

Nedostatci sadašnjeg snizivanja LDL kolesterola i uloga inhibitora PCSK9.

Matijević Rončević, Sanja; Gobić, David; Zaninović Jurjević, Teodora; Rubeša Miculinić, Željka; Zaputović, Luka

Source / Izvornik: **Cardiologia Croatica, 2016, 11, 330 - 342**

Journal article, Published version

Rad u časopisu, Objavljena verzija rada (izdavačev PDF)

<https://doi.org/10.15836/ccar.2016.330>

Permanent link / Trajna poveznica: <https://urn.nsk.hr/urn:nbn:hr:184:146775>

Rights / Prava: [In copyright / Zaštićeno autorskim pravom.](#)

Download date / Datum preuzimanja: **2024-05-17**



Repository / Repozitorij:

[Repository of the University of Rijeka, Faculty of Medicine - FMRI Repository](#)



Nedostatci sadašnjeg snizivanja LDL kolesterola i uloga inhibitora PCSK9

Disadvantages of Current LDL-cholesterol Lowering and the Role of PCSK9 Inhibitors

Sanja Matijević Rončević ,
David Gobić ,
Teodora Zaninović Jurjević ,
Željka Rubeša Miculinić ,
Luka Zaputović* 

Medicinski fakultet Sveučilišta u Rijeci, Klinički bolnički centar Rijeka, Rijeka, Hrvatska
 University of Rijeka School of Medicine, University Hospital Centre Rijeka, Rijeka, Croatia

RECEIVED:
 July 4, 2016

ACCEPTED:
 July 12, 2016



SAŽETAK: LDL kolesterol (LDL-K) snažan je nezavisni čimbenik kardiovaskularnog (KV) rizika na koji je moguće utjecati. Statini su danas terapija izbora u postizanju ciljnih vrijednosti LDL-K-a. Iako su u kontroliranim kliničkim ispitivanjima dokazano učinkoviti i sigurni, u praksi se često susrećemo s nepodnošenjem statina, a u značajnog dijela bolesnika i nepostizanjem ciljnih vrijednosti LDL-K-a unatoč maksimalnim dozama. U bolesnika s visokim i vrlo visokim KV rizikom i preostala eulipemijska farmakoterapija često nije dosta. Inhibitori PCSK9 (PCSK9-I) novi su, revolucionarni lijekovi s potentnim učinkom na LDL-K. Brzi razvoj PCSK9-I-a započeo je 2003. godine otkrićem mutacije gena PCSK9 u bolesnika s porodičnom hipercolesterolemijom. Produkt tog gena, enzim proprotein konvertaza subtilizin/keksin tip 9 (PCSK9) ima važnu ulogu u regulaciji ekspresije LDL receptora i u metabolizmu kolesterola. Istraživanjima na životinjama dokazano je da inaktivacija PCSK9 gena snizuje LDL-K-s regresijom aterosklerotskih promjena aorte. Heterozigoti i homozigoti s inaktivirajućom mutacijom gena PCSK9 imaju niske vrijednosti LDL-K-a u manju pojavnost ateroskleroze. Među različitim skupinama PCSK9-I-a, snažan razvoj doživjela su monoklonska protutijela (alirokumab, evolokumab i bococizumab). Klinička ispitivanja treće faze porodične i primarne hipercolesterolemije sa statinskom intolerancijom ili rezistenčnom pokazala su snažan povoljan učinak alirokumaba i evolokumaba na LDL-K (sniženje od 60 %), uz visoku sigurnost i dobru podnošljivost. OSLER studija s evolokumabom dokazala je i povoljne učinke na KV ishode. U tijeku je više kliničkih pokusa različitih PCSK9-I-a, u kojima se prati njihov učinak na KV pobol i smrtnost. Pozitivni rezultati tih studija potvrdili bi veliki potencijal PCSK9-I-a u boljoj prevenciji i liječenju KV bolesti.

SUMMARY: LDL cholesterol (LDL-C) is a strong independent cardiovascular (CV) risk factor that can be easily influenced. Today, statins are the therapy of choice for the achievement of target LDL-C values. Although controlled clinical trials have demonstrated their effectiveness and safety, in practice we are often met with statin intolerance as well as a failure to achieve target LDL-C values in a significant portion of the patients despite maximal doses. In patients with high and very high CV risk, other antilipemic pharmacotherapy is often also insufficient. PCSK9 inhibitors (PCSK9-I) are new revolutionary drugs with a potent effect on LDL-C. The rapid development of PCSK9-I began in 2003 with the discovery of a PCSK9 gene mutation in patients with familial hypercholesterolemia. The product of this gene, proprotein convertase subtilisin/kexin type 9 (PCSK9), has an important role in the expression of LDL receptors and cholesterol metabolism. Animal models demonstrated that inactivation of the PCSK9 gene lowers LDL-C with regression of atherosclerotic changes in the aorta. Heterozygotes and homozygotes with the inactivation mutation of PCSK9 have lower LDL-C values and lower incidence of atherosclerosis. Among the various groups of PCSK9-I, monoclonal antibodies saw strong development (alirocumab, evolocumab, bococizumab). Phase 3 clinical trials on familial and primary hypercholesterolemia with statin intolerance or resistance have demonstrated a strong positive effect of alirocumab and evolocumab on LDL-C values (a reduction of 60 %), with high safety and good tolerability. The OSLER study on evolocumab also demonstrated positive effects on CV outcomes. Multiple clinical trials on PCSK9-I are currently monitoring their effect on CV morbidity and mortality. Positive results from these studies would confirm the great potential of PCSK9-I for better prevention and treatment of CV diseases.

KLJUČNE RIJEĆI: kardiovaskularne bolesti, dislipidemije, LDL kolesterol, statini, PCSK9 inhibitori.

KEYWORDS: cardiovascular diseases, dyslipidemia, LDL-cholesterol, statins, PCSK9 inhibitors.

CITATION: Cardiol Croat. 2016;11(8):330–342. | DOI: <http://dx.doi.org/10.15836/ccar2016.330>

***ADDRESS FOR CORRESPONDENCE:** Luka Zaputović, Klinički bolnički centar Rijeka, Krešimirova 42, HR-51000 Rijeka, Croatia. / Phone: +385 51 407 149 / E-mail: luka.zaputovic@medri.uniri.hr

ORCID: Sanja Matijević Rončević, <http://orcid.org/0000-0003-0627-2114> • David Gobić, <http://orcid.org/0000-0001-9406-1127>
 Teodora Zaninović Jurjević, <http://orcid.org/0000-0001-8359-3910> • Željka Rubeša Miculinić, <http://orcid.org/0000-0002-1880-1493>
 Luka Zaputović, <http://orcid.org/0000-0001-9415-9618>

Uvod

LDL kolesterol (LDL-K) snažan je nezavisni čimbenik kardiovaskularnog (KV) rizika na koji je moguće djelovati. Postizanje ciljnih vrijednosti LDL-K-a jedan je od temeljnih zahtjeva primarne i sekundarne prevencije KV bolesti^{1,2}. Kliničkim je istraživanjima dokazano da smanjenje LDL-K-a za 1 mmol/L smanjuje rizik od neželjenih KV ishoda za 22%^{3,4}. Prema suvremenim smjernicama Europskoga kardiološkog društva, ciljne vrijednosti LDL-K-a određene su ukupnim KV rizikom te u osoba s vrlo visokim rizikom iznose <1,8 mmol/L, a u osoba s visokim rizikom <2,6 mmol/L^{5,6}. Posebno su rizična populacija bolesnici s nasljednom hipercolesterolemijom, u kojih se preuranjena KV bolest pojavljuje već u dvadesetim godinama života u homozigota te u četrdesetim godinama u heterozigota⁷⁻⁹. Terapija izbora u liječenju povišenog LDL-K-a dobro su poznati statini. Upravo su statini u brojnim kliničkim studijama i metaanalizama dokazali čvrstu povezanost između povišenog LDL-K-a i neželjenih KV događaja¹⁰. Iako su bili učinkoviti i sigurni u randomiziranim kontroliranim kliničkim ispitivanjima (1 – 5 % ukupnih mišićnih nuspojava), u svakodnevnoj kliničkoj praksi često se susrećemo s problemom nepodnošenja statina i terapijske rezistencije na statine¹¹.

Statinska intolerancija

Definicija statinske intolerancije, odnosno nepodnošenja statina, još uvijek nije usuglašena. Nekoliko je suvremenih definicija (EMA, NLA, Canadian Working Group Consensus), a zajedničko im je nepodnošenje statina uopće ili nepodnošenje punе terapijske doze statina zbog pojave različitih nuspojava¹²⁻¹⁴. Najčešće su mišićne nuspojave, od asimptomatskog povišenja vrijednosti kreatin kinaze (CK) do mijalgija, miopatija, miozitsa i rabdomiolize, kao najteže i vrlo rijetke mišićne nuspojave¹⁵. Uzroci statinske intolerancije nisu potpuno poznati, no zna se da postoji individualna, genetski uvjetovana podložnost bolesnika, ovisna o polimorfizmu gena i proteina uključenih u metabolizam ili transport statina u hepatocite (CYP 450, OATP)^{16,17}. Dobro je poznato i da podnošljivost statina ovisi o dozi statina i istodobno primjeni lijekova koji utječu na njihov metabolizam (povisujući razinu statina u plazmi – gemfibrozil, itrakonazol)¹⁸⁻²⁰ te da se kod nekih statina (pravastatina, fluvastatina i pitavastatina) zbog različitih putova metabolizma, mišićne nuspojave pojavljuju rijede^{21,22}. Nedavna istraživanja upućuju na mogućnost postizanja tolerancije statina u većine bolesnika koji ih ne podnose smanjenjem doze i intermitentnom primjenom lijeka^{23,24}. Niže doze statina u liječenju bolesnika s poznatom KV bolesti rijetko će dovesti do postizanja ciljnih vrijednosti LDL-K-a. Nerazmjer između učestalosti mišićnih nuspojava u kliničkim i postkliničkim ispitivanjima rezultat je strogih uključnih kriterija. Bolesnici s poznatim mišićnim bolestima, anamnezom mijalgija, povišenom vrijednosti CK i oni koji su uzimali lijekove s utjecajem na metabolizam statina nisu bili uključeni u klinička ispitivanja. Nadalje, većina randomiziranih statinskih studija nije detaljno procjenjivala blaže mišićne nuspojave. Naglasak je bio postavljen na rabdomiolizu, koja je, kao što je spomenuto, vrlo rijetka.

Druge po učestalosti jesu rijetke jetrene nuspojave koje se pojavljuju u do 3 % bolesnika na visokim dozama statina te u

Introduction

LDL cholesterol (LDL-C) is a strong independent cardiovascular (CV) risk factor that can be influenced. Achieving target LDL-C values is one of the fundamental goals of primary and secondary CV disease prevention^{1,2}. Clinical trials have demonstrated that a reduction in LDL-C values of 1 mmol/L reduces the risk of unwanted CV outcomes by 22%^{3,4}. According to current European Society of Cardiology guidelines, target LDL-C values are determined by the total CV risk, and are <1.8 mmol/L in persons with very high risk and <2.6 mmol/L for persons with high risk^{5,6}. Patients with hereditary hypercholesterolemia are a population with especially high risk, since the diseases manifests already in their twenties in homozygous patients and in the forties in heterozygous patients⁷⁻⁹. The treatment of choice for elevated LDL-C are the well-known statins. It was statins that demonstrated a strong association between elevated LDL-C and unwanted CV events in numerous clinical trials and meta-analyses¹⁰. Although they were shown to be safe and effective in randomized controlled clinical trials (1-5% total muscle side effects), in everyday clinical practice we are often faced with the issue of statin intolerance and resistance to statin treatment¹¹.

Statin intolerance

The definition of statin intolerance has still not been agreed upon. There are several modern definitions (EMA, NLA, Canadian Working Group Consensus), all of which include general statin intolerance or intolerance to full treatment doses due to the appearance of various side effects¹²⁻¹⁴. Most common are muscle side effects, ranging from asymptomatic elevation of creatine kinase (CK) levels to myalgia, myopathy, myositis, and rhabdomyolysis as the most severe and very rare muscle side effect¹⁵. The causes of statin intolerance are not fully known, but it has been shown that patients have individual, genetically determined susceptibility to statins, dependent on gene polymorphism and proteins involved in the metabolism or transport of statins to hepatocytes (CYP 450, OATP)^{16,17}. It is also well known that statin tolerance depends on the doses and the simultaneous application of drugs that affect statin metabolism (elevating plasma statin levels – gemfibrozil, itraconazole)¹⁸⁻²⁰ and that muscle side effects are more rare for some statins (pravastatin, fluvastatin, and pitavastatin) due to different metabolic pathways^{21,22}. Recent studies indicate the possibility of achieving statin tolerance in most patients with statin intolerance through dose reduction and intermittent drug application^{23,24}. Lower statin doses in the treatment of patients with an already diagnosed CV disease will rarely achieve target LDL-C values. The discrepancy in the incidence of muscle side effects in clinical and post-clinical trials is the result of strict inclusion criteria. Patients with already diagnosed muscle diseases, history of myalgia, elevated CK values, and those taking drugs that affect statin metabolism were not included in the clinical trials. Furthermore, most randomized statin studies did not include a detailed assessment of milder muscle side effects. The focus was on rhabdomyolysis, which is, as already mentioned, very rare.

manje od 1 % bolesnika na srednjim i niskim dozama²⁵. Utjecajem statina na jetru smatra se porast aminotransferaza od tri ili više puta iznad gornje granice normale, nakon započinjanja statinske terapije. Ovakva je pojava uglavnom prolazna²⁶. Teška oštećenja jetre statinima rijetka su i nepredvidiva. Novija su istraživanja dokazala da nealkoholna masna bolest jetre (nealkoholni steatohepatitis) nije kontraindikacija za liječenje statinima^{27,28}. Naprotiv, statini su u dijelu bolesnika s blagim do umjerenim porastom jetrenih enzima doveli do sniženja aminotransferaza.

