REVIEW ARTICLE

Current Drugs and Nutraceuticals for the Treatment of Patients with Dyslipidemias

Michele Scognamiglio[#,1], Dario Costa*[,#,1], Antonio Sorriento[1] and Claudio Napoli[1,2]

[1]*U.O.C. Division of Clinical Immunology, Immunohematology, Transfusion Medicine and Transplant Immunology, Clinical Department of Internal Medicine and Specialistics, Department of Medical, Surgical, Neurological, Metabolic and Geriatric Sciences, University of Campania "L. Vanvitelli", Naples, Italy;* [2]*IRCCS SDN, Naples, Italy*

Abstract: Coronary heart disease (CHD) remains the leading cause of disability and death in industrialized Countries. Among many conditions, which contribute to the etiology and progression of CHD, the presence of high low density lipoprotein-cholesterol (LDL-C) levels represents the major risk factor. Therefore, the reduction of LDL-C levels plays a key role in the management of patients with high or very high cardiovascular risk. Although statins represent the gold standard therapy for the reduction of cholesterol levels, these drugs do not allow to achieve target levels of LDL-C in all patients. Indeed, a significant number of patients resulted intolerants, especially when the dosage increased. The availability of new lipid-lowering drugs, such as ezetimibe and PCSK9 inhibitors, may represent an important alternative or complement to the conventional lipid-lowering therapies. However, long-term studies are still needed to define both efficacy and safety of use of these latter new drugs. Some nutraceuticals may become an adequate and effective support in the management of some patients. To date, several nutraceuticals with different mechanism of actions that provide a good tolerability are available as lipid-lowering agents. In particular, the most investigated are red yeast rice, phytosterols, berberine, beta-glucans and soy. The aim of this review was to report recent data on the efficacy and safety of principle hypocholesterolemic drugs available and to evaluate the possible role of some nutraceuticals as support therapy in the management of patients with dyslipidemias.

Keywords: Nutraceuticals, dyslipidemias. hypercholesterolemia, cardiovascular disease, LDL cholesterol, lipid-lowering agents.

1. INTRODUCTION

Despite the observed progressive reduction of cardiovascular mortality in industrialized Countries, atherosclerosis and subsequent coronary heart diseases (CHD) still remain the major causes of premature death and permanent disability in the Western population [1]. These diseases are multi-factorial because they are caused by multiple genes with the interaction of environmental factors (genetic and epigenetic causes), some are modifiable with lifestyle interventions such as stop smoking, accurate dietary interventions and increased physical activity to contrast the negative effects of overweight and obesity [2, 3]. However, changes in lifestyle are often considered as a deprivation for the patients and in the vast majority of cases are not able to significantly reduce alterations in the lipid profile. Other risk factors including dyslipidemias, hypertension and diabetes, require additional pharmacological treatments.

Dyslipidemias are the most important risk factors for cardiovascular disease, with over 30% of elders that are hypertriglyceridemics and alterations in cholesterol levels affect more than 50% of subjects in adulthood [4]. It has been widely demonstrated that alterations of the lipid profile, in particular of low-density lipoproteins cholesterol (LDL-C), in association with other risk factors can negatively influence the development and progression of CHD and cerebrovascular disease [5]. In the last decades, epidemiological studies characterized by very large populations such as MRFIT (Multiple Risk Factor Intervention Trial) [6], or with extremely long observational periods, such as the Framingham Study [7], have shown a linear relationship between LDL-C levels and incidence/mortality of such diseases.

An interesting survey, conducted in Italy on 7912 adults (aged 35-74 years) between 2008 and 2012 provided the prevalence data of hypercholesterolemia, resulting ≥240 mg/dl for 34% of men and 36% of women [8]. When compared with a previous survey realized in period 1998-2002, these data demonstrated an increase in prevalence in men (+39%) and women (+33%) [4]. The most worrying aspect was that about 40% of the subjects affected by dyslipidemias did not know that they had this pathology, and a similar percentage of patients with established diagnosis did not follow a specific diet or drug therapy. Furthermore, another problem for those who followed a lipid-lowering therapy is that they maintain normal exceeding eating habits comparable to that of subjects without pathology.

The treatment of hypercholesterolemia should be considered as an integral part in the prevention and management interventions of cardiovascular diseases. Different lipid-lowering drugs are now available that allow an effective reduction of cholesterol levels using pharmacological mono- or combined therapy [9, 10]. In particular, statins are the most commonly prescribed cholesterol-lowering agents worldwide to manage and prevent CHD and ischemic cerebrovascular [11-13]. However, there is a great proportion of patients that does not reach the desirable targets after pharmacological treatment or does not tolerate the drugs [14]. Instead, the other drugs introduced to replace or supplement the statins, need further clinical trials to verify the beneficial effect on the treatment of disease.

To overcome the limits presented by lipid-lowering drugs, dietary supplementation with natural compounds has been evaluated as a potential integrative treatment approach for hypercholesterolemia. To date, the number of nutraceuticals compounds that appear to have an effect on reducing LDL-C levels is continuously increasing [15]. Among that, the most investigated are red yeast rice (RYR), phytosterols, berberine, dietary fibers (*i.e.* beta-glucans), and soy. The main advantage of these nutraceuticals seems to be the differ-

*Address Corresponding to this author at the U.O.C. Immunohematology, Transfusion Medicine and Transplant Immunology, Azienda Ospedaliera Universitaria (AOU), University of Campania "L. Vanvitelli" Piazza Miraglia, 2 80138 Napoli; Tel: +390815665067; Fax: +390815665071; E-mail: dario.costa@policliniconapoli.it
[#]*These authors have contributed equally.*

ent mechanism of action and good tolerability when used at appropriate doses in comparison to conventional drugs [16].

The aim of this review was to report recent data on the efficacy and safety of main hypocholesterolemic drugs available and to evaluate the possible role of some nutraceuticals as support therapy in the management of patients with dyslipidemias.

2. LIPID-LOWERING DRUGS

2.1. Statins

Statins represent the first choice therapy for the treatment of hypercholesterolemia and mixed dyslipidemia in patients not responder to dietary treatment or increased physical activity. In addition, the use of statins is strongly recommended in patients with very high LDL-C and cholesterol levels [17, 18].

Statins act through the selective and competitive inhibition of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase, which is the key enzyme of endogenous cholesterol genesis, determining a reduction of circulating LDL. The inhibition of this enzyme determines a decrease of cholesterol synthesis in the liver, but successively as compensatory mechanism, there is an increased expression of LDLR and HMG-CoA reductase. The increase of LDLR leads to a fewer number of very low density lipoprotein (VLDL) and intermediate density lipoprotein (IDL) circulating, with consequently reduced conversion of these precursors into LDL molecules. As a consequence, there is increased endocytosis of the circulating LDL and a decrease of cholesterol transported by LDLs from 30% to over 50% [19].

Statins differ for pharmacokinetic parameters such as plasma protein binding, absorption, bioavailability and interactions with other drugs. In order to exert the hypocholesterolemia activity, lovastatin and simvastatin must be converted *in vivo* into active forms through hydrolysis reactions [20]. Compared to other statins, atorvastatin and rosuvastatin have the longest terminal half-life (11–20 h *vs.* 1-4 h) [21]. Moreover, particular attention should be given to the interaction between statins and other drugs such as fibrates, niacin, cyclosporine, and all inhibitors of cytochrome 3A4 (CYP3A4); the consequences of such combinations can be severe myopathy or rhabdomyolysis [22]. Likewise, co-administration of warfarin with fluvastatin, lovastatin and rosuvastatin can significantly increase anticoagulant activity [23]. The knowledge of these characteristics is necessary for an adequate personalization of drug therapy. Despite the existence of a class-effect common to all the statins, the hypocholesterolemic efficacy varies according to the different molecules and the relative dosages. The American College of Cardiology categorized statin regimens as low intensity (<30% LDL-C reduction with simvastatin 10 mg daily), moderate intensity (30%-50% reduction with simvastatin 20–40 mg, atorvastatin 10–20 mg, or rosuvastatin 5–10 mg daily), or high intensity (>50% reduction with atorvastatin 40–80 mg or rosuvastatin 20–40 mg daily) [24].

The benefit of treatment with statins have been confirmed by meta-analysis of several randomized controlled trials on more than 170,000 patients of Cholesterol Treatment Trialists' Collaboration [25, 26]. In particular, it was found that a reduction of 38 mg/dl of LDL-C is associated with a 20-25% reduction in the relative risk of new major cardiovascular events (cardiovascular mortality and nonfatal infarction) [27]. However, the absolute benefit of the treatment is greater in patients with higher baseline LDL-C levels (baseline effect). All these findings are confirmed by the considerable reduction in age-related cardiovascular mortality rates observed in recent years, passing from 62/100000 people per year in 1980 to approximately 19 in 2008 in men, and from 13 to 4 in women [28].

The most recent guidelines have clearly indicated that there are different categories of cardiovascular risk, which correspond to a specific target of LDL-C. For patients with very high cardiovascu-

lar risk the target of LDL-C is <70 mg/dl (or a reduction of more than 50% of basal LDL-C) providing the best benefits in the management of disease. In these cases, only high doses of atorvastatin (80 mg) and rosuvastatin (40 mg) may reduce of approximately 50% the values of LDL-C, greatly increasing the risk of incurring side effects and serious adverse reactions [29]. In patients with moderate or high risk, the target value of LDL-C could be respectively <100 mg/dl and 115 mg/dl. In asymptomatic subjects, the first step is to evaluate the overall cardiovascular risk and identify all modifiable risk factors such as diet, not smoking and sedentary lifestyle.

Despite the high hypocholesterolemic efficacy of statins, a significant proportion of patients on pharmacological treatment with statins does not reach cholesterol levels target. Others patients are intolerant to statins even at low daily doses, determining high rates of discontinuation in therapy as well as a proportion of subjects who refuse treatment with these drugs for the apprehension of possible adverse events [30]. Moreover, patients have been reported that while achieving target cholesterol levels already develop cardiovascular disease (residual cardiovascular risk) [31]. In these cases, a quantification of the residual risk in the population and an improved comprehension of molecular mechanisms at the base of vascular events could be essential to reduce the burden of cardiovascular disease.

