14th EFLM Symposium for Balkan Region

Neighbouring Countries: The Same Professional Aim

Plenary Sessions / Abstracts

UDK 577.1 : 61 **ISSN 1452-8258**

J Med Biochem 37: 240–257, 2018

Plenarne sekcije
Plenary sessions

LABORATORIJSKA MEDICINA: JUČE, DANAS I SUTRA

Ana-Maria Šimundić

Zavod za medicinsko-laboratorijsku dijagnostiku
Klinička bolnica Sveti Duh, Zagreb, Hrvatska

U toku je proces transformacije zdravstvene zaštite prema sistemu koji je orijentisan na pacijenta i kvalitet. Glavni cilj ove evolucije je maksimalno povećanje koristi za pacijente, kroz osiguranje najboljeg ishoda uz minimiziranje troškova, oštecenja i otpada. Neke zdravstvene discipline i organizacije su već u naprednoj fazi ovog procesa, dok se druge i dalje trude da pronađu svoj put. Koja je uloga laboratorijske medicine u ovom procesu i kako da ostanemo u korak sa ovom transformacijom? Štaviše, kako možemo da preuzmemo aktivnu ulogu u ovome, a ne samo da budemo posmatrači? Brojni izazovi stoje na našem putu, da pomenemo samo neke: povećanje obima posla, nedostatak osoblja, budžetska ograničenja, promena politike nadoknade i nedostatak javnih sredstava. Uz rame sa ekonomskim napretkom idu i brojne tehnološke inovacije, kao što su minijaturizacija, automatizacija, konsolidacija, informaciona tehnologija, »biobanking«, generisanje velikih količina podataka, nove molekularne dijagnostičke tehnike, »point-of-care« instrumenti itd. koji već sada, a nastaviće i u bliskoj budućnosti da dramatično menjaju našu profesiju. Moraćemo da pronađemo način da prihvatimo ove inovacije i da ih u potpunosti iskoristimo proaktivno radeći sa našim kolegama u sistemu zdravstvene zaštite, a pod okriljem laboratorijske medicine. Jedan od najvećih izazova u ovoj evolucionoj adaptaciji jeste kako pokazati značaj laboratorijske medicine i kako povezati značaj laboratorijskih ispitivanja sa ishodom pacijenta. Očigledno, to se može postići samo kroz kompetentnost, znanje i interdisciplinarno istraživanje. Takođe ćemo morati da naučimo da promovišemo našu profesiju i njegovu ulogu u zdravstvenoj zaštiti. Moraćemo da pređemo sa naše tradicionalne uloge koja zahteva samo tehničke veštine i kliničke kompetencije, i na druge neophodne i suštinske sposobnosti/veštine kao što su organizacione, ekonomske, liderstvo i menadžment. Obezbeđivanje usluge visokog kvaliteta treba

LABORATORY MEDICINE: YESTERDAY, TODAY AND TOMORROW

Ana-Maria Šimundić

Department of Medical Laboratory Diagnostics
Clinical Hospital Sveti Duh, Zagreb, Croatia

The transformation of healthcare towards value-based and patient centred system is ongoing. Main goal of this evolution is to maximize the value for patients, through ensuring best possible patient outcome while minimizing cost, damage and waste. Some healthcare disciplines and organizations are already in the advanced stage of this process, whereas others are still struggling to find their way through. Which is the role of laboratory medicine in this process and how do we stay abreast of this transformation? Moreover, how can we take the active role in this and not just be bystanders and observers? A number of challenges stand in our way: increasing workload, staff shortage, budget constraints, changing reimbursement policies and shortage of public funding, to name just a few. Side by side with economic drives, there are also numerous technological advances and breakthroughs, such as miniaturization, automation, consolidation, information technology, biobanking, generation of big data, emerging molecular diagnostic techniques, point-of-care instrumentation, etc., which have already and will continue to dramatically change the way we practice our profession in the near future. We will need to find a way to embrace these innovations and work proactively with our peers in the healthcare system to utilise them to their full potential under the broad umbrella of laboratory medicine. One of the greatest challenges in this evolutionary adaptation shall be to learn how to demonstrate the value of laboratory medicine and how to connect the value of laboratory testing with patient outcome. Obviously, this can only be achieved through competence, knowledge and interdisciplinary research. We will also have to learn to promote our profession and its role within the healthcare. We will have to move from our traditional role which requires only technical skills and build not only our clinical competences but also put a great emphasis on other necessary and essential compe-

da bude najveći prioritet našeg posla. Predviđanje kako će laboratorijska medicina izgledati za par decenija je intrigantno i izazovno, ako ne i gotovo nemoguće. Ipak, na nama je da preuzmemo aktivnu ulogu i budemo ključni akter u strateškom razvoju sistema zdravstvene zaštite.

tences such as organizational, economic, leadership and management. Delivering high value care should be on the top of our agenda. Predicting how laboratory medicine will look like in a couple of decades from today is intriguing and challenging, if not almost impossible. Nevertheless, it is up to us to take the active role and be the key player in the strategic developments of the healthcare evolution.

EFLM PROJEKAT »RAZMENA PRAKTIČNOG ZNANJA I VEŠTINA U LABORATORIJSKOJ MEDICINI« – »EFLMLabX«

Evgenija Homšak

Univerzitetski Klinički Centar Maribor, Odsek za laboratorijsku dijagnostiku, Maribor, Slovenija
EFLM WG-CPE, Predsedavajući

U mnogim društvima članicama EFLM postoji potreba za sticanjem dodatnih praktičnih znanja i veština u različitim oblastima Laboratorijske Medicine (LM), koje se mogu dobiti u drugim laboratorijama u zemlji ili inostranstvu. Do sada nije bilo moguće, bilo na formalnom ili neformalnom nivou, da se pronađe veza sa takvim dodatnim, ali vrlo važnim obrazovanjem, posebno za mlade pripravnike, ali i za sve druge evropske stručnjake za laboratorijsku medicinu (EuSpLM), koji žele deliti znanje LM na drugom nivou. Pod okriljem EFLM-a, kao glavne i centralne evropske profesionalne organizacije, sada postoji opcija koja bi mogla pomoći u rešavanju ovog problema: projekat »Razmena praktičnih znanja i veština u laboratorijskoj medicini« koju priprema Radna grupa za kongrese i postdiplomsko obrazovanje (WG-CPE). Prema prvim pozitivnim rezultatima ankete (146 odgovora) o potrebama i interesima da ponude različite prakse u oblasti LM, koje smo dobili od Nacionalnih društava EFLM (28) u 2015. godini, zapazili smo veliko interesovanje za obuku i razmenu praktičnih znanja i veština u LM među zemljama EFLM. Zbog toga smo bili stimulisani za naredni korak dalje, za kreiranje internet stranice programa kojim bismo realizovali projekat. Cilj ovog EFLM projekta je kreiranje i upravljanje mrežom medicinskih laboratorija koje žele i mogu da pruže praktičnu obuku u različitim oblastima / aspektima Laboratorijske medicine. Mogućnosti obuke mogu se kretati od posete, opšte specijalističke obuke za sticanje veština neophodnih za specijalizovane merne metode ili sisteme (uvođenje novih IVD sistema), istraživačke metode koje pružaju grupe u laboratorijama do praktičnih kurseva o temama vezanim za laboratorijsku medicinu. U okviru glavne internet stranice EFLM razvijen je namenski veb program EFLMLabKs, koji nudi mogućnost pretraživanja i prijave na gore pomenute ponude i uspostavljanje

EFLM PROJECT »EXCHANGE OF PRACTICAL KNOWLEDGE AND SKILLS IN LABORATORY MEDICINE« – EFLMLabX

Evgenija Homšak

University Clinical Centre Maribor, Department for Laboratory Diagnostics, Maribor, Slovenia
EFLM WG-CPE, Chair

In many EFLM Member Societies there is a need to acquire additional practical knowledge and skills in different fields of Laboratory Medicine (LM), which may be obtained in other laboratories in the country or abroad. Until now, there were no possibilities, on official and open way to find the link to such additional but very important education, especially for young trainees, but also for all other European Specialists of Laboratory Medicine (EuSpLM), who want to share the knowledge of LM on different level. Under the umbrella of EFLM, as a main and central European professional organization, there is now an option which could help to address this problem: the project »Exchange of practical knowledge and skills in Laboratory Medicine« which the Working Group for Congresses & Postgraduate Education (WG-CPE) is developing. According to the first very positive survey results (146 responses) about needs and interest to offer different practices on the field of LM, that we have obtained from EFLM National Societies (28) in 2015, we noted a great interest for training and exchange of practical knowledge and skills in LM amongst EFLM countries. Therefore, we were stimulated for next further step to create the website program to start and running the project. The aim of this EFLM project is to create and operate a network of medical laboratories willing and able to offer practical training in various fields/aspects of Laboratory Medicine. Training opportunities in the database can range from visiting, general specialist training to gaining skills necessary for specialized measurement methods or systems (introduction of new IVD systems), research methods provided by groups in laboratories and practical courses in laboratory medicine related topics. A dedicated website EFLMLabX program was developed within the frame of the main EFLM website, offering the possibility to search and

direktnih veza i komunikacija između laboratorija i korisnika prakse. Na kraju svake zaključene prakse, svaki učesnik ce dobiti sertifikat o pohađanju.

Koristeći ovakav projekat, moći ćemo na nivou EFLM:

- postici viši nivo iskustva u različitim oblastima laboratorijske dijagnostike,
- podeliti znanje i iskustvo u praksi u različitim laboratorijskim ustanovama,
- učiti i razmenjivati znanje i veštine različitih dijagnostičkih metoda (specifični-GC -MS, novi IVD sistemi ...),
- steći znanja i veštine specifične oblasti dijagnostike (u smislu razvoja i uvođenja nove dijagnostike u interesu svih korisnika laboratorije),
- pružiti mogućnost mladim pripravnicima i specijalistima da prošire kontakte sa (između) stručnjaka (motivacija za rad i istraživanje),
- da dobijemo priliku za istraživački rad i kao deo toga dobijemo dodatna znanja i veštine na akademskom nivou (npr. pisanje naučnih članaka),
- pronaći nove potencijalne saradnike na dijagnostičkim poljima / istraživačkoj nauci.

