Review Article

High-density lipoprotein: Quality versus quantity in type 2 diabetes mellitus

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Abstract

Dyslipidaemia, inflammation and oxidative stress play an important role in the development of atherosclerosis and increased cardiovascular disease (CVD) risk in patients with type 2 diabetes mellitus (T2DM). High-density lipoprotein-cholesterol (HDL-C) correlates negatively with CVD risk. However, drugs aiming to increase HDL-C have failed to show a beneficial effect on CVD risk. Further research in this area showed the importance of HDL-associated proteins in relation to CVD risk. Diabetes mellitus leads to alteration in the quantity as well as the quality of HDL. The modifications in HDL proteins convert an antiatherogenic HDL to a proatherogenic HDL termed dysfunctional HDL. This review focuses on the normal functions of HDL-associated proteins and their alterations in T2DM.

Keywords: Cardiovascular disease, dysfunctional high-density lipoprotein, high-density lipoprotein, type 2 diabetes mellitus

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INTRODUCTION

Diabetes mellitus (DM) is a metabolic-cum-vascular disorder of various aetiologies which is characterised by chronic hyperglycaemia resulting from variations in insulin secretion, insulin action or both.[1] Its prevalence is rapidly and progressively rising due to the increase in average life expectancy, growing prevalence of obesity and westernisation of lifestyles in developing countries.[2,3] Individuals with diabetes have a two-to-four-fold increased risk of developing cardiovascular disease (CVD) compared with non-diabetic individuals[4,5] and are a major contributor to morbidity and mortality among patients with type 2 DM (T2DM).[6] Therefore, the reduction of cardiovascular risk is vital in people with type 2 diabetes.

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CARDIOVASCULAR DISEASE RISK IN TYPE 2 DIABETES MELLITUS

Patients with T2DM have several traditional and non-traditional risk factors which contribute to their CVD risk apart from the presence of diabetes itself. The traditional risk factors include the presence of obesity, cigarette smoking, physical inactivity, hypertension and dyslipidaemia.[7] Other non-traditional risk factors present in patients with T2DM include elevated levels of homocysteine, high sensitivity C-reactive protein (hs-CRP), fibrinogen and lipoprotein (a). A synergism is said to result between hyperglycaemia with other CVD risk factors. Improved glycaemic control is known to reduce the cardiovascular complications in patients with

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DM.[8] Universal guidelines recommend that diabetes should be considered a CVD risk equivalent and that intensive multifactorial intervention is required to treat all cardiovascular risk factors.[9]

DYSLIPIDAEMIA IN TYPE 2 DIABETES MELLITUS

Among the traditional risk factors, dyslipidaemia is more frequently present in patients with diabetes than age- and gender-matched non-diabetic individuals.[10,11] Diabetic dyslipidaemia is most often characterised by high plasma triglyceride (TG) concentration, low high-density lipoprotein-cholesterol (HDL-C) concentration and increased concentration of small dense low-density lipoprotein-cholesterol (LDL-C) levels.[12] This dyslipidaemia is a result of elevated free fatty acid release from insulin-resistant fat cells.[12] The excess free fatty acids get converted to TGs in the liver, whose increased production, in turn, stimulates very LDL-C (VLDL-C) and apolipoprotein B synthesis. The reduced activity of lipoprotein lipase in the insulin-deficient state may also contribute to elevated TG and VLDL-C levels.[12] The consequence of these elevated lipid fractions is increased small dense LDL-C levels and decreased HDL-C.

DIABETES MELLITUS AND HIGH-DENSITY LIPOPROTEIN-CHOLESTEROL

Studies have shown a positive correlation between LDL-C[13] and CVD and an inverse correlation between HDL-C and CVD.[14] HDL-C is considered as a protective molecule against atherosclerosis. The potent antiatherogenic properties of HDL particles originate from their unique composition and structure, i.e. HDL-associated proteins.

FUNCTIONAL HIGH-DENSITY LIPOPROTEIN

Proteins that are associated with HDL include apolipoproteins (apoA-I, apoA-II, apoA-IV, apoE and apoC); lipid transfer proteins including cholesterol ester transfer protein (CETP) and phospholipid (PL) transfer protein; enzymes (lecithin–cholesterol acyltransferase (LCAT), glutathione peroxidase, paraoxonase and platelet-activating factor-acetylhydrolase) and other minor proteins such as apoD and apoM. The lipids include 3%–15% TGs, 26%–46% PLs, 15%–30% cholesterol esters (CEs) and 2%–10% cholesterol.[15]

The antiatherogenic functions of HDL include reverse cholesterol transport (RCT), anti-inflammatory role and antioxidant role.[16] Other antiatherogenic functions of HDL include antiplatelet, antithrombotic and regulation of systemic insulin sensitivity and protection against endothelial dysfunction.[16]

The most important antiatherogenic effect of HDL is contributed by its RCT which takes place in three stages. The first stage involves efflux of cellular cholesterol to HDL which is mediated by three known intracellular cholesterol transporters, which are ATP-binding cassette transporter A1, ATP-binding cassette transporter G1 and scavenger receptor class B type I. The second step involves esterification of cholesterol in HDL to cholesteryl esters by LCAT, thereby creating a concentration gradient for uptake of more cholesterol into HDL. In the third stage, the HDL molecule with CE in its core is carried to the liver where CE is finally excreted into bile.