Different adverse events associated with statin therapy have been reported such as myopathy, hypertransaminases, nephropathy and onset of type II diabetes [30, 32-34]. Myopathy typically manifests with muscle pain (more or less intense) and can be associated with up to 10-fold increase in serum creatine kinase (CK) concentration [35]. The severe form of myopathy is rhabdomyolysis characterized by marked CK and myoglobin levels, which can lead to acute renal failure. Hepatic intolerance to statins typically manifests with a significant increase in serum transaminases up to 3-fold compared to the normal values which disappears with treatment discontinuation and reappears after re-challenge. Furthermore, data emerging from randomized clinical trials demonstrated an increased risk of development of type II diabetes in predisposed subjects (obese, familiarity, sedentary lifestyle) treated with statins [36, 37] Anyway, even if confirmed this risk would be counterbalanced by the benefit of the reduction of cardiovascular events and mortality in subjects receiving statins.

2.2. Ezetimibe

When the treatment with statins result ineffective because not lead to LDL-C target values, the addition of ezetimibe may be an appropriate choice. The hypocholesterolemic action of ezetimibe is achieved through a selective reduction of intestinal cholesterol absorption at the duodenum-jejunum level mediated by the Niemann-Pick C1-like protein 1 (NPC1L1) sterol transporter block. [38] As a result, the reduction of cholesterol absorption leads to improved receptor-mediated LDL-C uptake; however, when the drug is used in monotherapy, in response to lower cholesterol absorption, an increased LDL-C biosynthesis can occur. Thus, the compensatory effect limit hypocholesterolemic activity of ezetimibe.

IMPROVE-IT is the first clinical trial that evaluated the efficacy of ezetimibe in combination with statins compared to statin monotherapy in hypercholesterolemic patients [39]. In patients intolerant to statins, the ezetimibe used alone leads to the reduction of LDL-C for values ranging between 15-20%. The association with statins would be able to guarantee a reduction of the LDL-C values by a further 20% [40] and also provides the decrease of Apoliprotein B (ApoB) in patients with primary hypercholesterolemia [41]. In addition to the potent lipid-lowering activity, the results of the IMPROVE-IT study have clearly demonstrated that statin-ezetimibe association guarantees a significant improvement in the clinical prognosis through reduction of cardiovascular events [42]. The benefit of adding ezetimibe was largely improved in pa-

tients with diabetes mellitus, but also in non-diabetic patients with high cardiovascular risk [43]. Moreover, several studies have reported the efficacy of ezetimibe combined with different statins in the reduction of coronary plaque volume in patients with chronic kidney disease (CKD) that have a higher cholesterol absorption compared to patients without CKD [44, 45]. The American College of Cardiology proposed the use of ezetimibe (daily dose recommended of 10 mg for adults) as the first choice in patients who fail to reach the corresponding LDL-C target levels at their cardiovascular risk level with statin therapy alone [46].

Interestingly, it has been observed that ezetimibe unlike other cholesterol-lowering agents (with a similar mechanism of action) not interfere with the absorption of other molecules such as triglycerides, liposoluble vitamins, fatty acids, bile acids, progesterone and also ethinyl estradiol [47]. Furthermore, the absence of hepatic metabolism involving CYP450 allows ezetimibe to significantly reduce all possible drug interactions, potentially dangerous.

2.3. PCSK9 Inhibitors

In addition to the well-known capacity of increase of LDLR expression, the use of statins also activates the expression of a particular protein of 692 amino acids, proprotein convertase subtlysin/kexina type 9 (PCSK9), which acts as a negative control mechanism by reducing the number of LDLR. This protein plays a key role in the metabolism of cholesterol, with principal expression in liver and bowel. Normally, the presence of a ApoB domain at the LDLR allows the binding to LDL-C and the subsequent internalization of the complex thus formed, in a clathrin-coated vesicle. In the presence of an acidic environment such as the inside of the endosomes, the dissociation of LDL-C from its receptor occurs [48]. As a consequence, LDLR can return to the cell surface, while its ligand is degraded to the lysosomal level. In the endoplasmic reticulum of hepatocyte, PCSK9 is first cleaved and then secreted into the circulatory stream. When LDL-C is fixed to the receptor in the presence of PCSK9, the recycling of LDLR on the surface is greatly reduced. In this way, the amount of LDLR on the cell surface decreases as well as the uptake of the LDL-C from the circulatory stream [48].

Different studies have observed that a loss-of-function mutations of the PCSK9 gene lead to a significant reduction in plasma levels of LDL-C and cardiovascular risk, proving the key role played by this protein in the metabolism of cholesterol [49, 50]. Contrarily, an inverse relationship between the density of LDLR expressed on the hepatocyte surface and the levels of PCSK9 have been observed [51]. Thus, it is evident that there is a direct relationship between the levels of circulating LDL-C and PCSK9. This observation have encouraged an intense research activity on PCSK9 with consequent development of a new class of hypocholesterolemic drugs represented by PCSK9 inhibitors.

The hypocholesterolemic efficacy of these drugs has been confirmed by several clinical trials both in monotherapy and in association with statins or ezetimibe, ensuring a mean reduction in cholesterol levels above 50% [52-54]. In the recent years, at least 6 different monoclonal antibodies have been developed as PCSK9 inhibitors, among which the most investigated are evolocumab (Repatha®) and alirocumab (Praluent®) [55] These antibodies, specifically binding to circulating PCSK9 inhibit the degradation of LDLR and increase its expression on the hepatocyte surface. The ability of these drugs to exercise control over the entire lipid profile is particular interesting. It has also been observed that the increased expression of LDLR is followed by a greater clearance of VLDL and IDL residues, thus determining a reduction of ApoB, non-high density lipoprotein (HDL) cholesterol, Lp(a) and triglycerides [53, 56]. PCSK9 inhibitors, in line with the recommendations of the main International regulatory organization (EMA, FDA) are indicated only for patients with primary hypercholesterolemia and mixed dyslipidemia, which do not satisfactorily respond to the

maximum tolerated dose of statins or in those patients for whom there are contraindications to the use of statins [57-59].

For its particular pharmacokinetic, alirocumab and evolocumab have a bioavailability approximately of 80% and a half-life of about 15 days (from 11 days to 20 days) which allows for subcutaneous or intravenous administration every two weeks. According to baseline cholesterol levels in patients, the recommended adult dose of alirocumab is 75 mg or 150 mg every two weeks. Similarly, the recommended dose of evolocumab is 140 mg every two weeks or 420 mg once a month [60]. However, its use in clinical practice is strongly conditioned by the high cost (100-fold greater than statins), which can only be partially justified by the possible reduction of cardiovascular events and days of hospitalization.

2.4. Nicotinic Acid (Niacin)

Nicotinic acid and its amide are pyridine derivatives with vitamin activity. Because of similarity between the terms nicotine and nicotinic acid, to avoid that the alkaloid contained in the tobacco was considered an essential nutrient, it was preferred to use nicotinic acid referring to niacin and its amide (niacinamide). The mechanism of action of the lipid-lowering activity of nicotinic acid has been unknown for a long time and also today remain many ambiguous aspects. In recent years, it has been shown that many cell types such as adipocytes, immune cells and keratinocytes express on their surface a specific nicotinic acid receptor (GPR109A) or HCA receptor 2 (HCA2) through which it exerts its different pharmacological effects [61]. The anti-lipolytic effect mediated by the activation of HCA2 is well known and consists into a reduced release of free fatty acids from adipose tissue to the liver [61]. This results in decreased hepatic production and secretion of VLDL-triglycerides as well as decreased circulating LDL-C levels [62-64]. Different experimental evidence has shown the ability of this agent to decrease also the clearance of HDL-ApoA1 [65, 66]. However, the mechanism underlying the increase in HDL-C concentration is less clear at the moment.

The nicotinic acid assumed at dosages between 1.5-3 g/day may reduce LDL cholesterol, triglycerides, free fatty acids and Lp(a) levels as well as markedly increase HDL-C concentration [67, 68]. Particularly, Lp(a) is an emerging cardiovascular risk factor independent of traditional markers such as total cholesterol, LDL cholesterol and ApoB. However, the reduction of Lp(a) levels by niacin result extremely variable, ranging between 10% and 40% in comparison to baseline levels [67, 68]. This is due to the differences in drug dose, duration of treatment and detection methods as well as inter-individual patients characteristics [69]. Interestingly, different studies conducted on patients with hyperlipidemia or dyslipidemia evaluated niacin/simvastatin combination therapy, resulting more efficacy and safety compared to statins monotherapy [70-72].

Although the anti-dyslipidemic action of nicotinic acid is widely documented, numerous safety and efficacy aspects remain to be evaluated. The most common adverse events reported were flushing (mainly located on the face and neck), dizziness, blurred vision, abnormal liver function and gastro-intestinal disorders such as diarrhea and nausea that require suspension of treatment [73]. Nicotinic acid was authorized in combination with laropripant. Laropripant has no effect on cholesterol but reduces hot flushes which are the principle side effect of nicotinic acid at cholesterol-lowering dosage. As the consequence of these numerous side effect, its use is contraindicated for diabetics and patients with reduced hepatic or renal function.

3. FROM LIPID-LOWERING DRUGS TO NUTRACEUTICALS

The first approach in the management of the patient with mild or moderate hypercholesterolemia is the improvement of lifestyle considering the optimization of dietary habits, increase of physical activity and smoking abolition. This approach must be maintained

and consolidated even in the case of pharmacological interventions. In recent years, there is an increased interest on nutraceuticals and in particular on their hypocholesterolemic activity.