Da bismo podržali ovu važnu razmenu znanja i veština u LM, posebno za mlade pripravnike, želeli bismo da stvorimo fondaciju za stipendiste, koju će podržati IVD partneri. Stipendijama bi se mogli pokriti troškovi putovanja, smeštaja i eventualne naknade za edukaciju učesnika. Na taj način ćemo moći da stimulišemo razmenu znanja između različitih profesionalaca / laboratorija / institucija i zemalja EFLM. Ovim projektom, u skladu sa dobijenim višim nivoom znanja / iskustva u različitim oblastima laboratorijske dijagnostike opšte profesionalne populacije (EuSpLM) i boljom mrežom između profesionalaca, stručnjaka i naučnika, dobićemo viši opšti kvalitet naše profesije.

apply to the above mentioned offer and to establish direct links and communications between both, providers and users /applicants of practices. At the end of each concluded practice, each participant will receive Certificate of attendance.

With such a project we will be able on EFLM level:

- to achieve higher level of experience on different field of laboratory diagnostics,
- to share the knowledge and experience among practice in different lab-institution,
- to learn and get/ exchange the knowledge and skills of different diagnostic methods (specific-GC –MS, new IVD systems...),
- to get the knowledge and skills of specific field of diagnostics (in terms of development and introduction of new diagnostics as an interest of participant/applicant's Laboratory-institution),
- to offer the opportunity for the young trainees and specialist to expanding contacts with (between) the experts (motivation for work and research),
- to get the opportunity for research work and as part of that to get additional knowledge and skills on academic level (e.g. writing scientific articles),
- to find the new potential co-workers on diagnostic fields/research science.

To support this important exchange of knowledge and skills in LM, especially for young trainees, we would like to create the fundation for Bursaries, that will be supported by IVD partners. With bursaries would be possible to cover expences for traveling, accomodation and possible fee for education of participants. On that way we would be able to stimulate the exchange of knowledge between different professionals/laboratories/institutions and EFLM countries. With this project, according to obtained higher level of knowledge/experience on different field of laboratory diagnostics of general professional population(of EuSpLM), better networks between professionals, experts, and scientists we will gain higher general quality of our profession.

ORGANIZACIJA VANJSKE KONTROLE KVALITETA U LABORATORIJSKOJ MEDICINI BOSNE I HERCEGOVINE

Jozo Ćorić, E. Kučukalić, E. Begović

OJ Klinička hemija i biohemija,
Klinički centar Univerziteta u Sarajevu,
Bosna i Hercegovina

Udruženje medicinskih biohemičara Bosne i Hercegovine (UMBBiH) je u proteklih 20 godina, dva puta godišnje provodila vanjsku kontrolu kvaliteta laboratorija za medicinsku biohemiju u Bosni i Hercegovini. Učešće u vanjskoj kontroli kvaliteta je na dobrovoljnoj osnovi i obuhvatalo je oko 50 laboratorija. Zahvaljujući kontroli kvaliteta rada, u Bosni i Hercegovini postaju dostupni biohemijski, hematološki, koagulacijski i imunološki programi. Poboljšanje kvaliteta medicinskih laboratorija, omogućeno od strane UMBBiH, važno je s obzirom da rezultirajuća analiza laboratorija učesnica obezbeđuje nezavisnu i objektivnu procjenu ponovljivosti primijenjenih analitičkih metoda u laboratorijama, upoređivanje različitih analitičkih metoda te kontinuiranog nadzora trenutnog stanja laboratorijske opreme i primjenjene tehnologije, kao i preporučene analitičke metode. Uzorci koji se koriste za kontrolu kvaliteta uključuju komercijalne materijale sa unaprijed definisanim vrijednostima za različite analitičke metode i različite analizatore. Laboratorije učesnice su odgovorne za rukovanje uzorcima kontrole na isti način kao sa uzorcima pacijenata. Laboratorije su bile obavezne da analiziraju neprihvatljive rezultate uključujući i rezultate koji utiču na pacijente, kao i da pokrenu korektivne akcije. U nacionalnoj šemi vanjske kontrole za medicinske biohemijske laboratorije, ciljne vrednosti laboratorijskih ispitivanja su definisane kao srednje ciljne vrednosti grupe ± 2SD prema različitim metodama i različitim analizatorima. Harmonizacija metode ocjene rezultata sa međunarodno priznatim kriterijima, glavni je cilj Komiteta UMBBiH za provođenje vanjske kontrole kvaliteta rada.

ORGANIZATION OF EXTERNAL QUALITY CONTROL IN LABORATORY MEDICINE IN BOSNIA AND HERZEGOVINA

Jozo Ćorić, E. Kučukalić, E. Begović

Department for Clinical Chemistry and Biochemistry,
Clinical Center University of Sarajevo,
Bosnia and Herzegovina

The Association of Medical Biochemists in Bosnia and Herzegovina (AMBBH) has conducted External Quality Assessment (EQA) of medical biochemistry laboratories in Bosnia and Herzegovina twice a year for the past 20 years. Participation in EQA has been voluntary and it includes circa 50 laboratories. Thanks to the EQA, clinical chemistry, haematology, coagulation and immunology Survey programs have become available in Bosnia and Herzegovina. Quality improvement of medical laboratories, enabled by the EQA is important given that the resulting analysis of participating laboratories provides an independent and objective evaluation of reproducibility of applied analytical methods on a large number of participating laboratories, comparison of different analytical methods and continuous surveillance of the current state of laboratory equipment and applied technology as well as recommended analytical methods. Samples used for EQA controls included commercial materials with predefined target values for different analytical methods and different analysers. Participating laboratories are responsible for handling the EQA samples in the same manner as patient samples. Furthermore, laboratories were obligated to analyse unacceptable results including results that impact patients and also to initiate corrective actions. In the national EQA scheme for medical biochemical laboratories, target values of laboratory test results are defined as mean target values of the group ± 2SD according to different methods and different analysers. Harmonisation of the result assessment method with internationally recognized criteria to the full extent, is the main goal of the EQA Committee of the AMBBH.

ULOGA ADIPOZNOG TKIVA U ODRŽAVANJU ENERGETSKE HOMEOSTAZE

Dragana Puhalo Sladoje

Univerzitetska bolnica Foča, Republika Srpska

Adipozno tkivo je centralni metabolički organ u regulisanju energetske homeostaze organizma. Bijelo masno tkivo funkcioniše kao ključni energetski rezervoar. Masno tkivo pored različitih metabolita, masnih kiselina, luči i veliki broj signalnih proteina (citokina) – adipokina. Do danas je izolovano više od 600 različitih adipokina. Uloge adipokina u humanom organizmu su brojne i različite, učestvuju u kontroli mnogih metaboličkih procesa, utiču na metabolizam ugljenih hidrata i lipida, krvni pritisak, sistem koagulacije krvi, kao i na iflamatorne procese. Adipocitima pripadaju leptin i adiponektin koje luče adipociti, zatim rezistin, visfatin, retinol vezujući protein-4, kao i niz različitih citokina kao interleukin-6 (IL-6), tumor nekrozis faktor-α (TNF-α), inhibitor aktivatora plazminogena-1 (PAI-1) i drugi. Adipokini koje sintetišu adipociti pokazuju različite endokrine funkcije, a adipocitokini koje proizvode makrofagi masnog tkiva djeluju parakrino i kontrolišu metabolizam adipocita. Poremećena sekrecija adipocitokina koja prati gojaznost ima važnu ulogu u razvoju različitih kardiometaboličkih poremećaja, ukuljučujući metabolički sindrom, dijabetes mellitus tip 2, zapaljenske i vaskularne promjene.

Ključne riječi: adipozno tkivo, adipocitokini, energetska homeostaza.

THE ROLE OF ADIPOSE TISSUE IN MAINTAINING ENERGY HOMEOSTASIS

Dragana Puhalo Sladoje

University Hospital Foča, Republic of Srpska

Adipose tissue is central metabolic organ in energy homeostasis regulation of the organism. White adipose tissue functions as a key energetic reservoir. Aside from different metabolic and fatty acids, adipose tissue excretes a great number of signal proteins as well (cytokines) which are called adipocytokines or adipocytes. So far, more than 600 different adipokines have been isolated which interact with central and peripheral organs such as brain, liver, pancreas and skeletal muscles. These adipokines control different processes, such as food intake, energy consumption, lipid and carbohydrate metabolism, blood pressure, inflammations of blood clots. The adipocytokine group contains: leptin and adiponectin which are excreted exclusively by adipocytes, resistin, visfatin, retinol binding protein 4, but also a large number of cytokines such as interleukin 6(IL-6), tumor necrosis factor- α (TNF-α), inhibitor activator plasminogen – 1 (PAI-1), etc. Although many of these adipocytokines are synthesized in adipocytes, they have different endocrine functions, others are being produced in macrophages of adipose tissue and act paracrine in order to control metabolism of adipocytes. It is also evident that dysregulation of adipocyte secretion, which happens in obesity, plays a crucial role in development of different cardiometabolic disorders, including metabolic syndrome, diabetes mellitus type 2, inflammatory and vascular changes.

Key words: adipose tissue, adipocytokines, energy homeostasis.

ANKETA O UNOSU JODA U MAKEDONIJI

Sonja Kuzmanovska

Institut za patofiziologiju i nuklearnu medicinu, Medicinski fakultet, Skoplje, Makedonija

Adekvatni unos joda je važan za homeostazu štitaste žlezde. I nedostatak joda i višak joda mogu uticati na funkciju štitne žlezde i njenu morfologiju posledično dovodeći do poremećaja u metabolizmu, rastu i razvoju. Poremećaji nedostatka joda uključuju ne samo gušavost i hipotiroidizam, već i mentalnu retardaciju, oštećenu reprodukciju i povećanu stopu mrtvorođenih. Preporučeni dnevni unos joda od 90–290 μg zavisi od starosti i fiziološkog statusa. Kako su

IODINE INTAKE SURVEY IN MACEDONIA – A TEN YEARS GAP

Sonja Kuzmanovska

Institute of Pathophysiology and Nuclear Medicine, Medical Faculty, Skopje, R Macedonia

