ksantan gume molekulima Tween 80, površinski napon smeše je konstantan i odgovara vrednostima rastvora čiste PAM. Dobijeni rezultati potvrđuju postojanje interakcija između ksantan gume i Tween 80. Ponašanje Tween 80 u prisustvu ksantan gume na granici faza voda-vazduh upućuje da su formirani kompleksi rezultat hidrofilnih i hidrofobnih interakcija.

## **PHYSICO-CHEMICAL EVALUATION OF POLYMER-SURFACTANT MIXTURES AS POTENTIAL DRUG DELIVERY SYSTEMS**

**Maja Milanović, Veljko Krstonošić, Milica Atanacković Krstonošić**

Department of Pharmacy, University of Novi Sad - Faculty of Medicine (Serbia)

Polymer and surfactants could be found in many drug delivery systems due to their individual properties. However, the characteristics of these systems may be influenced by the occurrence of interactions between polymer and surfactant which depend on nature, concentration, and net charge of both of them. Hence, the understanding of physico-chemical properties of polymer-surfactant mixtures is necessary in order to create appropriate and efficient drug delivery vehicles. The aim of this work was to determine the occurrence and possible mechanism of interactions between anionic polymer-xanthan gum and nonionic surfactant-Tween 80 using Fourier transform infrared spectrometry (FT-IR) and tensiometry.

After FT-IR analysis of pure substances that was performed in the wave number range 4000 to 600 cm<sup>-1</sup> (resolution of 4 cm<sup>-1</sup>), the surface tension measurements of xanthan gum-Tween 80 mixtures were done at 25°C ± 0.1 using a du Noüy ring method. The obtained FT-IR spectra imply that complexes between xanthan gum and Tween 80 could occur through hydrophobic interactions as well as hydrogen bonding.

The differences in the shape of curves with and without constant xanthan gum concentration, on the surface tension versus Tween 80 concentration plot, could be ascribed to the occurrence of polymer- surfactant interactions. The xanthan gum-Tween 80 complexes occurred in the bulk, and thus the amount of free surfactant monomer was decreased at the air-water interface resulting in the higher surface tension value of mixtures in compare to the pure surfactant solution of the same concentration. After the saturation of xanthan gum chains with Tween 80, the obtained surface tension values for mixtures corresponded to the values for pure Tween 80.

The obtained results confirmed the occurrence of xanthan gum-Tween 80 interactions. Based on the mixture behavior at air-water interface, xanthan gum and Tween 80 mainly form complexes through hydrophobic and hydrophilic association.

## ACE I $\alpha$ -GLUKOZIDAZNA INHIBITORNA AKTIVNOST METANOLNOG EKSTRAKTA *ALCHEMILLA VIRIDIFLORA* ROTHM. (ROSACEAE)

**Jelena Radović<sup>1</sup>, Nađa Grozdanić<sup>2</sup>, Tatjana Stanojković<sup>2</sup>,  
Relja Suručić<sup>3</sup>, Tatjana Kundaković-Vasović<sup>1</sup>**

<sup>1</sup>Katedra za farmakognoziju, Univerzitet u Beogradu - Farmaceutski fakultet,

<sup>2</sup>Institut za onkologiju i radiologiju Srbije, Beograd (Srbija), <sup>3</sup>Katedra za

farmakognoziju, Univerzitet u Banjoj Luci - Medicinski fakultet, Republika Srpska (Bosna i Hercegovina)

Tanini, polifenolni biljni metaboliti, značajno smanjuju postprandijalnu hiperglikemiju inhibicijom  $\alpha$ -glukozidaze, i stoga mogu biti efikasna strategija u kontroli dijabetesa tipa 2. Takođe, dokazano je i da nespecifično inhibiraju aktivnost angiotenzin-konvertujućeg enzima (ACE). Kako su tanini identifikovani samo u vrsti *Alchemilla vulgaris* L., cilj ovog istraživanja je da se odredi sadržaj tanina u do sada neistraženoj vrsti *A. viridiflora* Rothm. (Rosaceae), kao i inhibitorni uticaj na aktivnost ACE i  $\alpha$ -glukozidaze.

Ukupni sadržaj tanina u metanolnom ekstraktu *A. viridiflora* određen je prema propisu Ph. Eur. 9.0. Sivi metanolni ekstrakt, enzimski rastvor (400 mU/ml  $\alpha$ -glukozidaze u 0,1 M fosfatnom puferu) i supstrat, p-nitrofenil  $\alpha$ -D-glukopiranoid korišćeni su za kolorimetrijski test inhibitorne aktivnosti  $\alpha$ -glukozidaze. Kao pozitivna kontrola korišćena je akarboza. ACE inhibitorna aktivnost metanolnog ekstrakta ispitana je korišćenjem komercijalnog testa *ACE Kit- WST* (Dojindo Inc., Japan) prema uputstvu proizvođača. Procenat inhibicije enzima je izračunata  $IC_{50}$  vrednost, tj. procenjena koncentracija ekstrakta koja je izazvala 50% inhibicije aktivnosti enzima, koristeći linearnu regresionu analizu.

$IC_{50}$  vrednost metanolnog ekstrakta *A. viridiflora*, očitana sa dozno-zavisne krive iznosi  $2,6 \pm 0,5$   $\mu\text{g}/\text{ml}$ , i ekstrakt pokazuje bolju anti- $\alpha$ -glukozidazu aktivnost od standarda akarboze ( $IC_{50}=74,2 \pm 3,3$   $\mu\text{g}/\text{ml}$ ). Takođe, ispitivani ekstrakt pokazuje dozno-zavisnu inhibiciju ACE pri  $IC_{50}$  2  $\mu\text{g}/\text{ml}$ . Dobijeni rezultati su u korelaciji sa visokim sadržajem tanina u metanolnom ekstraktu *A. viridiflora* (3,74 %).

Pokazane inhibicije angiotenzin-konvertujućeg enzima i  $\alpha$ -glukozidaze čine metanolni ekstrakt vrste *A. viridiflora* pogodnim za dalje istraživanje u cilju pronaalaženja novih prirodnih proizvoda značajnih za terapiju kardiovaskularnih bolesti i dijabetesa.

*Istraživanje je podržano od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije (Projekat ON 173021).*

## **ACE AND $\alpha$ -GLUCOSIDASE INHIBITORY ACTIVITY OF METHANOL EXTRACT OF *ALCHEMILLA VIRIDIFLORA* ROTHM. (ROSACEAE)**

**Jelena Radović<sup>1</sup>, Nada Grozdanić<sup>2</sup>, Tatjana Stanojković<sup>2</sup>,  
Relja Suručić<sup>3</sup>, Tatjana Kundaković-Vasović<sup>1</sup>**

<sup>1</sup>Department of Pharmacognosy, University of Belgrade-Faculty of Pharmacy,

<sup>2</sup>Institute of Oncology and Radiology of Serbia, Belgrade (Serbia), <sup>3</sup>Department of Pharmacognosy, University of Banja Luka - Faculty of Medicine, Republic of

Srpska (Bosnia and Herzegovina)

Tannins, polyphenolic plant metabolites, significantly reduce postprandial hyperglycemia by inhibiting  $\alpha$ -glucosidase, and therefore can be an effective strategy for controlling type 2 diabetes. It has also been proven that they are non-specific inhibitors of the activity of angiotensin-converting enzyme (ACE). As tannins were identified only in the species *Alchemilla vulgaris* L., the aim of this study is to determine the content of tannins in the unexplored *A. viridiflora* Rothm. (Rosaceae), as well as the inhibitory effect on the activity of angiotensin-converting enzyme and  $\alpha$ -glucosidase.

The content of tannins in methanol extract of *A. viridiflora* was determined according to the Ph. Eur. 9.0. Dry methanol extract, enzyme solution (400 mU/ml of  $\alpha$ -glucosidase in 0.1 M phosphate buffer) and substrate, p-nitrophenyl  $\alpha$ -D-glucopyranoside were used for colorimetric  $\alpha$ -glucosidase inhibitory activity test. Acarbose was used as a positive control. The ACE inhibitory activity of the methanol extract was tested using the commercial *ACE Kit-WST* (Dojindo Inc., Japan) according to the manufacturer's instructions. The percentage of enzyme inhibition is the calculated IC<sub>50</sub> value, i.e. estimated concentration of the extract that caused 50% inhibition of enzyme activity using linear regression analysis.

The IC<sub>50</sub> of *A. viridiflora* methanol extract, read from the dose-dependent curve, was 2.6±0.5 µg/mL, and this extract demonstrated better anti- $\alpha$ -glucosidase activity than standard acarbose (IC<sub>50</sub>=74.2±3.3 µg/mL). In addition, the examined extract shows a dose-dependent inhibition of ACE with IC<sub>50</sub> 2 µg/mL. Obtained results were in correlation with high level of tannins in methanol extract of *A. viridiflora* (3.74%).

The proven inhibitions of ACE and  $\alpha$ -glucosidase make the methanol extract of *A. viridiflora* suitable for further scientific research in order to find a new natural product for the treatment of cardiovascular diseases and diabetes.

*The study was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (Project ON 173021).*

## PROCENA BEZBEDNOSNOG PROFILA ETARSKIH ULJA TAKSONA RODA *HERACLEUM* L. (APIACEAE) U ODNOSU NA UTVRĐENI SADRŽAJ FURANOKUMARINA

**Ljuboš Ušjak, Silvana Petrović**

Katedra za farmakognoziju, Univerzitet u Beogradu - Farmaceutski fakultet  
(Srbija)

Za etarska ulja izolovana iz različitih biljnih organa osam taksona roda *Heracleum* L. (*H. sphondylium* L., *H. sibiricum* L., *H. montanum* Schleich. ex Gaudin, *H. ternatum* Velen., *H. pyrenaicum* subsp. *pollinianum* (Bertol.) F. Pedrotti & Pignatti, *H. pyrenaicum* subsp. *orsinii* (Guss.) F. Pedrotti & Pignatti, *H. verticillatum* Pančić i *H. orphanidis* Boiss.), u prethodnim ispitivanjima pokazana je antimikrobnna, citotoksična i antioksidantna aktivnost. S obzirom da je u pojedinim od ovih etarskih ulja utvrđeno prisustvo potencijalno fototoksičnih furanokumarina (bergaptena, izobergaptena, pimpinelina i/ili izopimpinelina), cilj rada bio je da se izvrši kvantifikacija ukupnih furanokumarina i ustanoji maksimalni dozvoljeni dnevni unos ispitivanih ulja u skladu sa preporukama u odgovarajućem dokumentu Evropske agencije za lekove (Doc. Ref. EMEA/HMPC/317913/2006).

Furanokumarini su kvantifikovani gasnom hromatografijom, metodom eksternog standarda, na osnovu površina pikova detektovanih plameno-jonizacionim detektorom (FID). U skladu sa preporukom EMA, sadržaj ukupnih furanokumarina izražen je kao ksantotoksin (8-metoksipsoralen, 8-MOP).

Prema navedenom dokumentu EMA, smatra se da dnevni unos 1,5 mg furanokumarina izraženih kao 8-MOP putem biljnih lekovitih proizvoda ne doprinosi značajno ukupnom riziku, a da dnevni unos 15 µg ne predstavlja nikakav rizik. U skladu sa tim, dnevni unos ispitivanih etarskih ulja koji ne doprinosi značajno ukupnom riziku kreće se u opsegu 1,94-5,23 mL za 15 etarskih ulja korena, 5,23-15,68 mL za 14 ulja plodova i 2,90-15,68 mL za tri ulja listova ili cvasti, a unos koji ne predstavlja nikakav rizik kreće se u opsegu 0,02-0,05 mL za navedena etarska ulja korena, 0,05-0,16 mL za ulja plodova i 0,03-0,16 mL za ulja listova ili cvasti. U četiri ulja plodova i 24 ulja listova ili cvasti ispitivanih taksona roda *Heracleum* furanokumarini nisu detektovani.

Ovaj rad demonstrira praktičnu primenu aktuelnih preporuka EMA koje se odnose na maksimalni dozvoljeni dnevni unos furanokumarina u cilju utvrđivanja bezbednosnog profila biljnih preparata u kojima su ovi sastojci detektovani.

*Istraživanje je podržano od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije (Projekat ON 173021).*

## **EVALUATION OF SAFETY PROFILE OF THE ESSENTIAL OILS OF HERACLEUM L. TAXA (APIACEAE) RELATED TO DETERMINED FURANOCOUMARIN CONTENT**

**Ljuboš Ušjak, Silvana Petrović**

Department of Pharmacognosy, University of Belgrade - Faculty of Pharmacy  
(Serbia)

Essential oils of different plant parts of eight *Heracleum* L. taxa (*H. sphondylium* L., *H. sibiricum* L., *H. montanum* Schleich. ex Gaudin, *H. ternatum* Velen., *H. pyrenaicum* subsp. *pollinianum* (Bertol.) F. Pedrotti & Pignatti, *H. pyrenaicum* subsp. *orsinii* (Guss.) F. Pedrotti & Pignatti, *H. verticillatum* Pančić and *H. orphanidis* Boiss.) previously exhibited antimicrobial, cytotoxic and antioxidant activities. Considering that in some of these oils potentially phototoxic furanocoumarins were detected (bergapten, isobergapten, pimpinellin and/or isopimpinellin), the aim of this work was to quantify total furanocoumarins and estimate maximum daily intake of investigated oils, according to recommendations in corresponding document of European Medicines Agency (Doc. Ref. EMEA/HMPC/317913/2006).

Furanocoumarins were quantified using gas chromatography, by external standard method, based on peak areas obtained by flame ionization detector (FID). As proposed by EMA, the sum of furanocoumarins equivalent to xanthotoxin (8-methoxysoralen, 8-MOP) was calculated.

According to noted EMA document, daily exposure of 1.5 mg furanocoumarins expressed as 8-MOP through herbal medicinal products is not considered to contribute significantly to overall risk, and the intake of 15 µg is not considered to pose any unacceptable risk. Thus, daily intake of investigated essential oils, not contributing significantly to overall risk is in the range of 1.94-5.23 mL for 15 root essential oils, 5.23-15.68 mL for 14 fruit oils and 2.90-15.68 mL for three leaf or flower oils, and the intake, not posing any unacceptable risk is in the range of 0.02-0.05 mL for mentioned root oils, 0.05-0.16 mL for fruit oils and 0.03-0.16 mL for leaf or flower oils. In four fruit, and 24 leaf or flower oils of investigated *Heracleum* taxa furanocoumarins were not detected.

This work demonstrates practical application of current EMA recommendations, which refer to maximum daily intake of furanocoumarins in order to establish safety profile of herbal preparations containing these compounds.

*The study was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (Project ON 173021).*

## ISPITIVANJE POGODNOSTI GRANULATA OBLOŽENOG TOPLJENJEM ZA KOMPRIMOVANJE U TABLETE DEFINISANE DEBLJINE

**Ana Milanović<sup>1,2</sup>, Marija Bujišić<sup>1</sup>, Katarina Drezgić<sup>1</sup>,  
Jovana Drobnjak<sup>1</sup>, Ivana Aleksić<sup>1</sup>, Sandra Cvijić<sup>1</sup>**

<sup>1</sup>Agencija za lekove i medicinska sredstva Srbije, <sup>2</sup>Katedra za farmaceutsku tehnologiju i kozmetologiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Oblaganje topljenjem podrazumeva oblaganje biokompatibilnim lipidnim supstancama, bez primene rastvarača. Pored ostalog, dobijena lipidna obloga može uticati na tabletabilna svojstva dobijenog proizvoda (npr. granulata). Cilj ovog rada je da se ispitaju tabletabilna svojstva granula sa paracetamolom obloženih glicerol distearatom (Precirol® ATO 5) metodom topljenja, pri komprimovanju do zadate debljine tablete.

Granule sa paracetamolom (15 uzoraka obloženih topljenjem u modifikovanom uređaju tipa fluidizirajućeg sistema (Mycrolab, Huttlin, Nemačka) pod različitim procesnim uslovima), neobloženi granulat i smeša komponenti granulata komprimovani su pomoću uređaja Gamlen D serije (Gamlen Instruments Ltd, Velika Britanija), pri uslovima zadate debljine tablete (3 mm). Pritisak kompresije, rad kompresije, elastični oporavak i ejekcioni stres su obračunati na osnovu podataka koje generiše uređaj. Zatezna čvrstina je izračunata na osnovu čvrstine tableta određene pomoću uređaja Erweka TBH125D (Erweka GmbH, Nemačka). Sva merenja su urađena u triplikatu. Pri kompresiji neobloženog granulata do zadate debljine tablete primenjena je veća sila kompresije i veći rad (ukupni, neto, rad elastične sile) u odnosu na obložene granulate. Zatezna čvrstina komprimata izrađenih od neobloženog granulata (0,92 MPa) takođe je veća u odnosu na komprime obloženih uzoraka (0,53-0,83 MPa), sa izraženom varijabilnošću između neobloženih uzoraka. Zbog lubrikantnog svojstva lipidne oblage, ejekcioni stres komprimata obloženih granula smanjen je više od tri puta u odnosu na neobložene uzorke. Fizička smeša je pokazala najveći elastični oporavak, slabu kompaktibilnost i nedovoljnu čvrstinu dobijenih tableta (tablete su se lomile pre ispitivanja).

Tablete dobijene kompresijom obloženih granulata, pod uslovima zadate debljine, pokazale su prihvatljivu zateznu čvrstinu i nizak ejekcioni stres, ukazujući na dobre tabletabilne osobine uzoraka obloženih topljenjem. Tabletabilna svojstva neobloženog granulata su lošija, dok fizička smeša nije pogodna za komprimovanje u tablete zadate debljine.

## **ASSESSING THE ABILITY OF HOT MELT COATED GRANULES TO PRODUCE TABLETS OF CONTROLLED THICKNESS**

**Ana Milanović<sup>1,2</sup>, Marija Bujišić<sup>1</sup>, Katarina Drezgić<sup>1</sup>,**  
**Jovana Drobnjak<sup>1</sup>, Ivana Aleksić<sup>1</sup>, Sandra Cvijić<sup>1</sup>**

<sup>1</sup>Medicines and Medical Devices Agency of Serbia, <sup>2</sup>Department of Pharmaceutical Technology and Cosmetology, University of Belgrade - Faculty of Pharmacy (Serbia)

Hot-melt coating (HMC) refers to a solvent-free coating technique using biocompatible lipid materials. Among other properties, lipid coating can influence tableting properties of HMC product (e.g. granules). The aim of this study was to investigate tableting properties of paracetamol granules coated with glycerol distearate (Precirol® ATO5), when tablets are compressed to predetermined thickness.

HMC paracetamol granules (15 samples obtained in a modified fluid-bed apparatus (Mycrolab, Hüttlin, Germany) under different process parameters setups), uncoated granules and mixture of granules ingredients were compressed using Gamlen D series (Gamlen Instruments Ltd, UK) under fixed thickness (3 mm) operating mode. Compaction pressure, work of compaction, elastic recovery and ejection stress were calculated from the instrument generated data. Tensile strength was calculated from tablets hardness tested using Erweka TBH125D tester (Erweka GmbH, Germany). All measurements were done in triplicate. Fixed thickness compression of uncoated granules resulted in higher compression force and higher work of compaction (total, plastic, elastic) in comparison to HMC granules. Tensile strength of tablets obtained from uncoated samples (0.92 MPa) was also higher than for HMC granules (0.53-0.83 MPa), with notable variability between uncoated samples. Lubricating effect of lipid coating decreased ejection stress of tablets obtained from HMC granules more than three times, compared to uncoated samples. Physical mixture of ingredients showed the highest elastic recovery, poor compactibility and insufficient tablets hardness (tablets broke even before testing).

The assessment of HMC granules to produce tablets of controlled thickness indicated acceptable tablets tensile strength and low ejection force, demonstrating good tableting properties of all HMC samples. Tableting properties of uncoated granules were less favourable, while the tested physical mixture was not suitable for tableting under the target thickness.

## FORMULACIJA I OPTIMIZACIJA ORALNO-DISPERZIBILNIH TABLETA IZRAĐENIH DIREKTNOM KOMPRESIJOM SA VISOKIM UDELOM AKTIVNE SUPSTANCE

**Milica Drašković, Erna Turković, Jelena Đuriš, Jelena Paročić**

Katedra za farmaceutsku tehnologiju i kozmetologiju, Univerzitet u Beogradu-  
Farmaceutski fakultet (Srbija)

Najveći izazov u razvoju formulacije oralno-disperzibilnih tableta (ODT) je postizanje kratkog vremena raspadanja uz održavanje prihvatljivih mehaničkih karakteristika. Ovo se može postići primenom sistemskog pristupa formulaciji uz detaljnu analizu uticaja faktora formulacije i procesnih parametara na kritična svojstva kvaliteta tableta. Cilj ovog rada je razvoj i optimizacija formulacije ODT sa visokim udelom aktivne supstance namenjene direktnoj kompresiji koja poseduje prihvatljiva mehanička svojstva i kratko vreme raspadanja.

ODT su izrađene direktnom kompresijom smeše aktivne supstance (ibuprofen, odnosno kofein primjenjeni u masenom udelu 10-90%) i različitim komercijalno dostupnih koprocesovanih ekscipijenasa (Ludiflash®, Parteck® ODT, Disintequik™ ODT i Pharmaburst® 500). Odgovarajućim metodama ispitana je zatezna čvrstina i raspadljivost uzorka, nakon čega je primenom teorije perkolicije određen maksimalan udio aktivne supstance u formulaciji. Kako bi se procenio efekat procesnih parametara na kritična svojstva kvaliteta odabranih formulacija, kao i uticaj visokog u dela aktivne supstance na kompaktaciona svojstva koprocesovanih ekscipijenasa primenjena je dinamička analiza kompakcije.

Primenom teorije perkolicije uočeno je da raspadljivost predstavlja osetljiviji parametar kvaliteta ODT, u poređenju sa zateznom čvrstinom i da je najveći udio aktivnih supstanci moguće inkorporirati u uzorce sa Disintequik™ ODT i Pharmaburst® 500. Pomenuti uzorci su pokazali i najkraće vreme raspadanja uz optimalnu zateznu čvrstinu ( $> 1 \text{ MPa}$ ). Brzina kompresije je pokazala neznatan uticaj na ispitivana svojstva kvaliteta odabranih formulacija. Inkorporiranje aktivne supstance smanjilo je kompresibilnost koprocesovanih ekscipijenasa, međutim, pozitivno je uticalo na kompaktibilnost i tabletabilnost kod svih formulacija izuzev kod ODT sa Pharmaburst® 500 i ibuprofenum. Međutim, jedino kod pomenute formulacije povećanje pritiska kompresije nije značajno usporilo raspadanje, ukazujući na mogućnost primene nešto veće sile kompresije, kako bi se postigla prihvatljiva mehanička otpornost. Odabirom pogodnih koprocesovanih ekscipijenasa moguće je inkorporirati visok udio aktivne supstance u formulaciju ODT, bez narušavanja njihovih kompaktacionih svojstava i uz postizanje kratkog vremena raspadanja i optimalne mehaničke otpornosti.

# **FORMULATION AND OPTIMIZATION OF ORODISPERSIBLE TABLETS CONTAINING HIGH DRUG LOAD PREPARED BY DIRECT COMPRESSION**

**Milica Drašković, Erna Turković, Jelena Đuriš, Jelena Paročić**

Department of Pharmaceutical Technology and Cosmetology, University of Belgrade-Faculty of Pharmacy (Serbia)

The greatest challenge in orally disintegrating tablet (ODT) formulation development is achievement of fast disintegration, while maintaining the acceptable mechanical properties. Considering systematic approach to formulation development, it is necessary to gain knowledge about impact of formulation factors and process parameters on product quality. The aim of this study is formulation and optimization of ODT with high drug load, short disintegration time and acceptable tensile strength.

Samples are prepared by direct compression of tablet mixtures containing model drug (caffeine or ibuprofen in the range of 10-90%) and commercially available co-processed excipients (Ludiflash®, Parteck® ODT, Disintequik™ ODT and Pharmaburst® 500). After evaluation of tensile strength and disintegration time, percolation theory was applied to determine maximum drug load that can be incorporated in ODT formulation. Dynamic compaction analysis was used in order to assess effect of process parameters on ODT critical quality attributes, as well as influence of high drug load on compaction properties of co-processed excipients.

Based on the results obtained by percolation theory application, it can be assumed that disintegration is more critical ODT quality parameter than tensile strength. The highest drug load can be incorporated in samples containing Disintequik™ ODT and Pharmaburst® 500. Those samples have the shortest disintegration time and optimal mechanical properties. Compression speed did not influence evaluated critical quality attributes. Drug inclusion affected negatively compressibility, while compactability and tabletability in all formulations, except one containing Pharmaburst® 500 and ibuprofen, were improved. However, only in the mentioned formulation increase in compression pressure did not significantly prolong disintegration time, indicating to possibility of applying somewhat greater compression force in order to achieve acceptable mechanical characteristics. By selection of suitable co-processed excipient, it is possible to incorporate high drug load in ODT formulation, without disrupting compaction properties, and achieve optimal tensile strength followed by fast disintegration.

## SUPERKRITIČNA IMPREGNACIJA TABLETA MIKROKRISTALNE CELULOZE IBUPROFENOM

**Jovana Potpara<sup>1</sup>, Jasna Ivanović<sup>2</sup>, Svetlana Ibrić<sup>1</sup>**

<sup>1</sup>Katedra za farmaceutsku tehnologiju i kozmetologiju, Univerzitet u Beogradu - Farmaceutski fakultet, <sup>2</sup>Katedra za organsku hemijsku tehnologiju, Univerzitet u Beogradu - Tehnološko-metalurški fakultet (Srbija)

Bioaktivne komponente (antiinflamatorne, antimikrobne, antiseptične, itd.) se, u zavisnosti od svoje biološke uloge, mogu impregnirati u čvrste materijale različitim metodama. Jedna od takvih metoda je i superkritična impregnacija, koja podrazumijeva rastvaranje aktivne supstance u superkritičnom fluidu, pri čemu nastali rastvor dolazi u kontakt sa čvrstim matriksom (najčešće polimernim materijalom) koji će se impregnirati. Cilj rada je bio ispitivanje mogućnosti korišćenja mikrokristalne celuloze dobijene iz prirodnih sirovina, u formi tableta, kao nosača za aktivnu supstancu ibuprofen, metodom superkritične impregnacije uz pomoć ugljen-dioksida.

Mikrokristalna celuloza je iz pšeničkih ostataka dobijena metodom kisele hidrolize, koristeći 64% rastvor sumporne kiseline. Tablete mikrokristalne celuloze (mase 70 mg i prečnika 6 mm) su izrađene metodom direktnе kompresije pod opterećenjem od 100 kg, pomoću laboratorijskog simulatora kompakcije Gamlen Tablet Press-a (Gamlen Tableting Ltd, Velika Britanija). Impregnacija ibuprofena u izrađene tablete mikrokristalne celuloze pomoću superkritičnog ugljen-dioksida je izvršena u čeliji za rad pod visokim pritiscima (Eurotechnica GmbH, Njemačka). Superkritična impregnacija je izvedena pri pritisku od 10 MPa i temperaturi od 40°C, u trajanju od 2h, a masa impregniranog ibuprofena je određena gravimetrijski. Tablete mikrokristalne celuloze, dobijene iz pšeničnih ostataka, su uspješno impregnirane ibuprofenom pomoću superkritičnog ugljen-dioksida, pri odabranim uslovima (10 MPa, 40°C), pri čemu nisu pokazale znakove oštećenja. Iz razlike mase čvrstog nosača prije i posle impregnacije izračunat je prinos impregnacije ibuprofena, koji je iznosio 2,9%. U daljim eksperimentima neophodno je optimizovati uslove impregnacije, pri čemu bi se postigla bolja rastvorljivost ibuprofena u superkritičnom ugljen-dioksidu, kao i razmotriti korišćenje mikrokristalne celuloze u vidu praška umjesto tableta, sa ciljem dobijanja većih prinosa impregnacije.

## SUPERCRITICAL IMPREGNATION OF MICROCRYSTALLINE CELLULOSE TABLETS WITH IBUPROFEN

**Jovana Potpara<sup>1</sup>, Jasna Ivanovic<sup>2</sup>, Svetlana Ibric<sup>1</sup>**

<sup>1</sup>Department od Pharmaceutical Technology and Cosmetology, University of Belgrade - Faculty of Pharmacy, <sup>2</sup>Department od Organic Chemical Technology, University of Belgrade - Faculty of Technology and Metallurgy (Serbia)

Bioactive components (anti-inflammatory, antimicrobial, antiseptic, etc.), depending on their biological role, can be impregnated into solid materials by different methods. One of these methods is supercritical impregnation, which involves the penetration of supercritical fluid with a dissolved active substance into a solid matrix (usually polymer material) enabling distribution of the active substance through the whole material. The aim of this work was to investigate the possibility of using microcrystalline cellulose (MCC), obtained from natural raw materials, in the form of tablets as a carrier for the active substance ibuprofen by the method of supercritical impregnation with carbon dioxide.

MCC is obtained from wheat straw by acid hydrolysis method using 64% sulfuric acid solution. MCC tablets (70 mg weight and 6 mm diameter) were made using a direct compression method at a load of 100 kg, using the Gamlen tablet press laboratory simulator (Gamlen Tableting Ltd, UK). Impregnation of ibuprofen into manufactured MCC tablets was performed in a high pressure cell (Eurotechnica GmbH, Germany) using supercritical carbon dioxide. Supercritical impregnation was performed at a pressure of 10 MPa and a temperature of 40° C for a duration of 2 hours. Mass of the impregnated ibuprofen was determined gravimetrically. Microcrystalline cellulose tablets derived from wheat residues were successfully impregnated with ibuprofen using supercritical carbon dioxide under selected conditions (10 MPa, 40°C, 2h), without showing signs of damage. The yield of ibuprofen impregnation was 2.9% (calculated from the difference between the mass of the solid carrier before and after impregnation). In further experiments, it is necessary to optimize the conditions of impregnation in order to achieve better solubility of ibuprofen in supercritical carbon dioxide, as well as to consider the use of MCC as a powder instead of tablets in order to obtain higher yields of impregnation.

## UTICAJ SADRŽAJA LEKA NA DINAMIKU MEĐUPOVRŠINSKOG SLOJA NISKOENERGETSKIH NANOEMULZIJA – STUDIJA SA KURKUMINOM

**Ines Nikolić<sup>1</sup>, Evgenia Mitsou<sup>2</sup>, Dominique Jasmin Lunter<sup>3</sup>,**  
**Vassiliki Papadimitriou<sup>2</sup>, Aristotelis Xenakis<sup>2</sup>,**  
**Rolf Daniels<sup>3</sup>, Snežana Savić<sup>1</sup>**

<sup>1</sup>Katedra za farmaceutsku tehnologiju i kozmetologiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija), <sup>2</sup>Nacionalna helenska istraživačka fondacija, Institut za biologiju, medicinsku biohemiju i biotehnologiju, Atina (Grčka),

<sup>3</sup>Institut za farmaceutsku tehnologiju, Eberhard-Karls Univerzitet, Tübingen (Nemačka)

Niskoenergetske nanoemulzije (NE-NE) predstavljaju inovativne i multifunkcionalne nosače, čija su međufazna svojstva važna kako u kontekstu stabilnosti ovih sistema, tako i biofarmaceutskih postignuća. Kurkumin (KU), aktivna supstanca u ovom istraživanju, jeste molekula sa brojnim povoljnim efektima, ali zbog prilično zahtevnih fizičko-hemijskih osobina, njegovi potencijali i dalje ostaju neostvareni. Cilj rada bio je analiza dinamike međufaznog sloja razvijenih NE-NE, kao i procena lokalizacije KU unutar NE-NE i njegovog uticaja na organizaciju međupovršinske membrane, kao nagoveštaja mogućih performansi ovih nosača.

NE-NE su izrađene spontanoemulgajućim metodom, koristeći trigliceride srednje dužine lanca kao masnu fazu (10%), kombinaciju polisorbata 80 i lecitina soje u odnosu 9:1 kao stabilizatora (10%) i visokoprečišćenu vodu. Pripremljene su i formulacije sa 1, 2 i 3 mg/mL KU. Sprovedena je bazična fizičko-hemijska karakterizacija, praćena analizom termalnog ponašanja (diferencijalna skenirajuća kalorimetrija/DSC) i procenom dinamike međupovršinskog sloja (elektronska paramagnetska rezonantna spektroskopija/EPR).

Placebo NE-NE imale su prosečan dijametar kapi  $111,3 \pm 1,73$  nm, koji se povećavao srazmerno sadržaju KU, ostajući uvek ispod 150 nm, uz usku distribuciju veličina kapi u svim slučajevima. DSC merenja su pokazala izražen endotermni pik koji odgovara isparavanju vode iz uzorka, pomerajući se ka nižim temperaturama u formulacijama sa KU (koncentraciono zavisno). To može biti povezano sa interakcijom hidroksilnih grupa KU sa hidrofilnim delom međupovršine, uzrokujući određeno preuređenje u ovom regionu. Slična, ali konkretnija zapažanja zabeležena su primenom EPR, otkrivši 2 različite mikrosredine u međufaznom sloju, zavisno od rasporeda surfaktanata: regioni sačinjeni dominantno od polisorbata 80, i regioni sa prisutnim lecitinom - oba KU-interagujuća. Nakon inkapsulacije, KU je dominantno bio prisutan u lipofilnom delu membrane surfaktanata. Regioni bogati lecitinom su postali fluidniji sa povećanjem koncentracije KU.

Dobijeni rezultati mogu implicirati potencijalnu korist ovih nosača za KU, posebno u topikalnoj primeni, s obzirom na to da je međupovršinska lokalizacija aktivne molekule u ovom slučaju poželjna jer može obezbediti veću raspoloživost na mestu primene.

# **DRUG LOADING INFLUENCE ON THE INTERFACIAL MEMBRANE DYNAMICS OF THE LOW-ENERGY NANOEMULSIONS -A CURCUMIN CASE STUDY**

**Ines Nikolić<sup>1</sup>, Evgenia Mitsou<sup>2</sup>, Dominique Jasmin Lunter<sup>3</sup>,  
Vassiliki Papadimitriou<sup>2</sup>, Aristotelis Xenakis<sup>2</sup>,  
Rolf Daniels<sup>3</sup>, Snežana Savić<sup>1</sup>**

<sup>1</sup>Department of Pharmaceutical Technology and Cosmetology, University of Belgrade - Faculty of Pharmacy (Serbia), <sup>2</sup>National Hellenic Research Foundation, Institute for Biology, Medicinal Chemistry and Biotechnology, Athens (Greece), <sup>3</sup>Institute of Pharmaceutical Technology, Eberhard-Karls University Tübingen (Germany)

Low-energy nanoemulsions (LE-NEs) represent novel multifunctional carriers. Their interface characteristics are crucial not only in the context of stability, but also for biopharmaceutical behavior. Curcumin (CU), active substance used in this study, is a powerful pleiotropic molecule. Due to its physicochemical issues, its potentials are still beyond the reach. In the scope of this research was assessment of interfacial properties of developed LE-NEs with regard to CU's concentration, as a hint of prospective performances.

LE-NE were prepared via spontaneous emulsification, using medium-chain triglycerides as the oil phase (10%), combination of polysorbate 80 and soybean lecithin in the ratio 9:1 as stabilizers (10%) and ultrapure water. CU-loaded formulations contained 1, 2 and 3 mg/mL of CU. Basic physicochemical characterization was performed, followed by thermal behavior analysis (differential scanning calorimetry/DSC), and interfacial membrane dynamics assessment (electron paramagnetic resonance spectroscopy/EPR).

The placebo LE-NE exhibited mean droplet diameter of  $111.3 \pm 1.73$  nm, which augmented with increase in the CU content, but remained below 150 nm, with narrow distribution in all cases. DSC showed intense endothermic peak corresponding to the water evaporation, shifting towards lower temperatures for CU-loaded formulations (concentration dependent manner). This might be related to the interactions of CU's hydroxyl groups with the hydrophilic part of the interface, causing some rearrangements in this region. Similar, but more specific findings were captured by EPR, revealing 2 different interfacial microenvironments with respect to the surfactant distribution: regions with and without lecithin, both interacting with CU. Upon encapsulation, CU's participation closer to the lipophilic parts of the surfactant layer was disclosed. Lecithin-rich regions became more fluid with increase in CU concentration, but interface still remained rigid.

Obtained results may imply potentially beneficial role of developed LE-NEs for CU delivery, especially for topical application, because, in this case, interfacial localization is a preferred drug locus for higher availability.

## BIOKOMPATIBILNE NANOEMULZIJE ZA ISPORUKU ACEKLOFENAKA U/KROZ KOŽU PRIMENOM HEMIJSKIH POJAČIVAČA PENETRACIJE I ČVRSTIH MIKROIGALA

**Tanja Ilić<sup>1</sup>, Sanela Savić<sup>2</sup>, Bojan Batinić<sup>3</sup>, Jelena Đoković<sup>1</sup>, Bojan Marković<sup>4</sup>, Miroslav Savić<sup>5</sup>, Snežana Savić<sup>1</sup>**

<sup>1</sup>Katedra za farmaceutsku tehnologiju i kozmetologiju, Univerzitet u Beogradu - Farmaceutski fakultet, <sup>2</sup>DCP Hemigal, Leskovac, <sup>3</sup>Katedra za fiziologiju, Univerzitet u Beogradu - Farmaceutski fakultet, <sup>4</sup>Katedra za farmaceutsku hemiju, Univerzitet u Beogradu - Farmaceutski fakultet, <sup>5</sup>Katedra za farmakologiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Tokom poslednje decenije, neželjeni efekti udruženi sa hroničnom oralnom primenom aceklofenaka (ACF), podstakli su brojna istraživanja ka razvoju različitih metoda za poboljšanje isporuke ACF u/kroz kožu kako bi se postiglo efikasno lečenje bolesti koštano-mišićnog sistema. Otuda, cilj ove studije je bio da se ispita sposobnost nanoemulzija stabilizovanih smešom lecitina i saharoznih estara, sa/bez predtretmana kože čvrstim mikroiglama, da poboljšaju isporuku ACF u/kroz kožu.

Tri nanoemulzije ACF koje su se razlikovale u sadržaju lecitina, saharoza palmitata i stearata poredene su sa referentnim uzorkom stabilizovanim smešom lecitina i polisorbata 80 u pogledu fizičko-hemijskih karakteristika, dugoročne stabilnosti i *in vitro* oslobađanja/permeacije ACF. Stepen preuzimanja ACF u flikule dlake procenjen je primenom diferencijalnog *stripping*-a na koži uha svinje. Dodatno, određeni su farmakokinetički profili ACF u plazmi (uključujući i sadržaj ACF deponovanog u koži) pacova Wistar soja, nakon transdermalne primene odabranih nanoemulzija, sa/bez predtretmana čvrstim mikroiglama, kao fizičkim inhenserima.

Karakterizacija je pokazala zadovoljavajuć opseg veličina kapi (~180nm), relativno usku raspodelu veličina (<0,15), visoko površinsko nanelektrisanje (oko -40mV), i zadovoljavajuću dugoročnu stabilnost (godinu dana na 4±1°C) formulacija kostabilizovanih saharoza palmitatom i polisorbatom 80. *In vitro* ispitivanje oslobađanja/permeacije i diferencijalni *stripping* potvrdili su superiornost nanoemulzija na bazi saharoznih estara u odnosu na nanoemulziju sa polisorbatom 80. Međutim, rezultati dobijeni *in vitro* nisu bili u potpunosti u skladu sa nalazima *in vivo* - nisu uočene značajne razlike između ispitivanih formulacija u farmakokineticima i ukupnoj količini ACF deponovanog u koži 24h nakon primene, pri čemu su, istovremeno, ukazali na odloženu isporuku ACF u sistemsku cirkulaciju. Konačno, predtretman kože mikroiglama rezultovao je 1,4-2,1 puta povećanjem biološke raspoloživosti, kao i 1,2-1,7 puta povećanjem sadržaja aceklofenaka zadržanog u koži pacova.

Dobijeni rezultati ukazuju da je kombinacija mikroigla i nanoemulzije kostabilizovane saharoza palmitatom korisna za postizanje veće koncentracije ACF u koži, dok je kombinacija mikroigala i nanoemulzije kostabilizovane polisorbatom 80 pogodnija za postizanje veće koncentracije ACF u krvotoku.

# BIOCOMPATIBLE NANOEMULSIONS FOR ACECLOFENAC DELIVERY INTO/THROUGH THE SKIN USING CHEMICAL PENETRATION ENHANCERS AND SOLID MICRONEEDLES

**Tanja Ilić<sup>1</sup>, Sanela Savić<sup>2</sup>, Bojan Batinić<sup>3</sup>, Jelena Đoković<sup>1</sup>,  
Bojan Marković<sup>4</sup>, Miroslav Savić<sup>5</sup>, Snežana Savić<sup>1</sup>**

<sup>1</sup>Department of Pharmaceutical Technology and Cosmetology, University of Belgrade - Faculty of Pharmacy, <sup>2</sup>DCP Hemigal, Leskovac, <sup>3</sup>Department of Physiology, University of Belgrade-Faculty of Pharmacy, <sup>4</sup>Department of Pharmaceutical Chemistry University of Belgrade-Faculty of Pharmacy, <sup>5</sup>Department of Pharmacology University of Belgrade-Faculty of Pharmacy (Serbia)

Over the past decade, adverse effects associated with chronic oral administration of aceclofenac (ACF) enforced intensive research efforts towards exploring different penetration enhancement technologies aiming to ensure effective treatment of musculoskeletal disorders via skin. Hence, this study was designed to investigate the potential of lecithin-based nanoemulsions costabilized by sucrose esters, with/without skin pretreatment with solid microneedles, to improve delivery of ACF into/across the skin.

Three ACF-loaded nanoemulsions differing in the ratio of lecithin, sucrose palmitate and stearate, were compared, and with the reference stabilized with lecithin/polysorbate 80, regarding physicochemical properties, long-term stability and in vitro drug release/permeation. The extent of ACF follicular uptake was assessed using differential stripping on porcine ear skin. Additionally, the plasma pharmacokinetics of ACF (including quantification of ACF amount retained in the skin) after topical administration of formulated nanoemulsions, with/without skin perforation using solid microneedles, as physical enhancers, was investigated in Wistar rats.

The characterization revealed favorable droplet size (~180nm), narrow size distribution (<0.15), high surface charge (about -40mV) and satisfying long-term stability (one year at 4±1°C) of the formulations costabilized by sucrose palmitate and polysorbate 80. In vitro release/permeation testing and differential stripping proved the superiority of sucrose ester- over polysorbate-based nanoemulsion. However, in vitro findings were not fully indicative of the in vivo performances – no significant differences were observed between investigated formulations in pharmacokinetics and total amount of ACF deposited in the skin 24h after dosing, simultaneously pointing to delayed ACF delivery into the systemic circulation. Finally, skin pretreatment with microneedles led to 1.4–2.1-fold increased bioavailability and 1.2–1.7-fold enhanced level of ACF retained in the skin.

Obtained results suggest that combination of microneedles and sucrose palmitate-costabilized nanoemulsion could be useful to attain higher skin concentration, while combination of microneedles with polysorbate 80-costabilized one could be preferable for enhancing ACF delivery into the bloodstream.

## ISPITIVANJE VARIJABILNOSTI U KONCENTRACIJAMA METOTREKSATA IZMEĐU CIKLUSA TERAPIJE

**Biljana Škorić<sup>1</sup>, Marija Jovanović<sup>1</sup>, Branislava Miljković<sup>1</sup>,**  
**Miloš Kuzmanović<sup>2,3</sup>, Dragan Mićić<sup>3</sup>, Katarina Vučićević<sup>1</sup>**

<sup>1</sup>Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu - Farmaceutski fakultet, <sup>2</sup>Univerzitet u Beogradu - Medicinski fakultet, <sup>3</sup>Institut za zdravstvenu zaštitu majke i deteta Srbije „Dr Vukan Čupić”, Beograd (Srbija)

Primena intenzivnih terapijskih protokola u lečenju akutne limfomblastne leukemije (ALL) i non-Hodgkin limfoma (NHL), uz istovremenu primenu više različitih citotoksičnih agenasa, dovodi do visokog procenta izlečenja pedijatrijskih pacijenata. Metotreksat se primenjuje u visokim dozama i zbog značajne inter- i intra-individualne varijabilnosti u farmakokinetici indikovano je terapijsko praćenje leka (*Therapeutic drug monitoring, TDM*). Cilj ovog rada je bio da se ispita individualna varijabilnost u koncentracijama metotreksata između različitih ciklusa terapije.

U studiju su uključeni pedijatrijski pacijenati sa dijagnozom ALL ili NHL lečeni na Institutu za zdravstvenu zaštitu majke i deteta Srbije „Dr Vukan Čupić“. Podaci o pacijentima su prikupljeni retrospektivno iz medicinskih istorija. Etički odbor Instituta je odobrio sprovođenje studije. Statistička analiza podataka kod pacijenata koji su primenjivali dozu od  $5 \text{ g/m}^2$  metotreksata je izvršena primenom programa  $R^{\circledast}$ .

Kod 38 pacijenata je izmereno ukupno 122 koncentracije metotreksata 24 h nakon početka terapije ( $76,7 \pm 37,7 \text{ } \mu\text{mol/L}$ ), pri čemu je broj ciklusa bio od 1 do 4. Mediana koeficijenta varijacije izmernih koncentracija metotreksata, uzimajući u obzir individualnu varijabilnost između ciklusa kod svakog pacijenta, je bila 32,3%, dok je raspon bio od 6,69 do 106%. Kod 36 pacijenata je bilo dostupno 113 koncentracija nakon 48 h ( $0,47 \pm 1,26 \text{ } \mu\text{mol/L}$ ), a broj ciklusa je bio od 1 do 4. Koeficijent varijacije je iznosio od 7,07 do 152%, dok je mediana bila 44%. 72 h nakon početka terapije je izmereno 17 koncentracija metotreksata ( $0,263 \pm 0,442 \text{ } \mu\text{mol/L}$ ) kod 8 pacijenata, pri čemu je broj ciklusa bio od 1 do 4. Mediana koeficijenta varijacije je iznosila 26,2%, dok je raspon bio u opsegu od 10,9 do 129%.

Rezultati analize pokazuju značajnu varijabilnost u koncentracijama metotreksata uzimajući u obzir individualnu varijabilnost između ciklusa kod svakog pacijenta, ukazujući na značaj *TDM-a* i ispitivanje faktora koji doprinose varijabilnosti.

*Istraživanje je realizovano u okviru projekta broj 175023 finansiranog od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije.*

## INVESTIGATION OF VARIABILITY IN METHOTREXATE CONCENTRATIONS BETWEEN THERAPY CYCLES

**Biljana Škorić<sup>1</sup>, Marija Jovanović<sup>1</sup>, Branislava Miljković<sup>1</sup>,**  
**Miloš Kuzmanović<sup>2,3</sup>, Dragan Mićić<sup>3</sup>, Katarina Vučićević<sup>1</sup>**

<sup>1</sup>Department of Pharmacokinetics and Clinical Pharmacy, University of Belgrade - Faculty of Pharmacy, <sup>2</sup>University of Belgrade - School of Medicine,  
<sup>3</sup>Institute for Mother and Child Healthcare „Dr Vukan Čupić”, Belgrade (Serbia)

The application of intensive therapeutic protocols for the treatment of acute lymphoblastic leukemia (ALL) and non-Hodgkin lymphoma (NHL), with the simultaneous administration of several different cytotoxic agents, leads to good treatment outcomes in paediatric patients. Methotrexate is administered in high doses and due to significant inter- and intra-individual variability in the pharmacokinetics therapeutic drug monitoring (TDM) is indicated. The aim of the study was to evaluate individual variability in methotrexate concentration between different therapy cycles.

The study included paediatric patients diagnosed with ALL or NHL treated with 5 g/m<sup>2</sup> of methotrexate at the Institute for mother and child healthcare „Dr Vukan Čupić”. Data were retrospectively collected from medical charts. The Ethics Committee of the Institute approved the study protocol. Statistical analysis was performed with program R®.

In 38 patients, a total of 122 methotrexate concentrations were measured 24h after initiation of therapy ( $76.7 \pm 37.7 \text{ } \mu\text{mol/L}$ ), while the number of cycles was 1-4. Median value of coefficient of variation (CV) in methotrexate concentrations, taking into account the individual variability between cycles in each patient, was 32.3%, while the range was 6.69-106%. In 36 patients, 113 concentrations were available after 48h ( $0.47 \pm 1.26 \text{ } \mu\text{mol/L}$ ) and the number of cycles 1-4. Values of CV were from 7.07 to 152%, while the median was 44.0%. 72h after initiation of therapy, 17 concentrations of methotrexate ( $0.263 \pm 0.442 \text{ } \mu\text{mol/L}$ ) were measured in 8 patients, with the number of cycles from 1 to 4. The median value of CV was 26.2%, while the range was from 10.9 to 129%.

The results of the analysis present significant variability in methotrexate concentrations taking into account the individual variability between cycles in each patient, indicating the importance of TDM and assessment of the sources of variability.

*This work was conducted as a part of the project No. 175023 funded by the Ministry of Education, Science and Technological Development, Republic of Serbia.*

## OPTIMIZACIJA PROTOKOLA ZA SAKUPLJANJE UZORAKA KRVI ZA ISPITIVANJE FARMAKOKINETIKE ZONISAMIDA KOD PEDIJATRIJSKIH PACIJENATA

**Maša Roganović<sup>1</sup>, Branislava Miljković<sup>1</sup>, Marija Jovanović<sup>1</sup>,**  
**Andelija Malenović<sup>2</sup>, Nebojša Jović<sup>3</sup>, Katarina Vučićević<sup>1</sup>**

<sup>1</sup>Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu - Farmaceutski fakultet, <sup>2</sup>Katedra za analitiku lekova, Univerzitet u Beogradu - Farmaceutski fakultet, <sup>3</sup>Klinika za neurologiju i psihijatriju za decu i omladinu Kliničkog centra Srbije, Univerzitet u Beogradu - Medicinski fakultet (Srbija)

Pri terapijskom praćenju antiepileptika (*Therapeutic Drug Monitoring, TDM*) najčešće se uzima jedan uzorak biološkog materijala koji odgovara minimalnoj koncentraciji leka u krvi. Prilikom planiranja kliničke studije, sa ciljem procene farmakokinetičkih parametara za odgovarajući/e proces/e, neophodno je proceniti optimalan broj uzoraka biološkog materijala i vremena uzorkovanja u odnosu na primjenjenu dozu leka. Dodatno, sa aspekta razvoja i validacije bioanalitičke metode za određivanje koncentracije leka, od značaja je poznavanje očekivanog raspona koncentracija nakon uobičajenih režima doziranja. Cilj ovog rada jeste predviđanje raspona koncentracija zonisamida u plazmi i optimizacija protokola uzimanja uzorka krvi kod pacijenata na kombinovanoj terapiji zonisamidom kako bi se obezbedila adekvatna procena parametara u okviru populacione farmakokinetičke analize.

Za optimizaciju protokola uzimanja biološkog materijala, korišćen je softver PFIM (v.4. 0). Ulazni podaci obuhvataju literaturno dostupne vrednosti populacionih farmakokinetičkih parametara (ka, Vd, CL) zonisamida i njihove interindividualne varijabilnosti i rezidualne greške, očekivan broj pacijenata u planiranoj studiji i uobičajene režime doziranja zonisamida (100-400 mg/12 h). Na osnovu navedenih podataka, simulirani su koncentracija-vreme (C-t) profili zonisamida, a dodatno uz inicijalno predložen broj uzoraka i vremena uzorkovanja izvršena je optimizacija eksperimentalnog dizajna preko Fedorov-Wynn algoritma.

Očekivan raspon koncentracija zonisamida uplazmi je 2-65 µg/mL. Predložena vremena za sakupljanje uzoraka krvi su 0,5, 1, 1,5, 2, 4, 6, 8, 11,5, 12 h nakon primjenjene doze. Optimizacija je izvršena za tri, odnosno četiri uzorka po pacijentu. Ukoliko je maksimalan broj uzoraka po pacijentu (u grupi od 30 pacijenata) četiri, optimalno vreme uzorkovanja je u sledećim vremenskim tačkama: 1,5, 4, 8, 12 h. Za tri uzorka po pacijentu, optimalno uzorkovanje je 2, 6 i 12 h nakon primjenjene doze.

Simulacija C-t profila daje adekvatnu podršku u razvoju i validaciji bioanalitičke metode, dok optimizovani protokoli uzimanja uzoraka omogućavaju precizniju i tačniju procenu populacionih vrednosti farmakokinetičkih parametara, njihovih interindividualnih varijabilnosti kao i rezidualne greške.

*Istraživanje je realizovano u okviru projekta broj 175023 finansiranog od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije.*

# OPTIMIZATION OF THE BLOOD SAMPLING PROTOCOL FOR THE ZONISAMIDE PHARMACOKINETIC STUDY IN PEDIATRIC PATIENTS

**Maša Roganović<sup>1</sup>, Branislava Miljković<sup>1</sup>, Marija Jovanović<sup>1</sup>,**  
**Andelija Malenović<sup>2</sup>, Nebojša Jović<sup>3</sup>, Katarina Vučićević<sup>1</sup>**

<sup>1</sup>Department of Pharmacokinetics and Clinical Pharmacy, University of Belgrade - Faculty of Pharmacy, <sup>2</sup>Department of Drug Analysis, University of Belgrade - Faculty of Pharmacy, <sup>3</sup>Clinic of Neurology and Psychiatry for Children and Youth, University of Belgrade - School of Medicine (Serbia)

During the therapeutic drug monitoring (TDM) of antiepileptics, blood samples are taken just before the next oral dose. To adequately estimate specific pharmacokinetic parameter/s, it is necessary to estimate the optimal number of blood samples and sampling times. For the development and validation of a bioanalytical method for measuring drug levels, expected concentration ranges following the typical dosing regimens are required. The aim of this study is to predict the range of plasma concentrations, and to optimise blood sampling protocol in patients receiving zonisamide.

PFIM (v.4.0) was used to optimise the study protocol. The input data include available values of the population pharmacokinetic parameters ( $k_a$ ,  $V_d$ ,  $CL$ ) of zonisamide, the interindividual variabilities and residual errors, the expected number of patients in the planned study and the usual dosing regimens of zonisamide (100-400 mg/12 h). Based on the given data, concentration-time (C-t) zonisamide profiles were simulated. Additionally, optimization of the experimental design was carried out through the Fedorov-Wynn algorithm and using the initially proposed number of samples and the sampling times.

The expected range of zonisamide plasma concentrations is 2-65 µg/mL. The suggested times for blood samples collection were: 0.5, 1, 1.5, 2, 4, 6, 8, 11.5, 12 h after the dose. Optimization was done for three and four samples per patient. If the maximum number of samples per patient (in a group of 30 patients) is four, the optimum sampling times are at: 1.5, 4, 8, 12 h. For three samples per patient, optimal sampling was at 2, 6, 12 h.

C-t profile simulation provides adequate support for the development and validation of the bioanalytical method, while optimised sampling protocols allow accurate estimation of the population pharmacokinetic parameters, their interindividual variability, and residual errors.