HDL proteins important for its RCT function are thus apoA-I, LCAT and CETP. Further, this efficiency is dependent on the function of apoA-I. ApoA-I, the major HDL-associated protein, consists of 243 amino acids[17] and is encoded by apoA gene located on chromosome 11.[17] ApoA-I structure–function relationship has been studied and has shown a decreased capacity of apoA-I mutants in promoting cellular cholesterol efflux. Mutations in apoA-I affect the secondary and tertiary structure of the C-terminal domain of apoA-1 and thus the cholesterol efflux capacity of HDL. These mutations include Pro165 → Arg,[18] Arg173 → Cys[19] and Leu141 → Arg.[20] Two other natural variants of apoA-1 with the C-terminal mutations (Glu198 → Lys and Glu235 → 0) show differential effects. While Glu235 → 0 was shown to have a 54% reduced ability to efflux cholesterol (94),[21] Glu198 → Lys did not affect cholesterol efflux function of apoA-I (93).[18] The apoA-IMilano dimer has an increased ability to promote efflux from macrophages and Fu5AH compared to wild type.[22]

Human LCAT (EC 2.3.1.43) is a 63-kDa lipoprotein-associated enzyme consisting of 416 amino acids. It is encoded by LCAT gene located on 16q22. LCAT gene is a 4.5-kb long and contains 6 exons comprising a 1.5-kb coding sequence.[23] It is synthesised in the liver and to a small extent in other tissues, such as brain and testes.[23] LCAT is secreted into the plasma where it circulates reversibly bound to lipoprotein particles or in a lipid-free form.

LCAT is involved in the esterification of free cholesterol to cholesteryl esters in circulating plasma lipoproteins, especially in HDL.[23] It is capable of binding lipids directly. However, the reaction requires activation by exchangeable apoproteins mainly apoA-I. The regions in apoA-1 associated

with LCAT activation is the helix 144–165 with secondary contribution by helix 166–186 region. It acts preferentially on the discoidal, nascent, pre-β1-HDL particles, containing apoA-I, where it esterifies free cholesterol via α-LCAT activity.[23] This esterification involves a transfer of the sn-2 fatty acyl group of phosphatidylcholine to the 3-β hydroxyl group of free cholesterol, forming a cholesteryl ester and lysophosphatidylcholine. The cholesteryl esters are then incorporated into the HDL particles, resulting in the formation of the spherical, mature, α-migrating forms of HDL. LCAT plays an important role in the reverse cholesterol function, thus transferring excess free cholesterol from peripheral tissues to the liver for biliary excretion.

Cholesteryl ester transfer protein (CETP) is a glycoprotein responsible for the transfer of cholesteryl esters from HDL to VLDL, LDL and intermediate-density lipoprotein (IDL) in exchange for TGs. About two-thirds of cholesteryl esters from HDL are transferred to the liver by CETP, whereas the remaining one-thirds are transferred by scavenger receptor class B type 1 (SR-B1). It is composed of 476 amino acids with a molecular weight of ~53 kDa.[24] Its structure resembles a banana-shaped conformation composed of amino (N)-and carboxy (C)-terminal β-barrels. A central β-sheet lies between the two β-barrels which is said to accommodate two CE molecules which is plugged by an amphiphilic helix 'X' (Glu465-Ser476) present at the C-terminus.[25] Mutations (linker insertion mutations, i.e., insertion of two nucleotides) in the N-terminal end at residues 48, 53, 165, 373 and 379 could impair lipid transfer activity of CETP.[26]

PL transfer protein (PLTP) is a glycoprotein with 476 amino acids and a molecular weight of 81 kDa synthesised in the placenta, pancreas, lung, kidney, heart, liver, skeletal muscle and brain.[27] PLTP-mediated PL transfer between HDL subfractions converts HDL3 into larger HDL particles with loss of apoA-I leading to the generation of nascent HDL particles which lead to its accelerated uptake by the liver. PLTP is also considered as a positive acute-phase reactant with a role in the innate immune defence.[28]

OTHER PROTEINS

Paraoxonase

Paraoxonase-1 (PON-1) is a 45-kDa glycoprotein composed of 355 amino acids and is encoded by PON gene located on chromosome 7 (q21.22).[29] PON-1 is synthesised in the liver and secreted into the blood where it is associated with apoA-I of HDL particles. Paraoxonase exhibits various types of hydrolytic activities which include lactonase activity, arylesterase activity and organophosphatase activity. Associated with HDL, it exhibits antioxidant and antiatherogenic properties by inhibiting lipoprotein oxidation and inactivating the toxic peroxidation products (ox-LDL) and also prevents their accumulation. Paraoxonase by way of its antioxidant nature also protects LDL and HDL against oxidative damage and prevents the formation of atherogenic ox-LDL molecules.[29] It enhances cholesterol efflux from macrophages through HDL. PON-1 also inhibits the transformation of monocytes into macrophages and thus inhibits the process of foam cell formation and thus reduces atherosclerotic plaques.[29]