The adequate iodine intake is important for thyroid gland homeostasis. Both iodine deficiency and iodine excess can affect thyroid function and morphology, with an impact on metabolism, growth and development. Iodine deficiency disorders include not only goiter and hypothyroidism, but also mental retardation, impaired reproduction and increased rate of stillbirths. Recommended daily iodine intake from 90–290 μg is dependent on age and physiolog-

prirodni izvori joda niski i ograničeni, jodiranje hrane (soli, hleba) je jedini izvor dnevnog unosa. S obzirom na pojavu endemske gušavosti u nekim regijama zemlje, Makedonija, kao federalna jedinica unutar Jugoslavije, uvela je univerzalno jodiranje soli od 1953. godine sa 10 mg/kg KI. Ubrzo nakon sticanja nezavisnosti 1991. godine, pilot studija je pokazala da je deficit joda postojao u nekim regijama u zemlji. Od 1995. godine smo uspeli započeti projekat procene nedostatka joda na celoj teritoriji, finansijski podržan od strane UNICEF-a. Kao rezultat, 2000. godine novi profilaktički opseg od 20–30 mg/kg joda postao je obavezan za jodiranje soli. Uskoro, 2003. godine, Republika Makedonija je proglašena zemljom sa dovoljnim unosom joda, prema kriterijumima SZO/UNICEF/ICCIDD. Najpouzdaniji indikator unošenja joda u populaciji je procena koncentracije urinarnog joda (UI). UI analize se izvode u centralizovanoj laboratoriji za procenu joda u Institutu za patofiziologiju i nuklearnu medicinu. Koristi se spektrofotometrijska metoda klasifikovana kao »Metoda A« od strane SZO/UNICEF/ICCIDD, a stručnost Laba potvrđuje se putem EKUIP programa eksternog kvaliteta koji organizuje CDC, USA. Rezultati istraživanja su izraženi kao medijana koncentracija (μg/L). Počevši sa 117 μg/L 1995. godine, vrednosti u narednim istraživanjima 1999–2003. porasle su do 191 μg/L, pri čemu je vrednost postavljena kao »adekvatan unos joda« (100–199 μg/L). Nakon postizanja dovoljne količine joda, UNICEF je povukao finansijsku podršku, a mi smo uspeli da nastavimo sa istraživanjem samo još dve godine. Dobijeni rezultati (228 μg/L u 2005. i 241 μg/L u 2007.) pokazali su »više od adekvatnog unosa joda« (200–299 μg/L). Iako su ovi rezultati bili od posebnog interesa, zbog ograničenog finansiranja, istraživanja su prekinuta u narednih 10 godina. Rizik od pojave viška joda, uz zdravstvene posledice izazvane jodom, bio je prisutan. Na kraju, u 2016. godini uspeli smo da se pridružimo ogromnom Panevropskom projektu »EUThyroid« – koji ima za cilj da proceni status joda u Evropi kroz usklađivanje rezultata istraživanja. Što se tiče ocene UI, usklađivanje rezultata ispitivanja uključuje upoređivanje naših rezultata sa onima dobijenim od strane referentne laboratorije na Nacionalnom institutu za zdravlje i socijalni rad u Helsinkiju, Finska. Do kraja ovog projekta (sredinom 2018. godine) moći ćemo predstaviti pouzdan status unosa joda za našu zemlju i planirati dalje aktivnosti.

ical status. As iodine natural sources are low and restricted, food (salt, bread) iodination is the only source of daily intake. With the past history of endemic goiter in some regions of the country, Macedonia, as federal unit within Yugoslavia, has introduced universal salt iodination since 1953 with 10 mg/kg of KI. Soon after the independency in 1991, a pilot study indicated that iodine deficiency still existed in some regions of the country. Since 1995, we were able to start a project to assess the iodine deficiency for the whole territory, financially supported by UNICEF. As a result, in 2000 a new prophylactic range of 20–30 mg/kg iodine became obligatory for salt iodination. Soon after, in 2003, Republic of Macedonia has been declared iodine sufficient country, according the criteria of WHO/UNICEF/ICCIDD. The most reliable indicator of iodine intake in a population is the assessment of urinary iodine (UI) concentration. The UI analyses are performed in the centralized, dedicated Laboratory for Urinary Iodine Assessment at the Institute of Pathophysiology and Nuclear Medicine. A Spectrophotometric method, classified as »Method A« by WHO/UNICEF/ICCIDD is used and the proficiency of the Lab is confirmed through EQUIP, external quality program organized by CDC, USA. The results from the survey studies were expressed as median (μg/L) concentration. Starting with 117 μg/L in 1995, the median values from the consequent surveys performed 1999–2003 constantly increased up to 191 μg/L, a value categorized as »adequate iodine intake« (100–199 μg/L). After achieving iodine sufficiency, UNICEF withdrew the financial support, and we were able to continue the survey for only two more years. The results obtained (228 μg/L in 2005 and 241 μg/L in 2007) indicated »more than adequate iodine intake« (200–299 μg/L). Although these results should have gained special interest, due to limited funding, the surveys were discontinued for the next 10 years. The risk of reaching iodine excess, with iodine induced health consequences, was persistent. Finally, in 2016 we succeeded to join the huge Pan-European Project »EUThyroid« – intended to evaluate the iodine status in Europe through harmonized survey approach. With regards to UI assessment, the harmonization of the test results includes the comparison of our results with those obtained by the Reference Laboratory at the National Institute of Health and Welfare in Helsinki, Finland. By the end of this project (mid 2018), we would be able to present a reliable iodine intake status for our country and to plan further activities respectively.

INTERFERENCIJE U LABORATORIJSKIM TESTOVIMA

Najdana Gligorović Barhanović

Centar za kliničko-laboratorijsku dijagnostiku,
Klinički centar Crne Gore,
Podgorica, Crna Gora

Prisustvo interferirajućih supstanci u biološkim uzorcima može bitno uticati na tačnost i pouzdanost laboratorijskih rezultata i time pokrenuti cijlu kaskadu nepotrebnih dijagnostičkih testova, pogrešnih dijagnoza, pa čak dovesti i do uključivanje terapije sa potencijalno nepovoljnim efektima po zdravlje pacijenta. Interferencija se može definisati kao efekat supstance prisutne u uzorku koja mijenja stvarnu vrijednost rezultata, obično izraženu kao koncentracija ili aktivnost parametra. Iako im se često ne pridaje dovoljno značaja, mogu biti povezane sa bilo kojom fazom laboratorijskog procesa testiranja. Neadekavtno prikupljanje, transport i skladištenje uzorka u preanalitičkoj, interferencije vezane za samo izvođenje testa u analitičkoj i neblagovremeno saopštavanje, greške u komunikaciji i pogrešno tumačenje rezultata u postanalitičkoj fazi. Interferencije se najčešće klasifikuju kao egzogene i endogene. Egzogene su rezultat prisustvasupstanci koje se prirodnone nalaze u adekvatno prikupljenim i skladištenim uzorcima pacijenata. Uključuju lijekove i druge supstance koje se koriste u terapiji, radioaktivna ili fluorescentna jedinjenja, biljne preparate, prehrambene dodatke itd. Sistemi za uzorkovanje, odnosno različiti aditivi koje sadrže, takođe mogu nepovoljno utjecati na tačnost laboratorijskog rezultati. Svi postupci koji utiču na kvalitet uzorka (sakupljanje, transport, skladištenje, centrifugiranje) potencijalni su izvor interferirajućih komponenti. Iako noviji laboratorijski analizatori imaju deklarisane veoma niske vrijednosti caryovera; ne treba zanemariti ni taj potencijalni uzrok kontaminacije. Sa druge strane, endogene interfencije mogu biti posledica prisustva komponenti koje su sastavni dio kako zdravih, tako i patoloških uzoraka pacijenata. U njih spadaju hemoglobin, bilirubin, lipidi, različiti proteini, antitijela i supstance koje karakteriše ukrštena reaktivnost. Iako su efekti nekih interferirajućih supstanci relativno dobro poznati i nije ih zahtjevno identifikovati, prepoznavanje značajnog broja njih je veliki izazov. Specijalisti laboratorijske dijagnostike uvijek moraju imati na umu da neslaganje između kliničke slike i laboratorijskih nalaza nije samo problem klinicara, već da svaki takav događaj ukazuje na moguće prisustvo interferirajućih supstanci. Bez obzira na stavove da su, u precizno kontrolisanom cjelokupnom procesu testiranja, neblagovremeno identifikovane interferencije rijetke, činjenica da i njihov utjecaj može imati neželjene efekte po zdravlje pacijenta uvijek se mora uzeti u obzir. Detekcija interferirajućih supstanci često zahtijeva čitav niz koraka i nema uvijek za

INTERFERENCES IN LABORATORY ASSAYS

Najdana Gligorovic Barhanovic

Center for Clinical Laboratory Diagnostic,
Clinical Center of Montenegro, Podgorica,
Montenegro

Laboratory assays can be compromised with interference that can decrease the accuracy and reliability of the result, leading to inappropriate further diagnostic tests, incorrect diagnoses, and treatments with potentially unfavorable outcomes for the patient. Interference is defined as »the effect of a substance present in the sample that alters the correct value of the result, usually expressed as concentration or activity, for an analyte« i.e. it occurs when a substance or process falsely alters an assay result. Interference, serious but often underestimated problem, can be related to all the three phases of total testing process. Preanalytical can be consequence of inappropriate specimen collection, storage conditions and delays in transportation of samples to the laboratory. Analytical encompasses assay interference, while postanalytical can be caused by delays, errors in communication or misinterpretation of the results. By nature, interferences can be classified as exogenous or endogenous. Exogenous are result of presence of substances not naturally found in properly collected and stored patient samples, including drugs, substances used as therapy, radioactive or fluorescent compounds, herbal medicines, nutritional supplements etc. Blood collection devices components can adversely affect the accuracy of laboratory test results. All processes affecting the sample (collection, transport, storage, and centrifugation) and clots can potentially be a source of interfering substances. Newer laboratory analyzers declare a careful monitoring; but the »carryover« can be a source of contamination, too. Endogenous interference can be caused by substances that can occur in both healthy and pathological patient samples, like hemoglobin, bilirubin, lipids, proteins, antibodies and cross-reacting substances. Although some interferences are relatively well known and easy to identify, recognition of considerable number of them is a great challenge. Laboratory specialists should always keep in mind that the disagreement between the clinical picture and the laboratory findings is not only a clinician's problem, but every event of that type must raise doubts about the presence of interfering substances. Regardless of some viewpoints, considering that in the tightly controlled total testing process unidentified interference are rare, the fact that their impact may have adverse effects on patient's health should never be neglected. Detection of the interfering substances, often require a whole series of steps and does not always lead to their identification, but even in this case the laboratory should not report a definitive result, especially without com-

rezultat njihovu identifikaciju, ali čak i u tom slučaju laboratorija ne bi trebala izdavati konačni rezultat (izraženu kao koncentraciju ili aktivnost parametra), pogotovo ne bez komentara o njegovoj eventualnoj nepouzdanosti. Stalni nadzor i dobra komunikacija između laboratorije i kliničara, imaju centralno mjesto u minimalizaciji efekta interferencija.