The term nutraceutical was coined in 1989 from the fusion of the terms "nutrition" and "pharmaceutical". It represents the science that identifies and investigates foods or particular components with recognized health benefits. A large number of compounds are included in this group, such as food supplements, functional foods, and medicinal plant preparations. According to the recent scientific evidence and literature available, the European Food Safety Authority (EFSA) has authorized specific nutritional and healthy indications for nutraceuticals regulating the communication to the final

users [74]. Frequently, the nutraceuticals formulations for the control of cholesterol levels are proposed as associations; thus, it is possible to use the different mechanisms of action of the compounds and to reduce the quantities of the single components in order to limit their toxicity. Despite the growing interest of consumers and manufacturers on nutraceuticals, many aspects remain unclear. Indeed, in many cases the results obtained have proven conflicting, often showing methodological limitations. In particular, large clinical studies to evaluate their efficacy and safety as well as the assessment of the related health claims and successively marketing are needed. In Table **1** was listed the most important nutraceuticals compounds that was investigated in different clinical trials,

Table 1. **Dosage, duration of treatment and main features of single nutraceutical compounds evaluated in clinical studies.**

Compound	Dosage	Patients characteristics	Protective effects	Concerns	Mean duration of treatment	References
RYR	3-10 mg/day	Mild/moderate hypercholesterolemia and low CV risk	↓LDL-C ≤25% vs baseline;	Risk of contamination (citrinin);	12-24 weeks	[79]; [80]; [81]; [82]; [84];
			↓CV risk;	High costs;		
			Good tollerability;	No for statins intollerant;		
Phytosterols	1.5-3.0 g/day	Mild hypercholesterolemia and low CV risk	↓LDL-C ≤10% vs baseline;	↓Absorption of fat-soluble vitamins;	10-12 weeks	[86]; [87]; [88]; [90];
			Low cost			
Berberine	500-1500 mg/day	Mild/moderate hypercholesterolemia and low CV risk	↓LDL-C ≤20% vs baseline;	RCT only in asiatic people;	12 weeks	[94]; [95]; [100];
			↓TG;	High costs;		
			↑HDL-C;	Additive effect with PCSK9 inhibitors;		
			Improve of glucose metabolism			
Beta-glucans	3-5 g/day	Mild hypercholesterolemia and low CV risk	↓LDL-C 5-6% vs baseline;	High dose = intestinal discomfort;	4-6 weeks	[105]; [106]; [107]; [108];
			↓TG;	Possible anti-nutritional action;		
			Improve of glucose metabolism			
			Low cost			
			Support for dietary control			
Soy	25-30 g/day	Mild/moderate hypercholesterolemia and low CV risk	↓LDL-C ≤10% vs baseline;	Allergenic risk;	4 weeks-1 years	[111]; [112]; [113];
			↑HDL-C;	GMO?;		
Polyphenols	150 mg-13 g/day	Mild hypercholesterolemia and low CV risk	↓LDL-C ≤15% vs baseline;	Possible interaction with drugs;	4-12 weeks	[115]; [116]; [123]; [124];
			↑HDL-C;	Limited bioavailability;		
				Incomplete pharmacokinetic data;		
Policosanol	5-80 mg/day	Mild hypercholesterolemia and low CV risk	↓LDL-C ≤25% vs baseline;	Mixed data?;	8-24 weeks	[132]; [133]; [134]; [135];
			↑HDL-C;	RCT mainly in Cuban people;		
			↓Total cholesterol;			
			Good tollerability;			

CV: Cardiovascular Risk; GMO: Genetically Modified Organism; HDL-C: High Density Lipoprotein-Cholesterol; LDL-C: Low Density Lipoprotein-Cholesterol; Lp(a): Lipoprotein(a); RCT: Randomized Clinical Trials; RYR: Red Yeast Rice; TG: Triglycerides

Table 2. **Main food containing nutraceuticals with hypocholesterolemic activity and their healthy daily intake considering a balanced diet.**

Food source	Nutraceuticals	Daily intake
Whole grains (bran, barley, oats)	Beta-glucan/fibers	80-100g
Vegetable oils, dried fruits, nuts, legumes and enriched foods	Phytosterols	30-150ml of oil/ 80-100g of fruits or vegetables
Grapes and red wine	Resveratrol	300g of fruits/ 125-250ml of wine
Capers and onions	Quercetin	100g of raw vegetables
Apple, bergamot and pomegranate	Anthocianins and anthocyanidins	200g of fruits/ 240 ml of juice
Soy foods	Isoflavones, lecithin and PUFA	25-100g
Yoghurt and dairy products	Probiotics	125-200g

PUFA: Polyunsatured fatty acids. Data from http://www.euro.who.int/__data/assets/pdf_file/0017/150083/E79832.pdf?ua=1.

considering the advantages and relative contraindications for their use. Main food containing nutraceuticals with hypocholesterolemic activity and their healthy daily intake was reported in Table **2**.

4. NUTRACEUTICALS

4.1. Red Yeast Rice

The extract obtained from the rice (*Oryza Sativa*) fermentation by yeasts as *Monascus Purpureus* and other microorganisms of the same group is one of the most studied and effective hypocholesterolemic nutraceutical. During fermentation, the yeasts produce a wide series of metabolites that have important biological activity, including monacolins with a mean concentration of around 2% [75]. The most effective components of RYR extracts is represented by monacolin K, a molecule with a chemical structure very similar to lovastatin. It reduces endogenous cholesterol synthesis by inhibiting HMG-CoA reductase. In addition to the monacolin K, it is possible to find other molecules with important bioactivities such as sterols, isoflavones, mono-unsaturated fatty acids and other monacolins (J, L, X, M) that could increase the hypocholesterolemic activity and modify the monacolin K bioavailability [76].

Notably, compared to lovastatin, monacolin K may exist in two different chemical forms: the lactonic and acidic form (particularly active and bioavailable). The proportion between the two forms can vary considerably, depending on the fermentation conditions and the type of yeast used. The conversion from lactonic to acidic form can also occur in alkaline environment or enzymatically through the activity of intestinal or hepatic CYP3A4 [77]. However, despite the structural analogy with lovastatin, monacolin K has a different pharmacokinetic profile and different bioavailability. Instead, monacolin K shows greater bioavailability compared to purified lovastatin at the same doses. Therefore, the therapy with RYR extract using a minor quantity of active ingredient seems to guarantee reduced risk of adverse events and a greater adherence to therapy.

The hypocholesterolemic action of RYR has been confirmed by numerous studies [78], reporting efficacy data that can be compared to those of "older" generation statins. It has been observed that the daily dose intake of 3 mg of monacolin K may reduce LDL-C levels <4.14 mmol/L in 51% of the participants and 26% achieved the threshold level of homocysteine <10 μmol/L [79]. The additive synergistic action of other components present in RYR, including non-monacolin K seems contribute to the hypocholesterolemic effect of monacolin K with a greater efficacy compared to "old

generation" statins also at significant lower doses [80]. Randomized double-blind controlled trials have shown a reduction in total cholesterol and LDL-C of 15-20% and 22-25% respectively, with daily doses of 10 mg of monacolin K for periods of 8-12 weeks [81, 82]. Magno S *et al.* have confirmed the efficacy of a dietary supplements monacolin K-based (Argicolina®) on the significant reduction of LDL-C levels, also evaluating its association with L-arginine, coenzyme Q10 and ascorbic acid [83]. In addition, a significant reduction of triglycerides levels was observed, while it have not been found differences in HDL-C levels.

In addition to the hypocholesterolemic activity, RYR also seems to guarantee a significant reduction in C-reactive protein levels, an important inflammatory index and cardiovascular risk factor as well as other vascular remodeling plasma markers such as matrix metalloproteinases 2 and 9 [84].

The greater tolerability of the preparations based on RYR compared to the statins could be due to several reasons including the lower amount of monacolin K required and no less importantly also a psychological component related to the belief of assuming a natural product. In any case to prevent the occurrence of adverse events in particularly sensitive individuals (even at minimal doses of statins), it is preferable to avoid co-administration of RYR-based preparations with drug that inhibits CYP3A4. Particular attention must be paid to quality and purity of products based on RYR, because it is frequent the risk of contamination, especially from citrinin, a toxic mycotoxin for the liver and kidney [76].

4.2. Phytosterols (Plant Sterols and Stanols)

Plant sterols and stanols (also known as phytosterols) are compounds naturally and exclusively present in variable quantity in vegetables. Principal food sources of phytosterols are represented by refined vegetable oils (corn, rape, soybean, grape seed, etc.) and whole grains where these molecules are located in the germ of the seeds. Thanks to a molecular structure very similar to the cholesterol, they reduce its absorption by competing for cholesterol as components of mixed micelles [85]. As consequence of reduced cholesterol intestinal absorption, phytosterols promote a slight increase in its synthesis in the liver and at the same time a greater expression of LDLR on the hepatocyte surface, increasing their capacity of uptake. The hypocholesterolemic action of phytosterols is also enhanced by their ability to form crystals with cholesterol, increasing its fecal excretion.

As reported by different studies, the hypocholesterolemic efficacy of plant sterols contained in functional food at dosages of 1.5-2 g/day is generally around 10% compared to baseline values and a further moderate reduction of cholesterol levels (12.5%) has been observed at dosages of 2.5-3 g/day [86-88]. Unlike other nutraceuticals after the intake of plant sterols there are no changes in HDL-C and triglycerides plasma levels. It has been estimated that in Western countries, the intake of plant sterols through foods is around 500 mg/day [89]. Therefore, the therapeutically active dose in the cholesterol control of plant sterols is difficult to reach only with diet and require additional supplementation for hypocholesterolemic activity.

According to EFSA recommendation, the same effects and dosages are also necessary in the case of stanols considering a minimum period of supplementation at least 2-3 weeks [87, 88]. For a better treatment yield, the assumption of plant sterols and vegetable stanols must occur during main meals. Thus the greater amount of alimentary cholesterol in the intestine and the biliary secretion induced by the cholesterol-containing meal, increase the hypocholesterolemic action [90]. Although the intake of phytosterol supplements is well tolerated and without significant side effects, their repeated intake can reduce the absorption of essential nutrients such as carotenoids and fat-soluble vitamins [91]. Thus, in patients undergoing nutraceutical treatments it is recommended to increase intake of food containing these nutrients.