Platelet-activating factor-acetylhydrolase

Platelet-activating factor-acetylhydrolase (PAF-AH) belongs to a small family of related phospholipases A2 that hydrolyse the sn-2 acetyl residue of platelet-activating factor and inactivates it. Platelet-activating factor is an important inflammatory mediator which activates cells at picomolar concentration.[30] Macrophages contribute to the largest amount of circulating enzyme. About two-thirds of the plasma enzyme is associated with LDL, whereas only one-third is associated with HDL. The HDL-associated PAF-AH is not capable of hydrolysing PAF owing to the low levels of PAF present under normal physiological conditions. This has been attributed also to the physical location of PAF-AH in HDL molecule which limits the access of PAF to the enzyme's active site.[31] The enzyme rapidly transfers between LDL and HDL particles, and since only about one in a thousand LDL particles carries a molecule of PAF-AH, HDL may function to distribute the enzyme among individual lipoprotein particles. By inactivating PAF, PAF-AH suppresses inflammatory signalling and prevents the foam cell formation, thereby decreasing atherogenicity. This is brought about by removal of the oxidatively truncated PLs which prevent oxidative modification of the lipoprotein particles which are taken up by vascular cells to initiate foam cell formation.[32]

Glutathione peroxidase

Glutathione peroxidases (GPXs) are a group of important enzymes possessing antioxidant property through glutathione involved in neutralising peroxides involved in oxidative damage to cells. The type of GPXs identified include cytosolic (cGPX or GPX1), gastrointestinal (GI-GPX or GPX2), plasma (pGPX or GPX3), PL hydroperoxide (PHGPX or GPX4) glutathione peroxidase, GPX5 and GPX6.[33] GPX4 is associated with HDL and LDL.[34] Activation of GPX4 has been identified as an important anti-inflammatory target.[35]

Acute-phase response proteins

Certain positive acute-phase proteins elevated during acute inflammation constitute a class of HDL-associated proteins. These include serum amyloid A, fibrinogen, alpha-1-acid glycoprotein 2 and alpha-2-HS glycoprotein. On the other hand, apoproteins such as apoA-I and apoA-IV are reduced during inflammation and can be considered as negative acute-phase response proteins.

Complement components

Complement components such as complement 3 (C3)- and C4b-binding protein associate with HDL. While C3 is an activator of the classical and alternative activation pathways, C4 is an activator of the classical pathway. C9, a subunit of the membrane attack complex, is also associated with HDL. Vitronectin, a protein which is an inhibitor of the membrane damaging effect of cytolytic pathway, also associates with HDL.

Serine proteinases

Serine proteinases serve as regulators of important processes such as inflammation, coagulation, angiogenesis and matrix modelling. Some of these serine proteinases such as alpha-1-antitrypsin circulate exclusively in HDL, whereas alpha-2-antiplasmin circulates partly in HDL. HDL also carries proteins such as inter-alpha-trypsin inhibitor heavy chain H4 and bikunin which are components of the inter-alpha-trypsin inhibitors. Similarly, HDL carries proteins such as haptoglobin-related protein, kininogen-1, prothrombin, angiotensinogen and procollagen C-proteinase enhancer-2.[36,37]

Other minor proteins

HDL acts as a carrier for other proteins having diverse functions such as retinol-binding protein, serotransferrin, transthyretin, hemopexin, albumin, Vitamin D-binding protein, platelet basic protein, Wnt signalling molecules and progranulin.[38]

DYSFUNCTIONAL HIGH-DENSITY LIPOPROTEIN

In systemic inflammatory diseases such as diabetes, obesity, metabolic syndrome, rheumatic diseases and familial hypercholesterolemia, these antiatherogenic properties of HDL are altered.[15,39,40] This results in change in HDL from antiatherogenic to proatherogenic. The National Institutes of Health (NIH) described altered HDL as 'dysfunctional' with regard to the increased risk of developing atherosclerosis.[40] Increased CVD has been reported in individuals with high levels of HDL, thus suggesting that besides the quantity, the composition and the functional behaviour of HDL play a significant role in contributing to the CVD risk.[15] Dysfunctional HDL has been shown to correlate clinically with increased risk of atherosclerosis.[40,41] Thus, the quantitative measurement of HDL may not accurately represent the protective effects of HDL cholesterol.

DIABETES AND HIGH-DENSITY LIPOPROTEIN

Studies have shown diabetes to have an effect on both the quality and the quantity of HDL. These alterations are seen both in the lipid component and in the protein component [Table 1].

LIPID CHANGES IN HIGH-DENSITY LIPOPROTEIN ASSOCIATED WITH DIABETES MELLITUS

The qualitative changes in lipids are also accompanied by kinetic changes occurring in the lipidome of HDL. The lipid alterations in HDL include an increase in TG content along with a decrease in PL content.[46] In patients with diabetes, insulin deficiency leads to increased lipolysis which, in turn, leads to increased concentration of free fatty acids returning to the liver which are packed in VLDL as TGs.[57] Insulin deficiency further facilitates this process

Table 1: HDL proteins and functional alterations in type 2 diabetes mellitus

HDL protein	Normal function	Alteration in type 2 diabetes mellitus	Function in type 2 diabetes mellitus
Apoprotein A-I	Cholesterol efflux	Oxidative modification,[42] glycation[43]	Impaired cholesterol efflux[44]
Apoprotein E	Interferes with LDL binding to proteoglycans in the vessel wall[45]	Decreased[46]	Proatherogenic[46]
Apoprotein M	Increases endothelial NO production, inhibits monocyte recruitment into intima	Decreased[46]	Proatherogenic[46]
PLTP	Phospholipid transfer between HDL subfractions converts HDL3 into larger HDL particles[38]	Increased[47,48]	Proatherogenic[49]
CETP	Transfer of cholesteryl esters from HDL to VLDL, LDL and IDL in exchange for triglycerides[38]	Increased[50-52]	Proatherogenic[52]
LCAT	Esterification of free cholesterol to cholesteryl esters in circulating plasma lipoproteins, especially HDL[23]	Levels increased, activity decreased,[53] glycation[54]	Proatherogenic[54,55]
PON-1	Inhibits lipoprotein oxidation and inactivating the toxic peroxidation products[29]	Decreased[46]	Pro-oxidant[56]