NEKI TEŠKI METALI I ELEMENTI U TRAGOVIMA U HUMANOM MLEKU

Ayşegül Çebi

Fakultet zdravstvenih nauka, Univerzitet Giresun, Giresun, Turska

Humano majčino mleko je osnovni nutrijent koji sadrži esencijalne supstance za razvoj deteta. To je jedinstveni izvor hrane za bebu tokom prvih šest meseci laktacije. Humano mleko, pored makronutrijenata, sadrži i mikronutrijente, kao što su elementi u tragovima. Mnogi elementi u tragovima su potrebni za rast i razvoj novorođenčadi i dojenčadi. Promene u količini elemenata u tragovima mogu dovesti do promena u metabolizmu novorođenčadi i dojenčadi. Sastav majčinog mleka može se promeniti u zavisnosti od ishrane majki i faktora životne sredine. Žene koje doje i njihova dojenčad su više podložni neželjenim dejstvima toksičnih metala. Pored toga, gastrointestinalna apsorpcija toksičnih supstanci je brža kod novorođenčadi i dojenčadi. U više različitih zemalja, prijavljeno je da se neki toksični metali kao što su živa, olovo i kadmijum nalaze u humanom mleku. Cink ima važnu ulogu u transkripciji gena i metabolizmu i ulazi u sastav nekih enzima. Arsen, kadmijum i olovo su poznati kao potencijalno nefrotoksični, hemotoksični i neurotoksični elementi. Svetska zdravstvena organizacija (SZO) je 1989. godine u Ženevi objavila izveštaj o elementima u tragu u majčinom mleku. Objavljene su mnoge studije vezane za sadržaj teških metala i elemenata u tragovima humanog majčinog mleka.

Ključne reči: teški metali, elementi u tragovima, humano majčino mleko

ment on its possible unreliability. In order to minimalize interferences impact, is necessary to ensure constant surveillance of both the laboratory specialists and the clinicians.

SOME HEAVY METALS AND TRACE ELEMENTS IN HUMAN BREAST MILK

Ayşegül Çebi

Faculty of Health Sciences, Giresun University, Giresun, Turkey

Human breast milk is a basic nutrient containing essential substances for infant development. It is unique food source for suckling baby during the first six months of lactation. Human milk contains micronutrients such as trace elements in addition to macronutrients. Many of the trace elements are needed for the growth and development of newborns and infants. The changes in quantities of the trace elements may lead to different results in the metabolism for the newborns and infants. The composition of breast milk may change depending on the diet of the mothers and environmental factors. Breastfeeding women and infants are more susceptible to side effects of toxic metals. Furthermore, gastrointestinal absorption of the toxic substances is faster in newborns and infants. Some toxic metals such as mercury, lead and cadmium have been reported to found in human breast milk from different countries. Zinc has important role in transcription of genes and human metabolism, and is found in constituent of some enzymes. Arsenic, cadmium and lead are known as potentially nephrotoxic, hemotoxic and neurotoxic elements. In 1989, World Health Organization (WHO) declared a report about minor and trace elements in breast milk in Geneva. Many studies were published related with the content of heavy metal and trace elements in human breast milk from different countries.

Keywords: Heavy metal, trace element, human breast milk

EVROPSKA FEDERACIJA ZA KLINIČKU HEMIJU I LABORATORIJSKU MEDICINU – RADNA GRUPA ZA PREANALITIČKU FAZU: PREPORUKA ZA UZORKOVANJE VENSKE KRVI

Zorica Šumarac, Ana-Maria Šimundić, Karin Bolenius, Janne Cadamuro, Stephen Church, Michael P. Cornes, Edmée C. van Dongen-Lases, Pinar Eker, Tanja Erdeljanović, Kjell Grankvist, Joao Tiago Guimaraes, Roger Hoke, Mercedes Ibarz, Helene Ivanov, Svetlana Kovalevskaya, Gunn B.B. Kristensen, Giuseppe Lippi, Alexander von Meyer, Mads Nybo, Barbara de la Salle, Christa Seipelt, Pieter Vermeersch*

**Beograd, Srbija*

Radna grupa za preanalitičku fazu (WG-PRE) Evropske federacije za kliničku hemiju i laboratorijsku medicinu (EFLM) kao vodeći profesionalni entitet uključen u preanalitičku fazu, izradila je dokument pod nazivom: Evropska federacija za kliničku hemiju i laboratorijsku medicinu (EFLM) Radna grupa za preanalitičku fazu (WG-PRE): Preporuka za uzorkovanje venske krvi. Cilj ovog dokumenta je pružanje jednostavne, objedinjene, preporuke zasnovane na analizi rizika i naučnim dokazima za uzorkovanje venske krvi. Ovaj dokument pruža sveobuhvatan pregled najkritičnijih koraka za standardizovanu proceduru uzorkovanja venske krvi i daje praktična uputstva o tome kako uspešno prevazići potencijalne barijere i prepreke za njegovu široku primenu. Pored stručnjaka iz laboratorijske medicine, autori ovog dokumenta su predstavnici nacionalnih udruženja medicinskih sestara, bolničkih sestara, flebotomista i predstavnika proizvođača sistema za uzorkovanje krvi. Preporučena procedura zasnovana je na najboljim raspoloživim dokazima. Svaki korak je ocenjen korišćenjem sistema koji ocenjuje kvalitet dokaza i snagu preporuke. Proces ocenjivanja je obavljen na nekoliko sastanaka u kojima su učestvovali svi prethodno navedeni učesnici. Glavni delovi ove preporuke su: postupci pre uzorkovanja krvi, uzorkovanje, postupci posle uzorkovanja krvi i implementacija iste. Za uspešno sprovođenje preporuke u praksi od velikog značaja su: uspostavljanje sistema sertifikacije osoblja uključenog u postupak uzimanja uzoraka krvi, organizovanje kontinuirane edukacije, provere rada, ponovljene obuke osoblja, primena indikatora kvaliteta i ostalo. Sa ciljem implementacije ove preporuke, EFLM WG PRE je pripremila sledeće alate: power point prezentaciju, video sa opisom celog postupka, test znanja za procenu nivoa znanja i podizanje svesti osoblja pre i nakon edukacije, kontrolnu listu za proveru pri-

EUROPEAN FEDERATION OF CLINICAL CHEMISTRY AND LABORATORY MEDICINE – WORKING GROUP FOR PREANALYTICAL PHASE: RECOMMENDATION FOR VENOUS BLOOD SAMPLING

Zorica Šumarac, Ana-Maria Šimundić, Karin Bolenius, Janne Cadamuro, Stephen Church, Michael P. Cornes, Edmée C. van Dongen-Lases, Pinar Eker, Tanja Erdeljanović, Kjell Grankvist, Joao Tiago Guimaraes, Roger Hoke, Mercedes Ibarz, Helene Ivanov, Svetlana Kovalevskaya, Gunn B.B. Kristensen, Giuseppe Lippi, Alexander von Meyer, Mads Nybo, Barbara de la Salle, Christa Seipelt, Pieter Vermeersch*

**Belgrade, Serbia*

European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) Working Group for Preanalytical Phase (WG-PRE) as the leading professional entity involved in preanalytical phase, has provided a document named European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) Working Group for Preanalytical Phase (WG-PRE): Recommendation for venous blood sampling. The aim of this document is to provide a simple, condensed, risk and evidence based recommendation for venous blood sampling. The current document provides a comprehensive overview of the most critical steps for a standardized blood collection procedure and practical guidance on how to successfully overcome potential barriers and obstacles to its widespread implementation. In addition to specialists in laboratory medicine, the authors of this document are representatives of national nursing associations, hospital nurses, phlebotomists and representatives of manufacturers of blood collection systems. The recommended procedure is based on the best available evidence. Each step was graded using a system that scores the quality of the evidence and the strength of the recommendation. The process of grading was done at several face-to-face meetings involving the same mixture of stakeholders stated previously. The main parts of this recommendation are: pre-sampling procedures, sampling procedure, post-sampling procedures and implementation. For successful implementation in practice of great importance are establishing a system of certification of staff involved in the blood sampling procedure, organizing continuous education, auditing and re-training for all staff members, implementation of quality indicators and others. With this objective, EFLM WG PRE has prepared the following tools for the implementation of this recommendation: a power point presentation, a video

mene postupaka uzimanja uzoraka krvi tokom periodičnih provera i postere sa opisom cele procedure. Proces implementacije treba da se uradi kroz blisku multidisciplinarnu saradnju svih učesnika na nacionalnom nivou, kao što su nacionalna udruženja medicinskih sestara, stručna udruženja u laboratorijskoj medicini i nacionalni regulatorni organi. EFLM WG PRE želi da ohrabri stručnjake širom Evrope da usvoje i primene ovu preporuku, kako bi se poboljšao kvalitet postupka uzorkovanja krvi i povećala bezbednost pacijenata.

describing the entire procedure, a knowledge test to assess the level of knowledge and raise awareness of the staff prior and after the education, a checklist for auditing the blood sampling procedure during periodical observational audits and posters with a cartoon describing the entire procedure. The implementation process should be done as a joint effort in close multidisciplinary collaboration of all stakeholders at the national level such as national nursing associations, professional societies in laboratory medicine and national regulatory bodies. EFLM WG PRE would like to encourage professionals throughout Europe to endorse, adopt and implement this recommendation to improve the quality of blood collection practices and increase patient safety.

POVEZANOST POLIMORFIZMA TMPRSS6 A736V MATRIPTAZE-2 I MUTACIJA HFE SA KONCENTRACIJOM HEPCIDINA-25 I STATUSOM FE KOD PACIJENATA SA PRETERMINALNOM I TERMINALNOM BUBREŽNOM SLABOŠĆU

Miljan Savković, Danica Ćujić, Ivana Novaković, Sanja Simić-Ogrizović, Violeta Dopsaj

Centar za medicinsku biohemiju,
Klinika za nefrologiju Klinički centar Srbije,
Farmaceutski fakultet Univerzitet u Beogradu,
Institut za aplikaciju nuklearne energije INEP,
Medicinski fakultet Univerzitet u Beogradu,
Beograd, Srbija

Transkripcija Hepcidina-25, glavnog regulatora hemostaze gvožđa, je inhibirana od strane Matriptaze-2. Ova serinproteaza je kodirana TMPRSS6 genom i njegov polimorfizam A736V(rs 855791) u najvećoj meri određuje koncentraciju gvožđa kod zdravih individua. Najučestaliji »missense« polimorfizmi gena za hemohromatozu (HFE), C282Y (rs1799946) i H63D (rs1800562), su povezani sa nekontrolisanom apsorpcijom gvožđa iz hrane i posledičnim negativnim efektom slobodnih radikala usled nagomilavanja gvožđa. Cilj ove studije je da proceni da li TMPRSS6 A736V polimorfizami HFE mutacije imaju efekta na koncentraciju Hepcidina-25 i status gvožđa kod pacijenata sa preterminalnom i terminalnom bubrežnom slabošću. Bubrežni bolesnici na hroničnom dijaliznom tretmanu (HD grupa, n=107), preterminalni bubrežni bolesnici (CHD grupa, n=23) i kontrolna grupa (C grupa, n=29) su uključeni u ispitivanje. Hepcidin-25 je kvantitativno određivan u serumu korišćenjem Direktnog Hemiluminescentnog ELISA eseja (Corgenix, CO, USA).