*This work was conducted as a part of the project No.175023 funded by the Ministry of Education, Science and Technological Development, Republic of Serbia.*

## ISPITIVANJE UTICAJA FUNKCIONALNOG VOLUMENA ŠTITASTE ŽLIJEZDE NA VJEROVATNOĆU ISHODA TERAPIJE 131I KOD PACIJENATA SA BENIGNIM OBOLJENJIMA ŠTITASTE ŽLIJEZDE

**Valentina Topić Vučenović<sup>1</sup>, Dijana Jelić<sup>1</sup>, Zvezdana Rajkovača<sup>2</sup>,**  
**Branislava Miljković<sup>3</sup>, Katarina Vučićević<sup>3</sup>**

<sup>1</sup>Odsjek za farmaciju, Univerzitet u Banjoj Luci - Medicinski fakultet,

<sup>2</sup>Univerzitet u Banjoj Luci - Medicinski fakultet (Bosna i Hercegovina), <sup>3</sup>Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Cilj ove studije je bio ispitivanje i kvantifikovanje uticaja funkcionalnog volumena štitaste žlijezde na vjerovatnoću ishoda terapije 131I kod pacijenata sa benignim oboljenjima štitaste žlijezde razvojem binarnog logističkog regresionog modela.

Podaci za analizu su retrospektivno prikupljeni iz medicinskih kartona pacijenata. Dimenzije štitaste žlijezde su određene ultrazvučnom metodom. Za pacijente sa Grejvsovom bolešću (GB) i multinodularnom gušom (MNG) funkcionalni volumen je određen kao ukupni volumen štitaste žlijezde, dok je kod pacijenata sa toksičnim adenomom (TA) volumen autonomnog („vrućeg“) čvora uzet kao funkcionalni volumen. Za aproksimaciju volumena režnjeva, istmusa i čvorova štitaste žlijezde upotrijebljena je formula za volumen elipsoida ( $V = \pi/6 \times \text{dužina (cm)} \times \text{širina (cm)} \times \text{dubina (cm)}$ ). Klinički ishod je procijenjen godinu dana nakon terapije 131I, a kao uspješan ishod je razmatran eu ili hipotireoidizam. Analiza je sprovedena pomoću softvera NONMEM® (v7.3), PsN® (v4.6.0) i R Studio (v1.0.153).

Podaci za analizu su obuhvatili 95 kliničkih ishoda određenih godinu dana nakon primjene 131I kod 95 odraslih pacijenata (57 (60%) sa GB, 21 (22,1%) sa MNG i 17 (17,9%) sa TA). Prema dobijenom modelu, odnos šansi za uspješan ishod terapije se smanjuje za 19,3% (CI: 17,2 – 21,4%) za svakih 5 mL povećanja funkcionalnog volumena preko vrijednosti medijane (31,06 mL za GB, 46,13 mL za MNG i 10,13 mL za TA) pri medijani apsorbovane doze zračenja (199,43 Gy). Vjerovatnoća uspješnog ishoda pri vrijednostima medijane prediktorskih varijabli iznosi 0,695.

Analiza je pokazala da je funkcionalni volumen štitaste žlijezde statistički značajan prediktor vjerovatnoće ishoda terapije 131I i da ga je potrebno uzeti u obzir pri određivanju doze radioaktivnosti za uspješan ishod terapije kod pacijenata sa benignim oboljenjima štitaste žlijezde.

# INVESTIGATION OF THE INFLUENCE OF FUNCTIONAL THYROID VOLUME ON THE PROBABILITY OF $^{131}\text{I}$ THERAPY OUTCOME IN PATIENTS WITH BENIGN THYROID DISEASE

**Valentina Topić Vučenović<sup>1</sup>, Dijana Jelić<sup>1</sup>, Zvezdana Rajkovača<sup>2</sup>,**  
**Branislava Miljković<sup>3</sup>, Katarina Vučićević<sup>3</sup>**

<sup>1</sup>Department of Pharmacy, University of Banja Luka - Faculty of Medicine,

<sup>2</sup>University of Banja Luka - Faculty of Medicine (Bosnia and Herzegovina),

<sup>3</sup>Department of Pharmacokinetics and Clinical Pharmacy, University of Belgrade - Faculty of Pharmacy (Serbia)

The objective of the study was to investigate and quantify the influence of functional thyroidal volume on the probability of the outcome of  $^{131}\text{I}$  therapy in patients with benign thyroid disease by a development of a binary logistic regression model.

Data for analysis were retrospectively collected from patients' medical records. The dimensions of the thyroid gland were determined by ultra-sonography. For the patients with Graves' disease (GD) and multi-nodular goitre (MNG) functional volume was total thyroid volume, whereas in patients with toxic adenoma (TA) autonomous ("hot") nodule was considered as functional volume. The ellipsoid volume formula ( $V = \pi/6 \times \text{length (cm)} \times \text{width (cm)} \times \text{depth (cm)}$ ) was used for approximation of the volume of thyroid lobes, isthmus and nodules. The clinical outcome was evaluated 1 year after  $^{131}\text{I}$  therapy and a successful outcome was eu- or hypothyroidism. The analysis was performed using NONMEM® (v7.3), PsN® (v4.6.0) and R Studio (v1.0.153) software.

Data for analysis included 95 clinical outcomes obtained 1 year after  $^{131}\text{I}$  therapy from 95 adult patients (57 (60%) with GD, 21 (22.1%) with MNG and 17(17.9%) with TA). According to the model, the odds ratio of having successful outcome decreased by 19.3% (CI: 17.2 – 21.4%) for each 5 mL increase of the functional volume over the median value (31.06 mL, 46.13mL and 10.13 mL for GD, MNG and TA, respectively) at median value of absorbed radiation dose (199.43 Gy). Baseline probability of successful outcome at median values of the predictor variables was 0.695.

The analysis showed that the functional thyroidal volume is statistically significant predictor of the probability of  $^{131}\text{I}$  therapy outcome in hyperthyroid patients and should be considered when determining dose of radioactivity necessary for a successful outcome in patients with benign thyroid disease.

**Predavanja po pozivu**

**Invited Lectures**

# SADRŽAJ – CONTENTS

PP 1

- HOMOLOGY MODELING OF HUMAN HISTONE DEACETYLASE 10 AND DESIGN OF POTENTIAL SELECTIVE INHIBITORS  
- [Abdullahi Ibrahim Uba, Kemal Yelekçi](#) 103

PP 2

INHIBITORI EPIGENETSKIH ENZIMA IZ PRIRODNIH IZVORA

INHIBITORS OF EPIGENETIC ENZYMES FROM NATURAL SOURCES

- [Slavica Erić](#) 104

PP 3

FIZIČKO-HEMIJSKA I ADME KARAKTERIZACIJA ANALOGA ESTARA ETILENDIAMIN-N,N'-DI-2-(3-CIKLOHEKSIL) PROPANSKE KISELINE SA POTENCIJALNIM CITOTOKSIČNIM DEJSTVOM

PHYSICOCHEMICAL AND ADME CHARACTERIZATION OF ESTERS OF ETHYLENEDIAMINE-N,N'-DI-2-(3-CYCLOHEXYL)PROPIONIC ACID ANALOGS WITH POTENTIAL CYTOTOXIC ACTIVITY

- [Biljana Tubić, Bojan Marković,  
Sandra Vladimirov, Vladimir Dobričić,  
Jelena Poljarević, Aleksandar Savić, Tibor Sabo](#) 106

PP 4

SINTEZA I BIOLOŠKA AKTIVNOST PROPIOFENONSKIH DERIVATA

SYNTHESIS AND BIOLOGICAL ACTIVITY OF PROPIONOPHENONE DERIVATIVES

- [Branka Ivković, Nemanja Turković,  
Bojan Marković, Zorica Vujić](#) 108

PP 5

IZOKUMARINSKI DERIVATI-SINTEZA I ANTIFUNGALNA AKTIVNOST

ISOCOUMARIN DERIVATIVES-SYNTHESIS AND ANTIFUNGAL ACTIVITY

- [Milena Simić](#) 110

PP 6

**PRIMENA PAMPA TEHNIKE I QSPR ANALIZE U PROCENI  
GASTROINTESTINALNE APSORPCIJE I DIZAJNIRANJU NOVIH  
BIOLOŠKI AKTIVNIH JEDINJENJA**

**APPLICATION OF PAMPA TECHNIQUE AND QSPR ANALYSIS IN THE  
EVALUATION OF GASTROINTESTINAL ABSORPTION AND DESIGN OF  
NEW BIOLOGICALLY ACTIVE COMPOUNDS**

- **Vladimir Dobričić, Jelena Savić, Biljana Tubić, Katarina Nikolić,  
Jasmina Brborić, Bojan Marković, Olivera Čudina**

**112**

PP 7

**IN VITRO ISPITIVANJE INHIBITORNOG POTENCIJALA SINTETISANIH  
β -HIDROksi-β-ARILALKANSKIH KISELINA KORIŠĆENJEM  
KOMERCIJALNOG COX KITA**

**IN VITRO ASSESSMENT OF THE INHIBITORY POTENTIAL OF  
SYNTHEZIZED β-HYDROXY-β-ARYLALKANOIC ACIDS USING  
COMMERCIAL COX KIT**

- **Jelena Savić, Jelena Kotur-Stevuljević,  
Sanda Dilber, Sote Vladimirov, Jasmina Brborić**

**114**

PP 8

**EUROPEAN ASSOCIATION OF HOSPITAL PHARMACY (EAHP)  
COMPETENCY FRAMEWORK FOR HOSPITAL PHARMACY**

- **Petr Horak**

**116**

PP 9

**SAVREMENI PRISTUP OPTIMIZACIJI ANTIBIOTSKE TERAPIJE NA  
OSNOVU IZMERENIH KONCENTRACIJA**

**A MODERN APPROACH TO OPTIMIZING ANTIBIOTIC THERAPY  
BASED ON MEASURED CONCENTRATIONS**

- **Katarina Vučićević**

**117**

PP 10

**INTERVENCIJE KLINIČKOG FARMACEUTA NA ODELJENJU  
HEPATOLOGIJE**

**CLINICAL PHARMACIST'S INTERVENTIONS ON A HEPATOLOGY  
WARD**

- **Milica Ćulafić**

**119**

PP 11

**ZDRAVSTVENA ANALIZA VRSTE I ISHODA GREŠKE - *HEALTHCARE FAILURE MODE AND EFFECT ANALYSIS (HFMEA)* PRIMENJENA NA PROCES SUPSTITUCIJE ANTIBIOTSKE TERAPIJE TOKOM NESTAŠICE LEKOVA**

**HEALTHCARE FAILURE MODE AND EFFECT ANALYSIS (HFMEA)  
APPLIED TO ANTIBIOTIC SUBSTITUTION IN MEDICINE SHORTAGES**

- **Nenad Miljković, Karyofyllis Tsiakitzis,  
Cristina Garcia Yubero, Branislava Miljković**

**121**

PP 12

**MIKROBIOTA CREVA, ZNAČAJ ZA ETIOPATOGENEZU I TERAPIJU AUTOIMUNSKIH BOLESTI**

**THE ROLE OF GUT MICROBIOTA IN ETIOPATHOGENESIS AND THERAPY OF AUTOIMMUNE DISEASES**

- **Dorđe Miljković**

**123**

PP 13

**REGULATORNE T ĆELIJE - NOVI PRISTUP U LEČENJU AUTOIMUNSKIH BOLESTI**

**T REGULATORY CELLS - NEW APPROACH IN THE TREATMENT OF AUTOIMMUNITY**

- **Ivana Stojanović**

**125**

PP 14

**ADRENERGIČKI LEKOVI - KANDIDATI ZA NOVE NEKONVENCIONALNE IMUNOMODULATORNE LEKOVE?**

**ADRENERGIC DRUGS – CANDIDATES AS NOVEL NON-CONVENTIONAL IMMUNOMODULATORY DRUGS?**

- **Gordana Leposavić**

**127**

PP 15

**SAVREMENA TERAPIJA MULTIPLE SKLEROZE - OD IMUNOMODULACIJE DO SELEKTIVNE IMUNE REKONSTITUCIJE**

**CURRENT THERAPY OF MULTIPLE SCLEROSIS - FROM IMMUNOMODULATION TO IMMUNE RECONSTITUTION**

- **Dragana Obradović**

**129**

PP 16

**SAVREMENI PRISTUP U LEČENJU REUMATOIDNOG ARTRITISA**

**ADVANCES IN THE TREATMENT OF RHEUMATOID ARTHRITIS**

- **Mirjana Šefik Bukilica**

**131**

PP 17

**CARDIOPROTECTION DURING CANCER CHEMOTHERAPY WITH THE**

**USE OF NATURAL ANTIOXIDANTS: REVIEW OF LITERATURE AND**

**RESULTS OF OWN STUDIES**

- **Jolanta Lukowicz, Grażyna Peszyńska-Sularz,  
Anita Piasek, Stefan Popadiuk, Agata Kot-Wasik,  
Monika Janicka, Jacek Namieśnik, Włodzimierz Grajek,  
Agnieszka Bartoszek**

**133**

PP 18

**VITAMIN D I RIZIK ZA NASTANAK MALIGNIH BOLESTI**

**VITAMIN D AND RISK FOR CANCER DEVELOPMENT**

- **Aleksandra Zeljković**

**135**

PP 19

**PRIMENA TUMORSKIH MARKERA U KLINIČKOJ PRAKSI I  
PERSONALIZOVANOJ MEDICINI**

**APPLICATION OF TUMOR MARKERS IN CLINICAL PRACTICE AND  
PERSONALIZED MEDICINE**

- **Svetlana Ignjatović**

**137**

PP 20

**ENERGETSKI BALANS I ULOGA ADIPOCITOKINA U PATOGENEZI  
MALIGNIH BOLESTI**

**ENERGY BALANCE AND ADIPOCYTOKINES IN CANCER  
PATHOGENESIS**

- **Aleksandra Stefanović**

**139**

PP 21

**KADMIJUM KAO FAKTOR RIZIKA ZA RAZVOJ KARCINOMA PANKREASA: PODACI IZ STUDIJE NA LJUDIMA, EKSPERIMENTALNIM ŽIVOTINJAMA I ĆELIJSKIM KULTURAMA**

**CADMIUM AS A RISK FACTOR FOR PANCREATIC CANCER DEVELOPMENT: HUMAN, ANIMAL AND IN VITRO DATA**

- Aleksandra Buha Đorđević, Vesna Matović, Novica Boričić, Dejan Radenković, Vladimir Đorđević, David Wallace

141

PP 22

**OLIVE BIOACTIVE COMPOUNDS: CHEMISTRY AND BIOLOGY**

- Apostolis Angelis, Leandros Skaltsounis

143

PP 23

**HERBA CITRALNOG HEMOTIPA PANONSKOG TIMIJANA KAO POTENCIJALNO NOVA BILJNA LEKOVITA SIROVINA**

**THE HERB OF PANNONIAN THYME CITRAL CHEMOTYPE AS POTENTIALLY NEW HERBAL RAW MATERIAL WITH MEDICINAL PROPERTIES**

- Zoran Maksimović

144

PP 24

**PROCENA FARMAKOLOŠKE AKTIVNOSTI ODABRANIH VRSTA FAMILIJE ERICACEAE**

**PHARMACOLOGICAL SCREENING OF SELECTED SPECIES FROM ERICACEAE FAMILY**

- Dragana Pavlović

146

PP 25

**NOVIJE INFORMACIJE O LEKOVITOM POTENCIJALU VRSTA RODA HYPERICUM**

**UPDATES ON THERAPEUTIC POTENTIAL OF HYPERICUM SPECIES**

- Nebojša Kladar, Neda Gavarić, Biljana Božin

148

PP 26

**EFEKTI METANOLNIH EKSTRAKATA DVE BILJNE VRSTE IZ FLORE SRBIJE NA ISHEMIJSKO-REPERFUZIONU POVREDU IZOLOVANOG SRCA PACOVA: UTICAJ OKSIDACIONOG STRESA**

**EFFECTS OF OF METHANOL EXTRACTS OF TWO PLANT SPECIES FROM THE FLORA OF SERBIA ON ISCHEMIC/REPERFUSION INJURY OF ISOLATED RAT HEART: ROLE OF OXIDATIVE STRESS**

- Nevena Jeremić, Jovana Bradić, Vladimir Živković, Ivan Srejović,  
Jovana Jeremić, Tamara Nikolić-Turnić, Vladimir Jakovljević 150

PP 27

**OPTIMIZACIJA EKSTRAKCIJE PLODA ARONIJE, ARONIA MELANOCARPA (MICHX.) ELLIOTT, MIKROINKAPSULACIJA EKSTARKTA I ISPITIVANJE BIOLOŠKIH AKTIVNOSTI EKSTRAKTA**

**OPTIMIZATION OF CHOKEBERRY EXTRACTION, ARONIA MELANOCARPA (MICHX.) ELLIOTT, EXTRACT MICROENCAPSULATION AND BIOLOGICAL ACTIVITIES**

- Nada Ćujić, Katarina Šavikin, Gordana Zdunić,  
Branko Bugarski, Nevena Mihailović-Stanojević, Svetlana Ibrić 152

PP 28

**GENERIČKI LEKOVI OD PODNOŠENJA ZAHTEVA DO ODOBRENJA SAŽETKA KARAKATERISTIKA LEKA**

**GENERIC MEDICINAL PRODUCTS FROM APPLICATION TO FINAL SUMMARY OF PRODUCT CHARACTERISTICS**

- Branka Brzaković 154

PP 29

**BIOEQUIVALENCE REQUIREMENTS FOR LOCALLY ACTING DOSAGE FORMS**

- Alfredo Garcia-Arieta 156

PP 30	POTVRDA TERAPIJSKE EKVIVALENTNOSTI ORALNIH INHALACIONIH LEKOVA - REGULATORNI ASPEKTI	
	REGULATORY FRAMEWORK FOR DEMONSTRATION OF THERAPEUTIC EQUIVALENCE OF ORALLY INHALED PRODUCTS	
- Zorica Pejčić		157
PP 31	KADA (NI)JE MOGUĆA SUPSTITUCIJA GENERIČKIM LEKOM	
	WHEN GENERIC SUBSTITUTION IS (NOT) APPROPRIATE	
- Marija Jovanović		159
PP 32	AKUTNA TROVANJA ILEGALNIM PSIHOAKTIVnim SUPSTANCAMA - ISKUSTVA NACIONALNOG CENTRA ZA KONTROLU TROVANJA	
	ACUTE POISONING WITH ILLICIT PSYCHOACTIVE SUBSTANCES - EXPERIENCE OF THE NATIONAL POISON CONTROL CENTRE	
- Jasmina Jović-Stošić, Tomislav Režić, Nataša Perković-Vukčević, Slavica Vučinić, Gordana Brajković, Snežana Đorđević, Mirjana Đukić		161
PP 33	THE ANALYSIS OF PSYCHOACTIVE SUBSTANCES: CHALLENGES RELATED TO BIOLOGICAL SAMPLES AND ANALYTICAL TOOLS	
- Goran Mitulović		163
PP 34	PREGLED SITUACIJE NA TRŽIŠTU DROGA U SRBIJI I PREDIZIRANJA SUPSTANCAMA ZLOUPOTREBE LEČENIH U NACIONALNOM CENTRU ZA KONTROLU TROVANJA VMA	
	AN OVERVIEW OF THE DRUG MARKET AND SUBSTANCES OF ABUSE OVERDOSE TREATED IN THE NATIONAL POISON CONTROL CENTER MMA	
- Slavica Vučinić, Jasmina Jović-Stošić, Dragana Đorđević, Tomislav Režić, Snežana Đorđević, Vesna Kilibarda		164

PP 35	
<b>KURIKULARNE I EKSTRAKURIKULARNE AKTIVNOSTI U DOPRINOSU RAZUMEVANJU ZLOUPOTREBE PSIHOAKTIVNIH SUPSTANCI</b>	
<b>CURRICULAR AND EXTRA-CURRICULAR ACTIVITIES TO DEVELOP COMPREHENSION ON ABUSE OF PSYCHOACTIVE SUBSTANCES</b>	
- <a href="#">Mirjana Đukić</a>	<a href="#">166</a>
PP 36	
<b>THE EFFECTS OF NATURAL AND SYNTHETIC ENVIRONMENTAL POLLUTANTS ON HUMAN HEALTH: SOME CASE STUDIES</b>	
- <a href="#">Emanuela Testai</a>	<a href="#">168</a>
PP 37	
<b>METIL-ŽIVA U NAŠEM OKRUŽENJU: KLJUČNE ČINJENICE ZA SIGURNU BUDUĆNOST</b>	
<b>METHYLMERCURY IN OUR ENVIRONMENT: KEY FACTS FOR A SAFE FUTURE</b>	
- <a href="#">Danijela Đukić-Ćosić</a>	<a href="#">169</a>
PP 38	
<b>DOKAZI TOKSIČNOSTI USPORIVAČA GORENJA - POLIBROMOVANI DIFENILETRI</b>	
<b>EVIDENCE OF FLAME RETARDANTS TOXICITY - POLYBROMINATED DIPHENYL ETHERS</b>	
- <a href="#">Marijana Ćurčić</a>	<a href="#">171</a>
PP 39	
<b>OPASNE HEMIKALIJE U PROIZVODIMA ŠIROKE POTROŠNJE I REGULATORNI ASPEKT KAO MEHANIZAM KONTROLE U EU I REPUBLICI SRBIJI</b>	
<b>HAZARDOUS CHEMICALS IN ARTICLES FOR EVERYDAY USE AND REGULATORY ASPECT AS A CONTROL MECHANISM IN EU AND SERBIA</b>	
- <a href="#">Jasminka Randelović, Jelena Milić, Valentina Mart, Lazarija Šojić</a>	<a href="#">173</a>

PP 40

**PRIMENA 3D ŠTAMPE U FARMACIJI - IZAZOVI I PERSPEKTIVE**

**3D PRINTING FOR PHARMACEUTICAL APPLICATIONS – CHALLENGES AND PROSPECTS**

- **Svetlana Ibrić**

**175**

PP 41

**SAVREMENI PRISTUP ODABIRU FORMULACIJE I EKSCIPIJENASA**

**MODERN APPROACH TO FORMULATION AND EXCIPIENTS SELECTION**

- **Ružica Kolaković**

**177**

PP 42

**ULOGA KLINIČKOG FARMACEUTA U RAZVOJU FORMULACIJA LEKOVA ZA SPROVOĐENJE KLINIČKIH ISPITIVANJA**

**CLINICAL PHARMACIST'S ROLE IN CLINICAL TRIALS INVESTIGATIONAL DRUG DEVELOPMENT**

- **Marija Tubić - Grozdanis**

**179**

PP 43

**INKAPSULACIJA ODABRANIH SUPERKRITIČNIH EKSTRAKATA LEKOVITOG BILJA U LIPOSOME METODOM HOMOGENIZACIJE POD VISOKIM PRITISKOM**

**INCAPACULATION OF SELECTED MEDICINAL HERB'S SUPERCritical EXTRACTS IN LIPOSOMES USING THE HIGH PRESSURE HOMOGENIZATION METHOD**

- **Ivana Arsić, Vanja Tadić, Milica Stanković, Vesna Savić**

**181**

PP 44

**ANTIEPILEPTICI U SVETLU NOVIH INDIKACIJA**

**ANTIEPILEPTICS IN LIGHT OF NEW INDICATIONS**

- **Radica Stepanović-Petrović**

**183**

PP 45	
<b>BENZODIAZEPINES ARE ALL ALIKE - EXCEPT WHEN THE OPPOSITE COMES TRUE</b>	
- Margot Ernst	185
PP 46	
<b>ALFA 1, 2, 3, 4, 5, 6 GABA A RECEPTORI: ŠTO VIŠE TO BOLJE KAO CILJ ZA NOVE LEKOVE?</b>	
<b>ALPHA 1, 2, 3, 4, 5, 6 GABA A RECEPTORS: THE HIGHER THE BETTER AS A TARGET FOR NOVEL MEDICINES?</b>	
- Miroslav Savić	186
PP 47	
<b>KOMBINACIJE ANALGETIKA U SAVREMENOM LEČENJU BOLA</b>	
<b>COMBINATIONS OF ANALGESICS IN THE CONTEMPORARY TREATMENT OF PAIN</b>	
- Maja Tomić	188
PP 48	
<b>IS EDUCATING PHARMACISTS TO BE COMPETENT ENOUGH FOR THE FUTURE OF THE PROFESSION?</b>	
- Martin Henman	190
PP 49	
<b>STRUČNO OSPOSOBLJAVANJE I PROFESIONALNI RAZVOJ FARMACEUTA - AKADEMSKA PERSPEKTIVA</b>	
<b>QUALIFICATION AND PROFESSIONAL DEVELOPMENT OF PHARMACIST - ACADEMIC PERSPECTIVE</b>	
- Ljiljana Tasić	191
PP 50	
<b>STRUČNO OSPOSOBLJAVANJE I PROFESIONALNI RAZVOJ FARMACEUTA - PERSPEKTIVA APOTEKARSKE PRAKSE</b>	
<b>QUALIFICATION AND PROFESSIONAL DEVELOPMENT OF PHARMACIST - PHARMACY PRACTICE PERSPECTIVE</b>	
- Svetlana Stojkov	193

PP 51	
<b>WHY CLINICAL COMMUNICATION SKILLS REALLY MATTER? SOME EXAMPLES OF EFFECTIVE TEACHING AND LEARNING METHODS</b>	
- Afonso Miguel Cavaco	195
PP 52	
<b>VALUE FRAMEWORKS AND DECISION MAKING AROUND THE GLOBE</b>	
- Wija Oortwijn, Rob Baltussen, Maarten Janssen	196
PP 53	
<b>VREDNOST INOVACIJE PRILIKOM DONOŠENJA ODLUKA U ZDRAVSTVU</b>	
<b>THE VALUE OF INNOVATION IN HEALTH CARE DECISION MAKING</b>	
- Tanja Novaković	197
PP 54	
<b>ZAŠTO SISTEMATIČNI PREGLEDI LITERATURE?</b>	
<b>WHY SYSTEMATIC REVIEWS?</b>	
- Mark Parker	199
PP 55	
<b>INHIBITORI KOTRANSPORTERA ZA NATRIJUM I GLUKOZU TIPA 2 KOD OBOLELIH OD DIJABETES MELITUSA TIPA 2 I SRČANE INSUFICIJENCIJE: KLINIČKI POGLED NA TERAPIJU KOJA MOŽE DA SNIZI MORBIDITET I MORTALITET</b>	
<b>SODIUM GLUCOSE CONTRANSPORTER-2 INHIBITORS IN TYPE-2 DIABETES AND HEART FAILURE: THE CLINICAL STANDPOINT ON TREATMENT THAT CAN REDUCE MORBIDITY AND MORTALITY</b>	
- Marija Polovina	201
PP 56	
<b>NOVEL ANTIDIABETIC AGENTS AND CARDIOVASCULAR RISK</b>	
- Manfredi Rizzo	203

PP 57

**KONTINUIRANI SKOR ZA METABOLIČKI SINDROM U POPULACIJI  
DECE I ADOLESCENATA**

**CONTINUOUS METABOLIC SYNDROME SCORE FOR USE IN PEDIATRIC  
POPULATION**

- **Rade Vuković, Ivan Soldatović, Tatjana Milenković,  
Katarina Mitrović, Slađana Todorović, Ljiljana Plavšić**

**204**

PP 58

**OPSTRUKTIVNA APNEJA U SNU I KARDIOMETABOLIČKI RIZIK**

**OBSTRUCTIVE SLEEP APNEA AND CARDIOMETABOLIC RISK**

- **Jelena Vekić**

**206**

PP 59

**GENETIČKA ISPITIVANJA U METABOLIČKOM SINDROMU**

**GENETIC TESTING FOR METABOLIC SYNDROME**

- **Ana Ninić**

**208**

PP 60

**SKOR DISLIPIDEMIJE, INFILAMACIJE I OKSIDATIVNOG STRESA U  
PROCENI KARDIOVASKULARNOG RIZIKA**

**DYSLIPIDEMIA, INFLAMMATION AND OXIDATIVE STRESS SCORE IN  
CARDIOVASCULAR RISK ESTIMATION**

- **Jelena Kotur-Stevuljević, Nataša Bogavac-Stanojević,  
Jelena Vekić, Vesna Kalimanovska-Spasojević,  
Zorana Jelić-Ivanović, Slavica Spasić**

**210**

PP 61

**HRONIČNA TERAPIJA – OČEKIVANJA I ZABRINUTOST NAŠIH  
PACIJENATA**

**CHRONIC THERAPY – EXPECTATIONS AND CONCERNS OF OUR  
PATIENTS**

- **Branislava Miljković**

**212**

PP 62

**FARMACEUTI U SRBIJI IDENTIFIKUJU TERAPIJSKE PROBLEME KOD STARIJIH PACIJENATA - KOJE, KAKO, KOLIKO?**

**PHARMACISTS IN SERBIA IDENTIFY DRUG-RELATED PROBLEMS IN ELDERLY PATIENTS - WHICH, HOW, HOW MANY?**

- [Sandra Vezmar Kovačević](#)

**214**

PP 63

**PRIMENA KONCEPTA FARMACEUTSKE ZDRAVSTVENE ZAŠTITE KOD PACIJENATA SA ASTMOM I HOBP – MODEL PRIMARNE ZDRAVSTVENE ZAŠTITE**

**PHARMACEUTICAL CARE MODEL IN THE COMMUNITY PHARMACY SETTINGS – FOCUS ON ASTHMA AND COPD PATIENTS**

- [Milena Kovačević](#)

**216**

PP 64

**STOPP/START KRITERIJUMI ZA OPTIMIZACIJU TERAPIJE U GERIJATRIJSKOJ POPULACIJI**

**STOPP/START CRITERIA FOR OPTIMIZATION OF PHARMACOTHERAPY IN ELDERLY**

- [Aleksandra Catić-Đorđević, Nikola Stefanović,  
Radmila Veličković-Radovanović](#)

**218**

PP 65

**REGULATIVA O DODACIMA ISHRANI**

**REGULATION ON FOOD SUPPLEMENTS**

- [Ivan Stanković](#)

**220**

PP 66

**KORISTI SUPLEMENTACIJE U PROMOCIJI ZDRAVLJA**

**DIETARY SUPPLEMENT IN HEALTH PROMOTION**

- [Brižita Đorđević, Nevena Ivanović, Ivana Baralić](#)

**222**

PP 67  
**ZDRAVSTVENI RIZICI UPOTREBE DIJETETSKIH SUPLEMENATA**  
**DIETARY SUPPLEMENTS - HEALTH RISK**

- **Zorica Bulat** 224

PP 68  
**ANALIZA DIJETETSKIH SUPLEMENATA KOJE KORISTE SPORTISTI U SRBIJI**  
**ANALYSIS OF DIETARY SUPPLEMENTS USED BY SERBIAN ATHLETES**

- **Nenad Dikić, Marija Andelković, Milica Vukašinović Vesić, Brižita Đorđević** 226

PP 69  
**APOTHECARY PROFESSION AND PHARMACEUTICAL ACTIVITIES IN THE HEALTH CARE SERVICE AT THE END OF THE FIRST WORLD WAR**

- **Adriana Elena Taerel** 228

PP 70  
**JAČANJE PROFESIONALIZMA U APOTEKARSKOJ PRAKSI: ČEMU NAS UČE APOTEKARSKE ZAKLETVE OD NAJSTARIJIH DO SAVREMENIH**  
**REINFORCING PROFESSIONALISM IN APOTHECARY PRACTICE: WHAT COULD WE LEARN FROM THE APOTHECARIES' OATHS FROM THE PAST TO THE MOST CONTEMPORARY**

- **Dušanka Krajnović** 229

PP 71  
**QUALITY INDICATORS OF PHARMACEUTICAL CARE SERVICES**

- **Mitja Kos** 231

PP 72  
**ANALIZA FARMACEUTSKIH USLUGA U EVROPI I SRBIJI - MODALITETI RAZVOJA U SVETLU NOVIH TEHNOLOGIJA**  
**ANALYSIS OF PHARMACEUTICAL SERVICES PROVIDED IN COMMUNITY PHARMACIES IN EUROPE AND SERBIA - MODALITIES FOR FUTURE DEVELOPMENT IN THE LIGHT OF NEW TECHNOLOGIES**

- **Ivana Tadić** 232

PP 73

**UNAPREĐENJE ZDRAVSTVENE ZAŠTITE TRUDNICA I DOJILJA -  
ULOGA FARMACEUTA I DOPRINOS FARMACEUTSKIH USLUGA**

**PREGNANT AND BREASTFEEDING WOMEN HEALTHCARE  
IMPROVEMENT - THE ROLE OF PHARMACISTS**

- **Marina Odalović**

**234**

PP 74

**KOLIKO KOŠTA FARMACEUTSKA USLUGA?**

**HOW MUCH DOES THE PHARMACEUTICAL SERVICE COST?**

- **Dragana Lakić**

**236**

PP 75

**PATIENT CENTRIC DOSAGE FORM DESIGN**

- **Andreas Zimmer, Sven Stegemann**

**238**

PP 76

**INCREASED PATIENT SAFETY BY READY-TO-USE/READY-TO-  
ADMINISTER PARENTERALS PREPARED IN HOSPITAL PHARMACIES**

- **Irene Krämer**

**239**

PP 77

**FORMULACIJA FARMACEUTSKIH OBLIKA LEKOVA ZA PRIMENU U  
PEDIJATRIJSKOJ POPULACIJI - ASPEKTI  
PRIHVATLJIVOST/ADHERENCA**

**FORMULATION OF PAEDIATRIC DOSAGE FORMS - ACCEPTABILITY  
ISSUES/COMPLIANCE**

- **Jela Milić, Sandra Cvijić, Ivana Pantelić**

**240**

PP 78

**CANCER IMMUNOTHERAPY: WHERE DID ITS PRECISION  
COME FROM AND WHERE WILL IT GO?**

- **Farzin Farzaneh**

**242**

PP 79

**PRECIZNA MEDICINA U ONKOLOŠKOJ PRAKSI:  
PROCENA KORISTI I RIZIKA**

**PRECISION MEDICINE IN ONCOLOGY PRACTICE:  
BENEFIT-RISK ASSESSMENT**

- [Ivana Božović-Spasovjević](#)

[243](#)

PP 80

**MOLECULAR PATHWAYS THAT OPERATE IN MLL-ASSOCIATED  
LEUKEMIA TO OVERCOME RESISTANCE TO ANTICANCER DRUGS**

- [Boban Stanojević](#)

[245](#)

PP 81

**PRECIZNE ANTIKancerske TERAPIJE: KAKO FARMACEUTSKA  
TEHNOLOGIJA DAJE DOPRINOS?**

**PRECISE ANTI-CANCER THERAPIES: HOW DOES PHARMACEUTICAL  
TECHNOLOGY CONTRIBUTE TO THEM?**

- [Snežana Savić](#)

[246](#)

PP 82

**SAVREMENI PRISTUPI U KONTROLI KVALITETA  
BIOLOŠKIH LEKOVA**

**CONTEMPORARY APPROACHES IN BIOLOGICAL DRUGS QUALITY  
CONTROL**

- [Borut Štrukelj](#)

[248](#)

PP 83

**MICRO-PHOTOGRAMMETRY AS A NOVEL TOOL FOR  
CHARACTERISATION OF DISSOLUTION BEHAVIOUR OF  
PHARMACEUTICAL DOSAGE FORMS**

- [Alessandra D'Angelo, Mike Reading, Milan Antonijević](#)

[250](#)

PP 84

**KONCEPTUALNI MODEL ZA UNAPREĐENJE SISTEMATSKE  
KONTROLE**

**CONCEPTUAL MODEL FOR THE IMPROVEMENT OF MARKET  
SURVEILLANCE PROCESS**

- **Gordana Pejović**

**251**

PP 85

**ANALITIKA POLARNIH SUPSTANCI PRIMENOM METODE TEĆNE  
HROMATOGRAFIJE HIDROFILNIH INTERAKCIJA**

**HYDROPHILIC INTERACTION LIQUID CHROMATOGRAPHY (HILIC) AS  
A VALUABLE ALTERNATIVE FOR REVERSED-PHASE LIQUID  
CHROMATOGRAPHY (RP-LC) IN THE ANALYSIS OF POLAR  
COMPOUNDS**

- **Biljana Jančić Stojanović**

**253**

PP 86

**MOGUĆNOSTI PRIMENE EKOLOŠKI PRIHVATLJIVIH  
HROMATOGRAFSKIH METODA U KONTROLI LEKOVA**

**PROSPECTS OF ECOLOGICALLY ACCEPTABLE CHROMATOGRAPHIC  
METHODS IN DRUG CONTROL**

- **Ana Protić, Nevena Maljurić, Biljana Otašević, Mira Zečević**

**255**

## **HOMOLOGY MODELING OF HUMAN HISTONE DEACETYLASE 10 AND DESIGN OF POTENTIAL SELECTIVE INHIBITORS**

**Abdullahi Ibrahim Uba, Kemal Yelekçi**

Department of Bioinformatics and Genetics, Faculty of Engineering and Natural Sciences, Kadir Has University, Istanbul (Turkey)

Histone deacetylases (HDACs) are implicated in the pathology of various cancers and their pharmacological blockade has proven to be promising in reversing the malignant phenotypes. However, lack of crystal structures of some of the human HDAC isoforms (e.g., HDAC10) hinders the design of isoform-selective inhibitor. Here, the recently-solved X-ray crystal structure of *Danio rerio* (zebrafish) HDAC10 (PDB ID; 5TD7, release date 24-05-2017) was retrieved from the Protein Data Bank (PDB) and used as a template structure to model the 3D structure of human HDAC10. The overall quality of the best model (M0017) was assessed by computing its z-score a measure of the deviation of the total energy of the structure with respect to an energy distribution derived from random conformations, and by docking of known HDAC10 inhibitors to its catalytic cavity. Furthermore, to identify potential HDAC10-selective inhibitors ligand-based virtual screening was carried out against ZINC database. The free modeled structure of HDAC10, and its complexes with quisinostat and the highest-ranked compound, ZINC19749069 were submitted to molecular dynamics simulation. Comparative analysis of root-mean-squared deviation (RMSD), root-mean-squared fluctuation (RMSF), radius of gyration (Rg), and potential energy of these systems showed that HDAC10-ZINC19749069 complex remained the most stable over time. Thus, M0017 could be potentially used for structure-based inhibitor against HDAC10, and ZINC19749069 may provide a scaffold for further optimization.

## **INHIBITORI EPIGENETSKIH ENZIMA IZ PRIRODNIH IZVORA**

**Slavica Erić**

Katedra za farmaceutsku hemiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Epigenetske promene utiču na ekspresiju gena na nivou transkripcije putem gornje i donje regulacije, ili kompletним utišavanjem gena. Poremećena regulacija epigenetskih događaja može biti uzrok patoloških promena koje dovode do razvoja kardiovaskularnih, neuroloških, metaboličkih i kancerskih oboljenja. Poznavanje specifičnih epigenetskih promena karakterističnih za ove tipove bolesti je značajno za razvoj specifičnih epigenetskih lekova. S obzirom da pri razvoju ovih bolesti dolazi do poremećaja enzima koji učestvuju u epigenetskim reakcijama, oni predstavljaju pogodan target za dizajniranje lekova.

Inhibitori dve klase ovih enzima našli su primenu u kliničkoj praksi: inhibitori DNK metiltransferaze i histon deacetilaze. Pokazalo se da su druge klase epigenetskih enzima takođe veoma značajne za razvoj bolesti i trenutno se koriste kao targeti u otkrivanju novih lekova.

Jedan od aspekata u otkriću novih inhibitora epigenetskih enzima uključuje istraživanje aktivnih komponenata iz prirodnih izvora. Ove komponente mogu se primeniti za dalju karakterizaciju enzima koji modifikuju hromatin. U mnogim slučajevima predstavljaju samo prve agense koji su identifikovani kao inhibitori ili modulatori određenog enzima i dalje služe kao vodeće molekule za nove komponente sa osobinama lekova, ili mogu biti lekovi kandidati. S obzirom da mnogi prirodni proizvodi nisu dovoljno istraženi, kao i da mehanizmi dejstva mnogih agenasa nisu dovoljno razjašnjeni, prirodni izvori predstavljaju veliki potencijal u otkrivanju novih lekova na epigenetskom nivou.

U ovom radu prikazani su prirodni proizvodi koji deluju na epigenetske mehanizme inhibicijom enzima uključenih u patogenezu različitih bolesti. Mnogi od njih predstavljaju samo polaznu tačku u razvoju novih inhibitora epigenetskih enzima koji se mogu primeniti u hemoprevenciji i hemoterapiji različitih bolesti.

# **INHIBITORS OF EPIGENETIC ENZYMES FROM NATURAL SOURCES**

**Slavica Erić**

Department of Pharmaceutical Chemistry, University of Belgrade - Faculty of Pharmacy (Serbia)

Epigenetic changes alter gene expression at the level of transcription by upregulating, downregulating, or silencing genes completely. Dysregulation of epigenetic events can be pathological, leading to cardiovascular disease, neurological disorders, metabolic disorders, and cancer development. Knowledge of the specific epigenetic changes associated with these types of diseases facilitates the development of specific epigenetic drugs. Many of the enzymes that mediate these epigenetic reactions are dysregulated in human diseases, so could be suitable target for drug design.

Inhibitors against two classes of these enzymes have been approved for use in patients: DNA methyltransferase (DNMT) inhibitors and histone deacetylase (HDCA) inhibitors. Other classes of epigenetic enzymes have been demonstrated to have strong disease association and are currently being targeted for use in drug discovery.

One of the aspect in drug discovery of inhibitors of epigenetic enzymes is search for active compounds from natural sources. Those compounds could be important tools to further characterize the chromatin-modifying enzymes. In many cases they are the first agents identified as inhibitors or modulators of the particular enzyme and thereby serve as lead structures for new drug-like compounds or may be drug candidates themselves. Since many natural sources are not examined yet, as well as the mechanism of action of many of them is not still elucidated, natural sources represent a great potential in discovering new drugs on epigenetic level.

In this review, natural products that target epigenetic mechanisms by the inhibition of enzymes involved in patghogenesis of various diseases are presented. Many of them are just strating point in developing new inhibitors of epigenetic enzymes that might be used for chemoprevention and chemotherapy of various diseases.

## FIZIČKO-HEMIJSKA I ADME KARAKTERIZACIJA ANALOGA ESTARA ETILENDIAMIN-N,N'-DI-2-(3-CIKLOHEKSIL) PROPANSKE KISELINE SA POTENCIJALNIM CITOTOKSIČNIM DEJSTVOM

Biljana Tubić<sup>1</sup>, Bojan Marković<sup>2</sup>, Sandra Vladimirov<sup>2</sup>,  
Vladimir Dobričić<sup>2</sup>, Jelena Poljarević<sup>3</sup>, Aleksandar Savić<sup>3</sup>, Tibor Sabo<sup>3</sup>

<sup>1</sup>Agencija za lekove i medicinska sredstva Bosne i Hercegovine (Bosna i Hercegovina), <sup>2</sup>Katedra za farmaceutsku hemiju, Univerzitet u Beogradu - Farmaceutski fakultet, <sup>3</sup>Univerzitet u Beogradu - Hemijski fakultet (Srbija)

Estri (S,S)-1,2-etandiamin-N,N'-di-2-(3-cikloheksil)propanske kiseline (EDCP) i (S,S)-1,3-propandiamin-N,N'-di-2-(3-cikloheksil)propanske kiseline (PDCP) dizajnirani su kao ligandi za Pt(IV) komplekse. U in vitro ispitivanjima utvrđena je značajna citotoksična aktivnost navedenih kompleksa, ali i liganada u nevezanom obliku. Cilj rada je da se za navedene ligande izvrši in vitro i in silico biofarmaceutska karakterizacija.

Razvijena je i validirana UHPLC-MS/MS metoda za ispitivanje derivata EDCP i PDCP. Metoda je primenjena za određivanje koncentracija analita u toku ispitivanja rastvorljivosti, lipofilnosti i permeabilnosti. Primenom računarskog programa Metabolizer izvršeno je predviđanje potencijalnih metabolita. Predviđanje apsorpcije, distribucije, metabolizma, eliminacije i toksičnosti za sve ispitivane supstance i potencijalne metabolite izvršeno je primenom programa ADME(T) predictor. Za predviđanje mehanizma dejstva ispitivanih supstanci i njihove interakcije sa ciljnim mestima primjenjen je molekulski docking.

Kiseline (EDCP i PDCP) se dobro rastvaraju u izrazito kiseloj sredini i u izrazito baznoj sredini, dok je rastvorljivost estara najveća u izrazito kiseloj sredini. Ispitana je lipofilnost 14 derivata određivanjem LogD<sub>7,4</sub> koeficijenta primenom shake-flask metode, kao i određivanjem retencionog faktora (logk<sub>r</sub>), retencionog vremena (t<sub>R</sub>) i indeksa hidrofobnosti (CHI i φ<sub>0</sub>) primenom UHPLC-MS/MS metode. Postavljen je matematički model za predviđanje lipofilnosti za novosintetisane derivate ili potencijalne metabolite, a primenom leave-one-out validacione metode ocenjena je i pokazana dobra prediktivna moć predloženog matematičkog modela ( $Q^2=0,89$ ). Primenom paralelnog testa za ispitivanje permeabilnosti na veštačkim membranama (PAMPA) pokazano je da se estri značajno zadržavaju u membrani (30,09-99,89%), dok u niskom procentu prolaze kroz membranu (0,07-22,00%). In silico ispitivanja pokazuju da se antiproliferativno dejstvo ostvaruje putem više mehanizama i da su aktivne kiseline i potencijalni metaboliti (laktam-karboksilat i laktam alkil estar), dok su estri verovatno prodrug supstance. In vitro i in silico ispitivanja pokazuju da su estri EDCP i PDCP prodrug supstance sa povoljnog bioraspoloživošću na ciljnim mestima za citotoksično delovanje.

# **PHYSICOCHEMICAL AND ADME CHARACTERIZATION OF ESTERS OF ETHYLENEDIAMINE-N,N'-DI-2-(3-CYCLOHEXYL)PROPIONIC ACID ANALOGS WITH POTENTIAL CYTOTOXIC ACTIVITY**

**Biljana Tubić<sup>1</sup>, Bojan Marković<sup>2</sup>, Sandra Vladimirov<sup>2</sup>,  
Vladimir Dobričić<sup>2</sup>, Jelena Poljarević<sup>3</sup>, Aleksandar Savić<sup>3</sup>, Tibor Sabo<sup>3</sup>**

<sup>1</sup>Agency for medicines and medical devices of Bosnia and Herzegovina (Bosnia and Herzegovina), <sup>2</sup>Department of Pharmaceutical Chemistry, University of Belgrade - Faculty of Pharmacy, <sup>3</sup>University of Belgrade - Faculty of Chemistry (Serbia)

Esters of (S,S)-1,2-ethanediamine-N,N'-di-2-(3-cyclohexyl)propanoic acid (EDCP) and (S,S)-1,3-propanediamine-N,N'-di-2-(3-cyclohexyl)propanoic acid (PDCP) have been designed as ligands for Pt(IV) complexes. Results of in vitro investigations showed significant cytotoxic activity of complexes, as well as of ligands alone. The aim of this study was in vitro and in silico biopharmaceutical characterization of these ligands.

Novel UHPLC-MS/MS method for quantitative analysis of EDCP and PDCP derivatives was developed and validated. This method was used for quantification of tested compounds in solubility, lipophilicity and membrane permeability experiments. Metabolizer software was used for the prediction of potential metabolites. ADME(T) predictor software was used for absorption, distribution, metabolism, elimination and toxicity predictions. Molecular docking study was used for investigation of interactions with receptors and for the prediction of mechanisms of cytotoxic activity.

Investigated acids (EDCP and PDCP) and their esters have good solubility in water at low pH values, while acids have good water solubility at high pH values also. Lipophilicity was evaluated on the basis of LogD<sub>7.4</sub> coefficients, determined by shake-flask method, and on the basis of UHPLC-MS/MS chromatography parameters ( $\log k$ ,  $t_R$ , CHI and  $\varphi_0$ ). Based on obtained results, mathematical model for lipophilicity prediction of newly synthesized derivatives or potential metabolites was developed and its predictive power was confirmed by the leave-one-out method ( $Q^2=0.89$ ). Results of PAMPA test showed that esters possess high membrane retention (30.09-99.89%) and low membrane permeability (0.07-22.00%). On the basis of in silico results, it can be assumed that antiproliferative effects of investigated substances are result of several mechanisms, acids and potential metabolites (lactam carboxylate and lactam alkyl esters) are active, whereas esters are most likely prodrugs.

Results of in vitro and in silico experiments showed that esters of EDCP and PDCP are prodrug substances with favourable bioavailability at the sites of cytotoxic action.

## SINTEZA I BIOLOŠKA AKTIVNOST PROPIOFENONSKIH DERIVATA

**Branka Ivković<sup>1</sup>, Nemanja Turković<sup>2</sup>, Bojan Marković<sup>1</sup>, Zorica Vujić<sup>1</sup>**

<sup>1</sup>Katedra za farmaceutsku hemiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija), <sup>2</sup>Agencija za ljekove i medicinska sredstva Crne Gore (Crna Gora)

Strukturno jednostavna i biološki aktivna propiofenonska struktura nalazi se u osnovi velikog broja jedinjenja koja su našla primenu u terapiji kardiovaskularnih, malignih, infektivnih oboljenja. Biološka aktivnost, ali mala selektivnost čini strukturu interesantnom kao nosioca različitih grupa koje će uticati na selektivnost u delovanju. In silico i in vitro ispitivanja (antimikrobnog, antiproliferativnog, vozodilatatornog i antiaritmiskog) koja su sprovedena na do sada sintetisanim propiofenonskim derivatima (PD) pokazala su da prisustvo različitih grupa na PD strukturi utiče kako na aktivnost, tako i na selektivnost u delovanju. Aktivnost sintetisanih PD u najvećoj meri se zasniva na interakcijama sa aminokiselinama koje se nalaze u osnovi jonskih kanala kao esencijalnih struktura za funkciju svake ćelije, ali i virusnih čestica. Sintetisani PD u osnovi sadrže hidroksietilaminsku grupu kao farmakoformnu grupu antivirotika iz klase inhibitora HIV proteaze. S tim u vezi, cilj daljih istraživanja je da se ispita antiviralne aktivnosti PD.

Sprovedena in silico studija omogućila je filtraciju potencijalnih antivirotika među do sada sintetisanim PD, kao i filtraciju polaznih molekula za sintezu novih PD koji bi bili uključeni u dalja in vitro ispitivanja.

## **SYNTHESIS AND BIOLOGICAL ACTIVITY OF PROPIONOPHENONE DERIVATIVES**

**Branka Ivković<sup>1</sup>, Nemanja Turković<sup>2</sup>, Bojan Marković<sup>1</sup>, Zorica Vujić<sup>1</sup>**

<sup>1</sup>Department of Pharmaceutical Chemistry, University of Belgrade - Faculty of Pharmacy (Serbia), <sup>2</sup>Agency for Medicines and Medical Devices of Montenegro (Montenegro)

Structurally simple and biologically active propylphenonic structure is found in the basis of a large number of compounds that have found use in the treatment of cardiovascular, malignant, infectious diseases. Biological activity, but low selectivity makes the structure interesting as a carrier of different groups that will affect selectivity in effects. In silico and in vitro studies (antimicrobial, antiproliferative, vasodilatatory and antiarrhythmic) that have been conducted on the so far synthesized PD have shown that the presence of different groups on the PD structure affects both activity and selectivity in action. The activity of synthesized PDs is mostly based on interactions with the amino acids found in the basis of ion channels as the essential structures for the function of each cell, but also of viral particles. Synthesized PD basically contains a hydroxyethylamine group as a pharmacophoric group of antiviriotics from the class of HIV protease inhibitors. In this regard, the goal of further research is to investigate antiviral activities of the PD.

The conducted in silico study enabled the filtration of potential antiviriotics among the previously synthesized PD, as well as filtration of the starting molecules for the synthesis of new PDs that would be included in further in vitro studies.

## **IZOKUMARINSKI DERIVATI-SINTEZA I ANTIFUNGALNA AKTIVNOST**

**Milena Simić**

Katedra za organsku hemiju, Univerzitet u Beogradu - Farmaceutski fakultet  
(Srbija)

Izokumarini predstavljaju zanimljivu klasu sekundarnih metabolita biljaka i nekih gljiva. Osnovu strukture ovih derivata čini  $1H$ -benzo[ $c$ ]piran-1-on. Izokumarinski derivati, prirodni i sintetski, poznati su po različitim biološkim aktivnostima, kao što su antibakterijska, antifungalna, citotoksična, antiviralna, insekticidna. Zahvaljujući ovoj činjenici, istraživanja u oblasti prirodnih i sintetskih izokumarina privlače pažnju medicinskih hemičara. Cilj ovog istraživanja bio je razvoj nove sintetske metodologije za dobijanje izokumarinskih derivata, sinteza i evaluacija njihove antifungalne aktivnosti. Polazeći od 3-bromizokumarina, primenom paladijumom-katalizovanih reakcija kuplovanja, dobijena je serija novih 3-supstituisanih izokumarinskih derivata.

Ispitivana je *in vitro* antifungalna aktivnost serije novosintetisanih izokumarina. Na osnovu preliminarnog ispitivanja disk-difuzionim testom, derivati koji su pokazali značajnu zonu inhibicije rasta *C. albicans* odabrani su za dalja testiranja. Na osnovu određivanja MIC vrednosti odabranih derivata, najveći antifungalni potencijal pokazali su 3-heteroaril izokumarini. Najaktivnije jedinjenje je pokazalo *in vitro* antifungalnu aktivnost jednaku vorikonazolu.

Razvijena je nova sintetska metodologija za dobijanje 3-supstituisanih izokumarinskih derivata. Ispitivana je antifungalna aktivnost odabranih sintetisanih jedinjenja. Najveću *in vitro* antifungalnu aktivnost prema *C. albicans* pokazali su 3-heteroaril izokumarini.

# **ISOCOUMARIN DERIVATIVES-SYNTHESIS AND ANTIFUNGAL ACTIVITY**

**Milena Simić**

Department of Organic Chemistry, University of Belgrade - Faculty of Pharmacy  
(Serbia)

Isocoumarins represent very interesting class of secondary metabolites of plants and some fungi. Structure of these derivatives contains 1H-benzo[c]pyrane-1-on core. Isocoumarin derivatives, either natural or synthetic, are known to have various biological activities such as antibacterial, antifungal, cytotoxic, antiviral, insecticide. Because of these facts, investigations in area of natural and synthetic isocoumarins attract attention of medicinal chemists. The aim of this study was development of new synthetic methodology for synthesis of new isocoumarins and evaluation of their antifungal activity. Applying the palladium-catalysed coupling reaction to 3-bromoiso coumarin, a series of novel 3-substituted isocoumarin derivatives was prepared.

In vitro antifungal activity of series of novel synthesised isocoumarin derivatives was investigated and evaluated. Based on preliminary testing using a dics-diffusion assay, a series of compounds for further testing were selected. By determining MIC values of selected derivatives, the highest antifungal potential showed 3-heteroaryl isocoumarins. The most potent compound showed antifungal activity equal to voriconazole.