HDL=High-density lipoprotein; PLTP=Phospholipid transfer protein; CETP=Cholesterol efflux transfer protein; LCAT=Lecithin-cholesterol transfer protein; PON-1=Paraoxonase-1; LDL=Low-density lipoprotein; VLDL=Very-low-density lipoprotein; IDL=Intermediate-density lipoprotein

due to decreased catabolism of apoB-100 which, in turn, aides VLDL synthesis. Increased VLDL drives exchanges of TGs in VLDL for cholesteryl esters present in HDL due to increased activity of CETP.[58,59]

The kinetic changes include an increase in the catabolism of HDL. A decrease in cholesteryl esters is measured clinically as a decrease in HDL-C levels. The TG in HDL is a substrate for plasma lipases and hepatic lipase which is thus responsible for the increased catabolism of HDL.[60,61]

CHANGES IN PROTEINS ASSOCIATED WITH DM

Apoproteins

Hyperglycaemia seen in patients with DM can cause glycation of HDL proteins and their oxidative modification [Table 1].[62,63] Oxidative stress seen in patients with DM has been shown to cause oxidative modification of both apoA-I[42] and apoA-II.[64] Oxidation of methionine residue at 148 impairs the reverse cholesterol function of HDL.[44] Glycation of apoA-I has been shown to be associated with severity of CAD and plaque progression in patients with T2DM.[43]

ApoA-II is the second most predominant apoprotein in HDL. DM has been shown to modify the proteoforms of HDL as well as cause oxidative modification of methionine residues. The proportion of oxidised to native forms of apoA-II was shown to be increased in patients with DM. Oxidative modification of the methionine residues leads to alteration in the helical structure of the protein which, in turn, affects its binding to the lipid. This altered lipid binding has been shown to decrease the lipid-clearing ability of HDL.[65]

Using proteomic analysis, Gordon *et al.* showed changes in 7 of the 45 proteins in HDL. Changes were noted in apoA-II, apoE and paraoxonase. The authors noted a five-fold decrease in peptide counts of apoE [Table 1].[46] ApoE has been shown to be associated with antiatherogenic property of HDL as it interferes with LDL binding to proteoglycans in the vessel wall.[45] This could thus prevent atherosclerosis. A decrease in apoE in larger HDL molecules could thus be proatherogenic by facilitating LDL binding to proteoglycans in the vessel wall and promoting atherosclerosis.

Another antiatherogenic apoprotein altered in DM is apoM.[46] ApoM was reported to enhance sphingosine-1-phosphate content of HDL. Sphingosine-1-phosphate has been shown to induce endothelial nitric oxide production and thus favour arterial vasodilatation.[66] It has also been shown to inhibit monocyte recruitment into the intima. Both these effects contribute to the antiatherogenic effects of apoM. A decrease in apoM in patients with DM could thus contribute to the proatherogenic nature of altered HDL [Table 1].

Lipid transfer proteins

PLTP is considered to be proatherogenic. A positive association between PLTP activity and CAD has been shown in humans.[67] This is supported by studies in animal models which show a proatherogenic effect of PLTP.[49] Increased PLTP levels have been shown in patients with T2DM [Table 1].[47,48]

The lipid transfer activity of CETP has been considered to be proatherogenic considering the fact that the molecules, i.e., HDL, and other lipoproteins, i.e. VLDL, LDL and IDL, are subjected to remodelling by the activity of hepatic lipase which is enhanced in the presence of insulin resistance seen in patients with T2DM to more atherogenic particles, i.e., the smaller and denser HDL particles which are cleared from circulation by the liver receptors or by the kidney and the small dense LDL particles which are not taken up by LDL receptors but can be taken up by the macrophages in the subendothelial spaces, thereby promoting plaque formation [Table 1]. The CE resulting from action of LCAT in HDL is thus channelled to other lipoproteins rather than being taken up by the liver through SR-B1 or LDL receptors.[50-52]

LCAT is crucial for the maintenance and maturation of normal HDL metabolism. LCAT activity has been shown to be decreased in patients with T2DM along with an increase in LCAT levels.[53] A study showed that glycaemia-induced glycation of HDL is a strong predictor of decrease in LCAT activity in patients with DM [Table 1].[54]

Other proteins

HDL from diabetics was deficient in their ability to promote endothelial progenitor cell-dependent endothelial repair, increase endothelial nitric oxide expression and produce endothelium-dependent relaxation. These defects were associated with raised HDL lipid peroxidation and MPO content and could be improved with niacin therapy.[55] Antioxidative activity of small dense HDL (HDL_3) has been shown to be defective as evidenced by lower paraoxonase/arylesterase 1 (PON-1) and platelet activating factor acetylhydrolase (PAF-AH) activities in diabetic patients compared to controls.[68] A recent study conducted in young female patients with type 2 diabetes showed them to have a higher concentration of inflammatory molecule serum amyloid A

in the HDL fractions HDL_2 and HDL_3 along with higher PON-1 activity and CETP activity providing evidence of dysfunctional HDL due to inflammation.[56] This suggests that variation in HDL features may contribute to the loss of the cardioprotective properties in pre-menopausal women with T2DM.