4.3. Berberine

The plants of the *Berberis* species contain especially in the roots, but also in rhizomes and cortex, an interesting alkaloid known as berberine with documented hypocholesterolemic and hypoglycemic activity. Berberine seems to have different mechanisms of action, actually not fully understood. In particular, regarding its hypocholesterolemic activity, has been reported that berberine can reduce the levels of PCSK9 mRNA and at the same time promotes encoding of LDLR mRNA [92, 93]. As discussed above, PCSK9 promotes the internalization of LDLR from the hepatocyte surface into the cytosol and here their degradation by lysosomes occur. Thus, the action of berberine determine an increased expression of hepatic receptors with a higher LDL-C uptake.

Several studies have evaluated that the assumption of 0.5-1.5 g/day berberine may lead to a mean LDL-C reduction of 10-25% [94-96]. However, these results cannot be generalized because they have been predominantly conducted on Asian subjects, while its assumption in the Western population is always associated with RYR. In recent years, the numerous evidences support the role of nutraceuticals in the management of lipid profile especially as combinations of multiple compounds; this allowed to use different mechanisms of action that operate on the cholesterol levels, reducing the dosage of individual components without decreasing their effectiveness [97, 98]. Marazzi *et al.* have evaluated the efficacy of berberine when associated to RYR and polycosanols in elderly hypercholesterolemic patients that were intolerant to statins [99]. In these subjects treated with the combination of nutraceuticals, a reduction of about 20% of total cholesterol and over 30% of LDL-C was documented. Another interesting study have observed a reduction >32% of LDL-C in patients after co-administration of simvastatin and berberine, which in monotherapy reduced of 14.3% and 23.8%, respectively [100]. Also a significant reduction of triglycerides levels was found. This is the example of combination therapy that uses complementary actions of different agents.

In addition, different others beneficial properties of berberine have been reported such as reduction of triglyceride and glucose plasma levels as well as the increment of HDL-C concentration [93]. These actions could be particularly favorable in patients with metabolic syndrome, in which these alterations very often coexist. The hypoglycemic activity of berberine seems to be achieved through a dual mechanism: a reduction of intestinal glucose absorption and an increase of glucose uptake in muscle and liver [101]. Moreover, several studies both *in vitro* and *in vivo* has showed that berberine is able to exert vascular protective effect through an anti-oxidant and anti-inflammatory activity [102, 103].

However, although data related to efficacy of berberine-based compounds on hypocholesterolemic activity are encouraging, it is necessary to consider that the low bioavailability of berberine administered by *os* (<3%) can significantly reduce its effectiveness. For this reason, usually it is used in combination with other nutraceuticals including RYR and polycosanols.

4.4. Beta-Glucan

Beta-glucans are non-starch polysaccharides that consist of D-glucose units linked by β glycosidic bonds (1-3, 1-4 and 1-6 beta-D-glucans). Primary sources of β-glucans is represented by the cereal grains, particularly oats and barley, mushrooms and seaweeds. Due to their high molecular weight and high solubility in water, β-glucans are able to form viscous masses typical of soluble fiber (gel-forming). The mechanism of action of these polymers on the control of cholesterol is not fully understood; however, their anti-dyslipidemic effect seems to be achieved through a reduction in intestinal absorption and an increased fecal excretion of fats introduced with food [104].

It has been showed that β-glucans may reduce cholesterol and in particular LDL-C levels by 5-6% at dosages of about 3 g/day, without changing in HDL cholesterol [105-108]. As consequence, the consumption of cereal-based foods (representing the main source of β-glucans) may reduce the risk of develop cardiovascular diseases. Other dietary fibers such as psyllium or chitosan seems to have similar effect [109].

Similarly to soluble fibers as pectin, β-glucan in addition to the anti-dyslipidemic and anti-cholesterol action exert other beneficial metabolic effects of a prebiotic type, optimizing the growth and the selective development of certain bacterial strains. Moreover, it has been demonstrated the capacity of β-glucans to modulate circulating post-prandial glucose levels. This effect may results of particular interest in diabetic patients or in subject with impaired glucose tolerance [110]. At the same time, the gel-forming fiber can stimulate a sense of fullness resulting of particular interest in the course of dietary regimens required in patients with impaired glucose or lipid metabolism.

4.5. Other lipid-lowering Agents

In the recent years, other substances have been evaluated for their lipid-lowering capacity. Among these, the most important seems to be the soy (*Glycine max*) which represents an important source of fiber, phytosterols, isoflavone, lecithin and proteins. All these substances (phytocomplex) promote hypocholesterolemic effect through two distinct mechanisms: increased hepatic LDLR expression and reduced intestinal absorption of cholesterol. Data from clinical studies confirm that soy derivatives at a dose of 25-30 g/day for treatment periods between 4 weeks to one year lead to a reduction of 4.8 mg/dl (2-7%) [111-114].

Other compounds has attracted the attention of the scientific community for their lipid-lowering activity including several plant polyphenols such as flavonoids, epicatechin, anthocyanins, procyanidin or stilbenes (*i.e.* resveratrol) [115-118]. Although the effective mechanism of action on lipid metabolism is still unclear, they appear mainly to exert a competitive inhibition of HMG-CoA reductase [119]. Moreover, it has been demonstrated that resveratrol may exert a positive effect on LDLR gene expression, with a consequent reduction of cardiovascular risk [120]. Bondonno *et al.* have shown that also flavonoid-rich apples may influence cardiovascular health through the improvement of endothelial function and reduction of blood pressure [121]. Similarly, their strong antioxidant activity could help to inhibit LDL oxidation, playing an important cardioprotective role [122]. Another interesting study has

demonstrated that a particular cultivar of apple (*Malus pumila* Miller cv. Annurca) can be considered a functional food at a quantity of 200 g/day (2 apples) capable of decreasing total cholesterol and LDL-C by 8.3% and 14.5%, respectively [123]. The most considerable result is the simultaneous increase of HDL-C levels by 15.2%, since only few natural or synthetic substances have shown a similar capacity. Also, efficacy of supplementation with bergamot (*Citrus bergamia*) juice extract in patients with dyslipidemia for a short period (6 months) was observed [124]. In particular, a significant reduction of triglycerides, total and LDL cholesterol as well as an increase of HDL-C, was reported. The authors have attributed these beneficial effects to high amounts of flavonoids contained in bergamot fruit juice including neoeriocitrin, neohesperidin, naringin [124]. In addition to the compounds described above, other different plant-derivative agents through their high polyphenolic content may have beneficial effects on various risk factors of cardiovascular disease. It is the case of pomegranate, a fruit polyphenol-rich with different bioactivity as high anti-oxidant, anti-atherogenic, anti-hypertensive, and anti-inflammatory effects [125]. Several studies demonstrated that the assumption of pomegranate juice decrease significantly atherosclerotic lesion in patients with type 2 diabetes and reduce systolic blood pressure in atherosclerotic patients through the inhibition of serum angiotensin converting enzyme [126-128]. However, considering its controversial effect on the reduction of serum cholesterol in patients with hypercholesterolemia, pomegranate juice currently cannot be included in the category of hypocholesterolemic nutraceuticals [129-131].

Finally, also policosanols are included in the list of hypocholesterolemic agents. These compounds represent a mix of long chain aliphatic alcohols found in some vegetable sources such as sugarcane and potatoes. Several clinical trials have evaluated the efficacy of policosanols on patients with hypercholesterolemia in Cuba, showing encouraging clinical data. In particular, at daily dosage of policosanols between 5-10 mg, a significant decrease of total cholesterol (between 13-17%) and LDL-C (between 17-27%) was observed [132-134]. However, successive studies conducted on dyslipidemic patients outside Cuba, have shown that at various dosages (10-80 mg/day) their hypocholesterolemic activity remains extremely heterogeneous, although with a good tolerability profile [135]. It may depend on different sampling bias due to policosanol's origin, dosage and extraction method.

5. PROBIOTICS

An independent discussion deserve the probiotics, which have demonstrated a strong correlation between numerous systemic pathologies including alterations of the lipid profile and the loss of the qualitative-quantitative balance of different microbial species that harbor at intestinal level [136]. Several studies have emphasized the aspect that different probiotics can modulate the composition of intestinal microflora and act on some of the metabolic-cardiovascular risk factors such as hypercholesterolemia, obesity, arterial hypertension, and type 2 diabetes [137-141]. At the base of the lipid-lowering effect observed with probiotics there would be different mechanisms of action [142]. The first is related to the increased production of short-chain fatty acids, which include propionic and butyric acids with direct inhibitory effect on HMG-CoA reductase. Another important mechanism of probiotic strains is represented by enzymatic deconjugation of bile salts by bile-salt hydrolase. In particular, the hydrolysis of bile salts conjugated to glycine or taurine determines an increase in non-conjugated bile acids that are less soluble and reabsorbed less efficiently than the corresponding conjugates, resulting eliminated in the feces.

Numerous clinical studies have evaluated the effect of different probiotics predominantly inserted within a food matrix including fermented milk and yogurt also in association with prebiotics in subjects with normal or moderately increased cholesterol levels with a duration of treatment between 3 weeks [143] and 6 months [144]. However, the results of these studies appear in any case rather variable and modest reporting a reduction of total cholesterol of 0-5% and of LDL-C approximately of 0-10%, as well as a significant increase of HDL-C. Conversely, an interesting meta-analysis [145] observed that the intake of specific probiotics has a beneficial effect for total cholesterol and LDL-C, but report a neutral effect for HDL-C and triglycerides. Moreover, long-term (>4 week) probiotic supplementation resulted statistically more effective in reducing triglycerides and LDL-C compared to short-term (≤ 4-week) supplementation. Particular recommendations for the use of probiotics in patients with enterocolitis, diarrhea, inflammatory bowel disease, irritable bowel syndrome and *Clostridium difficile* diarrhea are needed [146].

CONCLUSION

Several scientific studies have confirmed that different lipid-lowering drugs such as statins reduce significantly cholesterol levels in both primary and secondary prevention, leading to a decrease in cardiovascular morbidity [24]. However, in clinical practice there are numerous conditions where conventional drugs should not be used for intolerance, cannot be prescribed, and also circumstances where not taken for fear of adverse events (nocebo effect) [30, 31]. For this last occurrence could be useful to comply with the patient's request by using natural substances, considered less harmful and more tolerated rather than completely renounce to pharmacological treatment of sure effectiveness.

