REVIEW ARTICLE

Drugs 2000 Jul; 60 (1): 55-93
0012-6667/00/0007-0055/$25.00/0

Current, New and Future Treatments in Dyslipidaemia and Atherosclerosis

Pang H. Chong[1] and *Bonnie S. Bachenheimer*[2]

1 College of Pharmacy, University of Illinois, and Cook County Hospital, Chicago, Illinois, USA
2 Chicago College of Pharmacy, Midwestern University, and Lutheran General Hospital, Park Ridge, Illinois, USA

Contents

Abstract . . . 55
1. Lipoprotein Metabolism and the Atherogenic Process . . . 57
2. Approaches to Treatment . . . 59
3. Current Hypolipidaemic Agents . . . 60
   3.1 Fibric-Acid Derivatives . . . 60
   3.2 Nicotinic Acid . . . 64
   3.3 Probucol . . . 65
   3.4 HMG-CoA Reductase Inhibitors . . . 66
   3.5 Resins and Bile Acid Sequestrants . . . 69
   3.6 Soluble Fibres . . . 69
   3.7 Hormone Replacement Therapy . . . 69
   3.8 Tamoxifen/Raloxifene . . . 70
4. New and Future Hypolipidaemic Agents . . . 71
   4.1 Newer Fibric Acid Derivatives . . . 71
   4.2 New Bile Acid Sequestrants . . . 71
   4.3 New Direct Low Density Lipoprotein (LDL)-Receptor Activators . . . 71
   4.4 Alteration of Cholesterol Metabolism . . . 72
   4.5 Inhibitors of Cholesterol Synthesis . . . 74
   4.6 Monocyte and Macrophage Inhibitors . . . 75
   4.7 Thyromimetic Therapy . . . 75
   4.8 Cholesterol Vaccination . . . 76
   4.9 Somatic Gene Therapy . . . 77
   4.10 Recombinant Proteins . . . 78
   4.11 Prevention of LDL Oxidation . . . 78
   4.12 Other Developing Modalities . . . 81
5. Nutritional Strategies . . . 82
6. Recommendations . . . 84
7. Conclusion . . . 84

Abstract

The new therapeutic options available to clinicians treating dyslipidaemia in the last decade have enabled effective treatment for many patients. The development of the HMG-CoA reductase inhibitors (statins) have been a major advance in that they possess multiple pharmacological effects (pleiotropic effects) resulting in potent reductions of low density lipoproteins (LDL) and prevention of the

atherosclerotic process. More recently, the newer fibric acid derivatives have also reduced LDL to levels comparable to those achieved with statins, have reduced triglycerides, and gemfibrozil has been shown to increase high density lipoprotein (HDL) levels. Nicotinic acid has been made tolerable with sustained-release formulations, and is still considered an excellent choice in elevating HDL cholesterol and is potentially effective in reducing lipoprotein(a) [Lp(a)] levels, an emerging risk factor for coronary heart disease (CHD). Furthermore, recent studies have reported positive lipid-lowering effects from estrogen and/or progestogen in postmenopausal women but there are still conflicting reports on the use of these agents in dyslipidaemia and in females at risk for CHD. In addition to lowering lipid levels, these antihyperlipidaemic agents may have directly or indirectly targeted thrombogenic, fibrinolytic and atherosclerotic processes which may have been unaccounted for in their overall success in clinical trials.

Although LDL cholesterol is still the major target for therapy, it is likely that over the next several years other lipid/lipoprotein and nonlipid parameters will become more generally accepted targets for specific therapeutic interventions. Some important emerging lipid/lipoprotein parameters that have been associated with CHD include elevated triglyceride, oxidised LDL cholesterol and Lp(a) levels, and low HDL levels. The nonlipid parameters include elevated homocysteine and fibrinogen, and decreased endothelial-derived nitric oxide production. Among the new investigational agents are inhibitors of squalene synthetase, acylCoA: cholesterol acyltransferase, cholesteryl ester transfer protein, monocyte-macrophages and LDL cholesterol oxidation. Future applications may include thyromimetic therapy, cholesterol vaccination, somatic gene therapy, and recombinant proteins, in particular, apolipoproteins A-I and E. Non-LDL–related targets such as peroxisome proliferator-activating receptors, matrix metalloproteinases and scavenger receptor class B type I may also have clinical significance in the treatment of atherosclerosis in the near future.

Before lipid-lowering therapy, dietary and lifestyle modification is and should be the first therapeutic intervention in the management of dyslipidaemia. Although current recommendations from the US and Europe are slightly different, adherence to these recommendations is essential to lower the risk of atherosclerotic vascular disease, more specifically CHD. New guidelines that are expected in the near future will encompass global opinions from the expert scientific community addressing the issue of target LDL goal (aggressive versus moderate lowering) and the application of therapy for newer emerging CHD risk factors.

According to the Framingham Heart Study,[1] dyslipidaemia, which can range from hypercholesterolaemia to hypoapolipoproteinaemia, is one of many modifiable major risk factors for coronary artery disease (CAD), stroke and peripheral vascular disease (PVD). Hypertension, diabetes mellitus and tobacco smoking are the other major risk factors. More specifically, dyslipidaemia can lead to the development of coronary atherosclerosis, which can be further accelerated in the presence of multiple risk factors. The clinically relevant manifestations of coronary atherosclerotic plaques are myocardial infarctions (MIs) and unstable angina pectoris. In the US, 800 000 new MIs and 450 000 recurrent MIs occur each year, and about one third of those patients affected die.[2] The economic impact of coronary atherosclerosis in the US is enormous. For example, in 1993, the cost of CAD alone to the nation's economy was $US51 billion, the largest contributor to the total cost of atherosclerotic cardiovascular diseases that year ($US210 billion).[2] More recently, in 1998, the American

Heart Association estimated that the total cost of cardiovascular disease which included coronary heart disease (CHD) and stroke was $US274 billion.[3]

The goal of this review is to introduce clinicians to new and upcoming therapeutic options, including those directed at targets unrelated to low density lipoprotein (LDL) cholesterol, for the treatment of dyslipidaemia and atherosclerosis to prevent CAD. Many of the new therapeutic options have been provoked by the clinical success of current agents, such as the HMG-CoA reductase inhibitors (statins). Despite aggressive LDL cholesterol lowering with the statins, a large proportion of individuals still develop CAD or, in secondary prevention, have a relapse of the disease.

1. Lipoprotein Metabolism and the Atherogenic Process

A brief review of lipoprotein metabolism and the atherogenic process is necessary to understand the emerging therapies that will be discussed.

Cholesterol and triglyceride from dietary fat are solubilised by bile acids into lipid-protein complexes and absorbed into the small intestine. The complexes appear in a spherical shape and contain triglyceride lipid droplets and cholesteryl esters surrounded by polar phospholipids and proteins called apolipoproteins.[4,5] Apolipoproteins play a major role in binding, solubilisation and transport of lipids. They are also important indicators for risk of CHD. In particular, apo A-I and –II are reduced in patients with dyslipidaemia as a result of decreased synthetic (transport) rate and increased fractional catabolic rate.

Chylomicrons function to transport cholesterol and triglyceride throughout the body and contain primarily triglyceride, apo B-48, apo A-I, -II and -IV. In addition, apo C-I, -II, -III and apo E are picked up by chylomicrons from circulating high-density lipoprotein (HDL). Lipoprotein lipase (LPL), located on endothelial cell wall, and hepatic lipase (HL), located on endothelium of the liver, are activated by apos C-I and –II (C-III may inhibit LPL), subsequently breaking down triglyceride into free fatty acids, which are then absorbed into tissues, converted back to triglyceride and stored. Apo A-I, -II, apo Cs and phospholipids are transferred to HDL, leaving chylomicron remnants containing cholesterol ester, apo E and apo B-48, which are taken up by the liver.[6] The increased uptake of these remnants reduces hepatic production of apo B/E receptors. This process also reduces uptake of LDL and remnant particles into cells and increases intracellular cholesterol.

Excess fat and carbohydrate intake is converted into triglycerides in the liver, which are packaged into very-low-density lipoproteins (VLDLs) and transported to extra-hepatic tissues for storage or production of energy. VLDLs contain triglycerides, cholesterol, cholesteryl esters, apo B-100, and apo Cs and apo E acquired from HDLs. The triglycerides in VLDL particles are hydrolysed via LPL, similar to chylomicrons. The resultant particle, remnant VLDL and intermediate density lipoprotein (IDL), contains apo B-100 and apo E, which are necessary for interaction with LDL receptors in the liver. The liver takes up approximately 60% of IDL via apo B/E receptors and the remaining is hydrolysed by HL to produce LDL particles. Therefore, only apo B-100 is returned in this process and thus, the loss of triglycerides from VLDL forms the LDLs.[6]

The major function of LDLs is to carry cholesterol in the blood to tissues. However, when intracellular cholesterol is required, there is an up-regulation of B/E receptors. The only apolipoprotein that LDLs contain is apo B-100, which is necessary for interaction with apo B/E receptors, and subsequent uptake into cells primarily in the liver, adrenal glands and fatty tissue. Approximately 60 to 80% of LDL particles are removed via hepatic and peripheral apo B/E receptors.[7] Once LDL uptake occurs, the cholesterol esters are hydrolysed to free cholesterol, which is used to synthesise cell membranes or hormones, as needed. Excess free cholesterol is re-esterified by acylCoA : cholesterol acyltransferase (ACAT) and stored. In addition, increased intracellular cholesterol inhibits HMG-CoA reductase, the rate limiting step for cholesterol biosyn-

thesis (*de novo*), and decreases the synthesis of apo B/E receptors in an attempt to limit the uptake into the cell.[4-6] LDL can be modified by oxidation and glycation (especially in diabetes mellitus) reactions, aggregation, association with proteoglycans or incorporation into immune complexes.[8] Modified LDL is associated with decreased recognition of LDL by the apo B/E (LDL) receptor and increased recognition by scavenger receptors on macrophages. Subsequently, the uptake of LDL into macrophages is increased, leading to the formation of lipid peroxides, accumulation of cholesterol esters, and foam cell formation.[9]

HDL is important for removal of cholesterol from peripheral tissues. Nascent HDLs are synthesised in the liver and small intestine, and contain the apo A-I, -II, C-I, -II, -III and E. The enzyme, lecithin-cholesterol acyl transferase (LCAT), which is activated by apo A-I, esterifies free cholesterol in HDLs and peripheral tissues to cholesteryl esters, resulting in the conversion of nascent HDL to HDL_3. Further addition of esterified cholesterol results in the conversion to HDL_2, which is also formed from catabolism of chylomicrons and VLDLs. Triglyceride-rich HDL_2 may be converted back into HDL_3 by HL. Cholesteryl esters in HDL are transferred to other lipoproteins containing apo B (VLDL, IDL, LDL) in exchange for triglycerides via cholesteryl ester transfer protein (CETP). Reverse cholesterol transport is the process whereby HDL rids excess cholesterol from the tissues to the liver, where it is excreted or reconstituted into cell membrane or VLDL.[4-6] Patients with low CETP appear to have a lower incidence of CHD (discussed later in section 4.4.2) since cholesterol is not transferred to the atherogenic IDL and LDL, and subsequently transported to arteries.