Rijetka je nuspojava na statine novonastala šećerna bolest tipa 2, osobito u osoba s prethodno oštećenom tolerancijom glukoze. Ipak, smanjenje ukupnog KV rizika postignuto je i u takvih bolesnika²⁹.

Statinska rezistencija

Statinska je rezistencija nemogućnost postizanja ciljnih vrijednosti LDL-K-a unatoč liječenju maksimalnom terapijskom dozom statina. Statinskom se rezistencijom može smatrati i nemogućnost prevencije aterosklerotskih promjena i smanjenja neželjenih KV događaja. Treba je razlikovati od mnogo češće pseudorezistencije, koja je posljedica bolesnikova nepridržavanja uputa za liječenje. Nepostizanje ciljnih vrijednosti LDL-K-a često je posljedica neopravdanog propisivanja nedovoljnih doza ili manje učinkovitih statina³⁰. Statinska rezistencija, kao i intolerancija, najvećim je dijelom uvjetovana genetskim čimbenicima. Do danas su istraživani brojni geni čiji polimorfizam izaziva različit individualni odgovor na statine te utjecaj na farmakokinetiku i farmakodinamiku lijeka³¹. Osim genetskih, dokazani su i stečeni čimbenici koji utječu na različit individualni odgovor na statine. Pušači i bolesnici s arterijskom hipertenzijom imaju slabiji odgovor od nepušača i osoba s normalnim arterijskim tlakom³². Upalni citokin IL-1 beta utječe na povratnu regulaciju LDL receptora (LDL-R) smanjujući njegovu ekspresiju pa je u uvjetima upale potrebna veća doza statina za postizanje jednakog učinka na LDL-K. Smanjeni ekspresiji LDL-R-a uzrokuju i hipotireoza te liječenje amiodaronom, neovisno o tireoidnom statusu bolesnika³³.

Drugi hipolipemici s povoljnim učinkom na LDL kolesterol

Za postizanje ciljnog LDL-K-a u bolesnika rezistentnih na statine ili u onih koji statine ne podnose, od ostalih su hipolipemika dostupni ezetimib, sekvestranti žučnih kiselina, fibrati i niacin. Ezetimib je selektivni inhibitor apsorpcije kolesterolja u sluznici tankoga crijeva. U usporedbi sa statinima ostvaruje mnogo skromniji učinak na LDL-K. U kliničkim istraživanjima u bolesnika s porodičnom hiperkolesterolemijom, ezetimib je kao monoterapija snizivao LDL-K za skromnih 7–11 %. U kombinaciji sa statinima ciljni LDL-K < 3,0 mmol/L postigao je mnogo veći broj ispitanika rezistentnih na statine (18 % na kombiniranoj terapiji u odnosu prema 5 % na statinima). Unatoč kombiniranoj terapiji ezetimiba sa statinom, najveći je broj ispitanika imao LDL-K > 3,0 mmol/L³⁴. Kombinacijom ezetimiba sa sekvestrantima žučnih kiselina postižu se bolji učinci na LDL-K, bez znatnog porasta nuspojava u usporedbi s

The second most common side effects are rare liver side effects that manifest in 3% of patients receiving high statin doses and in less than 1% of patients taking moderate or low doses²⁵. Statins are considered to be affecting the liver when aminotransferase levels increase by three or more times above the upper limit of normal levels after commencement of statin treatment. This is usually a transient effect²⁶. Severe liver damage caused by statins is rare and unpredictable. Newer studies have demonstrated that non-alcoholic fatty liver disease (non-alcoholic steatohepatitis) is not a contraindication for statin treatment^{27,28}. On the contrary, statins led to aminotransferase reduction in a portion of these patients with mild to moderate elevation of liver enzymes.

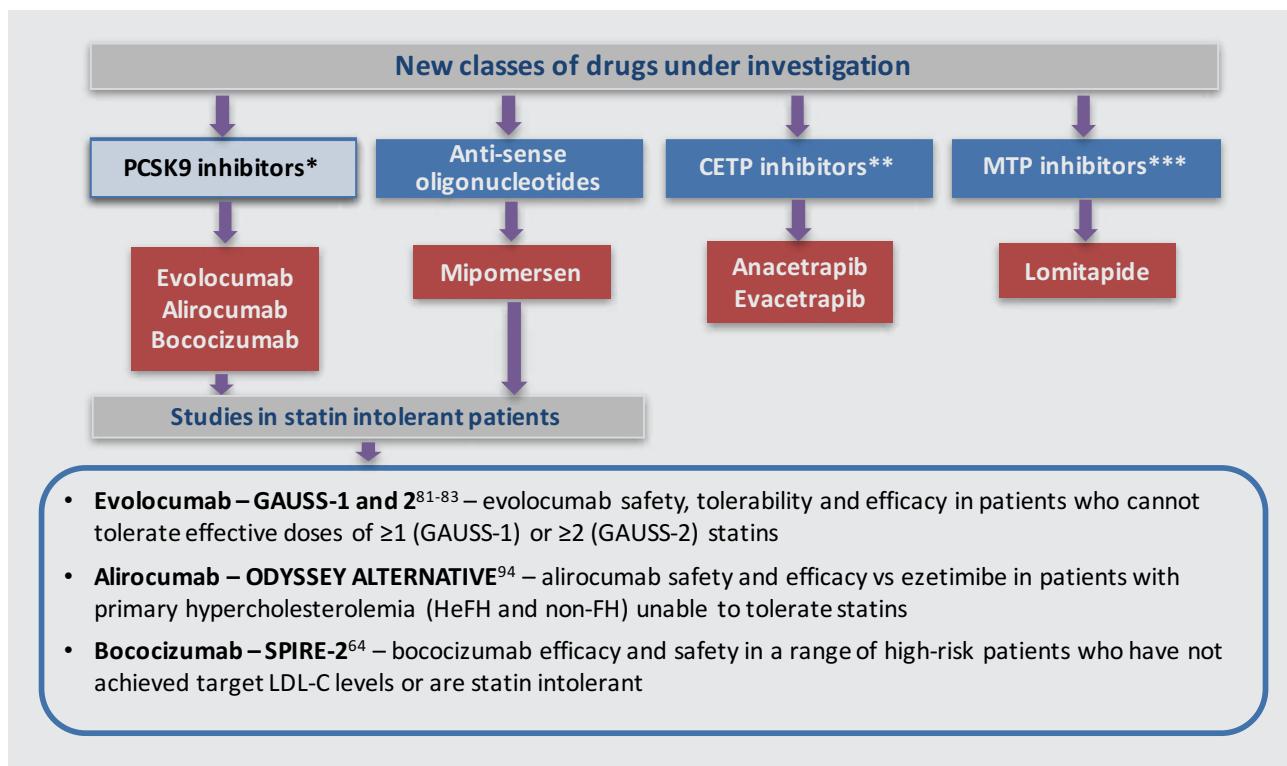
A rare statin side effect is newly developed diabetes type 2, especially in persons with previously impaired glucose tolerance. However, a reduction in total CV risk was achieved in these patients as well²⁹.

Statin resistance

Statin resistance is the inability to achieve target LDL-C values despite treatment with the maximum statin therapeutic dose. The inability to prevent atherosclerotic changes and reduce unwanted CV events using statins can also be considered statin resistance. This should be differentiated from the much more common pseudo-resistance, which is the consequence of the patient's lack of treatment compliance. Not achieving target LDL-C values is often a result of unjustified prescription of insufficient doses or less effective statins³⁰. Like statin intolerance, statin resistance is mostly determined by genetic factors. Research has shown that the polymorphism of numerous genes causes different individual responses to statins and affects the pharmacokinetics and pharmacodynamics of the medication³¹. In addition to genetic factors, other acquired factors have been shown to affect individual response to statins. Smokers and patients with arterial hypertension have weaker response than non-smokers and persons with normal arterial pressure³². The inflammatory cytokine IL-1 beta affects feedback regulation of LDL receptors (LDL-R), reducing its expression, so a larger statin dose is required under inflammatory conditions to achieve the same effect on LDL-C values. Reduced LDL-R expression is also caused by hypothyroidism and treatment with amiodarone, regardless of the thyroid status of the patient³³.

Other hypolipidemics with positive effects on LDL-cholesterol

Other hypolipidemics available for achieving target LDL-C values in patients with statin resistance or intolerance are ezetimibe, bile acid sequestrants, fibrates, and niacin. Ezetimibe is a selective inhibitor of cholesterol absorption in the small intestine mucosa. In comparison with statins, it has a significantly weaker effect on LDL-C. In clinical trials on patients with familial hypercholesterolemia, ezetimibe monotherapy reduced LDL-C by a humble 7–11%. In combination with statins, the target LDL-C reduction of <3.0 mmol/L was achieved by a significantly larger number of participants with statin resistance (18% with combined therapy in comparison with 5% on statins). Despite

**FIGURE 1. New pharmacological options for the treatment of dyslipidaemias.**

*PCSK9: proprotein convertase subtilisin/kexin type 9, **CETP: cholestrylo ester transfer protein, ***MTP: microsomal triglyceride transfer protein

monoterapijom sekvestrantima žučnih kiselina³⁵. Ograničenje te kombinacije česte su nuspojave na sekvestrante žučnih kiselina (konstipacija, proljev, nadutost, porast serumskih triglicerida) i višekratno doziranje lijeka, što smanjuje suradljivost bolesnika i izaziva česte prekide liječenja³⁰.

Fibrati i niacin veći učinak ostvaruju na tzv. ne-HDL kolesterol, dok je njihov učinak na LDL-K mnogo slabiji. Fibrati snizuju LDL-K za 13 – 35 %, a profil nuspojava sličan je onima na statine (prije svega miopatija) te stoga nisu povoljni za liječenje bolesnika koji statine ne podnose^{36,37}. U kombinaciji sa statinima u bolesnika rezistentnih na statine znatno dodatno snizuju LDL-K, no mnogi bolesnici i dalje ne postižu ciljne vrijednosti^{5,38}.

Niacin je dobro poznat lijek s povoljnim eulipemijskim učinkom, ali sa slabom podnošljivošću. U svrhu smanjivanja nuspojava istraživani su novi oblici lijeka - niacin s produženim otpuštanjem (ER-niacin) i kombinacija niacina s laropiprantom (studije AIM-HIGH i HPS2-THRIVE)^{39,40}. Iako je dokazano smanjenje LDL-K-a, nije bilo i manje neželjenih KV događaja, a, uz prije poznate, pojavile su se i nove ozbiljne nuspojave. Zbog toga je EMA (European Medicines Agency) nedavno preporučila povlačenje tih lijekova iz Europe.

Nove lijekove s povoljnijim učinkom na LDL-K, mipomersen i lomitapide, nedavno je odobrila američka FDA (Food and Drug Administration) u liječenju homozigotnog oblika nasljedne hipokolesterolijke (**slika 1**). Mipomersen je specifični oligonukleotid koji se veže za ApoB glasničku RNA i inhibira njezinu translaciju, odnosno sintezu ApoB (primarnoga strukturnog apoproteina LDL-K-a i drugih aterogenih lipoproteina) i, posljedično tomu, VLDL čestica, što konačno rezultira nižim LDL-K-

combined ezetimibe and statin treatment, the majority of the patients had LDL-C >3.0 mmol/L³⁴. Combining ezetimibe with bile acid sequestrants achieved better effects on LDL-C without a significant increase in side effects in comparison with bile acid sequestrants monotherapy³⁵. The limitations of this combined treatment are the high incidence of side effects of bile acid sequestrants (constipation, diarrhea, bloating, high serum triglycerides) and repeated drug administration, which reduces patient cooperation and causes frequent treatment interruptions³⁰.