Therefore, nutraceuticals could assume a role of "first actors" for some patients and in other cases effective support to conventional pharmacological therapies. In mild hypercholesterolemia subjects with low cardiovascular risk, nutraceuticals are indicated as pre-pharmacological treatment in addition to specific dietary and lifestyle interventions. The importance of physical activity in addition to a balanced nutritional intake in the reduction of elevated LDL-C levels as well as in the increase of low HDL-C levels, has already been highlighted [2]. Nutraceuticals may have also a health benefits in hypercholesterolemia patients with moderate-high cardiovascular risk, who did not reach LDL-C target in *add-on* therapy despite the maximum tolerated dose of statins assumed. Thus, during statin therapy is needed to choose nutraceuticals with mechanisms of action different to the inhibition of HMG-CoA reductase, to avoid the risk of serious adverse events.

Single nutraceutical compounds or their combination showed wide lipid-lowering efficacy (5%-25%), significantly below what is observed with statins in monotherapy or in association with ezetimibe. In particular, it has been observed that the treatment with RYR and berberine as single compound or in combination may impact favorably on LDL-C levels. Instead, the beneficial effects of the other nutraceutical compounds as phytosterols, β-glucan, niacin, soy and polyphenols in terms of LDL-C reduction, are widely debated. However, their benefits may exceed the hypocholesterolemic activity, extending also to other cardiovascular risk factors such as triglyceride reduction, increase of HDL-C levels, modulation of glucose metabolism or enhancing the sense of fullness (dietary fibers) useful in overweight/obese patients. The use of probiotics in patients with hypercholesterolemia is limited by their weak effect in the reduction of LDL-C, but their action at the level of intestinal microbiota could improve the efficacy of other drugs/nutraceuticals.

Currently, numerous combinations of nutraceuticals are commercially available. The purpose of these hypocholesterolemic co-formulations is to use different mechanisms of action in a synergistic-additive approach, reducing both the single active compound dosages and the pill burden with a lower risk of adverse events. Moreover, considering that the efficacy of these treatments is dependent on their continued assumption, also nutraceuticals may be subject to problems of poor adherence. Thus, the development of nutraceutical formulations with high tolerability, which ensure full compliance to treatment, is particularly important.

Despite many studies seem to confirm the efficacy and safety of different nutraceuticals, there are still many open questions regarding their use. In particular, successive post-marketing nutritional studies (similar to pharmacovigilance for conventional drugs) aimed to describe any unexpected side effects, as well as drugs-food or other nutraceutical interactions, are needed. Another important aspect is widespread use of nutraceutical supplements available *online* that could induce patients to self-medication interrupting or discontinuing pharmacological therapies, as a consequence of simple advertising tips or recommendations of nutritionist without medical consensus. Furthermore, daily cost of nutraceutical supplements that are not reimbursed by the NHS, may be higher than statin with low intensity such as simvastatin (5-10 mg) and pravastatin (20 mg).

Recently, a new class of therapeutic agents based on RNA interference strategy has been developed, which cleaves and inactivates mRNA encoding for PCSK9 [147, 148]. Despite preliminary data are encouraging, further studies are needed to confirm the safety and efficacy of these oligonucleotide-based drugs.

In conclusion, the targeted use of nutraceuticals according to the clinical characteristics of the subject associated with a healthy lifestyle can help to prevent or limit the pharmacological approach. Future clinical trials conducted on large populations may clarify the aspects related to dosage, time treatment and safety of the various nutraceuticals, considering the possible synergy between nutraceuticals and lipid-lowering drugs. Furthermore, the re-modulation of the intestinal microbiota by specific probiotics capable of modulating positively the different components of cardiovascular risk could be a promising field of research.

CONSENT FOR PUBLICATION

Not applicable.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

REFERENCES

[1]   Napoli C, Cacciatore F. Novel pathogenic insights in the primary prevention of cardiovascular disease. Prog Cardiovasc Dis 2009; 51: 503-23.

[2]   Crimi E, Ignarro LJ, Cacciatore F, Napoli C. Mechanisms by which exercise training benefits patients with heart failure. Nat Rev Cardiol 2009; 6: 292-300.

[3]   Napoli C. Developmental mechanisms involved in the primary prevention of atherosclerosis and cardiovascular disease. Curr Atheroscler Rep 2011; 13: 170-5.

[4]   Giampaoli S, Palmieri L, Donfrancesco C, et al. Cardiovascular health in Italy. Ten-year surveillance of cardiovascular diseases and risk factors: Osservatorio Epidemiologico Cardiovascolare/Health Examination Survey 1998-2012. Eur J Prev Cardiol 2015; 22: 9-37.

[5]   Ference BA, Ginsberg HN, Graham I, et al. Low-density lipoproteins cause atherosclerotic cardiovascular disease. 1. Evidence from genetic, epidemiologic, and clinical studies. A consensus statement from the European Atherosclerosis Society Consensus Panel. Eur Heart J 2017; 38: 2459-72.

[6]   Neaton JD, Blackburn H, Jacobs D, et al. Serum cholesterol level and mortality findings for men screened in the Multiple Risk Factor Intervention Trial. Multiple Risk Factor Intervention Trial Research Group. Arch Intern Med. 1992; 152: 1490-500.

[7]   D'Agostino RB Sr, Vasan RS, Pencina MJ, et al. General cardiovascular risk profile for use in primary care: the Framingham Heart Study. Circulation 2008; 117: 743-53.

[8]   De Nicola L, Donfrancesco C, Minutolo R, et al. Prevalence and cardiovascular risk profile of chronic kidney disease in Italy: results of the 2008-12 National Health Examination Survey. Nephrol Dial Transplant 2015; 30: 806-14.

[9]   Sahebkar A, Watts GF. New LDL-cholesterol lowering therapies: pharmacology, clinical trials, and relevance to acute coronary syndromes. Clin Ther 2013; 35: 1082-98.

[10]   Liu C, Liu Q, Xiao X. Effectiveness and safety of combinational therapy compared with intensified statin monotherapy in patients with coronary heart disease. Exp Ther Med 2018; 15: 4683-8.

[11]   Sacks FM, Pfeffer MA, Moye LA, et al. The effect of pravastatin on coronary events after myocardial infarction in patients with average cholesterol levels. Cholesterol and Recurrent Events Trial investigators. N Engl J Med. 1996; 335: 1001-9.

[12]   Heart Protection Study Collaborative Group. MRC/BHF Heart Protection Study of cholesterol lowering with simvastatin in 20,536 high-risk individuals: a randomised placebo-controlled trial. Lancet 2002; 360: 7-22.

[13]   Wang W, Zhang B. Statins for the prevention of stroke: a meta-analysis of randomized controlled trials. PLoS One 2014; 9: e92388.

[14]   Gulizia MM, Colivicchi F, Ricciardi G, et al. AN-MCO/ISS/AMD/ANCE/ARCA/FADOI/GICR-IACPR/SICI-GISE/SIBioC/SIC/SICOA/   SID/SIF/SIMEU/SIMG/SIMI/SISA Joint Consensus Document on cholesterol and cardiovascular risk: diagnostic-therapeutic pathway in Italy. Eur Heart J Suppl 2017; 19: D3-D54.

[15]   Poli A, Barbagallo CM, Cicero AFG, et al. Nutraceuticals and functional foods for the control of plasma cholesterol levels. An intersociety position paper. Pharmacol Res 2018; 134: 51-60.

[16]   Johnston TP, Korolenko TA, Pirro M, Sahebkar A. Preventing cardiovascular heart disease: Promising nutraceutical and non-nutraceutical treatments for cholesterol management. Pharmacol Res 2017; 120: 219-25.

[17]   Napoli C, Sica V. Statin treatment and the natural history of atherosclerotic-related diseases: pathogenic mechanisms and the risk-benefit profile. Curr Pharm Des 2004; 10: 425-32.

[18]   Bonetti PO, Lerman LO, Napoli C, Lerman A. Statin effects beyond lipid lowering--are they clinically relevant? Eur Heart J 2003; 24: 225-48.

[19]   Law MR, Wald NJ, Rudnicka AR. Quantifying effect of statins on low density lipoprotein cholesterol, ischaemic heart disease, and stroke: systematic review and meta-analysis. BMJ 2003; 326: 1423.

[20]   Tiwari V, Khokhar M. Mechanism of action of anti-hypercholesterolemia drugs and their resistance. Eur J Pharmacol 2014; 741: 156-70.

[21]   Gazzerro P, Proto MC, Gangemi G, et al. Pharmacological actions of statins: a critical appraisal in the management of cancer. Pharmacol Rev 2012; 64: 102-46.

[22]   Ezad S, Cheema H, Collins N. Statin-induced rhabdomyolysis: a complication of a commonly overlooked drug interaction. Oxf Med Case Reports 2018; 2018: omx104.

[23]   Shaik AN, Bohnert T, Williams DA, Gan LL, LeDuc BW. Mechanism of drug-drug interactions between warfarin and statins. J Pharm Sci 2016; 105: 1976-86.

[24]   Stone NJ, Robinson JG, Lichtenstein AH, et al 2013 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: 2889-934.

[25]   Cholesterol Treatment Trialists' (CTT) Collaboration, Baigent C, Blackwell L, 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: 1670-81.

[26]   Cholesterol Treatment Trialists' (CTT) Collaboration, Fulcher J, O'Connell R, et al. Efficacy and safety of LDL-lowering therapy among men and women: meta-analysis of individual data from 174,000 participants in 27 randomised trials. Lancet 2015; 385: 1397-405.

[27]   Baigent C, Keech A, Kearney PM, et al. Efficacy and safety of cholesterol-lowering treatment: prospective meta-analysis of data from 90,056 participants in 14 randomised trials of statins. Lancet 2005; 366: 1267-78.

[28]   Townsend N, Nichols M, Scarborough P, Rayner M. Cardiovascular disease in Europe--epidemiological update 2015. Eur Heart J 2015; 36: 2696-705.