Atherosclerosis is now well recognised as an inflammatory disorder. Various atherosclerotic lesions represent different stages of the chronic inflammatory process in the artery. If the process is not stopped or is overly aggressive, advanced complicated lesions will develop. Advanced lesions often have active inflammation, rendering them vulnerable to plaque rupture, thrombosis and acute coronary syndromes (i.e. unstable angina and non-Q-wave MI).[9]

Endothelial dysfunction is a key component of the atherosclerotic process and occurs when there is decreased production, release and/or activity of endothelial-derived nitric oxide (NO), also known as endothelial-derived relaxing factor (EDRF), an endogenous protective vasodilator.[10] The consequences of decreased NO and a subsequently dysfunctional endothelium are: impaired vascular relaxation of endothelium; increased endothelial permeability to leucocytes (mainly agranular monocytes), T lymphocytes and platelets; release of vasoactive substances such as prostaglandins that inhibit platelet activation as well as plasminogen that is effective in lysing fibrin clots, pro-inflammatory cell-cell signalling proteins called cytokines [e.g. tumour necrosis factor (TNF)-α, interleukin (IL)-1, interferon (IFN)-γ and numerous growth factors such as platelet-derived growth factor (PDGF) and macrophage-colony stimulating factor (M-CSF)]; adhesion of leucocytes; and increased collagen production by vascular smooth muscle cells (SMC).[11-13] Endothelial dysfunction is caused by many factors, which may include: oxidised LDL; free radical formation (e.g. from smoking, hypertension or diabetes mellitus); genetic mutations; increased plasma homocysteine, lipoprotein(a) and fibrinogens; infectious organisms (e.g. *Chlamydia pneumonia*, *Helicobacter pylori*, herpesviruses, cytomegalovirus, periodontitis); mechanical injury (i.e. percutaneous transluminal coronary angioplasty); and/or the release of vasoconstricting substances [e.g. endothelin-1, thromboxane (TX)A2, angiotensin II].[9,14-16]

The dysfunctional endothelium compensates by activating endothelial cells and SMC proliferation. These activated cells express cell-surface adhesion molecules (CAM) which, in turn, promote chemotaxis and recruit more circulating leucocytes.[11] Monocytes which adhere to the vessel wall and migrate into the extravascular space differentiate into macrophages which take up oxidised LDL via a scavenger receptor pathway and become foam cells. Oxidised LDL stimulates the secretion of mono-

cytic chemotactic proteins and M-CSF by the endothelium and SMCs, which leads to further attraction of monocytes, adhesion and differentiation into macrophages, as well as preventing the macrophages from leaving the intima and acting as cytotoxic agents that damage the endothelium and induce thickening of the intima.[11,12] During this process, some lipids are exported to plasma by macrophages, possibly via reverse cholesterol transport mediated by increased HDL. Therefore, this repetitive process initiates the creation of a lipid core with the existing fatty streak.

Unstable atherosclerotic lesions (advanced atheroma lesions) contain large amounts of oxidised LDL and lipid-laden macrophages, and a thin, fibrous cap. Areas of high foam cell density contribute to degradation and are prone to fissure, leading to acute inflammation, plaque rupture, thrombus formation and acute coronary events. Stable lesions, on the other hand, contain eccentric lipid cores and a thick, fibrous cap. They can grow and eventually occlude an artery, leading to occlusion and ischaemic episodes.[10,17,18] This accumulation of fibrous tissue is a hallmark of the atherosclerotic plaque. However, this does not occur in all individuals. Lipid reduction lowers lipid influx and stabilises the plaque, which restores the injured endothelium to produce NO and other vasodilatory secretions, resulting in normalisation of endothelial cells. This is still considered a fundamental event in the initiation of an atherosclerotic lesion.

Current and future treatments of atherosclerosis are centred around reducing inflammation, improving endothelial function and stabilising the atherosclerotic plaque. The major therapeutic approaches include aggressive lipid lowering, increasing NO activity, inhibiting inflammatory cytokines and treating infectious causes. This review article will briefly cover these therapeutic approaches, but will focus primarily on lipid-lowering therapy.

2. Approaches to Treatment

As discussed, lipoproteins remain a primary target for promoting and developing newer agents to combat dyslipidaemia,[19] particularly elevated LDL, which has conclusively been proven to be involved in atherogenesis and CAD. The current US treatment guidelines focus on LDL cholesterol, and aggressive treatment in the presence of CAD.[20] The development of the statins has been a major advancement in treating hypercholesterolaemia and their benefits have resulted in reduction of overall total mortality in clinical trials.[21-25] However, in an attempt to develop newer agents, the growing knowledge in the field of lipid disorders has further focused on other atherogenic lipoprotein and nonlipid parameters, and the process of atherosclerotic lesion development.

Through epidemiological studies, many lipoprotein and nonlipid parameters have been recently identified which are associated with an increased risk of cardiovascular events. To date, over 200 additional risk factors have been identified. Lipoprotein parameters associated with an increased risk include elevated Lp(a), increased triglycerides, remnant particles, reduced HDL, oxidised LDL and small dense LDL (pattern B).[26] However, the associations have been inconclusive and the need or benefit from diet or drug therapy is still unclear. These emerging CHD risk factors were unaccounted for in the original Framingham Heart Study and may be reclassified to be consistent with the National Cholesterol Education Program (NCEP) ATP II recommendations.[27] Elevated Lp(a) levels, 1 part LDL and 1 part apo(a), [>35 mg/dl; 90.5 mmol/L] may be involved in the recruitment of monocytes to vessel walls, and may participate in the atherogenic process and interfere with fibrinolysis.[28] High serum triglycerides may be a dependent and/ or independent risk factor for cardiovascular disease.[29-32] Triglyceride-rich lipoproteins include VLDL, IDL and chylomicron remnants. Like oxidised LDL, these lipoproteins can cross the vascular intima, may be retained in the arterial wall longer than LDL, and may cause detrimental effects to the wall similar to oxidised LDL. Another possible mechanism for the association of hypertriglyceridaemia and CHD may be related to increased plasma viscosity.[32] In addition, small, dense LDL which is not taken up by LDL receptors and is, therefore, sus-

ceptible to oxidation, has been associated with high serum triglyceride and apo B in LDL, low HDL and CHD. The increased small, dense LDL may be caused by an increased transfer of cholesteryl ester from LDL to triglyceride-rich lipoproteins and may be more atherogenic than pattern A [larger, buoyant LDL particle of normal diameter (>25nm) which is more frequent in most individuals]. The pattern B (<25nm) type accounts for about 50% of the patients with CHD. Isolated low HDL (<10% of patients with CHD) is frequently associated with elevated triglycerides, leading to low levels of HDL and subsequently an increased risk of CHD.[33] There is increased HDL-dependent transfer of esterified cholesterol from HDL to VLDL and LDL particles. Improvement in HDL levels may enhance reverse cholesterol transport, act as an antioxidant and profibrinolytic, and reduce adhesion of monocytes to endothelium.[34]

Among nonlipid parameters, increased levels of homocysteine and fibrinogen are strong indicators for CHD as well as C-reactive protein (CRP), an inflammatory marker, and elevated plasminogen activator inhibitor-1 (PAI-1).[35] Recently, much attention has focused on elevated homocysteine levels (≥16 μmol/L) and their association with an increased risk of atherosclerosis of coronary, cerebral and peripheral arteries, and cardiovascular death.[9] The cofactors, folic acid, pyridoxine (vitamin B6) and cynancobalamin (vitamin B12), are essential for homocysteine metabolism, and are inversely correlated with plasma levels of homocysteine. Homocysteine levels also correlate with kidney function, smoking, levels of fibrinogen and CRP.[36] Elevated CRP may be a significant predictor of a future coronary event, perhaps with or without overt hyperlipidaemia. Investigations into the relative importance of these emerging risk factors in predicting future cardiovascular events are ongoing and will help determine when the broader recommendations are needed in screening and treatment.

To improve morbidity and mortality associated with lipid disorders, the discovery of pleiotropic effects (multiple pharmacological effects) of existing agents and the discovery and development of different therapies that are potentially effective on lipoprotein metabolism and atherosclerosis are currently being evaluated in animals and humans. As a result, agents with direct action on the thrombogenic medium, fibrinolytic process, and changes to atherosclerotic processes on the arterial wall (e.g. endothelial dysfunction, vasospasm, and cytokine regulation of macrophages and smooth muscle function) have been identified.[19]

Research has identified novel therapies that directly enhance LDL receptor activity or that stimulate hepatic apo A-I production, which plays a major role in increasing HDL. Furthermore, inhibitors of different enzymes in cholesterol synthesis such as HMG-CoA reductase and squalene synthetase have been developed. In lipoprotein metabolism, inhibition of ACAT, CETP, monocytes and macrophages, prevention of *in vivo* LDL oxidation with novel antioxidants, and enhanced LPL activity has demonstrated favourable results.[19,35] Future therapies may include revised nutritional strategies, reintroduction of thyromimetic agents, and identifying certain types of patients for cholesterol vaccination, somatic gene therapy and recombinant apolipoproteins. In addition, there are ongoing investigations evaluating the role of non-LDL–related targets including peroxisome proliferator-activated receptors (PPARs), matrix metalloproteinases (MMPs) and scavenger receptor class B type I (SR-BI) [see table I].

3. Current Hypolipidaemic Agents

Clinical studies have exploited the advantages and discovered therapeutic niches for existing hypolipidaemic drugs, and some drugs have pleiotropic effects. Furthermore, research has led to their improved efficacy and safety, which may also improve patient compliance.

3.1 Fibric Acid Derivatives

3.1.1 Gemfibrozil

Gemfibrozil, an older fibric acid derivative (fibrate), has the ability to lower triglycerides, moderately lower LDL and increase HDL levels. All fibrates reduce hepatic synthesis of VLDL and facilitate

Table I. Current, new and future treatments of dyslipidaemia and atherosclerosis

Treatment strategies	Pharmacological agents or class[a]
Lipid parameters/mechanisms	
Directly stimulate hepatic apo B/E (LDL) receptor uptake activity	HMG-CoA RIs, HRT, tamoxifen/raloxifene, thyromimetics (CGS 26214, -23425), imidazolidinyl-pyrimidine derivative (HOE-402)
Decrease bile acid absorption or promote chelation of cholesterol	Resins and cholesterol sequestrants
Inhibit IBAT	IBAT inhibitor (S-8921)
Decrease dietary fat and cholesterol absorption	Resins and cholesterol sequestrants, soluble fibres, ACAT inhibitors
Inhibit pCEH	pCEH inhibitor (WAY-121,898)
Inhibit ACAT activity	ACAT inhibitors
Increase LPL activity	Fibric-acid derivatives (direct activator), NO-1886 (indirect activators)
Decrease CETP activity	CETP inhibitors, fish oil,[b] oleate (monosaturated fats), isoflavin (CGS-25159), hog plasma peptides, recombinant proteins (apo E, apo A-I, HDL), HMG-CoA RIs
Increase CETP activity	Probucol
Increase HDL levels	Nicotinic acid, fibric-acid derivatives, HMG-CoA RIs, HRT, tamoxifen/raloxifene, CETP inhibitors, ACAT inhibitors, recombinant proteins, phenytoin[c]
Decrease triglyceride levels	Fibric-acid derivatives, fish oil, HMG-CoA RIs, nicotinic acid
Increase conversion of small, dense LDL to large, buoyant LDL	Fibric-acid derivatives
Prevent LDL oxidation	HMG-CoA RIs, fibric-acid derivatives, probucol, ACAT inhibitors, fish oil, antioxidants [tocopherol (vitamin E) and ascorbic acid (vitamin C), β-carotene], dietary supplements (polyunsaturated fats, oleic acid, selenium, iron), NAC, aminoguanidine
Inhibit cholesterol synthesis	HMG-CoA RIs, squalene synthetase (squalestatin-1, RPR-107393) and epoxidase inhibitors (TU-2078, NB-598)
Decrease apo B-containing lipoproteins	Nicotinic acid, ACAT inhibitors, fish oil
Increase apo A-I levels	Thyromimetics
Induce antibodies to bind to lipoproteins for removal by scavenger macrophages	Cholesterol vaccination
'Liver-target' genomic application using viral vectors (adenovirus, retrovirus)	Somatic gene therapy
'Over or under expression' via transfer of desirable genes such as apo E, A-I and LCAT	Somatic gene therapy
Enhance Scavenger Receptor Class B Type I	Gene therapy, probucol, polyunsaturated fatty acids
Non-lipid parameters/mechanisms	
Inhibit thrombotic markers (e.g. fibrinogen, PAI-1, PAF-1)	HMG-CoA RIs, fibric-acid derivatives, HRT, nicotinic acid, fish oil
Inhibit inflammatory markers [C-reactive protein, cytokines (TNF-α, INF-γ, IL-1), TXA_2, PGE_2, LTB_4, CAMs]	Leukotriene and IL inhibitors, NSAIDs, CAM inhibitors, HMG-CoA RIs, fibric-acid derivatives, HRT, probucol, fish oil
Induce antiatherosclerotic activity [e.g. decrease SMC proliferation and migration, improve endothelial function (release NO)]	HMG-CoA RIs, fibric-acid derivatives, ACAT inhibitors, probucol, L-arginine, ACE inhibitors,[c] CAs[c], NO synthase gene therapy
Decrease homocysteine levels	Folinic acid, pyridoxine (vitamin B_6), cynancobalamin (vitamin B_{12}), diet
Decrease Lp (a) levels	Nicotinic acid, HRT, tamoxifene, raloxifene, fibric-acid derivatives, thyromimetic, somatic gene therapy, HMG-CoA RIs, acetylsalicylic acid (aspirin)[c]
Inhibit monocytes and macrophages	Leukotriene inhibitors, fish oil, HMG-CoA RIs, probucol, ACAT inhibitors