Fibrates and niacin have a larger effect on so-called non-HDL cholesterol, while their effect on LDL-C is much weaker. Fibrates reduced LDL-C by 13-35%, and the side effect profile is similar to that of statins (primarily myopathy), making them unsuitable for the treatment of patients with statin intolerance^{36,37}. In combined treatment with statins in patients with statin resistance, they provide a significant added reduction in LDL-C, but many patients still do not reach target values^{5,38}.

Niacin is a well-known drug with beneficial antilipemic effects but poor tolerance. To reduce side effects, new forms of the drug have been studied – niacin extended release (ER-niacin) and the combination of niacin with laropiprant (the AIM-HIGH and HPS 2-THRIVE studies)^{39,40}. Although a reduction in LDL-C was demonstrated, there was no reduction in unwanted CV events, and new serious side effects were present along with those already known. Thus, the European Medicines Agency (EMA) recently recommended withdrawing these drugs from the European market.

New drugs with beneficial effects on LDL-C, mipomersen and lomitapide, have been recently approved by the American Food

om. Lomitapide je oralni inhibitor mikrosomnog transportnog proteina za trigliceride (MTP-a), važne karike u produkciji VLDL-a u jetri⁴¹. Iako su u kliničkim ispitivanjima doveli do znatnog sniženja LDL-K-a, njihov učinak na KV ishode još nije ispitivan. Zbog učestalih nuspojava (pojačano nakupljanje masti u jetri, ozbiljne kožne reakcije na mjestu uboda kod mipomersena), malo je vjerojatno da će ovi lijekovi ikada biti odobreni u liječenju bolesnika sa statinskom intolerancijom i rezistencijom^{42,43}.

Protein konvertaza subtilizin/keksin tip 9 (PCSK9)

Inhibitori PCSK9 (engl. *proprotein convertase subtilisin-kexin type 9 inhibitors*) novi su, revolucionarni lijekovi u liječenju povišenih vrijednosti LDL-K-a. Razvoju terapijske PCSK9 inhibicije prethodilo je otkriće mutacije gena PCSK9 u bolesnika s dominantnim oblikom nasljedne hiperkolesterolemije 2003. godine⁴⁴. Dokazano je da u 10 – 25 % slučajeva heterozigotnog oblika nasljedne hiperkolesterolemije postoji mutacija gena PCSK9 koja rezultira pojačanom aktivnosti gena te povišenom koncentracijom PCSK9 u jetri i u perifernoj krvi^{7,45}.

PCSK9 je enzim iz grupe proteinazi K sličnih proteina, koji pripada porodici sekretnih subtilizina. Primarno se sintetizira u hepatocitima i secernira u jetri, gdje postiže i najviše koncentracije^{46,47}.

Ključna uloga PCSK9 jest regulacija ekspresije LDL-R-a u jetri. Vezanjem N-terminalnog kraja PCSK9 molekule za LDL-R dolazi do njegove internalizacije u stanicu i razgradnje putem lisosoma (**slika 2**). Rezultat su smanjena količina staničnih LDL-R-a, manji klirens i povišena plazmatska koncentracija LDL-K-a⁴⁸. Ekstrahepatični učinci PCSK9 uključuju pojačanu sekreciju hilomikrona i regulaciju apsorpcije kolesterola u enterocitima⁴⁷. Eksperimentalne studije na životinjama dokazale su snažan utjecaj PCSK9 na progresiju ili regresiju aterosklerotskih promjena⁴⁹. Rezultati tih studija sugeriraju da PCSK9 ne utječe samo na metabolizam kolesterola nego je uključen i u metabolizam glukoze, regeneraciju hepatocita i podložnost infekciji virusom hepatitis C⁵⁰⁻⁵³.

Suprotno pojačanoj aktivnosti PCSK9 gena u bolesnika s porodičnom hiperkolesterolemijom, kliničkim je studijama dokazano da mutaciju s posljedično smanjenom aktivnosti gena, prate niže vrijednosti LDL-K-a za 11 – 28 % te mnogo manja pojavnost KV događaja, manja intima – medija debljina karotidnih arterija i rijeda pojava periferne arterijske bolesti⁵⁴⁻⁵⁹.

Spomenuta su otkrića bila temelj za ideju farmakološke inhibicije PCSK9 u bolesnika intolerantnih ili rezistentnih na statine, zatim u onih s nasljednom ili primarnom hiperkolesterolemijom te u svih visokorizičnih KV bolesnika koji ne postižu ciljni LDL-K unatoč optimalnoj terapiji. Rezultati do sada provedenih kliničkih studija vrlo su obećavajući. Za razliku od prije spomenutih mipomersena i lomitapida, u PCSK9 inhibitora nije bilo značajnijih nuspojava, a postignuta je snažnija redukcija LDL-K-a. Istraženo je više načina inhibicije PCSK9. Dok je prva faza kliničkih ispitivanja s „antisense“ oligonukleotidima prekinuta⁶⁰, rezultati ispitivanja inhibicije PCSK9 monoklonalskim protutijelima ili malom interferirajućom RNA vrlo su obećavajući. Male interferirajuće RNA vežu se za PCSK9 glasničku RNA i onemogućuju njezinu translaciju te produkciju PCSK9.

and Drug Administration (FDA) for the treatment of the homozygous hereditary hypercholesterolemia (**Figure 1**). Mipomersen is a specific oligonucleotide that binds to ApoB messenger RNA and inhibits its translation, i.e. the synthesis of ApoB (the primary structural apoprotein for LDL-C and other atherogenic lipoproteins), and consequently also VLDL particles, which finally results in lower LDL-C values. Lomitapide is an oral inhibitor for the microsomal triglyceride transfer protein (MTP) and an important link in the production of VLDL in the liver⁴¹. Although clinical trials showed a significant reduction in LDL-C values, their effect on CV outcomes has not yet been examined. Due to common side effects (increased buildup of fats in the liver and severe skin reactions at the injection site for mipomersen), it is not likely that these drugs will ever be approved for the treatment of patients with statin intolerance and resistance^{42,43}.

Protein convertase subtilisin/kexin type 9 (PCSK9)

PCSK9 inhibitors are new revolutionary drugs for the treatment of elevated LDL-C values. The therapeutic use of PCSK9 inhibition was preceded by the discovery of a PCSK9 gene mutation in patients with the dominant form of hereditary hypercholesterolemia in 2003⁴⁴. It has been demonstrated that the mutation of the PCSK9 gene is present in 10-25% of cases of heterozygous hereditary hypercholesterolemia, resulting in increased activity of the gene and elevated concentrations of PCSK9 in the liver and in peripheral blood^{7,45}.

PCSK9 is an enzyme from the proteinase K similar proteins, belonging to the secretory subtilisin group. It is primarily synthesized in hepatocytes and secreted in the liver, where it reaches the highest concentrations^{46,47}.

The key role of PCSK9 is the regulation of LDL-R expression in the liver. Binding the N-terminal end of the PCSK9 molecule to LDL-R leads to it being internalized in the cell and broken down with lysosomes (**Figure 2**). The result is a reduction in the amount of cell LDL-R, lower clearance, and higher plasma LDL-C concentration⁴⁸. Extrahepatic effects of PCSK9 include increased secretion of chylomicrons and regulation of cholesterol absorption in enterocytes⁴⁷. Studies using animal models demonstrated a strong effect of PCSK9 on the progression or regression of atherosclerotic changes⁴⁹. The results of these studies suggest that PCSK9 does not only affect the metabolism of cholesterol but is also involved in glucose metabolism, hepatocyte regeneration, and susceptibility to hepatitis C virus infection⁵⁰⁻⁵³.

While the PCSK9 gene is more active in patients with familial hypercholesterolemia, clinical studies have shown that a mutation that results in reduced activity of the gene is accompanied by a reduction in LDL-C levels of 11-28% and a significantly lower incidence of CV events, lower carotid intima-media thickness, and less common peripheral arterial disease⁵⁴⁻⁵⁹.

These discoveries were the basis for the idea of applying pharmacological PCSK9 inhibition in patients with statin intolerance or resistance, and later in those with hereditary or primary hypercholesterolemia and in all high-risk CV patients who do not achieve target LDL-C values despite optimal therapy. The results of clinical trials have so far been very promising. As opposed to previously mentioned mipomersen and

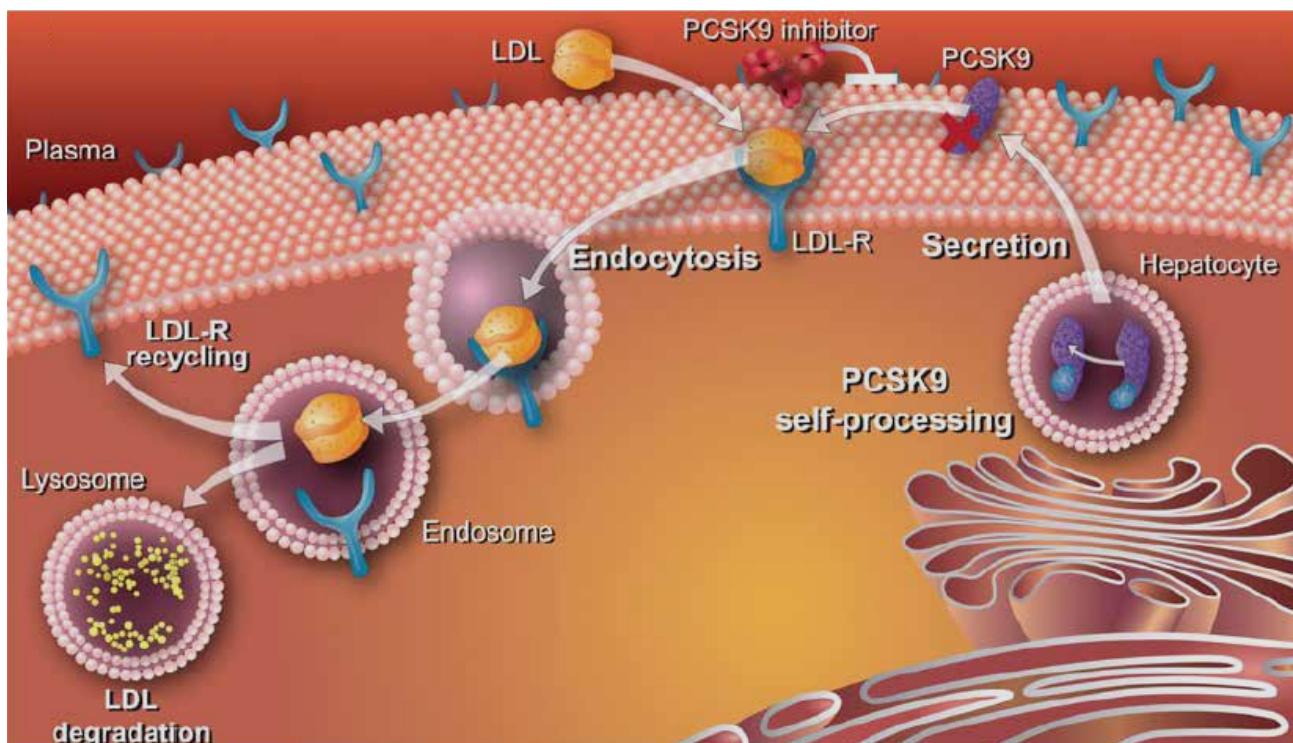


FIGURE 2. Regulation of hepatic LDL receptor expression and mechanism of LDL cholesterol reduction by PCSK9 inhibition (adapted from Shimada YJ et al94).

Nakon uspješnih pretkliničkih pokusa na životinjama, povođenjem učinak malih interferirajućih RNA dokazan je i u prvoj fazi kliničkih istraživanja, u kojima je postignuta 70 %-tina redukcija slobodnog PCSK9 u plazmi i 40 %-tina redukcija LDL-K-a^{61,62}. Trenutačno je najaktualnija monoklonska inhibicija PCSK9 molekule. Alirokumab i evolokumab humana su monoklonska protutijela koja vežu PCSK9, povećavaju broj LDL-R-a i snizuju koncentraciju LDL-K-a (slika 2). Bokocizumab je rekombinantno humanizirano mišje protutijelo na PCSK9. Dovršeno je više kliničkih istraživanja 3. faze s alirokumabom (ODYSSEY Mono, ODYSSEY COMBO I, ODYSSEY COMBO II) i evolokumabom (DECARTES, LAPLACE-2, GAUSS-2, MENDEL-2, RUTHERFORD-2, OSLER-2, TESLA Part B), a takva klinička istraživanja s bokocizumabom još su u tijeku^{63,64}.

Alirokumab

Učinkovitost je alirokumaba ispitana u više od 6000 ispitanika s primarnom hipercolesterolemijom koji nisu podnosiли statine ili nisu postizali ciljni LDL-K unatoč maksimalnoj podnošljivoj dozi hipolipemika.