The novel methodology for synthesis of 3-substituted isocoumarins was developed. A series of novel 3-substituted isocoumarin derivatives was designed, synthesised and tested against *C. albicans*. 3-Heteroaryl coumarins showed the highest in vitro antifungal activity.

## PRIMENA PAMPA TEHNIKE I QSPR ANALIZE U PROCENI GASTROINTESTINALNE APSORPCIJE I DIZAJNIRANJU NOVIH BIOLOŠKI AKTIVNIH JEDINJENJA

**Vladimir Dobričić, Jelena Savić, Biljana Tubić, Katarina Nikolić,  
Jasmina Brborač, Bojan Marković, Olivera Čudina**

Katedra za farmaceutsku hemiju, Univerzitet u Beogradu - Farmaceutski  
fakultet (Srbija)

PAMPA (*Parallel Artificial Membrane Permeability Assay*) je brza i jednostavna in vitro tehnika za procenu gastrointestinalne apsorpcije. Zasniva se na pasivnoj difuziji ispitivanih supstanci kroz veštačku membranu koja simulira gastrointestinalni trakt. QSPR (*Quantitative Structure- Permeability Relationship Analysis*) povezuje rezultate PAMPA testa sa fizičko-hemijskim osobinama ispitivanih jedinjenja, na osnovu čega je moguć dizajn novih derivata sa poboljšanom apsorpcijom. Cilj rada je procena gastrointestinalne apsorpcije trinaest  $\beta$ -hidroksi- $\beta$ -arilalkanskih kiselina sa antiinflamatornom aktivnošću i četrnaest derivata 1,2-etandiamina i 1,3-propandiamina sa antiproliferativnom aktivnošću primenom PAMPA testa, kao i dizajn novih jedinjenja na osnovu QSPR analiza.

Gastrointestinalna apsorpcija je procenjena na hidrofobnim PVDF PAMPA pločama, impregniranim 1% rastvorom lecitina jajeta u dodekanu (w/v). Molekulski deskriptori ispitivanih jedinjenja su izračunati u programu Dragon i pomoću platforme ChemDes. QSPR modeli su napravljeni u programima Simca 12+ P i STATISTICA.

Za sva ispitivana jedinjenja određeni su koeficijenti permeabilnosti ( $P_{app}$ ) primenom PAMPA testa, a za formiranje QSPR modela izračunati su i negativni logaritmi ovih koeficijenata (-log $P_{app}$ ). Izdvojene su  $\beta$ -hidroksi- $\beta$ -arilalkanske kiseline (1C, 1B i 2C), kao i derivati 1,2-etandiamina i 1,3-propandiamina (DM-EDCP, EDCP i DM-PDCP) sa najvećom permeabilnošću kroz PAMPA membranu. Formirani su ANN-, MLR-, PLS- i SVM-QSPR modeli, pri čemu su najpouzdaniji modeli za predviđanje permeabilnosti MLR(-log $P_{app}$ ) (za  $\beta$ -hidroksi- $\beta$ -arilalkanske kiseline), odnosno PLS(-log $P_{app}$ ) (za derive 1,2-etandiamina i 1,3-propandiamina). Na osnovu deskriptora koji formiraju izdvojene modele predložene su strukturne promene koje bi trebalo da poboljšaju permeabilnost kroz PAMPA veštačku membranu i gastrointestinalnu apsorpciju.

Primenom PAMPA tehnike procenjena je gastrointestinalna apsorpcija trinaest  $\beta$ -hidroksi- $\beta$ -arilalkanskih kiselina, kao i četrnaest derivata 1,2-etandiamina i 1,3-propandiamina. Izdvojeni su derivati sa najvećom permeabilnošću i formirani su QSPR modeli. Analizom najpouzdanijih modela, predložene su strukturne promene i dizajnirani su novi derivati od kojih se može očekivati bolja gastrointestinalna apsorpcija.

# **APPLICATION OF PAMPA TECHNIQUE AND QSPR ANALYSIS IN THE EVALUATION OF GASTROINTESTINAL ABSORPTION AND DESIGN OF NEW BIOLOGICALLY ACTIVE COMPOUNDS**

**Vladimir Dobričić, Jelena Savić, Biljana Tubić, Katarina Nikolić,  
Jasmina Brborač, Bojan Marković, Olivera Čudina**

Department of Pharmaceutical Chemistry, University of Belgrade - Faculty of Pharmacy (Serbia)

PAMPA (Parallel Artificial Membrane Permeability Assay) is a fast and simple in vitro technique used for the evaluation of gastrointestinal absorption. It is based on passive diffusion of tested substances through artificial membrane which simulates gastrointestinal tract. QSPR (Quantitative Structure-Permeability Relationship Analysis) relates PAMPA results to physico-chemical properties of tested compounds, which can be used for design of new derivatives with improved absorption. The aim of this work was evaluation of gastrointestinal absorption of thirteen  $\beta$ -hydroxy- $\beta$ -arylalkanoic acids with antiinflammatory activity and fourteen derivatives of 1,2-ethanediamine and 1,3-propanediamine with antiproliferative activity using PAMPA, as well as design of novel derivatives on the basis of QSPR analyses.

Gastrointestinal absorption was evaluated using hydrophobic PAMPA plates impregnated with 1% egg lecithin solution in dodecane (w/v). Molecular descriptors of tested compounds were calculated using Dragon software and ChemDes platform. QSPR models were created in Simca 12+P and STATISTICA programs.

Permeability coefficients ( $P_{app}$ ) of all tested compounds were determined using PAMPA, whereas for QSPR modelling negative logarithms of these coefficients (-log $P_{app}$ ) were calculated.  $\beta$ -hydroxy- $\beta$ -arylalkanoic acids (1C, 1B and 2C), as well as derivatives of 1,2-ethanediamine and 1,3-propanediamine (DM-EDCP, EDCP and DM-PDCP) with the highest PAMPA permeability were underlined. ANN-, MLR-, PLS- and SVM-QSPR models were created, and the most reliable for permeability prediction were MLR(-log $P_{app}$ ) ( $\beta$ -hydroxy- $\beta$ -arylalkanoic acids) and PLS(-log $P_{app}$ ) (derivatives of 1,2-ethanediamine and 1,3-propanediamine). On the basis of descriptors that form selected models, structural modifications that should improve PAMPA permeability and gastrointestinal absorption were proposed.

Gastrointestinal absorption of thirteen  $\beta$ -hydroxy- $\beta$ -arylalkanoic acids and fourteen derivatives of 1,2-ethanediamine and 1,3-propanediamine was evaluated using PAMPA technique. Derivatives with the highest permeability were underlined and QSPR models were created. After the analysis of the most reliable models, structural modifications were proposed and new derivatives with better expected gastrointestinal absorption were designed.

## IN VITRO ISPITIVANJE INHIBITORNOG POTENCIJALA SINTETISANIH β-HIDROKSI-β-ARILALKANSKIH KISELINA KORIŠĆENJEM KOMERCIJALNOG COX KITA

**Jelena Savić<sup>1</sup>, Jelena Kotur-Stevuljević<sup>2</sup>, Sanda Dilber<sup>3</sup>,  
Sote Vladimirov<sup>1</sup>, Jasmina Brborigić<sup>1</sup>**

<sup>1</sup>Katedra za farmaceutsku hemiju, Univerzitet u Beogradu - Farmaceutski fakultet,

<sup>2</sup>Katedra za medicinsku biohemiju, Univerzitet u Beogradu -

Farmaceutski fakultet, <sup>3</sup>Katedra za organsku hemiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Mehanizam delovanja nesteroidnih antiinflamatornih lekova (NSAIL) je inhibicija enzima ciklooksigenaze (COX) koji postoji u dve izoforme: COX-1 i COX-2. Selektivni COX-2 inhibitori ne izazivaju neželjene gastrointestinalne efekte koji predstavljaju glavni ograničavajući faktor za primenu NSAIL. Cilj rada je da se komercijalnim COX kitom, koji sadrži ovčiju izoformu COX-1 i humanu rekombinatnu izoformu COX-2, *in vitro* odrede IC<sub>50</sub> vrednosti prema svakoj izoformi i procene antiinflamatorni potencijal i selektivnost sintetisanih β-hidroksi-β-arylalkanskih kiselina.

Sedam odabralih β-hidroksi-β-arylalkanskih kiselina je rastvoren u dimetilsulfoksidu, tako da se dobiju koncentracije: 0,01 μM; 0,1 μM; 1 μM; 10 μM, 50 μM i 100 μM. Ostali reagensi su pripremljeni prema uputstvu proizvođača. Test se sastojao iz ciklooksigenazne reakcije, u kojoj je ispitivano jedinjenje inkubirano sa odgovarajućom COX izoformom, i ELISA testa, kojim je kvantifikovan proizvod prethodne COX reakcije.

Dobijene IC<sub>50</sub> vrednosti, koje se nalaze u opsegu 21-327 μmol za COX-1 i 5,2-62,8 μmol za COX-2 izoformu, pokazuju da testirana jedinjenja imaju umereni afinitet za obe izoforme. Izračunati indeks selektivnosti (odnos IC<sub>20(COX-1)</sub>/IC<sub>50(COX-2)</sub>) je u opsegu 1,7-62,7. Najveći indeks selektivnosti i najnižu IC<sub>50(COX-2)</sub> vrednost ima jedinjenje koje je derivat β-hidroksi-β,β-difenilpropionske kiseline sa polarnom nitro grupom na jednom od benzenovih prstenova. Jedinjenje sa nepolarnom trifluorometil grupom na jednom od benzenovih prstenova ima najvišu IC<sub>50(COX-2)</sub> i nizak indeks selektivnosti (3,7). Dve kiseline bez supstituenata na benzenovom prstenu imaju najniži indeks selektivnosti (1,7). *In vitro* testiranjem sedam odabralih β-hidroksi-β-arylalkanskih kiselina na komercijalnom COX kitu određene su IC<sub>50(COX-1)</sub> i IC<sub>50(COX-2)</sub> vrednosti, kao i indeksi selektivnosti. Zaključeno je da je jedinjenje sa polarnom nitro grupom na jednom od benzenovih prstenova najselektivniji COX-2 inhibitor, a da polarna nitro grupa doprinosi jačini inhibicije COX-2.

## **IN VITRO ASSESSMENT OF THE INHIBITORY POTENTIAL OF SYNTHESIZED $\beta$ -HYDROXY- $\beta$ -ARYLALKANOIC ACIDS USING COMMERCIAL COX KIT**

**Jelena Savić<sup>1</sup>, Jelena Kotur-Stevuljević<sup>2</sup>, Sanda Dilber<sup>3</sup>,  
Sote Vladimirov<sup>1</sup>, Jasmina Brborić<sup>1</sup>**

<sup>1</sup>Department of Pharmaceutical Chemistry, University of Belgrade - Faculty of Pharmacy, <sup>2</sup>Department of Medical Biochemistry, University of Belgrade - Faculty of Pharmacy, <sup>3</sup>Department of Organic Chemistry, University of Belgrade - Faculty of Pharmacy (Serbia)

Mechanism of action of nonsteroidal anti-inflammatory drugs (NSAIDs) is inhibition of enzyme cyclooxygenase (COX) which exists in two isoforms: COX-1 and COX-2. Selective COX-2 inhibitors do not cause gastrointestinal side effects which are limiting factor for NSAIDs administration. The aim of this work is to in vitro determine IC<sub>50</sub> values towards each isoform, asses anti-inflammatory potential and selectivity of synthetized  $\beta$ -hydroxy- $\beta$ -arylalkanoic acids using commercial COX kit which includes ovine COX-1 and human recombinant COX-2 isoform.

Seven selected  $\beta$ -hydroxy- $\beta$ -arylalkanoic acids were dissolved in dimethyl sulfoxide to obtain concentrations: 0.01  $\mu$ M; 0.1  $\mu$ M; 1  $\mu$ M; 10  $\mu$ M, 50  $\mu$ M and 100  $\mu$ M. The rest of the reagents is prepared according to the manufacturer's manual. The test has consisted of cyclooxygenase reaction in which tested compound was incubated with corresponding COX isoform and ELISA which quantifies the product of the previous reaction.

Obtained IC<sub>50</sub> values which are in the range 21-327  $\mu$ mol for COX-1 and 5.2-62.8  $\mu$ mol for COX-2 isoform shows that tested compounds exhibit moderate affinity for each isoform. Calculated selectivity index (ratio IC<sub>50(COX-1)</sub>/IC<sub>50(COX-2)</sub>) is in the range 1.7-62.7. A compound which is a derivative of  $\beta$ -hydroxy- $\beta$ , $\beta$ -diphenylpropionic acid with a polar nitro group on one benzene ring showed the highest selectivity index and the lowest IC<sub>50(COX-2)</sub> value. A compound with a nonpolar trifluoromethyl group on one benzene ring has the highest IC<sub>50(COX-2)</sub> value and low selectivity index (3.7). Two acids without substituents on benzene ring have the lowest selectivity index (1.7).

IC<sub>50(COX-1)</sub> and IC<sub>50(COX-2)</sub>, as well as selectivity indices of seven selected  $\beta$ -hydroxy- $\beta$ -arylalkanoic acids were determined using commercial COX kit. It was concluded that the compound with the polar nitro group on one benzene ring is the most selective COX-2 inhibitor and that polar nitro group contributes to the strength of COX-2 inhibition.

## **EUROPEAN ASSOCIATION OF HOSPITAL PHARMACY (EAHP) COMPETENCY FRAMEWORK FOR HOSPITAL PHARMACY**

**Petr Horak**

European Association of Hospital Pharmacy

The European Association of Hospital Pharmacists (EAHP), and its 35 member country platforms are creating a Common Training Framework (CTF) for hospital pharmacy education in Europe with a focus on competency-based approaches to training, staff development and assessment which are increasingly viewed as a central strategy for improving the effectiveness of those who provide care.

Through the CTF project, EAHP worked on securing voluntary agreement across countries about the knowledge, skills and attitudes/behaviours that underpin advanced practice in the hospital sector. This draft framework was reviewed through a Delphi Consultation in which a diverse group of stakeholders participated. Furthermore, EAHP conducted the labour mobility survey aiming at exploring the attitudes and perspectives of hospital pharmacists on labour mobility within Europe.

The final competency framework has 24 competencies, 87 knowledge items identified and 136 behaviour competencies. Clusters with individual outcomes defined are: patient care and clinical pharmacy skills competencies; medicines and their use related competencies; management competencies and professional competencies. Moreover, the results of the labour survey make it clear that hospital pharmacists are interested in pursuing their free movement rights, while 85% of the survey participants would support the creation of a CTF for the hospital pharmacy specialisation. This would loosen perceived barriers to labour mobility in Europe and make the comparability of competencies, knowledge and skills possible across the continent.

CTF would set an educational benchmark for all European countries to strive for. Development of the profession, facilitation of exchange of expertise, standardisation in the quality of education and increase of mobility opportunities are benefits to be gained by creating a new tool for automatic cross-border recognition of the hospital pharmacy specialisation.

## SAVREMENI PRISTUP OPTIMIZACIJI ANTIBIOTSKE TERAPIJE NA OSNOVU IZMERENIH KONCENTRACIJA

Katarina Vučićević

Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu-  
Farmaceutski fakultet (Srbija)

U fokusu terapijskog praćenja lekova (Therapeutic Drug Monitoring, TDM) su oduvek bili lekovi varijabilne farmakokinetike, uskog terapijskog raspona, za koje je dokazana i utvrđena veza između koncentracije leka u plazmi i farmakološkog efekta. Od svog uvođenja do danas TDM je ostao jedan od glavnih alata u kliničkoj praksi u procesu individualizacije režima doziranja antibiotika kod različitih populacija pacijenata (npr. kritično oboleli, pedijatrijski, gerijatrijski). Razvoj farmakometrije kao naučne discipline i intenzivni napredak u oblasti bioanalitičkih metoda, danas ojačava poziciju TDM-a i pruža mu dodatne mogućnosti u optimizaciji antiobiotske terapije. Stoga, ovaj rad ima za cilj da predstavi savremeni pristup TDM-a.

Populacioni farmakokinetičko-farmakodinamički modeli prošireni matematičkim modelima koji opisuju razvoj rezistencije, stepen adherence i ishode lečenja su najsloženiji alati koji se danas koriste u procesu individualizacije. Jedan od pristupa je da se na osnovu demografskih, farmakogenetičkih i kliničkih podataka pristupi optimizaciji režima doziranja antibiotika, dok se dodatno u okviru TDM-a sa ciljem individualizacije doziranja uzima u obzir izmerena/e koncentracija/e antibiotika u biološkom materijalu.

Kako bi modeli razvijeni na velikoj grupi pacijenata bili upotrebljivi u kliničkoj praksi ili kako bi se unapredili postojeći korišćenjem izmerenih koncentracija leka kod pacijenata neophodno je da budu jednostavnii za korišćenje u svakodnevnom radu. Stoga su oni inkorporirani u kompjuterske programe koji ne zahtevaju poznavanje složene farmakometrijske metodologije. Kliničkim farmaceutima su danas na raspolaganju mnogobrojne aplikacije za mobilne telefone, kompjuterski programi ili inter-/intra-net platforme. Unosom osnovnih podataka o pacijentu i izborom ostalih ključnih podataka mogu relativno brzo, jednostavno i pouzdano dobiti ključne smernice za optimalni izbor doze i dužine terapije antibioticima. Savremeni koncept TDM-a antibiotika baziran na integrisanim populacionim modelima leka, uzročnika i organizma pacijenta u kome se procesi odvijaju, povećava verovatnoću postizanja željenog ishoda terapije kod pacijenta.

*Istraživanje je realizovano u okviru projekta ON-175023 finansiranog od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije.*

# **A MODERN APPROACH TO OPTIMIZING ANTIBIOTIC THERAPY BASED ON MEASURED CONCENTRATIONS**

**Katarina Vučićević**

Department of Pharmacokinetics and Clinical Pharmacy, University of Belgrade-Faculty of Pharmacy (Serbia)

Drugs with highly variable pharmacokinetics, narrow therapeutic index, with the established relationship between plasma drug concentration and pharmacological effect have always been in the focus of therapeutic drug monitoring (TDM). Since its introduction to date, TDM remained one of the main tools in the clinical practice in the process of individualizing antibiotic dosing regimens in different patient populations (e.g. critically ill, pediatric and geriatric). Development of pharmacometrics as a scientific discipline and intensive advancement in the field of bioanalytical methods are strengthening TDM's position and provide additional opportunities in optimizing antibiotic therapy. Therefore, this paper aims to present a modern TDM approach.

Integrated population pharmacokinetic-pharmacodynamic models expanded by mathematical models describing the bacterial development of resistance, patient's adherence over the treatment course and the treatment outcomes are the most complex tools used in the process of dose individualization today. The basic approach utilizes demographic, pharmacogenetic and clinical patient's data, while TDM-based individualization additionally takes into account measured antibiotic level in the biological matrix.

The use of big data models developed on a large group of patients and their continuous improvement through measured drug concentration inputs ought to be straightforward in everyday clinical practice. Therefore, they are incorporated into computer softwares that do not require knowledge of a complex pharmacometric methodology. Nowadays, various applications for mobile phones, computer programs or inter/intranet platforms are accessible to clinical pharmacists. Entering basic patient's information and other relevant data, guidelines for optimal antibiotic dosing could be quickly available and reliable.

The modern concept of antibiotics' TDM is based on population models of the drug, integrated with models of the bacterial resistance and the patient's organism, aiming to increase the probability of achieving the desired therapy outcome in an individual patient.

*This work was conducted as a part of the project ON-175023) funded by the Ministry of Education, Science and Technological Development, Belgrade, Republic of Serbia.*

## **INTERVENCIJE KLINIČKOG FARMACEUTA NA ODELJENJU HEPATOLOGIJE**

**Milica Ćulafić**

Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu-  
Farmaceutski fakultet i Transplantaciona hepatologija, Klinika za  
gastroenterologiju i hepatologiju, Klinički centar Srbije, Beograd (Srbija)

Na odeljenju hepatologije zbrinjavaju se pacijenti sa različitim bolestima jetre, pacijenti koji se pripremaju za transplantaciju jetre, te vrši kontinuirano praćenje pacijenata nakon transplantacije. Pristup rešavanju problema vezanih za lekove kod pacijenata sa bolestima jetre zahteva znanja o farmakokineticici i farmakodinamici lekova koje je potrebno interpretirati u odnosu na individualne karakteristike pacijenta. Cilj studije je prikazati pregled aktivnosti farmaceuta u posmatranom periodu kod pacijenata sa oslabljenom funkcijom jetre te kod transplantiranih pacijenata. Praćene su aktivnosti farmaceuta tokom boravka na odeljenju hepatologije Klinike za gastroenterologiju i hepatologiju, Kliničkog centra Srbije (2x nedeljno), kao i konsultacije koje su usledile nakon toga. Procenjen je i stepen prihvaćenosti predloženih intervencija od strane nadležnih lekara uperiodu od juna 2017. do juna 2018. godine.

Zabeleženo je 519 intervencija u vezi sa lekovima od čega je farmaceut inicirao 71,9% intervencija. Preostale intervencije koje je imao farmaceut sprovedene su na osnovu upita lekara (22,5%), pacijenta (3,1%), medicinske sestre (2,5%). Najčešće intervencije su se ticale neželjenih reakcija na lek (24,0%), sledi izborleka/korekcija doze (18,3%), interakcije (16,5%), savetovanje pacijenata o lekovima posle transplantacije (16,0%), konverzija sa IV na oralnu primenu (11,7%), edukacija članova zdravstvenog tima o (novim) lekovima (9,9%). Intervencije su bile prihvачene u 91,6% slučajeva. Sproveden je značajan broj intervencija od kojih je većina inicirana od strane farmaceuta. Uključivanje farmaceuta je dovelo do optimizacije farmakoterapije što je reflektovano prihvatanjem od strane hepatološkog tima. Farmaceut poseduje znanja i veštine koje mogu unaprediti brigu o hepatološkim pacijentima, te poboljšati ishode lečenja.

## **CLINICAL PHARMACIST'S INTERVENTIONS ON A HEPATOLOGY WARD**

**Milica Ćulafić**

Department of Pharmacokinetics and Clinical Pharmacy, University of Belgrade-Faculty of Pharmacy and Hepatology and Liver Transplant Unit, Clinic for Gastroenterology and Hepatology, Clinical Center of Serbia, Belgrade (Serbia)

Hepatology unit is involved in the treatment of patients with different liver disorders but also provides care for patients who are preparing for a liver transplant, and conducts continuous monitoring of post liver transplant recipients. Interventions to identify and resolve drug-related problems in patients with liver diseases require knowledge of pharmacokinetic and pharmacodynamic medication characteristics that need to be interpreted in relation to the individual patient needs. This study aimed to present an overview of the activities of pharmacist directed to patients with hepatic impairment and liver transplant recipients during the study period. Pharmacist activities on the Hepatology ward at Clinic for Gastroenterology and Hepatology (2 days per week), as well as consultations that followed subsequently, were observed. The degree of acceptance for the proposed interventions by competent doctors was also recorded during the study period from June 2017 to June 2018.

We documented 519 interventions in regards to drug-related problems of which the majority was pharmacist-initiated (71.9%). Other interventions conducted by the pharmacist were a result of queries by physicians (22.5%), patients (3.1%) and nurses (2.5%). The most common interventions were related to the adverse drug reactions (23.0%), followed by choice of the drug/correction of the dose (19.3%), the interactions (16.5%), patient counseling for post liver transplant medications (16.0%), conversion from IV to oral administration (11.7 %), education of members of the healthcare team for the (new) drugs (9.9%). Physicians' acceptance rate of pharmacy interventions was 91.6%. Significant number of interventions (majority initiated by the pharmacist) was recorded. The inclusion of pharmacist has led to the optimisation of pharmacotherapy as reflected by the significant intervention acceptance rate of hepatology/transplant team. The pharmacists have the knowledge and skills that may enhance the care of patients with liver diseases and improve treatment outcomes.

## ZDRAVSTVENA ANALIZA VRSTE I ISHODA GREŠKE - *HEALTHCARE FAILURE MODE AND EFFECT ANALYSIS (HFMEA)* PRIMENJENA NA PROCES SUPSTITUCIJE ANTIBIOTSKE TERAPIJE TOKOM NESTAŠICE LEKOVA

**Nenad Miljković<sup>1</sup>, Karyofyllis Tsakitzis<sup>2</sup>, Cristina Garcia Yubero<sup>3</sup>,  
Branislava Miljković<sup>4</sup>**

<sup>1</sup>Institut za ortopedsko-hirurške bolesti „Banjica”, Beograd, <sup>2</sup>Nosokomeio Thessalonikis Georgios Papanikolaou (Grčka), <sup>3</sup>El Hospital Universitario Infanta Sofía Salud Madrid (Španija), <sup>4</sup>Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu- Farmaceutski fakultet (Srbija)

Zdravstvena analiza vrste i ishoda greške-*Healthcare Failure Mode and Effect Analysis* (HFMEA) predstavlja prospективnu analizu rizika, koja se sprovodi radi smanjenja rizika po zdravlje pacijenta, u toku pružanja zdravstvene zaštite. Cilj ovog istraživanja je komparativna procena primene HFMEA u određivanju i prioritizaciji rizika kod zamene antibiotske terapije tokom nestašice lekova.

HFMEA metoda je sprovedena u Institutu za ortopedsko-hirurške bolesti „Banjica” (bolnica A), Nosokomeio Thessalonikis Georgios Papanikolaou (bolnica B) i El Hospital Universitario Infanta Sofía Salud Madrid (bolnica C) u periodu od februara do juna 2018. Nakon identifikacije i kvantifikacije rizika na osnovu njihove ozbilnosti i verovatnoće nastanka, sprovedena je i komparativna hazard analiza grešaka, i njihovih uzroka.

HFMEA metodom je ustanovljeno 13 tipova grešaka u bolnici B, 12 u bolnici A i 10 u bolnici C. U bolnicama A i B identifikovano je 12 uzroka grešaka, dok je 9 određeno u bolnici C. Greške vezane za supstituciju antibiotika sa najvišim hazard brojem u bolnicama A, B i C su: nesprovođenje pregleda novonastalih interakcija lekova; nesprovođenje procene potrebe za dodatnim monitoringom pacijenta; neadekvatni prenos informacije o supstitucionoj terapiji; neodgovarajuća procena stanja pacijenta i nepostojanje liste alternativnih antibiotika kada nestašica nastupi. Uzroci grešaka zajednički za sve bolnice su nedostatak vremena, neadekvatna komunikacija među zdravstvenim radnicima i neadekvatna podrška u vidu informacionih tehnologija. Predložene mere kontrole rizika su efikasnija raspodela vremena zaposlenih, pisani protokoli za supstituciju antibiotika i validacija procesa zamene leka od strane zdravstvenih radnika koji učestvuju u donošenju odluke o antibiotskoj terapiji i primeni antimikrobne terapije.

Rezultati HFMEA metode, ukazuju na neophodnost potpunog pristupa zdravstvenim radniku informacijama o alternativnim terapijskim opcijama, efikasnije komunikacije među zdravstvenim radnicima i detaljnijeg uvida u zdravstvenu dokumentaciju pacijenta, te potrebe za dodatnim monitoringom, kao osnovnih mera za kontrolu rizika koji se javljaju prilikom zamene terapije tokom nestašice lekova.

## **HEALTHCARE FAILURE MODE AND EFFECT ANALYSIS (HFMEA) APPLIED TO ANTIBIOTIC SUBSTITUTION IN MEDICINE SHORTAGES**

**Nenad Miljković<sup>1</sup>, Karyofyllis Tsakritzis<sup>2</sup>, Cristina Garcia Yubero<sup>3</sup>,  
Branislava Miljković<sup>4</sup>**

<sup>1</sup>The Institute of Orthopaedic Surgery Banjica, <sup>2</sup>The George Papanikolau Hospital Thessaloniki, <sup>3</sup>The Infanta Sofia Hospital Madrid, <sup>4</sup>Department of Pharmacokinetics and Clinical Pharmacy, University of Belgrade- Faculty of Pharmacy (Serbia)

Healthcare Failure Mode and Effect Analysis (HFMEA) is a prospective risk assessment applied in healthcare settings to reduce patient risk while still providing health services. Therein, this study compares HFMEA as a risk assessment and prioritisation tool within antibiotic substitution in medicine shortages.

The HFMEA was carried out at the Institute of Orthopaedic Surgery Banjica in Belgrade (Hospital A), the George Papanikolau Hospital in Thessaloniki (Hospital B), and the Infanta Sofia Hospital in Madrid (Hospital C) from February to June, 2018. Upon identifying risks and quantifying them as based on their severity and probability, including their proposed control measures, a comparative-failure mode and cause hazard-score analysis was conducted.

13 failure modes in Hospital B, 12 in A, and 10 in C were detected. 12 failure-mode causes were identified in Hospitals A and B, and 9 in C. These linked to antibiotic substitution which scored highest among hazards in all hospitals were found to be the following: non-verification of new drug interactions; lack of additional patient monitoring; inadequately communicating the substitution's specifics to staff; not fully reviewing the patient's condition; and no available list of alternative antibiotics for medicine shortage. The failure mode causes for all hospitals were analysed as: lack of time, insufficient communication among healthcare professionals regarding the substitution; and insufficient IT support. The risk control measures suggested include: more efficient time management; written protocol for antibiotic substitution; and the substitution's validation by healthcare professionals involved in the decision making process and medicine administration.

The HFMEA results demonstrate that full access to information on treatment alternatives, more efficient inter-staff communication, a valid assessment of patient records, and the need for additional patient monitoring all represent main risk-control measures. To improve outcomes, these must be carried out in substituting a medicine while there is a shortage.

## **MIKROBIOTA CREVA, ZNAČAJ ZA ETIOPATOGENEZU I TERAPIJU AUTOIMUNSKIH BOLESTI**

**Đorđe Miljković**

Odeljenje za imunologiju, Institut za biološka istraživanja „Siniša Stanković”,  
Univerzitet u Beogradu (Srbija)

Mikrobiota creva ostvaruje intenzivnu interakciju sa organizmom čoveka i umnogome utiče na funkciju osnovnih homeostatskih organskih sistema čoveka, uključujući i imunski sistem. Smatra se da je evolucija adaptivnog imunskog sistema uslovljena uspostavljanjem komensalih i simbiotskih odnosa klčmenjaka sa mikroorganizmima creva. Zna se da mikroorganizmi creva na različite načine deluju na imunske ćelije, kako u samom gastrointestinalnom traktu, tako i na sistemskom nivou. Sve je više podataka koji ukazuju da bi mikrobiota creva mogla da ima presudnu ulogu u otpočinjanju i propagaciji, ali i sprečavanju i regulaciji neadekvatnih imunskih odgovora, kakvi se dešavaju u autoimunskim bolestima. U ovom izlaganju će biti predstavljeni ključni nalazi koji podržavaju hipotezu o značaju mikrobiote creva za etiopatogenezu autoimunskih bolesti, kao i oni koji ukazuju da bi modulacija mikrobiote creva mogla biti adekvatan pristup u terapiji autoimunskih bolesti. Takođe, biće predstavljena i istraživanja Odeljenja za imunologiju Instituta za biološka istraživanja „Siniša Stanković” koja se tiču uloge mikrobiote creva u patogenezi multiple skleroze i dijabetesa melitusa tip I.

## **THE ROLE OF GUT MICROBIOTA IN ETIOPATHOGENESIS AND THERAPY OF AUTOIMMUNE DISEASES**

**Đorđe Miljković**

Department of Immunology, Institute for Biological Research „Sinisa Stankovic”, University of Belgrade (Serbia)

Gut microbiota interacts intensively with human body, having enormous impact on the homeostatic systems, including immune system. The very origin of the adaptive immune system is closely related to the incorporation of commensal and symbiotic microorganisms into the gut. Gut microorganisms influence immune cells in humans in various ways, both within the gut-associated lymphoid tissue and at the systemic level. An increasing body of evidence suggests that gut microbiota has the decisive role in the initiation and the propagation, as well as in the prevention and the regulation of the inappropriate immune response that is the major feature of autoimmune disorder pathogenesis. The key data supporting a hypothesis that gut microbiota is the crucial element in the etiopathogenesis of autoimmune disorders will be presented. Moreover, results of the Department of Immunology, Institute for biological Research „Sinisa Stankovic”, related to the role of gut microbiota in multiple sclerosis and diabetes type 1 will be shown.

## REGULATORNE T ĆELIJE – NOVI PRISTUP U LEĆENJU AUTOIMUNSKIH BOLESTI

Ivana Stojanović

Odeljenje za imunologiju, Institut za biološka istraživanja „Siniša Stanković“,  
Univerzitet u Beogradu (Srbija)

Medicinski proizvodi za naprednu terapiju (od engl. *Advanced therapy medicinal products* - ATMP) se baziraju na genima, tkivima i ćelijama koje se koriste u terapiji oboljenja i povreda kod ljudi. Iako su neke od ATMP ušle u kliničku praksu, imunoterapija autoimunskih bolesti regulatornim T ćelijama (Treg) još je u fazi I kliničkih studija. Uobičajeno se koriste Treg iz krvi obolele osobe (iako postoji mogućnost disfunkcije Treg), i poliklonske Treg, tj. one koje prepoznaju različite antigene (naspram antigen-specifičnih koje su specifičnije). Da bi se prevazišao potencijalni problem disfunkcije Treg i obezbedila njihova specifičnost delovanja, cilj naše studije bio je da napravimo insulin-specifične Treg (za lečenje tip 1 dijabetesa) od efektorskih insulin-specifičnih T limfocita primenom različitih *in vitro* postupaka. Prva prepreka na tom putu bio je mali broj insulin-specifičnih efektorskih Treg u NOD miševima koji spontano razvijaju dijabetes. Ove ćelije smo umnožili kultivacijom CD4+ T ćelija na dendritskim ćelijama u prisustvu insulin peptida B9:23. Od inicijalnih 0,1% dobili smo  $5,4 \pm 2,1\%$  insulin-specifičnih ćelija nakon 24 sata, koje su dalje sortirane uz pomoć insulin-specifičnih MHC tetramera klase II i izložene stimulatorima i IL-2 iTGF-β. Bilo je neophodno samo 48 sati da efektorske ćelije konvertuju u Treg fenotip (CD4+CD25<sup>high</sup>FoxP3+GITR+CD127<sup>+</sup>). Ove ćelije su proizvodile TGF-β i suprimirale proliferaciju efektorskih ćelija *in vitro*. Takođe, eksprimirale su hemokinski receptor CXCR3 koji može da upravlja njihovu migraciju u pankreas. Ovi rezultati ukazuju na mogućnost *in vitro* konverzije efektorskih ćelija u regulatorne sa specifičnosću na zadati antigen, ali njihova efikasnost u terapiji tip 1 dijabetesa tek treba da bude ispitana.

*Finansirano od strane Porodične fondacije Jakoka (Boston, SAD) i delom sa projekta 173013 Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije. Obeleženi tetrameri su poklon NIH Tetramer Core Facility.*

## **T REGULATORY CELLS - NEW APPROACH IN THE TREATMENT OF AUTOIMMUNITY**

**Ivana Stojanović**

Department of Immunology, Institute for Biological Research „Sinisa Stankovic”, University of Belgrade (Serbia)

Advanced therapy medicinal products (ATMPs) are medicines that are based on genes, tissues or cells. Although several ATMPs entered clinical practice, immunotherapy of autoimmune diseases with CD4<sup>+</sup>CD25<sup>high</sup>FoxP3<sup>+</sup> regulatory T cells (Tregs) is still at the stage of phase I clinical trials. Usually, Tregs used for human therapy are isolated from individual's blood (concern about the function of Treg) and they are polyclonal i.e. they recognize a number of different antigens (antigen-specific Tregs are more specific). So, to overcome the problem of possible Treg malfunction and to provide antigen-specificity of Tregs, we aimed to generate insulin-specific Tregs (for the treatment of type 1 diabetes) from insulin-specific T effector cells using various *in vitro* manipulations. Due to low occurrence of insulin-specific CD4<sup>+</sup> T cells in NOD mice that develop type 1 diabetes spontaneously (0.1%), CD4<sup>+</sup> T cells were co-cultured with autologous mature dendritic cells (DC) in the presence of insulin B9:23. The proportion of activated insulin-specific CD4<sup>+</sup>CD69<sup>+</sup> T cells increased up to 5.4±2.1% after 24 h and they were then sorted using fluorescent insulin-loaded MHC class II tetramers and cultured with TCR stimulators, recombinant IL-2 and TGF-β. It took only 48 h for cells to fully convert to CD4<sup>+</sup>CD25<sup>high</sup>FoxP3<sup>+</sup>GITR<sup>+</sup>CD127<sup>-</sup> Tregs phenotype. These converted cells produced TGF-β and suppressed proliferation of T effectors *in vitro*. Also, they expressed CXCR3 that can direct cell migration toward the pancreas. Their proliferation was stimulated only in the case of exposure to immature DC. These results confirm that it is feasible to generate antigen-specific Tregs from antigen-specific effector cells, but the effectiveness of these cells *in vivo* remains to be determined.

*Funded by the Iacocca Family Foundation (Boston, USA) and partly by the Ministry of Science, Education and Technological Development, Republic of Serbia (173013). Fluorescent MHC class II tetramers were a donation from NIH Tetramer Core Facility.*

## **ADRENERGIČKI LEKOVI – KANDIDATI ZA NOVE NEKONVENCIONALNE IMUNOMODULATORNE LEKOVE?**

**Gordana Leposavić**

Katedra za fiziologiju, Univerzitet u Beogradu-Farmaceutski fakultet (Srbija)

Brojni nalazi pokazuju da su kateholamini, krajnji medijatori simpato-adrenalne osovine, uključeni u modulaciju urođenog i stečenog imunskog odgovora. Osim toga, pokazano je da imunske ćelije mogu da sintetišu noradrenalin (tzv. „adrenergičke ćelije”) koji deluje autokrino/parakrino. Ovo svojstvo imunskih ćelija je posebno važno onda kada je izmenjeno (smanjeno) neurokrino delovanje kateholamina (npr. u hroničnom stresu, inflamaciji). Pokazano je da skoro svi tipovi imunskih ćelija eksprimiraju  $\beta$ -adrenergičke receptore i da kateholamini deluju imunomodulatorno uglavnom posredstvom ovog tipa adrenergičkih receptora. Međutim, sve je više podataka da su i  $\alpha$ -adrenergički receptori uključeni u imunomodulaciju. Naša istraživanja pokazuju da  $\alpha$ -adrenergičke receptore ispoljavaju antigen prezentujuće ćelije, uključujući dendritske ćelije (koje imaju ključnu ulogu u iniciranju i usmeravanju stečenog imunskog odgovora) i CD4+CD25+FoxP3+ regulatorne T-ćelije, koje su važne za održavanje imunske homeostaze i prevenciju razvoja autoimunskih bolesti. Brojni nalazi ukazuju da se sinteza kateholamina menja u mononuklearnim ćelijama periferne krvi tokom razvoja autoimunskih bolesti, kao što su reumatoidni artritis i multipla skleroza. Takođe je pokazano da se sadržaj noradrenalina u nervnim vlaknima i njegova sinteza u imunskim ćelijama u sekundarnim limfoidnim organima i ciljnom organu menja tokom razvoja eksperimentalno indukovanih autoimunskih bolesti, kao što su kolagenom-indukovani artritis (model reumatoidnog artritisa) i eksperimentalni autoimunski encefalomijelitis (model multiple skleroze). Osim toga, farmakološke manipulacije delovanjem kateholamina tokom razvoja eksperimentalno indukovanih autoimunskih bolesti ukazuju da kateholamini učestvuju u njihovoј patogenezi. Naša skorašnja ispitivanja su ukazala na celularne i molekularne mehanizme koji bi mogli da objasne ulogu kateholamina u sklopu kompleksne imunopatogenze eksperimentalnog autoimunskog encefalomijelitisa. Imajući u vidu da su adrenergički lekovi jeftini i dobrog bezbednosnog profila, sagledavanje njihovog imunomodulatornog potencijala bi moglo da bude važno za proširenje njihovih terapijskih indikacija na adjuvantnu terapiju ne samo multiple skleroze, veći drugih autoimunskih bolesti.

*Rad je finansiralo MPNTR Republike Srbije, projekat 175050.*

## **ADRENERGIC DRUGS – CANDIDATES AS NOVEL NON-CONVENTIONAL IMMUNOMODULATORY DRUGS?**

**Gordana Leposavić**

Department of Physiology, University of Belgrade - Faculty of Pharmacy  
(Serbia)

There is a pile of evidence that catecholamines, the end-point mediators of sympathoadrenal axis, modulate innate and adaptive immune responses. Immune cells also synthesize noradrenaline, which then acts in autocrine/paracrine manner. Noradrenaline synthesis in immune cells („adrenergic” cells) is shown to be particularly important under condition affecting catecholamine neurocrine action (e.g. under stress, during inflammation). Catecholamine effects on immune cells are mediated mainly through  $\beta$ -adrenoceptors, which are expressed on almost all types of innate and adaptive immunity cells. However, emerging data indicate a role for  $\alpha$ -adrenoceptors in immunomodulation. Our finding showed that  $\alpha$ -adrenoceptors on antigen presenting cells, including dendritic cells, which are crucial in initiation and direction of adaptive immune response, but also on CD4+CD25+FoxP3+ regulatory T cells, which are important in maintaining immune homeostasis and preventing development of autoimmune diseases. An accumulating body of evidence indicate that synthesis of catecholamines in peripheral blood mononuclear cells changes during development of autoimmune diseases, such as multiple sclerosis and rheumatoid arthritis. There are also data indicating that noradrenaline release from sympathetic nerve fibres and its synthesis in immune cells in secondary lymphoid organs and target tissues change during development of experimentally induced autoimmune diseases, such as collagen-induced arthritis (rheumatoid arthritis model) and experimental autoimmune encephalomyelitis (multiple sclerosis model). Additionally, pharmacological manipulations with catecholamine action during development of experimentally induced autoimmune diseases strongly suggested that they are involved in their pathogenesis. Our recent studies pointed out to mechanisms underlying adrenoceptor-mediated immunomodulation during development of experimental autoimmune encephalomyelitis. Considering the fact that adrenergic drugs are cheap and have a good safety profile, understanding their role in development of autoimmune diseases could be important for extending their therapeutic indications to include adjuvant therapy of autoimmune diseases.

*This work was supported by research grant 175050, Ministry of Science, Education and Technological Development, Republic of Serbia.*

## SAVREMENA TERAPIJA MULTIPLE SKLEROZE - OD IMUNOMODULACIJE DO SELEKTIVNE IMUNE REKONSTITUCIJE

Dragana Obradović

Klinika za neurologiju, Vojnomedicinska akademija, Beograd (Srbija)

Multipla skleroza (MS) je imunološki posredovano oboljenje centralnog nervnog sistema koje u u osnovi karakteriše inflamacija, demijelinizacija i progresivan gubitak aksona. Najčešće se javlja u dobi između 20. i 40. godine života i ukoliko se ne leči dovodi do teškog invaliditeta. Postoji globalni porast prevalence oboljevanja od MS, naročito u ženskoj populaciji, oboljevaju čak tri puta češće. Faktori rizika za nastanak MS su pušenje, nizak nivo vitamin D3, gojaznost u dečijem dobu, infekcija EBV i prisustvo HLA DRB15 gena. Poslednjih dvadeset godina napravljen je ogroman napredak u lečenju MS i trenutno je na raspolaganju čak 18 različitih lekova. Istovremeno, dokazano je da rana terapija, na samom početku bolesti, bitno menja tok bolesti i značajno odlaže pojavu onesposobljenosti. Postoje imunomodulatorni lekovi: interferoni beta, glatiramer, dimetil fumarat i imunosupresivni lekovi – fingolimod, teriflunomid, kladribin, natalizumab, alemtuzumab, okrelizumab i mitiksantron koji se danas koriste u lečenju ove bolesti. Pojedini lekovi zahtevaju stalnu, kontinuiranu primenu da bi se njihov pozitivan efekat održao, dok druga grupa lekova izaziva kratkotrajnu imunosupresiju sa naknadnom rekonstitucijom imunog sistema i održivim kilničkim efektom (kladribin i alemtuzumab, limfoablacija i autologa transplantacija matičnih ćelija). Efekti terapije se odnose na redukciju broja relapsa, usporavanje/zaustavljanje razvoja onesposobljenosti, redukciju broja novih demijelinizacionih lezija, zaustavljanje atrofije mozga i čak poboljšanje neurološkog statusa. Osnovno pravilo u lečenju je da se lek bira prema karakteristikama samog pacijenta i prema karakteristikama njegove bolesti – aktivnosti i težine, tako da govorimo o pravoj personalizovanoj terapiji. Vrlo je značajan monitoring tokom terapije pojedinim lekovima, obzirom na moguće neželjene efekte. Potrebne su godišnje neurološke i MRI kontrole i u slučaju neefikasnosti leka, neophodna je promena terapije, odnosno prelazak na efikasniji lek. Rani početak i adekvatan izbor leka su ključni za dobru kontrolu bolesti i dobar kvalitet života obolelih od MS.

## **CURRENT THERAPY OF MULTIPLE SCLEROSIS – FROM IMMUNOMODULATION TO IMMUNE RECONSTITUTION**

**Dragana Obradović**

Department of Neurology, Military Medical Academy, Belgrade (Serbia)

Multiple sclerosis (MS) is immune mediated disease of the central nervous system, characterized by inflammation, demyelination and progressive axonal loss. Peak incidence of MS is between 20 and 40 year of life. There is global trend in MS prevalence increase, especially in female population, women being three times more affected. MS risk factors are smoking, vitamin D3 deficiency, EBV infection and obesity and HLADRB15 presence. Enormous progress in MS treatment was made in the last 20 years, and 18 drugs are available at the moment. Early treatment, just after the diagnosis of MS, has been proven to modify MS course and postpone disability. There are two groups of drugs, immunomodulatory ones, such as: interferons beta, glatiramer, dimethyl fumarate and immunosuppressive ones, such as: fingolimod, teriflunomid, cladribin, natalizumab, alemtuzumab, ocerlizumab and mitoxantron. They have different mode of action and different mode of application. Some of them need to be used continuously in order to maintain efficacy, while others cause short time immunosuppression, subsequent immune reconstitution and longer lasting clinical benefit. Goals of MS treatments are relapse reduction, slowing of disability, reduction of new demyelinated lesions and brain atrophy on MRI and even improvement of neurological status. Today's guidelines recommend selection of treatment according to patients characteristics and characteristics' of the disease (activity and severity), which means personalized treatment. Considering possible adverse events, regular follow-up is mandatory, specifically for some drugs (fingolimod, alemtuzumab, natalizumab). Yearly clinical and MRI follow-up is strongly recommended and in case of lack of treatment response, switching to more active therapy is obligatory. Early start and adequate choice of MS drug are crucial for disease control and satisfactory quality of life of MS patients.

## **SAVREMENI PRISTUP U LEĆENJU REUMATOIDNOG ARTRITISA**

**Mirjana Šefik Bukilica**

Institut za reumatologiju, Univerzitet u Beogradu - Medicinski fakultet (Srbija)

Reumatoidni artritis (RA) je hronično, destruktivno, zapaljensko oboljenje perifernih zglobova. Do kraja XX veka terapija RA se oslanjala na upotrebu lekova koji su razvijeni empirijskim pristupom bez detaljnog razumevanja molekularnih mehanizama koji su od značaja za patogenezu bolesti. Međutim, poslednjih decenija svedoci smo eksplozije našeg razumevanja zapaljenskog procesa i molekularnih puteva koji su uključeni u RA. To je dovelo do novog pristupa u razvoju lekova koji je omogućio primenu ciljane terapije uperene direktno na molekule za koje se smatra da su uključeni u proces zapaljenja. Primena tehnologije za proizvodnju monoklonskih antitela u terapijske svrhe bila je glavni preduslov za razvoj biološke terapije. Prvi biološki lekovi koji su uvedeni u terapiju RA bili su inhibitori faktora nekroze tumora glavnog citokina uključenog u zapaljenski proces (TNF-inhibitori). Terapijski uspeh TNF-inhibitora otvorio je vrata i za primenu bioloških lekova uperenih protiv drugih proinflamatornih citokina kao što su IL-6 i IL-1. Terapija biološkim lekovima unela je revoluciju u tretman RA, posebno kod bolesnika koji nisu imali dobar odgovor na konvencionalne sintetske lekove koji menjaju tok bolesti, kao što je metotreksat. Tokom poslednjih nekoliko godina, pojavio se potpuno novi pravac razvoja anti-zapaljenskih lekova. Brojne studije su pokazale da je JAK/STAT signalni put uključen u patogenezu i progresiju RA. Mnogi citokini koriste ovaj put za prenošenje signala u ciljnu ćeliju. Jaki inhibitori su nas uveli u eru „malih molekula” koji imaju sličnu efikasnost kao biološki lekovi.

## **ADVANCES IN THE TREATMENT OF RHEUMATOID ARTHRITIS**

**Mirjana Šefik Bukilica**

Institute of Rheumatology, University of Belgrade-Faculty of Medicine (Serbia)

Rheumatoid arthritis (RA) is a chronic, destructive, inflammatory disease of the peripheral joints. Until late in the 20th century, the therapy of RA relied on the use of drugs that had been developed through empirical approaches without detailed understanding of the molecular mechanisms involved. However, the last decades have witnessed an explosion of our understanding of the inflammatory process and the molecular pathways involved in RA. This led to a new approach of drug development whereby targeted therapies are developed by directly targeting molecules thought to be involved in the inflammatory process. A major approach for the development of targeted therapeutics has been the application of monoclonal antibody technologies for therapeutic purposes. The first targeted therapeutics in RA were antibodies and related molecules interfering with the function of tumor necrosis factor- $\alpha$  (TNF- $\alpha$  or TNF), a major cytokine involved in inflammatory process. The initial advance with anti-TNF biologic therapeutics opened up new avenues for targeting other proinflammatory targets by biologic agents (IL-6 and IL-1). The treatment with biological drugs has revolutionized the therapeutic approach of RA, particularly in patients resistant to standard treatment with *conventional disease-modifying antirheumatic drugs such as methotrexate*. During the last few years, an entirely different direction for developing novel anti-inflammatory agents has emerged. Numerous studies have implicated the Janus kinase/signal transducers and activators of transcription (JAK/STAT) pathway in the pathogenesis and progression of RA. Many cytokines involved in the pathogenesis of RA use JAKs and STATs to transduce intracellular signals. JAK inhibitors introduced us in the era of 'small molecules', which have similar efficacy to the biologic therapies.

## **CARDIOPROTECTION DURING CANCER CHEMOTHERAPY WITH THE USE OF NATURAL ANTIOXIDANTS: REVIEW OF LITERATURE AND RESULTS OF OWN STUDIES**

**Jolanta Łukowicz<sup>1</sup>, Grażyna Peszyńska-Sularz<sup>1</sup>, Anita Piasek<sup>2</sup>,  
Stefan Popadiuk<sup>1</sup>, Agata Kot-Wasik<sup>2</sup>, Monika Janicka<sup>2</sup>,  
Jacek Namieśnik<sup>2</sup>, Włodzimierz Grajek<sup>3</sup>, Agnieszka Bartoszek<sup>1</sup>**

<sup>1</sup>Chemical Faculty, Gdańsk University of Technology, <sup>2</sup>Medical University of Gdańsk, <sup>3</sup>University of Natural Sciences, Poznań (Poland)

Cardiotoxicity is a frequent side effect occurring during cancer chemotherapy, often responsible for long term heart failure in surviving cancer patients. The abnormalities range from small changes in blood pressure and arrhythmias to cardiomyopathy. This type of toxicity has been most widely investigated in the case of anthracyclines, doxorubicin (DOX) in particular, the effective anticancer drugs whose clinical use is limited by cumulative dose-dependent injury to cardiac tissue, often jeopardizing patients' life despite successful cancer eradication. Though best described for DOX, cardiotoxicity as a side effect has been observed during chemotherapy with majority of antineoplastic agents displaying different mechanisms of action: mitoxantrone (cardiomyopathy), fluorouracil (myocardial infarction), cyclophosphamide and vinca alkaloids (cardiac necrosis), trastuzumab (cardiac dysfunction), imatinib mesylate (congestive heart failure).

The cardiotoxicity of anthracyclines, at least in part, is attributed to their ability to redox cycle with molecular oxygen leading to the formation of superoxide radical that initiates cascade of reactive oxygen and nitrogen species. It has therefore been suggested that some phytochemicals with high antioxidant potential, when administered together with DOX (and perhaps other antitumor agents), could decrease the toxic side effects of chemotherapy and reduce the risk of heart failure. Cardioprotective properties have been shown for preparations obtained from such foods as grapes, garlic, tomato, spinach, as well as for melatonin (a hormone synthesized by the pineal gland, but also present in many edible plants), chalcones (precursors of all known flavonoids), some herbal dietary supplements and vitamins A, C, and E. However, in the majority of these studies natural antioxidants were administered i.v., thus in a way typical for pharmacological approach. In contrast, our studies were designed so as to represent truly nutritional approach in which animals undergoing chemotherapy were fed the diet enriched in a particular food item – red beetroot (*Beta vulgaris*) juice (RBJ). In these experiments, we checked whether the dietary intervention with RBJ might have any impact on therapeutic efficacy of DOX. For this purpose, leukaemia L1210 bearing mice were treated with DOX and fed RBJ ad

libitum (instead of water) for 7 or 14 days. DOX was very effective in prolonging survival time of leukaemia bearing mice (ILS about 400%). However, only in groups receiving DOX in combination with RBJ total cures were observed. These were not sporadic events but concerned about 50% of animals.

In accompanying experiments, healthy or leukaemia L1210 bearing mice were fed RBJ ad libitum instead of water for 7 days and then were treated with DOX applied in different schemes. Control mice received water to drink. From control and treated mice, bloods and hearts were collected and analysed for various markers of oxidative insult. In mice fed with RBJ prior to DOX treatment, the damage of DNA in cardiomyocytes and the content of isoprostanes in blood were decreased indicating marked protection offered by the employed dietary intervention. As RBJ on its own had no antitumor effect, one can speculate that the improved outcome of chemotherapy resulted from reduced cardiotoxicity. Our research suggests then that appropriately designed dietary intervention may offer very considerable benefits to cancer patients.

## **VITAMIN D I RIZIK ZA NASTANAK MALIGNIH BOLESTI**

**Aleksandra Zeljković**

Katedra za medicinsku biohemiju, Univerzitet u Beogradu-Farmaceutski fakultet (Srbija)

Savremeno razumevanje značaja koji vitamin D ima u humanom organizmu prevazilazi njegovu dobro poznatu ulogu regulatora metabolizma kalcijuma i fosfata. Prva istraživanja započela su pre više od 80 godina sa postavljanjem hipoteze koja povezuje nisku izloženost sunčevom zračenju sa povećanom incidencijom različitih tipova kancera. S obzirom da koncentracija vitamina D direktno zavisi od izloženosti UVB zracima, predložena je potencijalna protektivna uloga ovog vitamina u kancerogenezi.