Table I. Contd

Treatment strategies	Pharmacological agents or class[a]
Enhance angiogenesis of vascular growth factors (VEGF, FGF, PDGF)	Somatic gene therapy
Inhibition of MMPs	MMP inhibitors, TIMP gene therapy
Activate Peroxisome Proliferator Activated Receptors	Natural prostaglandins, antidiabetics (traglitazone, ciglitazone, rosiglitazone), fibric-acid derivatives, WY-14643

a Pharmacological agents or classes listed here may be represented by numerous or limited animal or human data and not necessarily act upon all listed parameters/mechanisms.

b Refers to 3-polyunsaturated fatty acids (3-PUFA), also known as ω-3 fatty acids.

c Nonhypolipidaemic agents.

ACAT = acylcoA:acyltransferse; **ACE** = angiotensin converting enzyme; **apo** = apolipoprotein; **CAM** = cell adhesion molecule; **CAs** = calcium antagonists; **CETP** = cholestryl ester transfer protein; **FGF** = fibroblast growth factor; **IBAT** = ileal Na^+/bile acid cotrasporter; **HDL** = high density lipoprotein; **HMG-CoA RIs** = HMG-CoA reductase inhibitors (statin); **HRT** = hormone replacement therapy; **IL** = interleukin; **INF-γ** = interferon-γ; **LCAT** = lecithin-cholesterol acyl transferase; **LDL** = low-density lipoprotein; **LPL** = lipoprotein lipase; **Lp(a)** = lipoprotein (a); **LTB_4** = leukotriene B4; **MMP** = matrix metallaproteinase; **NAC** = N-acetylcysteine; **NO** = nitric oxide; **NSAID** = nonsteroidal anti-inflammatory drug; **PAF-1** = plasminogen activating factor-1; **PAI-1** = plasminogen activator inhibitor-1; **pCEH** = pancreatic cholesteryl ester hydrolase; **PDGF** = platelet-derived growth factor; **PGE_2** = prostaglandin; **SMC** = smooth muscle cell; **TIMP** = tissue inhibitors of matrix metalloproteinases; **TNF-α** = tumour necrosis factor-α; **TXA_2** = thromboxane A_2; **VEGF** = vascular endothelial growth factor.

VLDL catabolism by stimulating LPL. The result is a reduction in triglyceride levels. They also increase HDL as a result of stimulation of apo A-I synthesis and reducing cholesterol transfer from HDL to VLDL (as a result of the decline in VLDL). In addition, they may prevent CHD by affecting LDL particle size, whereby the fibrate may convert the LDL particle from pattern B to pattern A. Fibrates may also reduce the lag-time of LDL oxidation. After 12 weeks of gemfibrozil 600 mg/day, 23 patients with combined hyperlipidaemia had marked decreases in levels of autoantibodies against oxidised LDL *in vivo*.[37] In addition, gemfibrozil lowers PAI-1 levels, another CAD risk factor.[38] However, gemfibrozil may have a prothrombotic effect, which may require coadministration with acetylsalicylic acid (aspirin).[39,40]

3.1.2 Newer Fibrates

The newer fibrates, bezafibrate and fenofibrate, also reduce triglycerides and increase HDL levels. They have a more potent LDL lowering effect, with fewer nonresponders compared with gemfibrozil, and may also lower plasma fibrinogen levels.[19,41] They may also decrease postprandial lipidaemia and reduce Lp(a) levels.[42-45] A micronised formulation of fenofibrate demonstrates increased absorption and more predictable plasma levels, allowing dose reductions and once-daily administration. Micronised fenofibrate significantly reduced the proinflammatory mediators (cytokines, TNF-α and IFN-γ) in addition to plasma lipids (total cholesterol, triglycerides, apo B, LDL) in 10 patients with type IIb hyperlipidaemia with peripheral atherosclerosis compared with a control group.[46] This small study confirmed that plasma levels of TNF-α and IFN-γ were higher in patients with atherosclerosis compared with healthy controls, and that fenofibrate lowered levels of these cytokines in the hyperlipidaemic patients by approximately 50%, after which they were comparable with levels in the controls.

Comparative studies of fenofibrate with other cholesterol lowering agents have demonstrated improved lowering of triglyceride levels, and increased HDL and decreased fibrinogen levels. In a 12-week study involving 130 patients with type IIa or IIb hyperlipidaemia, micronised fenofibrate 200mg was compared with simvastatin 20 mg/day.[47] Fenofibrate significantly decreased triglyceride (41.4 *vs* 16.5%) and increased HDL (18.5 *vs* 15%) levels compared with simvastatin, whereas simvastatin was significantly better at lowering LDL (34.9 *vs* 20.8%) and total cholesterol (24.5 *vs* 19.4%) levels. In addition, fenofibrate significantly lowered fibrinogen (10.2 *vs* +3.6%) and uric acid levels (25 *vs* 0%) compared with simvastatin. The combination of simva-

statin 40mg plus fenofibrate 200mg for 6 months was compared with previous treatment of simvastatin 40mg plus cholestyramine 32 g/day in 29 patients with severe familial hypercholesterolaemia.[48] The combination of fenofibrate and simvastatin was significantly better at reducing total cholesterol (35 *vs* 29.3%), LDL (40.6 *vs* 37.1%) and triglyceride (17.2 *vs* 12.5%) levels compared with simvastatin plus cholestyramine. HDL levels increased to a greater extent with fenofibrate, but the results were not significant (20 vs 5%). However, the LDL/HDL ratio was significantly improved with fenofibrate (4 *vs* 3.6). Similar results were shown when fenofibrate was compared with other statins, including atorvastatin and lovastatin in patients with combined hyperlipidaemia and type IIa and IIIb hyperlipidaemia, respectively.[49,50] Fenofibrate produced a significant reduction in triglycerides and increase in HDL levels, whereas the two statins had equal to better lowering effect on total LDL cholesterol levels when these agents were compared. In addition, fibrinogen levels were significantly increased by fenofibrate at one month compared with lovastatin, but the results were not significant at 3 months. These clinical trials support the drug's potential use in type IV hyperlipidaemia (hypertriglyceridaemia) as well.

Fibrates may also have beneficial effects on glucose haemostasis. For example, gemfibrozil, and perhaps the other fibrates, may improve insulin resistance in patients with hypertriglyceridemia who are not diabetic but have glucose intolerance, currently known as cardiovascular metabolic syndrome, formerly called syndrome X.[51]

Fibrates have demonstrated a positive clinical outcome as shown with gemfibrozil in the Helsinki Heart Study.[52] In this study, there was a 34% reduction in death due to cardiovascular disease, nonfatal MI, and fatal MI in patients treated with gemfibrozil. There was no difference in all-cause mortality. Two angiographic trials with third generation fibrates include the Progression and Regression of Minor Coronary Arterial Narrowing Fenofibrate Study,[53,54] in which angiographic regression correlated with lower LDL levels, and the Bezafibrate Coronary Artery Intervention Trial (BECAIT),[55] which demonstrated a significant difference of 0.13mm in lumen diameter compared with placebo, a decrease in the extent of stenosis (3.4%) and a reduction in the coronary event rate (3 versus 11; nearly 80%) in young patients with a history of MI treated with bezafibrate. The Lopid Coronary Angiography Trial (LOCAT) also showed decreased progression of coronary and bypass graft atherosclerosis in men with normal LDL and low HDL levels receiving gemfibrozil.[56]

New clinical trials are testing the effects of fibrates on clinical outcomes in patients with mild to moderate hypercholesterolaemia and low HDL levels. The Bezafibrate Infarction Prevention Trials'[57] primary end-point is fatal or nonfatal MI, or sudden death in patients with dyslipidaemias. The recently published Veterans Affairs High Density Lipoprotein Cholesterol Intervention Trial (HIT)[58] evaluated the time to occurrence of nonfatal MI or CHD death in men with CHD taking extended-release gemfibrozil. The intent-to-treat analysis from the HIT trial is the first to show that elevation in HDL may play a critical role in reducing risk for CHD and stroke. Between 1991 to 1993, gemfibrozil 1200 mg/day was compared with placebo in 2531 men <74 years of age with CHD and LDL <140 mg/dl (3.62 mmol/L) [to increase recruitment], HDL <40 mg/dl (1.3 mmol/L) and triglyceride <300 mg/dl (7.76 mmol/L). The gemfibrozil-treated patients had a 7.5% increase in HDL compared with 1.8 in placebo recepients (absolute increase of 2 mg/dl). There was no change in LDL and a 24.5% decrease in triglyceride levels. The relative risk reduction of CHD/nonfatal MI and stroke were 22% ($p = 0.006$) and 27%, ($p = 0.73$; NS), respectively. The reduction in death and cancer were not statistically significant. The relatively low increase in HDL levels indicate that there may be other factors that led to the observed clinical benefits. Of interest, a primary prevention clinical trial, the Air Force/Texas Coronary Atherosclerosis Prevention Study (AFCAPS/TexCAPS), showed a 7% increase in HDL levels with lovastatin.[23] This result allowed the expansion of the Food and Drug

Administration (FDA) indications for lovastatin to include the need to increase HDL levels. Recently, atorvastatin and simvastatin have also received FDA approval for elevating HDL levels. These results support the potential benefit of elevating HDL levels, an effect often seen with fibrates and statins.