HIGH-DENSITY LIPOPROTEIN AS TARGET FOR TREATMENT

The target for treating diabetic dyslipidaemia is a reduction of LDL-C levels. For this purpose, statins are the drugs of choice. Apart from achieving target lipid levels, statins have been shown to have a beneficial effect on both HDL quantity and quality. Statins have a beneficial role in improving the RCT function of HDL by increasing the cholesterol efflux capacity.[69] However, the results with respect to the effect of statins on apoA-1 levels have not been consistent.[70-72] While a few studies have shown the beneficial effect when used alone, others have shown statins to be effective in increasing apoA-1 levels only when used in combination with other agents (carnitine and ezetimibe)[71,72] some studies observed no change.[73,74] The differential results can be attributed to the dose of statin used, the duration as well as the number of subjects studied. Similarly, the results are mixed with respect to the effect of statins on LCAT activity with a few studies showing a decrease in activity,[55] whereas still others observed no change.[75]

Although statins help achieve target lipid levels in majority of the patients, some patients fail to achieve LDL-C targets. Such patients benefit from addition of non-statin drugs.[76] These drugs have an additional effect on HDL-C levels. Some of the drugs which target HDL include fibrates which also decrease serum TG levels along with increasing HDL-C levels through activation of peroxisome proliferator-activated receptor-α and are useful in patients with mixed dyslipidaemia with residual CVD risk.[76] Cholesterol ester transfer protein (CETP) inhibitors which inhibit CETP, an enzyme involved in exchange of cholesterol esters for TGs between HDL and VLDL/LDL is another drug target.[77] Niacin has been shown to be useful when used alone or in combination in patients with T2DM in a meta-analysis. However, close monitoring of fasting blood glucose is required as it tends to increase fasting blood glucose (FBG) levels.[78] Thus, drug targets which can modify the functions of HDL rather than the quantity could be useful in decreasing CVD risk in patients with DM.

Normal HDL molecule is antiatherogenic owing to its structural proteins which exhibit important functions such as reverse cholesterol function, anti-inflammatory and antioxidant activities. DM is associated with modifications in some of the important antiatherogenic proteins owing to the inflammatory and oxidative milieu along with elevated glucose levels which promote glycation of the proteins, thereby converting them to proatherogenic molecules. Drugs targeting HDL proteins and thus its function could be of use in decreasing CVD risk in patients with T2DM.

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REFERENCES

1. Kota SK, Meher LK, Jammula S, Kota SK, Krishna SV, Modi KD. Aberrant angiogenesis: The gateway to diabetic complications. Indian J Endocrinol Metab 2012;16:918-30.
2. Wild S, Roglic G, Green A, Sicree R, King H. Global prevalence of diabetes: Estimates for the year 2000 and projections for 2030. Diabetes Care 2004;27:1047-53.
3. Hossain P, Kawar B, El Nahas M. Obesity and diabetes in the developing world – a growing challenge. N Engl J Med 2007;356:213-5.
4. Kannel WB. Lipids, diabetes, and coronary heart disease: Insights from the Framingham study. Am Heart J 1985;110:1100-7.
5. Stamler J, Vaccaro O, Neaton JD, Wentworth D. Diabetes, other risk factors, and 12-yr cardiovascular mortality for men screened in the multiple risk factor intervention trial. Diabetes Care 1993;16:434-44.
6. Authors/Task Force Members, Rydén L, Grant PJ, Anker SD, Berne C, Cosentino F, *et al*. ESC guidelines on diabetes, pre-diabetes, and cardiovascular diseases developed in collaboration with the EASD: The task force on diabetes, pre-diabetes, and cardiovascular diseases of the European Society of Cardiology (ESC) and developed in collaboration with the European Association for the Study of Diabetes (EASD). Eur Heart J 2013;34:3035-87.
7. Gupta S, Gudapati R, Gaurav K, Bhise M. Emerging risk factors for cardiovascular diseases: Indian context. Indian J Endocrinol Metab 2013;17:806-14.
8. Martín-Timón I, Sevillano-Collantes C, Segura-Galindo A, Del Cañizo-Gómez FJ. Type 2 diabetes and cardiovascular disease: Have all risk factors the same strength? World J Diabetes 2014;5:444-70.
9. De Backer G, Ambrosioni E, Borch-Johnsen K, Brotons C, Cifkova R, Dallongeville J, *et al*. European guidelines on cardiovascular disease prevention in clinical practice. Third joint task force of European and other societies on cardiovascular disease prevention in clinical practice. Eur Heart J 2003;24:1601-10.
10. Krauss RM. Lipids and lipoproteins in patients with type 2 diabetes. Diabetes Care 2004;27:1496-504.
11. Elboudwarej O, Hojjat H, Safarpoor S, Vazirian S, Ahmadi S. Dysfunctional HDL and cardiovascular disease risk in individuals with diabetic dyslipidemia. J Diabetes Metab 2011;S4:1-9.
12. Chehade JM, Gladysz M, Mooradian AD. Dyslipidemia in type 2 diabetes: Prevalence, pathophysiology, and management. Drugs 2013;73:327-39.
13. Glass CK, Witztum JL. Atherosclerosis. The road ahead. Cell 2001;104:503-16.
14. D'Agostino RB Sr., Vasan RS, Pencina MJ, Wolf PA, Cobain M, Massaro JM, *et al*. General cardiovascular risk profile for use in primary care: The Framingham heart study. Circulation 2008;117:743-53.
15. Eren E, Yilmaz N, Aydin O. High density lipoprotein and it's dysfunction. Open Biochem J 2012;6:78-93.
16. Guo ZG, Li C, Zhong JK, Tu Y, Xie D. Laboratory investigation of