ODYSSEY-MONO prva je studija s alirokumabom. Ispitivane su učinkovitost i sigurnost alirokumaba kao monoterapije u odnosu prema ezetimibu u ispitniku s hipercolesterolemijom i umjerenim KV rizikom (10-godišnji rizik od smrti zbog KV događaja od 1 do 5 % prema SCORE alatu), prije toga neliječenih statinima ili drugim hipolipemikom. U 103 ispitnika s vrijednostima LDL-K-a od 2,6 do 4,9 mmol/L, alirokumab je u usporedbi s ezetimibom doveo do znatnog sniženja LDL-K-a. U vecine ispit-

lomitapide, PCSK9 inhibitors did not show significant side effects, while achieving stronger LDL-C reduction. Multiple ways of inhibiting PCSK9 have been studied. While the first phase of clinical trials with antisense oligonucleotides has been discontinued⁶⁰, trial results for PCSK9 inhibition with monoclonal antibodies or small interfering RNA have been very promising. Small interfering RNA binds to PCSK9 messenger RNA and prevent its translation and the production of PCSK9. After successful pre-clinical animal model experiments, the beneficial effect of small interfering RNA has been demonstrated in the first phase of clinical trials, where a 70% reduction in free plasma PCSK9 and a 40% reduction in LDL-C were achieved^{61,62}.

The most recent focus is monoclonal inhibition of the PCSK9 molecule. Alirocumab and evolocumab are human monoclonal antibodies that bind PCSK9, increase the number of LDL-Rs, and reduce LDL-C concentrations (Figure 2). Bococizumab is a recombinant humanized mouse antibody against PCSK9. Multiple phase 3 clinical trials with alirocumab (ODYSSEY Mono, ODYSSEY COMBO I, ODYSSEY COMBO II) and evolocumab (DECARTES, LAPLACE-2, GAUSS-2, MENDEL-2, RUTHERFORD-2, OSLER-2, TESLA Part B) have been completed, while such trials with bococizumab are still under way^{63,64}.

Alirocumab

The effectiveness of alirocumab was studied in over 6000 study participants with primary hypercholesterolemia that had statin intolerance or did not achieve target LDL-C values despite maximum tolerable hypolipidemic agent doses.

nika niža doza alirokumaba od 75 mg jednom u dva tjedna bila je dovoljna za redukciju početnog LDL-K-a za 50 %⁶⁵.

Studije ODYSSEY COMBO I (311 bolesnika) i ODYSSEY COMBO II (707 bolesnika) obuhvatile su ispitanike s hipercolesterolemijom i visokim KV rizikom, koji nisu postigli ciljne vrijednosti LDL-K-a unatoč liječenju maksimalno podnošljivim dozama statina (s dodatnom hipolipemijskom terapijom ili bez nje). Uspoređivan je učinak alirokumaba u odnosu prema ezetimibu. Oba su ispitivanja potvrdila mnogo bolji učinak alirokumaba na LDL-K i druge ispitivane parametre lipida. U ODYSSEY COMBO II studiji alirokumabom je nakon 24 tjedna liječenja postignuto sniženje LDL-K-a za 50,6 %, dok je ezetimib snizio LDL-K za 20,7 % ($p < 0,0001$). Također, mnogo je više ispitanika na alirokumabu postiglo ciljnu vrijednost LDL-K (< 1,8 mmol/L) u usporedbi s ispitanicima na ezetimibu (77 prema 45,6 %, $p < 0,0001$)^{66,67}. U ODYSSEY-OPTIONS dodatak alirokumaba statinu rezultirao je mnogo većom redukcijom LDL-K-a negoli dodatak ezetimiba, udvostručenje doze statina ili prebacivanje na potentniji rosuvastatin^{68,69}.

ODYSSEY FH I i ODYSSEY FH II ispitivale su učinkovitost i sigurnost alirokumaba u bolesnika s heterozogotnim oblikom porodične hipercolesterolemije. Ciljni LDL-K bio je definiran ovisno o KV riziku: u bolesnika bez poznate KV bolesti < 2,6 mmol/L, a kod poznate KV bolesti < 1,8 mmol/L. U 24. tjednu praćenja 72,2 % ispitanika na alirokumabu postiglo je ciljni LDL-K u FH I, a 81,4 % u FH II ($p < 0,0001$)^{70,71}. Povoljan učinak na LDL-K, uz dobru podnošljivost alirokumaba, održan je do 78. tjedna praćenja⁷².

ODYSSEY LONG TERM, do sada najopsežnije istraživanje s alirokumabom, procjenjivalo je dugotrajnu sigurnost i podnošljivost alirokumaba. Obuhvatilo je čak 2341 visoko rizičnog bolesnika (bolesnike s heterozigotnim oblikom porodične hipercolesterolemije s manifestnom KV bolesti ili bez nje te bolesnike s primarnom hipercolesterolemijom i koronarnom bolesti srca), koji nisu postigli ciljne vrijednosti LDL-K-a unatoč maksimalno podnošljivoj hipolipemijskoj terapiji, pri čemu je 44 % ispitanika primalo maksimalne dopuštene doze statina. Nakon 24 tjedna liječenja alirokumabom je postignuto smanjenje LDL-K-a za visokih 61,9 % te drugih značajnih lipidnih parametara rizika – ApoB-a za 54,0 %, ne-HDL kolesterol za 52,3 %, lipoproteina(a) za 25,6 %, uz porast HDL-K-a od 4,6 % ($p < 0,0001$). Ciljnu vrijednosti LDL-K-a < 2,6 mmol/L postiglo je 76 % ispitanika na alirokumabu, u usporedbi sa samo 2 % ispitanika u kontrolnoj skupini, a ciljnu vrijednost LDL-K-a < 1,8 mmol/L čak 81 % ispitanika na alirokumabu u odnosu prema 9 % u kontrolnoj skupini⁷³.

Evolokumab

Evolokumab je u nizu kliničkih ispitivanja potvrdio visoku učinkovitost i sigurnost. Iako je učinak lijeka bio sličan pri primjeni svaka dva ili svaka četiri tjedna⁷⁴, kod primjene svaka četiri tjedna postojale su veće fluktuacije vrijednosti LDL-K-a⁷⁵. U studiji MENDEL ukupno je 406 ispitanika s hipercolesterolemijom i nepodnošnjem statina randomizirano u podskupine koje su primale evolokumab od 70, 105 i 140 mg svaka 2 tjedna, podskupine koje su primale 280, 350 i 420 mg svaka 4 tjedna ili ezetimib. Evolokumab je pokazao veliku superiornost, a najsnajniji je učinak postignut dozom od 140 mg svaka 2 tjedna, bez značajnih nuspojava⁷⁶. Studija MENDEL-2 ispitivala je

ODYSSEY-MONO was the first study on alirocumab. The effectiveness and safety of alirocumab monotherapy was studied, in comparison with ezetimibe, in patients with hypercholesterolemia and moderate CV risk (10-year risk of death from CV events of 1-5% according to the SCORE tool) who had not been previously treated with statins or other hypolipidemic agents. In 103 patients with LDL-C values of 2.6-4.9 mmol/L, alirocumab led to a significant reduction in LDL-C values in comparison with ezetimibe. In most participants, the lower alirocumab dose of 75 mg once per two weeks was sufficient to reduce baseline LDL-C by 50%⁶⁵.

The studies ODYSSEY COMBO I (311 patients) and ODYSSEY COMBO II (707 patients) consisted of participants with hypercholesterolemia and high CV risk that did not achieve target LDL-C values despite treatment with maximum tolerable statin doses (with or without additional hypolipidemic therapy). The effectiveness of alirocumab in comparison with ezetimibe was studied. Both studies demonstrated significantly better effects of alirocumab on LDL-C values and other studied lipid parameters. In the ODYSSEY COMBO II, 24 weeks of treatment with alirocumab achieved a reduction of LDL-C values of 50.6%, while ezetimibe reduced LDL-C by 20.7% ($p < 0.0001$). Furthermore, significantly more participants treated with alirocumab achieved target LDL-C values (<1.8 mmol/L) in comparison with participants treated with ezetimibe (77.0% vs. 45.6%, $p < 0.0001$)^{66,67}. In the ODYSSEY-OPTIONS study, adding alirocumab to a statin resulted in significantly larger reduction of LDL-C values than adding ezetimibe, doubling the statin dose, or changing to the more potent rosuvastatin^{68,69}.

The studies ODYSSEY FH I and ODYSSEY FH II examined the effectiveness and safety of alirocumab in patients with heterozygous familial hypercholesterolemia. Target LDL-C values were defined based on CV risk: in patients with no diagnosed CV disease as <2.6 mmol/L, and as <1.8 mmol/L in those with already diagnosed CV diseases. At 24-week follow-up, 72.2% of participants using alirocumab achieved target LDL-C values in FH I and 81.4% in FH II ($p < 0.0001$)^{70,71}. The beneficial effect on LDL-C, along with good tolerance of alirocumab, was still present at 78-week follow-up⁷².

ODYSSEY LONG TERM, currently the broadest study on alirocumab, assessed the long-term safety and tolerability of alirocumab. It encompassed as many as 2341 high-risk patients (patients with heterozygous familial hypercholesterolemia with or without manifest CV diseases and patients with primary hypercholesterolemia and coronary heart disease) who did not achieve target LDL-C values despite maximum tolerable hypolipidemic therapy, with 44% of the participants taking maximum safe doses of statins. After 24 weeks of treatment, alirocumab achieved a high reduction in LDL-C values of 61.9%, as well as reduction of other significant lipid risk parameters – ApoB by 54.0%, non-HDL cholesterol by 52.3%, and lipoprotein(a) by 25.6%, along with an increase in HDL-C of 4.6% ($p < 0.0001$). Target LDL-C values of <2.6 mmol/L were achieved in 76% of participants taking alirocumab, in comparison with just 2% of participants in the control group, and the target value of <1.8 mmol/L was achieved by as much as 81% of participants taking alirocumab in comparison with 9% in the control group⁷³.

Evolocumab

Evolocumab has proven its high effectiveness and safety in a number of clinical trials. Although the effect of the drug was

ucinkovitost, sigurnost i podnošljivost evolokumaba u usporedbi s ezetimibom i placebom u ispitanika s hipercolesterolemijom i visokim KV rizikom. Evolokumab je snizio LDL-K za 55 – 57 % više negoli placebo te za 38 – 40 % više negoli ezetimib (u oba slučaja $p < 0,001$)⁷⁷. U studiji LAPLACE-TIMI 57 bolesnici s hipercolesterolemijom liječeni statinima bili su randomizirani u podskupine koje su primale različite doze evolokumaba u različitim vremenskim intervalima (svaka dva i svaka četiri tjedna). Nakon 12 tjedana postignuta je snažna redukcija LDL-K-a ovisna o dozi evolokumaba (42 – 66 % kod primjene svaka dva tjedna i 41 – 50 % kod primjene svaka četiri tjedna)⁷⁸. Studija LAPLACE-2 uspoređivala je evolokumab u različitim dozama i vremenu primjene (140 mg svaka dva tjedna i 420 mg jednom mjesечно) s placebom i ezetimibom, u 1896 ispitanika s primarnom hipercolesterolemijom i miješanom dislipidemijom, liječenih umjerenim i visokim dozama statina. Nađeno je da evolokumab dodan statinima mnogo bolje snizuje LDL-K (za 66 – 75 %, ovisno o dozi) nakon 10 do 12 tjedana u usporedbi s ezetimibom ili placebom⁷⁹.

Studija DESCARTES s evolokumabom randomizirala je ispitanike u četiri skupine, ovisno o KV riziku i inicijalnoj vrijednosti LDL-K-a: na ispitanike koji nisu primali dodatnu hipolipemijsku terapiju, na one koji su uz evolokumab primali atorvastatin 10 mg ili atorvastatin u dozi od 80 mg te na ispitanike koji su uz evolokumab primali dvojnu terapiju atorvastatom 80 mg i ezetimibom 10 mg. Snažni povoljni učinci evolokumaba na LDL-K dokazani su u svim četirima skupinama. Osim povoljnog učinka na LDL-K, dokazano je i znatno sniženje ostalih lipidnih čimbenika KV rizika – apolipoproteina B, lipoproteina(a) i triglicerida⁸⁰.

Studija GAUSS je ispitivala učinkovitost i podnošljivost evolokumaba u ispitanika koji nisu podnosili statine zbog mijalgije ili miopatije. Pritom je ispitivan evolokumab u dozi od 280 mg, 350 mg i 420 mg te evolokumab 420 mg u kombinaciji s ezetimibom 10 mg. Učinak evolokumaba i kombinacije uspoređivan je s ezetimibom 10 mg. Evolokumab je u svim dozama doveo do znatne redukcije LDL-K-a (za 40 – 65%, ovisno o dozi), pri čemu je mijalgiju imalo samo 7,4 % ispitanika na evolokumabu, 20 % ispitanika na kombinaciji evolokumaba i ezetimiba te 3,1 % ispitanika na ezetimibu i placebo⁸¹. Studija GAUSS-2 također je uključivala ispitanike s hipercolesterolemijom i statinskom intolerancijom^{82,83}. Ispitanici su primali evolokumab 140 mg svaka dva tjedna ili 420 mg jednom mjesечно, dok je kontrolna skupina primala ezetimib. Evolokumab je u usporedbi s ezetimibom doveo do znatnije redukcije vrijednosti LDL-K-a uz nižu pojavnost mišićnih nuspojava (12 % na evolokumabu, 23 % na ezetimibu).