Two ongoing angiographic studies are examining the progression of CHD in patients with type 2 diabetes mellitus with mild to moderate hypercholesterolaemia, including the Diabetes Atherosclerosis Intervention Study (DAIS),[59] an angiographic regression trial with micronised fenofibrate, and the Fenofibrate Intervention and the Event Lowering in Diabetes (FIELD) study in 8000 patients with mild to moderate lipid abnormalities and a history of cardiovascular disease.[60,61]

3.1.3 Clofibrate

Fibrates have also shown a negative clinical outcome with clofibrate in the Cooperative Trial on Primary Prevention of Ischaemic Heart Disease.[62] Clofibrate reduced the first major coronary event by 20% and nonfatal MI by 25% compared with control. However, there was no change in the incidence of fatal MI, and there was greater all-cause mortality in the clofibrate group. Moreover, the World Health Organization trial using clofibrate showed an increased incidence of cardiac arrhythmias and higher total mortality.[63] The drug is no longer recommended as a lipid-lowering agent because of this large increase in overall mortality

3.2 Nicotinic Acid

Nicotinic acid (niacin) inhibits hepatic production of VLDL and its metabolite LDL. In comparison with other currently available hypolipidaemic agents, the advantages of nicotinic acid are its potent ability to increasing HDL (HDL, HDL_2) and apo A-1 levels. This is accomplished by reducing lipid transfer of cholesterol from HDL to VLDL and delaying HDL clearance. Hypothetically, nicotinic acid may inhibit the HDL catabolism pathway involving apo A-1 (by inhibiting the removal of apo A-1 at the catabolic receptor or pathway - 'holoparticle uptake') but not the SR-BI that mediates selective cholesterol ester removal. This mechanism would increase circulating HDL levels without significantly affecting cholesterol removal, thereby augmenting reverse cholesterol transport and other functions of HDL (discussed in section 4.12.3).[64] Nicotinic acid also produces modest decreases in triglyceride and apo B levels, and significant decreases in Lp(a) levels. It has also demonstrated a significantly favourable correlation with changes in fibrinogen and LDL cholesterol levels.[65] Unfortunately, nicotinic acid is often poorly tolerated. Sustained-release formulations demonstrate similar efficacy to the crystalline forms but produce a lower incidence of flushing. However, sustained-release formulations have been associated with more gastrointestinal disturbances and hepatotoxicity.[66] These adverse effects may have been dose-related, and/or due to the type of preparation used.

A new extended-release, once daily, nicotinic acid formulation was recently approved by the FDA. Its efficacy and safety was maintained for up to 96 weeks in one study with minor effects on hepatic enzymes.[67] 225 patients receiving this new extended-release formulation as monotherapy completed the 96-week trial and received a mean dose of nicotinic acid 2000mg. A 20% decrease in LDL cholesterol, a 17% decrease in apo B, a 28% increase in HDL cholesterol, a 13% decrease in total cholesterol, a 28% decrease in triglyceride and a 39% decrease in Lp(a) levels, and a 31% favourable change in the total/HDL cholesterol ratio from baseline were seen. The incidence of flushing was about 75%. There were statistically significant increases in levels of hepatic transaminases, alkaline phosphatase, direct bilirubin, amylase, uric acid and leucocytes. The study also observed significant changes in phosphorus and glucose levels, and platelet counts. According to the authors, these changes did not appear to pose clinically significant safety issues. However, the decrease in phosphorus was a new finding in nicotinic acid therapy. The efficacy and safety profile of this new extended-release formulation in clinical trials has been similar to those observed with immediate-release nicotinic acid.[67-70]

Nicotinic acid can affect insulin resistance resulting in glucose intolerance, and should be used

cautiously in patients with diabetes. However, in one study, the combination of low-dose nicotinic acid with pravastatin in 14 patients with diabetes had little or no effects on glycaemic control and favourable effects on the lipid profile.[71] However, patients receiving combination therapy should be observed closely for the occurrence of myopathy, especially with concomitant statin therapy. Nicotinic acid can also decrease the secretion of uric acid, resulting in increased uric acid levels. The risk is dose-related and it should be used cautiously, if at all, in patients predisposed to gout.[72]

Nicotinic acid has demonstrated favourable outcomes in cardiovascular morbidity and mortality in 6 major clinical trials and, therefore, has had a solid foundation for its use.[73] Nicotinic acid monotherapy decreased the incidence of recurrent nonfatal MI by 27% and cerebrovascular events by 26% after 6 years, and decreased total mortality by 10.6% after 15 years of long term follow-up in the Coronary Drug Project (CDP).[74] Other trials showed benefits in coronary and total mortality, coronary events, and angiographic progression and/or regression. The Stockholm Ischaemic Heart Disease Secondary Prevention Study (IHD) with nicotinic acid plus clofibrate resulted in decreased risk of ischaemic heart disease mortality by 36% and decreased total mortality by 26%.[75] The Cholesterol-Lowering Atherosclerosis Study (CLAS)[76,77] and the Familial Atherosclerosis Treatment Study (FATS)[78] were angiographic trials of nicotinic acid plus colestipol, which demonstrated regression and decreased progression of lesions, and decreased cardiovascular events. In CLAS I, coronary angiography was performed 2 years after coronary artery-bypass graft surgery (CABG). The change in progression of lesions from baseline was 39 versus 61% and the change in regression was 16 versus 2% for the placebo group compared with the niacin/colestipol group, respectively. In CLAS II, an angiogram was performed 4 years after study initiation. The change in progression of lesions from baseline was 48 versus 85% and the change in regression was 18 versus 6% for the placebo group compared with the nicotinic acid/colestipol group, respectively. The FATS study demonstrated progressive disease in 25% of patients compared with 46% in the placebo group, regression in 39% of patients compared with 11% in the placebo group, and a 1.1% reduction from baseline in stenosis compared with a 2% increase in the placebo group. Although the effects on lesions with nicotinic acid plus colestipol were significant although marginal, the incidence of fatal/nonfatal MI with this treatment was 6 versus 19% in the placebo group. The combined results of nicotinic acid plus colestipol and lovastatin plus colestipol in this trial demonstrated a reduction in clinical events of 73%. The marked reduction in clinical events despite the minimal effect on stenosis led to the belief that the current lipid-lowering drugs have alternate mechanisms of reducing cardiovascular clinical events. The University of California-San Francisco Specialized Center of Research study was an angiographic trial with nicotinic acid plus colestipol and lovastatin (in approximately half the patients) which showed a regression of percent area stenosis of 1.5 ($p = 0.04$) with a 39% reduction in LDL.[79] The Harvard Atherosclerosis Reversibility Project was another angiographic trial using pravastatin, nicotinic acid, colestipol and gemfibrozil, which did not demonstrate significant differences in lesion progression or clinical coronary events but showed a 33% reduction in clinical events.[80]

3.3 Probucol

Probucol reduces LDL levels by 10 to 15% and HDL levels by approximately 30% in patients with hypercholesterolaemia. The mechanism by which probucol decreases HDL is unclear, but it may be partly as a result of increasing the activity of CETP, a lipid regulatory process that is involved in lipoprotein metabolism. A beneficial effect of probucol is that it prevents LDL oxidation. Therefore, probucol increases the resistance of LDL to oxidative modification to the more atherogenic form perhaps because the drug invades the inside of the lipid-rich core of LDL. It reduces lipid peroxides via a free radical scavenger action, and protects LDL and Lp(a) from oxidative damage *in vitro*. Thus, probucol

may have a niche in the treatment of homozygous familial hypercholesterolaemia (HFH).[19,81] One study showed that xanthoma (exposed body fat deposits) regression with probucol use in patients with HFH was directly related to reductions in HDL levels.[82] After 3 years, the results of the Probucol Quantitative Regression Swedish Trial (PQRST) showed that probucol 1 g/day plus cholestyramine 8 to 16 g/day compared with cholestyramine alone or placebo, reduced HDL and LDL levels by 24 and 12%, respectively, and produced LDL particles that were less susceptible to oxidation in 274 patients with relatively advanced femoral atherosclerosis.[83] However, the study failed to show a clinical benefit from probucol. Probucol decreased atherosclerotic lesions in Watanabe heritable hyperlipidaemic (WHHL) rabbits, which was attributed to its antioxidant effects rather than its lipid-lowering effects.[84]

The Probucol and Multivitamins in the Prevention of Restenosis After Coronary Angioplasty trial compared probucol alone with placebo, multivitamins [beta carotene, ascorbic acid (vitamin C) and tocopherol (vitamin E)], or probucol plus multivitamins with an end-point of reduction in the frequency of restenosis after coronary angioplasty in 317 patients undergoing elective angioplasty.[85] Interestingly, probucol alone, but not with multivitamins, significantly reduced the rate of restenosis after balloon angioplasty. The authors suggested that the positive effects observed from probucol may have been due to its powerful antioxidant effects which may have prevented endothelial damage and LDL oxidation secondary to angioplasty. The effects may also have been due to inhibition of IL-1 secretion by macrophages, which could have decreased the secretion of MMPs by SMCs. MMPs are proteinases that destroy collagen and connective tissues (discussed in section 4.12.2). Macrophages secrete MMP-1, which has the same destructive effects. Therefore, probucol may have indirectly prevented neointimal formation and/or affected the remodelling of the extracellular matrix of the artery wall.

It must be noted that although probucol appears to possess desirable properties for the treatment of hyperlipidaemia and atherosclerosis, the drug has been withdrawn from the US market because of its potential to induce serious ventricular arrhythmias. It should be avoided in patients, especially females, who have QT prolongation or are currently taking other drugs which prolong the QT interval.[86]

3.4 HMG-CoA Reductase Inhibitors

The clinical success of the HMG-CoA reductase inhibitors (statins) has led to newer synthetic molecules, which are more potent (atorvastatin) or equipotent (cerivastatin) on a milligram per milligram basis compared with synthetic fluvastatin. All statins are competitive inhibitors of HMG-CoA reductase, the rate-limiting step in cholesterol biosynthesis. The reduction of intrahepatic cholesterol stimulates synthesis of apo B/E (LDL) receptors in the liver, thereby enhancing removal of LDL and VLDL remnant particles from circulation and further lowering LDL. They also modestly increase HDL and lower triglyceride levels as VLDL remnant particles are cleared by newly synthesised apo B/E (LDL) receptors. At current FDA approved maximal doses, atorvastatin 80mg is the most efficacious statin with LDL reductions of 50 to 60%, followed by simvastatin 80mg (47%), lovastatin 80mg (40%), pravastatin 40mg (35%), cerivastatin 0.4mg (34%) and fluvastatin 40mg (30%).[87] Both atorvastatin and cerivastatin exhibit improved activity for reducing lipoproteins and improving tissue selectivity. After 54 weeks, compared with other statins, atorvastatin demonstrated higher liver selectivity, longer inhibitory effects and a potent reduction in LDL and triglyceride levels.[88] The longer inhibitory effect is probably the reason for its higher potency, rather than its liver selectivity. Atorvastatin demonstrated a significantly greater reduction in LDL levels at doses 10 to 40mg over milligram-equivalent doses of the other statins. The patients evaluated had baseline LDL levels of ≥160 mg/dl (4.14 mmo/L) and triglyceride levels of <400 mg/dl (10.35 mmol/L). The potency of atorvastatin was also demonstrated in the Target-To-Treat trial which compared the reduction in LDL levels with NCEP goals.[89] Approximately 32% of patients receiving atorva-

statin (starting dose 10 mg/day) reached LDL goals after 54 weeks compared with fluvastatin (1%), lovastatin (10%) and simvastatin (22%). Some patients receiving these statins required frequent titration and the addition of colestipol (5g twice daily) in order to achieve their LDL goals.

Cerivastatin, although studied in doses as high as 0.8 mg/day, has been approved by the FDA at a dose of 0.3mg and more recently, 0.4 mg/day. Although the number of completed clinical trials with cerivastatin is limited, it does not seem to offer any additional clinical advantage over the other statins at this time. However, it may be a good economical choice at equipotent doses of lovastatin, pravastatin and simvastatin when patients require <35% reduction of LDL to meet their goal.