dysfunctional HDL. Chem Phys Lipids 2012;165:32-7.
17. Dominiczak MH, Caslake MJ. Apolipoproteins: Metabolic role and clinical biochemistry applications. Ann Clin Biochem 2011;48:498-515.
18. von Eckardstein A, Castro G, Wybranska I, Theret N, Duchateau P, Duverger N, *et al*. Interaction of reconstituted high density lipoprotein discs containing human apolipoprotein A-I (ApoA-I) variants with murine adipocytes and macrophages. Evidence for reduced cholesterol efflux promotion by apoA-I(Pro165-->Arg). J Biol Chem 1993;268:2616-22.
19. Bielicki JK, McCall MR, Stoltzfus LJ, Ravandi A, Kuksis A, Rubin EM, *et al*. Evidence that apolipoprotein A-IMilano has reduced capacity, compared with wild-type apolipoprotein A-I, to recruit membrane cholesterol. Arterioscler Thromb Vasc Biol 1997;17:1637-43.
20. Miccoli R, Zhu Y, Daum U, Wessling J, Huang Y, Navalesi R, *et al*. A natural apolipoprotein A-I variant, apoA-I (L141R) Pisa, interferes with the formation of alpha-high density lipoproteins (HDL) but not with the formation of pre beta 1-HDL and influences efflux of cholesterol into plasma. J Lipid Res 1997;38:1242-53.
21. Han H, Sasaki J, Matsunaga A, Hakamata H, Huang W, Ageta M. A novel mutant, apoA-I nichinan(Glu235-->0), is associated with low HDL cholesterol levels and decreased cholesterol efflux from cells. Arterioscler Thromb Vasc Biol 1999;19:1447-55.
22. Calabresi L, Canavesi M, Bernini F, Franceschini G. Cell cholesterol efflux to reconstituted high-density lipoproteins containing the apolipoprotein A-IMilano dimer. Biochemistry 1999;38:16307-14.
23. Rousset X, Vaisman B, Amar M, Sethi AA, Remaley AT. Lecithin: Cholesterol acyltransferase – From biochemistry to role in cardiovascular disease. Curr Opin Endocrinol Diabetes Obes 2009;16:163-71.
24. Drayna D, Jarnagin AS, McLean J, Henzel W, Kohr W, Fielding C, *et al*. Cloning and sequencing of human cholesteryl ester transfer protein cDNA. Nature 1987;327:632-4.
25. Qiu X, Mistry A, Ammirati MJ, Chrunyk BA, Clark RW, Cong Y, *et al*. Crystal structure of cholesteryl ester transfer protein reveals a long tunnel and four bound lipid molecules. Nat Struct Mol Biol 2007;14:106-13.
26. Wang S, Deng LP, Brown ML, Agellon LB, Tall AR. Structure-function studies of human cholesteryl ester transfer protein by linker insertion scanning mutagenesis. Biochemistry 1991;30:3484-90.
27. Föger B, Santamarina-Fojo S, Shamburek RD, Parrot CL, Talley GD, Brewer HB Jr. Plasma phospholipid transfer protein. Adenovirus-mediated overexpression in mice leads to decreased plasma high density lipoprotein (HDL) and enhanced hepatic uptake of phospholipids and cholesteryl esters from HDL. J Biol Chem 1997;272:27393-400.
28. Albers JJ, Vuletic S, Cheung MC. Role of plasma phospholipid transfer protein in lipid and lipoprotein metabolism. Biochim Biophys Acta 2012;1821:345-57.
29. Furlong CE, Marsillach J, Jarvik GP, Costa LG. Paraoxonases-1, -2 and -3: What are their functions? Chem Biol Interact 2016;259:51-62.
30. McIntyre TM, Prescott SM, Stafforini DM. The emerging roles of PAF acetylhydrolase. J Lipid Res 2009;50:S255-9.
31. Stafforini DM, McIntyre TM, Carter ME, Prescott SM. Human plasma platelet-activating factor acetylhydrolase. Association with lipoprotein particles and role in the degradation of platelet-activating factor. J Biol Chem 1987;262:4215-22.
32. Jurkovic S, Osredkar J, Marc J. Molecular impact of glutathione peroxidases in antioxidant processes. Biochem Med 2008;18:162-74.
33. Sattler W, Maiorino M, Stocker R. Reduction of HDL- and LDL-associated cholesterylester and phospholipid hydroperoxides by phospholipid hydroperoxide glutathione peroxidase and ebselen (PZ 51). Arch Biochem Biophys 1994;309:214-21.
34. Li C, Deng X, Xie X, Liu Y, Friedmann Angeli JP, Lai L. Activation of glutathione peroxidase 4 as a novel anti-inflammatory strategy. Front Pharmacol 2018;9:1120.