[29]   Karlson BW, Palmer MK, Nicholls SJ, Lundman P, Barter PJ. Doses of rosuvastatin, atorvastatin and simvastatin that induce equal reductions in LDL-C and non-HDL-C: Results from the VOYAGER meta-analysis. Eur J Prev Cardiol 2016; 23: 744-7.

[30]   Pasternak RC, Smith SC Jr, Bairey-Merz CN, *et al.* ACC/AHA/NHLBI clinical advisory on the use and safety of statins. Stroke 2002; 33: 2337-41.

[31]   Lieb W, Enserro DM, Larson MG, Vasan RS. Residual cardiovascular risk in individuals on lipid-lowering treatment: quantifying absolute and relative risk in the community. Open Heart 2018; 5: e000722.

[32]   Vidt DG, Cressman MD, Harris S, Pears JS, Hutchinson HG. Rosuvastatin-induced arrest in progression of renal disease. Cardiology 2004; 102: 52-60.

[33]   Davidson MH, Clark JA, Glass LM, Kanumalla A. Statin safety: an appraisal from the adverse event reporting system. Am J Cardiol 2006; 97: 32C-43C.

[34]   McKenney JM, Davidson MH, Jacobson TA, Guyton JR; National Lipid Association Statin Safety Assessment Task Force. Final conclusions and recommendations of the National Lipid Association Statin Safety Assessment Task Force. Am J Cardiol 2006; 97: 89C-94C.

[35]   Armitage J. The safety of statins in clinical practice. Lancet 2007; 370: 1781-90.

[36]   Sattar N, Preiss D, Murray HM, *et al.* Statins and risk of incident diabetes: a collaborative meta-analysis of randomised statin trials. Lancet 2010; 375: 735-42.

[37]   Preiss D, Seshasai SR, Welsh P, *et al.* Risk of incident diabetes with intensive-dose compared with moderate-dose statin therapy: a meta-analysis. JAMA 2011; 305: 2556-64.

[38]   Garcia-Calvo M, Lisnock J, Bull HG, *et al.* The target of ezetimibe is Niemann-Pick C1-Like 1 (NPC1L1). Proc Natl Acad Sci USA 2005; 102: 8132-7.

[39]   Cannon CP, Giugliano RP, Blazing MA, *et al.* Rationale and design of IMPROVE-IT (IMProved Reduction of Outcomes: Vytorin Efficacy International Trial): comparison of ezetimbe/simvastatin *versus* simvastatin monotherapy on cardiovascular outcomes in patients with acute coronary syndromes. Am Heart J 2008; 156: 826-32.

[40]   Thongtang N, Lin J, Schaefer EJ, *et al.* Effects of ezetimibe added to statin therapy on markers of cholesterol absorption and synthesis and LDL-C lowering in hyperlipidemic patients. Atherosclerosis 2012; 225: 388-96.

[41]   Davidson MH, McGarry T, Bettis R, *et al.* Ezetimibe coadministered with simvastatin in patients with primary hypercholesterolemia. J Am Coll Cardiol 2002; 40: 2125-34.

[42]   Cannon CP, Blazing MA, Giugliano RP, *et al.* Ezetimibe added to statin therapy after acute coronary syndromes. N Engl J Med 2015; 372: 2387-97.

[43]   Torimoto K, Okada Y, Mori H, *et al.* Efficacy of combination of Ezetimibe 10 mg and rosuvastatin 2.5 mg *versus* rosuvastatin 5 mg monotherapy for hypercholesterolemia in patients with type 2 diabetes. Lipids Health Dis 2013; 12: 137.

[44]   Fujisue K, Nagamatsu S, Shimomura H, *et al.* Impact of statin-ezetimibe combination on coronary atheroma plaque in patients with and without chronic kidney disease - Sub-analysis of PRECISE-IVUS trial. Int J Cardiol 2018; 268: 23-26.

[45]   Stanifer JW, Charytan DM, White J, *et al.* Benefit of ezetimibe added to simvastatin in reduced kidney function. J Am Soc Nephrol 2017; 28: 3034-43.

[46]   Writing Committee, Lloyd-Jones DM, Morris PB, *et al.* 2016 ACC expert consensus decision pathway on the role of non-statin therapies for LDL-cholesterol lowering in the management of atherosclerotic cardiovascular disease risk: a report of the american college of cardiology task force on clinical expert consensus documents. J Am Coll Cardiol 2016; 68: 92-125.

[47]   Lipka LJ. Ezetimibe: a first-in-class, novel cholesterol absorption inhibitor. Cardiovasc Drug Rev 2003; 21: 293-312.

[48]   Fitzgerald G, Kiernan T. PCSK9 inhibitors and LDL reduction: pharmacology, clinical implications and future perspectives. Expert Rev Cardiovasc Ther 2018; 16: 567-78.

[49]   Abifadel M, Varret M, Rabès JP, *et al.* Mutations in PCSK9 cause autosomal dominant hypercholesterolemia. Nat Genet 2003; 34: 154-6.

[50]   Do RQ, Vogel RA, Schwartz GG. PCSK9 Inhibitors: potential in cardiovascular therapeutics. Curr Cardiol Rep 2013; 15: 345.

[51]   Navarese EP, Kolodziejczak M, Schulze V, *et al.* Effects of proprotein convertase subtilisin/kexin type 9 antibodies in adults with hypercholesterolemia: a systematic review and meta-analysis. Ann Intern Med 2015; 163: 40-51.

[52]   Koren MJ, Lundqvist P, Bolognese M, *et al.* Anti-PCSK9 monotherapy for hypercholesterolemia: the MENDEL-2 randomized, controlled phase III clinical trial of evolocumab. Anti-PCSK9 monotherapy for hypercholesterolemia: the MENDEL-2 randomized, controlled phase III clinical trial of evolocumab. J Am Coll Cardiol 2014; 63: 2531-40.

[53]   Robinson JG, Nedergaard BS, Rogers WJ, *et al.* Effect of evolocumab or ezetimibe added to moderate- or high-intensity statin therapy on LDL-C lowering in patients with hypercholesterolemia: the LAPLACE-2 randomized clinical trial. JAMA 2014; 311: 1870-82.

[54]   Stroes E, Colquhoun D, Sullivan D, *et al.* Anti-PCSK9 antibody effectively lowers cholesterol in patients with statin intolerance: the GAUSS-2 randomized, placebo-controlled phase 3 clinical trial of evolocumab. J Am Coll Cardiol 2014; 63: 2541-8.

[55]   Karatasakis A, Danek BA, Karacsonyi J, *et al.* Effect of PCSK9 inhibitors on clinical outcomes in patients with hypercholesterolemia: a meta-analysis of 35 randomized controlled trials. J Am Heart Assoc 2017; 6.

[56]   Rosenson RS, Jacobson TA, Preiss D, *et al.* Efficacy and safety of the PCSK9 inhibitor evolocumab in patients with mixed hyperlipidemia. Cardiovasc Drugs Ther 2016; 30: 305-13.

[57]   Kereiakes DJ, Robinson JG, Cannon CP, *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: 906-15.

[58]   Waters DD, Hsue PY, Bangalore S. PCSK9 Inhibitors for Statin Intolerance? JAMA 2016; 315: 1571-2.

[59]   Schreml J, Gouni-Berthold I. Role of anti-PCSK9 antibodies in the treatment of patients with statin intolerance. Curr Med Chem 2018; 25: 1538-48.

[60]   Chaudhary R, Garg J, Shah N, Sumner A. PCSK9 inhibitors: A new era of lipid lowering therapy. World J Cardiol 2017; 9: 76-91.

[61]   Feingold KR, Moser A, Shigenaga JK, Grunfeld C. Inflammation stimulates niacin receptor (GPR109A/HCA2) expression in adipose tissue and macrophages. J Lipid Res 2014; 55: 2501-8.

[62]   Wang W, Basinger A, Neese RA, *et al.* Effect of nicotinic acid administration on hepatic very low density lipoprotein-triglyceride production. Am J Physiol Endocrinol Metab 2001; 280: E540-7.

[63]   Le Bloc'h J, Leray V, Chetiveaux M, *et al.* Nicotinic acid decreases apolipoprotein B100-containing lipoprotein levels by reducing hepatic very low density lipoprotein secretion through a possible diacylglycerol acyltransferase 2 inhibition in obese dogs. J Pharmacol Exp Ther 2010; 334: 583-9.

[64]   Pang J, Chan DC, Hamilton SJ, Tenneti VS, Watts GF, Barrett PH. Effect of niacin on high-density lipoprotein apolipoprotein A-I kinetics in statin-treated patients with type 2 diabetes mellitus. Arterioscler Thromb Vasc Biol 2014; 34: 427-32.

[65]   Lamon-Fava S, Diffenderfer MR, Barrett PH, *et al.* Extended-release niacin alters the metabolism of plasma apolipoprotein (Apo) A-I and ApoB-containing lipoproteins. Arterioscler Thromb Vasc Biol 2008; 28: 1672-8.

[66]   Zhang LH, Kamanna VS, Ganji SH, Xiong XM, Kashyap ML. Niacin increases HDL biogenesis by enhancing DR4-dependent transcription of ABCA1 and lipidation of apolipoprotein A-I in HepG2 cells. J Lipid Res 2012; 53: 941-50.

[67]   Bays HE, Shah A, Lin J, Sisk CM, Dong Q, Maccubbin D. Consistency of extended-release niacin /laropiprant effects on Lp(a), ApoB, non-HDL-C, Apo A1, and ApoB/ApoA1 ratio across patient subgroups. Am J Cardiovasc Drugs 2012; 12: 197-206.

[68]   Cenarro A, Puzo J, Ferrando J, *et al.* Effect of Nicotinic acid/Laropiprant in the lipoprotein(a) concentration with regard to baseline lipoprotein(a) concentration and LPA genotype. Metabolism 2014; 63: 365-71.

[69]   Julius U. Niacin as antidyslipidemic drug. Can J Physiol Pharmacol 2015; 93: 1043-54.