Although atorvastatin and cerivastatin have yet to demonstrate a reduction in CHD events, ongoing clinical trials with these agents should provide the answers in the near future. The much-awaited results will add to the existing wealth of information regarding the statin group of drugs and CHD events. Notable clinical trials for primary prevention of CHD include The West of Scotland Coronary Prevention Study (WOSCOPS)[21] and the AFCAPS/TexCAPS.[23] Notable trials for secondary prevention include the Scandinavian Simvastatin Survival Study (4S),[25] the Cholesterol and Recurrent Events (CARE) study[24] and the Long term Intervention with Pravastatin in Ischaemic Disease (LIPID) study.[22]

The WOSCOPS, CARE and LIPID trials involved pravastatin, whereas the AFCAPS/TexCAPS trial involved lovastatin and the 4S trial evaluated simvastatin. WOSCOPS studied 6595 high-risk patients with average total cholesterol level of 272 mg/dl (7.03 mmol/L) and HDL of 44 mg/dl (1.14 mmol/L), in which the treatment reduced the risk for nonfatal MI or CHD by 31% and the risk reduction in all-cause mortality by 22%. The CARE study, which evaluated 4159 patients with CHD with an average baseline total cholesterol level of 209 mg/dl (5.4 mmol/L) and LDL of 139 mg/dl (3.6 mmol/L) demonstrated a 24% decreased risk of nonfatal MI and CHD death. The LIPID trial was the first to show a reduction in CHD mortality (24%) in patients (n = 9014) with a history of MI or unstable angina and normal cholesterol levels [total cholesterol level = 218 mg/dl (5.64 mmol/L)]. The study recruited more elderly, more females and more patients with diabetes than previous clinical trials. A meta-analysis will soon be available comparing the results of the WOSCOPS, CARE and LIPIDS trials. AFCAPS/TexCAPS studied 6605 low-risk patients with average baseline total cholesterol levels of 221 mg/dl (5.72 mmol/L) and HDL cholesterol levels of 37 mg/dl (0.96 mmo/L). Lovastatin 20 to 40 mg/day reduced the risk of a first major acute coronary event by 37%. Secondary prevention was also observed with simvastatin in the 4S trial.[25] In contrast to the CARE study, patients in 4S had elevated levels of both total and LDL cholesterol, and experienced a 30% decreased risk in total mortality.

Although the above statin trials did not specifically address optimal goals for LDL cholesterol level lowering, current trials are addressing this issue. An ongoing clinical trial [Study of Effectiveness of Additional Reductions of Cholesterol and Homocysteine (SEARCH)] is comparing aggressive therapy with simvastatin 80mg versus 20mg. Previously, the Post Coronary Artery Bypass Graft trial (Post-CABG)[90] trial involving lovastatin plus warfarin showed that aggressive lowering of LDL (to <100 mg/dl; 2.59 mmol/L) with lovastatin up to 80 mg/day plus cholestyramine up to 8g/day, if needed, may be better than moderate (to <130 mg/dl; 3.36 mmol/L) lowering with lovastatin 2.5 mg/day plus cholestyramine, if needed, at reducing the progression of bypass graft atherosclerosis. The low-dose warfarin 1 mg/day did not affect atherosclerosis. The Atorvastatin Versus Revascularisation Treatments (AVERT) showed that 87% of patients (n = 341) who were candidates for coronary angioplasty when randomised to atorvastatin 80 mg/day, were able to reduce LDL to below 100 mg/dl and remained on medical therapy for 18 months.[91] The incidence of primary end-point (death, nonfatal MI, stroke, need for subsequent revascularisation, hospitalisation for angina, and resuscitated cardiac

arrest) was 13% in the atorvastatin group versus 21% in the angioplasty group ($p = 0.045$), with a trend towards statistical significance. However, angioplasty provided better relief of angina (54 versus 41% with atorvastatin). The mean baseline LDL level of 140 mg/dl (3.62 mmol/L) was lowered to 77 mg/dl (1.99 mmol/L) in the atorvastatin group compared with 118 mg/dl (3.05 mmol/L) in the angioplasty group. It should be noted that the AVERT study design allowed the use of atorvastatin, according to clinical guidelines, for patients randomised to angioplasty; this explains the cholesterol reduction in the angioplasty arm of the trial.

The aggressive therapy studies, in addition to the primary and secondary trials, suggest that the relationship between lowering LDL levels and reduction in CHD events is nonlinear. In other words, the clinical benefits of cholesterol lowering with statins diminish once LDL falls below a certain level, suggesting that the relationship between LDL and relative risk for CHD may follow a curvilinear or a threshold model.[92] For example, in the CARE trial, no further risk reduction occurred once LDL levels fell below 125 mg/dl (3.23 mmol/L), supporting the threshold model. In the 4S trial, the data suggest that lower cholesterol levels give continuous but progressively smaller decrements in CHD risk, supporting the curvilinear model. In the Treating to New Targets (TNT) clinical trial, atorvastatin will be studied to determine whether lowering LDL levels below the current NCEP targets can further reduce the incidence of MI or death in patients with preexisting CHD. This 5-year trial will investigate whether there is greater clinical benefit in treating patients to a lower target LDL goal of 75 mg/dl (1.94 mmol/L) compared with 100 mg/dl (2.59 mmol/L).[93] The results from the primary and secondary prevention trials involving the statins suggests that they can reduce CHD events spanning the entire continuum of risk – high-risk patients at risk for CAD (4S), at intermediate-risk for CAD (CARE, LIPID, WOSCOPS) and at low-risk for CHD (AFCAPS/TexCAPS).

In addition to potent LDL lowering effects, the statins possess other pharmacological effects that may have contributed to the prevention of atherosclerosis and thrombosis and the reduction in total mortality reported in several clinical trials. These nonlipid effects can be categorised as antiatherosclerotic and antithrombogenic effects since they influence plaque stability and thrombosis. There is limited clinical evidence emerging which demonstrates that the statins may regulate coronary arterial tone by improving endothelial dysfunction associated with hypercholesterolaemia and CAD via NO formation.[94-96] This antiatherosclerotic regulation by the statins may improve coronary ischaemic symptoms and stabilise the atherosclerotic plaque, particularly if administered early in the course of an acute coronary event.[96] Statins may interfere with SMC proliferation[97,98] and migration.[98,99] This occurs independently of their cholesterol-lowering effect and differences among the various statins may be related to their ability to penetrate cells. Inhibition of monocyte-macrophages and their inflammatory cytokine activity, subsequent foam-cell formation and increased resistance to LDL oxidation are other potential mechanisms by which the statins may alter the atherosclerotic process.[100] Recently, the proinflammatory component that occurs during atherosclerosis was significantly diminished with a statin. Atorvastatin diminished not just lipids and lesion size, but macrophage infiltration and monocyte chemoattractant protein-1 (MCP-1) in neointimal inflammation. In addition, TNF-α-induced vascular SMC MCP-1 expression and nuclear fraction-$\kappa\beta$ (NF-$\kappa\beta$) were down-regulated.[101]

Statins possess antithrombotic mechanisms that may be associated with net atherosclerotic changes.[100] These mechanisms include the inhibition of platelet activating factors and platelet aggregation, reduction of fibrinogen levels, suppression of tissue factors and reduction of PAI-1 levels. These properties of statins may prevent or stabilise the atherosclerotic plaque, particularly the 'vulnerable plaque', in which the lipid core of the lesion is exposed to further platelet aggregation and LDL oxidation, and subsequent inflammation. These favourable nonlipid properties may have contributed to the re-

duction of CHD events reported in several atherosclerotic regression trials, such as the Regression Growth Evaluation Statin Study (REGRESS),[102] the Pravastatin, Lipids, and Atherosclerosis in the Carotid Arteries (PLAC) study,[103] the Multicentre Anti-atheroma Study (MAAS)[104] and the Canadian Coronary Atherosclerosis Intervention Trial (CCAIT).[105] These potential nonlipid mechanisms of action of statins suggest that their beneficial effects go beyond LDL reduction. This indicates that clinical decision making should not be based solely on surrogate end-points but on clinical outcomes, the gold standard for assessing the efficacy of any treatment.

3.5 Resins and Bile Acid Sequestrants

Cholestyramine and colestipol have been the agents most used in this class of drugs. They are desirable because of their nonsystemic mode of action. They bind bile acids in the intestine, resulting in interruption of reabsorption of bile acids. This lowers intrahepatic cholesterol and promotes the synthesis of apo B/E (LDL) receptors. These receptors bind LDL from plasma, resulting in a further lowering of cholesterol in the blood. These agents also minimally increase HDL via intestinal formation of nascent HDL. In the Lipid Research Clinics Coronary Primary Prevention Trial (LRC-CPPT), the cholestyramine-treated group demonstrated a 19% reduction in CHD, which was associated with each 11% decrement in LDL and 8% decrease in total cholesterol levels.[106] Unfortunately, intolerance to these agents makes compliance difficult for large numbers of patients. Newer formulations such as tablets, caplets, flavoured granules and 'lighter preparations' have been associated with fewer gastrointestinal adverse effects.

Inorganic cholesterol sequestrants include bismuth salicylate plus montmorillonite clay, aluminum hydroxide and calcium carbonate antacids. Bismuth salicylate with montmorillonite clay has a sequestering ability similar to cholestyramine.[107] Aluminum hydroxide has been reported to moderately decrease LDL levels.[108]

3.6 Soluble Fibres

Research has also been performed with natural and semi-synthetic water-soluble dietary fibres, which reduce the absorption of exogenous dietary cholesterol. Essentially, these agents are poorly digested polysaccharides that decrease passive transport of cholesterol substrates into the intestinal lumen.[33] Since most cholesterol in the intestine is biliary rather than dietary, these agents may also reduce serum LDL levels via increased LDL receptor uptake activity in the liver.

There are several reasons for using soluble fibre supplements to treat hypercholesterolaemia. First, the viscosity of the fibre may decrease absorption of cholesterol from the gut. Secondly, fibre may increase the excretion of bile acids in the faeces, forcing the liver to produce more bile acids from cholesterol to compensate. Thirdly, the fermentation of soluble fibre by colonic bacteria produces short-chain fatty acids including propionates. These in return are absorbed and converted to succinyl-coenzyme A in the liver where cholesterol synthesis may be inhibited.[109]

Examples of natural water soluble fibres include psyllium, oat, guar and pectin. These agents demonstrated a reduction in total cholesterol levels of 15 to 20% in healthy volunteers and in patients with type 2 diabetes.[110] The hypocholesterolaemic activity of these natural soluble fibres is modest at best in reducing absorption of cholesterol in animal and humans. In addition, the large doses required have the potential for significant adverse intestinal effects. Currently, semisynthetic soluble fibres are being evaluated for preventing cholesterol entry into the intestinal mucosa.