ASSOCIATION OF POLYMORPHISM TMPRSS6 A736V MATRIPTASE-2 AND HFE MUTATIONS WITH HEPCIDIN-25 AND IRON STATUS IN PATIENTS WITH PRETERMINAL AND TERMINAL RENAL FAILURE

Miljan Savković, Danica Ćujić, Ivana Novaković, Sanja Simić-Ogrizović, Violeta Dopsaj

Center for Medical Biochemistry,
Clinic of Nephrology Clinical Center of Serbia,
Faculty of Pharmacy University of Belgrade,
Institute for The Application of Nuclear Energy INEP,
School of Medicine, University of Belgrade,
Belgrade, Serbia

Transcription of the main iron regulatory hormone Hepcidin-25 decreases by Matriptase-2. This serin protease is encoded by TMPRSS6 gene and its polymorphism A736V(rs 855791)is a major iron determinant in healthy individuals. The most frequent missense polymorphism in the hemochromatosis (HFE) gene, C282Y(rs1799946) and H63D (rs1800562), are associated with redundant iron intake from the diet and consequently with free radical reactions due iron accumulation. The aim of this study was to evaluate whether the TMPRSS6 A736V polymorphism and HFE mutations influences Hepcidin-25 levels and iron status in patients with preterminal and terminal renal failure. Hemodialysis-dependent chronic kidney diseases patients (HD group, n=107), preterminal renal failure patients (CHD group, n=23) and control group (C group, n=29) were enroled in the study. Quantitative measurement of Hepcidin-25 in human serum is determined using Chemiluminescent Direct ELISA assay (Corgenix, CO, USA). Concentrations of serum iron, transferin, fer-

Koncentracija serumskog gvožđa, transferina, feritina, totalnog kapaciteta vezivanja gvožđa (TIBC) je određivana korišćenjem komercijalnih dijagnostičkih testova (Roche®, Swiss). Parametri kompletne krvne slike su dobijeni testiranjem na automatskom hematološkom analizatoru Advia 2120i (Siemens Healthcare Diagnostics®, Germany). HFE genotip (varijante C282Y i H63D) i polimorfizam TMPRSS6 A736V su određivani alel specifičnom polimeraza lančanom reakcijom u realnom vremenu (Applied Biosystems®, Foster City, CA, USA). Distribucije varijanti C282Y i H63D HFE kao i A736V TMPRSS6 varijante nisu narušile Hardy-Weinbergov ekvilibrijum i nisu se statistički značajno razlikovale između grupa (p = 0,848). Frekvence mutacija rs 855791 i rs1799946 gena su bile 58% i 11%, dok je rs1800562 gena bila 2%. Ispitanici sa prisustvom GA genotipa rs1800562 su imali statistički niže vrednosti TIBC (p = 0,02). Nismo uočili razliku u koncentraciji Hepcidina-25 između rs1799946 polimorfizma (p = 0,507). Medijana koncentracije bioaktivnog Hepcidina-25 je bila statistički značajno viša u HD i CHD grupama (53,22 ng/mL, 59,26 ng/mL) u poređenju sa C grupom (8,17 ng/mL, (p = 0,000)). Koncentracija hemoglobina je bila statistički značajno niža u grupi iznad medijane Hepcidina-25 (p = 0,000). Uočena je pozitivna korelacija između Hepcidina-25 i feritina unutar svih grupa (r = 0,501 za C, r = 0,833 za HDi r = 0,851 za CHD, p = 0,01). Kod HD grupe Hepcidin-25 je negativno korelirao sa TIBC, hemoglobinom i transferinom (r = -0,44, r = -0,395 i r = -0,583, p = 0,000). Logistička univarijantna analiza je indikovala feritin (p = 0,000), hemoglobin (p = 0,000) i istovremeno prisustvo genotipova GG rs855791 i GA rs1800562 (p = 0,037) kao nezavisne prediktore Hepcidina-25, R = 0,712. Ovi rezultati ukazuju na povezanost istovremenog prisustva polimorfizma rs 1800562 i rs 855791 sa koncentracijom Hepcidina-25, dok GA genotip rs1800562 ima uticaj naTIBC kod CHD i HD pacijenata.

ritin, total iron binding capacity were obtained using commercialy diagnostic tests (Roche®, Swiss). Parameters of complete blood count were determined using automated hematological analyser Advia 2120i (Siemens Healthcare Diagnostics®, Germany). HFE genotype (C282Y and H63D variants) and the TMPRSS6 A736V polymorphism were assessed by Real-Time allele specific polymerase chain reaction assays (Applied Biosystems®, Foster City, CA, USA). The frequency distribution of the C282Y and H63D of HFE and A736V of the TMPRSS6 variantsdid not violate Hardy-Weinberg equilibrium and was not significantly different among groups (p=0,848). Frequency of mutations in rs 855791 and rs1799946 genes were 58% and 11%, while inrs1800562 gene was 2%. Patients with presence of GA genotype rs1800562 had significantly lower TIBC (p = 0.02). There was no difference in level of Hepcidin-25 among rs1799946 polimorphism (p = 0.507). The median concentration of bioactive Hepcidin-25 were significaly higher in HD and CHD group (53.22 ng/mL, 59.26 ng/mL) compared to control group (8.17 ng/mL, p = 0,000). Hemoglobin concentration were significantly lower in group above median of Hepcidin-25 (p = 0.000). We observed positive correlation between Hepcidin-25 and ferritin witnin all groups (r = 0.501 for C, r = 0.833 for HD and r = 0.851 for CHD, p = 0.01). In HD group Hepcidin-25 were negatively correlated with total Iron binding capacity, hemoglobin and transferin (r = -0.627, r = -0.395 and r = -0.583, p = 0.01). Logistic univariate analysis indicated ferritin (p = 0.000), Hemoglobin (p = 0.000) and simultaneously presence of genotypes GG rs855791 and GA rs1800562 (p = 0.037) as independent predictors of Hepcidin-25, R = 0.712. These findings suggest that concurrently presence of rs 1800562 and rs 855791 polymorphism are associated with Hepcidin-25 concentration, while GA genotype ofrs1800562 have influence on TIBC in CHD and HD patients.

POVEZANOST NIVOA GENSKE EKSPRESIJE REZISTINA I KONCENTRACIJE REZISTINA SA ISHEMIJSKOM BOLEŠĆU SRCA

Jelena Munjas

Katedra za medicinsku biohemiju, Farmaceutski fakultet, Univerzitet u Beogradu

Humani rezistin je proinflamatorni protein, koji može imati značajnu ulogu u razvoju ateroskleroze i IBS. Kako bi se doprinelo boljem razumevanju uloge rezistina u IBS, ciljevi ovog istraživanja su bili određivanje nivoa iRNK rezistina i njegovog receptora CAP1 u mononuklearnim ćelijama periferne krvi i koncentracija rezistina u krvi pacijenata sa IBS i kontrolne grupe (KG), kao i ispitivanje njihove povezanosti sa lipidnim i nelipidnim faktorima rizika za razvoj ateroskleroze. U ovu studiju uključeno je 95 pacijenata sa indikacijama za koronarnu angiografiju i 33 KG. Nivoi iRNK rezistina, CAP1 i CD36 receptora čistača su određeni metodom kvantitativnog PCR-a, koncentracije rezistina ELISA metodom, a subklase lipoproteinskih čestica elektroforezom na gradijentu nedenaturišućeg poliakrilamidnog gela. Koncentracije rezistina i nivoi iRNK CAP1 su bili značajno viši kod svih grupa pacijenata sa IBS u odnosu na KG ($p<0,001$; $p<0,001$). Pacijenti sa akutnim koronarnim sindromom su imali značajno više koncentracije rezistina u odnosu na druge grupe pacijenata ($p<0,05$). Nivoi iRNK rezistina se nisu značajno razlikovali između ispitivanih grupa. Pacijenti sa sadržajem malih, gustih LDL (mgLDL) čestica ≥50% su imali značajno više koncentracije rezistina ($p=0,031$) i nivoe iRNK rezistina ($p=0,004$) u odnosu na pacijente sa sadržajem mgLDL čestica<50%. Nezavisni prediktori koncentracija rezistina subili kreatinin i ukupni holesterol, adj$R^2=0,298$; nivoa iRNK rezistina dijametar LDL čestica i nivoi iRNK CD36, adj$R^2=0,192$; nivoa iRNK CAP1 trigliceridi i nivoi iRNK CD36, adj$R^2=0,352$; nivoa iRNK CD36 hsCRP, kreatinin i nivoi iRNK CAP1, adj$R^2=0,505$. U ovoj studiji uočena je značajna povezanost rezistina i IBS, posebno sa kliničkim manifestacijama IBS. Povišene vrednosti rezistina i nivoa iRNK rezistina su udružene sa proaterogenim lipidnim statusom, pri čemu dislipidemija može imati uticaj na ekspresiju gena za rezistin.