Studija RUTHERFORD dokazala je visoku učinkovitost evolokumaba u bolesnika s heterozigotnim oblikom nasljedne hipercolesterolemije (167 ispitanika) i visokim LDL-K-om unatoč uzimanju maksimalno podnošljivih doza statina⁸⁴. U studiji RUTHERFORD-2 trećina je ispitanika uzimala maksimalno podnošljive doze statina, a dvije trećine uz statine i ezetimib. Randomizirani su u skupine na evolokumabu 140 mg svaka dva tjedna i evolokumabu 420 mg jednom mjesечно te na kontrolnu skupinu na placebo. U objema skupinama s evolokumabom nakon 12 tjedana postignuto je znatno sniženje LDL-K-a (za 59,2 % na 140 mg i 61,3 % na 420 mg) ($p < 0,0001$)⁸⁵.

similar when taken every two and every four weeks⁷⁴, larger fluctuations in LDL-C values were present in four-week doses⁷⁵. The MENDEL study included a total of 406 participants with hypercholesterolemia and statin intolerance randomized into subgroups taking evolocumab in doses of 70, 105, and 140 mg every 2 weeks, subgroups taking 280, 350, and 420 mg every 4 weeks, and taking ezetimibe. Evolocumab was shown to be greatly superior, and the strongest effect was achieved with the dose of 140 mg every 2 weeks, with no significant side effects⁷⁶. The MENDEL-2 study assessed the effectiveness, safety, and tolerability of evolocumab in comparison with ezetimibe and placebo in participants with hypercholesterolemia and high CV risk. Evolocumab reduced LDL-C values by 55-57% more in comparison with placebo and 38-40% more in comparison with ezetimibe ($p < 0,0001$ in both cases)⁷⁷. The LAPLACE-TIMI study randomized 57 patients with hypercholesterolemia who were treated with statins into subgroups taking different doses of evolocumab in different intervals (every two and every four weeks). A significant reduction in LDL-C values was achieved after 12 weeks, depending on the evolocumab dose (42-66% when taken every two weeks and 41-50% when taken every four weeks)⁷⁸. The LAPLACE-2 study compared evolocumab at different doses and intervals (140 mg every two weeks and 420 once per month) with placebo and ezetimibe in 1896 participants with primary hypercholesterolemia and mixed dyslipidemia that were treated with moderate and high doses of statins. It was found that adding evolocumab to statin treatment reduces LDL-C values significantly more (by 66-75%, depending on the dose) after 10 to 12 weeks in comparison with ezetimibe or placebo⁷⁹.

The DESCARTES study with evolocumab randomized participants into four groups based on CV risk and initial LDL-C values, consisting of participants receiving no additional hypolipidemic therapy, those that in addition to evolocumab also received atorvastatin 10 mg or atorvastatin 80 mg, and participants that received combination therapy consisting of atorvastatin 80 mg and ezetimibe 10 mg in addition to evolocumab. The strong beneficial effects of evolocumab were demonstrated in all four groups. In addition to the beneficial effect on LDL-C values, the study also found a significant reduction in other lipid CV risk factors – apolipoprotein B, lipoprotein(a), and triglycerides⁸⁰.

The GAUSS study examined the effectiveness and tolerability of evolocumab in participants with statin intolerance due to myalgia or myopathy. Evolocumab doses of 280 mg, 350 mg, and 420 mg were tested, as well as evolocumab 420 mg in combination with ezetimibe 10 mg. The effect of evolocumab and the combination treatment was compared with ezetimibe 10 mg. At all doses, evolocumab achieved a significant reduction of LDL-C (40-65%, depending on the dose), with myalgia being present in only 7.4% participants using evolocumab, 20% of participants taking combination evolocumab and ezetimibe treatment, and 3.1% of participants receiving ezetimibe and placebo⁸¹. The GAUSS-2 study also included participants with hypercholesterolemia and statin intolerance^{82,83}. The study participants received evolocumab 140 mg every two weeks or 420 mg once per month, while the control group was given ezetimibe. Evolocumab led to a more significant reduction in LDL-C values with a lower incidence of muscle side effects in comparison with ezetimibe (12% for evolocumab, 23% for ezetimibe).

Studija TESLA dokazala je povoljan učinak evolokumaba u bolesnika s homozigotnim oblikom porodične hiperkolesterolemije, liječenih maksimalnim podnošljivim dozama statina i drugom hipolipemijskom terapijom, pri čemu je nakon 12 tjedana liječenja evolokumabom postignuto dodatno sniženje LDL-K-a od 31 % u odnosu prema placebo, bez značajnih nuspojava⁸⁶. Studija OSLER ispitivala je učinak evolokumaba na LDL-K, ali i na KV ishode. Ukupno 4465 ispitanika randomizirano je u skupine koje su primale 140 mg evolokumaba svaka 2 tjedna i 420 mg jednom mjesечно. Tijekom 11,1 mjeseca praćenja evolokumab je snizio LDL-K za 61 % (s prosječnim 3,1 mmol/L na 1,2 mmol/L, p < 0,001). Zajednički KV ishodi (smrtnost, akutni koronarni sindrom, zatajivanje srca, moždani udar ili TIA) nakon godinu dana u kontrolnoj skupini iznosi 2,18 %, a u skupini na evolokumabu 0,95 % (HR 0,47; 95 % CI 0,28 – 0,78; p = 0,003). Veći je dio bolesnika prije randomizacije primao statine (69,7 % u skupini s evolokumabom i 70,9 % u kontrolnoj skupini na placebo)⁸⁷.

Studija GLAGOV s evolokumabom procijenit će učinke niske koncentracije LDL-K-a na regresiju volumena aterosklerotske plaka u bolesnika s poznatom koronarnom bolesti srca⁸⁸.

Bokocizumab

U fazi 2b kliničkih istraživanja s bokocizumabom postignuto je znatno sniženje vrijednosti LDL-K-a u ispitanika s hiperkolesterolemijom liječenih statinima. Uključena su 354 ispitanika, a ispitivane su doze od 50 mg, 100 mg i 150 mg bokocizumaba svaka dva tjedna te doze od 200 mg i 300 mg jednom mjesечно kroz 12 tjedana. Bokocizumab je u svim dozama i učestalosti davanja ostvario značajan povoljan učinak na LDL-K u usporedbi s placeboom, pri čemu su najsnadniji redukciji LDL-K-a ostvarile doze od 150 mg svaka dva tjedna i 300 mg jednom mjesечно⁸⁹. Klinička istraživanja 3. faze s bokocizumabom započeta su u listopadu 2013. godine. Uključuju dvije studije koje primarno ispituju KV ishode (SPIRE-I i SPIRE-II) te više studija s ispitivanjem učinka bokocizumaba na različite lipidne parametre, a uključuju ispitivanje s hiperkolesterolemijom i visokim KV rizikom, rezistentne na statine. Primarni je cilj studije SPIRE-I ispitati da li sniženje LDL-K-a ispod vrijednosti preporučenih u postojećim smjernicama dodatno povoljno djeluje na KV ishode u visokorizičnih ispitanika s inicijalnim LDL-K-om od 1,81 do 2,59 mmol/L⁶³. Studija SPIRE-II ispitivat će učinkovitost i sigurnost bokocizumaba u visokorizičnih bolesnika koji nisu postigli ciljnu vrijednost LDL-K-a < 2,59 mmol/L unatoč terapiji statinima ili u onih koji statine ne podnose⁶⁴. U objema navedenim studijama početna doza bokocizumaba je 150 mg svaka dva tjedna.

Dodatni učinci inhibitora PCSK9

Osim povoljnog učinka na LDL-K, inhibitori PCSK9 pokazali su i dodatne povoljne hipolipemijske učinke. Povišeni lipoprotein(a) neovisan je čimbenik KV rizika u bolesnika liječenih statinima s niskim LDL-K-om⁹⁰. Inhibitori PCSK9 snizuju Apo(a) za oko 30 %, što implicira moguće dodatne kardioprotektivne učinke povrh onih postignutih utjecajem na LDL-K.

The RUTHERFORD study demonstrated the high effectiveness of evolocumab in patients with the heterozygous hereditary hypercholesterolemia (167 participants) and high LDL-C despite taking the maximum tolerable statin dose⁸⁴. In the study RUTHERFORD-2, a third of the participants took maximum tolerable statin doses, and two thirds received ezetimibe in addition to statins. The participants were randomized into groups that received evolocumab 140 mg every two weeks, evolocumab 420 mg once per month, and the control group taking placebo. In both groups on evolocumab, significant reduction in LDL-C values was achieved after 12 weeks (59.2% for 140 mg and 61.3% for 420 mg doses) (p<0.0001)⁸⁵.

The TESLA study demonstrated the positive effect of evolocumab in patients with homozygous familial hypercholesterolemia treated with maximum tolerable statin doses and other hypolipidemic therapy, with evolocumab treatment achieving an additional LDL-C reduction of 31% after 12 weeks in comparison with placebo, with no significant side effects⁸⁶. The OSLER study examined the effect of evolocumab on LDL-C values as well as CV outcomes. A total of 4465 participants were randomized in groups taking 140 mg evolocumab every 2 weeks and 420 mg once per month. During 11.1 months of follow-up, evolocumab reduced LDL-C values by 61% (from an average of 3.1 mmol/L to 1.2 mmol/L, p<0.001). Combined cardiovascular outcomes (mortality, acute coronary syndrome, heart failure, stroke, or TIA) were 2.18% after one year in the control group, and 0.95% in the evolocumab group (HR 0.47; 95% CI 0.28-0.78; p=0.003). Before randomization, most patients were receiving statins (69.7% in the evolocumab group and 70.9% in the control group receiving placebo).⁸⁷

The GLAGOV study with evolocumab will assess the effects of low LDL-C concentrations on volume regression of atherosclerotic plaque in patients with already diagnosed coronary heart disease⁸⁸.

Bococizumab

Phase 2b clinical trials with bococizumab achieved significant reduction of LDL-C values in participants with hypercholesterolemia treated with statins. The trials included 354 participants receiving doses of bococizumab 50 mg, 100 mg, and 150 mg every two weeks and doses of 200 mg and 300 mg once per month over the course of 12 weeks. Bococizumab achieved a significant beneficial effect on LDL-C values in comparison with placebo at all doses and intervals, with the strongest reduction in LDL-C being achieved by doses of 150 mg every two weeks and 300 mg once per month⁸⁹. Phase 3 clinical trials with bococizumab began in October 2013. They include two studies which primarily assess CV outcomes (SPIRE-I and SPIRE-II) and multiple studies assessing the effects of bococizumab on various lipid parameters, consisting of participants with hypercholesterolemia and high CV risk who were resistant to statins. The primary aim of the SPIRE-I study is to examine whether LDL-C reduction below values recommended in current guidelines has an added beneficial effect on CV outcomes in high-risk participants with initial LDL-C values of 1.81 to 2.59 mmol/L⁶³. The SPIRE-II study will examine the effectiveness and safety of bococizumab in high-risk patients who have not achieved the target LDL-C value of <2.59mmol/L despite statin therapy or in those with statin intoler-

Sigurnost inhibitora PCSK9

U navedenim studijama, uz učinkovitost, ispitivana je i sigurnost inhibitora PCSK9. Nije bilo značajne razlike u javljanju blagih i težih nuspojava između ispitivanih monoklonskih protutijela i komparativnih lijekova ili placebo^{79,80}. U manje od 2% ispitnika na inhibitorima PCSK9 pojavljivao se porast aminotransferaza za više od tri puta od gornje granice normalne. Jednako je rijedak bio i porast CK⁹¹.

Najčešće nuspojave inhibitora PCSK9 bile su nazofaringitis, infekcije gornjega dišnog sustava, simptomi nalik gripi i bol u leđima. Reakcije na ubodnom mjestu supuktane primjene lijeka pojatile su se u 2% ispitnika na alirokumabu i 4% na evolokumabu⁹². Učestalost prekida liječenja zbog nuspojava nije se znatno razlikovala između inhibitora PCSK9 (alirokumaba i evolokumaba) i komparatora (2–10%)⁹³.

Zaključak

Vrijednosti LDL-K-a povezane s najpovoljnijim KV ishodima u osoba s visokim i vrlo visokim rizikom vrlo su niske (< 2,6 i < 1,8 mmol/L). Postizanje niskih vrijednosti veliki je izazov kakav dosadašnji hipolipemici često ne dostižu. Otkriće novih regulatornih mehanizama uključenih u metabolizam lipoproteina otvorilo je put novim farmakološkim pristupima u liječenju povišenog LDL-K-a.