3.7 Hormone Replacement Therapy

The estrogens directly stimulate LDL receptor activity, leading to marked reductions in total and LDL cholesterol levels, and moderate increases in HDL cholesterol levels.[111] In a study comparing estrogen/progestogen with simvastatin 10mg daily in 58 postmenopausal women with hypercholesterolaemia, hormone replacement therapy (HRT) de-

creased total cholesterol by 14%, LDL cholesterol by 24% and Lp(a) by 27%, and increased HDL cholesterol by 7% and triglycerides by 29%. In comparison, simvastatin reduced total cholesterol by 26%, LDL by 36% and triglycerides by 14%, increased HDL by 7% and had no effect on Lp(a).[112]

Observational studies have shown that the effects of estrogens on the lipid profile may partly account for their cardioprotective effect in postmenopausal women with significant risk factors for CHD. The Postmenopausal Estrogen/Progestin Interventions (PEPI) Trial demonstrated marked increase in HDL cholesterol levels with estrogen alone as compared with the combination of estrogen plus progestogen. However, estrogen alone was associated with a higher rate of endometrial hyperplasia.[109] Several other studies have shown that estrogen may lower Lp(a), a CHD risk factor and exhibit beneficial effects on the arterial wall in women.[113-116] In a recent study in 28 women taking conjugated estrogen 0.625mg and/or simvastatin 10mg for 6 weeks, estrogen lowered PAI-1 and CAM E-selectin levels, in addition to lowering LDL and Lp(a) and increasing HDL levels.[117] This effect is unique in that it targets markers of fibrinolysis and vascular inflammation.

In the case of primary or secondary prevention, studies of HRT have yielded conflicting results. The Heart and Estrogen/progestin Replacement Study (HERS) Research Group concluded that estrogen 0.625mg plus medroxyprogesterone 2.5 mg/day had no significant effect on primary (nonfatal MI or CHD death) or secondary (coronary revascularisation, unstable angina, congestive heart failure, MI, stroke, transient ischaemic attack, PVD and overall mortality) outcomes compared with placebo.[118] The study involved 2763 postmenopausal women with CHD who were followed for approximately 4 years. There was a lack of an overall effect despite an 11% reduction in LDL and a 10% increase in HDL levels compared with placebo ($p < 0.001$). However, there was a trend toward statistical significance if the study had continued. The majority (45%) of these patients were receiving lipid-lowering agents. The treatment group experienced a higher rate of thromboembolic events and gallbladder disease. The results of this study contradict previous studies regarding HRT and the prevention of CAD. The discrepancy may be due to previous studies being observational, different characteristics of study populations and differences in treatment regimens, including the use of different progestogen formulations or of unopposed estrogen. Another recent study from Duke University, North Carolina, in 1857 postmenopausal women with CAD found similar results to the HERS trial.[119] 33% of 111 women who started HRT post-MI were hospitalised within 12 months for unstable angina with no deaths. 17% of women not taking HRT were hospitalised within 1 year and and 4% died. In addition, 21% of 413 women taking HRT before their first MI and who continued taking HRT afterwards, were hospitalised with unstable angina within 12 months. Four women (1%) in this group died. The authors suggest that the thrombogenic properties of estrogens may partly explain the findings in these studies. Currently, estrogen does not have an FDA approved indication for either the regulation of lipids or the reduction of CHD.

The Women's Health Initiative (WHI), begun in 1992 and to be completed in 2007, is a large clinical investigation currently enrolling 63 000 postmenopausal women 50 to 79 years of age to determine the major causes of morbidity and mortality in older women. One of the objectives of the clinical trial is to compare estrogen with the combination of estrogen/progestogen to prevent CHD, which will include 27 500 women.[120] Hopefully, the results of this trial will answer some questions with respect to the use of estrogen alone versus combination therapy for the prevention of unfavourable outcomes in postmenopausal women with CHD. Some of the unresolved issues with HRT are the lack of controlled clinical trials, endometrial effects, preferred progesterone dosage regimen and resumption of menses (with cyclical therapy).

3.8 Tamoxifen/Raloxifene

Other sterol therapies used in postmenopausal women such as tamoxifen and raloxifene, used in

breast cancer and for the prevention of osteoporosis, respectively, have been demonstrated to reduce total and LDL cholesterol. Tamoxifen has been associated with lower rates of MI, although higher rates of thromboembolic diseases have been reported.[121] Raloxifene has been shown to lower fibrinogen and Lp(a) levels in addition to increasing HDL_2 without raising triglycerides.[122]

4. New and Future Antihyperlipidaemic Agents

Future advances may include refining existing lipid-lowering agents, altering activity of other enzymes involved in lipoprotein metabolism, inhibiting other enzymes of cholesterol synthesis, developing drugs to target specific risk factors and drugs directed at protecting the arterial wall against atherosclerosis. As there is limited availability of human data, much of the information provided in this section involves animal data.

4.1 Newer Fibric Acid Derivatives

Lifibrol is a compound that combines all of the desirable properties of a fibric acid derivative. Lifibrol markedly lowers LDL, apo B, postprandial lipids and Lp(a) levels.[123-125] Ciprofibrate is the latest addition to this class, which is currently undergoing clinical evaluation.

4.2 New Bile Acid Sequestrants

To further improve efficacy, patient tolerance and compliance, a large number of bile acid sequestrants have been derived from cholestyramine and colestipol by modifying their organic structures through a process called quaternisation.[107,126] Colesevelam, a nonabsorbed hydrogel with bile acid-sequestering properties has recently become available. It been show to lower LDL cholesterol at a dosage of 1.5 to 3.75 g/day without constipating effects.[127] Water soluble derivatives include 3,3-ioene, N-(cycloalkyl) alkylamines and poliglusam (chitosan; CP-88488).[33,107] Oral poliglusam has shown promising results. In poliglusam-fed mice without apo E, there was a 64% reduction in blood cholesterol and a 40 to 50% inhibition of atherogenesis of the aorta and aortic arch after 20 weeks.[128] Insoluble quaternised polystyrenes are also undergoing clinical testing. Other strategies are to functionalise polysaccharides or to use cross-linked poliglusam, kanamycin or neomycin.[33] However, the clinical applications of these strategies are not yet known.

Natural and synthetic bile acid sequestrants such as saponins have been extensively investigated and may be effective in the treatment of moderate hypercholesterolaemia.[33] These agents cause cholesterol precipitation, interference with micelle formation or bile acid absorption.[19] The synthetic saponin, tiqueside (CP-88818), similar to pamaqueside (CP-148623), is a β-tigogenin cellobioside which inhibits dietary and biliary cholesterol absorption and reduces cholesterol in animals and humans.[129,130] Pamaqueside is a modified derivative of tiqueside with advanced efficacy and 10 to 30 times greater potency.[131]

A novel inhibitor of cholesterol absorption, the ileal Na^+/bile acid cotransporter (IBAT) inhibitor, S-8921, demonstrates similar properties to bile acid sequestrants. After 2 weeks, serum cholesterol was reduced by 29 to 37% and faecal excretion of bile acid increased by 60 to 80% in WHHL rabbits compared with controls. In addition, aortic atherosclerotic lesions were reduced.[132] IBAT inhibitors appear promising but require future clinical investigations for the treatment of hypercholesterolaemia.

4.3 New Direct Low-Density Lipoprotein (LDL)-Receptor Activators

HOE-402 is an imidazolidinyl-pyrimidine derivative representing a novel agent that directly stimulates LDL receptor activity. HOE-402 at a dose of 100 mg/kg, reduced cholesterol levels by 50% after one month of treatment in heterozygous WHHL rabbits.[133] In male hamsters fed cholesterol diets plus HOE-402, whole body LDL production was 20% lower in the treatment group compared with controls, and plasma clearance of LDL was 2.1-fold higher. The study further demonstrated that HOE-402 had a more direct action on hepatic LDL receptor activity rather than a in-

direct intestinal effect on cholesterol absorption after 3 weeks.[134] When observed in human hepatic cells (Hep G2 cells), LDL receptor activity was significantly increased, independent of the HMG-CoA reductase pathway.[33] However, the clinical significance of this particular agent needs confirmation from human clinical trials.

4.4 Alteration of Cholesterol Metabolism

4.4.1 AcylCoA:Cholesterol Acyltransferase Inhibitors

A new investigational approach to treat and prevent atherosclerosis is to alter the activity of enzymes or proteins involved in regulating lipoprotein metabolism.[6] One approach is to inhibit ACAT, which is a ubiquitous enzyme responsible for esterifying excess intracellular cholesterol. Excess cholesterol is stored in the core of lipoprotein particles and, subsequently, may develop into an atheroma. The activity of ACAT is therefore enhanced by the presence of intracellular cholesterol. However, whether inhibition of ACAT will prevent atherosclerosis is not yet clear.

ACAT inhibition may also reduce the synthesis of VLDL, which is a product of cholesterol esterification.[33,35] Inhibition of cholesterol esterification would allow more cholesterol to come to the cell surface and subsequently be shunted into the bile acid pool. In rabbits with endogenous hypercholesterolaemia, ACAT inhibition reduced LDL levels by 43% and VLDL by 62% in cholesterol-fed rabbits compared with those receiving placebo.[135]

ACAT inhibitors have also reduced the overproduction of apo B-100–containing lipoproteins.[136,137] Apo B-100 is the major protein in LDL that binds to LDL receptors. Thus, in the presence of elevated LDL levels, the liver increases production of apo B-100–containing lipoproteins, leading to CAD. One study showed that ACAT inhibition with FCE-27677 decreased secretions of apo B-100–containing lipoproteins and cholesteryl ester load in human HepG2 cells, whereas gemfibrozil and lovastatin had no effect.[136] Similar results were demonstrated in casein-fed rabbits given FCE-27677[137] and in miniature pig hepG2 cells with avasimibe (CI-1011).[138] ACAT inhibitors may be useful in patients with overproduction of apo B-1003–containing lipoproteins.

Another ACAT inhibitor HL-004 showed prevention of cholesterol ester-rich foam cells and the stimulation of large amounts of free cholesterol from pre-established foam cells in the presence of HDL in mice peritoneal macrophages.[139] It also reduced serum cholesterol levels and simultaneously decreased hepatic cholesteryl ester content and increased free cholesterol in cholesterol-fed rats.[140] These studies suggest that the inhibition of foam cell formation and regression by the ACAT inhibitors may prevent the atherosclerotic process on the arterial wall.

ACAT is also necessary for absorption of cholesterol from the intestines. The ACAT inhibitors lecimibide (DuP-128) and CL-277082, reduce intestinal cholesterol absorption but do not provide reductions in elevated serum cholesterol.[141,142] However, other ACAT inhibitors, such as the dedecamanide derivative CI-976, the methylphenyl urea derivative E-532, and the N-arylurea derivative FR-186054, have reduced serum cholesterol with concomitant antiatherosclerotic activity at enzyme inhibiting doses in cholesterol-fed rabbits and normolipidaemic volunteers.[143-146] More recently, avasimibe (CI-1011) and YM-17E demonstrated prevention and regression of atherosclerosis in animals.[147,148] HL-004 prevented progression of atherosclerosis in cholesterol-fed rabbits as a result of inhibition of cholesterol absorption in the intestine.[149] The synthetic cholesterol absorption inhibitor, SCH-48461 showed marked inhibition of cholesterol absorption in cholesterol-fed hamsters, rats, dogs, rabbits and rhesus monkeys, but not in normal chow-fed animals. Although more studies are required, preliminary data indicate that this agent can potentiate the activity of statins when combined with them without interfering with lipid processing proteins.[150] The ACAT inhibitor, CL-283796 is being evaluated in phase II studies. These and other potential ACAT inhibitors may enhance the activity of 7-α-hydroxylase, the rate-limiting step in the

conversion of cholesterol to bile acid, without synthesising cholesterol at the same time.