ASSOCIATION OF RESITIN GENE EXPRESSION LEVELS AND CONCENTRATIONS OF PLASMA RESTININ WITH CORONARY ARTERY DISEASE

Jelena Munjas

Department of Medical Biochemistry, Faculty of Pharmacy, University of Belgrade

Human resistin is a proinflammatory cytokine with a potential role in atherosclerosis and CAD development. The aims of this study were to determine resistin mRNA levels and mRNA levels of resistin receptor, CAP1, in peripheral blood mononuclear cells (PBMC) and circulating resistin concentrations in CAD patients and control group (CG), and to evaluate their association with lipid and non-lipid atherosclerosis risk factors. This study included 95 patients requiring coronary angiography and 33 CG. Circulating resistin was measured by ELISA; PBMC resistin, CAP1 and CD36 mRNA was determined by qPCR. LDL and HDL subclasses were determined by gradient gel electrophoresis. Plasma resistin and CAP1 mRNA were significantly higher in all groups of CAD patients compared to CG ($P<0.001$; $P<0.001$). Acute coronary syndrome patients had significantly higher plasma resistin compared to other groups of patients ($P<0.05$). Resistin mRNA levels were not significantly different between any of the investigated groups. Plasma resistin ($P=0.031$) and resistin mRNA ($P=0.004$) were significantly higher in patients with the proportion of sdLDL particles 50%, compared to the group with the relative proportion of sdLDL particles <50%. Multiple linear regression analysis identified creatinine and total cholesterol as independent predictors of plasma resistin (adj$R^2=0.289$); LDL particle diameter and CD36 mRNA levels of resistin mRNA (adj$R^2=0.192$); TG and CD36 mRNA of CAP1 mRNA (adjR2=0.376); hsCRP, creatinine and CAP1 mRNA levels of CD36 mRNA, (adj$R^2=0,505$). A significant association was found between resistin and CAD, especially with clinical severity of the disease. Elevated circulating resistin and resistin mRNA were associated with proatherogenic lipid status. Also, thepresence of proatherogenic lipid status could induce resistin gene expression.

REZISTENCIJA NA KLOPIDOGREL I GENETIČKE OSNOVE REZISTENTNOSTI

Dragana Bačković

Makler, Beograd, Srbija

Trombociti igraju ključnu ulogu u razvoju trombotičkih komplikacija kod pacijenata sa aterosklerotskim vaskularnim oboljenjima. Oralni antitrombocitni lekovi, aspirin i klopidogrel, predstavljaju »kamen temeljac« u tretmanu arterijskih tromboza. Međutim, i pored dokazane kliničke efikasnosti ovih lekova, značajan broj bolesnika nema dobar terapijski odgovor na aspirin i/ili klopidogrel. U literaturi, prevalenca rezistencije na klopidogrel varira od 5% do 44% i najveća je kod bolesnika sa moždanim udarom. CYP2C19 enzim je uključen u oba oksidaciona procesa metabolizma klopidogrela. Nekoliko farmakogenetičkih studija je pokazalo da osobe koje su nosioci CYP2C19*2 alela imaju smanjeno stvaranje aktivnog metabolita klopidogrela I visoku reaktivnost trombocita, što dovodi do povećanja rizika od razvoja neželjenih kardiovaskularnih događaja. Međutim, postoje studije koje nisu ukazale na uticaj CYP2C19 genotipa na klinički efekat klopidogrela niti na prediktivnu vrednost CYP2C19*2 varijante gena. Glavni cilj ove studije je bio da se ispita povezanost terapijskog odgovora na antitrombocitni lek klopidogrel, koji je praćen laboratorijskim ispitivanjem agregacije trombocita, i prisustva CYP2C19*2 varijante gena i da se utvrdi uticaj genetskih i negenetskih faktora na reaktivnost trombocita kod bolesnika sa stenozom karotidnih arterija kod kojih je izvršena karotidna endarterektomija. Zastupljenost CYP2C19*2 alela u ispitivanoj populaciji je 26,8%. Nosioci CYP2C19*2 varijante gena imali su značajno veće vrednosti ADP-indukovane agregacije trombocita u odnosu na bolesnike koji nisu nosioci pomenutog alela tokom perioda praćenja. 46,7% bolesnika među nosiocima CYP2C19*2 alela imali su loš terapijski odgovor na klopidogrel posle 30 dana terapije klopidogrelom, odnosno 17,1% bolesnika u grupi nosioca wild type genotipa (P=0,001). Pokazano je da su prisustvo CYP2C19*2 varijante gena i visok nivo ukupnog holesterola nezavisni faktori rizika lošeg terapijskog odgovora na klopidogrel. Bolesnici koji su nosioci CYP2C19*2 varijante gena imaju skoro 4,5 puta veći rizik da će nakon 30 dana uzimanja klopidogrela imati loš odgovor na lek u odnosu na bolesnike sa wild type genotipom. Utvrđeno je da model, koji uključuje nivo ukupnog holesterola i prisustvo CYP2C19*2 varijante gena, može biti prediktor lošeg odgovora na klopidogrel 30 dana nakon uzimanja leka. U ovom istraživanju, 25,4% interindividualne varijabilnosti u terapijskom odgovoru na lek objašnjeno je holesterolom i prisustvom pomenutog alela. Dakle, značajan deo varijabilnosti je ostao neobjašnjen, odnosno faktori rizika su ostali nepoznati.

THE ROLE OF GENETIC FACTORS IN RESISTANCE TO CLOPIDOGREL THERAPY

Dragana Bačković

Makler, Belgrade, Serbia

Platelets play a key role in the development of thrombotic complications in patients with atherosclerotic vascular disease. Oral antiplatelet drugs, aspirin and clopidogrel are the cornerstone therapy in the conditions characterized by the risk for arterial thrombosis. Despite their proven clinical effect, a considerable number of patients don't have an adequate response to aspirin, clopidogrel or both. According to published data, the prevalence of clopidogrel resistance varies from 5% and may be as high as 44% in patients with acute ischemic stroke. The CYP2C19 enzyme is involved in both oxidative steps of clopidogrel metabolism. Several pharmacogenomics studies have demonstrated that individuals who are carriers of CYP2C19*2 allele have a reduced conversion of clopidogrel into its active metabolite and higher platelet reactivity, which increases the risk of adverse cardiovascular events. However, there are contrasting conclusions in the literature regarding the predictive value of the CYP2C19*2 variant allele. The aim of our study was to determine the prevalence of low responsiveness to clopidogrel and to identify risk factors, both genetic (CYP2C19*2) and non-genetic, for low responsiveness based on platelet function testing in patients with carotid artery stenosis undergoing KE. In the study group, CYP2C19*2 genotype frequency was 26.8%. The platelet aggregation was significantly higher in patients carrying CYP2C19*2 allele than in the group of non-carriers in all three measurements. 46.7% of those who were carriers of this allele had low response to clopidogrel after 30 days of taking the drug, in contrast to 17.1% of patients who were not carriers of the CYP2C19*2 variant (P=0.001).The risk for being a clopidogrel low-responder is 4.5-fold higher for heterozygous for the CYP2C19*2 allele compared to homozygous for wild type after 30 days of taking the drug. Also, it has been found that a model that includes total cholesterol levels and the presence of CYP2C19*2 gene variant may be a predictor of a low clopidogrel responsiveness 30 days after taking the drug. However, a combination of these factors was responsible for 25.4% of the interindividual variability and most of the variation in platelet response to clopidogrel remains unexplained.

STATUS VITAMINA B12 KOD PACIJENATA SA POVEĆANIM RIZIKOM ZA DEFICIT – POVEZANOST SERUMSKIH BIOMARKERA I MORFOMETRIJSKIH PARAMETARA LEUKOCITA

Dragana Totošković[1], Violeta Dopsaj[2,3], Jelena Martinović[4]

[1]Farmaceutski fakultet Novi Sad, Novi Sad, Srbija
[2]Farmaceutski fakultet, Beograd, Srbija
[3]Klinički centar Srbije, Beograd, Srbija
[4]Dom zdravlja Rakovica, Beograd, Srbija

Morfometrijski parametri neutrofila i monocita (volumen, provodljivost, rasipanje svetlosti i standardne devijacije) ispitivani su kod 104 pacijenta sa rizikom za deficit kobalamina (>60 godina, simptomi dispepsije), prosečnim volumenom eritrocita 80–100 fL, C-reaktivnim proteinom 10 mg/L i normalnom funkcijom bubrega. Krvna slika i morfometrijski parametri su dobijenina Coulter® LH 750 Hematology Analyzer. Vitamin B12 i folat su određeni na Access Immunoassay System®. Homocistein (Hcy) je analiziran na UniCel DxC 800 Synchron Clinical Systems®. Metilmalonska kiselina (MMA) je urađena na AB Sciex QTRAP® 5500 System. Pacijenti su podeljeni u: Grupa 1 kobalamin>221 pmol/L, MMA 210 nmol/L, Grupa 2–kobalamin>221 pmol/L, MMA>210 nmol/L, Grupa 3–kobalamin 221 pmol/L, MMA 210 nmol/L, Grupa 4–kobalamin 221 pmol/L, MMA>210 nmol/L. Hemoglobin, hematokrit, širina distribucije volumena eritrocita, morfometrijski parametri, folat i Hcy su upoređeni između grupa. Veće vrednosti Hcy bile su u Grupi 4 u poređenju sa Grupom 1 (P<0,05) i Grupom 3 (P<0,05). Standardna devijacija provodljivosti neutrofila (NeC-SD) bila je veća u Grupi 2 (P<0,05), Grupi 3 (P<0,05) i Grupi 4 (P<0,05) u poređenju sa Grupom 1. Koristeći granične vrednosti od 221 pmol/L za kobalamin i 210 nmol/L za MMA, pacijenti sa izolovano niskim kobalaminom ili povećanom MMA nisu imali veće vrednosti Hcy u odnosu na pacijente sa adekvatnim statusom vitamina B12, iako su uočene promene u unutrašnjoj morfologiji neutrofila na osnovu NeC-SD.

VITAMIN B12 STATUS IN PATIENTS AT INCREASED RISK OF DEFICIENCY – THE RELATIONSHIP BETWEEN SERUM BIOMARKERS AND MORPHOMETRIC PARAMETERS OF LEUKOCYTES

Dragana Totošković[1], Violeta Dopsaj[2,3], Jelena Martinović[4]

[1]Faculty of Pharmacy Novi Sad, Novi Sad, Serbia
[2]Faculty of Pharmacy, Belgrade, Serbia
[3]Clinical Centre of Serbia, Belgrade, Serbia
[4]Health Center Rakovica, Belgrade, Serbia

Morphometric parameters of neutrophils and monocytes (volume, conductivity, light scatter and standard deviations) were evaluated in 104 patients at risk of cobalamin deficiency (>60 years, symptoms of dyspepsia), mean corpuscular volume 80–100 fL, C-reactive protein 10 mg/L and normal renal function. Complete blood count and morphometric parameters were obtained by Coulter® LH750 Hematology Analyzer. Cobalaminand folate were determined on Access Immunoassay System®. Homocysteine (Hcy) was measured by UniCel DxC 800 Synchron Clinical Systems®. Methylmalonic acid (MMA) was obtained byAB SciexQTRAP® 5500 System. Patients were classified into: Group 1–cobalamin>221 pmol/L and MMA 210 nmol/L, Group 2–cobalamin>221 pmol/L and MMA>210 nmol/L, Group 3–cobalamin 221 pmol/L and MMA 210 nmol/L and Group 4–cobalamin 221 pmol/L and MMA>210 nmol/L. Hemoglobin, hematocrit, red blood cell distribution width, morphometric parameters, folate and Hcy were compared between groups. The results showed that Group 4had higher levels of Hcy compared to Group 1 (P<0.05) and Group 3 (P<0.05). Standard deviation of neutrophil conductivity (NeC-SD) was higher in Group 2 (P<0.05), Group 3 (P<0.05) and Group 4 (P<0.05) compared to Group 1. Using following cutoffs: 221 pmol/L for cobalaminand 210 nmol/L for MMA, patients with separately low vitamin B12 or high MMA did not have higher Hcy levels compared to patients with adequate vitamin B12 status, althoughchanged internal morphology of neutrophils was observed according to NeC-SD.