[70]   Ballantyne CM, Davidson MH, McKenney J, Keller LH, Bajorunas DR, Karas RH. Comparison of the safety and efficacy of a combination tablet of niacin extended release and simvastatin *vs.* simvastatin monotherapy in patients with increased non-HDL cholesterol (from the SEACOAST I study). Am J Cardiol 2008; 101: 1428-36.

[71]   Karas RH, Kashyap ML, Knopp RH, Keller LH, Bajorunas DR, Davidson MH. Long-term safety and efficacy of a combination of niacin extended release and simvastatin in patients with dyslipidemia: the OCEANS study. Am J Cardiovasc Drugs 2008; 8: 69-81.

[72]   Toth PP, Thakker KM, Jiang P, Padley RJ. Niacin extended-release/simvastatin combination therapy produces larger favorable changes in high-density lipoprotein particles than atorvastatin monotherapy. Vasc Health Risk Manag 2012; 8: 39-44.

[73]   Song WL, FitzGerald GA. Niacin, an old drug with a new twist. J Lipid Res 2013; 54: 2586-94.

[74]   EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA). Scientific opinion on the substantiation of health claims related to eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA), docos-apentaenoic acid (DPA) and maintenance of normal cardiac function (ID 504, 506, 516, 527, 538, 703, 1128, 1317, 1324, 1325), maintenance of normal blood glucose concentrations (ID 566), maintenance of normal blood pressure (ID 506, 516, 703, 1317, 1324), maintenance of normal blood HDL-cholesterol concentrations (ID 506), maintenance of normal (fasting) blood concentrations of triglycerides (ID 506, 527, 538, 1317, 1324, 1325), maintenance of normal blood LDL-cholesterol concentrations (ID 527, 538, 1317, 1325, 4689), protection of the skin from photo-oxidative (UV-induced) damage (ID530), improved absorption of EPA and DHA (ID 522, 523), contribution to the normal function of the immune system by decreasing the levels of eicosanoids, arachidonic acid-derived mediators and pro-inflammatory cytokines (ID 520, 2914), and "immunomodulating agent" (4690) pursuant to Article 13(1) of Regulation (EC) No 1924/2006. EFSA Journal 2010; 8(10): 1796.

[75]   Ma J, Li Y, Ye Q, et al. Constituents of red yeast rice, a traditional Chinese food and medicine. J Agric Food Chem 2000; 48: 5220-5.

[76]   Gordon RY, Cooperman T, Obermeyer W, Becker DJ. Marked variability of monacolin levels in commercial red yeast rice products: buyer beware! Arch Intern Med 2010; 170: 1722-7.

[77]   Chen CH, Uang YS, Wang ST, Yang JC, Lin CJ. Interaction between red yeast rice and CYP450 enzymes/p-glycoprotein and its implication for the clinical pharmacokinetics of lovastatin. Evid Based Complement Alternat Med 2012; 2012: 127043.

[78]   Li Y, Jiang L, Jia Z, et al. A meta-analysis of red yeast rice: an effective and relatively safe alternative approach for dyslipidemia. PLoS ONE 2014; 9: e98611.

[79]   Heinz T, Schuchardt JP, Möller K, Hadji P, Hahn A. Low daily dose of 3 mg monacolin K from RYR reduces the concentration of LDL-C in a randomized, placebo-controlled intervention. Nutr Res 2016; 36: 1162-70.

[80]   Becker DJ, Gordon RY, Halbert SC, French B, Morris PB, Rader DJ. Red yeast rice for dyslipidemia in statin-intolerant patients: a randomized trial. Ann Intern Med 2009; 150: 830-9.

[81]   Bogsrud MP, Ose L, Langslet G, et al. HypoCol (red yeast rice) lowers plasma cholesterol - a randomized placebo controlled study. Scand Cardiovasc J 2010; 44: 197-200.

[82]   Verhoeven V, Lopez Hartmann M, Remmen R, Wens J, Apers S, Van Royen P. Red yeast rice lowers cholesterol in physicians - a double blind, placebo controlled randomized trial. BMC Complement Altern Med 2013; 13: 178.

[83]   Magno S, Ceccarini G, Pelosini C, et al. LDL-cholesterol lowering effect of a new dietary supplement: an open label, controlled, randomized, cross-over clinical trial in patients with mild-to-moderate hypercholesterolemia. Lipids Health Dis 2018; 17: 124.

[84]   Cicero AF, Derosa G, Parini A, et al. Red yeast rice improves lipid pattern, high-sensitivity C-reactive protein, and vascular remodeling parameters in moderately hypercholesterolemic Italian subjects. Nutr Res 2013; 33: 622-8.

[85]   Marangoni F, Poli A. Phytosterols and cardiovascular health. Pharmacol Res 2010; 61: 193-9.

[86]   Hallikainen M, Lyyra-Laitinen T, Laitinen T, et al. Endothelial function in hypercholesterolemic subjects: Effects of plant stanol and sterol esters. Atherosclerosis 2006; 188: 425-32.

[87]   Ras RT, Geleijnse JM, Trautwein EA. LDL-cholesterol-lowering effect of plant sterols and stanols across different dose ranges: a meta-analysis of randomised controlled studies. Br J Nutr 2014; 112: 214-9.

[88]   Ras RT, Fuchs D, Koppenol WP, et al. Effect of a plant sterol-enriched spread on biomarkers of endothelial dysfunction and low-grade inflammation in hypercholesterolaemic subjects. J Nutr Sci 2016; 5: e44.

[89]   AbuMweis SS, Marinangeli CP, Frohlich J, Jones PJ. Implementing phytosterols into medical practice as a cholesterol-lowering strategy: overview of efficacy, effectiveness, and safety. Can J Cardiol 2014; 30: 1225-32.

[90]   Sarkkinen E, Lyyra M, Nieminen S, Kuusisto P, Wester I. Cereal-based snack bar with added plant stanol ester (Benecol®) consumed between meals lowers serum total and LDL cholesterol effectively in mildly to moderately hypercholesterolemic subjects. Cholesterol 2018; 2018: 1463628.

[91]   Gylling H, Plat J, Turley S, et al. Plant sterols and plant stanols in the management of dyslipidaemia and prevention of cardiovascular disease. Atherosclerosis 2014; 232: 346-60.

[92]   Momtazi AA, Banach M, Pirro M, Katsiki N, Sahebkar A. Regulation of PCSK9 by nutraceuticals. Pharmacol Res 2017; 120: 157-69.

[93]   Cameron J, Ranheim T, Kulseth MA, Leren TP, Berge KE. Berberine decreases PCSK9 expression in HepG2 cells. Atherosclerosis 2008; 201: 266-73.

[94]   Kong W, Wei J, Abidi P, et al. Berberine is a novel cholesterol-lowering drug working through a unique mechanism distinct from statins. Nat Med 2004; 10: 1344-51.

[95]   Derosa G, D'Angelo A, Bonaventura A, Bianchi L, Romano D, Maffioli P. Effects of berberine on lipid profile in subjects with low cardiovascular risk. Expert Opin Biol Ther 2013; 13: 475-82.

[96]   Lan J, Zhao Y, Dong F, et al. Meta-analysis of the effect and safety of berberine in the treatment of type 2 diabetes mellitus, hyperlipemia and hypertension. J Ethnopharmacol 2015; 161: 69-81.

[97]   Gonnelli S, Caffarelli C, Stolakis K, Cuda C, Giordano N, Nuti R. Efficacy and tolerability of a nutraceutical combination (red yeast rice, policosanols, and berberine) in patients with low-moderate risk hypercholesterolemia: a double-blind, placebo-controlled study. Curr Ther Res Clin Exp 2014; 77:1-6.

[98]   Millán J, Cicero AF, Torres F, Anguera A. Effects of a nutraceutical combination containing berberine (BRB), policosanol, and red yeast rice (RYR), on lipid profile in hypercholesterolemic patients: A meta-analysis of randomised controlled trials. Clin Investig Arterioscler 2016; 28: 178-87.

[99]   Marazzi G, Cacciotti L, Pelliccia F, et al. Long-term effects of nutraceuticals (berberine, red yeast rice, policosanol) in elderly hypercholesterolemic patients. Adv Ther 2011; 28: 1105-13.

[100] Kong WJ, Wei J, Zuo ZY, et al. Combination of simvastatin with berberine improves the lipid-lowering efficacy. Metabolism 2008; 57: 1029-37.

[101] Liu C, Wang Z, Song Y, et al. Effects of berberine on amelioration of hyperglycemia and oxidative stress in high glucose and high fat diet-induced diabetic hamsters in vivo. Biomed Res Int 2015; 2015: 313808

[102] Meng S, Wang LS, Huang ZQ, et al. Berberine ameliorates inflammation in patients with acute coronary syndrome following percutaneous coronary intervention. Clin Exp Pharmacol Physiol 2012; 39: 406-11.

[103] Li Z, Geng YN, Jiang JD, Kong WJ. Antioxidant and anti-inflammatory activities of berberine in the treatment of diabetes mellitus. Evid Based Complement Alternat Med 2014; 2014: 289264.

[104] Thandapilly SJ, Ndou SP, Wang Y, Nyachoti CM, Ames NP. Barley β-glucan increases fecal bile acid excretion and short chain fatty acid levels in mildly hypercholesterolemic individuals. Food Funct 2018; 9: 3092-6.

[105] Reyna-Villasmil N, Bermúdez-Pirela V, Mengual-Moreno E, et al. Oat-derived beta-glucan significantly improves HDLC and diminishes LDLC and non-HDL cholesterol in overweight individuals with mild hypercholesterolemia. Am J Ther 2007; 14: 203-12.

[106] Keenan JM, Goulson M, Shamliyan T, Knutson N, Kolberg L, Curry L. The effects of concentrated barley beta-glucan on blood lipids in a population of hypercholesterolaemic men and women. Br J Nutr 2007; 97: 1162-8.

[107] Queenan KM, Stewart ML, Smith KN, Thomas W, Fulcher RG, Slavin JL. Concentrated oat beta-glucan, a fermentable fiber, lowers serum cholesterol in hypercholesterolemic adults in a randomized controlled trial. Nutr J 2007; 6: 6.