Research continues to improve the antiatherogenic potential of ACAT inhibitors. For example, one ACAT inhibitor has the ability to inhibit TXA_2 formation, and several others have been modified with the addition of an antioxidant moiety.[19] The development of eldacimibe (ACA-147) may be the first ACAT inhibitor to demonstrate these pleiotropic effects. Eldacimibe has been reported to prevent atherosclerosis in cholesterol-fed rabbits with the ability to lower LDL levels, prevent LDL oxidation and increase HDL levels.[151] The ureidophenol derivative T-2591 demonstrated marked inhibition *in vitro* of copper ion and endothelial cell-induced LDL oxidation compared with probucol. This agent will be studied in animal models to evaluate its effects on atherogenesis.[152] Another method to enhance ACAT activity is to combine ACAT inhibitors with existing hypolipidaemic agents. Coadministration of atorvastatin plus CI-976 synergistically lowered total cholesterol and lipoprotein cholesterol in addition to reducing atherosclerotic lesions in cholesterol-fed rabbits.[153]

There are many ongoing clinical studies with ACAT inhibitors. Preliminary reports in humans suggest poor gastrointestinal tolerability, although the development of water-soluble ACAT inhibitors may solve this problem.[154] Overall, the use of ACAT inhibitors in the future appears promising. They may modify the atherosclerotic process by multiple mechanisms: preventing storage of lipids in the arterial wall, blocking cholesterol absorption in the intestines, preventing LDL oxidation and increasing serum levels of HDL.

4.4.2 Cholesteryl Ester Transfer Protein Inhibitors

Although the clinical implications have not been delineated, CETP inhibition has opened up a new pathway to study for drug development in an attempt to alter the regulation of lipid transport/metabolism. CETP is responsible for the transfer or exchange of cholesteryl ester carrying HDL and triglycerides in VLDL.[155] CETP allows for the disposal of cholesteryl esters which are deposited in HDL, thus enabling the lipoproteins to return to the smaller HDL_3, which is more suitable for interaction with the cell membrane. However, HDL_3 is more dense (higher protein content) and may possess a higher degree of atherogenicity. Cholesteryl ester transfer to VLDL is followed by lipolysis of VLDL into LDL, from which cholesteryl esters are eliminated by the high affinity receptor route.[33]

There is ambiguity in the understanding of the mechanism of CETP. The transfer of cholesteryl ester from HDL to LDL and VLDL by way of CETP occurs normally in reverse cholesterol transport. In some instances, this process may be detrimental since it transfers more cholesterol from HDL to the atherogenic LDL and VLDL. A support to this atherogenic hypothesis of CETP comes from its absence in rats, which exhibit a low incidence of atherosclerosis. A cohort of Japanese patients with CETP deficiency had high HDL levels and life longevity.[156] Inhibition of CETP may provide elevated HDL levels and shunt cholesterol away from atherogenic lipoproteins. CETP inhibition in rabbits demonstrated these marked increases in HDL levels.[35,157] In contrast, one study observed that Japanese men with reduced CETP levels were at increased risk for CHD.[158] The rationale was that they had inefficient HDL function because of their incapacity to dispose of stored cholesteryl esters, and on poor recognition of triglyceride-enriched LDL by the LDL receptors.[159] In spite of the ambiguity, CETP inhibition has been studied in humans. Future studies will need to address whether, and in whom, such agents can produce a net benefit.

Although the development of compounds to inhibit CETP is ongoing, current drugs such as probucol have been shown to raise CETP levels and reduce HDL_2 levels.[160] A synthetic isoflavan, CGS-25159, has produced favourable changes in lipoprotein profiles in hamsters by down-regulating CETP and subsequently increasing HDL levels.[161] HDL_2 is a larger lipid and contains more cholesterol than the smaller HDL_3. Therefore, inhibition of CETP increases the cholesterol-carrying capacity of HDL and may increase the efficacy of reverse cholesterol transport.

Among the various mechanisms exhibited by marine fish oils [such as 3-polyunsaturated fatty acids (3-PUFA)], they decelerate CETP activity and lower triglyceride levels, which may protect against CHD (discussed in section 5). In addition, oleate, a free-fatty acid, enhances binding of CETP to lipoproteins to be displaced later from the surface by lipid transfer inhibitor protein (LTIP). LTIP is a second plasma protein that binds to lipoproteins and inhibits CETP activity by displacing CETP from the lipoprotein surface. In laboratory experiments, oleate suppressed LTIP activity and allowed maximum CETP–mediated lipid transfer between all lipoproteins.[162] After 12 weeks, pravastatin 40 mg/day reduced CETP activity without changes in transfer of HDL-cholesteryl ester toward the denser LDL subfraction in 6 patients with HFH.[163]

One study demonstrated that the hog plasma peptides, P28 and P20, suppressed activity against CETP. P20 was injected into cholesterol-fed rabbits and one hour later 75% of CETP activity was suppressed for 30 hours. Cholesterol levels were reduced by 30% and HDL levels were increased by 32%. Cholesterol levels returned to pre-treatment levels after 48 hours.[164]

4.4.3 Pancreatic Cholesteryl Ester Hydrolase Inhibitors

The development of a novel pancreatic cholesteryl ester hydrolase (pCEH) inhibitor, WAY-121898, may have clarified that both pCEH and ACAT play a role in cholesterol absorption and may vary among different animal species and different dietary intakes. After parenteral administration of WAY-121898 in cholesterol-fed or chow-fed rabbits, inhibition of pCEH reduced liver cholesteryl ester content and cholesterol absorption but had relatively little effect on ACAT. However, when administered orally in cholesterol-fed hamsters, cholesterol was lowered without altering cholesterol absorption suggesting another mode of action, possibly inhibition of liver CEH.[165]

4.4.4 Enhancers of Lipoprotein Lipase Activity

Understanding the role of LPL activity has opened up pathways in the development of lipid regulation modifying agents. LPL deficiency has been associated with an increased risk of CHD.[166-168] In a process preceding CETP activity, LPL is responsible for VLDL catabolism with a subsequent loss of triglyceride and increase in HDL. Animal data have suggested that increased VLDL catabolism might lead to increased LDL levels resulting in an atherogenic lipid profile. However, fibric acid derivatives are indirect activators of LPL and they have favourable effects on serum lipids. In addition, the direct activator, NO-1886, enhances LPL activity and may exhibit antiatherosclerotic activity. This agent has lowered triglyceride levels, increased HDL levels and reduced atheroma formation in animals.[169]

4.5 Inhibitors of Cholesterol Synthesis

As already elucidated (in section 3.4), the statins inhibit the early rate-limiting step in cholesterol synthesis, the conversion of HMG-CoA to mevalonic acid. Mevalonic acid is necessary for the synthesis of cholesterol as well as nonsterol products such as dolichols and ubiquinones. Dolichols are necessary for glycoprotein synthesis. Ubiquinones are antioxidants that are involved in cell electron transport, serving as important component for muscle cell function. The inhibition of ubiquinones might block their beneficial antioxidant effects yet may explain the occurrence of myositis associated with the statins.[35,170]

A number of potential pharmacological strategies have been investigated in an attempt to inhibit cholesterol biosynthesis at a later step than HMG-CoA reductase in order to preserve the synthesis of non-sterol products. The inhibition of various steps in cholesterol synthesis requires effective LDL receptor activity. Inhibition of squalene synthetase has been reported to up-regulate LDL receptor activity, thereby enhancing removal of LDL cholesterol.[171] Squalene synthetase catalyses sterol biosynthesis by converting farnesyl pyrophosphate to squalene, which is later converted into cholesterol.[35,170] Squalestatin-1 (zaragozic acid A), isolated from phoma species, is a potent inhibitor of squalene synthetase in rat hepatocytes *in vitro* and in rat liver *in vivo*. Squalestatin-1 reduces the pro-

duction of farnesol formation in mice,[172] and it has reduced serum cholesterol up to 75% in marmosets.[170,173] Oral administration of RPR-107393 to rats and marmosets at a dosage of 30 mg/kg twice daily and 20 mg/kg twice daily, respectively, produced a greater reduction of cholesterol than lovastatin or pravastatin.[174] Squalene synthetase inhibitors or squalestatins have poor bioavailability and are therefore difficult to deliver to the desired site of action.

Another strategy is to inhibit squalene epoxidase, which prevents the conversion of squalene to cholesterol. Two examples of this class of agents, TU-2078 and NB-598, have been reported to inhibit squalene epoxidase formation leading to decreased intracellular concentration of cholesterol by activation of LDL receptors.[170,175,176] However, this increases the levels of squalene in the human liver, and the consequences of this are unknown at this time.

Other potential targets of cholesterol biosynthesis include oxidosqualene cyclase or feedback inhibition of HMG-CoA reductase expression with polar sterol derivatives.[177] Another potential area of cholesterol reduction is inhibition at an earlier step through the interference with the conversion of acetyl-CoA.[178] Specific inhibitors of sterol production of farnesyl/pyrophosphate:protein farnesyl transferase is another approach in slowing SMC proliferation, thus regulating vascular growth in the atherosclerotic process.[179] Although not fully elucidated, the consequences of depleting squalene-derived products may also alter the rate of transcription, HMG-CoA reductase, mRNA stability or translation of key proteins involved in cholesterol homeostasis.[180]

4.6 Monocyte and Macrophage Inhibitors

Monocyte recruitment to areas of endothelial damage, subsequent penetration through the intima into extravascular space to form macrophages, and secretion of cytokines (IL-1, TNF-α) are early steps in the atherosclerotic process. The vast majority of the macrophages then engulf oxidised LDL to produce foam cells. More specifically, the pathway involves inflammatory activation of monocytes, which synthesise secretory metabolites of the arachidonic acid cascade (activation of eicosanoids) including prostaglandins (PGE_2) and leukotriene B4 (LTB_4). Monocytic production of TXA_2 and PGE_2 is via the cyclooxygenase (COX) pathway as opposed to leukotrienes which are formed from the lipoxygenase pathway, and function as regulators of allergic and inflammatory reactions. LTB_4 is the most potent chemoattractant and activator of other monocytes and leucocytes.

Although alteration in dietary lipids may modulate the monocytic response, specific therapeutic strategies that target monocyte function or endothelium-provoked factors to inhibit atherosclerotic development have been investigated. PUFA and their derivative eicosapentaenoicacid (EPA) antagonise TXA_2, PGE_2 and LTB_4 production by monocytes because they serve as substrates or compete with arachidonic acid for metabolism products of both the COX and lipoxygenase pathways. Thus, the production of EPA metabolites has reduced pro-inflammatory activity. In addition, 3-PUFA may inhibit IL-1 and platelet activating factors which may affect the atherosclerotic process.[35,181] After 8 weeks administration in rabbits of PD-146176, a specific leukotriene inhibitor, the progression of monocyte-macrophage enrichment of an atherosclerotic lesion was decreased without changes in cholesterol levels.[182] Current leukotriene antagonists are promising agents in the treatment of other inflammatory conditions including asthma; however, future interventional studies may provide alternative options in the treatment of atherosclerosis.