Spoznanje uloge PCSK9 u ekspresiji LDL-R-a i u klirensu LDL-K-a potaknule su brzi razvoj novih skupina lijekova⁹⁴. Od nekoliko istraživanih skupina inhibitora PCSK9 najsnažniji razvoj doživjela su monoklonska protutijela. Alirokumab, evolokumab i bococizumab snizuju vrijednost LDL-K-a za 50–70%, uz dobru podnošljivost i visoku sigurnost. Veliki izazov u njihovoj praktičnoj primjeni svakako će biti cijena, a dijelom i supuktani način primjene. Povoljan utjecaj na KV ishode registriran je u studiji OSLER s evolokumabom te se ispituje u nizu kliničkih istraživanja. Rezultati spomenutih studija sigurno će dati jasniju sliku o kliničkoj djelotvornosti i ekonomskoj isplativosti inhibitora PCSK9 u odabranih skupina bolesnika.

erance⁶⁴. Both of these studies use a starting bococizumab dose of 150 mg every two weeks.

Additional effects of PCSK9 inhibitors

In addition to a beneficial effect on LDL-C, PCSK9 inhibitors have also showed other beneficial hypolipidemic effects. Elevated lipoprotein(a) is an independent CV risk factor in patients treated with statins with low LDL-C⁹⁰. PCSK9 inhibitors reduced Apo(a) by approximately 30%, implying possible additional cardioprotective effects beyond those achieved by influencing LDL-C values.

PCSK9 inhibitor safety

The abovementioned studies also assessed the safety of PCSK9 inhibitors in addition to their effectiveness. There was no significant difference in the incidence of mild and more severe side effects between the assessed monoclonal antibodies and comparative drugs or placebo^{79,80}. Less than 2% of participants receiving PCSK9 inhibitors presented with aminotransferase elevations of three or more times the upper limit of normal values. CK elevation was equally rare⁹¹.

The most common PCSK9 inhibitor side effects were nasopharyngitis, upper respiratory tract infections, flu-like symptoms, and back pain. Reactions at the subcutaneous site of drug injection were present in 2% of the study participants on alirocumab and 4% of those on evolocumab⁹². The incidence of treatment termination due to side effects did not differ significantly between PCSK9 inhibitors (alirocumab and evolocumab) and comparison drugs (2–10%)⁹³.

Conclusion

LDL-C values associated with the most positive CV outcomes in persons with high and very high risk are very low (<2.6 and <1.8 mmol/L). Achieving low values is a great challenge, and currently patients often fail to achieve them with hypolipidemic therapy. The discovery of new regulatory mechanisms involved in lipoprotein metabolism has opened the way to new pharmaceutical approaches for the treatment of elevated LDL-C.

Discovering the role of PCSK9 in LDL-R expression and LDL-C clearance has spurred the rapid development of new groups of drugs⁹⁴. Of the several groups of PCSK9 inhibitors, monoclonal antibodies have seen the most development. Alirocumab, evolocumab, and bococizumab reduce LDL-C values by 50–70%, with good tolerance and high safety. A great challenge in their practical application will undoubtedly be their price, and also in part their subcutaneous application. A beneficial effect on CV outcomes has been found in the OSLER study with evolocumab and is being assessed in a series of clinical trials. The results of these studies will certainly give a clearer picture of the clinical effectiveness and cost-effectiveness of PCSK9 inhibitors in chosen groups of patients.

LITERATURE

- Boekholdt SM, Arsenault BJ, Mora S, Pedersen TR, LaRosa JC, Nestel PJ, et al. Association of LDL cholesterol, non-HDL cholesterol, and apolipoprotein B levels with risk of cardiovascular events among patients treated with statins: a meta-analysis. *JAMA*. 2012;307(12):1302-9. DOI: <http://dx.doi.org/10.1001/jama.2012.366>
- Sniderman AD, Williams K, Contois JH, Monroe HM, McQueen MJ, de Graaf J, et al. A meta-analysis of low-density lipoprotein cholesterol, non-high density lipoprotein cholesterol, and apolipoprotein B as markers of cardiovascular risk. *Circ Cardiovasc Qual Outcomes*. 2011;4(3):337-45. DOI: <http://dx.doi.org/10.1161/CIRCOUTCOMES.110.959247>
- Cholesterol Treatment Trialists' (CTT) Collaboration, Baigent C, Blackwell L, Emberson J, Holland LE, Reith C, Bhala N, et al. Efficacy and safety of more intensive lowering of LDL cholesterol: a meta-analysis of data from 170 000 participants in 26 randomised trials. *Lancet*. 2010;376(9753):1670-81. DOI: [http://dx.doi.org/10.1016/S0140-6736\(10\)61350-5](http://dx.doi.org/10.1016/S0140-6736(10)61350-5)

Disadvantages of Current LDL-cholesterol Lowering and the Role of PCSK9 Inhibitors

4. Mihaylova B, Emberson J, Blackwell L, Keech A, Simes J, Barnes EH, et al; Cholesterol Treatment Trialists C. The effects os lowering LDL cholesterol with statin therapy in people with low risc of vascular disease: meta-analysis of individual data from 27 randomised trials. *Lancet.* 2012;380(9841):581-90. DOI: [http://dx.doi.org/10.1016/S0140-6736\(12\)60367-5](http://dx.doi.org/10.1016/S0140-6736(12)60367-5)
5. Reiner Ž, Catapano AL, De Backer G, Graham I, Taskinen MR, Wiklund O. ESC/EAS guidelines for the management of dyslipidaemias: the task force for the management of dyslipidaemias of the European Society of Cardiology (ESC) and the European Atherosclerosis Society (EAS). *Eur Heart J.* 2011;32:1769-818. DOI: <http://dx.doi.org/10.1093/eurheartj/ehr158>
6. Piepoli MF, Hoes AW, Agewall S, Albus C, Brotons C, Catapano AL, et al. 2016 European Guidelines on cardiovascular disease prevention in clinical practice: The Sixth Joint Task Force of the European Society of Cardiology and Other Societies on Cardiovascular Disease Prevention in Clinical Practice (constituted by representatives of 10 societies and by invited experts): Developed with the special contribution of the European Association for Cardiovascular Prevention & Rehabilitation (EACPR). *Eur J Prev Cardiol.* 2016 Jun 27. pii: 2047487316653709. [Epub ahead of print]. DOI: <http://dx.doi.org/10.1177/2047487316653709>
7. Maxwell KN, Breslow JL. Proprotein convertase subtilisin kexin 9: the third locus implicated in autosomal dominant hypercholesterolemia. *Curr Opin Lipidol.* 2005;16(2):167-72. PubMed: <http://www.ncbi.nlm.nih.gov/pubmed/15767856>
8. Humphries SE, Whittall RA, Hubbard CS, Maplebeck S, Cooper JA, Soutar AK, et al. Simon Broome Familial Hyperlipidaemia Register Group and Scientific Steering Committee. Genetic causes of familial hypercholesterolaemia in patients in the UK: relation to plasma lipid levels and coronary heart disease risk. *J Med Genet.* 2006;43(12):943-9. DOI: <http://dx.doi.org/10.1136/jmg.2006.038356>
9. Humphries SE, Cranston T, Allen M, Middleton-Price H, Fernandez MC, Senior V, et al. Mutational analysis in UK patients with a clinical diagnosis of familial hypercholesterolaemia: relationship with plasma lipid traits, heart disease risk and utility in relative tracing. *J Mol Med (Berl).* 2006;84(3):203-14. DOI: <http://dx.doi.org/10.1007/s00109-005-0019-z>
10. Stone NJ, Robinson J, Lichtenstein AH, Bairey Merz CN, Lloyd-Jones DM, Blum CB, et al. ACC/AHA guideline on the treatment of blood cholesterol to reduce atherosclerotic cardiovascular risk in adults: a report of the American College of Cardiology/American Heart Association Task Force on practice guidelines. *J Am Coll Cardiol.* 2014;63(25 Pt B):2889-934. DOI: <http://dx.doi.org/10.1016/j.jacc.2013.11.002>
11. Fernandez G, Spatz ES, Jablecki C, Phillips PS. Statin myopathy: a common dilemma not reflected in clinical trials. *Cleve Clin J Med.* 2011;78(6):393-403. DOI: <http://dx.doi.org/10.3949/ccjm.78a.10073>
12. European Medicines Agency. Committee for Medicinal Products for Human Use (CHMP). Guideline on clinical investigation of medicinal products in the treatment of lipid disorders, Available at: http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2014/01/WC500159540.pdf
13. Guyton JR, Bays HE, Grundy SM, Jacobson TA, The National Lipid Association Statin Intolerance Panel. An assessment by the Statin Intolerance Panel: 2014 update. *J Clin Lipidol.* 2014;8(3 Suppl):S72-81. DOI: <http://dx.doi.org/10.1016/j.jacl.2014.03.002>
14. Mancini GB, Baker S, Bergon J, Fitchett D, Frohlich J, Genset J, et al. Diagnosis, prevention, and management of statin adverse effects and intolerance: proceedings of a Canadian Working Group Consensus Conference. *Can J Cardiol.* 2011;27(5):635-62. DOI: <http://dx.doi.org/10.1016/j.cjca.2011.05.007>
15. Abd TT, Jacobson TA. Statin induced myopathy: a review and update. *Expert Opin Drug Saf.* 2011;10(3):373-87. DOI: <http://dx.doi.org/10.1517/14740338.2011.540568>
16. Carr DF, O'Meara H, Jorgensen AL, Campbell J, Hobbs M, McCann G, et al. SLC01B1 genetic variant associated with statin-induced myopathy: a proof-of-concept study using the clinical practice research datalink. *Clin Pharmacol Ther.* 2013;94(6):695-701. DOI: <http://dx.doi.org/10.1038/clpt.2013.161>
17. Patel J, Superko HR, Martin SS, Blumenthal RS, Christopher-Sine L. Genetic and immunologic susceptibility to statin-related myopathy. *Atherosclerosis.* 2015 240(1):260-71. DOI: <http://dx.doi.org/10.1016/j.atherosclerosis.2015.03.025>
18. Egan A, Colman E. Weighing the benefits of high-dose simvastatin against the risk of myopathy. *N Engl J Med.* 2011;365(4):285-7. DOI: <http://dx.doi.org/10.1056/NEJMmp1106689>
19. Mancini GB, Baker S, Bergeron J, Fitchett D, Frohlich J, Genest J, et al. Diagnosis, prevention, and management of statin adverse effects and intolerance: proceedings of a Canadian Working Group Consensus Conference. *Can J Cardiol.* 2011;27(5):635-62. DOI: <http://dx.doi.org/10.1016/j.cjca.2011.05.007>
20. Hirota T, Ieiri I. Drug-drug interactions that interfere with statin metabolism. *Expert Opin Drug Metab Toxicol.* 2015;11(9):1435-47. DOI: <http://dx.doi.org/10.1517/17425255.2015.1056149>
21. Murinson BB, Maragakis NJ, Jacobson TA. Fluvastatin, rhabdomyolysis, and myotoxicity. *Mayo Clin Proc.* 2008;83(11):1296; author reply 1297. DOI: <http://dx.doi.org/10.4065/83.11.1296>
22. Catapano AL. Statin-induced myotoxicity: pharmacokinetic differences among statins and the risk of rhabdomyolysis, with particular reference to pitavastatin. *Curr Vasc Pharmacol.* 2012;10(2):257-67. DOI: <http://dx.doi.org/10.2174/157016112799305021>
23. Mampuya WM, Frid D, Rocco M, Huang J, Brennan DM, Hazen SL, et al. Treatment strategies in patients with statin intolerance: the Cleveland clinic experience. *Am Heart J.* 2013;166(3):597-603. DOI: <http://dx.doi.org/10.1016/j.ahj.2013.06.004>
24. Keating AJ, Campbell KB, Guyton JR. Intermittent nondaily dosing strategies in patients with previous statin-induced myopathy. *Ann Pharmacother.* 2013;47(3):398-404. DOI: <http://dx.doi.org/10.1345/aph.R509>
25. Calderon RM, Cubeddu LX, Goldberg RB, Schiff ER. Statins in the treatment of dyslipidemia in the presence of elevated liver aminotransferase levels: a therapeutic dilemma. *Mayo Clin Proc.* 2010;85(4):349-56. DOI: <http://dx.doi.org/10.4065/mcp.2009.0365>
26. Argo CK, Loria P, Caldwell SH, Lonardo A. Statins in liver disease: a molehill, an iceberg, or neither? *Hepatology.* 2008;48(2):662-9. DOI: <http://dx.doi.org/10.1002/hep.22402>
27. Oni ET, Sinha P, Karim A, Martin SS, Blaha MJ, Agatston AS, et al. Statin use is not associated with presence of and severity of nonalcoholic fatty liver disease. *Arch Med Res.* 2014;45(1):52-7. DOI: <http://dx.doi.org/10.1016/j.arcmed.2013.12.003>
28. Younoszai Z, Li Z, Stepanova M, Erario M, Cable R, Younossi ZM. Statin use is not associated with liver related mortality. *Ann Hepatol.* 2013;13(1):84-90. PubMed: <http://www.ncbi.nlm.nih.gov/pubmed/24378207>
29. Maki KC, Ridker PM, Brown WV, Grundy SM, Sattar N, The Diabetes Subpanel of the National Lipid Association Expert Panel. An assessment by the Statin Diabetes Safety Task Force: 2014 update. *J Clin Lipidol.* 2014;8(Suppl 3):S17-29. DOI: <http://dx.doi.org/10.1016/j.jacl.2014.02.012>
30. Reiner Ž. Resistance and intolerance to statins. *Nutr Metab Cardiovasc Dis.* 2014;24(10):1057-66. DOI: <http://dx.doi.org/10.1016/j.numecd.2014.05.009>
31. Bitto A, Pallio G, Messina S, Arcoraci V, Pizzino G, Russo GT, et al. Genomic Variations Affecting Biological Effects of Statins. *Curr Drug Metab.* 2016;17(6):566-72. DOI: <http://dx.doi.org/10.2174/138920021766160219114116>
32. Simon JA, Lin F, Hulley SB, Blanche PJ, Waters D, Shibuski S, et al. Phenotypic predictors of response to simvastatin therapy among African-Americans and caucasians: the cholesterol and pharmacogenetics (CAP) Study. *Am J Cardiol.* 2006;97(6):843-50. DOI: <http://dx.doi.org/10.1016/j.amjcard.2005.09.134>
33. Al-Sarraf A, Li M, Frohlich J. Statin resistant dyslipidemia in a patient treated with amiodarone. *BMJ Case Rep.* Published online 2011. pii: bcr0820114620. DOI: <http://dx.doi.org/10.1136/bcr.08.2011.4620>
34. Wierzbicki AS, Doherty E, Lumb PJ, Chik G, Crook MA. Efficacy of ezetimibe in patients with statin-resistant and statin-intolerant familial hyperlipidaemias. *Curr Med Res Opin.* 2005;21(3):333-8. DOI: <http://dx.doi.org/10.1185/030079905X28872>
35. Xydakis AM, Guyton JR, Chiou P, Stein JL, Jones PH, Ballantyne CM. Effectiveness and tolerability of ezetimibe add-on therapy to a bile acid resin-based regimen for hypercholesterolemia. *Am J Cardiol.* 2004;94(6):795-7. DOI: <http://dx.doi.org/10.1016/j.amjcard.2004.06.008>
36. Zaputović L. Kako možemo povećati HDL kolesterol lijekovima. Posebna izdanja HAZU. "Prevencija ateroskleroze: smanjeni HDL-kolesterol kao čimbenik rizika". 2011;67-80.