Biološki efekti aktivnog oblika vitamina D - kalcitriola ostvaruju se vezivanjem za odgovarajuće receptore u jedru ciljnih ćelija i prevashodno se odnose na regulaciju homeostaze kalcijuma i fosfata. Međutim, danas se zna da osim klasičnog metaboličkog puta postoji i alternativni mehanizam u kojem ključnu ulogu ima hidroksilacija na položajima C-20 i C-22, pod uticajem enzima CYP11A1. Utvrđeno je da 20-hidroksi metaboliti imaju jednak, ili čak i veći kapacitet delovanja u odnosu na kalcitriol. Istraživanja su pokazala da vitamin D ostvaruje protektivne efekte u svim fazama razvoja tumora: od inicijacije do pojave metastaza. Predloženi mehanizmi uključuju uticaj na proliferaciju, diferencijaciju, apoptozu, autofagiju i epitelno-mezenhimnu tranziciju ćelija. Osim toga, smatra se da vitamin D učestvuje u modulaciji mikrookruženja tumora, delujući na angiogenezu, redoks status, inflamaciju i imuni odgovor. Međutim, mnoga pitanja su još uvek otvorena. Ona se pre svega tiču adekvatne procene statusa vitamina D u organizmu, kao i mogućnosti terapijske primene različitih metabolita.

## **VITAMIN D AND RISK FOR CANCER DEVELOPMENT**

**Aleksandra Zeljković**

Department of Medical Biochemistry, University of Belgrade-Faculty of Pharmacy (Serbia)

Contemporary understanding of the role of vitamin D in the human body largely exceeds a traditional perception of this vitamin as a regulator of calcium and phosphate. Eighty years ago, preliminary researches pointed toward the association between low exposure to sunlight and increased incidence of different malignancies. Given that the concentration of vitamin D directly depends on exposure to UVB lights, the potential protective role of this vitamin in tumorigenesis is proposed.

Biological effects of the active metabolite - calcitriol are achieved through binding to the corresponding receptors in the nuclei of the target cells and are predominantly related to the regulation of calcium and phosphate homeostasis. However, it is revealed that there is an alternative metabolic pathway characterized by the CYP11A1 - driven hydroxylation of vitamin D at C-20 and C-22. It has been demonstrated that 20-hydroxy metabolites have the same, or even higher capacity of action, compared to calcitriol. Vitamin D exhibits beneficial effects at all stages of cancerogenesis: from the initiation to the onset of metastases. The proposed mechanisms include modulation of proliferation, differentiation, apoptosis, autophagy, and epithelial-mesenchymal cell transitions. In addition, it is considered that vitamin D provokes changes of tumor microenvironment, by modifying angiogenesis, redox status, inflammation and immune response. However, numerous issues are still open. In particular, adequate assessments of vitamin D status, as well as possibilities for its therapeutic use are still under debate.

## PRIMENA TUMORSKIH MARKERA U KLINIČKOJ PRAKSI I PERSONALIZOVANOJ MEDICINI

Svetlana Ignjatović

Katedra za medicinsku biohemiju, Univerzitet u Beogradu-Farmaceutski fakultet i Centar za medicinsku biohemiju, Klinički centar Srbije (Srbija)

Cirkulišući tumorski markeri (TM) u krvi i drugim tečnostima klinički se koriste već više od 50 godina sa izvesnim uspehom i predstavljaju osnovni deo menadžmenta pacijentima sa kancerom, a takođe su uključeni u veći broj kliničkih vodiča. TM se lako mere u uzorcima seruma/plazme i drugim tečnostima, rezultati su brzo dostupni, a pripadajući troškovi su relativno mali. Iako TM nisu dijagnostički za kancer, postoji mnogo TM koji su prilično specifični za određene kancere - na primer, AFP i hCG kod tumora germinativnih ćelija, kalcitonin u medularnom kanceru tiroidne žlezde, PSA kod kancera prostate i imunoglobulini u limfomu i multiplom mijelomu. Korišćenje TM u menadžmentu pacijenata sa solidnim tumorima ima nekoliko utvrđenih indikacija za upotrebu kod kancera germinativnih ćelija, prostate, jajnika i pankreasa, kao i kolorektalnog, hepatocelularnog i neuroendokrinog kancera. TM su korisni alati u diferencijalnoj dijagnozi, određivanju faze/prognozi, praćenju tretmana, praćenju nadzora i rekurencije pacijenata sa kancerom. Nedavna istraživanja pokazala su poboljšanje dijagnostičkih performansi TM ukoliko se koristi kombinacija više TM kao panel za procenu (npr. kombinovani panel od šest serumskih TM za karcinom pluća), serijsko merenje ili procena TM u određenom vremenskom periodu. Napredak u molekularnoj i ćelijskoj biologiji u protekloj deceniji doveli su do uvođenja novih dijagnostičkih alata u onkologiji koja mere ćelijsku DNK, cirkulišuće tumorske ćelije, egzozome, mikroRNK, vanćelijske vezikule ili razjašnjavaju molekularne događaje kancera na pojedinačnom pacijentu, što dovodi do promene paradigme u načinu na koji se razvijaju antikancer terapije i pacijenti se biraju za specifične ciljane terapije. Optimalan menadžment pacijentima sa kancerom u budućnosti će integrisati nove i uspostavljene alate, uključujući i „tradicionalne“ TM kao test prve linije kako bi se na odgovarajući način pokrenulo dalja obrada i invazivnija dijagnostika.

## **APPLICATION OF TUMOR MARKERS IN CLINICAL PRACTICE AND PERSONALIZED MEDICINE**

**Svetlana Ignjatović**

Department of Medical Biochemistry, University of Belgrade-Faculty of Pharmacy and Center for Medical Biochemistry, Clinical Centre of Serbia (Serbia)

Circulating tumor markers (TMs) in blood and other body fluids have been used clinically for over 50 years with some success and have become an established part of management of cancer patients and are also included in a number of clinical guidelines. TMs are easily measured in serum/plasma samples and other body fluids, the results are rapidly available, and the associated costs are relatively low. Although TMs are not diagnostic for cancer, there are many TMs that are quite specific for certain cancers--for example, AFP and hCG in germ cell tumors, calcitonin in medullary thyroid cancer, PSA in prostatic cancer, and immunoglobulins in lymphoma and multiple myeloma. The use of TMs in the management of patients with solid tumors has several established indications for use in germ cell, prostate, ovarian and pancreatic cancers, colorectal, hepatocellular, and neuroendocrine cancers. TMs are useful tools in differential diagnosis, staging/prognosis, treatment monitoring, surveillance and recurrence monitoring of cancer patients. Recent research has shown improvement diagnostic performance of TMs using a combination of multiple TMs as a panel for assessment (e.g., combined panel of six serum TMs for lung cancer), serial measurement or assess the TM trend over defined period of time. Advances in molecular and cellular biology over the past decade have led to the introduction of novel diagnostic tools in oncology which measure cell-free DNA, circulating tumor cells, exosomes, microRNAs, extracellular vesicles or elucidate the molecular events of tumors on a single patient level, leading to a paradigm shift in how antitumor therapies are developed and patients are selected for specific targeted therapies. The optimal management of cancer patients in future will integrate novel and established tools, including „traditional” TMs as a first-line test to appropriately trigger further workup and more invasive diagnostics.

## **ENERGETSKI BALANS I ULOGA ADIPOCITOKINA U PATOGENEZI MALIGNIH BOLESTI**

**Aleksandra Stefanović**

Katedra za medicinsku biohemiju, Univerzitet u Beogradu-Farmaceutski fakultet (Srbija)

Prevalenca malignih bolesti poslednjih decenija je u stalnom porastu. U našoj zemlji maligne bolesti su odgovorne za oko 20% ukupne smrtnosti i nalaze se na drugom mestu, iza bolesti srca i krvnih sudova. Etiologija malignih bolesti je složena i temelji se, kako na genetskim činiocima, tako i na životnim navikama pojedinaca. Jedan od faktora rizika koji se povezuje sa razvojem, kao i lošom prognozom malignih bolesti jeste gojaznost. Gojaznost poprima karakteristike masovne metaboličke bolesti sa ozbiljnim posledicama na opšte zdravstveno stanje populacije. Rezultati velikog broja studija su nedvosmisleno doveli u vezu gojaznost sa umerenim rizikom za razvoj kolorektalnog karcinoma, karcinoma endometrijuma, bubrega, pankreasa i postmenopauzalnog karcinoma dojke. Hipertrofija adipoznog tkiva u gojaznosti karakteriše se hroničnom inflamacijom niskog stepena. Adipociti i makrofage adipoznog tkiva sekretuju peptide, adipocitokine, od kojih su se posebno proinflamatorni adipocitokini [faktor nekroze tumora  $\alpha$  (TNF - $\alpha$ ), leptin, rezistin, interleukin 6 (IL-6), inhibitor aktivatora plazminogena tip 1 (PAI-1)] pokazali značajnim molekularnim medijatorima složenih patofizioloških puteva kako razvoja, tako i progresije malignih bolesti. Kod razvoja i progresije tumora dojke, bubrega, jajnika, kao i kod kolorektalnog karcinoma poseban značaj ima činjenica da se ovi tumori razvijaju okruženi masnim tkivom. Tokom molekularnih interakcija adipocita sa malignim ćelijama dolazi do promena samih adipocita koji tako izmenjeni sekretuju adipocitokine koji stimulišu adheziju, migraciju i invaziju malignih ćelija. Takođe, ovi izmenjeni adipociti lipolizom oslobođaju slobodne masne kiseline koje maligne ćelije koriste kao značajan izvor energije čime se podstiče progresija i nekontrolisani rast tumora. Razumevanje složenih mehanizama kojim adipozno tkivo utiče na nastanak i razvoj malignih bolesti može dovesti do razvoja potencijalnih novih terapijskih pristupa malignim bolestima.

## **ENERGY BALANCE AND ADIPOCYTOKINES IN CANCER PATHOGENESIS**

**Aleksandra Stefanović**

Department of Medical Biochemistry, University of Belgrade-Faculty of Pharmacy (Serbia)

Over the recent decade, the cancer prevalence rate has been increasing. In our country, cancers are responsible for about 20% of total mortality, right behind cardiovascular disease. Etiology of malignant changes in tissue is complex and it is based on genetic, as well as environmental factors. Obesity is one of the important risk factors connected with the development and bad prognosis for some cancers. According to the results of the great number of studies, obesity is associated with increased risk of developing colorectal, endometrial, renal and postmenopausal breast cancer. Actually, hypertrophy of adipose tissue in obesity is followed by low grade inflammation processes. Proinflammatory adipocytokines [tumor necrosis factor  $\alpha$  (TNF- $\alpha$ ), leptin, rezisitin, interleukin 6 (IL-6), plasminogen activatorinhibitor 1 (PAI - 1)] secreted by adipocytes and macrophages are significant molecular mediators of complex pathophysiological pathways involved in cancer initiation and progression. During the initiation and progression of some tumor types (breast, renal, ovarian, colon) it is important to notice that tumor grow in specific vicinity of adipose tissue. Interactions between cancer cells and adipose tissue lead to specific adipocytes changes. They become cancer-associated adipocytes which are important sources of specific adipocytokines which stimulate the adhesion, migration and invasion of cancer cells. Also, cancer-associated adipocytes ensure fatty acids by lipolysis, and these fatty acids become important energy source for tumor progression and uncontrolled growth. Understanding the complex mechanisms of interaction between adipose tissue and cancer cells is the possible course of the development of potential new therapeutic approaches for treatment of malignant diseases.

## KADMIJUM KAO FAKTOR RIZIKA ZA RAZVOJ KARCINOMA PANKREASA: PODACI IZ STUDIJE NA LJUDIMA, EKSPERIMENTALNIM ŽIVOTINJAMA I ĆELIJSKIM KULTURAMA

**Aleksandra Buha Đorđević<sup>1</sup>, Vesna Matović<sup>1</sup>, Novica Boričić<sup>2</sup>,  
Dejan Radenković<sup>3</sup>, Vladimir Đorđević<sup>3</sup>, David Wallace<sup>4</sup>**

<sup>1</sup>Katedra za toksikologiju „Akademik Danilo Soldatović”, Univerzitet u Beogradu-Farmaceutski fakultet, <sup>2</sup>Institut za patologiju, Univerzitet u Beogradu - Medicinski fakultet, <sup>3</sup>Prva hirurška klinika, Klinički centar Srbije, Beograd (Srbija), <sup>4</sup>Katedra za farmakologiju i toksikologiju, Oklahoma State University Center for Health Sciences Tulsa, Oklahoma (SAD)

Iako intenzivno proučavana, etiologija karcinoma pankreasa i dalje predstavlja nepoznanicu. Neki od utvrđenih faktora rizika ovog oboljenja (poput godina starosti, pušenja) povezani su sa povećanim nivoima kadmijuma (Cd) u organizmu. Kako ovaj toksični metal danas predstavlja jednog od najprisutnijih zagadivača životne i radne sredine, sve je više studija koje ispituju njegovu ulogu u nastanku karcinoma pankreasa. Cilj ovog rada bio je da istraži potencijalnu ulogu Cd u nastanku ovog oboljenja i to sprovodenjem opservacione humane, eksperimentalne i in vitro studije.

Prospektivna studija slučaja obuhvatila je 31 pacijenta sa histološki baziranom dijagnozom karcinoma pankreasa podvrgnutih radikalnoj hirurškoj intervenciji (slučajevi) i 29 slučajnih smrtnih ishoda ili subjekata čije smrti nisu bile posledice karcinoma (kontrole).

Eksperimentalna studija obuhvatila je dve tretirane grupe Wistar pacova (15 i 30 mg/kg t.m.) i netretiranu kontrolnu grupu, žrtvovane 24 sata nakon jednokratnog oralnog izlaganja. Nivoi Cd u mineralizovanim pankreasnim tkivima određeni su atomskom apsorpcionom spektrofotometrijom sa grafitnom kivetom. In vitro studija vršena je na hTERT-HPNE pankreasnim ćelijama izloženim različitim koncentracijama Cd, koje odgovaraju nivoima Cd izmerenim u ljudskom karcinomskom pankreasnom tkivu, i ispitivana je aktivnost enzima kaspaze i nivo oksidativnog stresa.

Sadržaj Cd u tkivu karcinoma značajno se razlikovao od sadržaja u zdravim kontrolama. Neočekivano visoke koncentracije Cd (1,27-18,64 µg/g) izmerene su u karcinomskom tkivu u poređenju sa kontrolnim (0,27-2,50 µg/g). Odnosi šansi za razvoj karcinoma pankreasa bili su 2.79 (95% IP 0.91-8.50) i 3.44 (95% IP 1.19-9.95) u trećem i četvrtom kvartilu distribucije Cd. Eksperimentalna studija potvrdila je tkivo pankreasa kao mesto deponovanja Cd u organizmu. In vitro studija ukazala je na značajno smanjenje aktivnosti kaspaza 3/7 pri izlaganju najvišim koncentracijama Cd, te povećanje oksidativnog stresa koje je bilo izraženije pri višim koncentracijama Cd. Ovo istraživanje daje tri linije dokaza koji upućuju na Cd kao faktor rizika u nastanku karcinoma pankreasa.

## CADMIUM AS A RISK FACTOR FOR PANCREATIC CANCER DEVELOPMENT: HUMAN, ANIMAL AND IN VITRO DATA

**Aleksandra Buha Đorđević<sup>1</sup>, Vesna Matović<sup>1</sup>, Novica Boričić<sup>2</sup>,  
Dejan Radenković<sup>3</sup>, Vladimir Đorđević<sup>3</sup>, David Wallace<sup>4</sup>**

<sup>1</sup>Department of Toxicology „Akademik Danilo Soldatović”, University of Belgrade - Faculty of Pharmacy, <sup>2</sup>Department of Pathology, University of Belgrade - School of Medicine, <sup>3</sup>First Surgical Clinic, Clinical Center of Serbia, Belgrade (Serbia), <sup>4</sup>Department of Pharmacology & Toxicology, Oklahoma State University Center for Health Sciences Tulsa, Oklahoma (USA)

Although profoundly studied, etiology of pancreatic cancer (PC) is still rather scarce. Some of established risk factors of PC are connected to an increased cadmium (Cd) body burden (age, smoking, etc). This toxic metal is nowadays regarded as one of the most abundant occupational and environmental pollutants. Hence, an increasing number of studies are investigating its role in PC and the data is somewhat conflicting. The aim of this study was to investigate Cd possible role in PC development by conducting human observational, experimental and in vitro studies.

The case-control prospective study included 31 patients with a histologically based diagnosis of PC subjected to radical surgical intervention as cases and 29 accidental fatalities or subjects who died of a nonmalignant illness as controls. Animal study included two treated groups of Wistar rats (15 and 30 mg Cd/kg b.w) and untreated control group, sacrificed 24 hours after single oral exposure. Cadmium content in digested pancreatic tissues was assessed by GFAAS. In in vitro study pancreas hTERT-HPNE cells were exposed to different Cd concentrations corresponding to levels measured in human cancerous pancreatic tissue and oxidative stress and caspase activity were determined.

Cd content in cancer tissue significantly differed from the content in healthy controls. Unexpectedly high Cd concentrations (1.27-18.64 $\mu$ g/g) were found in cancerous tissue in comparison to controls (0.27-2.50 $\mu$ g/g). Odds ratio levels for PC development were 2.79 (95% CL 0.91-8.50) and 3.44 (95% CL 1.19-9.95) in the third and fourth quartiles of Cd distribution, respectively. Animal study confirmed Cd deposition in pancreatic tissue. Exposure of hTERT-HPNE cells to highest Cd concentration resulted in significant reduction in caspase 3/7 activity while observed increase in oxidative stress was concentration dependent. This study presents three different lines of evidence pointing towards Cd as an agent responsible for the development of PC.

## **OLIVE BIOACTIVE COMPOUNDS: CHEMISTRY AND PHARMACOLOGY**

**Apostolis Angelis, Leandros A. Skaltsounis**

Department of Pharmacognosy & Natural Products Chemistry, Faculty of Pharmacy, University of Athens (Greece)

The olive tree, closely connected to the Mediterranean region has provided a wealth of goods. Research on the olive has started early but it has proven inexhaustible revealing mainly a vast array of nutritional and health properties. Apart from olive oil and table olives, the by-products coming from olive processing industry have been proven attractive materials for research. The aim of this communication is to present a holistic research strategy towards the multifaceted exploitation of the olive tree including activities such as extraction, fractionation, isolation, analysis of olive tree products as well as investigation of processes related to olive industry and valorization of by-products. The main products of the olive tree, olive oil and table olives as well as by-products such as leaves, paste, mill wastes and table olive wastewater have been used as sources for the recovery of valuable secondary metabolites. This has been performed with conventional techniques and also by adsorptive resin technology. In addition standardized enriched fractions have been prepared with various techniques, such as MPLC, HPLC, and CCC. Isolation of promising lead compounds with emphasis to olive polyphenol oleuropein (leaves), hydroxytyrosol & tyrosol (olive oil, by-products), oleacein & oleocanthal (olive oil) and lactones (by-products), has been achieved. Additionally advanced analytical techniques and methodologies (UPLC/HPLC-DAD, HPLC-DAD-HR/MSn, and HPTLC) have been developed and applied for the qualitative and quantitative determination of secondary metabolites in all the above mentioned materials. The lab scale processes have been also adapted to pilot scale systems. The biological profile and the therapeutic potential of olive extracts and compounds is explored and supported by several in vitro and in vivo studies while their possible application as nutraceuticals, dietary supplements and cosmetics is also investigated.

## HERBA CITRALNOG HEMOTIPA PANONSKOG TIMIJANA KAO POTENCIJALNO NOVA BILJNA LEKOVITA SIROVINA

Zoran Maksimović

Katedra za farmakognoziju, Univerzitet u Beogradu-Farmaceutski fakultet  
(Srbija)

Panonski timijan (*Thymus pannonicus* All. Lamiaceae) je rasprostranjen u srednjoj i istočnoj Evropi na sušnim livadama i kamenjarima. U Srbiji raste pretežno u Vojvodini. Literaturni podaci ukazuju na značajne razlike u sastavu etarskog ulja samoniklog panonskog timijana. Potvrđeno je postojanje hemotipova; npr. timolnog i citralnog. Stabilna populacija citralnog hemotipa locirana je u Srbiji samo na Vršačkim planinama. Herba ovog hemotipa se, u južnom Banatu, koristi za pripremanje čajnog napitka specifičnog i prijatnog mirisa koji podseća na limun, ali i protiv nekih respiratornih i digestivnih oboljenja. Ovo je bio i osnov za detaljnije ispitivanje ove biljne vrste sa područja Vršačkih planina.

Hromatografskom analizom (GC FID/MS; HPLC), ispitivana je polimorfnost samoniklih populacija citralnog hemotipa panonskog timijana i procenjivan uticaj ekoloških faktora na njihove morfološke, anatomske i hemijske karakteristike. Testirana je antimikrobnja, antioksidantna, antitumorska i hepatoprotektivna aktivnost. Izvedeni su ogledi planskog gajenja i odabранe linije sa poželjnim osobinama. Utvrđeni su parametri kvaliteta herbe panonskog timijana, kao nove biljne sirovine.

Rezultati ukazuju da se panonski timijan sa Vršačkih planina može smatrati dobrim izvorom biljne sirovine bogate citralom. Najvažniji sastojci polarnih ekstrakata herbe samoniklog panonskog timijana bile su fenolske kiseline (rozmarinska, salvianolna) i flavonoidi (glukuronidi luteolina i apigenina). Vodeni ekstrakt je ispoljio umereni antioksidantni efekat in vivo, uz značajno smanjenje intenziteta lipidne peroksidacije i održavanje fizioloških koncentracija glutationa. Prema ćelijama Erlihovog ascitnog tumora kod miša, vodeni ekstrakt je delovao citotoksično, ispoljivši prooksidantni efekat kojim je indukovana apoptoza. Uočena je značajna antimikrobnja aktivnost prema testiranim mikroorganizmima, a naročito prema *Candida albicans*. Etarsko ulje i metanolni ekstrakt ispoljili su izuzetnu inhibitornu aktivnost prema kliničkim izolatima *Helicobacter pylori* rezistentnim na metronidazol i klaritromicin. Oplemenjivanjem, dobijena je sorta željenih tehnoloških svojstava koja daje veći prinos biljne mase, veći i stabilniji sadržaj etarskog ulja u odnosu na samoniklu biljku.

Citralni hemotip panonskog timijana se pokazao kao izdašan biološki izvor droge *Thymi pannonicici* herba. Zahvaljujući visokom sadržaju polifenolnih jedinjenja i etarskog ulja bogatog citralom, kao i ispoljenoj antioksidantnoj, antimikrobnoj i antitumorskoj aktivnosti, ona predstavlja potencijalno novu biljnu lekovitu sirovinu, koja bi se mogla primenjivati u savremenoj fitoterapiji.

*Istraživanje je podržalo Ministarstvo prosvete, nauke i tehnološkog razvoja (Projekat TR 31089 I ON 173021).*

# THE HERB OF PANNONIAN THYME CITRAL CHEMOTYPE AS POTENTIALLY NEW HERBAL RAW MATERIAL WITH MEDICINAL PROPERTIES

Zoran Maksimović

Department of Pharmacognosy, University of Belgrade-Faculty of Pharmacy  
(Serbia)

Pannonian thyme (*Thymus pannonicus* All. Lamiaceae) is distributed in central and eastern Europe, over dry meadows, grasslands and rocks. In Serbia, it can be found mostly in Vojvodina province. Reference data reveals high variability in the composition of wild-growing pannonian thyme. A number of chemotypes (e.g. thymol and citral) were confirmed. In Serbia, a stable population of citral chemotype has been found at Mt. Vršačke planine only. In southern Banat, dried herb of this chemotype is used to make tasty and refreshing herbal teas with peculiar and pleasant lemon-like scent; also, against some respiratory and gastrointestinal disorders. This provided the ground for further and detailed investigations on this plant species from Mt. Vršačke planine.

By chromatographic analysis (GC FID/MS; HPLC), polymorphism within wild populations of this species was studied, as well as dominant ecological factors that influenced their morphologic, anatomic and chemical properties. Antimicrobial, antioxidant, antitumour and hepatoprotective activity were also tested. Planned cultivation was attempted and lines with desirable traits were chosen. Parameters of quality for pannonian thyme herb as a new herbal raw material were defined.

The results indicate that pannonian thyme from Mt. Vršačke planine could be considered as a plentiful source of herbal raw material rich in citral. Principal constituents of polar extracts of wild growing pannonian thyme were phenolic acids (rosmarinic, salvianolic) and flavonoids (luteolin and apigenin glucuronides). Aqueous extract expressed moderate antioxidant effect in vivo, along with a significant decrease of lipid peroxidation intensity and preservation of physiological levels of glutathione. Against Ehrlich ascites tumor cells in mice, aqueous extract expressed significant cytotoxic activity, through prooxidant effect that induced apoptosis. Significant antimicrobial activity against tested microorganisms was observed; in particular, against *Candida albicans*. Essential oil and methanolic extract expressed remarkable inhibitory activity against clinical strains of *Helicobacter pylori* resistant to metronidazol and claritromycine. Finally, a variety with desired technological characteristics was bred, yielding superior biomass quantity, as well as higher and more stable essential oil contents in comparison to wild-growing plants.

Pannonian thyme citral chemotype appeared to be a plentiful biological source of herbal substance *Thymi pannonicici herba*. Owing to high contents of polyphenols and essential oil rich in citral, as well as to expressed antioxidant, antimicrobial and antitumor activity, it is a potentially new herbal raw material that could be used in contemporary phytotherapy.

*The study was supported by the Ministry of Education, Science and Technological Development (Projects ON 173021 and ON 173021).*

## PROCENA FARMAKOLOŠKE AKTIVNOSTI ODABRANIH VRSTA FAMILIJE ERICACEAE

Dragana Pavlović

Katedra za farmaciju, Univerzitet u Nišu-Medicinski fakultet (Srbija)

Pored vrste *Arctostaphylos uva-ursi*, najpoznatijeg predstavnika familije Ericaceae, i druge vrste ove familije se pominju u tradicionalnoj medicini. Cilj studije je procena i poređenje farmakološke aktivnosti pet vrsta familije Ericaceae sa Balkanskog poluostrva: *Arbutus unedo L.*, *Bruckenthalia spiculifolia*, *Calluna vulgaris*, *Erica arborea* i *Erica carnea*.

Farmakološko ispitivanje suvih etanolnih ekstrakata je uključilo procenu antioksidativne, antimikrobne, antiiritantne, spazmolitičke i antiinflamatorne aktivnosti. Svi uzorci su ispoljili odličnu antioksidativnu aktivnost u 4 komplementarna test sistema. Najveću sposobnost uklanjanja slobodnih radikala i redukcije jona gvožđa su pokazali ekstrakti *A. unedo* (verovatno usled prisustva arbutina koji nije detektovan u ostalim uzorcima), dok je ekstrakt herbe vrste *B. spiculifolia* bio najjači inhibitor lipidne peroksidacije. Antimikrobnu aktivnost ispitivana na 10 bakterijskih sojeva je bila veoma slaba. Hidrogelovi formulirani sa po 2 % jednog od ispitivanih ekstrakata su ispoljili pozitivne efekte na kožu dobrovoljaca. Svi uzorci su značajno snižavali nivo iritacije i vraćali pH i nivo hidriranosti kože na vrednosti pre iritacije. Ekstrakt vrste *A. unedo* pokazuje značajnu spazmolitičku aktivnost, posredovanu inhibicijom kalcijumovih kanala, pri čemu je ekstrakt listova sakupljenih u Grčkoj ispoljio snažniji efekat od listova sakupljenih u Crnoj Gori. Ekstrakt vrste *B. spiculifolia* (0.001–0.3 mg/ml) opušta spontane i indukovane kontrakcije izolovanog ileuma i distalnog kolona, dok inhibicija kontraktilnosti fundusa, duodenuma, jejunuma i proksimalnog kolona nije statistički značajna. Efekti ekstrakta vrste *B. spiculifolia* (50, 100 i 200 mg/kg per os) u karageninom-indukovanom edemu šapice pacova su značajni, dozno-zavisni i uporedivi sa efektima indometacina (4 mg/kg per os). Etilacetatna frakcija i etanolni ekstrakt herbe su ispoljile najsnažnije antioksidativno i antiinflamatorno dejstvo od testiranih etanolnih ekstrakata različitih organa vrste *B. spiculifolia* (koren, list, herba i cvet) i frakcija metanolnog ekstrakta njene herbe. Rezultati izvedenih istraživanja ukazali su da je *B. spiculifolia* ispoljila odlične efekte u sprovedenim farmakološkim testovima.

## **PHARMACOLOGICAL SCREENING OF SELECTED SPECIES FROM ERICACEAE FAMILY**

**Dragana Pavlović**

Department of Pharmacy, University of Niš-Faculty of Medicine (Serbia)

Beside *Arctostaphylos uva-ursi*, the famous member of the Ericaceae family, other species of this family are also mentioned in traditional medicine. The present study estimates and compares pharmacological activity of five Ericaceae species native to Balkan Peninsula: *Arbutus unedo*, *Bruckenthalia spiculifolia*, *Calluna vulgaris*, *Erica arborea* and *Erica carnea*.

Pharmacological screening of dry ethanol extracts included the studies of antioxidant, antimicrobial, antiirritant, spasmolytical and antiinflamatory activity. All samples exerted excellent antioxidant effects in four complementary test systems. The highest scavenging activity and feri-ion reduction were obtained with *A. unedo* extracts (probably due to the presence of arbutin, which is absent in other samples). *B. spiculifolia* herb extract was the most potent inhibitor of lipid peroxidation. Antimicrobial activity against 10 tested strains of bacteria was generally weak. Hydrogels, each containing 2 % of one investigated extract, showed positive effects on the human skin. All samples significantly decreased the skin irritation level and reversed the pH of the skin disturbed by pre-irritation alongside with its hydration. *A. unedo* extracts showed significant spasmolytic activity (mediated through calcium channel inhibition) in isolated rat ileum, with stronger effects of extract from leaves collected in Greece. *B. spiculifolia* extract (concentration range 0.001–0.3 mg/ml) decreased spontaneous and induced contractions of ileum and distal colon while there were no statistically significant inhibition of contractions of fundus, duodenum, jejunum and proximal colon. *B. spiculifolia* extract (50, 100 and 200 mg/kg/day per os) showed significant and dose-dependent activity comparable to that of indomethacin (4 mg/kg per os) in carrageenan-induced rat paw oedema model. Ehylacetat fraction and ethanol extract of herb demonstrated the strongest antioxidant and anti-inflammatory activities among tested ethanol extracts of different *B. spiculifolia* organs (root, leaf, herb and flower) and fractions of methanol extract of its herb. The results of the performed studies indicated that *B. spiculifolia* exhibited very good effects in the conducted pharmacological tests.

## NOVIJE INFORMACIJE O LEKOVITOM POTENCIJALU VRSTA RODA HYPERICUM

**Nebojša Kladar, Neda Gavarić, Biljana Božin**

Katedra za farmaciju, Univerzitet u Novom Sadu - Medicinski fakultet (Srbija)

Rod Hypericum obuhvata više od 500 vrsta klasifikovanih u 36 filogenetskih sekcija. Tokom istorije, ali i u današnje vreme kantarion (*Hypericum perforatum*, *Hypericaceae*) je najčešće proučavan predstavnik roda. Međutim, u poslednje tri decenije zabeležen je porast istraživačkog interesovanja i za ostale predstavnike roda *Hypericum*. Uzimajući u obzir aktivne principe *H. perforatum* koji uključuju jedinjenja iz klase naftodiantrona, floroglucinola, fenola, flavonoida, biflavonoida i ksantona, može se zaključiti da predstavnici filogenteski mlađih sekcija roda pokazuju značajan stepen sličnosti u pogledu kvalitativnog hemijskog profila, dok je sadržaj određenih komponenti izuzetno podložan varijacijama. Međutim, pored uočenih sličnosti između različitih vrsta u pogledu hemijskog sastava, ne smeju se zanemariti nove izolovane komponente iz različitih predstavnika sa obećavajućim biološkim potencijalom. Generalno, veliki broj vrsta roda *Hypericum* je okarakterisan visokim sadržajem fenolnih i flavonoidnih jedinjenja, što u značajnoj meri objašnjava snažan antioksidantni potencijal, uglavnom pokazan u in vitro istraživanjima. Takođe, nadaleko poznat antidepresivni efekat kantariona je prema poslednjim hipotezama posledica sinergističkog delovanja nekoliko klase jedinjenja i nije isključivo odlika *H. perforatum*. Trenutna istraživanja ukazuju na snažan potencijal predstavnika roda *Hypericum* da inhibišu biološki aktivne enzime što bi moglo biti od koristi u terapiji određenih patoloških stanja kao što su Alchajmerova i Parkinsonov bolest, odnosno dijabetes tip 2. Takođe, uzimajući u obzir porast incidence kancera na svetskom nivou, značajni su rezultati zabeleženi za ekstrakte predstavnika roda *Hypericum*, ali i izolovana jedinjenja. Naime, istraživanja ukazuju na njihovo antiproliferativno delovanje i mogućnost inhibicije procesa angiogeneze kada su primenjeni u obliku monoterapije ili ko-terapije sa konvencionalnim antikancerskim lekovima.

## **UPDATES ON THERAPEUTIC POTENTIAL OF SPECIES FROM GENUS HYPERICUM**

**Nebojša Kladar, Neda Gavarić, Biljana Božin**

Department of Pharmacy, University of Novi Sad - Faculty of Medicine (Serbia)

The genus *Hypericum* includes more than 500 species classified into 36 phylogenetic sections. During history, as well as nowadays, St John's wort (*Hypericum perforatum*, *Hypericaceae*) is the most studied species of the genus. However, in the last three decades an increasing trend of researches concerning other *Hypericum* species can be noticed. Regarding the identified active principles of *H. perforatum*, which include compounds belonging to classes of naphtodianthrones, phloroglucinols, phenolics, flavonoids, biflavonoids and xantones, it can be noticed that species belonging to phylogenetically younger sections mostly share the qualitative chemical profile of *H. perforatum*, while the quantities of the specific compounds are subjective to significant variations. However, beside the observed interspecies chemical similarities, new valuable compounds with biological activities isolated from different representatives must not be neglected. Generally, a large number of *Hypericum* species has been suggested as a rich source of phenolic and flavonoid compounds which can explain strong antioxidant potential, mostly recorded in vitro. Also, the well-known antidepressant activity of St. John's wort is by latest hypotheses a result of synergistic action of several classes of compounds and is not exclusively related to *H. perforatum*. Current studies suggest strong potential of *Hypericum* species to inhibit biologically important enzymes, which has the power to be utilized in treatment of certain pathological conditions such as Alzheimer's and Parkinson's disease and diabetes type 2. Furthermore, considering the worldwide growing incidence of cancer, the promising effects of *Hypericum*-based extracts, as well as isolated compounds, have been recorded. Specifically, data suggest their antiproliferative and anti-angiogenesis activities when applied as monotherapy, or co-therapy with conventional antica ncer drugs.

## EFEKTI METANOLNIH EKSTRAKATA DVE BILJNE VRSTE IZ FLORE SRBIJE NA ISHEMIJSKO-REPERFUZIONU POVREDU IZOLOVANOG SRCA PACOVA: UTICAJ OKSIDACIONOG STRESA

**Nevena Jeremić<sup>1</sup>, Jovana Bradić<sup>1</sup>, Vladimir Živković<sup>2</sup>, Ivan Srejović<sup>2</sup>,**  
**Jovana Jeremić<sup>1</sup>, Tamara Nikolić-Turnić<sup>1</sup>, Vladimir Jakovljević<sup>2</sup>**

<sup>1</sup>Odsek za farmaciju, Univerzitet u Kragujevcu - Fakultet Medicinskih nauka,

<sup>2</sup>Katedra za fiziologiju, Univerzitet u Kragujevcu - Fakultet Medicinskih nauka  
(Srbija)

Pored široke tradicionalne primene ivanjskog cveća (*Galium verum* L.- *G. verum*) i sremuša (*Allium ursinum* L.- *A. ursinum*) u terapiji brojnih bolesti i stanja njihovi efekti na funkciju srca i redoks status jošuvek nisu u potpunosti razjašnjeni. Cilj naše studije bio da ispita efekte metanolnih ekstrakata *G. verum* i *A. ursinum* na ishemisko-reperfuziona oštećenja na izolovanom srcu pacova.

30 muških Wistar albino pacova su nasumično podeljeni u tri grupe: kontrolnu i grupe *G.verum* i *A. ursinum*, koje su obuhvatile životinje tretirane sa 500 mg/kg telesne mase metanolnog ekstrakta *G. verum* i *A.ursinum* per os tokom perioda od 4 nedelje, redom. Nakon završenog tretmana srca životinja iz svih grupa su bila izolovana i retrogradno perfundovana prema Langendorff tehnicu pri konstantnom perfuzionom pritisku od 70 cm H<sub>2</sub>O. Nakon perioda stabilizacije srca su bila podvrgнутa ishemiji u trajanju od 20minuta, koja je praćena reperfuzijom u trajanju od 30 minuta. Registrovani su parametri srčane funkcije uključujući maksimalnu i minimalnu stopu razvoja pritiska, sistolni i dijastolni pritisak u levoj komori i srčanu frekvencu. Koronarni protok je meren floumetrijski. Nivoi superoksid anjon radikala, vodonik peroksida, nitrita i indeks lipidne peroksidacije (meren kao thiobarbituric acid reactive substances- TBARS) određivani su spektrofotometrijski u koronarnom venskom efluentu. Naši rezultati su pokazali da je tretman metanolnim ekstraktima *G. verum* i *A. ursinum* tokom 4 nedelje očuvao kontraktilnusnagu srca, sistolnu i dijastolnu funkciju i umanjio produkciju prooksidanasa.

Obećavajući potencijal *G. verum* i *A.ursinum* u ovoj studiji na modelu farmakološkog prekondicioniranja može biti polazna osnova za buduća istraživanja koja bi u potpunosti otkrila njihove efekte na funkciju srca i redoks ravnotežu na različitim modelima ishemisko-reperfuzionog oštećenja.

## EFFECTS OF OF METHANOL EXTRACTS OF TWO PLANT SPECIES FROM THE FLORA OF SERBIA ON ISCHEMIC/REPERFUSION INJURY OF ISOLATED RAT HEART: ROLE OF OXIDATIVE STRESS

**Nevena Jeremić<sup>1</sup>, Jovana Bradić<sup>1</sup>, Vladimir Živković<sup>2</sup>, Ivan Srejović<sup>2</sup>,**  
**Jovana Jeremić<sup>1</sup>, Tamara Nikolić-Turnić<sup>1</sup>, Vladimir Jakovljević<sup>2</sup>**

<sup>1</sup>Department of Pharmacy, University of Kragujevac - Faculty of Medical Sciences,

<sup>2</sup>Department of Physiology, University of Kragujevac - Faculty of Medical Sciences (Serbia)

Beside the widespread traditional use of *Galium verum* (G. verum) and *Allium ursinum* (A. ursinum) in the treatment of numerous diseases and conditions, their effects on heart function and redox status has still not been fully clarified. The aim of our study was to examine the effects of methanol extract of G. verum and A. ursinum on ischemia-reperfusion injury in isolated rat heart.

Thirty male Wistar albino rats were randomly divided into three groups: control and G. verum and A. ursinum groups, which included animals treated with 500 mg/kg body weight of the methanol extract of G. verum and A. ursinum per os for 4 weeks, respectively. At the end of the treatment hearts from animals in all groups were excised and retrogradely perfused according to the Langendorff technique at constant perfusion pressure of 70 cm H<sub>2</sub>O. After stabilization period hearts were subjected to 20 minutes ischemia followed by 30 minutes reperfusion. The parameters of cardiac function including the maximum and minimum rate of pressure development, systolic and diastolic left ventricular pressure and heartrate were registered. Coronary flow was measured flowmetrically. Levels of superoxide anion radical, hydrogen peroxide, nitrites and index of lipid peroxidation (measured as thiobarbituric acid reactive substances-TBARS) were determined spectrophotometrically in coronary venous effluent. Our results demonstrated that treatment with methanol extracts of G. verum and A. ursinum preserved cardiac contractility, systolic and diastolic function and diminished production of pro-oxidants. Promising potential of G. verum and A. ursinum in the present study in a model of pharmacological preconditioning may be a starting point for future researches, which would fully reveal their effects on cardiac function and redox status in various models of I/R injury.

## OPTIMIZACIJA EKSTRAKCIJE PLODA ARONIJE, ARONIA MELANOCARPA (MICHX.) ELLIOTT, MIKROINKAPSULACIJA EKSTAKTA I ISPITIVANJE BIOLOŠKIH AKTIVNOSTI EKSTAKTA

**Nada Ćujić<sup>1</sup>, Katarina Šavikin<sup>1</sup>, Gordana Zdunić<sup>1</sup>, Branko Bugarski<sup>2</sup>,**  
**Nevena Mihailović-Stanojević<sup>3</sup>, Svetlana Ibrić<sup>4</sup>**

<sup>1</sup>Odsek za farmaceutska istraživanja i razvoj, Institut za proučavanje lekovitog bilja „Dr Josif Pančić“, <sup>2</sup>Odsek za hemijski inženjeriing, Univerzitet u Beogradu - Tehnološko-metalurški fakultet, <sup>3</sup>Odesek za kardiovaskularnu fiziologiju, Univerzitet u Beogradu – Institut za medicinska istraživanja, <sup>4</sup>Katedra za farmaceutsku tehnologiju i kozmetologiju, Univerzitet u Beogradu – Farmaceutski fakultet (Srbija)

Aronija je jedan od najbogatijih izvora polifenolnih jedinjenja (antocijani, proantocijanidini, fenolne kiseline, flavanoli). Upravo ovi polifenoli predstavljaju bogati potencijal prirodnih antioksidanasa i njihova primena može imati povoljne efekte na smanjenje rizika od nastanka kardiovaskularnih, malignih i različitih degenerativnih bolesti. Primena ekstrakata bogatih polifenolima ima i niz ograničenja, a pre svega nestabilnost. Cilj rada je bila optimizacija ekstrakcije polifenola iz suvog ploda aronije, mikroinkapsulacija ekstrakta u cilju poboljšanja stabilnosti i bioraspoloživosti, kao i in vivo ispitivanje antihipertenzivnog delovanja ekstrakta na modelu esencijalne hipertenzije.

Eksperimentalni rad sadrži tri faze. 1) Optimizacija ekstrakcije i primena eksperimentalnog dizajna za dobijanje ekstrakta sa najvećom količinom polifenola. 2) Ekstrakt sa najvećom količinom polifenola je mikroinkapsuliran, elektrostatičkom ekstruzijom i sušenjem raspršivanjem. Mikročestice su fizičkohemijski i biofarmaceutski okarakterisane: efikasnost inkapsulacije, ispitivanje brzine oslobađanja polifenola, FTIR, SEM, veličina čestica (optički mikroskop i Mastersizer). 3) Ispitivanje antihipertenzivnog delovanja ekstrakta. Kod eksperimentalnih grupa određivani su sistemski i regionalni hemodinamski parametri, stepen lipidne peroksidacije (TBARS u plazmi i eritrocitima) i aktivnosti enzima antioksidativne zaštite.

Ekstrakt sa najvećom količinom aktivnih principa, dobijen je maceracijom sa 50% etanolom, 1:20 odnosom droga-rastvarač, 0,75 mm stepenom usitnjenošti, u trajanju od 60 minuta. HPLC analiza je potvrdila da se pod istim ekstrakcionim uslovima postiže najveći prinos polifenola. Obe tehnike za mikroinkapsulaciju ekstrakta su pogodne za proizvodnju malih i uniformih mikročestica, sa produženim oslobađanjem polifenola, kao efikasni sistemi za isporuku i očuvanje stabilnosti. Četvoronedeljnou primenom ekstrakta kod SHR pacova, značajno je redukovana sistolni pulsni pritisak, koji su povezani sa povećanom diurezom. Redukovan je TBARS u plazmi i eritrocitima, kao i lipidna peroksidacija, usled smanjenog oksidativnog stresa kod životinja koje su primale ekstrakt.

Ekstrakt aronije, sa maksimalnom količinom aktivnih principa se može koristiti kao blag antihipertenziv u početnim fazama bolesti ili kao dopuna konvencionalnoj antihipertenzivnoj terapiji. Mikroinkapsulacione metode su se pokazale kao efikasne u očuvanju stabilnosti ekstrahovanih jedinjenja.

# **OPTIMIZATION OF CHOKEBERRY EXTRACTION, ARONIA MELANOCARPA (MICHX.) ELLIOTT, EXTRACT MICROENCAPSULATION AND BIOLOGICAL ACTIVITIES**

**Nada Ćujić<sup>1</sup>, Katarina Šavikin<sup>1</sup>, Gordana Zdunić<sup>1</sup>, Branko Bugarski<sup>2</sup>,**  
**Nevena Mihailović-Stanojević<sup>3</sup>, Svetlana Ibrić<sup>4</sup>**

<sup>1</sup>Department for Pharmaceutical Research and Development, Institute for Medicinal Plant Research „Dr. Josif Pančić“; <sup>2</sup>Department of Chemical Engineering, University of Belgrade - Faculty of Technology and Metallurgy;

<sup>3</sup>Department for Cardiovascular Physiology, University of Belgrade - Institute for Medical Research, <sup>4</sup>Department of Pharmaceutical Technology and Cosmetology, University of Belgrade - Faculty of Pharmacy (Serbia)

Chokeberry is one of the richest sources of polyphenolic compounds (anthocyanins, proanthocyanidins, phenolic acids, flavanols). These polyphenols represent the rich potential of natural antioxidants and their application can have beneficial effects on reducing the risk of developing cardiovascular, malignant and various degenerative diseases. The use of extracts rich in polyphenols has a number of constraints, and above all, instability. The aim of the paper was to optimize the extraction of polyphenol from dry chokeberry, microencapsulation of the extract in order to improve stability and bioavailability, as well as in vivo testing the antihypertensive activity of the extract on the model of essential hypertension.

The experimental work consists of three phases. 1) Optimization of the extraction and application of experimental design to produce the extract with the highest amount of polyphenols. 2) Extract with the highest polyphenols content was encapsulated, by the electrostatic extrusion and spray drying. Microparticles were physicochemical and biopharmaceutical characterized: encapsulation efficiency, release rate of encapsulated polyphenols, FTIR, SEM, particle sizes (optical microscope and Mastersizer). 3) Antihypertensive effect examination of the extract. In experimental groups were determined the systemic and regional hemodynamic parameters, lipid peroxidation by TBARS method in plasma and erythrocytes and antioxidative enzymes activities.

Extract with the greatest amount of active principles was obtained by maceration using 50% ethanol, 1:20 solid-solvent ratio, 0.75 mm particle size, during 60 minutes. HPLC analysis confirmed that with the same, selected extraction conditions was achieved the highest yield of polyphenols. Both microencapsulation techniques were suitable technique for small and uniform particles production, as efficient systems for the prolonged polyphenols delivery and increased stability. A four week extract administration in SHR rats significantly reduced systolic and pulse pressure, associated with increased diuresis. Plasma and erythrocytes TBARS were decreased, as well as lipid peroxidation as a consequence of decreased oxidative stress in the extract treated experimental group. Chokeberry extract, with a maximum amount of active principles, could be used as a mild antihypertensive agent in the early stages of disease or as a supplement to conventional antihypertensive therapy. Microencapsulation methods have been proven to be effective in preserving the stability of the extracted compounds.

## **GENERIČKI LEKOVI OD PODNOŠENJA ZAHTEVA DO ODOBRENJA SAŽETKA KARAKATERISTIKA LEKA**

**Branka Brzaković**

Agencija za lekove i medicinska sredstva Srbije (Srbija)

Generički lekovi se često razmatraju u okviru strategija za smanjenje troškova zdravstvene zaštite. Razvoj generičkog leka ima za cilj dobijanje leka ekvivalentnog već registrovanom leku koji se naziva referentni lek. Aktivna supstanca je ista, koristi se u istoj dozi za lečenje iste bolesti, farmaceutski oblik je isti, dok se pomoćne supstance, ime, izgled i pakovanje mogu razlikovati. Generički lekovi se proizvode prema istim standardima kvaliteta kao i drugi lekovi i poštovanje smernica Dobre proizvođačke prakse (GMP) je neophodno.

Posle podnošenje zahteva za dobijanje dozvole za stavljanje leka u promet, regulatorno telo kao što je u našoj zemlji Agencija za lekove i medicinska sredstva Srbije, spovodi stručnu procenu kvaliteta, efikasnosti i bezbednosti leka. Proizvođač generičkog leka u obavezi je da dostavi potpunu dokumentaciju o kvalitetu ovog leka. U većini slučajeva neophodno je priložiti i podatke o studiji biološke ekvivalentnosti, čiji rezultati treba da pokažu da generički lek postiže iste nivoje aktivne supstance u telu kao referentni lek, dok se sprovođenje toksikoloških, farmakoloških i kliničkih studija po pravilu ne zahteva za generički lek. Informacije o generičkom leku sadržane u Sažetku karakteristika leka, Uputstvu za lek i nacrtu pakovanja su iste kao za referentni lek, sa izuzetkom indikacija ili farmaceutskog oblika, koji su još uvek pokriveni patentnom zaštitom. Posle registracije, regulatorno telo će nastaviti da prati bezbednost odobrenog generičkog leka.

## **GENERIC MEDICINAL PRODUCTS FROM APPLICATION TO FINAL SUMMARY OF PRODUCT CHARACTERISTICS**

**Branka Brzaković**

Medicines and Medical Devices Agency of Serbia (Serbia)

Generic medicinal products are often regarded as one of strategies to reduce healthcare expenses. They are developed to be equivalent to a medicine that has already been authorised (the reference medicine) having the same active substance, used at the same doses to treat the same diseases, and the same pharmaceutical form. Excipients, name, appearance and packaging can be different. Generic medicines are manufactured according to the same quality standards as all other medicines and good manufacturing practices (GMP) are required.

After application for marketing authorisation, a regulatory authority such as Medicines and Medical Devices Agency of Serbia, will conduct a scientific evaluation of the medicine's efficacy, safety and quality. A producer of a generic medicine needs to provide complete information on the quality of the medicine. In most cases data from a bioequivalence study are required to demonstrate that the generic medicine produces the same levels of the active substance in the body as the reference medicine. For a generic medicinal product it is not necessary to provide the results of toxicological and pharmacological tests or the results of clinical trials. The same information will appear in the product information (the summary of product characteristics, the labelling and the package leaflet) of the generic medicinal product, as in the product information of the reference medicine, except for indications or dosage forms still covered by patent law. After authorization, the regulatory authorities continue to monitor the safety of generic medicines.

## **BIOEQUIVALENCE REQUIREMENTS FOR LOCALLY ACTING DOSAGE FORMS**

**Alfredo García Arieta**

Agencia Española de Medicamentos y Productos Sanitarios (España)/Spanish Agency of Medicines and Medical Device (Spain)

The requirements to demonstrate equivalence between locally acting medicinal products has evolve in the European Union since the overarching guideline „Clinical requirements for locally applied, locally acting products containing known constituents” published in 1995 and the new guidelines for:

Orally inhaled products („Requirements for clinical documentation for orally inhaled products (OIP) including the 96 requirements for demonstration of therapeutic equivalence between two inhaled products for use in the treatment of Asthma and Chronic Obstructive Pulmonary Disease (COPD)”), which is presently under revision.

Gastrointestinal products („Guideline on equivalence studies for the demonstration of therapeutic equivalence for locally acting products in the gastrointestinal tract”), which is to be approved finally in 2018.

Cutaneous products („Guideline on quality and equivalence of topical products”), which is expected to be released for consultation in 2018.

The presentation summarises the requirements of these three new guidelines on locally acting products, whose basic principles can be used for similar products without an specific guideline like locally acting nasal products.

## POTVRDA TERAPIJSKE EKVIVALENTNOSTI ORALNIH INHALACIONIH LEKOVA - REGULATORNI ASPEKTI

Zorica Pejčić

Agencija za lekove i medicinska sredstva Srbije (Srbija)

Za potvrdu terapijske ekvivalentnosti oralnih inhalacionih lekova koji deluju lokalno u plućima, u EU se primenjuje stupnjeviti pristup u skladu sa smernicom CPMP/EWP/4151/00 Rev1. Prvi korak uključuje *in vitro* ispitivanja leka, drugi farmakokinetička (FK), a treći farmakodinamska (FD) ili klinička ispitivanja. *In vitro* ispitivanja vrše se uz pomoć uređaja koji *in vitro* simulira ponašanje inhaliranog leka u respiratornom traktu (npr. Andersenovog kaskadnog impaktora). Ukoliko su ispunjeni svi definisani uslovi navedeni u smernici, terapijska ekvivalentnost dva inhalaciona leka može se potvrditi već u ovom prvom koraku, a ako nisu ispunjeni, pristupa se *in vivo* FK ispitivanjima.

Za potvrdu ekvivalentne efikasnosti u FK studijama potrebno je pokazati da su generički i referentni lek ekvivalentni u pogledu plućne depozicije, odnosno količine leka dospelog u pluća i rasporeda čestica leka unutar pluća. U ove svrhe sprovode se FK studije u kojima je resorpcija aktivne susptance iz gastrointestinalnog trakta (GIT) blokirana *p.o.* primenom aktivnog uglja, te se prati samo resorpcija leka iz pluća. Za potvrdu ekvivalentne bezbednosti generičkog i referentnog leka potrebno je meriti i porebiti ukupnu sistemsku izloženost leku, odnosno ukupnu količinu leka resorbovanog iz GIT i pluća, za šta treba sprovesti FK studiju bez *p.o.* primene aktivnog uglja. Ukoliko se u FK studijama dobiju zadovoljavajući rezultati, terapijska ekvivalentnost može se smatrati potvrđenom.

Ukoliko terapijska ekvivalentost nije pokazana FK studijama neophodno je sprovesti FD ili kliničke studije u kojima se generički inhalacioni lek poredi sa referentnim na osnovu odgovarajućih parametara praćenja efikasnosti i bezbednosti. Najveći broj generičkih inhalacionih lekova u EU dobija dozvolu na osnovu FK studija. U toku je revizija smernice CPMP/EWP/4151/00 Rev1, od koje se očekuje da dodatno precizira i pojasni pojedine detalje u pogledu ispitivanja terapijske ekvivalentnosti inhalacionih lekova.

## **REGULATORY FRAMEWORK FOR DEMONSTRATION OF THERAPEUTIC EQUIVALENCE OF ORALLY INHALED PRODUCTS**

**Zorica Pejčić**

Medicines and Medical Devices Agency of Serbia (Serbia)

When concluding therapeutic equivalence for orally inhaled products which are locally acting in lungs, a step-wise approach is applied in the EU according to guideline CPMP/EWP/4151/00 Rev1. In vitro approach is recommended as the first step, the second step are pharmacokinetic (PK) studies and the third step are pharmacodynamic (PD) or clinical studies. In vitro investigations are conducted using an apparatus that in vitro simulates behaviour of the inhaled drug in the respiratory tract (e.g., Andersen cascade impactor). If all conditions recommended in the guideline are met, therapeutic equivalence can be shown in this first step, and if they are not met, PK studies should be conducted.

In PK studies, equivalent efficacy between generic and reference product is assumed if pulmonary deposition and pattern of deposition within the lungs is shown to be equivalent. This can be assessed if PK studies are performed in the conditions when drug absorption through the gastrointestinal tract (GIT) is blocked (e.g., by p.o administration of active charcoal), and the drug absorption occurs only through the lungs. For demonstration of equivalent safety, total systemic drug exposure should be measured, i.e., the portion of the dose absorbed both through the lungs and GIT in the PK study without charcoal blockage. If the results of PK studies are acceptable, therapeutic equivalence can be confirmed in this step.