[108] Wolever TM, Gibbs AL, Brand-Miller J, et al. Bioactive oat β-glucan reduces LDL cholesterol in Caucasians and non-Caucasians. Nutr J 2011; 10: 130.

[109] Cicero AFG, Fogacci F, Colletti A. Food and plant bioactives for reducing cardiometabolic disease risk: an evidence based approach. Food Funct 2017; 8: 2076-88.

[110] Tessari P, Lante A. A multifunctional bread rich in beta glucans and low in starch improves metabolic control in type 2 diabetes: a controlled trial. Nutrients 2017; 9(3): E297.

[111]   Gardner CD, Messina M, Kiazand A, Morris JL, Franke AA. Effect of two types of soy milk and dairy milk on plasma lipids in hypercholesterolemic adults: a randomized trial. J Am Coll Nutr 2007; 26: 669-77.

[112]   Clerici C, Setchell KD, Battezzati PM, *et al*. Pasta naturally enriched with isoflavone aglycons from soy germ reduces serum lipids and improves markers of cardiovascular risk. J Nutr 2007; 137: 2270-8.

[113]   Wofford MR, Rebholz CM, Reynolds K, *et al*. Effect of soy and milk protein supplementation on serum lipid levels: a randomized controlled trial. Eur J Clin Nutr 2012; 66: 419-25.

[114]   Ramdath DD, Padhi EM, Sarfaraz S, Renwick S, Duncan AM. Beyond the cholesterol-lowering effect of soy protein: a review of the effects of dietary soy and its constituents on risk factors for cardiovascular disease. Nutrients 2017; 9(4): E324.

[115]   Baba S, Natsume M, Yasuda A, *et al*. Plasma LDL and HDL cholesterol and oxidized LDL concentrations are altered in normo- and hypercholesterolemic humans after intake of different levels of cocoa powder. J Nutr 2007; 137: 1436-41.

[116]   Zhu Y, Xia M, Yang Y, *et al*. Purified anthocyanin supplementation improves endothelial function via NO-cGMP activation in hypercholesterolemic individuals. Clin Chem 2011; 57: 1524-33.

[117]   Qin Y, Xia M, Ma J, *et al*. Anthocyanin supplementation improves serum LDL- and HDL-cholesterol concentrations associated with the inhibition of cholesteryl ester transfer protein in dyslipidemic subjects. Am J Clin Nutr 2009; 90: 485-92.

[118]   Tomé-Carneiro J, Visioli F. Polyphenol-based nutraceuticals for the prevention and treatment of cardiovascular disease: Review of human evidence. Phytomedicine 2016; 23: 1145-74.

[119]   Leopoldini M, Malaj N, Toscano M, Sindona G, Russo N. On the inhibitor effects of bergamot juice flavonoids binding to the 3-hydroxy-3-methylglutaryl-CoA reductase (HMGR) enzyme. J Agric Food Chem 2010; 58: 10768-73.

[120]   Yashiro T, Nanmoku M, Shimizu M, Inoue J, Sato R. Resveratrol increases the expression and activity of the low density lipoprotein receptor in hepatocytes by the proteolytic activation of the sterol regulatory element-binding proteins. Atherosclerosis 2012; 220: 369-74.

[121]   Bondonno CP, Yang X, Croft KD, *et al*. Flavonoid-rich apples and nitrate-rich spinach augment nitric oxide status and improve endothelial function in healthy men and women: a randomized controlled trial. Free Radic Biol Med 2012; 52: 95-102.

[122]   Dower JI, Geleijnse JM, Gijsbers L, Zock PL, Kromhout D, Hollman PC. Effects of the pure flavonoids epicatechin and quercetin on vascular function and cardiometabolic health: a randomized, double-blind, placebo-controlled, crossover trial. Am J Clin Nutr 2015; 101: 914-21.

[123]   Tenore GC, Caruso D, Buonomo G, *et al*. Annurca (Malus pumila Miller cv. Annurca) apple as a functional food for the contribution to a healthy balance of plasma cholesterol levels: results of a randomized clinical trial. J Sci Food Agric 2017; 97: 2107-15.

[124]   Toth PP, Patti AM, Nikolic D, *et al*. Bergamot reduces plasma lipids, atherogenic small dense LDL, and subclinical atherosclerosis in subjects with moderate hypercholesterolemia: a 6 months prospective study. Front Pharmacol 2016; 6: 299.

[125]   Basu A, Penugonda K. Pomegranate juice: a heart-healthy fruit juice. Nutr Rev 2009; 67: 49-56.

[126]   de Nigris F, Williams-Ignarro S, Lerman LO, *et al*. Beneficial effects of pomegranate juice on oxidation-sensitive genes and endothelial nitric oxide synthase activity at sites of perturbed shear stress. Proc Natl Acad Sci USA 2005; 102: 4896-901.

[127]   de Nigris F, Williams-Ignarro S, Botti C, Sica V, Ignarro LJ, Napoli C. Pomegranate juice reduces oxidized low-density lipoprotein downregulation of endothelial nitric oxide synthase in human coronary endothelial cells. Nitric Oxide 2006; 15: 259-63.

[128]   Aviram M, Rosenblat M, Gaitini D, *et al*. Pomegranate juice consumption for 3 years by patients with carotid artery stenosis reduces common carotid intima-media thickness, blood pressure and LDL oxidation. Clin Nutr 2004; 23: 423-33.

[129]   Esmaillzadeh A, Tahbaz F, Gaieni I, Alavi-Majd H, Azadbakht L. Concentrated pomegranate juice improves lipid profiles in diabetic patients with hyperlipidemia. J Med Food 2004; 7: 305-8.

[130]   Mirmiran P, Fazeli MR, Asghari G, Shafiee A, Azizi F. Effect of pomegranate seed oil on hyperlipidaemic subjects: a double-blind placebo-controlled clinical trial. Br J Nutr 2010; 104: 402-6.

[131]   Hosseini B, Saedisomeolia A, Wood LG, Yaseri M, Tavasoli S. Effects of pomegranate extract supplementation on inflammation in overweight and obese individuals: A randomized controlled clinical trial. Complement Ther Clin Pract 2016; 22: 44-50.

[132]   Más R, Castaño G, Illnait J, *et al*. Effects of policosanol in patients with type II hypercholesterolemia and additional coronary risk factors. Clin Pharmacol Ther 1999; 65: 439-47.

[133]   Castaño G, Más R, Fernández JC, Illnait J, Fernández L, Alvarez E. Effects of policosanol in older patients with type II hypercholesterolemia and high coronary risk. J Gerontol A Biol Sci Med Sci 2001; 56: M186-92.

[134]   Castaño G, Más R, Fernández L, *et al*. A comparison of the effects of D-003 and policosanol (5 and 10 mg/day) in patients with type II hypercholesterolemia: a randomized, double-blinded study. Drugs Exp Clin Res 2005; 31: 31-44.

[135]   Kassis AN, Jones PJ. Lack of cholesterol-lowering efficacy of Cuban sugar cane policosanols in hypercholesterolemic persons. Am J Clin Nutr 2006; 84: 1003-8.

[136]   Yoo JY, Kim SS. Probiotics and prebiotics: present status and future perspectives on metabolic disorders. Nutrients 2016; 8: 173.

[137]   Miele L, Giorgio V, Alberelli MA, De Candia E, Gasbarrini A, Grieco A. Impact of gut microbiota on obesity, diabetes, and cardiovascular disease risk. Curr Cardiol Rep 2015; 17: 120.

[138]   Bernini LJ, Simão AN, Alfieri DF, *et al*. Beneficial effects of Bifidobacterium lactis on lipid profile and cytokines in patients with metabolic syndrome: A randomized trial. Effects of probiotics on metabolic syndrome. Nutrition 2016; 32: 716-9.

[139]   Costabile A, Buttarazzi I, Kolida S, *et al*. An *in vivo* assessment of the cholesterol-lowering efficacy of Lactobacillus plantarum ECGC 13110402 in normal to mildly hypercholesterolaemic adults. PLoS ONE 2017; 12: e0187964.

[140]   Tonucci LB, Olbrich Dos Santos KM, Licursi de Oliveira L, Rocha Ribeiro SM, Duarte Martino HS. Clinical application of probiotics in type 2 diabetes mellitus: A randomized, double-blind, placebo-controlled study. Clin Nutr 2017; 36: 85-92.

[141]   Naito E, Yoshida Y, Kunihiro S, *et al*. Effect of Lactobacillus casei strain Shirota-fermented milk on metabolic abnormalities in obese prediabetic Japanese men: a randomised, double-blind, placebo-controlled trial. Biosci Microbiota Food Health 2018; 37: 9-18.

[142]   Thushara RM, Gangadaran S, Solati Z, Moghadasian MH. Cardiovascular benefits of probiotics: a review of experimental and clinical studies. Food Funct 2016; 7: 632-42.

[143]   Agerholm-Larsen L, Raben A, Haulrik N, Hansen AS, Manders M, Astrup A. Effect of 8 week intake of probiotic milk products on risk factors for cardiovascular diseases. Eur J Clin Nutr 2000; 54: 288-97.

[144]   Kiessling G, Schneider J, Jahreis G. Long-term consumption of fermented dairy products over 6 months increases HDL cholesterol. Eur J Clin Nutr 2002; 56: 843-9.

[145]   Shimizu M, Hashiguchi M, Shiga T, Tamura HO, Mochizuki M. Meta-analysis: effects of probiotic supplementation on lipid profiles in normal to mildly hypercholesterolemic individuals. PLoS ONE 2015; 10: e0139795.

[146]   Floch MH, Walker WA, Sanders ME, *et al*. Recommendations for probiotic use--2015 update: proceedings and consensus opinion. J Clin Gastroenterol 2015; 49: S69-73.

[147]   Fitzgerald K, White S, Borodovsky A, *et al*. A highly durable RNAi therapeutic inhibitor of PCSK9. N Engl J Med 2017; 376: 41-51.

[148]   Ray KK, Landmesser U, Leiter LA, *et al*. Inclisiran in Patients at High Cardiovascular Risk with Elevated LDL Cholesterol. N Engl J Med 2017; 376: 1430-40.