37. Matijević S, Malić D, Zaputović L. Kako možemo utjecati na hipertrigliceridemiju lijekovima? Posebna izdanja HAZU. „Prevencija ateroskleroze: hipertrigliceridemija kao čimbenik rizika“. 2011;71-89.
38. Keating GM, Ormrod D. Micronised fenofibrate: an updated review of its clinical efficacy in the management of dyslipidaemia. *Drugs*. 2002;62(13):1909-44. **PubMed:** <http://www.ncbi.nlm.nih.gov/pubmed/12215067>
39. HPS2-THRIVE Collaborative Group, Landray MJ, Haynes R, Hopewell JC, Parish S, Aung T, Tomson J, et al. Effects of Extended-Release Niacin with Laropiprant in High-Risk Patients. *N Engl J Med*. 2014;371:203-12. **DOI:** <http://dx.doi.org/10.1056/NEJMoa1300955>
40. Guyton JR, Slee AE, Anderson T, Fleg JL, Goldberg RB, Kashyap ML, et al. Relationship of lipoproteins to cardiovascular events: the AIM-HIGH Trial (Atherothrombosis Intervention in Metabolic Syndrome With Low HDL/High Triglycerides and Impact on Global Health Outcomes). *J Am Coll Cardiol*. 2013;62(17):1580-4. **DOI:** <http://dx.doi.org/10.1016/j.jacc.2013.07.023>
41. Cuchel M, Meagher EA, du Toit Theron H, Blom DJ, Marais AD, Hegele RA, et al. Efficacy and safety of a microsomal triglyceride transfer protein inhibitor in patients with homozygous familial hypercholesterolemia: a single-arm, open-label, phase 3 study. *Lancet*. 2013;381(9860):40-6. **DOI:** [http://dx.doi.org/10.1016/S0140-6736\(12\)61731-0](http://dx.doi.org/10.1016/S0140-6736(12)61731-0)
42. Stein EA, Dufour R, Gagne C, Gaudet D, East C, Donovan JM, et al. Apolipoprotein B synthesis inhibition with mipomersen in heterozygous familial hypercholesterolemia: results of a randomized, double-blind, placebo-controlled trial to assess efficacy and safety as add-on therapy in patients with coronary artery disease. *Circulation*. 2012;126(19):2283-92. **DOI:** <http://dx.doi.org/10.1161/CIRCULATIONAHA.112.104125>
43. Thomas GS, Cromwell WC, Ali S, Chin W, Flaim JD, Davidson M. Mipomersen, an apolipoprotein B synthesis inhibitor, reduces atherogenic lipoproteins in patients with severe hypercholesterolemia at high cardiovascular risk: a randomized, double blind, placebo-controlled trial. *J Am Coll Cardiol*. 2013;62(23):2178-84. **DOI:** <http://dx.doi.org/10.1016/j.jacc.2013.07.081>
44. Abifadel M, Varret M, Rabes JP, Allard D, Ouguerram K, Devillers M, et al. Mutations in PCSK9 cause autosomal dominant hypercholesterolemia. *Nat Genet*. 2003;34(2):154-6. **DOI:** <http://dx.doi.org/10.1038/ng1161>
45. Cariou B, Ouguerram K, Zair Y, Guerrier R, Langhi C, Kourimate S, et al. PCSK9 dominant negative mutant results in increased LDL catabolic rate and familial hypobetalipoproteinemia. *Arterioscler Thromb Vasc Biol*. 2009;29(12):2191-7. **DOI:** <http://dx.doi.org/10.1161/ATVBAHA.109.194191>
46. Maxwell KN, Breslow JL. Adenoviral-mediated expression of PCSK9 in mice results in a low-density lipoprotein receptor knockout phenotype. *Proc Natl Acad Sci USA*. 2004;101(18):7100-5. **DOI:** <http://dx.doi.org/10.1073/pnas.0402133101>
47. Seidah NG, Prat A. The biology and therapeutic targeting of the proprotein convertases. *Nat Rev Drug Discovery*. 2012;11(5):367-83. **PubMed:** <http://www.ncbi.nlm.nih.gov/pubmed/22679642>
48. Cariou B, Le May C, Costet P. Clinical aspects of PCSK9. *Atherosclerosis*. 2011;216(2):258-65. **DOI:** <http://dx.doi.org/10.1016/j.atherosclerosis.2011.04.018>
49. Denis M, Marcinkiewicz J, Zaid A, Gauthier D, Poirier S, Lazure C, et al. Gene inactivation of proprotein convertase subtilisin/kexin type 9 reduces atherosclerosis in mice. *Circulation*. 2012;125(7):894-901. **DOI:** <http://dx.doi.org/10.1161/CIRCULATIONAHA.111.057406>
50. Levy E, Ouadha ABD, Spahis S, Sane AT, Garofalo C, Grenier E, et al. PCSK9 plays a significant role in cholesterol homeostasis and lipid transport in intestinal epithelial cells. *Atherosclerosis*. 2013;227(2):297-306. **DOI:** <http://dx.doi.org/10.1016/j.atherosclerosis.2013.01.023>
51. Farnier M. PCSK9: from discovery to therapeutic applications. *Archives of cardiovascular diseases*. 2014;107(1):58-66. **DOI:** <http://dx.doi.org/10.1016/j.acvd.2013.10.007>
52. Farnier M. PCSK9 inhibitors. *Curr Opin Lipidol*. 2013;24(3):251-8. **DOI:** <http://dx.doi.org/10.1097/MOL.0b013e3283613a3d>
53. Bridge SH, Sheridan DA, Felmlee DJ, Crossey MM, Fenwick FI, Lanyon CV, et al. PCSK9, apolipoprotein E and lipopiral particles in chronic hepatitis C genotype 3: evidence from genotype-specific regulation of lipoprotein metabolism. *J Hepatol*. 2015;62(4):763-70. **DOI:** <http://dx.doi.org/10.1016/j.jhep.2014.11.016>
54. Cohen J, Pertselidis A, Kotowski IK, Graham R, Garcia CK, Hobbs HH. Low LDL cholesterol in individuals of African descent resulting from frequent nonsense mutations in PCSK9. *Nat Genet*. 2005;37(2):161-5. **DOI:** <http://dx.doi.org/10.1038/ng1509>
55. Cohen JC, Boerwinkle E, Mosley TH Jr, Hobbs HH. Sequence variations in PCSK9, low LDL, and protection against coronary heart disease. *N Engl J Med*. 2006;354(12):1264-72. **DOI:** <http://dx.doi.org/10.1056/NEJMoa054013>
56. Benn M, Nordestgaard BG, Grande P, Schnohr P, Tybjaerg-Hansen A. PCSK9 R46L, low-density lipoprotein cholesterol levels, and risk of ischemic heart disease: 3 independent studies and meta-analyses. *J Am Coll Cardiol*. 2010;55(25):2833-42. **DOI:** <http://dx.doi.org/10.1016/j.jacc.2010.02.044>
57. Hooper AJ, Marais AD, Tanyanyiwa DM, Burnett JR. The C679X mutation in PCSK9 is present and lowers blood cholesterol in a Southern African population. *Atherosclerosis*. 2007;193(2):445-8. **DOI:** <http://dx.doi.org/10.1016/j.atherosclerosis.2006.08.039>
58. Huang CC, Fornage M, Lloyd-Jones DM, Wei GS, Boerwinkle E, Liu K. Longitudinal association of PCSK9 sequence variations with low-density lipoprotein cholesterol levels: the Coronary Artery Risk Development in Young Adults Study. *Circ Cardiovasc Genet*. 2009;2(4):354-61. **DOI:** <http://dx.doi.org/10.1161/CIRGENETICS.108.828467>
59. Folsom AR, Peacock JM, Boerwinkle E. Atherosclerosis Risk in Communities (ARIC) Study Investigators. Variation in PCSK9, low LDL cholesterol, and risk of peripheral arterial disease. *Atherosclerosis*. 2009;202(1):211-15. **DOI:** <http://dx.doi.org/10.1016/j.atherosclerosis.2008.03.009>
60. Do RQ, Vogel RA, Schwartz GG. PCSK9 Inhibitors: potential in cardiovascular therapeutics. *Curr Cardiol Rep*. 2013;15(3):345. **DOI:** <http://dx.doi.org/10.1007/s11886-012-0345-z>
61. Frank-Kamenetsky M, Grefhorst A, Anderson NN, Racine TS, Bramlage B, Akinc A, et al. Therapeutic RNAi targeting PCSK9 acutely lowers plasma cholesterol in rodents and LDL cholesterol in nonhuman primates. *Proc Natl Acad Sci USA*. 2008;105:11915-20. **DOI:** <http://dx.doi.org/10.1073/pnas.0805434105>
62. Fitzgerald K, Frank-Kamenetsky M, Shulga-Morskaya S, Liebow A, Bettencourt BR, Sutherland JE, et al. Effect of an RNA interference drug on the synthesis of proprotein convertase subtilisin/kexin type 9 (PCSK9) and the concentration of serum LDL cholesterol in healthy volunteers: a randomised, single-blind, placebo-controlled, phase 1 trial. *Lancet*. 2014;383(9911):60-8. **DOI:** [http://dx.doi.org/10.1016/S0140-6736\(13\)61914-5](http://dx.doi.org/10.1016/S0140-6736(13)61914-5)
63. Pfizer. The evaluation of bococizumab (PF-04950615;RN316) in reducing the occurrence of major cardiovascular events in high risk subjects (SPIRE-1). In: ClinicalTrials.gov [Internet]. 2013-2016. May 02. Available from: <https://clinicaltrials.gov/ct2/show/NCT01975376>
64. Pfizer. The evaluation of bococizumab (PF-04950615;RN316) in reducing the occurrence of major cardiovascular events in high risk subjects (SPIRE-2). In: ClinicalTrials.gov [Internet]. 2013-2016 May 12. Available from: <https://clinicaltrials.gov/ct2/show/NCT01975389>
65. Roth EM, McKenna JM. ODYSSEY MONO: effect of alirocumab 75 mg subcutaneously every 2 weeks as monotherapy versus ezetimibe over 24 weeks. *Future Cardiol*. 2015;11(1):27-37. **DOI:** <http://dx.doi.org/10.2217/fca.14.82>
66. Kereiakes DJ, Robinson JG, Cannon CP, Lorenzato C, Pordy R, Chaudhari U, et al. Efficacy and safety of the proprotein convertase subtilisin/kexin type 9 inhibitor alirocumab among high cardiovascular risk patients on maximally tolerated statin therapy: The ODYSSEY COMBO I study. *Am Heart J*. 2015;169(6):906-915.e13. **DOI:** <http://dx.doi.org/10.1016/j.ahj.2015.03.004>
67. Colhoun HM, Robinson JG, Farnier M, Cariou B, Blom D, Kereiakes DJ, et al. Efficacy and safety of alirocumab, a fully human PCSK9 monoclonal antibody, in high cardiovascular risk patients with poorly controlled hypercholesterolemia on maximally tolerated doses of statin: rationale and design of the ODYSSEY COMBO I and II trials. *BMC Cardiovasc Disord*. 2014 Sep 20;14:121. **DOI:** <http://dx.doi.org/10.1186/1471-2261-14-121>
68. Schwartz GG, Bessac L, Berdan LG, Bhatt DL, Bittner V, Diaz R, et al. Effect of alirocumab, a monoclonal antibody to PCSK9, on long term cardiovascular outcomes following acute coronary syndromes: rationale and design of the ODYSSEY outcomes trial. *Am Heart J*. 2014;168(5):682-9. **DOI:** <http://dx.doi.org/10.1016/j.ahj.2014.07.028>