HOMEOSTAZA HOLESTEROLA I KARDIOMETABOLIČKI RIZIK

Tamara Gojković[1], Sandra Vladimirov[1], Vesna Spasojević-Kalimanovska[1], Aleksandra Zeljković[1], Jelena Vekić[1], Dimitra Kalimanovska-Oštrić[2], Zorana Jelić-Ivanović[1]

[1]Katedra za medicinsku biohemiju, Farmaceutski fakultet, Univerzitet u Beogradu
[2]Institut za kardiovaskularne bolesti, Klinički centar Srbije, Medicinski fakultet, Univerzitet u Beogradu

Poremećaji homeostaze holesterola mogu izazvati dislipidemiju, aterosklerozu i razvoj koronarne arterijske bolesti (KAB). Praćenje neholesterolskih sterola (NHS) kao markera sinteze i apsorpcije, može ukazati na rani razvoj dislipidemije. Ova studija ispitivala je povezanost obrazaca homeostaze holesterola sa tradicionalnim lipidnim parametrima. Studija je obuhvatila 47 KAB pacijenata bez terapije statinima i 31 kontrolu (KG). NHS su određeni metodom gasne hromatografije sa plamenojonizacijonom detekcijom (GCFID). Koncentracije ukupnog holesterola (TC), triglicerida (TG), LDL-holesterola (LDL-C), HDL-holesterola (HDL-C), apolipoproteina AI (apoA) i B100 (apoB) su merene rutinskim metodama. Za procenu homeostaze holesterola ispitanici su podeljeni na dobre i/ili loše sintetizere i/ili apsorbere prema medijalnim vrednostima latosterola i β-sitosterola (L/β). U KG, ispitanici sa povećanom apsorpcijom imali su više koncentracije apoB ukoliko im je sinteza holesterola bila povećana, u poređenju sa onima sa smanjenom sintezom ($p<0,05$). Koncentracija apoA bila je niža u podgrupi loših sintetizera/loših apsorbera u poređenju sa lošim sintetizerima/dobrim apsorberima ($p<0,05$). Pacijenti koji su imali povećanu sintezu, bez obzira na nivoe apsorpcije, imali su veće vrednosti TC i LDL-C ($p<0,01$, u oba slučaja). Rezultati ukazuju da su različiti obrasci homeostaze holesterola povezani sa različitim nivoima lipoproteina. Odnos L/β može se koristiti za procenu individualne sklonosti ka razvoju dislipidemije i odgovora na terapiju hipolipidemicima.

CHOLESTEROL HOMEOSTASIS AND CARDIOMETABOLIC RISK

Tamara Gojković[1], Sandra Vladimirov[1], Vesna Spasojević-Kalimanovska[1], Aleksandra Zeljković[1], Jelena Vekić[1], Dimitra Kalimanovska-Oštrić[2], Zorana Jelić-Ivanović[1]

[1]Department of Medical Biochemistry, Faculty of Pharmacy, University of Belgrade, Serbia
[2]Institute for Cardiovascular Diseases, Clinical Center of Serbia, Serbia, School of Medicine, University of Belgrade, Serbia

Disturbance of cholesterol homeostasis may cause dyslipidemia, atherosclerosis progression and coronary artery disease (CAD) development. Determination of noncholesterol sterols (NCSs), as synthesis and absorption markers, may indicate early development of dyslipidemia. This study investigates associations of different cholesterol homeostasis patterns with traditional lipid parameters. We included 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), triglyceride (TG), LDL-cholesterol (LDL-C), HDLcholesterol (HDL-C), apolipoproteins AI (apoA) and B100 (apoB) were measured by routine methods on an ILab 300+ analyzer. For cholesterol homeostasis estimation, each group was divided into good and/or poor synthetizers and/or absorbers according to lathosterol and β-sitosterol median values (L/β). In CG participants with elevated cholesterol absorption, apoB was higher in those with increased cholesterol synthesis compared to those with reduced synthesis ($p<0.05$), while apoA concentration was significantly lower in poor synthetizers/poor absorbers subgroup compared to poor synthetizers/good absorbers ($p<0.05$). Statin-untreated patients with increased cholesterol synthesis, regardless of absorption, had increased levels of TC and LDL-C ($p<0.01$, for both). The results suggest the existence of different lipoprotein abnormalities according to various patterns of cholesterol homeostasis. L/β ratio could be used for estimating individual propensity toward dyslipidemia development and direct the future treatment.

DISTRIBUCIJA AKTIVNOSTI PARAOKSONAZE 1 (PON1) U HDL LIPOPROTEINSKIM SUBFRAKCIJAMA KOD BUBREŽNIH BOLESNIKA

Milica Miljkovic[1], Aleksandra Stefanovic[1], Sanja Simic-Ogrizovic[2], Jelena Vekic[1], Natasa Bogavac-Stanojevic[1], Darko Cerne[3], Zorana Jelic-Ivanovic[1], Vesna Spasojevic-Kalimanovska[1], Jelena Kotur-Stevuljevic[1]

[1]Katedra za medicinsku biohemiju, Farmaceutski fakultet, Univerzitet u Beogradu
[2]Klinika za nefrologiju, Klinički centar Srbije, Beograd
[3]Katedra za kliničku biohemiju, Farmaceutski fakultet, Univerzitet u Ljubljani

Paraoksonaza (PON1) predstavlja jedan od najvažnijih antioksidativnih enzima na HDL česticama koji ima primarnu ulogu u zaštiti LDL od oksidacije. Brojne studije ukazuju na nisku aktivnost i koncentraciju PON1 kod pacijenata sa hroničnom bolešću bubrega (HBB). Ova studija je imala za cilj da poredi aktivnost PON1 između različitih HDL subklasa kod pacijenata sa HBB i kod pacijenata na hemodijalizi. Takođe, ova studije se bavila ispitivanjem uticaja hemodijalize na aktivnost PON1 u serumu, na distribuciju HDL subklasa i na promene u aktivnost PON1 na HDL_2 i HDL_3 subklasama. Rezultati su pokazali značajno viši odnos HDL_2/HDL_3 kod zdravih ispitanika u odnosu na pacijentena dijalizi ($p<0,05$). Merenje aktivnosti PON1 direktno na gelu pokazalo je višu aktivnost PON1 na HDL_2 subklasama kod zdravih ispitanika u poređenju sa pacijentima sa HBB ($p<0,05$) i pacijentimana dijalizi ($p<0,01$). S druge strane, kod bubrežnih bolesnika aktivnost PON1 je uglavnom bila koncentrisana na HDL_3 subklasama (HBB pacijentip $<0,05$; pacijenti na dijalizi $p<0,01$). Aktivnost PON1 u serumu ($p<0,01$) kao i aktivnost PON1 na HDL_{2a} česticama ($p<0,05$) značajno su bile više posle procesa dijalize. Promene u distribuciji HDL subklasa, mogu biti jedan od potencijalnih objašnjenja za smanjenu aktivnost PON1 kod bubrežnih bolesnika.

PON1 ACTIVITY DISTRIBUTION AMONG HDL SUBCLASSES IN RENAL PATHOLOGY

Milica Miljkovic[1], Aleksandra Stefanovic[1], Sanja Simic-Ogrizovic[2], Jelena Vekic[1], Natasa Bogavac-Stanojevic[1], Darko Cerne[3], Zorana Jelic-Ivanovic[1], Vesna Spasojevic-Kalimanovska[1], Jelena Kotur-Stevuljevic[1]

[1]Department of Medical Biochemistry, Faculty of Pharmacy, University of Belgrade, Serbia
[2]Clinic of Nephrology, Clinical Center of Serbia, Belgrade, Serbia
[3]Department of Clinical Biochemistry, Faculty of Pharmacy, University of Ljubljana, Slovenia

Paraoxonase 1 (PON1) is one of the most important antioxidative enzymes at HDL particles, which is primary involved in protection of LDL particles against oxidation. Numerous studies suggest low activity and concentration of PON1 in patients with chronic kidney disease (CKD).This study aimed to compare PON1 activity among HDL subclasses in patients with CKD as well as inpatients using hemodialysis procedure. Also this study tested influence of hemodialysis on serum PON1 activity, HDL subclasses distribution and PON1 activity between HDL_2 and HDL_3 particles. Results showed significantly higher HDL_2 /HDL_3 ratio in control subjects compared to dialysis patients ($p<0.05$). Measurement of PON1 activity directly on gel showed higher PON1 activity at HDL_2 subclasses in control subjects compared toCKD ($p<0.05$) and dialysis patients ($p<0.01$). On the other hand, in renal patients PON1 activity was mainly concentrated at HDL_3 subclasses (CKD patients $p<0.05$; dialysis patients $p<0.01$). Serum PON1 activity ($p<0.01$) and PON1 activity at HDL_{2a} particles ($p<0.05$) were significantly higher after hemodialysis procedure. Alternation in HDL distribution could be one of possible explanations for decreased PON1 activity in renal patients.