If therapeutic equivalence is not confirmed by PK studies, it is necessary to conduct PD or clinical studies in which generic and reference products are compared based on appropriate efficacy and safety endpoints. Most generic orally inhaled products are now being approved in the EU based on PK studies. Currently, the guideline CPMP/EWP/4151/00 Rev1 is under revision with the aim of its updating in order to reflect the knowledge gained from regulatory experience in the previous period.

## **KADA (NI)JE MOGUĆA SUPSTITUCIJA GENERIČKIM LEKOM**

**Marija Jovanović**

Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu-  
Farmaceutski fakultet (Srbija)

Generički lekovi se pojavljuju na tržištu posle isteka patentnih prava originalnog leka. Dokumentacija koja se podnosi za izdavanje dozvole za stavljanje generičkog leka u promet je u manjem obimu u odnosu na originalni lek i, pored dokumentovanog kvaliteta, sadrži podatke o biološkoj ekvivalentnosti u odnosu na referentni lek. Generički lek ima isti kvalitativni i kvantitativni sastav aktivnih supstanci kao i referentni lek. Takođe, generički lek ima isti farmaceutski oblik kao referentni lek, ali se mogu razlikovati u pogledu imena, pomoćnih supstanci, izgleda i pakovanja.

Generička supstitucija se zasniva na pretpostavci da je generički lek ekvivalentan referentnom u pogledu efikasnosti i bezbednosti. Iako se politika vezana za generičku supstituciju razlikuje od zemlje do zemlje, često se ohrabruje kao pokušaj smanjenja troškova zdravstvene zaštite. Međutim, u slučaju određenih lekova ili određenih okolnosti možda je bolje da se izbegne rizik. Posebno je problematična grupa lekova sa uskim terapijskim opsegom kao što su određeni antiepileptici, antikoagulansi, imunosupresivi i drugi, što može rezultovati nepovoljnim kliničkim ishodima. Male varijacije u koncentraciji leka mogu dovesti do izostanka terapijskog odgovora ili povećane toksičnosti. Pored toga, i druge aspekte vezane za lek ili pacijenta treba pažljivo razmotriti. Na kraju, kada postoje razlozi za zabrinutost, adekvatnost generičke supstitucije treba procenjivati individualno, za određenog pacijenta. Svakako je korisno konsultovati validne i pouzdane izvore informacija prilikom donošenja odluke o zameni.

## **WHEN GENERIC SUBSTITUTION IS (NOT) APPROPRIATE**

**Marija Jovanović**

Department of Pharmacokinetics and Clinical Pharmacy, University of  
Belgrade-Faculty of Pharmacy (Serbia)

Generic drugs appear on the market after the expiry of patent rights of the original drug. The documentation submitted for marketing authorisation of generic medicine is reduced compared to the original medicine, and besides documented quality, contains data on bioequivalence with the reference medicinal product. A generic drug has the same qualitative and quantitative composition in active substances as the reference medicinal product. Also, it has the same pharmaceutical form as the reference drug, but it can be different in terms of the name, inactive ingredients, appearance and packaging.

Generic substitution rests on the assumption that the generic medicine is equivalent to the reference product in terms of efficacy and safety. Although policies on generic substitution vary from country to country, it is often encouraged as an attempt to reduce healthcare costs. However, in conjunction with certain medicines or certain circumstances, it may be better to avoid the risk. Drugs with narrow therapeutic ranges such as certain antiepileptics, anticoagulants, immunosuppressants and others are particularly problematic and may result in adverse clinical outcomes. Small variations in drug concentration may result in lack of therapeutic response or increased toxicity. In addition, other aspects related to the medicinal product or the patient should be carefully considered. Finally, when there are reasons to be concerned, the appropriateness of generic substitution should be assessed on an individual basis, for a specific patient. Undoubtedly, valid and trusted sources of information should be consulted when making a decision about the substitution.

## AKUTNA TROVANJA ILEGALNIM PSIHOAKTIVnim SUPSTANCAMA - ISKUSTVA NACIONALNOG CENTRA ZA KONTROLU TROVANJA

**Jasmina Jović-Stošić<sup>1</sup>, Tomislav Režić<sup>1</sup>, Nataša Perković-Vukčević<sup>1</sup>,  
Slavica Vučinić<sup>1</sup>, Gordana Brajković<sup>1</sup>, Snežana Đorđević<sup>1</sup>, Mirjana Đukić<sup>2</sup>**

<sup>1</sup>Nacionalni centar za kontrolu trovanja, Medicinski fakultet Vojnomedicinske akademije, Univerzitet odbrane, Beograd, <sup>2</sup>Katedra za toksikologiju „Akademik Danilo Soldatović”, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Zloupotreba ilegalnih psihoaktivnih supstanci može dovesti do neželjenih efekata i predoziranja sa ozbiljnim akutnim poremećajima i eventualno smrtnim ishodom. Godišnje se u Nacionalnom Centru za kontrolu trovanja zbog predoziranja drogama zbrine 300–400 pacijenata koji su doveženi kao hitni slučajevi, uglavnom sa teritorije Beograda i obližnjih gradova. Prikazujemo aktuelne epidemiološke podatke i ukazujemo na osnovne karakteristike ovih trovanja u pogledu dijagnostike, terapije i ishoda. Retrospektivno su analizirani slučajevi predoziranja ilegalnim drogama u petogodišnjem periodu (2013-2017).

U posmatranom periodu su lečene 1683 osobe, životne dobi od 14 do 60 godina, pri čemu je zastupljenost mlađih od 18 godina iznosila 12,6%. Predoziranja su bila značajno češća kod muškaraca (u oko 75% slučajeva). Najčešće se radilo o heroinu, kod 51% bolesnika. Tegobe nakon konzumiranja marihuane su bile razlog javljanja lekaru kod 18% bolesnika. Derivati amfetamina [3,4-metilendioksimetamfetamin (MDMA, metamfetamin i dr.) su bili uzročnici trovanja u 11%, a kokain u 7% slučajeva. „Nove” psihoaktivne supstance [sintetski kanabinoidi, γ-hidroksibuterna kiselina (GHB)] su registrovane kod 4% bolesnika, dok kod 9% nije potvrđen etiološki agens. Kod većine pacijenata za potvrdu dijagnoze su korišćene standardne metode, dok su za nove psihoaktivne supstance razvijene LC/MS ili HPLC/PDA metode.

Velika većina (skoro 90%) pacijenata je zbrinuta u dnevnoj bolnici. Opijatni sindrom uzrokovani heroinom, sa depresijom disanja, hipotenzijom i ARDS-om je bio najčešći uzrok teških trovanja i produženog hospitalnog lečenja. Letalitet u trovanju heroinom je iznosio 1%. Kod ostalih agenasa je zabeležen samo jedan letalni ishod u trovanju amfetaminima čije su glavne manifestacije bile hipertermija i konvulzije. Kod predoziranja marihanom su dominirali tahikardijski i psihički simptomi, a ovi poremećaji su bili izraženiji nakon zloupotrebe sintetskih kanabionoida.

Iskustvo NCKT ukazuje da heroin i dalje predstavlja najznačajniji uzrok teških i letalnih trovanja, dok zloupotreba amfetaminskih psihostimulansasa i drugih novih psihoaktivnih supstanci, predstavlja sve veći izazov u pogledu dijagnostike i terapije.

## **ACUTE POISONING WITH ILLICIT PSYCHOACTIVE SUBSTANCES – EXPERIENCE OF THE NATIONAL POISON CONTROL CENTRE**

**Jasmina Jović-Stošić<sup>1</sup>, Tomislav Režić<sup>1</sup>, Nataša Perković-Vukčević<sup>1</sup>,  
Slavica Vučinić<sup>1</sup>, Gordana Brajković<sup>1</sup>, Snežana Đorđević<sup>1</sup>, Mirjana Đukić<sup>2</sup>**

<sup>1</sup>National Poison Control Centre, Military Medical Academy, Medical Faculty,  
University of Defence, Belgrade, <sup>2</sup>Department of Toxicology „Akademik Danilo  
Soldatović”, University of Belgrade-Faculty of Pharmacy (Serbia)

Abuse of illicit psychoactive substances may result in adverse effects and overdoses with serious disorders and sometimes fatal outcomes. National Poison Control Centre admits per year about 300-400 emergency cases due to illicit drugs overdoses, mainly from the territory of Belgrade and nearby cities. We present the current epidemiological data and point to the basic characteristics of these poisonings in terms of diagnostics, therapy and outcomes.

During the observed period, 1683 persons, aged from 14 to 60 were treated. The share of younger than 18 was 12.6%. Overdoses were significantly more common in men (in about 75% of cases). The most commonly used drug was heroin, in 51% of patients. The symptoms after consuming marijuana were the cause of medical help asking in 18% of patients. Amphetamine derivatives [3,4-methylenedioxymethamphetamine (MDMA), methamphetamine, etc.] were toxic in 11%, and cocaine in 7% of cases. „New” psychoactive substances [synthetic cannabinoids, γ-hydroxybutyric acid (GHB)] were registered in 4% of patients, while in 9% the etiological agent was not confirmed. In the majority of cases, standard methods were used to confirm the diagnosis, while LC/MS or HPLC/PDA methods were developed for new psychoactive substances.

The vast majority (almost 90%) of patients are treated in a daily hospital. Opioid syndrome caused by heroin, with respiratory depression, hypotension and ARDS was the most common cause of severe poisoning with prolonged hospital treatment. Mortality in heroin poisoning was 1%. Among the other agents, only one lethal outcome was recorded, in case of amphetamine poisoning, manifested by hyperthermia and convulsions. Adverse effects of marijuana were mainly tachycardia and psychotic symptoms, and these disorders were more pronounced after abuse of synthetic cannabinoids.

The experience of the NCKT indicates that heroin remains the most significant cause of severe and lethal poisoning, while the abuse of amphetamine psychostimulants and other new psychoactive substances is an increasing challenge in terms of diagnosis and therapy.

## THE ANALYSIS OF PSYCHOACTIVE SUBSTANCES: CHALLENGES RELATED TO BIOLOGICAL SAMPLES AND ANALYTICAL TOOLS

**Goran Mitulović**

Medical University of Vienna, Clinical Institute of Laboratory Medicine (Austria)

Blood, urine, hair, and saliva are the main and most commonly used biological materials for drug testing. Other matrices can also be used but have been applied only in limited amount and number of cases. Depending on clinical and analytical question to be answered, but also on legal restraints and environmental and technological settings, the choice of matrix and the analytical approach for analysis must be carefully chosen. For example, blood reflects a recent drug intake and is used to assess users' impairment. Analysis of saliva will also show a recent drug intake and indirectly show drug presence in blood but it is no possible to assess the concentration of the drug in blood. As for urine and hair, these matrices can be applied for detecting the drug (mis)use during a longer period of time.

Drug detection must not always be difficult and modern analytical technologies allow for fast detection of very low amounts of substances in biological matrices. However, before analyzing such a matrix one must consider the following:

- the timeframe of drugs' use,
- sampling methods,
- sample storage/transport,
- type of biological matrix (blood, urine, saliva, tissue etc.),
- steps for sample preparation prior to analysis and detection (protein precipitation, extraction etc.),
- the use of sample – court, screening etc.

Furthermore, the analytical method to be used shall be robust, reproducible, and relatively insensitive toward biological matrix applied. All these requirements pose a challenge for method development and validation, and for later quality control (think of blanks and controls).

## PREGLED SITUACIJE NA TRŽIŠTU DROGA U SRBIJI I PREDIZIRANJA SUPSTANCAMA ZLOUPOTREBE LEČENIH U NACIONALNOM CENTRU ZA KONTROLU TROVANJA VMA

**Slavica Vučinić, Jasmina Jović-Stošić, Dragana Đorđević,  
Tomislav Režić, Snežana Đorđević, Vesna Kilibarda**

Nacionalni centar za kontrolu trovanja, Vojnomedicinska akademija, Medicinski fakultet Univerzitet odbrane, Beograd (Srbija)

Evropski centar za monitoring droga i zavisnosti od droga i EUROPOL beleži sve veću pojavu droga uslovljenu veoma dinamičnim promenama na tržištu droga, novim tehnikama proizvodnje i načina distribucije. Na osnovu brojnih indikatora se zaključuje da je upotreba kanabisa i ilegalnih stimulanasa tipa kokaina, amfetamina i njemu sličnih supstanci (ATS) i novih psihoaktivnih supstanci (NPS) u Evropi u porastu, dok je broj korisnika heroina relativno stabilan. Cilj rada je analizirati učestalost i težinu trovanja različitim psihoaktivnim supstancama (PAS) lečenih u Nacionalnom centru za kontrolu trovanja (NCKT), VMA, Srbija.

Primenjena je retrospektivna studija pacijenata lečenih u NCKT zbog predoziranja supstancama zloupotrebe u petogodišnjem periodu. Od ukupnog broj trovanja, PAS su registrovane u 162 slučaja (4,06%) u 2011., 224 (5,36%) u 2012., 281 (6,69%) u 2013., 312 (7,07%) u 2014. i 442 (9,31%) u 2015. godini što je značajan porast broja trovanja ( $p<0.001$ ). U poređenju sa 2011.g., sledećih godina broj pacijenata lečenih zbog predoziranja heroinom (67,3%; 60,3%; 61,9%; 54,5%; 50,9%) je značajno manji ( $p<0,001$ ), dok raste broj pacijenata sa predoziranjem ATS. Ipak nije bilo razlike u stepenu težine trovanja u analiziranom periodu. Neki od potencijalno zabrinjavajućih trendova na tržištu droga povezani su sa porastom broja pacijenata koji se zbog efekata ATS i NPS leče u NCKT. Ponovni interes za MDMA praćen je odgovarajućim preventivnim akcijama NCKT. U Srbiji je ustanovljen zakonski okvir za organizovanu akciju unutar Sistema ranog upozoravanja na NPS u kome se podaci iz NCKT koriste za reagovanje.

# **AN OVERVIEW OF THE DRUG MARKET AND SUBSTANCES OF ABUSE OVERDOSE TREATED IN THE NATIONAL POISON CONTROL CENTER MMA**

**Slavica Vučinić, Jasmina Jović-Stošić, Dragana Đorđević,  
Tomislav Režić, Snežana Đorđević, Vesna Kilibarda**

National Poison Control Centre, Military Medical Academy, Medical faculty  
University of Defense, Belgrade (Serbia)

As the drug phenomenon continues to evolve, highly dynamic changes in the drug market, production techniques, and methods of distribution are noted by the European Monitoring Centre for Drugs and Drug Addiction and EUROPOL. Multiple indicators suggest that cannabis and illicit stimulant drugs, such as cocaine, amphetamine type substances (ATS), and new psychoactive substances (NPS) use are on the rise in Europe, while the number of heroin users is relatively stable. The objective of the current study was to analyze the frequency and severity of poisoning by different psychoactive substances (PAS) treated at the National Poison Control Centre (NPCC), MMA, Serbia. Retrospective study of patients treated for substances of abuse overdose at the NPCC in five-year period has been performed.

Out of all poisonings, PAS were represented with 162 (4.06%) in 2011, 224 (5.36%) in 2012, 281 (6.69%) in 2013, 312 (7.07%) in 2014 and 442 (9.31%) in 2015 which is a significant increase ( $p<0.001$ ). Compared to 2011, in the following years, the number of patients treated for heroin overdose (67.3%; 60.3%; 61.9%; 54.5%; 50.9%) was significantly reduced ( $p<0.001$ ), while the number of ATS overdose increased. However there was no differences in the severity of poisoning in the analyzed time period. Some potentially worrying changes in the drug market are associated with the increase of patients with ATS and NPS effects, treated at the NPCC. Resurgence of MDMA was followed by appropriate preventive actions of the NPCC. The framework for organized action within the Early Warning System on NPS is established in Serbia and the data from the NPCC will be used in the response.

## KURIKULARNE I EKSTRAKURIKULARNE AKTIVNOSTI U DOPRINOSU RAZUMEVANJU ZLOUPOTREBE PSIHOAKTIVNIH SUPSTANCI

Mirjana Đukić

Katedra za toksikologiju „Akademik Danilo Soldatović”, Univerzitet u Beogradu  
- Farmaceutski fakultet (Srbija)

Izborni predmet (kurs) „Sredstva koja izazivaju zavisnost sa analitikom” je uveden 2008. u dodiplomske studije na Farmaceutskom fakultetu Univerziteta u Beogradu. Kurikulum je postavljen po ugledu na svetske i evropske (EU) obrazovne programe na temu zloupotrebe droga (ZD). Kurs evoluira u skladu sa politikom, planovima i programima protiv ZD, od globalnog do nacionalnog nivoa. Cilj kursa je da upozna studenate farmacije sa složenošću ovog problema, sa medicinskog, analitičkog, ali i sa aspekta društva.

Nastavne aktivnosti su podeljene između profesora, kliničkog toksikologa, psihijatra, socijalnog radnika policije i samih studenata. Studenti izlažu sopstvene seminarске radove, referišu prikaze slučajeva (situacije iz realnog života), obavljaju laboratorijski rad i koriste platformu za e-učenje.

Kurikulum kursa pokriva sledeće oblasti: i) struktura-doza-efekat, mehanizmi toksičnosti, toksikokinetika i analitika droga (i u biološkim uzorcima); ii) medikamentozna terapija (prva pomoć, detoksikacioni protokoli, kliničke zbrinjavanje zavisnika); iii) tzv. „društveni detoksikacioni protokoli” (program rehabilitacije i reintegracije u 12 koraka i ostali pojedinačni ili grupni psihoterapijski programi); iv) nacionalna i EU politika protiv zloupotrebe droga i njihova klasifikacija (izvori: programi i godišnji izveštaji EU članica o zloupotrebi droga, a bazirano na ustanovljenim kriterijumima i pokazateljima od strane Evropskog centra za praćenje i zloupotrebu droge); i v) aktuelnosti o novim drogama, debate na temu legalizacije kanabisa, itd.

Za sada, farmaceuti u Srbiji nisu uključeni u kreiranje, razvoj i sprovođenje nacionalnog sistema za obrazovanje, strategiju i politiku ZD. U ovom domenu obavljaju dva tipa usluga: sudska- i kliničko-toksikološke analize (u referentnim institucijama) i kontrolu zloupotrebe recepata i lekova (pogotovo analgetika i na bazi efedrina/amfetamina) u slobodnoj prodaji (u apotekama).

## **CURRICULAR AND EXTRA-CURRICULAR ACTIVITIES TO DEVELOP COMPREHENSION ON ABUSE OF PSYCHOACTIVE SUBSTANCES**

**Mirjana Đukić**

Department of Toxicology „Akademik Danilo Soldatović”, University of Belgrade-Faculty of Pharmacy (Serbia)

The undergraduate elective course „Substance abuse” was established 2008, at the University of Belgrade-Faculty of Pharmacy. The curriculum was established in accordance with global and European Union (EU) educational system on drug abuse (DA). It has been evolving with trends in anti-drug policies, plans and programs, from global to national level.

The goal of the course is to teach students of pharmacy about the DA complexity from the medical, analytical and social prospective.

Teaching activities are shared between professors, clinical toxicologists, police social workers, psychiatrists and students themselves. Students present seminar papers, report on case studies (depict real-life situations), perform toxicological analysis and use e-learning platform.

The course curriculum includes the following topics: i) assessing of structure-dose-effect relationship, toxicity mechanisms, toxicokinetics and drug analysis; ii) medical treatment of SA (first aid, drug detox protocols and innovations, clinical case studies, etc.); iii) social programs (12- step rehabilitation and reintegration program and socio detox protocols-individual or group psychotherapy sessions); iv) classifications of SA and National and EU anti-drug policies (sources: programming documents and EU countries' annual surveys on SA, grounded on the criteria and indicators established by European Monitoring Centre for Drugs and Drug Addiction (EMCDDA); and v) actualities on novel drugs, debates on cannabis legalization, etc.

Pharmacists in Serbia have not been participating in creating, developing and providing the national educating system, strategy and policy on DA. So far, they manage two types of DA services: forensic and clinico-toxicological analysis (within referring institutions) and controlling the abuse of prescription and over-the-counter medications (emphasis on: analgesics and ephedrine/amphetamine containing drugs), in pharmacies.

## **THE EFFECTS OF NATURAL AND SYNTHETIC ENVIRONMENTAL POLLUTANTS ON HUMAN HEALTH: SOME CASE STUDIES**

**Emanuela Testai**

National Institute for Health Environment and Health Dept. (Italy)

Among environmental pollutants, those known as 'contaminants of emerging concern' have received an increasing attention, especially when chronic exposure is concerned. This is particularly true for water bodies contaminants, used as drinking water supplies, considering the frequency of exposure for a large part of the population. Beside some 'well known' chemicals which are routinely monitored, the improved analytical methods sensitivity has allowed to detect chemicals that had not previously been detected: among them microcystins (MCs), produced by cyanobacteria naturally present in many water bodies, and per- and polyfluorinated alkyl substances (PFAS), persistent environmental chemicals used in a wide range of industrial applications and commercial products. Dense blooms of cyanobacteria increasingly occurring worldwide, due to eutrophication and climate changes. MCs are among the most studied cyanotoxins; the group consists of more than 100 hepatotoxic variants, with different potency, associated to differences in their kinetic behaviour, acting through inhibition of PPA1 and PP2A triggering a cascade of events leading to hepatotoxicity and tumor promotion. Some variants showed *in vitro* also a neurotoxic potential. Also in the case of PFAS kinetics play a crucial role, determining their bioaccumulation potential by binding to plasma proteins paralleled by a lack of biotransformation and a very slow urinary excretion with renal resorption. Some epidemiological studies carried out in highly contaminated areas showed a positive association of PFAS with increased total cholesterol and low and high density lipoproteins in blood, suggesting dysfunction in lipid metabolism. Other effects were described but they are still under discussion. Both class of contaminants consist of a group of variants, to which it is possible to be simultaneously exposed, but unfortunately data gaps in knowledge about the toxicity of different MC variants or PFAS family members limit the application of a cumulative risk assessment procedure.

## METIL-ŽIVA U NAŠEM OKRUŽENJU: KLJUČNE ČINJENICE ZA SIGURNU BUDUĆNOST

Danijela Đukić-Ćosić

Katedra za toksikologiju „Akademik Danilo Soldatović”, Univerzitet u Beogradu  
- Farmaceutski fakultet (Srbija)

Povećano prisustvo žive (Hg) u životnoj sredini je globalni problem zbog njene perzistentnosti, potencijala za bioakumulaciju i toksičnosti za ljude. U životnu sredinu Hg dolazi iz prirodnih i antropogenih izvora i prisutna je u tri oblika: elementarna Hg, neorganska jedinjenja Hg i organska jedinjenja Hg. Pomoću bakterija neorganska jedinjenja Hg se transformišu u organski oblik, metil-živu (MeHg) koja se akumulira najviše u ribama. Izloženost MeHg putem kontaminirane ribe i proizvoda od njih može predstavljati opasnost po zdravlje ljudi. Metil-živa ima poznato štetno dejstvo na nervni sistem jer se apsorbuje oko šest puta lakše od neorganskih jedinjenja Hg i prolazi krv-moždanu barijeru. Novija istraživanja pokazuju da čak izlaganje malim količinama MeHg može izazvati ozbiljne zdravstvene probleme i predstavljati opasnost za in utero razvoj i razvoj male dece. Stoga je SZO MeHg svrstala među prvi deset hemikalija od značaja za javno zdravlje. Unos MeHg putem ribe i proizvoda od ribe se procenjuje širom sveta u osetljivim populacijama, poput trudnica i male dece i ovakvi preliminarni rezultati postoje i u Srbiji. Kod osetljivih populacija unos MeHg treba svesti na najmanju moguću meru uzimajući u obzir da riba predstavlja neophodnu namirnicu u uravnoteženoj ishrani. Međunarodne organizacije (FDA i EPA) dale su preporuke u vezi sa vrstama ribe koje su bezbedne za ishranu kako bi informisale trudnice i roditelje male dece.

Globalna situacija o prisustvu MeHg u lancu ishrane i pokazani negativni efekti kod ljudi zabrinjavajuće je delovala na vlade zemalja širom sveta koje su se 2013. godine složile o donošenju Minamata konvencije. Konvencija obuhvata niz akcija, uključujući smanjenje emisije Hg u vazduh eliminisanjem upotrebe Hg za dobijanje zlata, eliminisanjem iskopavanja Hg ruda i ograničenjima i zabranama određenih proizvoda koji sadrže Hg.

# **METHYLMERCURY IN OUR ENVIRONMENT: KEY FACTS FOR A SAFE FUTURE**

**Danijela Đukić-Ćosić**

Department of Toxicology „Akademik Danilo Soldatović”, University of Belgrade-Faculty of Pharmacy (Serbia)

Mercury (Hg) pollution is a global problem due to its persistence in the environment, potential to bioaccumulate, and toxicity to humans. In the environment, it comes from the natural and anthropogenic sources and exists in three forms: elemental, inorganic, and organic Hg. Once there, Hg can be transformed by bacteria into methylmercury (MeHg) which bioaccumulates mainly in fish. Human exposure to MeHg from contaminated fish and fish products can pose a variety of health risks. Methylmercury is a known neurotoxin. It is absorbed into the body about six times more easily than inorganic Hg and has the ability to cross the blood-brain barrier. Moreover, new investigations indicate that even exposure to small amounts of MeHg may cause serious health problems, especially in utero and early in life. Thus, WHO classified MeHg as one of the top ten chemicals of major public health concern.

Intake of MeHg through fish and fish products is assessed in sensitive populations worldwide, as well as in Serbia, where the preliminary results exist. The vulnerable populations are pregnant women, their unborn children and children. Their exposure to MeHg should therefore be minimized, while recognizing that fish constitutes an important part of a balanced diet. International organizations (FDA and EPA) have issued recommendations regarding fish species that are healthy and safe to eat in order to inform pregnant women and parents of young children.

The global situation about the presence of MeHg in the food chain and demonstrated adverse effects on human health have raised the concern of the governments worldwide and led to the adoption of the Minamata Convention in 2013. The Convention covers a range of actions, including decrease in mercury emissions into the air by stopping the use of mercury in gold mining, eliminating the mining of mercury and phasing out certain mercury-containing products.

## DOKAZI TOKSIČNOSTI USPORIVAČA GORENJA - POLIBROMOVANI DIFENILETRI

Marijana Čurčić

Katedra za toksikologiju „Akademik Danilo Soldatović”, Univerzitet u Beogradu  
- Farmaceutski fakultet (Srbija)

Polibromovani difeniletri (PBDEs) su industrijske hemikalije koje su korišćene u različitim vrstama proizvoda kao što su računari, mobilni telefoni, bela tehnika, predmeti od plastike i poliuretanske pene, itd. I pored dobrih karakteristika kao usporivača gorenja, utvrđeno je da ove hemikalije izazivaju brojne toksične efekte na zdravlje ljudi i životnu sredinu.

PBDEs se mogu transportovati na velike udaljenosti i perzistentni su zagađivači životne sredine s obzirom na dugo poluvreme razgradnje. Najviše koncentracije PBDEs izmerene su u krvi stanovništva koje živi u blizini deponija opasnog električnog i elektronskog otpada ili zaposlenih na poslovima njihove reciklaže. Epidemiološke i eksperimentalne studije ukazuju da izloženost PBDEs može ispoljiti toksičnost na brojne organe i sisteme organa. Dokazana je toksičnost PBDEs na štitastu žlezdu i to najčešće u vidu sniženja nivoa hormona T4, T3 i TSH što može dalje rezultirati i poremećajima u razvoju centralnog nervnog sistema. Pored štitaste Studije ukazuju i na hepato toksičnosti ovih hemikalija, kao i toksične efekte na imuni system, muški reproduktivni sistem i pankreas (nastanak Diabetes mellitus-a). Međunarodna agencija za istraživanje karcinoma (IARC) klasificuje PBDEs u grupu 3 (ne klasifikuju se kao karcinogeni za ljude), a *in vitro* ispitivanja svedoče i o cito- i genotoksičnom potencijalu ovih hemikalija. Mechanizmi toksičnosti PBDEs nisu u potpunosti razjašnjeni, a neki od predloženih mehanizama su vezivanje za receptore za aromatične ugljovodonike, nastanak oksidativnog stresa, uticaj na aktivnost enzima kao što su monooksigenaza i transferaza, itd. Dokazan neprihvatljiv rizik PBDEs za zdravlje ljudi i životnu sredinu inicirao je zabranu ili ograničenje njihove proizvodnje, stavljanja u promet i korišćenja, a regulatorni osnov za sprovođenje ovih aktivnosti je dala Stokholmska konvencija.

## **EVIDENCE OF FLAME RETARDANTS TOXICITY - POLYBROMINATED DIPHENYL ETHERS**

**Marijana Čurčić**

Department of Toxicology „Akademik Danilo Soldatović”, University of Belgrade-Faculty of Pharmacy (Serbia)

Polybrominated diphenyl ethers (PBDEs) are industrial chemicals which were used in different type of products such as PCs, cell phones, appliances, plastics, polyurethane foams, etc. Despite good characteristics as flame retardants, it was found that PBDEs cause numerous toxic effects in human and environment.

PBDEs could be transported to long distances and they are persistent environmental pollutants having in mind their long degradation half-time. The highest PBDEs concentrations were measured in blood of citizens living in the proximity of electronical and electrical waste landfills or who work in recycling factories. Epidemiological and experimental studies imply that exposure to PBDEs may exert toxicity on numerous organs and organ systems. Toxicity for the thyroid gland has been proven and is most frequently manifested as decreased T4, T3 and TSH levels which results in central nervous system developmental disorders. Studies also give evidence of their hepatotoxicity, immunotoxicity, male reproductive toxicity and toxic effects on pancreas (Diabetes mellitus development). The International Agency for Research on Cancer (IARC) has classified PBDE as a Group 3 carcinogen (not classifiable as to its carcinogenicity to humans) and *in vitro* studies point to cyto – and genotoxic potential of these chemicals. Mechanisms of toxicity are still rather unexplained, but some of the suggested mechanisms are bounding to aryl hydrocarbon receptor, oxidative stress induction, interference with the activity of enzymes such as monooxygenase and transpherase, etc. Proven unacceptable risk for human health and environment initiated bans and restrictions of their production, placing on the market and use, while regulatory basis for these activities was set by the Stockholm convention.

## OPASNE HEMIKALIJE U PROIZVODIMA ŠIROKE POTROŠNJE I REGULATORNI ASPEKT KAO MEHANIZAM KONTROLE U EU I REPUBLICI SRBIJI

**Iasminka Randelović, Jelena Milić, Valentina Mart, Lazarija Šojić**

Alternativa za bezbednije hemikalije - ALHem, Beograd (Srbija)

Jedan od važnih ciljeva ALHem-a, kao organizacije civilnog društva koja se bavi bezbednim upravljanjem hemikalijama, jeste praćenje primene zakonske regulative koja se odnosi na hemikalije i u tom smislu ALHem redovno sprovodi ispitivanja prisustva opasnih hemikalija u proizvodima za široku potrošnju koji se nalaze na tržištu RS. U toku 2017/2018 godine sproveli smo kampanju pod nazivom „Toksični račun” sa ciljem da skrenemo pažnju javnosti na prisustvo bisfenola A (BPA) u termalnim papirima, pre svega fiskalnim računima i bankovnim isečcima, sa kojima svi svakodnevno dolazimo u kontakt.

BPA se od januara 2018. godine nalazi na Listi kandidata supstanci koje izazivaju zabrinutost (Substances of very high concern – SVHC) u EU, s obzirom da je dokazano da može štetno da utiče na plodnost i da ometa rad endokrinog sistema. U decembru 2016. godine Evropska komisija je donela odluku o zabrani, odnosno ograničenju upotrebe bisfenola A u termalnom papiru, ukoliko je koncentracija BPA veća od ili jednaka 0,02 masenih procenata. Ova odluka počinje da se primenjuje od 2. januara. 2020. godine. Republika Srbija je u aprilu 2018. godine preuzeila ovu odredbu u domaće zakonodavstvo. Prisustvo BPA je laboratorijski testirano u ukupno 33 uzorka, od čega: 20 termalnih papira (fiskalnih računa i drugih termalnih papira iz javnog i privatnog sektora), kao i papirne i plastične ambalaže za hranu.

Rezultati su pokazali da su svi uzorci uvezenih fiskalnih rolni termalnog papira pozitivni na sadržaj BPA, u koncentraciji od 0,63 do 0,91 masenih %. Imajući u vidu da na evropskom tržištu postoje snabdevači termalnog papira koji ne sadrže ovu opasnu hemikaliju, apelujemo na institucije u državnom i javnom sektoru, kao i na kompanije u privatnom sektoru, naročito na trgovачke lance da zamene ovaj proizvod bezbednjom alternativom bez BPA kako bi doprineli očuvanju zdravlja svojih zaposlenih, naročito kasirki, ali i svih građana Srbije.

## **HAZARDOUS CHEMICALS IN ARTICLES FOR EVERYDAY USE AND REGULATORY ASPECT AS A CONTROL MECHANISM IN EU AND SERBIA**

**Iasminka Randelović, Jelena Milić, Valentina Mart, Lazarija Šojić**

Safer Chemicals Alternative - ALHem, Belgrade (Serbia)

One of the important goals of ALHem, as a civil society organization dealing with the safe chemicals management, is to monitor the implementation of chemicals legislation, and in this aspect ALHem regularly tests the presence of hazardous chemicals in consumer goods placed on the Serbian market. During 2017/2018, we conducted a campaign called „Toxic Cash Receipts” with the goal of drawing attention of the public to the presence of bisphenol A (BPA) in thermal papers, primarily cash receipts and banking slips, with which we are in the contact every day.

Since January 2018, BPA has been on the Candidate List of Substances of Very High Concern (SVHC) in the EU, because it was proved to be harmful for fertility and disruptive for endocrine system. In December 2016, the European Commission made a decision to ban, i.e. restrict use of BPA in thermal paper, if concentration of BPA equals or exceeds 0.02 mass %. This decision shall be applied as of 2 January 2020. The Republic of Serbia has transposed this provision in April this year into the national legislation. The presence of BPA was laboratory tested in a total of 33 samples, of which: 20 thermal papers (cash receipts and other thermal paper from public and private sectors), as well as on plastic and paper food packaging.

The results indicated that all samples of imported cash receipts rolls were positive on BPA, in the range of 0.63 and 0.91 mass %. Taking into account that on European market exists suppliers of BPA free thermal paper, ALHem calls upon institutions from state and public sectors, as well as upon companies from private sector, especially upon trade chains, to replace this product with safer alternative, so as to contribute to health protection of their staff, especially cashiers, but also of all citizens of Serbia.

## PRIMENA 3D ŠTAMPE U FARMACIJI - IZAZOVI I PERSPEKTIVE

Svetlana Ibrić

Katedra za farmaceutsku tehnologiju i kozmetologiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Tehnologija 3D štampe se intezivno razvijala u protekloj deceniji, a primena ove tehnologije u medicinske svrhe postaje veoma aktuelna. Nakon registracije SPIRITAM® tableta, koje je FDA registrovala 2015. godine, a koje su proizvedene tehnologijom 3D štampe, postoji povećan interes za primenu aditivne proizvodnje u farmaciji, posebno kada su u pitanju čvrsti oralni farmaceutski oblici. Uvedena je i nova reč za tablete pripremljene korišćenjem ove tehnike – „printlete“. Glavni pokretači za 3D štampanje lekova su personalizovana terapija za posebne grupe pacijenata (pedijatrijska ili gerijatrijska populacija); precizne doze lekova, kao i proizvodnja na zahtev pojedinačnog pacijenta, sa fleksibilnošću doze. Pored toga, moguće je proizvoditi „polypill“ - jedan dozirani oblik sa dve ili više lekovite supstance i više različitih kinetika oslobađanja leka. Magistralna izrada lekova u apoteci se čini odgovarajućim mestom gde bi se mogli pripremati 3D štampani lekovi, kada su propisani za potrebe pojedinačnog pacijenta, a sadrže personalizovane doze sa unapred određenim količinama leka. Postoji nekoliko 3D tehnologija štampe koje su na raspolaganju istraživačima: (1) „ink-jet“ štampa u obliku mlaznica; (2) modeliranje fuzionim taloženjem - ekstruzija materijala; (3) stereolitografija – fotopolimerizacija; (4) fuzija praška - selektivno lasersko sinterovanje; (5) ekstruzija gela/paste. Prednosti 3D štampe u poređenju sa konvencionalnom proizvodnjom lekova ogledaju se u preciznoj kontroli doziranja, reproduktivnosti, inovativnosti i mogućnosti modelovanja kinetike oslobađanja leka.

Uprkos napretku, 3D štampanje i dalje ostaje nova tehnologija i zbog toga je suočena sa brojnim etičkim i regulatornim pitanjima. Postoji niz značajnih pitanja koja moraju biti razmotrena u budućnosti, kao što su problem falsifikovanja lekova, promet neregistrovanih lekova, kao i brojna regulatorna pitanja.

## **3D PRINTING FOR PHARMACEUTICAL APPLICATIONS – CHALLENGES AND PROSPECTS**

**Svetlana Ibrić**

Department of Pharmaceutical Technology and Cosmetology, University of Belgrade – Faculty of Pharmacy (Serbia)

3D printing technology has become very attractive in medicine in the past decade. With FDA approval of SPIRITAM® tablets manufactured by 3D printing technology in 2015, there is increased interest in the field of additive manufacturing, especially for solid oral dosage forms. Even the new word for the tablets prepared using this technique is adopted – printlets. Main drivers for 3D printing of medicines are personalized therapy for special groups of patients (pediatric or geriatric population); precise dose medication as well as on-demand, point-of-care manufacturing (with dose flexibility). Additionally, using additive manufacturing, it is possible to manufacture „*polypill*”- one dosage form with two or more drugs. The compounding pharmacy seems to be the appropriate site where 3D printing medications can be prepared when prescribed/ordered, and packaged, handled, administered and charged in personalized dose units containing a predetermined amount of drug. There are several 3D printing technologies that are available for researchers: (1) powder bed ink-jet head printing - binder jetting; (2) fused deposition modeling – material extrusion; (3) stereolithography – vat photopolymerisation; (4) powder bed fusion – selective laser sintering; (5) gel/paste extrusion - material extrusion. The advantages and obstacles for each method will be discussed.

Considering the advantages of 3D printing, i.e. accurate control of droplet size and dosage, high reproducibility, complex standardized drug manufacturing processes, and the ability to produce dosage forms characterized by innovative and unique dosage forms, personalized drug dosing, and complex drug-release profiles, undoubtedly, the challenge to conventional drug production is imminent. Despite progress, 3D printing still remains a new technology and therefore suffers from ethical and regulatory problems. There are a number of significant concerns related to possibility of counterfeit drugs, proliferation of illegal medicines, mislabelling and regulatory vacuum, that must be addressed.

## SAVREMENI PRISTUP ODABIRU FORMULACIJE I EKSCIPIJENASA

Ružica Kolaković

Janssen Pharmaceutica, Johnson & Johnson (Belgija)

Većina novih aktivnih supstanci koje ulaze u fazu razvoja poseduju slabu rastvorljivost i/ili brzinu rastvaranja u vodi što dovodi do niske bioraspoloživosti. Dva najčešća pristupa rešenju ovog problema su formulacija aktivne supstance u obliku mekih želatinskih kapsula i konverzija kristalne supstance u amorfnu čvrstu disperziju (tehnikom sušenja raspršivanjem ili ekstruzijom). Veliki broj faktora mora biti razmotren u toku razvoja mekih kaspula i čvrstih disperzija pri čemu je odabir ekscipijenasa i njihove količine u formulaciji jedan od najvažnijih.

Trenutno dostupne tehnike za selekciju, unapređene od strane mnogobrojnih svetskih laboratorija, zasnovane su na različitim teorijskim i eksperimentalnim principima, ali u svrhu ostvarenja zajedničkog cilja - odabir formulacije koja će obezbediti potrebnu bioraspoloživost i istovremeno posedovati fizičku stabilnost. Izbor optimalne formulacije često zahteva veliko finansijsko i vremensko ulaganje, kao i utrošak značajne količine aktivne supstance koja je u ranim fazama razvoja često nedostupna. U cilju rešenja ovog problema unapređen je automatizovani skrining visokog operativnog kapaciteta (High throughput screening - HTS) koji se zasniva na upotrebi male zapremine uzorka. Skrining se koristi u cilju odabira ekscipijenasa za formulaciju mekih kapsula i čvrstih disperzija i izvodi se u pločama sa 96-bazenčića (96 well-plate). Za čvrste disperzije skrining se bazira na izradi tankih filmova koji sadrže 25-75 µg aktivne supstance. Odabir najbolje formulacije izvodi se na osnovu testiranja oslobađanja supstance iz filma, kao i stabilnosti filma (na povišenoj temperaturi i vlažnosti). Istovremeno se mogu testirati 43 formulacije uz utrošak 1 g aktivne supstance. Za formulaciju mekih kapsula rastvorljivost aktivne supstance u 96 različitih ekscipijenasa i njihovih kombinacija se testira istovremeno uz korišćenje 300 µl ekscipijensa po uzorku. Primenom ove tehnike značajno se redukuju vreme i novac potrebeni za unapređenje nove formulacije uz minimalan utrošak aktivne supstance.

## **MODERN APPROACH TO FORMULATION AND EXCIPIENTS SELECTION**

**Ružica Kolaković**

Janssen Pharmaceutica, Johnson & Johnson (Belgium)

Vast majority of new active pharmaceutical ingredients (APIs) entering development pipeline possess insufficient water solubility and/or low dissolution rate. These cause challenges in achieving desired bioavailability. Two most frequently used approaches to overcome this problem are incorporation of API in a lipid based drug delivery system (LBDDs) (i.e. soft-gel capsule formulation) and conversion of crystalline API to its amorphous form by means of spray drying or hot-melt extrusion (i.e. amorphous solid dispersions, ASD).

Variety of formulation parameters can affect the development of ASDs and LBDDs with selection of appropriate excipients and their ratio being one of them. Number of research laboratories have been developing their own screening methodologies, either based on experimental data or based on theoretical fundamentals with one common goal - choosing the right formulation composition which will ensure both desired bioavailability and physical stability of drug product over intended shelf-life.

Selecting the optimal formulation can take a vast amount of time, be extremely costly and, importantly, require significant amount of API which is not readily available in the early phase of development. To tackle this, Janssen Pharmaceutica has developed low-volume automated high throughput screening platform for selection of ASD and lipid based formulations. Such a screen is performed in 96 well-plate using automated liquid and solid dispensing. For ASDs a film casting method is used resulting in films containing 25-75 µg of API. Produced films are evaluated for their dissolution rate by small scale dissolution method and their stability assessed by exposing them to stressing conditions (elevated temperature and/or relative humidity). In that way, 43 ASD formulations can be tested simultaneously with use of only 1 g of API. For LBDDs solubility of API is performed in 96 prototype formulations simultaneously. Using this approach significant reduction in development time and cost, improved success rate and minimized API consumption are achieved.

## **ULOGA KLINIČKOG FARMACEUTA U RAZVOJU FORMULACIJA LEKOVA ZA SPROVOĐENJE KLINIČKIH ISPITIVANJA**

**Marija Tubić - Grozdanis**

Univerzitetski klinički centar, Johan Gutenberg Univerzitet u Majncu (Nemačka)

Tim kliničkog farmaceuta se svakodnevno suočava sa različitim izazovima tokom sprovođenja kliničkih studija različitih faza. Adekvatno obučeno osoblje i odgovarajući resursi kvalifikuju bolničku apoteku za učešće u kliničkim ispitivanjima na različite načine: od pripreme i izdavanja leka, popunjavanja propratne dokumentacije, pa sve do preuzimanja uloge koordinatora studije i glavnog istraživača. Tokom kliničkog ispitivanja (sponzorisanog od strane industrije ili pokrenutog od strane istraživača) klinički farmaceut učestvuje u procesu revizije protokola kliničke studije, izrade, pakovanja i obeležavanja leka koji se ispituje, kao i u definisanju strategije u placebo kontrolisanim studijama. Klinički farmaceut je takođe uključen u sagledavanje mogućnosti nabavke kvalitetnih aktivnih i pomoćnih supstanci, proceni troškova razvoja formulacije i izrade preparata, proces pribavljanja saglasnosti za izvođenje kliničke studije, kao i u određivanju mogućih rizika primene i problema u izradi preparata za sprovođenje kliničkih ispitivanja. ADKA (Udruženje kliničkih farmaceuta Nemačke) je objavila smernice o učestvovanju kliničkih farmaceuta u komercijalnim kliničkim ispitivanjima prema GCP pravilima, koje utvrđuju standarde u pogledu rekonstitucije, čuvanja i distribucije preparata koji se koriste u kliničkim ispitivanjima. Odeljenje za kliničke studije bolničke apoteke Univerzitetskog kliničkog centra u Majncu poseduje GMP dozvolu od 2006. godine i učestvuje u razvoju formulacija, izradi i distribuciji lekova za klinička ispitivanja u različitim centrima širom sveta. Proizvodnja obuhvata pripremu kapsula, punjenje ili prepakivanje ispitivanih, kao i placebo preparata. Analitičke metode i testovi stabilnosti su važna potpora razvoju i izradi preparata koji se koriste u kliničkim studijama. Postojanje Odeljenja za kliničke studije u bolničkoj apoteci, kao i mogućnost što ranijeg uključenja kliničkog farmaceuta u kliničko istraživanje je presudno za uspešnu saradnju kliničkog centra (pacijenti i medicinsko osoblje), bolničke apoteke i farmaceutske industrije, čime se kliničko istraživanje usmerava ka brzoj i uspešnoj primeni optimizovane inovativne terapije.

## **CLINICAL PHARMACIST'S ROLE IN CLINICAL TRIALS INVESTIGATIONAL DRUG DEVELOPMENT**

**Marija Tubić - Grozdanis**

University Medical Center, Johannes Gutenberg University Mainz (Germany)

Clinical pharmacists team daily faces different challenges in terms of running multiple clinical studies at multiple stages. With adequate trained staff and resources, hospital pharmacy can be involved in clinical trial research in a variety of ways, from providing drug and record keeping for drug accountability to taking on the roles ranging from study coordinator to principal investigator. In clinical trials (industry-sponsored or investigator-initiated trials), clinical pharmacist is able to advise on review process of clinical trial protocols, pharmacy manuals, manufacturing, packaging and labeling of investigational drug, as well as the techniques, challenges and strategies in comparator blinding. Clinical pharmacist is also involved in the issues such as: the source, quality and costs of investigational drug, the regulatory approval process, the identification of possible clinical risk and safety aspects of drug handling, dispensing and reconstitution. The ADKA (Association of German Hospital Pharmacists) released a guideline on the participation of hospital pharmacies in commercial clinical trials in adherence to GCP (Good Clinical Practice). The guideline constitutes quality standards for pharmacy services in Germany regarding: reconstitution, storage, logistics, documentation and destruction. Clinical trial Unit of the Pharmacy Department in the University Medical Center in Mainz, as a holder of a GMP (Good Manufacturing Practice) manufacturing licence since 2006, provides formulation assistance, manufacturing and distribution to national and international clinical sites. Manufacturing comprises encapsulation, filling or re-packaging of investigational and placebo products. Formulation development is supported by analytical and stability testing (including microbiological assays). The existence of Clinical trial unit within a hospital pharmacy and early as possible involvement of clinical pharmacists in clinical trial research is crucial for successful partnership between clinical site (patients and health care professionals), hospital pharmacy and pharmaceutical industry, support clinical trial research in the right direction and lead to less time consuming and cost optimization.

## INKAPSULACIJA ODABRANIH SUPERKRITIČNIH EKSTRAKATA LEKOVITOГ BILJA U LIPOSOME METODOM HOMOGENIZACIJE POD VISOKIM PRITISKOM

Ivana Arsić<sup>1</sup>, Vanja Tadić<sup>2</sup>, Milica Stanković<sup>1</sup>, Vesna Savić<sup>1</sup>

<sup>1</sup>Katedra za farmaciju, Univerzitet u Nišu - Medicinski fakultet, <sup>2</sup>Institut za proučavanje lekovitog bilja „Dr Josif Pančić”, Beograd (Srbija)

Liposomi su fosfolipidni, biodegradabilni i biokompatibilni vezikularni sistemi koji se koriste za stabilizaciju, poboljšanje penetracije i permeacije, produženo/kontrolisano oslobađanje, kao i povećanje bioraspoloživosti teško rastvornih lekovitih supstanci. Za proizvodnju biljnih i tradicionalnih biljnih lekova sve češće se koriste ekstrakti dobijeni procesom superkritične ekstrakcije koja ima niz prednosti u odnosu na konvencionalne metode: dobijaju se ekstrakti sa značajno većom koncentracijom aktivnih principa, povećane stabilnosti, bez teških metala i drugih zagađivača (mikroorganizama), bez ugrožavanja integriteta termolabilnih supstanci, uz očuvanje životne sredine (brzo uklanjanje ekstragensa-gasa u superkritičnom stanju) i mogućnost višestrukog frakcionisanja ekstrakta. Ekstrakti su visoke lipofilnosti, pa se radi poboljšanja rastvorljivosti u vodi, resorpcije i bioraspoloživosti mogu inkapsulirati u liposome. Proces se može izvesti korišćenjem prečišćenih frakcija fosfolipida, metodom homogenizacije pod visokim pritiskom. Cilj rada bio je utvrđivanje uticaja prirode ekstrakta i parametara procesa (pritisak i broj ciklusa homogenizacije) na efikasnost inkapsulacije, veličinu (srednji prečnik, RL) i indeks polidisperziteta (IP) liposoma iz prečišćenog fosfolipida Phosal-a 75SA (Lipoid GmbH, Nemačka) i natkritičnih ekstrakata: talusa lišaj brade, herbe timijana, smilja i majkine dušice, lista matičnjaka i šišarica hmelja. Liposomi su formirani primenom pritiska od 300, odnosno 500 bara, a broj ciklusa homogenizacije iznosio je 7, odnosno 10. Proces je izведен korišćenjem homogenizatora pod visokim pritiskom (EmulsiFlex-C3, Avestin, Kanada). Dobijene disperzije liposoma čuvane su u staklenim bočicama, dobro zatvorene, na sobnoj temperaturi u periodu od 3 meseca a stabilnost je praćena merenjem RL i IP u definisanim test terminima. RP i IP mereni su na aparatu Zetasizer Nano ZS90 (Malvern Instruments, Velika Britanija). Rezultati ukazuju na značajan uticaj vrste superkritičnog ekstrakta, primjenjenog pritiska i broja ciklusa homogenizacije na RL i IP i njihovu fizičku stabilnost. Postupkom homogenizacije (10 ciklusa, 500 bar), korišćenjem Phosal-a 75SA mogu se inkapsulirati ispitivani superkritični ekstrakti u liposome zadovoljavajućih karakteristika (srednji prečnik u rasponu 179-235 nm, efikasnost inkapsulacije 45-55%) i stabilnosti u posmatranom periodu.

## **INCAPACULATION OF SELECTED MEDICINAL HERB'S SUPERCRITICAL EXTRACTS IN LIPOSOMES USING THE HIGH PRESSURE HOMOGENIZATION METHOD**

**Ivana Arsić<sup>1</sup>, Vanja Tadić<sup>2</sup>, Milica Stanković<sup>1</sup>, Vesna Savić<sup>1</sup>**

<sup>1</sup>Department of Pharmacy, University of Niš - Faculty of Medicine, <sup>2</sup>Institute for Medicinal Plant Research „Dr Josif Pančić”, Belgrade (Serbia)

Liposomes are phospholipid, biodegradable and biocompatible vesicular systems which are used to stabilize, improve penetration and permeation, for prolonged/controlled release and increase the bioavailability of medicinal substances that are difficult to dissolve. The usage of extracts obtained by the supercritical extraction process for the production of herbal and traditional herbal remedies is increasing, due to a number of advantages of supercritical extraction in comparison to conventional extraction methods: it gives extracts with significantly higher concentration of active principles with better stability, without heavy metals and other pollutants (microorganisms), without compromising the integrity of thermolabile substances, with the preservation of the environment (rapid removal of extragent - gas in supercritical state) and with the possibility of multiple fractionation of the extracts. Because the extracts have a high level of lipophilicity, they can be encapsulated in the liposomes, in order to improve solubility in water and to enable better absorption and bioavailability. The process can be carried out using purified phospholipid fractions and a high pressure homogenization method. The aim of the work was to determine the influence of extract characteristics and process parameters (pressure and number of homogenization cycles) on the efficiency of encapsulation, size (mean diameter, RL) and index of polydispersity (IP) of liposomes from purified phospholipids Phosal 75SA (Lipoid GmbH, Germany) and supercritical extracts: thallus of beard lichen, wild thyme herb, curry plant herb, common thyme herb, lemon balm leaves and hops. Liposomes were formed using a pressure of 300 and 500 bar and the number of homogenization cycles was 7 and 10. The process was carried out using a high pressure homogenizer (EmulsiFlex-C3, Avestin, Canada). The obtained liposomal dispersions were stored in glass bottles, well closed, at room temperature for a period of 3 months, and the stability was tested by measuring RL and IP at defined time intervals. RL and IP were determined using the Zetasizer Nano ZS90 (Malvern Instruments, UK). The results showed that type of supercritical extract, the applied pressure and the number of the homogenisation cycle had an influence on the RL, IP and their physical stability. Using the homogenization procedure (10 cycles, 500 bar), with Phosal 75SA, investigated supercritical extracts can be encapsulated in liposomes with satisfactory characteristics (mean diameter in the range of 179-235 nm, efficiency of encapsulation 45-55%) and stability in the observed period.

## ANTIEPILEPTICI U SVETLU NOVIH INDIKACIJA

**Radica Stepanović-Petrović**

Katedra za farmakologiju, Univerzitet u Beogradu-Farmaceutski fakultet  
(Srbija)

U poslednjih 30-tak godina u upotrebu je ušlo oko 20-tak novih antiepileptika (AE). Imajući u vidu da je incidenca epilepsije 0,5-1%, jasno je da sa ovakvim napretkom u razvoju novih AE za farmaceutsku industriju ne može samo epilepsija biti atraktivna kao indikacija, mada je sa kliničkog aspekta problem rezistentnih epilepsija još uvek nerešen. S druge strane, mnogi AE imaju multimodalalan mehanizam dejstva koji često uključuje modulaciju GABA-ergičke i glutamatergičke neurotransmisije kao i promene u aktivnosti voltažno zavisnih kanala za natrijum i kalcijum i intracelularnih signalnih puteva. Zbog svega ovoga, poslednjih decenija je indikacijsko polje pojedinih AE postalo bogatije za izvesne neurološke i psihiatrijske bolesti. Konkretno, najznačajnije nove indikacije za AE su neuropatski bol, profilaksa migrene, fibromijalgija, bipolarni poremećaj i generalizovani anksiozni poremećaj. Pregabalin je upravo zbog proširenja indikacijskog područja (neuropatski bol, dodatna terapija u lečenju fokalnih epileptičkih napada, generalizovani anksiozni poremećaj) postao u 2018. godini jedan od lekova koji je ostvario enormne prihode (blockbuster).