Disadvantages of Current LDL-cholesterol Lowering and the Role of PCSK9 Inhibitors

69. Bays H, Gaudet D, Weiss R, Ruiz JL, Watts GF, Gouni-berthold I, et al. Alirocumab as add-on to atorvastatin versus other lipid treatment strategies: ODYSSEY OPTIONS I randomised trial. *J Clin Endocrinol Metabol.* 2015;100(8):3140-8. DOI: <http://dx.doi.org/10.1210/jc.2015-1520>
70. Kastelein JJP, Robinson JG, Farnier M, Krempf M, Langslet G, Lorenzato C, et al. Efficacy and safety of alirocumab in patients with heterozygous familial hypercholesterolemia not adequately controlled with current lipid-lowering therapy: design and rationale of the ODYSSEY FH studies. *Cardiovasc Drugs Ther.* 2014;28(3):281-9. DOI: <http://dx.doi.org/10.1007/s10557-014-6523-z>
71. Ginsberg HN, Rader D, Raal FJ, Guyton J, Lorenzato C, Pordy R, et al. ODYSSEY high FH: efficacy and safety of alirocumab in patients with severe heterozygous familial hypercholesterolemia. *Circulation.* 2014; 28(3): 281-9. DOI: <http://dx.doi.org/10.1007/s10557-014-6523-z>
72. Kastelein JJP, Ginsberg HN, Langslet G, Hovingh GK, Ceska R, Dufour R, et al. ODYSSEY FH I and FH II: 78 week results with alirocumab treatment in 735 patients with heterozygous familial hypercholesterolemia. *Eur Heart J.* 2015;36(43):2996-3003. DOI: <http://dx.doi.org/10.1093/euroheartj/ehv370>
73. Robinson JG, Farnier M, Krempf M, Bergeron J, Luc G, Averna M, et al. Efficacy and safety of alirocumab in reducing lipids and cardiovascular events. *N Engl j Med.* 2015;372(16):1489-99. DOI: <http://dx.doi.org/10.1056/NEJMoa1501031>
74. Robinson JG, Nedergaard BS, Rogers WJ, Fialkow J, Neutel JM, Ramstad D, et al. LAPLACE-2 Investigators. Effect of evolocumab or ezetimibe added to moderate or high-intensity statin therapy on LDL-Cholesterol in patients with hypercholesterolemia: the LAPLACE-2 randomized clinical trial. *JAMA.* 2014;311(18):1870-82. DOI: <http://dx.doi.org/10.1001/jama.2014.4030>
75. McKenney JM, Koren MJ, Kereiakes DJ, Hanotin C, Ferrand AC, Stein EA. Safety and efficacy of a monoclonal antibody to proprotein convertase subtilisin/kexin type 9 serine protease, SAR236553/REGN727, in patients with primary hypercholesterolemia receiving ongoing stable atorvastatin therapy. *J Am Coll Cardiol.* 2012;59(25):2344-53. DOI: <http://dx.doi.org/10.1016/j.jacc.2012.03.007>
76. Koren MJ, Scott R, Kim JB, Knausek B, Liu T, Lei T, et al. Efficacy, safety and tolerability of a monoclonal antibody to proprotein convertase subtilisin/kexin type 9 as monotherapy in patients with hypercholesterolemia (MENDEL): a randomised, double-blind, placebo controlled, a phase 2 study. *Lancet.* 2012;380(9858):1995-2006. DOI: [http://dx.doi.org/10.1016/S0140-6736\(12\)61771-1](http://dx.doi.org/10.1016/S0140-6736(12)61771-1)
77. Koren MJ, Lundquist P, Bolognese M, Neutel JM, Monsalvo ML, Yang J, et al. Anti PCSK9 monotherapy for hypercholesterolemia: the MENDEL 2 randomised controlled phase III clinical trial of evolocumab. *J Am Coll Cardiol.* 2014;63(23):2531-40. DOI: <http://dx.doi.org/10.1016/j.jacc.2014.03.018>
78. Giugliano RP, Desai NR, Kohli P, Rogers WJ, Somaratne R, Huang F, et al. Efficacy, safety and tolerability of a monoclonal antibody to proprotein convertase subtilisin/kexin type 9 in combination with a statin in patients with hypercholesterolemia (LAPLACE-TIMI 57): a randomised, placebo controlled, dose ranging, phase 2 study. *Lancet.* 2012;380(9858):2007-17. DOI: [http://dx.doi.org/10.1016/S0140-6736\(12\)61770-X](http://dx.doi.org/10.1016/S0140-6736(12)61770-X)
79. Robinson JG, Nedergaard BS, Rogers WJ, Fialkow J, Neutel JM, Ramstad D, et al. LAPLACE-2 Investigators. Effect of evolocumab or ezetimibe added to moderate or high-intensity statin therapy on LDL-Cholesterol in patients with hypercholesterolemia: the LAPLACE-2 randomized clinical trial. *JAMA.* 2014;311(18):1870-82. DOI: <http://dx.doi.org/10.1001/jama.2014.4030>
80. Blom DJ, Hala T, Bolognese M, Lillestol MJ, Toth PD, Burgess L, et al. DESCARTES Investigators. A 52-week placebo controlled trial of evolocumab in hyperlipidemia. *N Engl J Med.* 2014;370(19):1809-19. DOI: <http://dx.doi.org/10.1056/NEJMoa1316222>
81. Sullivan D, Olsson AG, Scott R, Kim JB, Xue A, Gebski V, et al. Effects of a monoclonal antibody to PCSK9 on low-density lipoprotein cholesterol levels in statin-intolerant patients: the GAUSS randomized trial. *JAMA.* 2012;308(23):2497-506. DOI: <http://dx.doi.org/10.1001/jama.2012.25790>
82. Stroes E, Colquhoun D, Sullivan D, Civeira F, Rosenson RS, Waatts GF, et al. Anti-PCSK9 antibody effectively lowers cholesterol in patients with statin intolerance: the GAUSS-2 randomised, placebo controlled, phase 3 clinical trial of evolocumab. *J Am Coll Cardiol.* 2014;63(23):2541-8. DOI: <http://dx.doi.org/10.1016/j.jacc.2014.03.019>
83. Cho L, Rocco M, Colquhoun D, Sullivan D, Rosenson RS, Dent R, et al. Design and the rational of the GAUSS-2 study trial: a double-blind, ezetimib controlled phase 3 study of the efficacy and tolerability of evolocumab (AMG 145) in subjects with hypercholesterolemia who are intolerant of statin therapy. *Clin Cardiol.* 2014;37(3):131-9. DOI: <http://dx.doi.org/10.1002/clc.22248>
84. Raal F, Scott R, Somaratne R, Bridges I, Li G, Wasserman SM, et al. Low-density lipoprotein cholesterol-lowering effects of AMG 145, a monoclonal antibody to proprotein convertase subtilisin/kexin type 9 serine protease in patients with heterozygous familial hypercholesterolemia: the reduction of LDL-C with PCSK9 inhibition in heterozygous familial hypercholesterolemia disorder (RUTHERFORD) randomised trial. *Circulation.* 2012;126(20):2408-17. DOI: <http://dx.doi.org/10.1161/CIRCULATIONAHA.112.144055>
85. Raal FJ, Stein EA, Dufour R, Turner T, Civeira F, Burgess L, et al. PCSK9 inhibition with evolocumab (AMG 145) in heterozygous familial hypercholesterolemia (RUTHERFORD-2): a randomised, double-blind, placebo controlled trial. *Lancet.* 2015;385(9965):331-40. DOI: [http://dx.doi.org/10.1016/S0140-6736\(14\)61399-4](http://dx.doi.org/10.1016/S0140-6736(14)61399-4)
86. Raal FJ, Honarpour N, Blom DJ, Hovingh GK, Xu F, Scott R, et al. Inhibition of PCSK9 with evolocumab in homozygous familial hypercholesterolemia (TESLA Part B): a randomised, double-blind, placebo controlled trial. *Lancet.* 2015;385(9965):341-50. DOI: [http://dx.doi.org/10.1016/S0140-6736\(14\)61374-X](http://dx.doi.org/10.1016/S0140-6736(14)61374-X)
87. Koren MJ, Giugliano RP, Raal FJ, Sullivan D, Bolognese M, Langslet G, et al. OSLER Investigators. Efficacy and safety of longer-term administration of evolocumab (AMG 145) in patients with hypercholesterolemia: 52-week results from the open-label study of long-term evaluation against LDL-C (OSLER) randomized trial. *Circulation.* 2014;129(2):234-43. DOI: <http://dx.doi.org/10.1161/CIRCULATIONAHA.113.007012>
88. Amgen. Global Assessment of Plaque reGression With a PCSK9 antibody as Measured by intraVascular Ultrasound (GLAGOV). In: ClinicalTrials.gov [Internet]. 2013-2015 Mar 17. Available at: <https://clinicaltrials.gov/ct2/show/NCT01813422>
89. Ballantyne CM, Neutel J, Cropp A, Duggan W, Wang EQ, Plowchalk D, et al. Results of Bococizumab, A Monoclonal Antibody Against Proprotein Convertase Subtilisin/Kexin Type 9, from a Randomized, Placebo-Controlled, Dose-Ranging Study in Statin-Treated Subjects With Hypercholesterolemia. *Am J Cardiol.* 2015;115(9):1212-21. DOI: <http://dx.doi.org/10.1016/j.amjcard.2015.02.006>
90. Khera AV, Everett BM, Caulfield MP, Hantash HM, Wohlgemuth J, Ridker PM, et al. Lipoprotein (a) concentrations, rosuvastatin therapy, and residual vascular risk: an analysis from the JUPITER trial (Justification for the use of statins in prevention: an intervention trial evaluating rosuvastatin). *Circulation.* 2014;129(6):635-42. DOI: <http://dx.doi.org/10.1161/CIRCULATIONAHA.113.004406>
91. Cicero AF, Tartagni E, Ertek S. Safety and tolerability of injectable lipid-lowering drugs: a review of available clinical data. *Exp Opin Drug Saf.* 2014;13(8):1023-30. DOI: <http://dx.doi.org/10.1517/14740338.2014.932348>
92. Roth EM, Taskinen MR, Ginsberg HN, Kastelein JJ, Colhoun HM, Robinson JG, et al. Monotherapy with the PCSK9 inhibitor alirocumab versus ezetimibe in patients with hypercholesterolemia: results of a 24 week, double-blind, randomized Phase 3 trial. *Int J Cardiol.* 2014;176(1):55-61. DOI: <http://dx.doi.org/10.1016/j.ijcard.2014.06.049>
93. Li C, Lin L, Zhang W, Zhou L, Wang H, Luo X, et al. Efficiency and Safety of Proprotein Convertase Subtilisin/Kexin 9 Monoclonal Antibody on Hypercholesterolemia: A Meta-Analysis of 20 Randomized Controlled Trials. *J Am Heart Assoc.* 2015;4(6):e001937. DOI: <http://dx.doi.org/10.1161/JAHNR.115.001937>
94. Shimada YJ, Cannon CP. PCSK9 (Proprotein convertase subtilisin/kexin type 9) inhibitors: past, present and future. *Eur Heart J.* 2015;36(36):2415-24. DOI: <http://dx.doi.org/10.1093/euroheartj/ehv174>