35. Vaisar T, Pennathur S, Green PS, Gharib SA, Hoofnagle AN, Cheung MC, *et al*. Shotgun proteomics implicates protease inhibition and complement activation in the antiinflammatory properties of HDL. J Clin Invest 2007;117:746-56.
36. Heinecke JW. The HDL proteome: A marker – And perhaps mediator – Of coronary artery disease. J Lipid Res 2009;50 Suppl: S167-71.
37. Navab M, Ananthramaiah GM, Reddy ST, Van Lenten BJ, Ansell BJ, Fonarow GC, *et al*. The oxidation hypothesis of atherogenesis: The role of oxidized phospholipids and HDL. J Lipid Res 2004;45:993-1007.
38. Kontush A, Lindahl M, Lhomme M, Calabresi L, Chapman MJ, Davidson WS. Structure of HDL: Particle subclasses and molecular components. In: von Eckardstein A, Kardassis D, editors. High Density Lipoproteins. Handbook of Experimental Pharmacology. Vol 224. Springer; 2015. p. 3-51.
39. Feng H, Li XA. Dysfunctional high-density lipoprotein. Curr Opin Endocrinol Diabetes Obes 2009;16:156-62.
40. Rosenson RS, Brewer HB Jr., Ansell BJ, Barter P, Chapman MJ, Heinecke JW, *et al*. Dysfunctional HDL and atherosclerotic cardiovascular disease. Nat Rev Cardiol 2016;13:48-60.
41. Ragbir S, Farmer JA. Dysfunctional high-density lipoprotein and atherosclerosis. Curr Atheroscler Rep 2010;12:343-8.
42. Yassine HN, Jackson AM, Reaven PD, Nedelkov D, Nelson RW, Lau SS, *et al*. The application of multiple reaction monitoring to assess apo A-I methionine oxidations in diabetes and cardiovascular disease. Transl Proteom 2014;4-5:18-24.
43. Pu LJ, Lu L, Zhang RY, Du R, Shen Y, Zhang Q, *et al*. Glycation of apoprotein A-I is associated with coronary artery plaque progression in type 2 diabetic patients. Diabetes Care 2013;36:1312-20.
44. Shao B, Cavigiolio G, Brot N, Oda MN, Heinecke JW. Methionine oxidation impairs reverse cholesterol transport by apolipoprotein A-I. Proc Natl Acad Sci U S A 2008;105:12224-9.
45. Umaerus M, Rosengren B, Fagerberg B, Hurt-Camejo E, Camejo G. HDL2 interferes with LDL association with arterial proteoglycans: A possible athero-protective effect. Atherosclerosis 2012;225:115-20.
46. Gordon SM, Davidson WS, Urbina EM, Dolan LM, Heink A, Zang H, *et al*. The effects of type 2 diabetes on lipoprotein composition and arterial stiffness in male youth. Diabetes 2013;62:2958-67.
47. Riemens SC, van Tol A, Sluiter WJ, Dullaart RP. Plasma phospholipid transfer protein activity is related to insulin resistance: Impaired acute lowering by insulin in obese type II diabetic patients. Diabetologia 1998;41:929-34.
48. Tan KC, Shiu SW, Wong Y, Tam S. Plasma phospholipid transfer protein activity and subclinical inflammation in type 2 diabetes mellitus. Atherosclerosis 2005;178:365-70.
49. Yang XP, Yan D, Qiao C, Liu RJ, Chen JG, Li J, *et al*. Increased atherosclerotic lesions in apoE mice with plasma phospholipid transfer protein overexpression. Arterioscler Thromb Vasc Biol 2003;23:1601-7.
50. Packard CJ, Shepherd J. Lipoprotein heterogeneity and apolipoprotein B metabolism. Arterioscler Thromb Vasc Biol 1997;17:3542-56.
51. Berneis KK, Krauss RM. Metabolic origins and clinical significance of LDL heterogeneity. J Lipid Res 2002;43:1363-79.
52. Chen GC, Liu W, Duchateau P, Allaart J, Hamilton RL, Mendel CM, *et al*. Conformational differences in human apolipoprotein B-100 among subspecies of low density lipoproteins (LDL). Association of altered proteolytic accessibility with decreased receptor binding of LDL subspecies from hypertriglyceridemic subjects. J Biol Chem 1994;269:29121-8.
53. Nakhjavani M, Morteza A, Karimi R, Banihashmi Z, Esteghamati A. Diabetes induces gender gap on LCAT levels and activity. Life Sci 2013;92:51-4.
54. Nakhjavani M, Esteghamati A, Esfahanian F, Ghanei A, Rashidi A, Hashemi S. HbA1c negatively correlates with LCAT activity in type 2 diabetes. Diabetes Res Clin Pract 2008;81:38-41.
55. Masana L, Cabré A, Heras M, Amigó N, Correig X, Martínez-Hervás S, *et al*. Remarkable quantitative and qualitative differences in HDL after niacin or fenofibrate therapy in type 2 diabetic patients. Atherosclerosis 2015;238:213-9.