Nisu svi AE efikasni u navedenim indikacijama koje nisu u vezi sa epilepsijom. Samo oni AE čiji se mehanizam dejstva dobro uklapa sa patofiziologijom određenih bolesti imaju potencijal da budu efikasni u ne-epileptičnim indikacijama. Tako su karbamazepin, valproinska kiselina i lamotrigin odobreni za primenu u bipolarnom poremećaju, dok karbamazepin i gabapentinoidi (gabapentin i pregabalin) predstavljaju osnovne lekove u lečenju različitih neuropatskih bolnih sindroma. Za profilaksu migrene se koriste valproati, topiramat i gabapentin, dok se gabapentin koristi još i za ublažavanje simptoma menopauze.

Posebno se novi AE ispituju preklinički i klinički u nekim ne-epileptičnim oboljenjima. I dok velika većina novih AE pokazuju efikasnost u prekliničkim i manjim otvorenim kliničkim studijama, u randomizovanim placebo kontrolisanim kliničkim studijama neki AE pokazuju odsustvo efikasnosti. Zato prednost treba dati kontrolisanim kliničkim studijama kako bi njihovi pozitivni rezultati vodili ka čvrsto utemeljenoj indikaciji a ne „off-label“ upotrebi.

*Ovaj rad je finansiran od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije (Projekat br. 175045).*

## **ANTIEPILEPTICS IN LIGHT OF NEW INDICATIONS**

**Radica Stepanović-Petrović**

Department of Pharmacology, University of Belgrade-Faculty of Pharmacy  
(Serbia)

In the last 30 years, 20 new antiepileptic drugs (AEDs) have been introduced in the market. Bearing in mind that the incidence of epilepsy is 0.5-1%, it is clear that with such progress in developing new AEDs, epilepsy alone could not be attractive for the pharmaceutical industry, although the clinical needs of refractory epilepsy remain unmet. On the other hand, most AEDs have multiple mechanisms of action which include modulation of GABA-ergic and glutamatergic neurotransmission, and alteration of voltage-gated sodium and calcium channels or intracellular signaling pathways. As a result of all this, in recent decades there has been an expansion of indications of individual AED for certain neurological and psychiatric diseases. In particular, the most significant new indications for AEDs are neuropathic pain, migraine prophylaxis, fibromyalgia, bipolar disorder and generalized anxiety disorder. Because of the widening of indication area (neuropathic pain, adjunctive therapy in the treatment of focal seizures, generalized anxiety disorder) pregabalin has become one of the drugs that generates enormous revenues (blockbuster) in 2018.

Not all AEDs are effective in treating nonepileptic conditions. Only those AEDs whose mechanism of action fits well with the pathophysiology of certain diseases have the potential to be effective in nonepileptic indications. In that way, carbamazepine, valproic acid and lamotrigine are approved for use in bipolar disorder, and carbamazepine and gabapentinoids ( gabapentin and pregabalin) are central drugs in the treatment of various neuropathic pain syndromes. Valproate, topiramate and gabapentin are used for prophylaxis of migraine, and gabapentin is used also for the relief of menopausal symptoms.

New AEDs are particularly tested preclinically and clinically in some nonepileptic conditions. The vast majority of new AEDs show efficacy in preclinical and small open-label clinical studies, but fail to provide strong evidence in randomized, placebo-controlled clinical trials. Therefore, priority should be given to controlled clinical trials whose positive results could lead to a firmly grounded indication rather than off-label use.

*This work was supported by the Serbian Ministry of Education, Science and Technological Development (grant 175045).*

## **BENZODIAZEPINES ARE ALL ALIKE - EXCEPT WHEN THE OPPOSITE COMES TRUE**

**Margot Ernst**

Medical University of Vienna - Department of Molecular Neurosciences  
(Austria)

Pharmacology textbooks to this date still state that all benzodiazepine based medicines act by the same mechanism, namely the unselective allosteric modulation of GABA-A receptors by interaction with four high affinity binding sites. This view has also led to efforts for developing agents that are selective for only one of these four sites, where „Z-drugs” are considered prototypical „alpha-1-selective” agents.

Careful scrutiny of the literature suggests a more complex picture, and clinicians are convinced that the assumption of a „common mechanism” falls short of any objectively useful and correct model. Here we approach the issue from two perspectives, namely the molecular viewpoint which reveals a much larger number of potentially important binding sites that may account for in vivo effects, and from the clinical perspective. The case of a patient who displays massive paradoxical effects to bromazepam, but not to several other benzodiazepines, suggests that the notion of „positive allosteric modulation” also should be refined and revisited.

Thus, we will provide an overview of all known benzodiazepine binding sites on the large family of benzodiazepine- sensitive GABA-A receptor subtypes. Based on this molecular multitude of sites, we will discuss the need to close gaps in the knowledge about the interactions of approved medications with so far less studied binding sites. We will further provide a literature review which clearly indicates that acute and chronic benzodiazepine effects indicate complexity well beyond any „four-site- model”. We finally will discuss the molecular nature of „positive allosteric modulation” and suggest refined views onto the terms of „benzodiazepine-site agonists, inverse agonists and antagonists”.

## ALFA 1, 2, 3, 4, 5, 6 GABA A RECEPTORI: ŠTO VIŠE TO BOLJE KAO CILJ ZA NOVE LEKOVE?

Miroslav Savić

Katedra za farmakologiju, Univerzitet u Beogradu-Farmaceutski fakultet  
(Srbija)

Jonotropni  $\text{GABA}_A$  receptori su najzastupljenija populacija receptora u centralnom nervnom sistemu, koju karakteriše izuzetna raznovrsnost. Ne samo da čak 19 podjedinica ( $6\alpha$ ,  $3\beta$ ,  $3\gamma$ ,  $\delta$ ,  $\epsilon$ ,  $\pi$ ,  $\theta$ ,  $3\rho$ ) može da učestvuje u formiraju pentamernog agregata, već su one različito eksprimirane u različitim reginima neurona (sinaptička prema ekstrasinaptičkoj lokalizaciji) i osim toga u različitim strukturama mozga. Pojedinačna karakteristika  $\text{GABA}_A$  receptora koja u najvećoj meri određuje ulogu odgovarajuće populacije receptora jeste prisustvo jednog od šest podtipova alfa podjedinice. Dok su prve tri populacije (alfa1  $\text{GABA}_A$ , alfa2  $\text{GABA}_A$  i alfa3  $\text{GABA}_A$  podtip) lokalizovane gotovo u potpunosti sinaptički, naredne tri populacije (alfa4  $\text{GABA}_A$ , alfa5  $\text{GABA}_A$  i alfa6  $\text{GABA}_A$  podtip) prisutne su pretežno, ako ne i isključivo, ekstrasinaptički. Mnogo napora je ulagano da se sintetišu novi ligandi koji bi posedovali selektivnost afiniteta ili funkcionalnu selektivnost za samo jednu populaciju  $\text{GABA}_A$  receptora. Aktuelno stanje znanja sugeriše da fiziološka aktivacija i/ili farmakološka potencijacija odgovarajuće populacije  $\text{GABA}_A$  receptora mogu da dovedu do sledećih bihevioralnih efekata: sedacije, amnezije, antikonvulzivne aktivnosti i podložnosti razvoju zavisnosti preko alfa1  $\text{GABA}_A$  receptora; anksiolize, smanjene reaktivnosti na stres, miorelaksacije i antihiperalgezije preko alfa2  $\text{GABA}_A$  receptora; izmenjene aktivnosti senzornomotornih vratnica, miorelaksacije i antihiperalgezije preko alfa3  $\text{GABA}_A$  receptora; kognitivnog oštećenja i regulacije unosa alkohola preko alfa4  $\text{GABA}_A$  receptora; anksiolize, smanjenog odgovora na stres, izmenjene aktivnosti senzornomotornih vratnica, kognitivnog oštećenja u zadacima niske interferencije i kognitivnog poboljšanja u testovima visoke interferencije preko alfa5  $\text{GABA}_A$  receptora, i izmenjene aktivnosti senzornomotornih vratnica i anithiperalgezije preko alfa6  $\text{GABA}_A$  receptora. Sveukupni razvoj koncepta selektivne modulacije populacija  $\text{GABA}_A$  receptora upućuje na veću verovatnoću postizanja terapijskih probrova u sklopu istraživanja sa ligandima koji deluju preko onih receptora čija je distribucija ograničenja i pretežno ekstrasinaptička, što je slučaj sa alfa5 i alfa6  $\text{GABA}_A$  receptorima.

# **ALPHA 1, 2, 3, 4, 5, 6 GABA A RECEPTORS: THE HIGHER THE BETTER AS A TARGET FOR NOVEL MEDICINES?**

**Miroslav Savić**

Department of Pharmacology, University of Belgrade-Faculty of Pharmacy  
(Serbia)

The ionotropic GABA<sub>A</sub> receptors are most abundant population of receptors in the central nervous system, characterized by an exceptional diversity. Not only that as many as 19 subunits ( $\alpha_6$ ,  $\beta_3$ ,  $\gamma_3$ ,  $\delta$ ,  $\varepsilon$ ,  $\pi$ ,  $\theta$ ,  $\beta_3$ ) can contribute to formation of the pentameric aggregate, but they are differentially expressed in various regions of a neuron (synaptic vs. extrasynaptic localization) and also in different structures of the brain. The single characteristic of GABA<sub>A</sub> receptors that mostly governs the role of the respective population of receptors is the presence of one of six alpha subunit subtypes. While the first three populations (alpha1 GABA<sub>A</sub>, alpha2 GABA<sub>A</sub> and alpha3 GABA<sub>A</sub> subtype) are located nearly completely synaptically, the latter three (alpha4 GABA<sub>A</sub>, alpha5 GABA<sub>A</sub> and alpha6 GABA<sub>A</sub> subtype) are predominantly if not exclusively present extrasynaptically. Much effort has been directed towards synthesis of novel ligands with affinity- or functional selectivity to only one population of GABA<sub>A</sub> receptors. The present state of knowledge suggests that physiological activation and/or pharmacological potentiation of the respective population of GABA<sub>A</sub> receptors may result in the following behavioral effects: sedation, amnesia, anticonvulsant activity and dependence liability through alpha1 GABA<sub>A</sub> receptors; anxiolysis, diminished reactivity to stress, myorelaxation and antihyperalgesia through alpha2 GABA<sub>A</sub> receptors; changed sensorimotor gating, myorelaxation and antihyperalgesia through alpha3 GABA<sub>A</sub> receptors; cognitive impairment and regulation of alcohol intake through alpha4 GABA<sub>A</sub> receptors; anxiolysis, diminished response to stress, changed sensorimotor gating, cognitive impairment in low interference tasks and cognitive improvement in high interference tasks through alpha5 GABA<sub>A</sub> receptors; and changed sensorimotor gating and antihyperalgesia through alpha6 GABA<sub>A</sub> receptors. The overall development of concept of selective modulation of GABA<sub>A</sub> receptor populations implies that it is more realistic to expect therapeutic breakthroughs at the level of novel ligands acting through those receptors which distribution is more limited and predominantly extrasynaptic, such is the case with alpha5 and alpha6 GABA<sub>A</sub> receptors.

## KOMBINACIJE ANALGETIKA U SAVREMENOM LEČENJU BOLA

Maja Tomić

Katedra za farmakologiju, Univerzitet u Beogradu-Farmaceutski fakultet  
(Srbija)

Ublažavanje bola je jedan od najzahvalnijih zadataka farmakoterapije. Primena kako klasičnih analgetika (opioida, nesteroidnih antiinflamatornih lekova i paracetamola), tako i alternativnih analgetika (prevashodno antiepileptika i antidepresiva), u pojedinačnoj terapiji, često je nedovoljno efikasna i/ili je praćena neželjenim efektima koji se ne mogu podneti. Dugotrajna primena opioida povezana je i sa razvojem tolerancije i zavisnosti. S obzirom da poslednjih nekoliko decenija nije bilo otkrića novih klasičnih analgetika koje bi značajno unapredilo lečenje bola, traganje za novim, efikasnijim i bezbednijim tretmanima predstavlja jedan od najznačajnijih ciljeva savremene farmakoterapije bola.

Jedan od načina za postizanje ovog cilja jeste multimodalna analgezija, koja podrazumeva kombinovanu primenu analgetika iz različitih farmakoloških grupa, tj. sa različitim mehanizmima dejstva. Koncept multimodalne analgezije zasniva se na poznavanju mehanizama transmisije i modulacije bola i delovanju na više različitih ciljnih mesta značajnih u ovim procesima i/ili na više nivoa duž bolnog puta (periferija, spinalni, supraspinalni nivo), čime se mogu postići sinergizam ili aditivnost u dejstvu. U prekliničkim uslovima, sinergizam se karakteriše značajnom, više nego dvostrukom redukcijom doza lekova primenjenih u dvokomponentnoj kombinaciji, i može se manifestovati kao postizanje određenog nivoa analgezije mnogo manjim dozama lekova od onih potrebnih za postizanje istog nivoa analgezije u monoterapiji, ili kao povećanje analgetičke efikasnosti kombinacije u odnosu na monoterapiju. Iako aditivnost podrazumeva manju redukciju doza lekova primenjenih u kombinaciji nego u slučaju sinergizma, ona i dalje može biti značajna sa aspekta redukovanja dozno-zavisnih neželjenih efekata.

Postoje brojni dokazi iz kliničkih studija i veliko kliničko iskustvo sa multimodalnom analgezijom u lečenju postoperativnog i malignog bola. Međutim, kod drugih akutnih, a posebno kod hroničnih bolnih stanja ne-maligne etiologije (kao što su neuropatski bol, artritis, fibromijalgija), čije je lečenje veliki izazov, dokazi su oskudniji, a primena kombinacija analgetika je često empirijska. Potrebna su dalja preklinička i klinička ispitivanja kako bi se identifikovale najpovoljnije kombinacije analgetika za različita bolna stanja.

*Ovaj rad je finansiran od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije (Projekat br. 175045).*

# **COMBINATIONS OF ANALGESICS IN THE CONTEMPORARY TREATMENT OF PAIN**

**Maja Tomić**

Department of Pharmacology, University of Belgrade-Faculty of Pharmacy  
(Serbia)

Alleviating pain is one of the most rewarding tasks of pharmacotherapy. The use of both classic analgesics (opioids, nonsteroidal antiinflammatory drugs and paracetamol) and alternative analgesics (mainly antiepileptics or antidepressants), as a single-drug treatment, is often not efficient enough and/or is accompanied by intolerable side-effects. Long-term administration of opioids is also associated with the development of tolerance and dependence. Since there was no discovery of new classic analgesics in the last few decades that would significantly improve the treatment of pain, the search for new, more effective and safer treatments is one of the most important goals of modern pain pharmacotherapy.

One of the ways to achieve this goal is to use multimodal analgesia, which involves the combined administration of analgesics from various pharmacological groups, i.e. with different mechanisms of action. The concept of multimodal analgesia is based on knowledge of the mechanisms of pain transmission and modulation, and acting on different targets involved in this processes and/or at several levels along the pain pathway (periphery, spinal, supraspinal level), which can result in synergism or additivity. In preclinical conditions, synergism is characterized by significant, more than a double reduction of doses of the drugs administered in two-drug combination, and can be manifested as achieving a certain level of analgesia with much lower doses of components than those required to achieve the same level of analgesia in monotherapy, or as an increase in the analgesic efficacy of the combination, compared to monotherapy. Although additivity implies less reduction of the doses of drugs administered in combination than in the case of synergism, it still may be meaningful from the aspect of dose-dependent side effects reduction.

There is ample evidence from clinical studies and large clinical experience with multimodal analgesia in the treatment of postoperative and malignant pain. However, for other types of acute and particularly chronic non-malignant pain states (such as neuropathic pain, arthritis, fibromyalgia), whose treatment is a major challenge, evidence is scarce, and the use of a combination of analgesics is often empirical. Further preclinical and clinical studies are needed to identify the most favourable combinations of analgesics for different painful conditions.

*This work was supported by the Serbian Ministry of Education, Science and Technological Development (grant 175045).*

## **IS EDUCATING PHARMACISTS TO BE COMPETENT ENOUGH FOR THE FUTURE OF THE PROFESSION?**

**Martin Henman**

School of Pharmacy and Pharmaceutical Sciences, Trinity College, University of Dublin (Ireland)

In response to the changing needs of patients, of health practitioners in managing medicines in complex cases and of health services in optimising the use of medicines, the profession of Pharmacy has revised pharmacy education to meet these needs. From the example of other health professions, pharmacists have sought to define their activities in terms of competencies. Each competency can be described in detail by what the pharmacist should be able to do, by behaviours, and the competencies can be grouped together as domains or areas containing the related competencies and their behaviours.

Co-ordinated by the International Pharmacy Federation (FIP), in 2012 the Pharmacy Education Taskforce, launched 'A Global Competency Framework'. With four domains, Pharmaceutical Care, Pharmaceutical Public Health, Organisation and Management and Personal/Professional, and 17 competencies, the document provided concise guidance for, in its own words, 'an overview of how practice at a foundation level can be translated into 'what' and 'how' students should learn'.

However, competencies and, in particular, the behaviours that are used to describe them, can become a list of elements to be assessed and one that is limited by the vision used to create the competency framework and the purpose for which the framework is used. Yet pharmacy education is about the student becoming a professional, it is a process not an itemised list of outcomes, it is about exercising judgement under difficult circumstances, when more than one option may appear to be appropriate. If the profession is to have a future in patient care, it must consider these aspects as well.

## STRUČNO OSPOSOBLJAVANJE I PROFESIONALNI RAZVOJ FARMACEUTA - AKADEMSKA PERSPEKTIVA

Ljiljana Tasić

Katedra za socijanu farmaciju i farmaceutsko zakonodavstvo, Univerzitet u Beogradu – Farmaceutski fakultet (Srbija)

Farmaceutski kadrovi su ključni za očuvanje i unapređenje zdravlja, a posebno za bezbednost pacijenata shodno izazovima novih tehnologija. Farmaceuti treba da su kvalifikovani i visoko kompetentni stručnjaci, aktivni učesnici zdravstvene zaštite orijentisani na pacijenta, spremni za kolaborativnu praksu, profesionalni i lični razvoj i kontinuirano unapredjenje. Ovo zahteva promene u pristupu obrazovanju koje akademska zajednica treba da prepozna, prihvati, sprovodi i unapređuje. Stručno osposobljavanje farmaceuta podrazumeva stepenasti sistem koji obuhvata najmanje četiri godine teorijske i praktične nastave na univerzitetu i najmanje šest meseci profesionalne prakse u apoteci u toku dodiplomskih studija ili u okviru pripravničkog staža, pre sticanja licence za samostalan rad. Nakon licenciranja farmaceuti treba da se celoživotno usavršavaju, u okviru formalnog sistema kontinuirane edukacije (KE), kao i u neformalnom sistemu edukacije (sopstveno iskustveno učenjena radnom mestu koje integriše profesionalni i lični razvoj). Visokoobrazovne ustanove prate, istražuju i aktivno učestvuju u ovim procesima i susreću se sa mnogim izazovima počev od: (i) usklađenosti nastavnih sadržaja sa potrebama zdravstvenih ustanova vs. potrebama građana (formalno i fizičko radno mesto farmaceuta se menja; građani su zahtevniji); (ii) novom organizacijom zdravstvenih sistema i novih usluga (fokus na pacijentu i primarnoj zdravstvenoj zaštiti); (iii) novom ulogom edukatora/savetnika u farmakoterapiji i prevenciji bolesti i promociji zdravlja; (iv) ulogom agenta/advokata u bezbednoj upotrebi lekova i upravljanju lekovima. Iskustva i publikacije o ovoj temi razvijenih zemalja (UK, SAD, Australija) ukazuju da se akademska zajednica menja i razvija shodno potrebama, jasnim političkim porukama, te da regulativa intezivno prati i uređuje odnose akademije i profesionalnih udruženja, a da Ministarstvo zdravlja prati tržište farmaceutskih kadrova. Sveukupno ovo značajno utiče na profesionalni razvoj farmaceuta, te je time kvalitet usluga i bezbednost pacijenata unapređena. U srednje i niže razvijenim zemljama formalno obrazovanje i usavršavanje (KE) je više zastupljeno od neformalnog, modaliteti profesionalnog razvoja su različiti, prisutan je nedostatak političke volje, nedovoljno istraživanja ovih fenomena, i inertnost/nemotivisanost nastavnika, kao i pojačano interesovanje (pritisak) građana. Rezultati istraživanja sprovedenih u okviru Erasmus+ projekta ReFEEHS u Srbiji, pojedinih pilot studija sprovedenih u Sloveniji, Hrvatskoj i BiH (Republici Srpskoj) će biti prikazani.

## **QUALIFICATION AND PROFESSIONAL DEVELOPMENT OF PHARMACIST - ACADEMIC PERSPECTIVE**

**Ljiljana Tasić**

Department of Social Pharmacy and Pharmaceutical Legislation, University of Belgrade - Faculty of Pharmacy (Serbia)

Pharmacists as health workers are essential for the preservation and improvement of health with emphasis on the patient safety in line with the challenges of new technologies. Pharmacists should be qualified and highly competent professionals, active participants in health care oriented towards patients, prepared for collaborative practice, professional and personal development and continuous improvement. This requires changes in the approach to education that the academic community should recognize, accept, implement and promote. Qualification as a pharmacist requires step-by-step system which includes at least four years of full-time theoretical and practical training at a university and six-month traineeship in a pharmacy during the undergraduate study program, or as internship before professional licensure. After licensing pharmacists are expected to conduct lifelong learning, either through the formal system of continuing education (CE) or in the informal system of work-based learning which integrates professional and personal development. Higher education institutions supervise, research and actively participate in these processes and are facing many challenges starting from: (i) alignment of curriculum with the health institutions needs vs. citizens' needs (pharmacist's workplace is changing; citizens are more demanding); (ii) a new organization and services in health care systems (focus on patient and primary health care); (iii) the new role of pharmacists as educators/advisors in pharmacotherapy, disease prevention and health promotion; (iv) the role of an agent/advocate in the safe use of medicines and drug management. Developed countries (UK, USA, Australia) experiences and publications on this subject indicate that the academic community is changing and developing in accordance with needs, clear political messages, regulation that intensively monitors and regulates relation: academy-professional associations and with Ministry of Health which monitors the pharmaceutical workforce. This significantly influences pharmacists' professional development and quality services and patient safety improvement. In the developing countries, formal education and training (CE) is more common than informal, the modalities of professional development are different, there is a lack of political will, insufficient research of these phenomena, and teachers' and preceptors' inertia with lack of motivation followed by increased citizens' pressure. The results of the research conducted within the Erasmus+ project ReFEEHS in Serbia, some pilot studies conducted in Slovenia, Croatia and Bosnia and Herzegovina (Republic of Srpska) will be presented.

## **STRUČNO OSPOSOBLJAVANJE I PROFESIONALNI RAZVOJ FARMACEUTA - PERSPEKTIVA APOTEKARSKE PRAKSE**

**Svetlana Stojkov**

Univerzitet Privredna akademija u Novom Sadu - Farmaceutski fakultet Novi Sad, Apoteka Subotica (Srbija)

Poslednjih decenija XX veka farmaceutska profesija značajno je pomerila aspekte i polja svog delovanja, čime je generisana potreba ka novim edukacionim sadržajima, programima i modelima, uključujući dostignuća informacionih tehnologija (IT). Cilj rada je da se prikažu stavovi i preferencije farmaceuta o područjima u kojima smatraju da treba da se razvijaju, formatima i modalitetima edukacije, sa akcentom na primenu IT.

Studijom preseka je za period 2014-2017. godina izvršeno istraživanje putem on-line ankete dostupne na sajtu Farmaceutske komore Srbije. Kreiran je upitnik koji je, pored ostalog, uključio preferencije i stavove farmaceuta o edukaciji uz primenu Likertove skale od 1 do 5 (1-najmanje interesantno, 5- najviše interesantno). Anketa je bila anonimna i dobrotvorna. U studiju je uključeno 565 diplomiranih farmaceuta/magistara farmacije.

Ispitanicima je bio ponuđen set pitanja u kojima su mogli da izraze svoju zainteresovanost za razvoj u određenim oblastima: 28,4% je odabralo bezbednost i delotvornost dijetetskih suplemenata u politerapiji, 26,2% posebne populacije i bezbednost lekova, 21,8% oblast novih odobrenih lekova, dok je dobra praksa farmakovigilance i nova EMA (European Medical Agency) bila najzanimljivija za 18,7% ispitanika. Istraživanje preferencija farmaceuta prema određenim formatima učenja, na skali od 1 do 5, je kod oko 60% ispitanika pokazalo najveći afinitet prema „prikazu slučaja“. Od ponuđenih modaliteta učenja, prvi izbori ispitanicima su edukacija putem Interneta (57,7%) i digitalni formati dostupni na internetu sa mogućnošću automatskog preuzimanja podkastova (49,4%). Telekonferencije, live streams webcastovi i on-demand webcastovi nisu prvi izbor za većinu ispitanika (7,4%; 17,4%; 17,5%, redom).

Kvalitet zdravstvene zaštite i bezbednost pacijenata zahteva stručne i logističke preduslove, kao i modele učenja koji su usklađeni sa potrebama i mogućnostima praktičara. Razvoj IT i dostupnost odgovarajućih edukativnih sadržaja, prilagođenih savremenom trenutku odnosno potrebama pacijenata, prepoznat je od farmaceuta u Srbiji kao poželjan put u profesionalnom razvoju.

## **QUALIFICATION AND PROFESSIONAL DEVELOPMENT OF PHARMACIST - PHARMACY PRACTICE PERSPECTIVE**

**Svetlana Stojkov**

University Business Academy in Novi Sad - Faculty of Pharmacy Novi Sad,  
Pharmacy Subotica (Serbia)

In last decades of the XX century the pharmaceutical profession significantly altered its aspects and area of operation, whereby a need for new educational content, programmes and models, including advancements in IT were generated. The objective of the paper is to demonstrate pharmacists' attitudes and preferences referring to the areas which they feel should be developed, via educational formats and modalities, with emphasis on IT application.

In the period from 2014 to 2017, a cross-sectional study was conducted via an online survey available on the website of the Pharmaceutical Chamber of Serbia. The survey, inter alia, included pharmacists' preferences and attitudes on education with the application of a Likert scale from 1 to 5 (1-least interesting, 5-most interesting). The survey was anonymous and voluntary. 565 BPharms/MPharms were included in the study.

Those surveyed was offered a set of questions where they were able to express their interest in the development of certain areas: 28.4% chose safety and the efficacy of nutritional supplements in polypharmacy, 26.2% chose specific populations and drug safety, 21.8% the area of newly approved drugs, while good pharmacovigilance practices and the new EMA (European Medical Agency) was of greatest interest to 18.7% of respondents. Researching pharmacists' preferences in terms of specific educational formats, on a scale of 1 to 5, approx. 60% demonstrated the greatest affinity towards „case review”. Of the available modalities of learning, the first choice of the respondents is education via the Internet (57.7%), and digital formats available on the Internet with the option of downloading podcasts (49.4%). Teleconferences, livestream webcasts and on demand webcasts are not the first choice of the majority of respondents (7.4%; 17.4%; 17.5%, respectively).

The quality of healthcare and patient safety requires expert and technical preconditions, as well as models of learning that comply with the needs and capabilities of practitioners. IT development and the availability of corresponding educational content, adapted to modern times i.e. patient needs, has been recognised by pharmacists in Serbia as the desired path in professional development.

## **WHY CLINICAL COMMUNICATION SKILLS REALLY MATTER? SOME EXAMPLES OF EFFECTIVE TEACHING AND LEARNING METHODS**

**Afonso Miguel Cavaco**

Department of Social Pharmacy, Faculty of Pharmacy, University of Lisbon  
(Portugal)

Pharmacy practice encompasses a number of professional activities, usually associated with medicines usage by patients. Pharmacists' duties should comprise the optimization of the clinical effectiveness and safety of medicines taken by patients. To be able to manage pharmacotherapy and evaluate its success with each patient, pharmacists need to collect and provide information. The individual patient is the main source of information, both objective and subjective. However, patients are more than data providers or recipients. Human nature is complex, suggesting the use of appropriate clinical communication skills. Although widely recognized as a key element in healthcare, these skills are usually less covered in pharmacy education and research. This presentation aims to address and debate how clinical communication skills are essential to take full advantage of the existing pharmaceutical knowledge, translating science into caring pharmacy practice. Some examples of teaching and learning resources for communication and clinical interaction skills will be presented, including options based on virtual reality.

## VALUE FRAMEWORKS AND DECISION MAKING AROUND THE GLOBE

**Wija Oortwijn, Rob Baltussen, Maarten Janssen**

Radboud University Medical Centre (Netherlands)

Health technology assessment (HTA) practices all employ so-called value frameworks for priority setting, i.e. making recommendations and/or reimbursement decisions regarding new health technologies. This includes a judgment on the relative importance of certain assessment criteria, such as clinical benefit and the incremental cost-effectiveness of (new) health technologies.

Some HTA practices focus on the development and use of evidence (e.g. Argentina), while others explicitly combine the use of evidence with procedural aspects, involving relevant stakeholders (e.g. the Netherlands). Although the practical application of a value framework is context-dependent, it is important to note that the underlying design has implications for the way in which priorities are set. Currently, some value frameworks may seriously comprise the legitimacy of reimbursement decisions. This indicates the need but also potential to improve HTA practices.

A way forward are evidence-informed deliberative processes (EDPs). EDPs is a new conceptual framework based on two validated frameworks, multi-criteria decision analysis and the Accountability for Reasonableness framework. EDPs provides a practical tool for HTA agencies aiming to set priorities regarding what is relevant and meaningful from a broader health system's perspective. It includes several steps related to the HTA process (scoping, assessment, appraisal, dissemination). These steps should not be considered as a blueprint but rather as an aspirational goal – organizations can take incremental steps. We will present the steps to undertake EDPs, substantiated with real world examples to enhance legitimate decision-making.

EDPs can facilitate democratic decision-making in various ways. It supports organizations to be more systematic, explicit and transparent, by making recommendations/decisions sensitive to a wider range of needs and values, and by promoting consistency across decisions.

## **VREDNOST INOVACIJE PRILIKOM DONOŠENJA ODLUKA U ZDRAVSTVU**

**Tanja Novaković**

ZEM Solutions, Beograd (Srbija)

Pružaoci zdravstvene zaštite u Centralnoj i Istočnoj Evropi (CEE) su suočeni sa nizom izazova koji se javljaju usled sve većeg broja zahteva i rastućih očekivanja. Kao i kod mnogih zdravstvenih sistema širom sveta, neophodno je da se poboljša pristup inovativnim tehnologijama u okviru sve ograničenijih budžeta. Ograničeno donošenje odluka i kašnjenje u procesu procene novih tehnologija mogu imati značajan uticaj na zdravlje pacijenata.

Inovacije mogu dovesti do povećane potrošnje u zdravstvenoj zaštiti kao rezultat supstitucije terapija sa nižom cenom novim tehnologijama čija je cena viša, efektima komplementarnosti, tj. novim i starim proizvodima koji se koriste istovremeno, i pružanjem terapije za bolesti za koje prethodno nisu bile dostupne terapije. Da bi se postigla finansijska stabilnost, potrebno je odgovoriti na dva ključna izazova. Prvi je odlučivanje o nivou raspoloživih resursa; drugi, osigurati optimalnu alokaciju resursa unutar ograničenih budžeta. Da bi bili relevantni za donošenje odluka u regionu CEE, vlade i HTA agencije moraju se baviti ovim ključnim izazovima.

Nakon nekoliko godina ograničenog odlučivanja u Srbiji i zaustavljenih rasprava postavlja se pitanje da li je budžet važniji od spašavanja i unapređenja kvaliteta života pacijenata?

## **THE VALUE OF INNOVATION IN HEALTH CARE DECISION MAKING**

**Tanja Novaković**

ZEM Solutions, Belgrade (Serbia)

Central and Eastern European (CEE) health care providers are faced with a number of challenges as a result of increased demand and rising expectations. As with many health systems across the world improving access to innovative technologies within increasingly constrained budgets is required. Limited decision-making and delays in evaluations of new technologies may have a significant health impact on patients.

Innovations can drive increased spending in health care as a result of substitution of lower priced products with new higher priced technologies, complementarity effects, i.e. new and old products used concurrently, and by providing treatments for conditions for which previously no treatments were available. To achieve financial stability, two key challenges need addressing. Firstly, deciding on the level of available resources; secondly, ensuring optimal resource allocation within finite budgets. To be relevant to decision making in the CEE region, governments and HTA agencies must address these key challenges.

After several years of limited decision making in Serbia and stalled discussions at what point is budget more important than saving and improving lives?

## ZAŠTO SISTEMATIČNI PREGLEDI LITERATURE?

**Mark Parker**

ZEM Solutions, Beograd (Srbija)

Randomizovane kontrolisane studije predstavljaju zlatni standard u definisanju kliničkih dokaza lečenja. Ove studije su dizajnirane da minimiziraju različite vrste pristrasnosti i druge probleme koji prate procenu kliničkog benefita. Međutim, ovakve studije su ograničene vremenom i prostorom, dosta je skupo da se realizuju. Klinička praksa, tzv., „real world“ je mnogo komplikovaniji od onog predstavljenog u studiji. Trenutna tehnološka dostignuća su dovela do eksplozije dostupnih dokaza koji su prikupljeni iz realnog života, iz bolnica i opšte prakse, i specifičnih registara pacijenata koji sadrže mnoštvo podataka o nizu terapija i primera iz prakse. Iako su ovi podaci značajni da se omogući znanje o pružanju zdravstvene zaštite, sposobnost obrade ovih podataka je još uvek u povoju. Cilj ovog predavanja je da se pomoći inovativnog alata koji predstavlja analizu „velikih podataka“ medicinske naučne literature pod originalnim nazivom *Publication Ocean*, pokažu problemi i rešenja izazova koji prate medicinu zasnovanu na dokazima (engl. *Evidence based medicine - EBM*). Pristup informacijama baziranim na dokazima je ključno za donošenje odluka koje koriste koristi (benefite) tehnologije da bi se postigla njena najbolja vrednost za određeno zdravstveno stanje i naj taj način da bi se postigao efikasan i kvalitetan zdravstveni sistem.

## **WHY SISTEMATYC REVIEWS?**

**Mark Parker**

ZEM Solutions, Belgrade (Serbia)

Randomised controlled trials represent the gold standard in defining clinical evidence for treatments. These trials are designed to minimise the various biases and other problems which accompany an assessment of clinical benefit. However, such trials are limited in time and place, extremely expensive to conduct and the real world is infinitely more complicated than is represented by the trials. Recent advancements in technology have resulted in an explosion of available evidence collected in real world settings, from hospital and general practice, to specific patient registries collecting a wealth of data on a range of treatments and practices. While this evidence is vital to support our knowledge of healthcare delivery, the ability to analyse it is still in its infancy. The goal of this lecture is to demonstrate problems and solutions to the challenges of evidence-based medicine (EBM) with the innovative tool „Publication Ocean“ which presents Big Data analytics of the Medical scientific literature. Access to evidence-based information is crucial for making decisions using the benefits of technology in order to achieve its best value for a particular health condition and in that way to achieve an efficient and qualitative health care system.

## **INHIBITORI KOTRANSPORTERA ZA NATRIJUM I GLUKOZU TIPO 2 KOD OBOLELIH OD DIJABETES MELITUSA TIPO 2 I SRČANE INSUFICIJENCIJE: KLINČKI POGLED NA TERAPIJU KOJA MOŽE DA SNIZI MORBIDITET I MORTALITET**

**Marija Polovina**

Klinika za kardiologiju Kliničkog centra Srbije, Univerzitet u Beogradu-  
Medicinski fakultet (Srbija)

Dijabetes melitus tipa 2 (T2DM) i srčana insuficijencija (SI) se smatraju pandemijama sa ozbiljnim javno-zdravstvenim posledicama. Kod pacijenata sa T2DM, SI je prisutna u 10 do 30% obolelih, a udruženo prisustvo obe bolesti je povezano sa 1,5-2 puta većim ukupnim i kardiovaskularnim mortalitetom i 2,5 puta većim rizikom od kardiovaskularnih hospitalizacija.

Optimalno lečenje T2DM u SI i dalje predstavlja klinički izazov. Primena preparata sulfonylureje i tiazolidinendiona se povezuje sa porastom mortaliteta kod osoba pod rizikom od SI ili sa već ispoljenom SI. Terapija insulinom dovodi do povećana telesne mase i retencije tečnosti što može da umanji pozitivne efekte dobre glikoregulacije u SI. Od novih oralnih hipoglikemika, inhibitor dipeptidil peptidaze-4 (DPP-4), saksagliptin, je povezan sa 27% višim rizikom za hospitalizaciju zbog pogoršanja SI, dok drugi DPP-4 inhibitori i inkretinski mimetici imaju neutralan efekat na SI.

U skorije vreme, u kliničku praksu je uvedena nova klasa oralnih hipoglikemika, a to su inhibitori kotransportera za natrijum i glukozu tipo 2 (SGLT-2 inhibitori). U do sada završenim randomizovanim studijama, primena empagliflozina i kanagliflozina bila je povezana sa značajnim sniženjem nepovoljnih kardiovaskularnih događaja kod pacijenata sa T2DM. Od posebnog je značaja da su oba leka bila povezana sa >30% nižim rizikom od hospitalizacije zbog pogoršanja SI. Takođe, u poređenju sa placebom, postignuto je i značajno sniženje rizika od novo-nastale SI, kao i smrtnost zbog SI kod pacijenata sa T2DM. Stoga se SGLT2 inhibitori ispituju u nekoliko randomizovanih studija kod bolesnika sa SI nezavisno od prisustva T2DM, sa ciljem utvrđivanja efikasnosti u snižavanju kardiovaskularnog mortaliteta i sprečavanju pogoršanja SI. Oboleli od T2DM i SI imaju visok rizik od nepovoljnih kardiovaskularnih događaja, a primena SGLT-2 inhibitora može da sniziti rizik od hospitalizacija zbog SI, kao i kardiovaskularni mortalitet. Iz kliničke perspektive, važno je revidirati politiku participacije kako bi ovi lekovi postali dostupniji većini obolelih.

## **SODIUM GLUCOSE CONTRANSPORTER-2 INHIBITORS IN TYPE-2 DIABETES AND HEART FAILURE: THE CLINICAL STANDPOINT ON TREATMENT THAT CAN REDUCE MORBIDITY AND MORTALITY**

**Marija Polovina**

Department of Cardiology, Clinical Center of Serbia, University of Belgrade – Faculty of Medicine (Serbia)

Type-2 diabetes mellitus (T2DM) and heart failure (HF) are considered pandemics with serious public health consequences. HF is a common comorbidity in T2DM, affecting 10-30% of individuals. Concomitant T2DM and HF portend 1.5 to 2-fold higher risk of all-cause and cardiovascular mortality, and increase the risk of cardiovascular hospitalization by 2.5-fold.

The optimal management of T2DM in HF remains challenging. Treatment with sulphonylureas and thiazolidinediones has been associated with increased mortality in patients at risk of, or with known HF. Insulin may promote weight gain and fluid retention, thereby potentially offsetting benefit of glycemic control in HF. Among the novel oral anti-diabetic medications, a dipeptidyl peptidase-4 (DPP-4) inhibitor, saxagliptin, has been associated with a 27% higher risk of hospitalization for worsening HF, while other DPP-4 inhibitors and incretin mimetics demonstrate a neutral effect on HF outcomes.

Recently, a new class of anti-diabetic medications, the sodium glucose co-transporter-2 (SGLT2) inhibitors, has been introduced. In the two completed randomized trials that, empagliflozin and canagliflozin have demonstrated a substantial reduction in adverse cardiovascular outcomes in T2DM patients. In particular, both medications significantly reduced the risk of HF hospitalization by >30%. Also, the risk of new-onset HF and HF-related mortality were significantly reduced with SGLT-2 inhibitors compared with placebo. Hence, several ongoing randomized trials, including patients with HF, are assessing the efficacy of SGLT2 inhibitors for the reduction of cardiovascular mortality or HF hospitalization irrespective of T2DM. In conclusion, patients with T2DM and HF are at high risk of adverse cardiovascular outcomes, and treatment with SGLT-2 inhibitors could decrease HF hospitalizations and cardiovascular death. From the clinical perspective, it is of great importance that reimbursement policies be revised so that these medications could be offered to the majority of the affected patients.

**NOVEL ANTIDIABETIC AGENTS AND CARDIOVASCULAR RISK****Manfredi Rizzo**

BioMedical Department of Internal Medicine and Medical Specialties, University of Palermo (Italy)

Numerous clinical trials have reported that treatment with novel classes of type 2 diabetes agents reduce and potentially prevent events in patients with type 2 diabetes (T2DM) and cardiovascular (CV) disease. The available scientific data indicates their effects on body weight and various cardiometabolic markers, such as lipids, blood pressure, inflammatory markers, oxidative stress, but also endothelial dysfunction, and subclinical atherosclerosis. A particular attention is on recent CV outcome studies (CVOTs) and the potential mechanisms beyond glucose-lowering effects. Briefly, empagliflozin, the sodium-glucose cotransporter (SGLT) 2 inhibitor, in the EMPA-REG CVOT study, markedly and rapidly reduced CV death and heart failure hospitalization. Similarly, liraglutide and semaglutide, the glucagon-like peptide (GLP)-1 receptor agonists, in the LEADER and SUSTAIN-6 CVOT study, respectively, reduced CV death and major adverse CV events, but did not influence heart failure risks, suggesting different underlying mechanisms. On the other hand, other GLP-1 receptor agonist, exenatide, in the EXSCEL study, met the goal of CV safety, but failed to show any significant CV benefit. We are waiting for the results from other 2 CVOTs, with the use of dulaglutide (REWIND study) and albiglutide (HARMONY study). However, the underlying mechanisms remain largely unknown. Recently, we have hypothesized the mechanism by which liraglutide may have direct effects on the atherosclerosis plaque, at the early stage of atherosclerosis, impacting its formation and progression. Based on the available CVOT results, liraglutide and semaglutide should be the preferred second-line medication in T2DM subjects in secondary CV prevention, as add-on to metformin therapy. All these novel therapeutic strategies allow customization of antidiabetic treatment to each patient's need, providing better metabolic control with reduced CV risk. Overall, their usage should be expanded in other cardiometabolic disorders as well.

## KONTINUIRANI SKOR ZA METABOLIČKI SINDROM U POPULACIJI DECE I ADOLESCENATA

**Rade Vuković<sup>1</sup>, Ivan Soldatović<sup>2</sup>, Tatjana Milenković<sup>1</sup>,  
Katarina Mitrović<sup>1</sup>, Sladana Todorović<sup>1</sup>, Ljiljana Plavšić<sup>1</sup>**

<sup>1</sup>Institut za zdravstvenu zaštitu majke i deteta „Dr Vukan Čupić”, Beograd,

<sup>2</sup>Univerzitet u Beogradu - Medicinski fakultet (Srbija)

Epidemija gojaznosti u populaciji mlađih u Srbiji predstavlja značajan javno zdravstveni problem, na šta ukazuje trostruki porast prevalencije gojaznosti među mladima u našoj zemlji tokom poslednjih dve decenije. Uporedo sa porastom prevalencije gojaznosti kod mlađih uočava se porast učestalosti i ranija pojava komplikacija gojaznosti kao što su tip 2 dijabetesa melitusa i metabolički sindrom. Definicija metaboličkog sindroma Internacionalne federacije za dijabetes (IDF) je dihotomnog karaktera, što za posledicu ima gubitak podataka i onemogućava kvantifikovanje metaboličkog sindroma, odnosno procenu težine stanja. Do sada korišćeni kontinuirani skorovi za metabolički sindrom poput sume Z skorova i PCA analize omogućavaju kvantifikovanje metaboličkog sindroma, ali su veoma kompleksni i zahtevaju upotrebu naprednih računarskih statističkih programa.

Novi kontinuirani skor za metabolički sindrom upopulaciji dece i adolescenata (PsiMS skor) računa se prema sledećoj formuli:  $(2 \times \text{obim struka (cm)} / \text{telesna visina (cm)}) + (\text{glikemija (mmol/l}) / 5,6) + (\text{trigliceridi (mmol/l}) / 1,7) + (\text{sistolni krvni pritisak (mmHg}) / 130) - (\text{HDL (mmol/l}) / 1,02$ . Na ovaj način izračunat skor ima odličnu korelaciju sa kompleksnim skorovima, a izuzetno je jednostavan za računanje, što ga čini pouzdanim i pogodnim za svakodnevnu primenu u kliničkoj praksi, kao i u kliničkim istraživanjima. PsiMS skor predstavlja praktičan i precizan dijagnostički metod za kvantifikovanje metaboličkog sindroma u populaciji gojazne dece i adolescenata.

## **CONTINUOUS METABOLIC SYNDROME SCORE FOR USE IN PEDIATRIC POPULATION**

**Rade Vuković<sup>1</sup>, Ivan Soldatović<sup>2</sup>, Tatjana Milenković<sup>1</sup>,  
Katarina Mitrović<sup>1</sup>, Sladana Todorović<sup>1</sup>, Ljiljana Plavšić<sup>1</sup>**

Mother and Child Health Care Institute „Dr Vukan Čupić”, Belgrade, <sup>2</sup>University of Belgrade – Faculty of Medicine (Serbia)

Rising prevalence of obesity in youth in Serbia emphasizes the public health issue of the obesity epidemics. An increase in the obesity comorbidities, such as type 2 diabetes and metabolic syndrome, as well as earlier occurrence, has also been noted in children and adolescents. The International Diabetes Federation (IDF) metabolic syndrome (MS) definition is dichotomous, which results in loss of data, without the possibility of quantifying the severity of the syndrome. Previously used continuous MS scores, such as sum of Z-scores and PCA analysis, overcame these issues allowing continuous quantification of MS, however these scores are complex, requiring advanced statistical software for calculation of the scores.

Novel continuous metabolic syndrome score for use in the population of children and adolescents (PsiMS score) is calculated using the following formula:  $(2 \times \text{waist circumference (cm)}) / \text{height (cm)} + (\text{glycemia (mmol/l}) / 5.6) + (\text{triglycerides (mmol/l}) / 1.7) + (\text{systolic blood pressure (mmHg}) / 130) - (\text{HDL cholesterol (mmol/l}) / 1.02)$ . PsiMS score correlates highly with more complex continuous MS scores, while being simple to calculate, making it a reliable and accurate score for the evaluation of MS in everyday clinical practice, as well as in clinical research. PsiMS score represents a practical and accurate diagnostic method for quantification of MS in the population of obese children and adolescents.

## OPSTRUKTIVNA APNEJA U SNU I KARDIOMETABOLIČKI RIZIK

Jelena Vekić

Katedra za medicinsku biohemiju, Univerzitet u Beogradu-Farmaceutski fakultet (Srbija)

Opstruktivna apneja u snu (OSA) je hronično progresivno oboljenje sa visokom prevalencijom u populaciji, koje, bez pravovremene dijagnoze i terapije, može dovesti do značajnih posledica po kvalitet života pacijenata. Razvoj ovog poremećaja je povezan sa gojaznošću, a karakteriše se rekurentnim epizodama potpunog ili parcijalnog kolapsa gornjih disajnih puteva tokom sna, što se manifestuje apnejom ili hipopnejom i učestalim buđenjima. Tokom epizoda respiratornog kolapsa dolazi do aktivacije simpatičkog nervnog sistema, što potencira vazokonstrikciju, te dovodi do razvoja hipertenzije i poremećaja srčanog rada. Procenjuje se da je 60-90% pacijenata sa OSA gojazno, a utvrđeno je da sa povećanjem telesne mase za 10% rizik za razvoj OSA raste 6 puta. OSA je čest komorbiditet kod pacijenata sa metaboličkim sindromom (MS) i kardiovaskularnim bolestima (KVB) i predstavlja faktor rizika za njihov nastanak. Studije su ukazale na vezu između MS i OSA, te je ovaj fenomen opisan kao poseban poremećaj - sindrom Z. Istraživanje uzročno-posledične veze između OSA i KVB je u velikoj meri otežano kompleksnom prirodnom samog oboljenja. Kardiometabolički rizik u OSA je udružen sa arterijskom hipertenzijom, insulinskog rezistencijom, endotelnom disfunkcijom, inflamacijom, dislipidemijom i oksidativnim stresom. Lečenje OSA se danas najefikasnije sprovodi neinvazivnom ventilacijom, pomoću uređaja koji obezbeđuje pozitivan pritisak u gornjim disajnim putevima (*continuous positive airway pressure, CPAP*) i na taj način sprečava pojavu apneja tokom spavanja. Rezultati kliničkih studija su pokazali da CPAP terapija značajno poboljšava hemodinamske parametre, reguliše hipertenziju, povećava osjetljivost na insulin i koriguje dislipidemiju. Buduća istraživanja bi trebalo da rasvetle da li je apneja u snu faktor rizika za KVB pre ili je ta veza posledica šireg patofiziološkog procesa, čiji je deo i OSA.

## **OBSTRUCTIVE SLEEP APNEA AND CARDIOMETABOLIC RISK**

**Jelena Vekić**

Department of Medical Biochemistry, University of Belgrade-Faculty of Pharmacy (Serbia)

Obstructive sleep apnea (OSA) is a chronic, progressive disorder with a high prevalence in the population. Without timely diagnosis and therapy OSA can significantly affect the quality of life of the patients. Development of OSA is associated with obesity. It is characterized by recurrent episodes of complete or partial collapse of the upper airway during sleep, which is manifested by apnea or hypopnoea and frequent waking. During the episodes of respiratory collapse activated sympathetic nervous system exacerbates vasoconstriction, leading to development of hypertension and cardiovascular disorders. It is estimated that 60-90% of patients with OSA are obese. Also, the risk for developing OSA increases six times if body weight increases by 10%. OSA is a common co-morbidity in patients with metabolic syndrome (MS) and cardiovascular disease (CVD) and is a risk factor for their development. Studies have pointed to the relationship between MS and OSA, and this phenomenon was described as syndrome Z. Investigation of the causal relationship between OSA and CVD has been greatly confounded by the complex nature of the disease itself. Cardiometabolic risk in OSA is associated with arterial hypertension, insulin resistance, endothelial dysfunction, inflammation, dyslipidemia, and oxidative stress. The treatment of OSA is now most effectively performed by continuous positive airway pressure (CPAP), a type of non-invasive ventilation which prevents the onset of sleep apnea. The results of clinical studies have shown that CPAP therapy significantly improves haemodynamic parameters, regulates hypertension, increases insulin sensitivity, and corrects dyslipidemia. Future investigations should clarify whether sleep apnea is a risk factor for CVD per se or is a consequence of a broader pathophysiological process, of which OSA is part.

## **GENETIČKA ISPITIVANJA U METABOLIČKOM SINDROMU**

**Ana Ninić**

Katedra za medicinsku biohemiju, Univerzitet u Beogradu-Farmaceutski fakultet (Srbija)

Metabolički sindrom (MetS) je heterogeni poremećaj koji se karakteriše višestrukim metaboličkim abnormalnostima kao što su abdominalna gojaznost, dislipidemija, insulinska rezistencija, hipertenzija i poremećena tolerancija glukoze. Ovo proaterogeno i proinflamatorno stanje, kao glavni faktor rizika za tip 2 dijabetes melitus i kardiovaskularne bolesti, nastaje pod uticajem genetičkih faktora i faktora životne sredine. Mnoge genetičke varijante zajedno i sa faktorima životne sredine mogu doprineti razvoju MetS. Genomske studije su utvrdile jaku vezu između regiona hromozoma 1q41, 2p22.3, 3q27, 7q31.3, 9p13.1, 9q21.1, 10p11.2, 17p12 i 19q13.4 i kliničkih markera (telesne mase, obima struka i kuka, leptina, insulina). 99,9% ljudskog genoma je identično za sve ljude, tako da su genomske studije asocijacije fokusirane na genetičke varijacije kao što su polimorfizmi pojedinačnih nukleotida (SNP). Genomske studije asocijacije su testirale veliki broj SNP-ova kao faktora rizika za MetS. Identifikovale su značajne SNP-ove koji predstavljaju 15 lokusa povezanih sa jednom ili pet komponenata MetS. Varijante gena adiponektina i rezistina se sve više proučavaju sa ciljem da se pokaže koja od njih ili zajedno najviše utiču na nastanak MetS. Mnoge od njih su i povezane sa ispoljavanjem MetS. Takođe, nekoliko SNP-ova povezanih sa raspodelom telesnih masti pokazalo je snažnu interakciju sa faktorima životne sredine i efekte na ispoljavanje MetS. Mnoge studije su izvedene kako bi se utvrdilo koje varijante imaju jak efekat interakcije sa faktorima okoline, ali njihovi rezultati nisu bili konzistentni. Profili ekspresije gena uključenih u energetsku i metaboličku homeostazu su, takođe, intenzivno ispitivani. Iako je poznato da je MetS poligenski poremećaj sa svakom genetičkom varijantom koja doprinosi malim efektima, nauka i dalje ima veliki zadatak da identificuje uzročne gene i njihove efekte na rizik za razvoj MetS.

## **GENETIC TESTING FOR METABOLIC SYNDROME**

**Ana Ninić**

Department of Medical Biochemistry, University of Belgrade-Faculty of Pharmacy (Serbia)

Metabolic syndrome (MetS) is heterogenous disorder characterized by the multiple metabolic abnormalities such as abdominal obesity, dyslipidemia, insulin resistance, hypertension and impaired glucose tolerance. This proatherogenic and proinflammatory state, as a major risk factor for type 2 diabetes mellitus and cardiovascular disease, is determined by the genetic and environmental factors. Many genetic variants together and with environmental factors may contribute to MetS development. Genome studies have confirmed strong link between chromosome regions 1q41, 2p22.3, 3q27, 7q31.3, 9p13.1, 9q21.1, 10p11.2, 17p12, and 19q13.4 and clinical markers (weight, waistand hip circumferences, leptin, insulin). 99.9% of human genome is the same for all people, so genetic association studies are focused on genetic variations such as single nucleotide polymorphisms (SNPs). Genome-wide association studies tested a large number of SNPs for association with risk forMetS. They identified significant SNPs representing 15 loci associated either to one or five MetS components. Adiponectin and resistin gene variants are extensively studied in order to determine which of them or together mostly influence MetS development. Many of them were, indeed, related to MetS occurrence. Also, several SNPs associated to body fat distribution showed a strong interaction with environmental factors. Many studies were performed in order to determine which variants had a strong interaction effect with environmental factors but their results were inconsistent. Gene expression patterns of genes involved in energy and metabolic homeostasis have been also extensively studied. Although it is known that MetS is polygenic disorder with each genetic variant contributing with small effects, science still have a major task to identify the causal genes and their effects on risk for MetS development.