NEALKOHOLNA MASNA JETRA I KVALITATIVNE I KVANTITATIVNE KARAKTERISTIKE LIPOPROTEINA VISOKE GUSTINE

Jelena Janać[1], Aleksandra Zeljković[1], Zorana Jelić-Ivanović[1], Vesna Dimitrijević-Srecković[2], Milica Miljković[1], Aleksandra Stefanović[1], Jelena Munjas[1], Jelena Vekić[1], Jelena Kotur-Stevuljević[1], Vesna Kalimanovska-Spasojević[1]

[1]Katedra za medicinsku biohemiju, Farmaceutski fakultet, Univerzitet u Beogradu, Beograd, Republika Srbija
[2]Klinika za endokrinologiju, dijabetes i bolesti metabolizma, Klinički centar Srbije, Medicinski fakultet, Univerzitet u Beogradu, Beograd, Republika Srbija

Nealkoholna masna jetra (NAFLD) je učestalo oboljenje koje nosi rizik od kardiovaskularnih komplikacija. Ova studija je imala za cilj procenu promena metabolizma i funkcije lipoproteina visoke gustine (HDL) koji su u vezi sa NAFLD. Učesnici ove studije su bile osobe bez istorije aterosklerotskih kardiovaskularnih bolesti, dijabetes melitusa i hepatitisa B, koje nisu na medikamentnoj terapiji lipidskih poremećaja. Klasifikacija je izvršena na osnovu vrednosti indeksa masne jetre (FLI): FLI<30 grupa bez NAFLD (N=38), FLI≥ 60 grupa sa NAFLD (N=49) i 30≤FLI <60 je definisana kao intermedijarna grupa (N=43). Pored osnovnih biohemijskih analiza, određene su aktivnosti lecitin-holesterol-aciltransferaze (LCAT) i holesterol-estar-transfernog proteina (CETP) i razdvojene su HDL subfrakcije. LCAT aktivnost je određena merenjem brzine smanjenja koncentracije supstrata, slobodnog holesterola, tokom inkubacije plazme. Transfer estara holesterola koji posreduje CETP je određen kao razlika između brzine nastajanja holesterol estara, što je posredovano aktivnošću LCAT, i brzine promene koncentracije holesterolestara u HDL sloju tokom inkubacije plazme. HDL subfrakcije su razdvojene metodom vertikalne nedenaturišuće gradijent gel elektroforeze. Pored niže koncentracije HDL-holesterola u FLI≥60 grupi u poređenju sa preostale dvegrupe, primećen je trend snižavanja koncentracije HDL-holesterola paralelno sa povećanjem FLI. Aktivnost LCAT se povećavala u svakoj uzastopnoj grupi sa porastom vrednosti FLI ($p<0.001$). Nije dokazana razlika u aktivnosti CETP između grupa. Relativni udeo najveće HDL2b subfrakcije je bio niži u FLI≥60 u poređenju sa FLI<30 grupom ($p<0,001$), dok je udeo najmanje HDL3c subfrakcije bio veći u FLI≥60 grupi u poređenju sa preostale dve grupe ($p<0,001$). Rezultati studije su pokazali povezanost NAFLD i kvalitativnih i kvantitativnih promena HDL čestica. Remodelovanje HDL čestica je pokazano kroz redistribuciju ka manjim HDL subfrakcijama, uz povećanu aktivnost LCAT.

NON-ALCOHOLIC FATTY LIVER DISEASE AND QUALITATIVE AND QUANTITATIVE CHARACTERISTICS OF HIGH-DENSITY LIPOPROTEINS

Jelena Janać[1], Aleksandra Zeljković[1], Zorana Jelić-Ivanović[1], Vesna Dimitrijević-Srecković[2], Milica Miljković[1], Aleksandra Stefanović[1], Jelena Munjas[1], Jelena Vekić[1], Jelena Kotur-Stevuljević[1], Vesna Kalimanovska-Spasojević[1]

[1]Department of Medical Biochemistry, Faculty of Pharmacy, University of Belgrade, Belgrade, Republic of Serbia
[2]Clinic for Endocrinology, Diabetes and Metabolic Diseases, Clinical Center of Serbia, Faculty of Medicine, University of Belgrade, Belgrade, Republic of Serbia

Non-alcoholic fatty liver disease (NAFLD) is a frequent liver disorder carrying high risk of cardiovascular complications. This study goal was to evaluate alterations in high-density lipoprotein (HDL) metabolism and function related to the NAFLD. We included subjects without a history of the atherosclerotic cardiovascular disease, diabetes mellitus, current intake of lipid modulating drugs and hepatitis B infection. Patients were classified according to the fatty liver index (FLI): FLI<30 without NAFLD (N=38), FLI≥60 with NAFLD (N=49), and 30≤FLI<60 was an intermediate group (N=43). In addition to basic biochemical analyses, we assessed lecithin-cholesterol acyltransferase (LCAT) and cholesteryl ester transfer protein (CETP) activity, and separated HDL subclasses. LCAT activity was determined by measuring the decrease of the concentration of substrate, free plasma cholesterol, during incubation of the whole plasma. CETP mediated cholesteryl ester transfer was assessed as the difference between the rate of cholesteryl esters formation by LCAT activity and the rate of cholesteryl esters exchange in HDL layer during incubation of plasma. HDL subclasses were separated using vertical no denaturing gradient gel electrophoresis. HDL-cholesterol concentration was lower in the FLI≥60 compared to the other two groups ($p<0.001$), but we also observed a trend of decreasing HDL levels in parallel with an increase of FLI. With each successive group with increasing FLI values, there was an increase in the LCAT activity ($p<0.001$), without a significant difference in the CETP activity. The relative proportion of the largest HDL 2b was lower in FLI≥60 compared to the FLI< 30 group ($p<0.001$), whereas the proportion of the smallest HDL3c subclass was higher in the FLI≥60 group compared to other two groups ($p<0.001$).This study results demonstrated a relationship between NAFLD and qualitative and quantitative HDL changes. HDL particles remodeling was verified as a shift toward smaller HDL subclasses, together with increased LCAT activity.

KVALITATIVNA ANALIZA LDL I HDL HOLESTEROLA U PROCENI RIZIKA ZA RAZVOJ KARCINOMA KOLONA I REKTUMA

Milica Stevanović[1], Jelena Vekić[1], Nataša Bogavac-Stanojević[1], Jelena Janać[1], Željka Stjepanović[2], Dejan Zeljković[3], Bratislav Trifunović[3], Vesna Spasojević-Kalimanovska[1], Aleksandra Zeljković[1]

[1]Univerzitet u Beogradu – Farmaceutski fakultet, Beograd, Srbija
[2]Medigroup opšta bolnica, Beograd, Srbija
[3]Klinika za opštu hirurgiju, Vojnomedicinska akademija, Beograd, Srbija

Prethodne studije koje su bile zasnovane uglavnom na kvantitativnom ispitivanju lipidnih parametara, dale su kontradiktorne rezultate o povezanosti izmenjenog lipidnog statusa i nastanka kolorektalnog karcinoma (CRC). Kako bismo dobili precizniji uvid u ovu vezu, odredili smo dominantne dijametre lipoproteinskih čestica niske gustine (LDL) ili poproteinskih čestica visoke gustine (HDL), i utvrdili smo raspodelu LDL i HDL subfrakcija kod pacijenata sa CRC. Ova studija je obuhvatila 84 pacijenta sa CRC i 92 zdrava ispitanika. Razdvajanje lipoproteinskih subfrakcija je vršeno metodom vertikalne gradijent gel elektroforeze, dok su lipidni parametri određeni upotrebom rutinskih enzimskih metoda. Nivo ukupnog holesterola, LDL i HDL holesterola je bio značajno niži kod pacijenata (P<0,001). Razdvajanjem subfrakcija, utvrđeno je da pacijenti imaju značajno manje dominantne dijametre LDL i HDL čestica (P<0,001), kao i da je raspodela subfrakcija pomerena u korist manjih čestica, odnosno da pacijenti imaju veći udeo manjih, gušćih LDL čestica (LDL IIB, P=0,002; LDL IIIA, P<0,001; LDL IIIB, P=0,001; LDL IVA, P=0,027), veći udeo manjih, gušćih HDL 3b (P<0,001), i manji udeo većih HDL 2b čestica (P=0,013). Manji dominantni dijametri LDL i HDL čestica su prepoznati kao nezavistan prediktor CRC (OR=0,489, 95%CI: 0,315–0,757, P=0,001 i OR=0,487, 95%CI: 0,286–0,828, P=0,008, respektivno). Ova studija je pokazala da manji dominantni dijametri LDL i HDL čestica zajedno sa koncentracijom HDL holesterola i godinama čine optimalno ekonomski isplativ model koji bi mogao da se koristi u rutinskoj kliničkoj praksi za procenu rizika za razvoj CRC i za tačnu klasifikaciju pacijenata sa CRC.

THE QUALITATIVE ANALYSIS OF LDL AND HDL CHOLESTEROL IN CRC DEVELOPMENT RISK ASSESSMENT

Milica Stevanović[1], Jelena Vekić[1], Nataša Bogavac-Stanojević[1], Jelena Janać[1], Željka Stjepanović[2], Dejan Zeljković[3], Bratislav Trifunović[3], Vesna Spasojević-Kalimanovska[1], Aleksandra Zeljković[1]

[1]University of Belgrade, Faculty of Pharmacy, Belgrade, Serbia
[2]Medigroup General Hospital, Belgrade, Serbia
[3]Clinic for General Surgery, Military Medical Academy, Belgrade, Serbia

Previous studies based mainly on the quantitative analysis of the blood lipid parameters gave inconsistent results on their relationship with the onset of colorectal cancer (CRC). In order to provide a more precise insight into the subject, we determined dominant low-density lipoprotein (LDL) and high-density lipoprotein (HDL) cholesterol particle diameters, and the distribution of LDL and HDL subclasses in CRC patients. This study included 84 patients with CRC and 92 healthy subjects. Separation of lipoprotein subclasses was performed by vertical gradient gel electrophoresis, while lipid parameters were measured using routine enzymatic methods. The levels of total cholesterol, LDL and HDLcholesterol were significantly lower in patients (P<0.001). By separating all subclasses, it was revealed that patients had significantly smaller dominant LDL and HDL particle diameters (P<0.001), and that the subclasses distribution was shifted towards smaller particles,i.e., patients expressed higher proportion of smaller, denser LDL particles (LDL IIB, P=0.002; LDL IIIA, P<0.001; LDL IIIB, P=0.001; LDL IVA, P=0.027), higher proportion of smaller, denser HDL 3b (P<0.001) and lower proportion of larger HDL 2b particles (P=0.013). Smaller dominant LDL and HDL particle diameters were recognized as independent predictors of CRC (OR=0.489, 95%CI: 0.315–0.757, P=0.001 and OR=0.487, 95%CI: 0.286–0.828, P=0.008, respectively). This study demonstrated that smaller dominant LDL and HDL particle diameters, alongside with HDL cholesterol level and age form the optimal cost-effective model that could be used in routine clinical practice for assessing the risk of CRC development and for accurate classification of CRC patients.