56. Nakhjavani M, Imani M, Larry M, Aghajani-Nargesi A, Morteza A, Esteghamati A. Metabolic syndrome in premenopausal and postmenopausal women with type 2 diabetes: Loss of protective effects of premenopausal status. J Diabetes Metab Disord 2014;13:102.
57. Ginsberg HN. Diabetic dyslipidemia: Basic mechanisms underlying the common hypertriglyceridemia and low HDL cholesterol levels. Diabetes 1996;45 Suppl 3:S27-30.
58. Femlak M, Gluba-Brzózka A, Ciałkowska-Rysz A, Rysz J. The role and function of HDL in patients with diabetes mellitus and the related cardiovascular risk. Lipids Health Dis 2017;16:207.
59. Hayek T, Azrolan N, Verdery RB, Walsh A, Chajek-Shaul T, Agellon LB, *et al*. Hypertriglyceridemia and cholesteryl ester transfer protein interact to dramatically alter high density lipoprotein levels, particle sizes, and metabolism. Studies in transgenic mice. J Clin Invest 1993;92:1143-52.
60. Horowitz BS, Goldberg IJ, Merab J, Vanni TM, Ramakrishnan R, Ginsberg HN. Increased plasma and renal clearance of an exchangeable pool of apolipoprotein A-I in subjects with low levels of high density lipoprotein cholesterol. J Clin Invest 1993;91:1743-52.
61. Chatterjee C, Sparks DL. Hepatic lipase, high density lipoproteins, and hypertriglyceridemia. Am J Pathol 2011;178:1429-33.
62. Ferretti G, Bacchetti T, Nègre-Salvayre A, Salvayre R, Dousset N, Curatola G, *et al*. Structural modifications of HDL and functional consequences. Atherosclerosis 2006;184:1-7.
63. Ansell BJ, Fonarow GC, Fogelman AM. The paradox of dysfunctional high-density lipoprotein. Curr Opin Lipidol 2007;18:427-34.
64. Azizkhanian I, Trenchevska O, Bashawri Y, Hu J, Koska J, Reaven PD, *et al*. Posttranslational modifications of apolipoprotein A-II proteoforms in type 2 diabetes. J Clin Lipidol 2016;10:808-15.
65. Anantharamaiah GM, Hughes TA, Iqbal M, Gawish A, Neame PJ, Medley MF, *et al*. Effect of oxidation on the properties of apolipoproteins A-I and A-II. J Lipid Res 1988;29:309-18.
66. Karuna R, Park R, Othman A, Holleboom AG, Motazacker MM, Sutter I, *et al*. Plasma levels of sphingosine-1-phosphate and apolipoprotein M in patients with monogenic disorders of HDL metabolism. Atherosclerosis 2011;219:855-63.
67. Schlitt A, Bickel C, Thumma P, Blankenberg S, Rupprecht HJ, Meyer J, *et al*. High plasma phospholipid transfer protein levels as a risk factor for coronary artery disease. Arterioscler Thromb Vasc Biol 2003;23:1857-62.
68. Nobécourt E, Jacqueminet S, Hansel B, Chantepie S, Grimaldi A, Chapman MJ, *et al*. Defective antioxidative activity of small dense HDL3 particles in type 2 diabetes: Relationship to elevated oxidative stress and hyperglycaemia. Diabetologia 2005;48:529-38.
69. Sviridov D, Hoang A, Ooi E, Watts G, Barrett PH, Nestel P. Indices of reverse cholesterol transport in subjects with metabolic syndrome after treatment with rosuvastatin. Atherosclerosis 2008;197:732-9.
70. Save V, Patil N, Moulik N, Rajadhyaksha G. Effect of atorvastatin on type 2 diabetic dyslipidemia. J Cardiovasc Pharmacol Ther 2006;11:262-70.
71. Galvano F, Li Volti G, Malaguarnera M, Avitabile T, Antic T, Vacante M, *et al*. Effects of simvastatin and carnitine versus simvastatin on lipoprotein(a) and apoprotein(a) in type 2 diabetes mellitus. Expert Opin Pharmacother 2009;10:1875-82.
72. Uemura Y, Watarai M, Ishii H, Koyasu M, Takemoto K, Yoshikawa D, *et al*. Atorvastatin 10 mg plus ezetimibe 10mg compared with atorvastatin 20 mg: Impact on the lipid profile in Japanese patients with abnormal glucose tolerance and coronary artery disease. J Cardiol 2012;59:50-6.
73. Vergès B, Florentin E, Baillot-Rudoni S, Petit JM, Brindisi MC, Pais de Barros JP, *et al*. Rosuvastatin 20 mg restores normal HDL-apoA-I kinetics in type 2 diabetes. J Lipid Res 2009;50:1209-15.
74. Jung KY, Kim KM, Han SK, Yun HM, Oh TJ, Choi SH, *et al*. Effect of rosuvastatin on cholesterol efflux capacity and endothelial function in type 2 diabetes mellitus and dyslipidemia. Circ J 2018;82:1387-95.
75. Bhatnagar D, Durrington PN, Kumar S, Mackness MI, Dean J, Boulton AJ. Effect of treatment with a hydroxymethylglutaryl coenzyme A reductase inhibitor on fasting and postprandial plasma lipoproteins and cholesteryl ester transfer activity in patients with NIDDM. Diabetes 1995;44:460-5.
76. Filippatos TD, Florentin M, Georgoula M, Elisaf MS. Pharmacological management of diabetic dyslipidemia. Expert Rev Clin Pharmacol 2017;10:187-200.
77. Kosmas CE, DeJesus E, Rosario D, Vittorio TJ. CETP inhibition: Past failures and future hopes. Clin Med Insights Cardiol 2016;10:37-42.
78. Ding Y, Li Y, Wen A. Effect of niacin on lipids and glucose in patients with type 2 diabetes: A meta-analysis of randomized, controlled clinical trials. Clin Nutr 2015;34:838-44.