## SKOR DISLIPIDEMIJE, INFLAMACIJE I OKSIDATIVNOG STRESA U PROCENI KARDIOVASKULARNOG RIZIKA

**Jelena Kotur-Stevuljević, Nataša Bogavac-Stanojević, Jelena Vekić,  
Vesna Kalimanovska-Spasojević, Zorana Jelić-Ivanović, Slavica Spasić**

Katedra za medicinsku biohemiju, Univerzitet u Beogradu-Farmaceutski fakultet (Srbija)

Kod oko 20% pacijenata koji dožive akutno ispoljavanje KVB (akutni infarkt miokarda, moždani udar), kardiovaskularni rizik procenjen preko tradicionalnih faktora rizika je mali ili umeren. Kod trećine ovih osoba bi mogla da se popravi predikcija rizika uključivanjem odnosa urinarnog albumina/kreatinina ili koncentracije hsCRP. Ovi podaci ukazuju da je značajno ispitivati prisustvo takozvanih netradicionalnih faktora rizika, pre svega biohemijskih biomarkera (markera u krvi). Predmet ovog istraživanja je grupa biomarkera, biohemijskih parametara od kojih neki spadaju u tradicionalne faktore rizika (parametri lipidnog statusa), neki se ubrajaju u nove biomarkere čiji je značaj ispitana, potvrđen u velikim studijama i čak uključen u neke kalkulture kardiovaskularnog rizika (marker inflamacije - hsCRP), a neki još uvek nisu potvrđeni kao nezavisni faktori rizika za kardiovaskularne bolesti (markeri oksidativno-stresnog statusa). Cilj ovog rada je prikaz načina računanja zbirnog kardiovaskularnog rizika koji uključuje tri vrste biohemijskih markera: dislipidemiju, inflamaciju i oksidativni stres. Statistička metoda koja je primenjena da bi se računao rizik koji potiče od svakog od faktora je Z-skor (standardni skor) statistika. Skor dislipidemije se računa kao razlika između srednje vrednosti Z skorova za koncentracije LDL-holesterola i triglicerida i Z-skora za HDL-holesterol. Inflamatorni skor se računa kao Z skor za hsCRP. Oksidativno stresni skor je razlika između prooksidativnog skora (srednja vrednost Z skorova prooksidativnih parametara) i antioksidativnog skora (srednja vrednost Z skorova antioksidativnih parametara). Ukupni kardiovaskularni skor rizika (KVSR) je zbir dislipidemija skora, inflamatornog skora i oksidativno-stresnog skora, nazvan DOI skor (dislipidemija, oksidativni stres, inflamacija). Populacione srednje vrednosti i standardne devijacije su dobijene iz baze podataka zdravih sredovečnih osoba čiji su uzorci analizirani tokom prethodnih 16 godina u našoj laboratoriji. Kao ilustracija ovako izračunatog KVSR biće prikazane vrednosti pojedinih elemenata skora u populacijama pacijenata sa akutnim infarktom miokarda i moždanim udarom.

## DYSLIPIDEMIA, INFLAMMATION AND OXIDATIVE STRESS SCORE IN CARDIOVASCULAR RISK ESTIMATION

**Jelena Kotur-Stevuljević, Nataša Bogavac-Stanojević, Jelena Vekić,  
Vesna Kalimanovska-Spasojević, Zorana Jelić-Ivanović, Slavica Spasić**

Department of Medical Biochemistry, University of Belgrade-Faculty of Pharmacy (Serbia)

In about 20% of patients with acute cardiovascular (CV) event (acute myocardial infarction, stroke), cardiovascular risk estimated through the traditional risk factors is small or moderate. In one third persons with this underscored risk the risk prediction could be improved by urinary albumin/creatinine rate or hsCRP inclusion. These data suggest that it is important non-traditional risk factors assessment, especially blood-born markers – biochemical parameters. Theme of this investigation are biomarkers' group, some belongs to the traditional risk factors (lipid status parameters), some to new, emerging biomarkers but with documented importance and predictive capability in CV disease, even implemented in risk calculators (inflammation markers – hsCRP), and some still not confirmed as independent factors for CV disease (oxidative stress markers). The aim of this paper is the explanation of the logic and the equation for the summary CV risk calculation, which includes three different kinds of the biochemical markers: dyslipidemia, inflammation and oxidative stress. Statistical method is based on the Z-score (standardized score statistics). According to our proposal dyslipidemia score is calculated as the difference between mean of the Z-scores for the LDL-cholesterol and triglycerides and Z-score for the HDL-cholesterol. Inflammatory score is Z-score for the hsCRP. Oxy score is difference between prooxidative score (mean of the Z-scores for the different prooxidants) and the antioxidative score (mean of the Z-scores for the antioxidative parameters). Summary CV risk score (CVRS) is sum of the dyslipidemia score, inflammatory score and oxy score, entitled DOI score (**dyslipidemia, oxidative stress, inflammation**). Population means and standard deviations values are calculated from our database generated during the last 16 years in our laboratory from the samples of the healthy middle-aged subjects. As an illustration for the DOI score calculation here will be presented results for the patients with acute myocardial infarction and acute ischemic stroke.

## **HRONIČNA TERAPIJA – OČEKIVANJA I ZABRINUTOST NAŠIH PACIJENATA**

**Branislava Miljković**

Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu-  
Farmaceutski fakultet (Srbija)

Stavovi pacijenta i njegovo uključivanje u proces farmaceutske zdravstvene zaštite mogu značajno uticati na adherencu i ishode terapije. Cilj studije bio je ispitati očekivanja i zabrinutost pacijenata na hroničnoj terapiji i starijih pacijenata na polifarmaciji. Prospektivna, opservaciona studija u trajanju od 4 meseca je sprovedena u apotekama na teritoriji Srbije. Svaki farmaceut je regrutovao 10 uzastopnih odraslih pacijenata sa uvedenim lekom za hroničnu terapiju i/ili 10 starijih pacijenata na polifarmaciji od kojih je traženo da popune formular (7 pitanja koji obuhvataju: znanje, očekivanja, problem, zabrinutost, i razlog za prestanak terapije). Druga zakazana konsultacija zasnovana na odgovorima pacijenata sprovedna je nakon 2-4 sedmice. Primenjena je deskriptivna statistika.

Popunjenu dokumentaciju je poslalo 44 (od 73) farmaceuta za 391 pacijenta na hroničnoj terapiji i 440 starijih pacijenata na polifarmaciji. Većina pacijenata na hroničnoj terapiji i starijih pacijenata na polifarmaciji je bila zainteresovana da dobije dodatne informacije (85% vs 59%, redom). Najčešće kategorije očekivanja kod pacijenata na hroničnoj terapiji i starijih pacijenata na polifarmaciji su: kontrola zdravstvenog stanja (46,36% vs 36,1%), unapređenje kvaliteta života (24,32% vs 28,4%) i efektivnost (9,32% vs 15,7%), redom. Polovina pacijenata na hroničnoj terapiji i starijih pacijenata na polifarmaciji iskazali su zabrinutost u vezi sa terapijom (55% vs 46%, redom). Najzastupljenija kategorija zabrinutosti kod pacijenata na hroničnoj terapiji i starijih pacijenata na polifarmaciji bila je bezbednost terapije (32,5% vs 19,8%), dužina trajanja terapije (9,8% vs 6,1%), neefikasnost (3,1% vs 4,5%), redom. Savetovanje pacijenata unapredilo je razumevanje primene terapije za 59,0% pacijenata na hroničnoj terapiji i 75,5% starijih pacijenata na polifarmaciji. Strukturirano savetovanje zasnovano na identifikovanim potrebama i očekivanjima pacijenata može unaprediti pravilnu primenu lekova kod pacijenata koji započinju hroničnu terapiju i kod starijih pacijenata na polifarmaciji.

*Istraživanje je realizovano u okviru Projekta 175023 finansiranog od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije.*

## **CHRONIC THERAPY – EXPECTATIONS AND CONCERNS OF OUR PATIENTS**

**Branislava Miljković**

Department of Pharmacokinetics and Clinical Pharmacy, University of Belgrade-Faculty of Pharmacy (Serbia)

Patients' attitudes and involvement in the process of pharmaceutical care could substantially influence the adherence and outcomes of therapy. The aim of the study was to explore patients' expectations and concerns with medication regarding chronic therapy and polypharmacy.

A four-month prospective observational study was conducted in community pharmacies across Serbia. Every pharmacist recruited 10 consecutive adult patients with the new medication for chronic conditions and/or 10 elderly polypharmacy patients who were asked to fill in the Checklist (7 questions covering: knowledge, expectations, problems, concerns and reason to stop treatment). The consultation appointment based on the patient's answers was scheduled within two-to-four weeks. Descriptive statistical analyses was performed.

Forty-four pharmacists sent complete documentation from 391 patients on chronic therapy and 440 elderly polypharmacy patients. Majority of the patients on chronic therapy and elderly polypharmacy patients were interested to receive additional information (85% vs 59%, respectively). The most prevalent expectations category in patients on chronic therapy and elderly polypharmacy patients were: control of the condition (46.36% vs 36.1%), quality of life improvement (24.32% vs 28.4%) and effectiveness (9.32% vs 15.7%), respectively. The half of the patients on chronic therapy and elderly polypharmacy patients was concerned about the therapy (55% vs 46%, respectively). The most prevalent concerns categories among the patients on chronic therapy and elderly polypharmacy patients were: side effects (32.5% vs 19.8%), duration of therapy (9.8% vs 6.1%), ineffectiveness (3.1% vs 4.5%), respectively. The counselling with pharmacists improved patients' understanding of the medication use in 59.0% patients on chronic therapy and in 75.5% elderly polypharmacy patients. Structured pharmacist counselling based on identified patients' concerns and needs could improve the appropriate use of therapy in patients starting with chronic therapy and elderly polypharmacy patients.

*This work was conducted as a part of the Project No. 175023 funded by the Ministry of Education, Science and Technological Development, Republic of Serbia.*

## FARMACEUTI U SRBIJI IDENTIFIKUJU TERAPIJSKE PROBLEME KOD STARIJIH PACIJENATA - KOJE, KAKO, KOLIKO?

Sandra Vezmar Kovačević

Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu-  
Farmaceutski fakultet (Srbija)

U kliničkoj praksi razvijenih zemalja uočeni su brojni terapijski problemi kod starijih pacijenata. Cilj istraživanja je bio da se utvrdi da li, i u kojoj meri farmaceuti u Srbiji identifikuju terapijske probleme kod starijih pacijenata u poređenju sa razvijenim zemljama. Dve studije preseka sprovedene su u apotekama primarne zdravstvene zaštite u periodu od dve godine u cilju identifikacije terapijskih problema kod starijih pacijenata. U okviru jedne studije korišćeni su STOPP/START kriterijumi za identifikaciju terapijskih problema dok su u drugoj studiji farmaceuti sprovodili klinički pregled lekova.

U istraživanju je učestvovalo 49 farmaceuta i 897 pacijenata, prosečne starosti  $73,1 \pm 6,4$  godina, od kojih je 56,8% bilo ženskog pola. U proseku su pacijenti primenjivali  $6,2 \pm 2,1$  leka, dok su najčešće indikacije za primenu lekova bile arterijska hipertenzija (91,4%), dijabetes melitus (36,3%), primarna prevencija kardiovaskularnog događaja (16,8%) i srčana insuficijencija (12,8%). Ukupno je identifikovano 1.567 problema u vezi sa terapijom (u proseku  $1,8 \pm 1,3$  po pacijentu). Značajno veći broj problema u vezi sa terapijom je identifikovani pri kliničkom pregledu lekova ( $p < 0,001$ ). Neodgovarajuća primena nesteroidnih antiinflamatornih lekova u terapiji bola (34,9%), dugotrajna primena benzodiazepina (27,7%) i neodgovarajuća primena acetilsalicilne kiseline u primarnoj prevenciji kardiovaskularnih događaja (22,5%) bili su najčešće identifikovani terapijski problemi sa prisutnim lekovima u obe studije. Takođe, identifikovana je nedovoljna primena statina u terapiji dijabetes melitusa (44,3%), acetilsalicilne kiseline u sekundarnoj prevenciji kardiovaskularnih događaja (38,6%) i inhibitora protonskе pumpe u cilju zaštite sluznice gastrointestinalnog trakta (22,6%). U poređenju sa istraživanjima u razvijenim zemljama, farmaceuti u Republici Srbiji su identifikovali slične terapijske probleme, ali je u našoj zemlji češće identifikovan nedostatak pojedinih lekova u terapiji, posebno statina i acetilsalicilne kiseline. Problemi u vezi sa terapijom pacijenata su često zastupljeni u našoj kliničkoj praksi, a farmaceuti su kompetentni stručnjaci koji mogu učestvovati u njihovoј identifikaciji i rešavanju.

*Istraživanje je realizovano u okviru Projekta 175023 finansiranog od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije.*

# **PHARMACISTS IN SERBIA IDENTIFY DRUG-RELATED PROBLEMS IN ELDERLY PATIENTS - WHICH, HOW, HOW MANY?**

**Sandra Vezmar Kovačević**

Department of Pharmacokinetics and Clinical Pharmacy, University of Belgrade-Faculty of Pharmacy (Serbia)

Many drug-related problems (DRPs) have been reported in older patients in the ambulatory care setting. The goal of this research was to establish if, and to what extent, pharmacists in Serbia participate in identifying DRPs in older patients, compared to developed countries.

Two cross-sectional studies were conducted in primary care pharmacies during two years with the aim of identifying DRPs in older patients. STOPP/START criteria were used in one study, whereas clinical medication review was performed to identify DRPs in the other.

Fourty-nine pharmacists participated in the research as well as 897 patients, average age  $73.1 \pm 6.4$ , of whom 56.8% were female. In average, patients were treated with  $6.2 \pm 2.1$  medications, most commonly for arterial hypertension (91.4%), diabetes mellitus (36.3%), primary prevention of cardiovascular events (16.8%) and heart failure (12.8%). In total, 1.567 DRPs were identified (in average  $1.8 \pm 1.3$  per patient). Significantly more DRPs were identified by clinical medication review ( $p < 0.001$ ). Inappropriate use of nonsteroidal anti-inflammatory drugs in the treatment of pain (34.9%), long-term use of benzodiazepines (27.7%) and inappropriate use of aspirin in primary prevention of cardiovascular events (22.5%) were the most frequently identified DRPs with existing treatment in both studies. Moreover, lack of use of statins in the treatment of diabetes mellitus (44.3%), aspirin in secondary prevention of cardiovascular events (38.6%) and proton pump inhibitors for gastrointestinal protection (22.6%) were commonly identified. Compared to studies in developed countries, pharmacists in Serbia identified similar DRPs but the occurrence of treatment omission was more frequent, particularly associated with the use of statins and aspirin. DRPs occur frequently in the primary care setting and pharmacists are competent professionals who can participate in their identification and solving.

*This work was conducted as a part of the Project No. 175023 funded by the Ministry of Education, Science and Technological Development, Republic of Serbia.*

## PRIMENA KONCEPTA FARMACEUTSKE ZDRAVSTVENE ZAŠTITE KOD PACIJENATA SA ASTMOM I HOBP – MODEL PRIMARNE ZDRAVSTVENE ZAŠTITE

Milena Kovačević

Katedra za farmakokinetiku i kliničku farmaciju, Univerzitet u Beogradu-  
Farmaceutski fakultet (Srbija)

Model primarne zdravstvene zaštite prepoznat je kao veoma značajna strategija za poboljšanje ishoda pacijenata sa hroničnim oboljenjima. Cilj studije bila je procena uticaja farmaceuta na implementaciju i efektivnost modela samokontrole bolesti kod pacijenata sa astmom i hroničnom opstruktivnom bolešću pluća (HOBP).

Prospektivna studija sprovedena u periodu april-septembar 2015. uključila je dvanaest samostalnih javnih apoteka/apotekarskih ustanova. Inicijalno, pacijenti su popunjavali zadate upitnike, nakon čega im je pruženo strukturirano savetovanje od strane farmaceuta. Nakon 3 meseca pacijenti su ponovo popunjavali validirane upitnike: (1) *Morisky 8-item*; (2) *Beliefs about Medicines*; (3) *Knowledge of Asthma and Asthma Medicines; Bristol COPD Knowledge*; (4) *Asthma Control Test; COPD Assessment Test i Modified Medical Research Council Dyspnea Scale*.

U studiji je učestvovalo 90 pacijenata sa astmom i 83 sa HOBP. Procenat pacijenata sa dobrom kontrolom astme povećan je za 18%, dok je za HOBP iznosio 6%, odnosno 12%, procenjeno preko CAT i mMRC skora, respektivno ( $p<0,05$ ). Broj pacijenata sa niskim stepenom adherence smanjen je za 14% u astmi i 17% u grupi sa HOBP ( $p<0,05$ ). Pacijenti su pokazali unapređenje znanja o bolesti i lekovima, sa 58,7% na 73,6% kod astme, i 41,6% na 63,1% tačnih odgovora za HOBP ( $p<0,05$ ). Značajan rezultat bilo je i smanjenje zabrinutosti pacijenta u pogledu štetnih efekata terapije, dok je uverenje o koristi primene terapije unapređeno ( $p<0,05$ ). Uticaj uverenja pacijenata identifikovan je kao bolji prediktor stepena adherence u brojnim studijama, nego što su to sociodemografski ili klinički faktori pacijenta.

Strukturirano savetovanje od strane farmaceuta dovelo je do povećanja stepena adherence, unapređenja znanja o bolesti i lekovima, unapređenja stavova pacijenata u vezi sa primenom terapije, i konačno do poboljšanja kontrole astme i HOBP. Pacijentima je pružena podrška za sprovođenje samokontrole bolesti edukacijom, što je dovelo do poboljšanja ishoda. Model primarne farmaceutske zdravstvene zaštite pokazan je kao efektivan, sa dodatnom prednošću lako dostupne zdravstvene zaštite.

*Istraživanje je realizovano u okviru projekta 175023 finansiranog od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije.*

# **PHARMACEUTICAL CARE MODEL IN THE COMMUNITY PHARMACY SETTINGS – FOCUS ON ASTHMA AND COPD PATIENTS**

**Milena Kovačević**

Department of Pharmacokinetics and Clinical Pharmacy, University of Belgrade-Faculty of Pharmacy (Serbia)

The primary care model has been recognized as a valuable strategy to improve the quality of chronic disease management. The study sought to assess the impact of community pharmacist education on patients self-management feasibility.

A prospective study was conducted from April 1st to September 30th 2015 in 12 independent/chain pharmacies in Serbia. The main inclusion criteria was a prescription for asthma/COPD medicine. After recruiting, patients filled out the questionnaires, received a structured counselling, and after three months completed the questionnaires again. Four groups of validated questionnaires were used: (1) Morisky 8-item; (2) Beliefs about Medicines; (3) Knowledge of Asthma and Asthma Medicines; Bristol COPD Knowledge; (4) Asthma Control Test; COPD Assessment Test and Modified Medical Research Council Dyspnea Scale.

A total of 90 asthma and 83 COPD patients enrolled the study. The proportion of patients with controlled asthma increased for 18%, and controlled COPD for 6%, or 12%, regarding to CAT and mMRC score, respectively (all  $p<0.05$ ). The proportion of low adherence decreased for 14% in asthma, and 17% in COPD patients ( $p<0.05$ ). Patients improved their knowledge on the disease and medications - in asthma from 58.7% to 73.6%, whereas in COPD from 41.6% to 63.1% of correct answers ( $p<0.05$ ). A decrease in harm and concern score, as well as an increase in necessity score was observed in both asthma and COPD patients ( $p<0.05$ ). It was shown previously that patients' beliefs were much stronger adherence predictors, than sociodemographic or social factors.

Structured pharmacist-delivered counselling resulted in improvement of adherence level, knowledge on the disease and medications used, beliefs and attitude towards the therapy, and finally in better disease control. Improved patients outcomes were achieved through self-management support. Community pharmacists are the most accessible health professionals, able to provide necessary information and empower patients to control their disease.

*This work was conducted as a part of the Project No. 175023 funded by the Ministry of Education, Science and Technological Development, Republic of Serbia.*

## **STOPP/START KRITERIJUMI ZA OPTIMIZACIJU TERAPIJE U GERIJATRIJSKOJ POPULACIJI**

**Aleksandra Catić-Đorđević, Nikola Stefanović,  
Radmila Veličković-Radovanović**

Katedra za farmaciju, Univerzitet u Nišu – Medicinski fakultet (Srbija)

Starenje prati pojava komorbiditeta i posledično uvođenje većeg broja lekova u terapiju. Potencijal za negativne ishode terapije i pojavu problema udruženih sa primenom lekova usled polipragmazije je poznati fenomen u gerijatriji. Kriterijumi za procenu propisane terapije starijim osobama (STOPP) i kriterijumi za upozorenje lekarima o potrebnom tretmanu starijih (START) razvijeni su kako bi se omogućilo identifikovanje potencijalno neadekvatno propisane terapije i uočila potreba za dopunom terapije. Cilj ovog rada bio je procena pojave polifarmacije i potencijalnih neadekvatno propisanih lekova kod ambulantnih (OP) i starijih pacijenata koji su smešteni u dom za brigu o starima (RHP) upotrebom STOPP/START kriterijuma. Izvršena je i procena stepena adherence ispitanika.

Upotrebom STOPP/START kriterijuma obavljen je pregled terapije 45 pacijenata smeštenih u Gerontološkom centru Niš, u Nišu, Srbija i 60 ambulantnih pacijenata. Kod svih pacijenata stepen adherence utvrđen je upotrebom Morisky 8 skale. Dobijeni podaci obrađeni su pomoću SPSS softvera.

RHP su pokazali veći broj lekova u terapiji ( $8,11 \pm 3,59$  vs.  $3,67 \pm 1,40$ ,  $p=0,001$ ), prisutnih komorbiditeta ( $2,8 \pm 1,34$  vs.  $1,72 \pm 0,85$ ,  $p=0,001$ ), kao i STOPP (26 vs. 9,  $p=0,001$ ) i START (11 vs. 2,  $p=0,002$ ) kriterijuma. Ipak, gledajući broj STOPP kriterijuma po broju propisanih lekova, učestalost STOPP kriterijuma je manja kod RHP ( $0,18 \pm 0,14$  vs.  $0,23 \pm 0,05$ ,  $p=0,035$ ). Za START kriterijume nije nađena značajna razlika između posmatranih grupa. Grupa RHP je pokazala viši stepen adherence u odnosu na OP ( $p=0,001$ ). Bez obzira na uočen veći broj lekova i komorbiditeta kod pacijenata smeštenih u domu za stare, učestalost uočavanja STOPP kriterijuma po broju propisanih lekova je manji uz istovremeno viši stepen adherence u odnosu na grupu ambulantnih pacijenata. Konstantan medicinski nadzor je važan u dobijanju predviđenih zdravstvenih ishoda, a pregled lekova od strane farmaceuta može pomoći u rukovođenju terapijom i vodi njenoj optimizaciji u svakodnevnoj kliničkoj praksi.

## **STOPP/START CRITERIA FOR OPTIMIZATION OF PHARMACOTHERAPY IN ELDERLY**

**Aleksandra Catić-Đorđević, Nikola Stefanović,  
Radmila Veličković-Radovanović**

Department of Pharmacy, University of Niš - Faculty of Medicine (Serbia)

The aging is often associated with comorbidities and following the introduction of numerous drugs in everyday therapy. The potential for negative outcomes and drug-related problems regarding multiple medications in older people is well documented. Screening Tool of Older Person's Prescriptions (STOPP) and Screening Tool to Alert doctors to Right Treatment (START) have been developed to identify potential inappropriate prescriptions and prescribing omissions. The aim was to measure the prevalence rates of polypharmacy and potential inappropriate prescriptions in residential home care patients (RHP) and outpatients (OP) over 65 using STOPP/START criteria. Secondly, we evaluated the level of medication adherence between those two groups of patients.

We reviewed therapy of 45 RHP in Gerontological center Nis, Serbia, and 60 OP, using the STOPP/START criteria. Also, we determined their level of medication adherence by Morisky 8 scale. All data were statistically processed by SPSS software.

RHP had higher number of drugs ( $8.11 \pm 3.59$  vs.  $3.67 \pm 1.40$ ,  $p=0.001$ ), more comorbidities ( $2.8 \pm 1.34$  vs.  $1.72 \pm 0.85$ ,  $p=0.001$ ), more patients identified with STOPP (26 vs. 9,  $p=0.001$ ) and START (11 vs. 2,  $p=0.002$ ) criteria. Still, RHP had a significantly lower rate of STOPP criteria per drug ( $0.18 \pm 0.14$  vs.  $0.23 \pm 0.05$ ,  $p=0.035$ ). There was no difference in START criteria per drug between defined groups. Additionally, RHP had higher level of medication adherence compared to OP ( $p=0.001$ ).

In spite of the presence of polypharmacy in older patients in the residential home center and the number of STOPP criteria, a rate of STOPP criteria per drug was low and level of medication adherence was high. Constant medical supervision is important for health outcomes, but medication review by a pharmacist can be a reliable tool for optimization and management of pharmacotherapy in everyday practice.

## **REGULATIVA O DODACIMA ISHRANI**

**Ivan Stanković**

Katedra za bromatologiju, Univerzitet u Beogradu-Farmaceutski fakultet  
(Srbija)

Dodaci ishrani (dijetetski suplementi) su namirnice koje dopunjuju normalnu ishranu i predstavljaju koncentrovane izvore vitamina, minerala ili drugih supstanci sa hranljivim ili fiziološkim efektom, pojedinačno ili u kombinaciji, a u prometu su u doziranim oblicima dizajnirane da se uzimaju u odmerenim pojedinačnim količinama (kapsule, tablete, kesice praška, ampute tečnosti, boćice za doziranje u kapima i sl.). U EU dodaci ishrani regulisani su Direktivom 2002/46/EC i njenim dopunama. EU regulativa koja još uvek nije kompletna sadrži listu dozvoljenih vitamina i minerala, jedinice za njihovo označavanje, listu supstanci koje se mogu koristiti kao njihovi izvori i posebne zahteve za deklarisanje dodataka ishrani. Na nivou EU još uvek nisu harmonizovane maksimalno dozvoljene količine vitamina i minerala u dnevnoj dozi dijetetskih suplemenata, ostale supstance sa nutritivnim ili fiziološkim efektom kao ni biljni dijetetski suplementi i za njih se primenjuju nacionalni propisi zemalja članica koji se razlikuju. Naš Pravilnik o zdravstvenoj ispravnosti dijetetskih proizvoda („Sl. Glasnik RS“ br. 45/2010, 27/2011, 50/2012, 21/2015, 75/2015 i 07/2017) usklađen je sa regulativom EU u delu koji se odnosi na dodatke ishrani. Propisana je obavezna procedura notifikacija, odnosno upis u bazu podataka koju vodi Ministarstvo zdravlja RS koji se vrši za period od 5 godina, a uključuje dobijanje stručnog mišljenja, kategorizacije i odobrenja teksta deklaracije od strane Farmaceutskog fakulteta, stručnog mišljenja o zdravstvenoj ispravnosti od ovlašćenih laboratorija Zavoda za javno zdravlje ili Vojnomedicinske akademije i podnošenje zahteva sa kompletном dokumentacijom Ministarstvu zdravlja Republike Srbije. Predviđenom izmenom Zakona o bezbednosti hrane biće omogućeno regulisanje dodataka ishrani posebnim propisom koji je potrebno usklađivati za izmenama i dopunama regulative EU o dodacima ishrani.

## **REGULATION ON FOOD SUPPLEMENTS**

**Ivan Stanković**

Department of Bromatology, University of Belgrade-Faculty of Pharmacy  
(Serbia)

Food supplements are foodstuffs the purpose of which is to supplement the normal diet and which are concentrated sources of nutrients or other substances with a nutritional or physiological effect, alone or in combination, marketed in dose form, namely forms such as capsules, tablets, sachets of powder, ampoules of liquids, drop dispensing bottles, and other similar forms of liquids and powders designed to be taken in measured small unit quantities. In the EU, food supplements are regulated by the Directive 2002/46/EC and its amendments. EU regulation on food supplements, that is not yet completed, includes a list of permitted vitamins and minerals, their labeling units, a list of substances that can be used as their sources and specific requirements for the labeling of food supplements. At the EU level, the maximum allowable amounts of vitamins and minerals in the daily dose of dietary supplements, other substances with nutritional or physiological effects, as well as herbal dietary supplements, are still not harmonized and they are subject to the national regulations of the Member States. Our rule book on the health safety of dietary products („Official Gazette of RS“ No. 45/2010, 27/2011, 50/2012, 21/2015, 75/2015 and 7/2017) is in line with the EU regulation in the part referring to food supplements. The mandatory notification procedure for a period of 5 years includes the obtaining of expert opinion, categorization and approval of the text on the label by the Faculty of Pharmacy, opinion on the food safety from the authorized laboratories of the Institutes for Public Health or Military Medical Academy and submitting of application with complete documentation to the Ministry of Health of the Republic of Serbia.

The foreseen amendment to the Food Safety Act will allow for the new separate rule book on food supplements that need to be periodically harmonized with amendments to the EU regulation on food supplements.

## KORISTI SUPLEMENTACIJE U PROMOCIJI ZDRAVLJA

**Brižita Đorđević<sup>1</sup>, Nevena Ivanović<sup>1</sup>, Ivana Baralić<sup>2,3</sup>**

<sup>1</sup>Katedra za bromatologiju, Univerzitet u Beogradu - Farmaceutski fakultet,

<sup>2</sup>Kliničko-bolnički centar Zvezdara, Beograd, <sup>3</sup>Udruženje za medicinu sporta,  
Beograd (Srbija)

Poslednjih decenija dijetarne intervencije pod kojim se podrazumeva potenciranje ili eliminacija određenih nutrijenata ili namirnica iz ishrane, kao i primena određenih nutrijenata u koncentrovanom obliku, u formi dijetetskih suplemenata, dobijaju sve više na značaju. Iako se stručna javnost uglavnom slaže da se odgovarajući unos nutrijenata može obezbediti dobro izbalansiranom ishranom, postoje stanja u kojima je upotreba suplemenata korisna i poželjna, kao što su starije osobe, pedijatrijska populacija, osobe sa poremećajem ishrane... U skladu sa izvornom definicijom, dijetetski suplementi obezbeđuju dodatni unos nutrijenata. Naime, pored vitamina i minerala koji su i najčešći sastojci suplemenata, u ovim proizvodima mogu biti prisutni i čitav niz drugih nutrijenata, kao što su pojedine masne kiseline, aminokiseline i izolovani proteini i peptidi, pojedini ugljeni hidrati, ali i veliki broj nenutritivnih supstanci za koja naučna istraživanja pružaju dokaze da su korisna i da povoljno deluju na organizam (karotenoidi, probiotički mikroorganizmi, koenzim Q10, bioflavonoidi i dr.). Kako se poslednjih godina u opštoj populaciji beleži izuzetno visoka prevalenca korišćenja dijetetskih suplemenata, veoma često se postavlja pitanje koja je stvarna korist od suplementacije. Ono na šta rezultati istraživanja ukazuju jeste da je u zemljama u kojima je upotreba dijetetskih suplemenata raširena, nutritivni status opšte populacije povoljniji, a pojava deficita ređa. Takođe, veliki broj studija govori u prilog dijetarnih intervencija suplementacijom. Kod najvećeg broja ovih studija osnovni cilj je korigovanje deficita nutrijenata, ali i opšta promocija zdravlja u smislu smanjenja rizika od hroničnih nezaraznih bolesti. Tako u literaturi postoji jasni dokazi o promotivnoj ulozi određenih biološki aktivnih jedinjenja - vitamina D, gvožđa, kalcijuma, cinka, omega-3 masnih kiselina, dok, s druge strane, za neka druga jedinjenja protektivna uloga još nije sasvim razjašnjena. Na kraju, zaključak o efikasnosti određenog suplementa, ali i sigurnosti i uslovima njegove primene, mora biti donet na osnovu vrste i količine dokaza dobijenih u adekvatno sprovedenim studijama.

## **DIETARY SUPPLEMENT IN HEALTH PROMOTION**

**Brižita Đorđević<sup>1</sup>, Nevena Ivanović<sup>1</sup>, Ivana Baralić<sup>2,3</sup>**

<sup>1</sup>Department of Bromatology, University of Belgrade-Faculty of Pharmacy,  
<sup>2</sup>Zvezdara University Medical Centar, Belgrade, <sup>3</sup>Sports medicine association of Serbia, Belgarde(Serbia)

In the former few decades the dietary interventions designed to potentiate or eliminate certain nutrients or whole foods from the diet, yet another designed to include some nutrients in a concentrated, dietary supplement form, are becoming more and more important. Although professional public in the most cases agree that appropriate intake of nutrients could be satisfied by well balanced diet, there are conditions in which use of supplement is useful and desirable such as use in older population, pediatric population, people with nutrition disorders. In accordance with the original definition, dietary supplements should provide additional intake of nutrients. Namely, besides vitamins and minerals which are the most common ingredients of supplements, a variety of other nutrients can be present in these products, such as certain fatty acids, amino acids and isolated proteins and peptides, certain carbohydrates, as well as a large number of non-nutritive substances for which scientific research provides evidence that they are useful and favorable for the human well-being (carotenoids, probiotic microorganisms, coenzyme Q10, bioflavonoids, etc.). As in recent years in the general population has been reported an extremely high prevalence of dietary supplements use, the question arises whether there are real benefits of supplements use. Literature data indicate that in countries where use of dietary supplements is widespread, the dominant status of the general population is more favorable, and the occurrence of the deficit is lower. Also, a large number of existing studies point to the benefit of supplement usage. In most of these studies, the main goal was correction of nutrient deficiencies, but also a general health promotion in terms of reducing the risk of chronic non-communicable diseases. Thus, there is clear evidence in the literature for role of certain biologically active compounds in health promotion - vitamin D, iron, calcium, zinc, omega-3 fatty acids. On the other hand, for some other compounds, the protective role has not been yet clarified. In the end, conclusion on the effectiveness of certain supplements, as well as the safety and conditions of its application, must be based on the type and amount of evidence obtained in adequately conducted studies.

## ZDRAVSTVENI RIZICI UPOTREBE DIJETETSKIH SUPLEMENATA

**Zorica Bulat**

Katedra za toksikologiju „Akademik Danilo Soldatović”, Univerzitet u Beogradu  
- Farmaceutski fakultet (Srbija)

Primena suplemenata se generalno smatra sigurnom što doprinosi njihovoj široko rasprostranjenoj upotrebi. Rizik od pojave eventualnih štetnih ili toksičnih efekata je veoma mali i najčešće potiče od nepravilne primene, upotrebe falsifikovanih preparata koji sadrže nedozvoljene supstance npr. steroide, prisustva nečistoća kao što su toksični metali itd.

Izvestan broj trovanja, koja su ujedno i najteža, posledica su akutne izloženosti visokim dozama suplemenata, bilo da je reč o slučajnim trovanjima ili samoubilačkim namerama. Centri za kontrolu trovanja konstantno beleže izvestan broj slučajeva ovih trovanja, pri čemu se većina slučajeva relativno lako zapažaju i karakteriše ih blaga do umereno teška klinička slika. Verovatno veći problem predstavlja nepravilna, slučajna ili tendenciozna, upotreba jednog ili više suplemenata, samih, ili zajedno sa lekovima. U ovim slučajevima eventualni štetni i/ili toksični efekti se teško zapažaju i retko dovode u vezu sa upotrebotom suplemenata. Nepravilna upotreba se ogleda, kako u unosu viših doza, tako i u primeni u dužem vremenskom periodu u odnosu na preporuke, a sa ciljem da se postigne željeni efekat. U ovim slučajevima, osobe koje ih koriste, često svesno ili nesvesno ne informišu lekara o upotrebi ovih preparata. Nekontrolisana upotreba pojedinih suplemenata koji mogu da interaguju, među sobom ili sa esencijalnim supstancama, pored rizika od prekomernog unosa per se, mogu rezultovati i suprotnim efektima, odnosno interakcijama koje vode ka smanjenoj raspoloživosti supstanci neophodnih organizmu.

Da bi se sprečili eventualni štetni efekti upotrebe suplemenata, pored pažljive kontrole ovih proizvoda od strane proizvođača i regulatornih tela, neophodno je da se podigne svest, kako osoba koji ih upotrebljavaju, tako i zdravstvenih radnika, o značaju njihove pravilne upotrebe. Naročito treba imati u vidu da istovremena primena većeg broja suplemenata može proizvesti brojne teško sagledive efekte, od deficita supstanci neophodnih organizmu, do pojave toksičnih efekata.

## **DIETARY SUPPLEMENTS - HEALTH RISK**

**Zorica Bulat**

Department of Toxicology „Akademik Danilo Soldatović”, University of Belgrade-Faculty of Pharmacy (Serbia)

Use of supplements is generally regarded as safe; hence their use is widespread nowadays. The risk of harmful or toxic effects is very low and most often arises from the improper use, intake of counterfeit preparations containing unauthorized substances, e.g. steroids, or the presence of impurities such as toxic metals, etc.

A certain number of poisoning cases are the consequence of acute ingestion of high doses, whether accidental or suicidal, and these cases are the most severe ones. The Poison Control Centers are consistently recording certain number of such cases, and majority can be easily recognized and are characterized by mild to moderately severe clinical presentations. Inappropriate, unintentional or intentional, use of one or more supplements, alone or in combination with drugs represents bigger issue. In such cases, harmful and/or toxic effects are difficult to detect and can rarely be associated with the use of supplements. In addition to overdose, improper use of supplements is also reflected by the duration of use which is in some instances longer than recommended. Moreover, rarely do these patients inform their medical doctors about the use of such products. Uncontrolled use of supplements that can interact among themselves, or with some essential substances, in addition to the risk of excessive intake per se, also can result in interactions leading to reduced availability of substances essential for physiological functions.

To prevent possible harmful effects of the supplements' use, in addition to careful control of these products by manufacturers and regulatory bodies, it is necessary to raise patient and health professional's awareness on the importance of their appropriate use. Special attention should be given to the fact that simultaneous intake of many supplements can produce several effects which are difficult to foresee such as deficit of essential substances necessary for physiological functions and various toxic effects.

## ANALIZA DIJETETSKIH SUPLEMENATA KOJE KORISTE SPORTISTI U SRBIJI

**Nenad Dikić<sup>1</sup>, Marija Andđelković<sup>2</sup>, Milica Vukašinović Vesić<sup>2</sup>,  
Brižita Đorđević<sup>3</sup>**

<sup>1</sup>Univerzitet Singidunum - Fakultet za fizičku kulturu i menadžment u sportu,

<sup>2</sup>Antidoping agencija Republika Srbije, <sup>3</sup>Katedra za bromatologiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Antidoping agencija Republike Srbije (ADAS) je sa ciljem informisanja sportista po pitanju prisustava supstance sa Liste zabranjenih supstanci, od 2011. godine uvela institucije davanja mišljenja o lekovima i suplementima. Da bi se izbegli nesporazumi, sva mišljenja o lekovima i suplementima su data pisano, a nakon elektronskog upita. Cilj ovog rada je prikaz suplemenata za koje su bili zainteresovani sportisti. Urađena je analiza svih izdatih mišljenja u periodu od 2011. do 2017. godine prema grupama lekova i suplemenata. U ovom radu će biti predstavljeni suplementi.

Prvih deset grupa suplemenata od 380 izdatih mišljenja na zahtev sportista prema različitim grupama suplemenata su: sagorevači masti (9,47%), whey protein (8,95%), NO reaktori (8,68%), multikomponentni preparati (6,58%), kreatin (6,05%), tribulus (5,79%), biljni preparati (4,74%), aminokiseline – kompleks (4,21%), povećanje energije - ugljeni hidrati (3,9%), povećanje telesne težine – gejneri (3,95%) itd. Kompletna logistika od strane ADAS koja je pružena sportistima nije samo servis, već, i pre svega, autentičan vid saradnje u borbi protiv dopinga u sportu, koji kao takav, ne postoji na svetu. Kompleksnost problema sigurno zahteva dublju farmakološku analizu, ali i samo nasumični pogled na tabelu sa grupama suplemenata koje koriste sportisti Srbije ukazuje na neophodnost razumevanja suplemenata koji sportistu mogu da uvedu u ozbiljan zdravstveni problem, i prekršaj doping pravila.

## **ANALYSIS OF DIETARY SUPPLEMENTS USED BY SERBIAN ATHLETES**

**Nenad Dikić<sup>1</sup>, Marija Andelković<sup>2</sup>, Milica Vukašinović Vesić<sup>2</sup>,**  
**Brižita Đorđević<sup>3</sup>**

<sup>1</sup>Singidunum University - Faculty of Physical Education and Management in Sport, <sup>2</sup>Antidoping agency of Serbia, <sup>3</sup>Department of Bromatology, Univeristy of Belgrade - Faculty of Pharmacy (Serbia)

The Anti-Doping Agency of the Republic of Serbia (ADAS) has introduced service on giving opinions on medicines and supplements since 2011, with the aim of informing the athlete about the presence of the substance from the List of prohibited substances. In order to avoid misunderstandings, all opinions on drugs and supplements are given in writing, after an electronic inquiry. The aim of this paper is to present the supplements issued at the request of athletes. An analysis of all published opinions in the period from 2011. to 2017. was carried out according to groups of drugs and supplements. In this work supplements will be presented.

The top ten groups of supplements of 380 opinions issued at the request of athletes according to different groups of supplements in the period from 2011 to 2017 are: fat burners (9.47%), whey protein (8.95%), NO reactors (8.68%), multicomponent preparations (6.58%), Creatine (6.05%), Tribulus (5.79%), herbal products (4.74%), amino acids - complex (4.21%), energy gainers (3.9%), body weight gainers (3.95%) etc. The complete ADAS logistics provided to athletes is not just a service, but above all an authentic type of cooperation in the fight against doping in sport, which as such does not exist in the world. The complexity of the problem certainly requires a deeper pharmacological analysis, but even a random view of the table with the groups of supplements used by athletes in Serbia points the necessity of understanding the supplements that can bring the athlete into a serious health problem and a violation of doping rules.

## **APOTHECARY PROFESSION AND PHARMACEUTICAL ACTIVITIES IN THE HEALTH CARE SERVICE AT THE END OF THE FIRST WORLD WAR**

**Adriana Elena Taerel**

„Carol Davila” University of Medicine and Pharmacy, Bucharest (Romania)

The innovative trends in the pharmaceutical industry at the end of the 19th century and before the First World War have become significant in the present-day pharmacy, determining pharmaco-historians to consider the 20th century as the age of medicine at European and international level. Favoured by the development of the chemistry and pharmaceutical technology, as well as by the modernisation of the manufacturing processes, the drug industry has seen an increasing development. It had generated the appearance of the large factories and chemical-pharmaceutical monopolies in developed European countries like Germany (Bayer, Hoechst), England, Netherlands etc.

Materials referring to the period under review were studied, with a focus on aspects related to the progress and stagnation, respectively, of the European pharmacy practice and apothecary service.

As regards the pharmaceutical industry, production laboratories are starting to open in many European countries, Romania as example, with up to 176 labs functioning by mid-20th century. The modern pharmacist education required the introduction of new subjects in the curriculum plan. The modern healthcare laws enacted in several European countries before 1914 and between the two World Wars were not fully enforced. Although they promoted modern organisation and pharmaceutical assistance, the healthcare laws in Romania had serious drawbacks.

During the historical period we observed, the overall pharmacy development followed an uneven upwards trend, however marked by setbacks and lagging. This was the case in many countries across the Europe. The pharmaceutical research has gained momentum on an international scale although the two World Wars devastated the social and political life on every level.

## JAČANJE PROFESIONALIZMA U APOTEKARSKOJ PRAKSI: ČEMU NAS UČE APOTEKARSKE ZAKLETVE OD NAJSTARIJIH DO SAVREMENIH

Dušanka Krajnović

Katedra za socijanu farmaciju i farmaceutsko zakonodavstvo, Univerzitet u  
Beogradu – Farmaceutski fakultet (Srbija)

Rutter i Duncan (2010) upućivali su na to da je „profesionalizam skup strukturalnih, karakternih i bihevioralnih osobina”, dok Hammer (2006) profesionalizam definiše njegovim ispoljavanjem u praksi, koje se „očituje kroz profesionalnu socijalizaciju, gde pojedinac može da usvoji vrednosti, osobine i praktično ponašanje koje je odraz profesionalizma”. Krenuli smo od istorijske analize koja je pokazala da se razvoj farmaceutske prakse, a posebno apotekarske prakse, menjao i morao da se prilagodi sve zahtevnijim društvenim promenama.

U radu je istraženo kako su se očekivanja, veštine, karakteristike i zahtevane moralne osobine apotekara menjale kroz vreme. Metoda koju smo koristili uključuje analizu sadržaja odabranih apotekarskih zakletvi, od onih prvih iz 12. veka do savremenih, poput FIP-ove zakletve farmaceuta. Izabrali smo zakletvu kao formu etičkog normativa zbog toga što je najstarija i zbog toga što se održala do danas. Od srednjeg veka do početka prošlog veka, karakterne osobine apotekara, kao hipotetička polazišta za prihvatljivo ponašanje, bile su u osnovi zahteva svih analiziranih tekstova zakletvi. U njima se može pratiti i promena profesionalne filozofije i društvene uloge apotekara, od majstora umeća izrade lekova, preko snabdevača i onoga ko izdaje lekove, do pružaoca usluge za pacijenta. Farmaceuti su shodno ovom preokretu imali tu prednost da budu osnaženi nizom novih zahteva i veština koje je trebalo usvojiti. Kao što nas upućuju tekstovi savremenih apotekarskih zakletvi, ovaj posao mora da se obavlja dostojanstveno, celishodno i s ponosom kako bi se ostvarila dva značajna moralna principa: kao prvo „ne naneti štetu” (primum non nocere) i drugo, dužnost da se staraju za pacijenta. Princip „raditi u najboljem interesu pacijenta” u osnovi je svih analiziranih tekstova i iskazan je kroz više zahteva, od kojih je samo jedan konstantno prisutan - a to je očuvanje poverenja koje pacijent ima u farmaceuta kao zdravstvenog profesionalca.

*Istraživanje je realizovano u okviru Projekta 14004 finansiranog od strane Ministarstva prosvete, nauke i tehnološkog razvoja Republike Srbije.*

# **REINFORCING PROFESSIONALISM IN APOTHECARY PRACTICE: WHAT COULD WE LEARN FROM THE APOTHECARIES' OATHS FROM THE PAST TO THE MOST CONTEMPORARY**

**Dušanka Krajnović**

Department of Social Pharmacy and Pharmaceutical Legislation, University of Belgrade - Faculty of Pharmacy (Serbia)

Rutter and Duncan (2010) suggested that „professionalism is a complex composite of structural, attitudinal and behavioral attributes”, while Hammer (2006) argued that professionalism is defined by its demonstration in practice and is „manifested through professional socialization, when one can learn to adopt to the values, attitudes and practical behaviors that are seen as representing professionalism.” We took the point of history analysis which showed that pharmacy profession, apothecary in particular, has had to adapt to accommodate to the imposed requirements of society.

This paper examined how expectations, skills, characteristics and the requisite moral character of apothecarists changed over time, by using the text analysis of several apothecary oaths. We chose oaths as the oldest form of ethical normative which has been persisted for almost two millennia. The oldest analyzed apothecary oath is from the 12<sup>th</sup> century, while the contemporary one is the FIP oath/promise of a pharmacist. From the middle ages the concept of „character” was emphasized as the set of assumptions about the proper behavior rather historically and culturally specific. Shifting of the role of pharmacist from a technician, compounder and seller of drugs to a career of patients, led to the evolution of pharmacy from occupational to professional status and the adoption of a new philosophy of practice accountable for pharmaceutical care in an ethical context. Pharmacists benefited from this change with a set of expectations and a set of skills to be acquired. As reflected in contemporary oaths this service must be carried out with dignity, integrity and honor in order to achieve two important moral principles: „not to do harm” and to provide „duty of care”.

*This research was supported by the grant of Ministry of Education, Science and Technological Development in Serbia, Grant Number 41004.*

## **QUALITY INDICATORS OF PHARMACEUTICAL CARE SERVICES**

**Mitja Kos**

Faculty of Pharmacy, University of Ljubljana (Slovenia)

A basic concept which should underlie all health care services and pharmacy practice is that of assuring the quality of patient care activities. Donabedian defined the three elements of quality assurance in health care as being structure, process and outcome (Donabedian A., 1980). Quality indicators address measurable quality aspects of the three elements. They provide an insight into the performance of care providers and are used to stimulate continuous improvement of patient care. They are informative to healthcare providers, payers and patients. With the development and implementation of new pharmacy-led services, pharmacists are challenged to form specific indicators that will reflect the main goals of their services. For example, medication use review service aims to improve medication adherence of patients, their knowledge about medicines and the appropriateness of use. Therefore, we would expect quality indicators reflect the above stated goals of the service. Quality indicators should be implemented into the system step by step and should begin with the most actual and professionally relevant ones.

## **ANALIZA FARMACEUTSKIH USLUGA U EVROPI I SRBIJI – MODALITETI RAZVOJA U SVETLU NOVIH TEHNOLOGIJA**

**Ivana Tadić**

Katedra za socijanu farmaciju i farmaceutsko zakonodavstvo, Univerzitet u Beogradu – Farmaceutski fakultet (Srbija)

Farmaceutske usluge koje se pružaju u apotekama razlikuju se u državama Evrope usled kulturno-različitih sistema zdravstvene zaštite. Promene u farmaceutskoj regulativi i primena novih zdravstvenih tehnologija utiču na promene u pružanju farmaceutskih usluga, kao i uloge farmaceuta u društvu. Velika studija sprovedena tokom 2012. i 2013. godine, sa ciljem da se analizira stepen pružanja farmaceutskih usluga, obuhvatila je farmaceute iz 16 država Evrope. Podaci o uslugama prikupljeni su pomoću „Behavioral Pharmaceutical Care Scale“ (BPCS) upitnika. Učešćem u studiji, po prvi put je omogućeno da se farmaceutske usluge koje se pružaju u Srbiji porede sa farmaceutskim uslugama drugih zemalja. Prosečan BPCS skor farmaceuta ( $n=374$ ) iz Srbije iznosio je  $77,5 \pm 25,5$ . Najveći BPCS skor zabeležen je u Švajcarskoj ( $82,7 \pm 22,8$ ) dok je najniži zabeležen u Litvaniji ( $60,4 \pm 20,8$ ). Mali procenat farmaceuta Srbije (3,7%) pripao je kategoriji „pružaoci farmaceutskih usluga“ (farmaceuti čiji je skor bio u rasponu najviših 20% vrednosti ukupnog BPCS skora). U ponovljenoj studiji sprovedenoj 2018. godine u Apoteci Beograd (učestvovalo 59 farmaceuta) ukupan prosečan BPCS skor iznosio je  $84,3 \pm 22,1$ , dok je 8,5% farmaceuta pripao kategoriji „pružaoci farmaceutskih usluga“. U poslednjih pet godina zabeležen je porast ukupnog BPCS skora farmaceuta iz Srbije. Tokom ovog perioda mnogi zakonski akti od značaja za farmaceutsku praksu pretrpeli su izmene ili su usvojeni novi. Implementirane zdravstvene tehnologije omogućile su izdavanje lekova putem obnovljivih elektronskih recepata, olakšano pružanje informacija o lekovima (načinu upotrebe lekova, interakcijama između lekova, kao i interakcijama lekova sa hranom ili dijetetskim suplementima, adherencij), olakšanu saradnju farmaceuta sa drugim zdravstvenim radnicima, kao i edukaciju farmaceuta putem elektronskih platformi. Sve ove promene značajno su uticale na promenu uloge farmaceuta u društvu.

# **ANALYSIS OF PHARMACEUTICAL SERVICES PROVIDED IN COMMUNITY PHARMACIES IN EUROPE AND SERBIA - MODALITIES FOR FUTURE DEVELOPMENT IN THE LIGHT OF NEW TECHNOLOGIES**

**Ivana Tadić**

Department of Social Pharmacy and Pharmaceutical Legislation, University of Belgrade - Faculty of Pharmacy (Serbia)

The pharmaceutical services are different across European countries due to country specific practice, culture and systems of health delivery. Changes in pharmaceutical regulations and implementation of new health technologies influence the evolution of pharmaceutical services and community pharmacists' role. In the years 2012 and 2013 the large study, with the aim to assess the degree of provision of pharmaceutical services by community pharmacists, was conducted across Europe using the Behavioral Pharmaceutical Care Scale - BPCS. For the first time, pharmaceutical services in Serbian community pharmacies were compared with the services of other 15 European countries that participated in the study. The total BPCS average score achieved by Serbian pharmacists (n=374 pharmacists) was  $77.5 \pm 25.5$  (the highest score was  $82.7 \pm 22.8$  of Switzerland pharmacists and the lowest was  $60.4 \pm 20.8$  of Lithuanian pharmacists). Only 3.7% Serbian pharmacists were categorized as „providers of pharmaceutical care” according to the classification of the top 20% of the total BPCS score. In repeated study conducted in 2018 year in Serbia within the Pharmacy Belgrade (n=59 pharmacists), the overall BPCS average score was  $84.3 \pm 22.1$  and 8.5% Serbian pharmacists were categorized as „providers of pharmaceutical care”.

The overall BPCS score of the Serbian pharmacists increased in the last five years. During this period many pharmaceutical regulations have been changed, some have been adopted and new technologies have been implemented. New health technologies enabled dispensing medicines on repeatable prescriptions, easier provision of information about medicines (medication use; interactions with other medicines, food and dietary supplements; adherence), easier referral and consultation activities as well as on-line education of pharmacists. All of these changes significantly influenced the change of the pharmacists' role in the society.

## **UNAPREĐENJE ZDRAVSTVENE ZAŠTITE TRUDNICA I DOJILJA – ULOGA FARMACEUTA I DOPRINOS FARMACEUTSKIH USLUGA**

**Marina Odalović**

Katedra za socijanu farmaciju i farmaceutsko zakonodavstvo, Univerzitet u Beogradu – Farmaceutski fakultet (Srbija)

Značajan doprinos farmaceuta u unapređenju zdravlja majki prepoznat je od strane Svetske farmaceutske federacije (International Pharmaceutical Federation - FIP). Uloga farmaceuta opisana je u okviru više intervencija i uključuje sledeće: informisanje trudnica i dojilja o vitaminskim i drugim dijetetskim proizvodima, uključujući folnu kiselinu i proizvode koji sadrže gvožđe; promovisanje odvikavanja od pušenja i upotrebe alkohola; evaluacija primene potencijalno teratogenih lekova i savetovanje adekvatne promene terapije (npr. kod terapije epilepsije). Dodatno, opisano je više farmaceutskih usluga tokom postpartalnog perioda kao što su: pomoć kod dojenja (preporuka odgovarajuće formule za odojče kada majka nije u mogućnosti da doji dete, i kada je ovakav način ishrane prihvatljiv, izvodljiv i bezbedan za bebu); skrining žena pod rizikom od postpartalne depresije; obezbeđenje suplemenata sa vitaminom A. Uprkos preporukama FIP-a, tradicionalno, farmaceuti nisu direktno uključeni u strukturirane usluge zdravstvene zaštite namenjene trudnicama i dojiljama. Međutim, trudnice navode farmaceute kao jedan od najkorisnijih izvora informacija u vezi sa različitim zdravstvenim problemima u trudnoći. Sa druge strane, farmaceuti opisuju različite barijere sa kojima se susreću pri savetovanju trudnica. Nedostatak znanja o terapiji specifičnih stanja u trudnoći, kao i nedostatak relevantnih izvora informacija o bezbednosti lekova i drugih proizvoda pri primeni u trudnoći prepoznati su kao najčešći problemi u praksi. Kursevi kontinuirane edukacije posvećeni upotrebi lekova u trudnoći predstavljaju za farmaceute značajan izvor korisnih informacija. Dobro informisani farmaceuti mogu dati značajan doprinos u unapređenju zdravlja majki i beba. Razvoj informacionih centara o teratogenosti lekova zajedno sa specifičnim kursevima kontinuirane edukacije mogu biti veoma korisni za farmaceute.

# **PREGNANT AND BREASTFEEDING WOMEN HEALTHCARE IMPROVEMENT - THE ROLE OF PHARMACISTS**

**Marina Odalović**

Department of Social Pharmacy and Pharmaceutical Legislation, University of Belgrade - Faculty of Pharmacy (Serbia)

Significant contribution of pharmacists in improvement of maternal health has been well recognized by the International Pharmaceutical Federation (FIP). The role of pharmacists has been described within the following interventions for pregnant women: educate mothers on and supply vitamins and nutritional supplements, including folic acid and iron supplements; promote cessation of alcohol and nicotine use; evaluation of potential teratogenic medicines; advice on alternative drug regimens if teratogenicity of current treatment is known or a reduction in risk is required (e.g. in epilepsy). Additionally, pharmacists interventions for postnatal period has also been described and include: support breastfeeding (when replacement feeding is acceptable, feasible, affordable, sustainable and safe); identify women at risk of postpartum depression; providing of vitamin A supplementation. In spite of FIP recommendations, traditionally, pharmacists have not provided speciality services to pregnant and breastfeedinf women. However, pregnant women has reported that they see pharmacist as one of the most useful sources of information about health issues related to pregnancy. On the other side, different areas of concerns and barriers related to counselling of pregnant women have been described by pharmacists. Gaps in knowledge related to specific condition treatment in pregnancy, and deficit of relevant sources of drug safety data for use in pregnancy have been reported as a common problems in practice. Continuing professional development courses have been recognized as important for contribution to the knowledge base related to this specific and demanding issue. Well informed pharmacists could give important contribution to improvement of maternal and newborn health. Development of teratogenic information service along with countinuing education courses could be very useful for pharmacists.

## **KOLIKO KOŠTA FARMACEUTSKA USLUGA?**

**Dragana Lakić**

Katedra za socijanu farmaciju i farmaceutsko zakonodavstvo, Univerzitet u Beogradu – Farmaceutski fakultet (Srbija)

Prema sadašnjem Pravilniku o cenama zdravstvenih usluga (bez obzira da li se radi o primarnom, sekundarnom ili tercijarnom nivou), gotovo nijedna farmaceutska usluga nije prepoznata za plaćanje od strane Republičkog fonda za zdravstveno osiguranje.

Farmaceutske usluge predstavljaju sve usluge koje pružaju farmaceuti u cilju obezbeđenja adekvatne farmaceutske zdravstvene zaštite. Ovo uključuje kako aktivnosti usmerene na nabavku, snabdevanje i izdavanje lekova i drugih proizvoda koji se mogu naći u apoteci, tako i aktivnosti usmerene u cilju promocije zdravlja, kao i proces komunikacije i pružanje saveta o adekvatnoj primeni leka bilo pacijentima bilo drugim zdravstvenim radnicima. Cilj pružanja farmaceutske usluge je dostizanje optimalnih ishoda terapije pacijenta i poboljšanje pacijentovog kvaliteta života.

U prezentaciji će biti prikazan ekonomski aspekt pružanja farmaceutske usluge u apoteci. Istraživanja iz sveta, ali i Srbije pokazuju da su pacijenti spremni za izdvajanjem određenog novčanog iznosa za farmaceutske usluge. Neophodno je pokazati značaj i vrednost farmaceuta i farmaceutske usluge i donosiocima odluka u zdravstvu.

## **HOW MUCH DOES THE PHARMACEUTICAL SERVICE COST?**

**Dragana Lakić**

Department of Social Pharmacy and Pharmaceutical Legislation, University of Belgrade - Faculty of Pharmacy (Serbia)

According to the current Ordinance on prices of health services (regardless of whether it is a primary, secondary or tertiary level), almost none of the pharmaceutical services are recognized for payment by the National Health Insurance Fund.

Pharmaceutical services represent all services provided by pharmacists in order to provide adequate pharmaceutical care. This includes both activities aimed at the procurement, supply and dispensing of medicines and other products that can be found in the pharmacy, as well as activities aimed at promoting health, and also the process of communication and providing advice on the adequate use of the drug to patients or other health professionals. The goal of providing pharmaceutical services is to achieve the optimal outcome of the patient's treatment and to improve the patient's quality of life.

The presentation will show the economic aspect of providing pharmaceutical services in the pharmacy. Research from the world as well as Serbia shows that patients are ready to allocate a certain amount of money for pharmaceutical services. It is necessary to show the importance and value of pharmacists and pharmaceutical services to decision-makers in healthcare.

## **PATIENT CENTRIC DOSAGE FORM DESIGN**

**Andreas Zimmer<sup>1</sup>, Sven Stegemann<sup>2</sup>**

<sup>1</sup>University of Graz, Institute of Pharmaceutical Sciences, Department of Pharmaceutical Technology and Biopharmacy, <sup>2</sup>Graz University of Technology, Institute of Process and Particle Engineering (Austria)

Since there is growing evidence that patient centric dosage form design plays a key role in achieving effectiveness and minimizing medication errors, many drug delivery systems are considered to meet patients needs. When reviewing the literature with regard to the clinical evidence for such claims, the results are very disappointing. The main reasons are a focus on a single product feature outside the entire contextual framework and the lack of specific methodology to investigate patient centric product design.

Patient centric dosage form design follows the logic of a patient as a medical lay person, based on prior learnings, perception, intuition and capabilities within the personal daily context and environment. Even though there is not a single or a set of methodology specifically developed for patient centric drug product design, several methods are applied by other disciplines and industries successfully.

Understanding the patient journey from symptom recognition through to effective disease management will be the starting point to identify and prioritize patient needs with regard to drug product design development. The patient needs will have to be integrated into the Target Product Profile (TPP). Potential approaches to address these needs have to be verified within the relevant patient medication use and process in comparative trials to achieve the desired endpoints. Other industries have a long time established process to design products for specific user groups, several methodologies can be used in a similar way by the pharmaceutical industry. While patient centric drug product design is considered being a mind-set, pharmaceutical organizations might have to adapt their existing development processes early on to increase effectiveness and patient safety through patient centric drug product design without impacting on development time lines.

## **INCREASED PATIENT SAFETY BY READY-TO-USE/READY-TO-ADMINISTER PARENTERALS PREPARED IN HOSPITAL PHARMACIES**

**Irene Krämer**

University Medical Center, Johannes Gutenberg-University Mainz (Germany)

Most medicinal products which are administered parenterally are approved and marketed as injection concentrate or powder for injection. Prior to administration, these products must be reconstituted, diluted and equipped with an administration device by health care professionals. According to the European Resolution CM/Res 2016, the risks associated with the reconstitution should be assessed. High-risk products should be prepared in the pharmacy department either as ready-to-administer or ready-to-use parenterals in patient-individual or standard doses. Antineoplastic drug solutions and parenteral nutrition admixtures are commonly prepared for individual patients in individualized doses. Antibiotics, antifungals, continuous injections for intensive care patients, analgesics, and emergency drugs are mostly prepared batch-wise in standardized doses or concentrations. Premises, facilities and pharmaceutical knowledge must be appropriate for the reconstitution or the preparation of unlicensed medicinal products for the special needs of patients. Appropriate quality assurance systems are to be implemented.

Examples for the different types of pharmacy preparations and their added value, especially increased patient safety, will be given.

## **FORMULACIJA FARMACEUTSKIH OBLIKA LEKOVA ZA PRIMENU U PEDIJATRIJSKOJ POPULACIJI - ASPEKTI PRIHVATLJIVOST/ADHERENCA**

**Iela Milić, Sandra Cvijić, Ivana Pantelić**

Katedra za farmaceutsku tehnologiju i kozmetologiju, Univerzitet u Beogradu -  
Farmaceutski fakultet (Srbija)

Formulacija lekova za pedijatrijske pacijente predstavlja poseban izazov za farmaceute. Mnogi lekovi nisu dostupni u odgovarajućem farmaceutskom obliku za pedijatrijski uzrast i često se propisuju na način koji nije predviđen sažetkom karakteristika leka, odnosno, pristupa se neodobrenoj (off-label) upotrebi leka. Takođe, registrovani lekovi za odrasle se često različitim postupcima „prilagođavaju“ za primenu kod dece, čime se povećava rizik od pojave neželjenih reakcija, greški u doziranju ili neprihvatljivosti leka.

Prilikom formulacije lekova za decu potrebno je odabrati odgovarajući farmaceutski oblik leka za određeni uzrast i pažljivo razmotriti izbor pomoćnih supstanci i „nosača“ za isporuku leka. „Lek prilagođen uzrastu“ treba da bude prihvatljiv od strane pacijenta i dovoljno jednostavan za primenu od strane roditelja/staratelja ili medicinskog osoblja. Pri tome treba razmotriti put primene leka, lakoću primene, preciznost i fleksibilnost doziranja, veličinu i disperzibilnost farmaceutskog oblika leka i mogućnost korigovanja/„maskiranja“ ukusa kod preparata za oralnu primenu. Značaj ovakvog pristupa je prepoznat od strane regulatornih organa koji su dali preporuke za procenu prihvatljivosti i odabir farmaceutskog oblika/puta primene leka u zavisnosti od uzrasta deteta.

Uporedno sa razvojem farmaceutskih oblika lekova za pedijatrijski uzrast, razvijaju se i uređaji/pribor koji omogućavaju precizno doziranje. No, uprkos naporima da se poveća dostupnost komercijalnih preparata za decu, i dalje postoji velika potreba za njihovom izradom u apotekama, za određene grupe pedijatrijskih pacijenata. U cilju unapređenja ovog segmenta rada u apoteci nedavno je pokrenut projekat za izradu Panevropskih pedijatrijskih formula namenjenih farmaceutima koji se bave izradom magistralnih lekova za pedijatrijske pacijente. Pedijatrijske formule treba da pruži informacije o sastavu, načinu izrade, pakovanju i uslovima čuvanja lekova za decu prilagođenih uzrastu, koji nisu dostupni na tržištu. Očekuje se da će nova istraživanja i odgovarajuće smernice regulatornih/stručnih tela pomoći u naporima da se u budućnosti proizvede/izradi veći broj bezbednih i efikasnih lekova, prihvatljivih za primenu kod dece.

## **FORMULATION OF PAEDIATRIC DOSAGE FORMS - ACCEPTABILITY ISSUES/COMPLIANCE**

**Iela Milić, Sandra Cvijić, Ivana Pantelić**

Department of Pharmaceutical Technology and Cosmetology, University of Belgrade-Faculty of Pharmacy (Serbia)

Formulation of paediatric medicines is associated with numerous challenges. Many drugs are not available in suitable dosage forms for paediatric patients, which often leads to off-label and unlicensed use of adult medicines. Also, adult dosage forms are often modified/manipulated to enable drug administration in children, which increase the risk of adverse drug reactions, dosing errors and non-compliance issues.

When developing paediatric formulations, it is necessary to select appropriate age-adapted dosage forms, and carefully consider the choice of excipients and delivery devices. An „age-appropriate medication” should be acceptable/suitable for use in children and easy to administer by parents/caregivers or healthcare professionals. Some points to consider include dosing route, ease of use, dose accuracy/flexibility, and also size, dispersibility and taste masking ability for oral dosage forms. In order to assist in the development of paediatric formulations, regulatory authorities have issued recommendations on acceptability rating and the selection of most appropriate dosage form/dosing route in relation to child age.

In parallel with the development of paediatric dosage forms, new dosing devices are being designed. But despite the efforts to improve availability of commercial drug products for children, there is still a widespread need for compounded preparations for certain paediatric age groups. In order to facilitate compounding of paediatric formulations, a project to develop Pan-European formulary has been launched recently. This formulary is intended to provide clinicians and pharmacists with compiling information on the composition, preparation, container systems and storage conditions regarding extemporaneous preparation of non-licensed medicines for children.

Future research on pediatric formulations and appropriate regulatory/scientific guidelines are expected to result in an increased number of safe and effective authorised/compounded, age-appropriate medicines for children.

## **CANCER IMMUNOTHERAPY: WHERE DID ITS PRECISION COME FROM AND WHERE WILL IT GO?**

**Farzin Farzaneh**

Division of Cancer Studies, Department of Haematological Medicine, Kings College London (United Kingdom)

The Nobel Prize in Physiology or Medicine 2018 was awarded to James P. Allison and Tasuku Honjo "for their discovery of cancer therapy by inhibition of negative immune regulation". By this accomplishment, cancer immunotherapy has come of age, approximately half a century after classical experiments that distinguished "self" from "non-self", and the seminal findings that humoral immunity, effected by B lymphocytes, is inextricably complemented by cellular immunity, effected by T lymphocytes. The major goal of cancer immunotherapy is to reverse the tolerant state that enables cancer cells to evade immune detection and destruction. Contemporary immunologic strategies for targeting tumors encompass mechanisms that target T cells: directly activating them, or inhibiting molecules that suppress T-cell activation, or modifying T cells genetically to allow them to recognize and kill tumor cells either in an MHC-dependent (TCR-modified T cells) or MHC-independent manner, by genetically engineered Chimeric Antigen Receptor Modified T cells (CAR T cells). In this field, major breakthroughs in clinical practice have been achieved with immune checkpoint blockade. This refers to the suppression of inhibitory pathways activated by cancer cells, and comprises antibodies directed against the components of the pathway involved in adaptive immune suppression, namely Programmed Cell Death -1 receptor (PD-1, expressed on activated T cells) and Ligand (PD-L1, expressed on antigen presenting cells). Currently, five PD-1/PD-L1 immune checkpoint inhibitors are approved for cancer immunotherapy: atezolizumab, avelumab, durvalumab, nivolumab and pembrolizumab. Another approach with substantial potential is to develop therapeutic cancer vaccines. By employing the strategies of adjuvant activation of toll-like receptors, such vaccines would aim to predominantly engage the cellular immunity in a way that can control the location, magnitude and duration of the elicited response, and are hoped to lead to further breakthroughs in the field of cancer immunotherapy.

## PRECIZNA MEDICINA U ONKOLOŠKOJ PRAKSI: PROCENA KORISTI I RIZIKA

Ivana Božović-Spasojević

Institut za onkologiju i radiologiju, Beograd (Srbija)

"Zavisnost od onkogena", kada nastanak i rast tumora kontinuirano zavise od signala onkogena, predstavlja ključno načelo u onkologiji. Koncept je potvrđen na brojnim onkogenima u kliničkim i pretkliničkim ispitivanjima, i smatra se opštim načelom odabira ciljane terapije u onkologiji. Međutim, dostupni vodići i dalje ne preporučuju rutinsko genomsко testiranje tumora zbog ograničenih dokaza o kliničkoj upotrebi i selekciji sistemske terapije kod pacijenata sa odmaklom malignom bolešću. S druge strane, rezultati genomskog testiranja mogu biti relevantni za utvrđivanje podobnosti pacijenata za klinička ispitivanja sa agensima od interesa. Precizna onkologija u pristupu utvrđivanja varijabilnosti gena koristi različite metode, od imunohistohemije do sekvensiranja tumora, ispitivanja molekularne abnormalnosti u DNK tumora u cilju utvrđivanja terapijske mete. Klinička primenjivost ovih metoda suočava se sa važnim izazovima: odabira najreprezentativnijeg tkiva za testiranje, ponavljanja testiranja tokom lečenja i praćenja pacijenta (vremenska i prostorna heterogenost tumora), tumačenja i razumevanja rezultata i njihovog stavljanja u klinički kontekst koji je relevantan za pacijenta, prioritizacije onkogenih signala kada ih ima više, cene testiranja i razumevanja pacijenata o neophodnosti i značaju testiranja.

Nove neinvazivne metode obećavaju prevazilaženje navedenih mana, dajući kompleksniju i sveobuhvatniju sliku maligne bolesti. To su tečne biopsije koje iz krvi mogu ispitivati cirkulišuću tumorsku DNK i tumorske ćelije, analize urina, pljuvačke, stolice ali i izdaha u kome se ispituju isparljive organske komponente. Ove metode prevazilaze tradicionalnu analizu tumorskog tkiva i omogućavaju ponavljanje, prospektivno, longitudinalno praćenje tumorskog odgovora, rezistencije i progresije na ordiniranu terapiju.

Uporedo sa tehnološkim napretkom desio se pomak u sprovođenju kliničkih ispitivanja koja zahtevaju novi, adaptabilni dizajn i način sprovođenja. S obzirom da je količina podataka dobijena bioinformatičkom analizom *omics* tehnologija velika, trend je da podaci budu sačuvani na zajedničkim platformama i zatim javno dostupni za dalje analize.

Svedoci smo razvoja imunoterapije u onkologiji. Od uvođenja anti-CTLA-4 i anti-PD-1/PD-L1 monoklonskih antitela kao standardne terapije za lečenje mnogih tumora, validirano je nekoliko bioloških markera. To su nivo ekspresije PD-L1, mikrosatelitska nestabilnost, i u skorije vreme opterećenje tumorskom mutacijom, kao prediktivni markeri odgovora na imunoterapiju.

Uzveši u obzir odnos koristi i rizika, kliničarima ostaje izazov odabira najboljih genetskih i molekularnih oruđa u odabiru i sprovođenju terapije.

## **PRECISION MEDICINE IN ONCOLOGY PRACTICE: BENEFIT-RISK ASSESSMENT**

**Ivana Božović-Spasojević**

Institute for Oncology and Radiology, Belgrade (Serbia)

A key principle underlying treatment selection is "oncogene addiction." This means that tumors are continuously addicted to the driving growth signal from an oncogene. This concept is validated across plentiful oncogenes in clinical and preclinical contexts, and represents general principle underlying selection of targeted therapy. Available clinical practice guidelines do not recommend genomic testing of tumors in routine practice because of limited evidence of clinical utility to guide selection of systemic therapy for patients with advanced cancer. However, genomic testing results may be relevant to determine eligibility for clinical trials with investigational agents. Precision oncology uses multiple testing techniques to identify molecular abnormalities in a patient's DNA with the aim of identifying therapeutic targets. However, clinical practice faces barriers in implementing precision medicine, such as understanding the best tissue for molecular tests, identifying the most precise molecular tests and when tissue should be tested, interpreting the test results, understanding the role of genetic counselling, patient attitudes and financial concerns.

Novel noninvasive cancer diagnostics platforms, like liquid biopsy, used to interrogate ctDNA or circulating tumor cells, urine, saliva and stool and breath biopsy, which measures volatile organic compounds, have continued to evolve in recent years. These noninvasive molecular diagnostics assays fundamentally transform the potential utilities of cancer diagnostics to enable repeat, prospective, and serial longitudinal biopsies to monitor disease response, resistance and progression on therapies.

Alongside these technology advances, a lot of trials with precision medicine principles were initiated. Precision medicine trials raise new and different challenges in their design and conduction, requiring new strategies for successful implementation in clinical practice.

We also face the recent advent of cancer immunotherapies. Since the arrival of anti-CTLA-4 and anti-PD-1/PD-L1 cancer monoclonal antibodies, a few genomic and tissue biomarkers have been validated and approved for cancer immunotherapy. These include tumoral PD-L1 expression levels and microsatellite instability status. Furthermore, strong evidence of tumor mutational load has been found to support its use as a predictive immuno-oncology biomarker.

For physicians, determining when and how to incorporate genetic and molecular tools into clinic in a cost-effective manner is critical.

## **MOLECULAR PATHWAYS THAT OPERATE IN MLL-ASSOCIATED LEUKEMIA TO OVERCOME RESISTANCE TO ANTICANCER DRUGS**

**Boban Stanojević**

Department of Haematological Medicine, Division of Cancer Studies, King's College London (United Kingdom), Laboratory for Radiobiology and Molecular Genetics, „Vinča“ Institute of Nuclear Sciences, University of Belgrade (Serbia)

Deregulation of gene expression can cause devastating cancers. Fusion of the mixed-lineage leukemia (MLL) gene (also known as KMT2A, MLL1, ALL-1, or HTRX) and various partners causes aggressive leukemia. To date, more than 70 genes have been reported to fuse with MLL. The sequence of the MLL gene was revealed in the early 1990s. Since then, a series of technological breakthroughs such as DNA arrays, shRNA library screening, proteomic and deep sequencing have provided us with a much deeper understanding of the molecular basis of leukemogenesis caused by MLL mutations. Advances in technology and the ongoing development of new targeted therapies have opened up new opportunities to combat drug resistance as well. We are now able to characterize the signalling pathways involved in regulating tumour cell response to chemotherapy more completely than ever before. Based on the understanding of these molecular mechanisms, several small molecules that inhibit critical processes of leukemogenesis have been developed as molecularly-targeted drug candidates. I herein review the normal biological roles of MLL1 and its fusion partners, how these roles are hypothesized to be dysregulated in the context of MLL1 rearrangements, the clinical manifestations of this group of leukemias with a special focus on molecular pathways that operate to prevent resistance to anticancer drugs.

## PRECIZNE ANTIKancerske terapije: Kako farmaceutska tehnologija daje doprinos?

Snežana Savić

Katedra za farmaceutsku tehnologiju i kozmetologiju, Univerzitet u Beogradu - Farmaceutski fakultet (Srbija)

Precizna/personalizovana medicina teži da individualizuje terapijske intervencije/isporuku leka na osnovu ex vivo i in vivo informacija o pacijentu i specifičnih karakteristika bolesti. Primena nanotehnologije, povezano sa humanim zdravljem (nanomedicina), u svetu precizne dijagnoze/tretmana različitih tumora ima već duže vreme istaknuto mesto u ovom kontekstu. Dok su hirurgija i radioterapija primarni tretmani koji se koriste za lokalizovane, nemetastatske kancere, antikancerski lekovi (hemioterapeutici, hormoni, biološki lekovi, imunoterapija) predstavljaju aktuelni pristup u terapiji metastatskih kancera. Neselektivna destrukcija zdravih ćelija, toksičnost konvencionalnih citotoksičnih lekova, kao i razvoj multiple rezistencije podržavaju ideju za pronalaženjem novih efektivnih opcija ciljne terapije, koji su zasnovani na izmeni molekulskih bioloških mehanizama tumorskih ćelija. Osnovni koncept za dizajniranje nano-antikancerskih lekova kroz niz prethodnih godina zasnovan na „efektu poboljšanja permeabilnosti i zadržavanja“ (EPR), opšteprihvaćenom kao „zlatni standard“ u polju ciljne isporuke antikancerskih lekova (koncept pasivne ciljne isporuke), u međuvremenu je delom osporen, pošto klinički rezultati ne podržavaju prekliničke nalaze na animalnim modelima. Stoga, sve češće se postavlja pitanje validnosti tvrdnji o efikasnosti terapije na bazi EPR efekta, odnosno pitanje o budućnosti nanomedicine bez EPR efekta, te razvoja klinički relevantnog modela EPR efekta?

Ipak, interes na polju daljeg specijalizovanja nano-antikancerskih lekova ne opada, što podržava i određeni (mada mali) broj FDA-odobrenih ili terapija u različitim fazama kliničkih ispitivanja iz grupe PEG-ilovanih liposoma, polimernih micela/konjugata/nanočestica, dendrimera, karbonskih nanotuba, koji uglavnom teže da isporuče hemioterapeutik do molekulskih meta koje su specifično eksprimirane na površini tumorskih ćelija; ovi nanonosači treba da transportuju hemioterapeutik do tumora, izbegavajući normalna tkiva i redukujući mu toksičnost, citostatik štite od degradacije, produžuju polu-vreme eliminacije, poboljšavaju rastvorljivost i efektivnost leka, smanjuju renalni klirens. Dodatno, u obzir se uzimaju posebnosti tumorskog mikro-okruženja, kroz razvoj nanonosača responsivnih na endogenu stimulaciju (pH varijacije, redoks gradijent, koncentracija enzima, promena temperature), ili uz udruženu primenu sa egzogenim stimulacijama (ultrazvuk, magnetno polje, svetlost), što zahteva skupu opremu.

# **PRECISE ANTI-CANCER THERAPIES: HOW DOES PHARMACEUTICAL TECHNOLOGY CONTRIBUTE TO THEM?**

**Snežana Savić**

Department of Pharmaceutical Technology and Cosmetology, University of Belgrade-Faculty of Pharmacy (Serbia)

Precision/Personalized medicine aims to individualize therapeutic interventions/drug delivery on the basis of ex vivo and in vivo information on patient- and disease-specific characteristics. Use of nanotechnology associated with human health (nanomedicine) in the light of precise diagnosis/treatment of various tumors has for a long time an indispensable place in such context. While surgery and radiotherapy are the primary treatments used for local and non-metastatic cancers, anti-cancer drugs (chemotherapeutics, hormones, biologics, immunotherapies) present the current approach in metastatic cancers. The indiscriminate destruction of normal cells, the toxicity of conventional chemotherapeutics, the development of multidrug resistance support the idea to find new effective targeted therapies, based on the changes in the molecular biology of tumor cells. The major underlying concept in the design of anti-cancer nanomedicines for longer was based on Enhanced Permeability and Retention effect (EPR), widely accepted as „gold standard” in the field of cancer passive targeting, but in meantime was partially handed down, as it worked in rodents but not in humans. Therefore, as the question on validity of claiming the efficacy of anti-cancer therapy via the EPR effect came out, it is probably time to ask what is the future of nanomedicine without the EPR effect and possibility to develop a new clinically relevant EPR effect model?

Still, the interest in the field of further specialized development of nano-anti-cancer drugs don't subside, supported by several (although only few) FDA-approved targeted therapies or those undergoing different phases of clinical trials (PEGylated liposomes, polymeric micelles/conjugates/nanoparticles, dendrimers, carbon nanotubes), mainly tending to deliver cytotoxic drugs to molecular targets overexpressed on the tumor cells surface; these nanocarriers should not only transport chemotherapeutic to tumor, avoiding normal tissues and reducing its toxicity, but also protect it from degradation, increase the half-life, payload and solubility and reduce renal clearance. In addition, some peculiarities of tumor microenvironment are taken into consideration through the development of endogenous stimuli-responsive nanocarriers (pH variations, redox gradient, enzyme concentration, temperature change), or accompanied with exogenous stimuli application (ultrasound, magnetic field, light), that requires expensive equipment.

## SAVREMENI PRISTUPI U KONTROLI KVALITETA BIOLOŠKIH LEKOVA

Borut Štrukelj

Farmaceutski fakultet, Univerzitet u Ljubljani (Slovenija)

Za razvoj proizvodnog procesa i za razvoj rutinskih analiza koje se primenjuju tokom proizvodnje bioloških lekova potrebno je primeniti veliki broj specifičnih analitičkih metoda, kao što su različite gel elektroforeze (PAGE), jono-izmjenjivačka hromatografija (IEC), cirkularni dihroizam (CD), ekskluziona hromatografija (SEC), kapilarna zonska elektroforeza (SDS-PAGE), florescentna spektroskopija, natrijum-dodecil sulfat poliakrilamid gel elektroforeza (SDS-PAGE), cepanje laserskim snopom i masena spektrometrija uz matriks potpomognutom jonizacijom laserskom desorpcijom (MALDI-TOF), micelarna elektrokinetska hromatografija, hidrofobna interakcija i reverzno fazne hromatografije (HIC, RPC), čelijski i enzimsko – imunski testovi (ELISA). Nabrojane analitičke metode koriste se za analizu biološke aktivnosti i u ovom radu će biti predstavljen kritički osvrt na iste. Biološki lekovi mogu izazvati imunološke reakcije koje mogu dati ozbiljnu kliničku sliku i ozbiljne neželjene reakcije. Potencijalna imunogenost kao i neželjene reakcije koje su posledica heterogene strukture mAbs predstavljaju najnepoželjnije reakcije izazvane primenom bioloških lekova. Razvili smo opštu platformu zasnovanu na čelijskoj tehnologiji, koja se oslanja na ispitivanje ranih DC-vodenih dogadaja kojima se inicira CD-4 T-čelijski zavistan humani adaptivni imuni odgovor, uključen u praćenje potencijalne imunogenosti.

U prvom delu istraživanja pretražena je literatura. Za otkrivanje potencijalne imunogenosti primenjeno je in vitro određivanje DC-sazrevanje i DC-stimulacija na mAbs i PBMC, praćenjem proliferacije mAbs strukturnih varijanti. Određen je potencijalni kapacitet agregata mAbs da podstaknu (indikuju) sazrevanje dendritičnih ćelija. DC i PBMC čelijsko određivanje bilo je optimizovano, a mAbs strukturne varijante proizvedene su podvrgavanjem mAb1 uzoraka stresnim uslovima svetlosti i zamrzavanja. Testirani su sledeći markeri: CD40, CD80, CD86, CCR7 i HLA-DR, kao i citokini: IL-1 $\beta$ , IL-6, IL-8, IL-10, IL-12p70 i TNF- $\alpha$ . Pokazano je da razvijena platforma pruža dovoljno informacija na osnovu kojih je moguće predvideti imunogenost. Za određivanje fizičko-hemijskih, strukturnih i bioloških karakteristika bioloških lekova potrebno je primeniti čitav set modernih analitičkih tehnika. Mi smo razvili brzu, specifičnu i pouzdanu metodu za detekciju potencijalne imunogenosti bioloških lekova, koja je posledica varijabilnosti u njihovoj strukturi.

# **CONTEMPORARY APPROACHES IN BIOLOGICAL DRUGS QUALITY CONTROL**

**Borut Štrukelj**

Faculty of Pharmacy, University of Ljubljana (Slovenia)

Production process development and routine analysis during the production of biologicals incorporate a large number of specific analytical methods, such as various gel electrophoresis (PAGE), ion-exchange chromatography (IEC), circular dichroism (CD), size-exclusion chromatography (SEC), capillary zone electrophoresis (CZE), fluorescence spectroscopy, sodium-dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE), laser-light scattering and matrix-assisted laser-desorption ionization time of flight (MALDI-TOF) mass spectrometry, micellar electrokinetic chromatography, hydrophobic interactions and reversed phase chromatographies (HIC, RPC) and cell-based and enzyme-linked immunoassays (ELISA) to study biological activity. In present paper, the critical review of such methods will be elucidated. Biological medicinal products may induce immune responses leading to serious clinical side effects. Potential immunogenicity as well as other side-effects due to the heterologous mAbs structure is one of the most undesirable effects. We developed the general platform using a cell-based technology, which relies on interrogation of the early DC-driven events that initiate CD-4 T-cell dependent humoral adaptive immune responses, integrated into the monitoring of potential immunogenicity.

For the first part of the study, the literature survey was applied. For the detection of potential immunogenicity, In-vitro DC maturation and DC stimulation assays on mAb and PBMC proliferation assay on mAb structural variants was used. Potential capacity of the mAb aggregates to induce maturation of dendritic cells has been determined. Both DC and PBMC cell assay was optimised and mAB structure variants were produced by using light stressed and freeze/thaw stressed mAb1 samples. The following markers were tested: CD40, CD80, CD86, CCR7 and HLA-DR, as well as cytokines: IL-1 $\beta$ , IL-6, IL-8, IL-10, IL-12p70 and TNF- $\alpha$ . It was demonstrated that the developed platform is enough informative for the prediction of immunogenicity. The complex toolbox of modern analytical techniques are required for the determination of physico-chemical, structural and biological characteristic of biological medicinal products. We developed fast, specific and reliable method for the detection of potential immunogenic effect of biologicals due to their variation in structure.

## **MICRO-PHOTOGRAMMETRY AS A NOVEL TOOL FOR CHARACTERISATION OF DISSOLUTION BEHAVIOUR OF PHARMACEUTICAL DOSAGE FORMS**

**Alessandra D'Angelo<sup>1</sup>, Mike Reading<sup>2</sup>, Milan Antonijevic<sup>1</sup>**

<sup>1</sup>Department of Pharmaceutical, Chemical and Environmental Sciences,  
University of Greenwich, <sup>2</sup>University of Huddersfield (United Kingdom)

Many novel and enhanced drug delivery systems are in the development or have been developed in recent years. Such rapid development of formulations demands a better and more rigorous analytical support and assessment. Work presented here aims at the development of an innovative analytical technique for the 3Dchemical mapping of pharmaceutical dosage forms, via simultaneous topographic characterisation and dissolution analysis of solid drug delivery systems.

The system is composed of micro-photogrammetry apparatus, dissolution system and software for data analysis. Micro-photogrammetry consists of a microscope which is held by magnets in position on a semi-circular support. The sample is placed on rotating platform that moves under computer control as the images are taken. Obtained images are processed by software which delivers 3D image of the object from which further analysis can be carried. Dissolution system consists of assembly that allows periodic ie. stepwise dissolution where sample is subjected for certain period of time to a medium after which medium is collected and analysed by HPLC and sample left to dry and analysed using photogrammetry. These steps are then repeated multiple times to gain a full dissolution profile. This methodology allows in-depth analysis of fast dissolution processes by subjecting sample to a small portion of medium for a very limited period of time, usually 10-60seconds. Dissolution of Ibuprofen sugar coated tablets (Bristol Laboratories) was conducted in seven discontinuous steps. Analysed 3D models of the formulations revealed a small volume displacement induced by the dissolution of the Sucrose outer layer, followed by a significant variation of tablet volume after exposure of the product components underlying the outer sugar layer. Volumetric analysis revealed that diffusion controlled the initial stages of the dissolution process, followed by erosion. Novel methodology presents a great potential in evaluating dissolution profiles of diverse pharmaceutical systems.

## **KONCEPTUALNI MODEL ZA UNAPREĐENJE SISTEMATSKE KONTROLE**

**Gordana Pejović**

Agencija za lekove i medicinska sredstva Srbije, Fakultet organizacionih nauka,  
Univerzitet u Beogradu (Srbija)

Substandardni lekovi na tržištu su veliki rizik u lečenju. Stoga nacionalni regulatorni autoriteti za lekove preduzimaju različite mere za kontrolu lekova na tržištu. Posebna pažnja se poklanja sistematskoj kontroli, koja se sprovodi nasumičnim uzorkovanjem lekova sa nacionalnih tržišta.

Izvršena je sveobuhvatna analiza regulative u oblasti lekova u regionu jugoistočne Evrope kako bi se sagledalo regulatorno okruženje za nadzor nad tržištem.

Predložen je konceptualni model za unapređenje procesa sistematske kontrole na regionalnom tržištu lekova, kojim su opisane i obuhvaćene sve važne regulatorne institucije na evropskom i regionalnom nivou. Opisana je i moguća harmonizacija regulatornih mehanizama koja bi usledila nakon usvajanja opisanog konceptualnog modela. Mogući pristup za unapređenje sistematske kontrole na regionalnom tržištu je osnivanje regionalnog centra za ispitivanje ospozobljenosti, koji će upravo biti namenjen sistematskoj kontroli. To bi mogla biti jedna od agencija za lekove u region, koji bi razvila posebne šeme za ispitivanje ospozobljenosti za kontrolu kvaliteta lekova na tržištu u region. Sistematska kontrola je ključna regulatorna mera za unapređenje kvaliteta lekova na tržištu, kojom se osigurava da lekovi substandardnog kvaliteta ne budu dostupni pacijentima.

## **CONCEPTUAL MODEL FOR THE IMPROVEMENT OF MARKET SURVEILLANCE PROCESS**

**Gordana Pejović**

Medicines and Medical Devices Agency of Serbia, Faculty of Organisational Sciences, University of Belgrade (Serbia)

Substandard quality of medicines on the market poses significant risk for disease treatment. Therefore, national medicines regulatory authorities (NMRAs) are applying various regulatory mechanisms to control authorised medicines on the market. The special emphasis is given to the market surveillance process, which is intended for the quality control of randomly sampled medicines from the national market.

A comprehensive review of medicines legislation in South East Europe (SEE) countries was undertaken to summarize regulatory framework for national market surveillance.

The conceptual model for the improvement of regional market surveillance process was developed, describing all the important regulatory institutions at European and regional level. The possible harmonisation of regulatory mechanisms resulting from the implementation of described conceptual model is described. The possible approach is to improve the regional market surveillance process through the introduction of regional proficiency testing (PT) centre, aimed at market surveillance. This centre could be one of NMRAs in the region, which would develop a specific PT schemes aimed at quality control of marketed medicines in the region. Market surveillance is the key regulatory measure for the improvement of medicines quality on the market, assuring that the substandard drugs are not available to patients.

## **ANALITIKA POLARNIH SUPSTANCI PRIMENOM METODE TEČNE HROMATOGRAFIJE HIDROFILNIH INTERAKCIJA**

**Biljana Jančić Stojanović**

Katedra za analitiku lekova, Univerzitet u Beogradu-Farmaceutski fakultet  
(Srbija)

Poslednjih godina, tečna hromatografija hidrofilnih interakcija (eng. Hydrophilic interaction liquid chromatography-HILIC) pojavljuje se kao korisna alternativa za revezno-faznu tečnu hromatografiju u analizi polarnih jedinjenja. Cilj ovog rada je da prikaže primenljivost HILIC metode u farmaceutskoj analizi različitih polarnih analita.

U HILIC metodi korsite se umereno polarne kolone i polarne mobilne faze sa visokim procentom organskog rastvarača (obično više od 50% organskog rastvarača). HILIC retencioni mehanizmi su veoma kompleksni i mogu uključiti više procesa kao što su: raspodela analita između adsorbovane vode na površini stacionarne faze i bulk-a mobilne faze, adsorpcije analita na površini stacionarne faze i jonske izmene između nanelektrisanog analita i suprotno nanelektrisanih grupa na površini stacionarne faze. U kojoj meri učestvuju navedeni procesi zavisi od značajnog broja faktora kao što su: vrsta stacionarne faze, polarnost i stepen ionizacije analita, kao i sastav mobilne faze. Iz tog razloga, retencioni procesi u HILIC-u još nisu dobro proučeni i predmet su brojnih kontraverznih tumačenja. Uzimajući u obzir navedene činjenice neophodan je pažljiv izbor HILIC uslova za analizu svakog polarnog analita. Kao rezultat rastuće popularnosti HILIC-a poslednjih godina je uloženo puno truda kako u teorijska istraživanja HILIC-a tako i u process razvoja HILIC metode. Time, u ovoj studiji kroz više eksperimentalnih primera prikazana je specifičnost razvoja HILIC metode.

# **HYDROPHILIC INTERACTION LIQUID CHROMATOGRAPHY (HILIC) AS A VALUABLE ALTERNATIVE FOR REVERSED-PHASE LIQUID CHROMATOGRAPHY (RP-LC) IN THE ANALYSIS OF POLAR COMPOUNDS**

**Biljana Jančić Stojanović**

Department of Drug Analysis, University of Belgrade-Faculty of Pharmacy  
(Serbia)

Hydrophilic interaction liquid chromatography (HILIC) has emerged in recent years as a valuable alternative to reversed-phase liquid chromatography (RP-LC) in the analysis of polar compounds. The aim of this study is to present applicability of HILIC method in pharmaceutical analysis of different polar analytes. Analysis of certain polar analytes using different polar stationary phases has been performed.

In HILIC, polar or moderately polar columns and less polar aqueous-highly organic mobile phases (usually above 50% of the organic solvent) are used. HILIC retention mechanism is rather complex and can involve several processes: partition of the analyte between the adsorbed water-enriched layer on the surface of the stationary phase and the bulk mobile phase, adsorption of the analyte on the stationary phase surface and ion-exchange between the charged analyte and oppositely charged groups on the stationary phase surface. The involvement of each process depends on considerable number of factors that are related to: the type of the stationary phase, the polarity and ionization of the analytes and the mobile phase composition. Therefore, HILIC retention process is still insufficiently elucidated and a subject of many controversial interpretations. Considering all those facts, careful investigation of HILIC conditions for any polar analyte is necessary.

As a result of a growing popularity of HILIC in recent years a lot of effort is put into both, theoretical investigations of HILIC systems and HILIC method development process. Thus, in this study through several experimental examples specificity of HILIC method development is presented.

## MOGUĆNOSTI PRIMENE EKOLOŠKI PRIHVATLJIVIH HROMATOGRAFSKIH METODA U KONTROLI LEKOVA

**Ana Protić, Nevena Maljurić, Biljana Otašević, Mira Zečević**

Katedra za analitiku lekova, Univerzitet u Beogradu-Farmaceutski fakultet  
(Srbija)

Upotreba toksičnih organskih rastvarača, u prvom redu acetonitrila zbog njegovog povoljnog uticaja na hromatografsku efikasnost, je nezaobilazna kod metoda reverzno-fazne tečne hromatografije (RP-HPLC). Na žalost, acetonitril pored povoljnih fizičko-hemijskih osobina poseduje i druge, ne tako dobre osobine, kao što su toksičnost, zapaljivost i štetnost po životnu sredinu. Iz tog razloga sve više se radi na pronaalaženju različitih strategija kako bi se ove metode prevele u ekološki prihvatljive i kako bi se utrošak toksičnog organskog rastvarača sveo na najmanju moguću meru. Cilj ovog rada je ispitivanje različitih načina na koje RP-HPLC metode mogu postati ekološki prihvatljive, kao i mogućnost primene ovog koncepta u kontroli lekova posmatrano sa regulatornog aspekta.

Gradijentna RP-HPLC metoda za ispitivanje stabilnosti dronedaron-hidrohlorida modifikovana je u UHPLC i micelarnu HPLC metodu (MLC). Thermo scientific calculator primjenjen je pri geometrijskom transferu HPLC u UHPLC metodu, dok je metodologija površine odgovora primenom Design Expert 7.0.0. softvera iskorišćena za vizuelizaciju eksperimentalnog prostora i razvoj MLC metode. Stepen ekološke prihvatljivosti novo predloženih metoda ispitana je pomoću vrednosti analitičkog ekološkog skora.

Prilikom geometrijskog transfera HPLC u UHPLC metodu ukazala se potreba za dodatnim modifikacijama kako bi se zadržala efikasna separacija ispitivanih supstanci. Sa druge strane MLC metoda je zahtevala potpuno novi razvoj shodno drugaćijim retencionim mehanizmima. Konstruisanje 3D-grafikona omogućilo je predlaganje najoptimalnijih uslova. Ekološki analitički skor za gradijentu UHPLC metodu iznosio je 83 poena, dok je za MLC metodu iznosio 90 poena od ukupnih 100. Sa druge strane, modifikacija hromatografske metode uslovljava prijavu varijacije kao i određivanje vrste varijacije i obim izmene registracione dokumentacije.

Pokazano je da je MLC metoda ekološki prihvatljivija na osnovu većeg ekološkog analitičkog skora, mada su oba bila veća od 70 što obe metode kvalifikuje kao ekološki prihvatljive. Bilo kakva promena metode zahteva prijavu varijacije regulatornim organima. Ukoliko se dokaže njen značajno poboljšanje varijacija bi bila tipa IA.

## **PROSPECTS OF ECOLOGICALLY ACCEPTABLE CHROMATOGRAPHIC METHODS IN DRUG CONTROL**

**Ana Protić, Nevena Maljurić, Biljana Otašević, Mira Zečević**

Department of Drug Analysis, University of Belgrade-Faculty of Pharmacy  
(Serbia)

Toxic organic solvents are widely used when working with reversed-phase liquid chromatography (RP-HPLC). Acetonitrile is the most commonly used regarding its outstanding chromatographic efficiency and other beneficial physical-chemical characteristics. Unfortunately, acetonitrile is very toxic, flammable and harmful for living world. For this reason the scientists are looking for different ways of limiting the consumption of this solvent. The aim of this work is to investigate different possibilities of greening RP-HPLC methods and to evaluate whether this concept could be incorporated into the analytical methods used for drug control.

Stability-indicating RP-HPLC method with gradient elution for the analysis of dronedarone-hydrochloride was modified to UHPLC and micellar HPLC method (MLC). Thermo scientific calculator was used during geometrical transfer of HPLC to UHPLC method, while Response Surface Methodology (RSM), performed in Design Expert 7.0.0, was applied for the visualization of the experimental space and development of MLC method. Ecological acceptability of these methods was evaluated using eco-scale score.

During geometrical transfer UHPLC method, the additional modifications of the chromatographic conditions were necessary in order to preserve efficient separation of all investigated compounds. On the other side, different retention mechanisms involved in MLC demanded development of new chromatographic method. 3D-graphicon construction enabled definition of optimal chromatographic conditions and eco-scale score was 83 and 90 points out of 100 for UHPLC and MLC method, respectively. From the regulatory point of view, modification of chromatographic conditions involves denunciation of the variation, determining type of the variation and level of registration documentation change. Eco-scale score pointed out that MLC is more ecologically acceptable than UHPLC method. However, eco-scale score above 70 points for both methods indicates its green character. Any change of the analytical method leads to denunciation of the variation to the regulatory affairs. If method's significant improvement is demonstrated the variation type could be IA.

**Plenarna predavanja**

**Plenary Lectures**

# SADRŽAJ – CONTENTS

PL1

**LONGEVITY EXTENSION, HEALTHSPAN AND HEALTHY AGING: THE ROLE OF PHARMACOLOGICAL AND NUTRITIONAL INTERVENTION**

**ZDRAVO STARENJE I PRODUŽETAK DUŽINE I KVALITETA ŽIVOTA: ULOGA FARMAKOLOŠKE I NUTRICIONE INTERVENCIJE**

- **Janko Nikolić-Žugić**

**79**

PL2

**PUTEVI I STRANPUTICE U LEČENJU PARKINSONOVE BOLESTI: KUDA DALJE?**

**PATHWAYS AND DEAD-ENDS IN TREATMENT OF PARKINSON'S DISEASE**

- **Vladimir S. Kostić**

**81**

PL3

**TRADITIONAL AND INNOVATIVE TECHNOLOGIES FOR MANUFACTURING ORAL FIXED DOSE COMBINATION DOSAGE FORMS**

- **Paolo Colombo**

**83**

PL4

**ANALYZING THE EVIDENCE OF CLINICAL PHARMACY SERVICES: HOW TO IMPROVE**

- **Fernando Fernandez-Llimos**

**84**

## **ZDRAVO STARENJE I PRODUŽETAK DUŽINE I KVALITETA ŽIVOTA: ULOGA FARMAKOLOŠKIH I NUTRICIONIH INTERVENCIJA**

**Janko Nikolić-Žugić**

Katedra za imunobiologiju, Medicinski fakultet i Centar za starenje,  
Univerzitet Arizone u Tusonu (SAD)

Broj osoba starijih od 65 godina, koji u mnogim zemljama označava doba odlaska u penziju, raste velikom brzinom širom sveta, pa je procenjeno da će do 2050. godine dostići preko 1,2 milijarde ljudi. U mnogim industrijskim zemljama, procenat stanovništva u ovomu zrastu dostiže i prevazilazi trećinu celokupnog stanovništva. I dok ovo produženje dužine života predstavlja značajno dostignuće savremenih zdravstvenih nauka, kvalitet života zaostaje za dužinom u značajnom segmentu starije populacije. Ovo se ogleda u nagomilavanju hroničnih bolesti, koje ne samo da smanjuju kvalitet života, već donose veliki trošak zdravstvenom i privrednom sistemu pojedinih zemalja s obzirom na visoke troškove kompleksnog lečenja i nege, te izgubljene radne sate, kako obolelih tako i članova njihovih porodice, odnosno onih koji ih neguju. Ovo predavanje će razmotriti koncept Geronauke, koji se zasniva na ideji da bazični molekularni i celularni procesi starenja predstavljaju osnovu za razvoj hroničnih bolesti, kao što su neurodegenerativne, srčane, metaboličke i maligne ebolesti. U drugom delu će se diskutovati kako se ovi procesi mogu modulirati nutricionim i farmakološkim metodama, i koliko su ta saznanja primenljiva kod ljudi.

# **LONGEVITY EXTENSION, HEALTHSPAN AND HEALTHY AGING: THE ROLE OF PHARMACOLOGICAL AND NUTRITIONAL INTERVENTION**

**Janko Nikolic-Žugich**

Department of Immunobiology and the University of Arizona Center on Aging,  
University of Arizona College of Medicine-Tucson (USA)

Demographic explosion of aging, or the “silver tsunami”, has engulfed the globe, and it has been predicted that by 2050, the world will have more than 1.2 billion people older than 65. These older adults will be making one third or more of the population in many industrial countries between 2030 and 2050. While this is certainly a great societal success, driven primarily by health sciences, a great number of this population suffers from multiple chronic conditions, and therefore does not enjoy a healthspan (defined as the number of years spent in good health) to match its lifespan. The net effect of poor healthspan is not only affecting the person suffering from it, but rather the health and economic system of entire countries (if not the world). Older adults with multiple comorbidities exact a high health cost due to complex care, but even more, are both a direct and indirect economic burden due to lost direct working hours of themselves and their caretakers. Finally, caretakers themselves are more prone to illness. This lecture will discuss the concept of Geroscience, the idea that the fundamental processes underlying the aging process simultaneously set stage or drive many, if not all, of the age-related chronic conditions, including neurodegenerative, cardiovascular, metabolic, inflammatory and malignant diseases. I will further review current knowledge on how to manipulate the aging process, and the chronic diseases that accompany it, by dietary and pharmacologic means, and discuss how far these concepts are from broad application in humans.

## **PUTEVI I STRANPUTICE U LEČENJU PARKINSONOVE BOLESTI: KUDA DALJE?**

**Vladimir S. Kostić**

Klinika za neurologiju, Klinički centar Srbije, Univerzitet u Beogradu -  
Medicinski fakultet (Srbija)

U skorašnjem revijskom radu analizirane su 143 studije koje su ispitivale efikasnost u lečenju Parkinsonove bolesti (PB). Zaključeno je da su u monoterapiji početnih faza PB efikasni neergotski agonisti dopamina (DA), oralni preparati levodope, selegilin i rasagilin, dok se kao moguća dopunska terapija stabilnih formi PB preporučuju neergotski DA, rasagilin i zonisamid, kao i fizioterapija, uz rivastigmin koji je verovatno koristan, posebno kod bolesnika sa poremećenim hodom. U kontroli motornih fluktacija preporučuju se neergotski DA, pergolid, levodopa ER, intestinalne infuzije levodope, enta- i opikapon, rasagilin, zonisamid, safinamid i obostrana DMS STJ i GPi, dok su amantadin, klozapin i obostrana DMS STJ i GPi dejstveni u kontroli diskinezija. Međutim, najveći problem je nepostojanje terapije koja usporava neizbežnu progresiju neurodegenerativnog procesa u PB. U ovom radu diskutovaćemo takve mogućnosti u okviru 4 kategorije: (1)  $\alpha$ -sinuklein, (2) patogenetski mehanizmi koji nisu direktno vezani za  $\alpha$ -sinuklein, (3) ne-SNCA genetske subtipove PB i (4) moguće intervencije koje modifikuju progresiju PB, a ne utiču direktno na njenu patobiologiju. Nažalost, nije poznato da li se relevantni patofiziološki mehanizmi odigravaju u identičnom vremenskom sekvenciјalnom sledu kod svih klinički zahvaćenih bolesnika ili se razlikuju zavisno od molekularnog podtipa ove bolesti, što bi neumitno diverzifikovalo i terapijski pristup.

## **PATHWAYS AND DEAD-ENDS IN TREATMENT OF PARKINSON'S DISEASE**

**Vladimir S. Kostić**

Institute of Neurology, Clinical Centre of Serbia, University of Belgrade - School of Medicine (Serbia)

In a recent review a total of 143 new studies have been analyzed for their effectiveness in the treatment of Parkinson's disease (PD). It has been shown that clinically useful as a monotherapy of early PD were nonergot dopamine agonists (DAs), oral levodopa preparations, selegiline, and rasagiline, as adjunct therapy in early/stable PD, nonergot DAs, rasagiline, and zonisamide, for adjunct therapy in optimized PD for general or specific motor symptoms including gait, rivastigmine was possibly and physiotherapy clinically useful. In a control of motor fluctuations, most nonergot DAs, pergolide, levodopa ER, levodopa intestinal infusion, entacapone, opicapone, rasagiline, zonisamide, safinamide, and bilateral STN and GPi DBS proved to be clinically useful, while amantadine, clozapine, and bilateral STN DBS and GPi DBS were clinically useful for dyskinesia. The greatest unmet therapeutic need in PD is the development of treatment that slows the relentless progression of the neurodegenerative process. Herein we discuss these possibilities in a frame of the following 4 categories: (1)  $\alpha$ -synuclein, (2) pathogenic mechanisms distinct from  $\alpha$ -synuclein (most also potentially triggered by  $\alpha$ -synuclein toxicity), (3) non-SNCA genetic subtypes of "PD," and (4) possible disease-modifying interventions not directly influencing the underlying PD pathobiology. It is still unknown whether the relevant pathophysiological mechanisms occur in a sequential fashion across most clinically affected individuals or manifest differentially in independent molecular subtypes of PD.

## **TRADITIONAL AND INNOVATIVE TECHNOLOGIES FOR MANUFACTURING ORAL FIXED DOSE COMBINATION DOSAGE FORMS**

**Paolo Colombo**

University of Parma (Italy)

Last AAPS conferences presented a number of sessions on drug product manufacturing technologies, exploring the possibility to construct personalized oral drug products. These technologies open major opportunities for pharmaceutical companies that in the combination in one dosage form of known drugs could find novel products. This lecture intends to illustrate a novel manufacturing technology, useful for adapting the drug product to patient requirements.

Pharma industry is looking to innovation and portfolio improvement through processes that simplify the fabrication and provide proprietary products. At the same time, the Regulatory Agencies are promoting the innovation and the combination of drugs, in front of recognized advantages for patients and payers. My personal experience attempted to cover this innovation need by developing a module assembling technology as an instrument for achieving the construction of personalized medicines characterized by adaptable fixed dose combination. This process was based on the typical solid dosage form manufacturing i.e., tablet compression. The new technology will be compared with the existing classical process.

## **ANALYZING THE EVIDENCE OF CLINICAL PHARMACY SERVICES: HOW TO IMPROVE**

**Fernando Fernandez-Llimos**

Faculty of Pharmacy - University of Lisbon (Portugal)

Pharmacists can improve patients' health outcomes with a series of interventions that constitute the clinical pharmacy services. But, in a highly competitive world with limited financial resources, reporting that a service is effective with a good designed study is not sufficient. A single study, even with the best design and rigorous conduct, will always have influence of known or unknown confounders that limit its generalizability. To solve this situation, the concept of evidence-based practice emerged. Synthesizing evidence means gathering the results from different studies with different confounders that biased the results differently, with the aim of balancing these biases and obtaining a picture that represents a closer image to the reality.

Many studies assessing the impact of clinical pharmacy services have been published, with different results. The quality of these studies is also very heterogeneous. When measured in terms of risk of bias, the way that we should evaluate the reliability of a randomized controlled trial, one can find room for improvement.

But this is especially true when evaluating the impact of clinical pharmacy services by the means of evidence-based methods, such as systematic reviews with or without meta-analyses. In this presentation, we will explore the results of several of these systematic reviews, and highlight their weaknesses as well as the primary studies weaknesses. Definitely, pharmacists can improve patients' health outcomes, but researchers and practitioners should make a joint effort to demonstrate this impact under the evidence-based procedures.