With regard to other studied agents, the use of oleic acid or ACAT inhibitors to reduce IL-1 in the monocyte-macrophage cell line has been unsuccessful to date.[183] The statins may have a more pivotal role in reducing macrophages, which may have contributed to their success in altering the atherosclerotic process.[100]

4.7 Thyromimetic Therapy

Thyroid hormones have potent cholesterol-lowering action due to the up-regulation of LDL recep-

tors. Dextrothyroxine (DT4) was among the first commercialised agents used for this effect. However, the large doses required led to potentially serious cardiovascular adverse events and, therefore, limited its use.[184] Contamination of DT4 with thyroxine may have sensitised the myocardium to epinephrine (adrenaline) and induced life-threatening arrhythmias. Recently, research has dissociated the thyromimetic effects from the hypolipidaemic properties, leading to thyroxine analogues with little to no adverse cardiovascular effects.[19]

One such example is CGS-26214, a thyroxine compound with a fluorinated ring. This compound is hepatoselective, sparing the drug from cardiac and thermogenic effects.[185] It produced a reduction in LDL of 31% in hypercholesterolaemic rats at a dose of 1 μg/kg, which was equivalent to that obtained with 25 μg/kg of liothyronine.[186] It produced a reduction in cholesterol in the rats and in normocholesterolaemic dogs in a dose-dependent manner. In normal chow-fed monkeys, CGS-26214 reduced Lp(a) levels by 43% at 30 μg/kg as well as LDL levels. This is consistent with what is observed in studies of hypothyroid patients receiving thyroid hormones.[187] In patients with hyperthyroidism receiving drugs such as thiamazole (methimazole), Lp(a) levels have been observed to increase once a euthyroid state is achieved.[188]

Another thyroxine analogue with cardiac sparing effects, CGS-23425, reduced LDL levels by 44% in hypercholesterolaemic rats at a dose of 10 μg/kg. In addition, there was a dose-dependent increase in apo A-I, an activator of LCAT which promotes cholesterol transport from peripheral tissues.[189] This feature may be useful for both prevention and reversal of atherosclerosis. Both CGS-23425 and CGS-26214 promote the clearance of chylomicrons after a fat load, perhaps because of their selectivity for the liver.[19,185]

Thyroxine analogues under investigation may be useful because of their ability to lower LDL and Lp(a) levels, increase apo A-1, and reduce postprandial fat load in patients with hypercholesterolaemia without any serious cardiovascular adverse effects as observed with earlier compounds.

4.8 Cholesterol Vaccination

Cholesterol vaccination has been proposed and tested as a technique for lowering serum cholesterol by enhancing clearance of lipoproteins via the reticuloendothelial system (RES).[190] Antibodies to cholesterol occur naturally in human plasma. The theory behind vaccination is to induce antibodies, which will bind to cholesterol in circulating VLDL, LDL or IDL, thereby opsonising them for removal by scavenger macrophages, especially Kuppfer cells. The up-regulation of LDL receptors would further lower serum LDL levels.[191,192]

Since cholesteryl esters are the predominant form of cholesterol found in both serum and atherosclerotic plaques, synthetic antigens containing cholesterol esters covalently coupled to various carrier proteins as haptens have been synthesised. One study demonstrated the effects using both bovine and human albumin cholesterol-sebacate antigens in atherogenic fat-fed rabbits. The rabbits were immunised weekly for 6 weeks, followed by booster shots at 3 to 4 week intervals for up to 15 weeks. The immunised rabbits showed a 25 to 35% decrease in total cholesterol levels and 90% suppression of aortic atherosclerotic plaques.[190] Another part of this study demonstrated beneficial effects of immunisation administered during 9 months even when the rabbits were fed cholesterol for the entire period. However, plaque lesions gradually returned after the 9-month period. The maximum reduction in serum total cholesterol by these synthetic antigens was an average of 330 mg/dl (8.54 mmol/L). This capacity is of interest since serum cholesterol levels in cholesterol-fed rabbits may exceed 1000 mg/dl (25.86 mmol/L) whereas in human's, serum cholesterol levels are in the 200 to 450 mg/dl (5.17 to 11.64 mmol/L) range. The reduction observed may be the maximum capacity of the RES to eliminate serum cholesterol.

Another study demonstrated inhibition of neointimal response to balloon injury in hypercholesterolaemic rabbits after immunisation with homologous oxidised LDL.[193] The 2 studies described indicate that cholesterol immunisation should be tested under conditions that do not exceed the capacity of

the RES, and that are more typical of the human population. In addition, a strategy for vaccination against atherosclerosis and restenosis is necessary.

4.9 Somatic Gene Therapy

It may take several years before somatic gene therapy makes an impact as a lipid-regulating agent. One method is to insert the gene coding for normal LDL receptors into liver cells. This is a 'liver-target' genomic treatment using viral vectors such as retroviruses or adenoviruses implanted into cells drawn from the patient. The gene expression must be stable and nontoxic, and the delivery mechanism should be safe and efficient.[194]

The absence of LDL receptors in HFH serves as a model for investigating gene therapy in CHD. One study, using retrovirus-mediated LDL receptor gene therapy, reduced LDL by 6 to 23% in 3 of 5 patients with HFH.[195]

Adenoviruses may prove more useful than retroviruses as viral vectors because of their high efficiency at transfection and their effectiveness in nonreplicating cells. Delivery of human LDL receptor DNA via adenovirus *in vivo* to WHHL rabbits resulted in decreased serum cholesterol.[196] However, the transgene expression was stable for only 7 to 10 days and cholesterol levels returned to baseline within 3 weeks. Subsequent doses were largely ineffective because of the development of neutralising antibodies.

Halting the atherosclerotic process may be accomplished by regulating vascular growth or tone without specifically altering lipoprotein concentrations. Vascular endothelial growth factor (VEGF), a regulator of angiogenesis, may be used to stimulate collateral vessel growth in patients who are not eligible for angioplasty, but who have occluded superficial arteries. The effects of VEGF are particularly mediated by stimulated NO and epoprostenol (prostacyclin) secretion in the vascular wall. Gene delivery of VEGF has been accomplished in this setting using a coated balloon catheter.[197,198] Gene transfer studies using VEGF are of great interest in the treatment of coronary atherosclerosis, postangioplasty restenosis, postbypass atherosclerosis, PVD and graft failures.[199,200] Fibroblast growth factors (FGF) act on vascular endothelium and angiogenesis. Adenovirus–mediated expression of these FGF have induced cellular proliferation and angiogenesis in mice.[201] Administration of antibodies against PDGF that cause cell proliferation have also limited neointimal response to coronary balloon injury in rats.[202]

Another possibility is transfection of apo E, which is capable of directly interacting with non-LDL-receptor-regulated pathways, such as lipoprotein receptor-related proteins.[203] The removal of the apo E gene in animals has caused abrupt, non-diet-induced, severe atherosclerosis, whereas the overexpression of apo E has made the animals resistant to diet-induced atherosclerosis.[204] Adenovirus-mediated gene transfer of human LPL ameliorates hyperlipidaemia associated with apo E and LDL-receptor deficiencies in mice.[205]

A simpler approach is by an *in vivo* system, in which viral DNA is directly injected or genes are positioned in specific territories. Viral DNA may be directly delivered at the arterial level onto plaques or artery thromboses via angioplasty.[33] Gene delivery may also include adventitial, intravascular and intramuscular gene transfer. However, current efforts are directed toward improving gene transfer and vector development making it more efficient and less immunogenic.

Another strategy is to inhibit the expression of certain genes. Antisense oligonucleotides can be used to control gene expression and protein synthesis at several points. Potential applications include inhibiting SMC proliferation in the arterial wall. Studies have shown that antisense oligonucleotides inhibit SMC proliferation *in vitro* and in animal models directed at *c-myb* and *c-myc* genes.[206,207] Perhaps another possibility is the overexpression of apo A-I or LCAT by adenovirus-mediated gene transfer in patients with atherosclerosis associated with low HDL levels. Overexpression of apo A-I and LCAT may increase serum HDL levels to improve reverse cholesterol transport.

Current clinical trials have shown that gene therapy (therapeutic angiogenesis) may provide a

promising new approach to dyslipidaemia, atherosclerosis and restenosis.[208] Two studies are currently enrolling patients treated with adenovirus-mediated $VEGF_{121}$ and FGF-4 gene transfer in CAD and/or CABG, and CAD, respectively. These findings are exciting in that the results emphasise the potential importance of targeting growth factors. In addition, NO synthesis gene therapy may provide a rapid amelioration of vascular markers of inflammation involved in atherosclerosis, which was demonstrated in rabbits.[209]

4.10 Recombinant Proteins

Research in molecular biology has led to the use of recombinant proteins for parenteral administration. The administration of recombinant apo E may significantly improve lipoprotein metabolism. As previously discussed, apo E is a scavenger molecule that has a role in reverse cholesterol transport. Apo E-containing lipoproteins have a high affinity for liver cell membranes, which are responsible for the uptake of dietary cholesterol via chylomicron remnants. Injections of apo E into WHHL rabbits increased chylomicron remnant clearance, thereby reducing cholesterol levels.[210,211] One study infused apo E 70mg into rabbits and demonstrated a reduction in serum cholesterol of 20 to 40% within 2 to 3 hours.[212] Apo E 30mg 3 times per week for 8.5 months in rabbits resulted in a 40% reduction of aortic lesions.[213] Although parenteral administration of apo E appears to be promising, the reduction in lesions has not been associated with significant falls in cholesterol levels.

The association between elevated HDL and reduced cardiovascular risks are well established. The use of whole HDL or of apolipoproteins dramatically reduced aortic lesions in rabbits in homologous HDL-very high density lipoprotein (VHDL) lipoprotein fraction-treated groups. The elevated HDL also caused a regression of atherosclerosis in rabbits with established lesions.[214] The responsible component for the resistance to form atherosclerotic lesions in the aortic vessel wall in rabbits fed a high-fat diet was thought to be due to apo A-I. In addition, HDL exerts a epoprostenol-stabilising property attributed to the apo A-I component,[215] and apo A-I may have direct fibrinolytic activity as seen *in vitro*.[216]

Studies have demonstrated the direct effects of apo A-I and HDL in inhibiting atherosclerotic progression and stimulating regression of atherosclerosis.[217,218] More specifically, the use of the recombinant dimer, apo $A\text{-}I_{Milano}$ ($A\text{-}I_M$), which is a natural mutant for apo A-1, has been associated with a reduced risk of arterial disease.[219] In addition, the dimer may have fibrinolytic activity.[220] Two cholesterol-fed rabbit studies using recombinant $A\text{-}I_M$ dimer showed similar reductions in restenosis and decreased SMC proliferation, one in peripheral angioplasty and the other in carotid perivascular manipulation.[221-223]

The effects of apo A-I and HDL in humans requires extensive and well-controlled clinical studies. Other recombinant proteins under study to reduce atherosclerotic lesions include monoclonal antibodies against arterial graft factors and direct antagonists of cell proliferation, such as leukaemia inhibiting factor.[224]

4.11 Prevention of LDL Oxidation

As briefly discussed in sections 1 and 2, in addition to limiting circulating lipids and altering lipoprotein metabolism, therapies are directed at protecting the arterial wall against atherogenesis. Multiple mechanisms are required to halt or prevent the atherosclerotic process. Among them are slowing LDL oxidation in the subintima and enhancing epoprostenol production. Others include blocking the production of growth factors associated with atherogenesis, such as PDGF, and increasing the production of growth factors that improve vascular tone, such as NO. These mechanisms are briefly discussed here.

Oxidation can damage lipid and protein components of LDL particles.[8] Free radicals *in vitro* damage unsaturated fatty acid side chains, which generate more free radicals leading to a continuous cycle of oxidative damage and rancidity in the core of the LDL particle. Furthermore, this process damages apolipoproteins causing both fragmentation and

aggregation. Oxidised LDL is cytotoxic to endothelial cells, SMCs and fibroblasts,[225] and more potent than glycated LDL in stimulating foam cell formation after uptake by macrophage scavenger receptors. Oxidised LDL, acting as a potent chemotactic factor also promotes leucocyte recruitment and adhesion to the endothelium.[226] LDL glycation is a nonenzymatic reaction of glucose with apo-B, the surface protein of LDL. It is accelerated in patients with diabetes thereby contributing to the atherogenic process. Atherosclerosis may be associated with oxidative modifications of both protein and LDL particles. One study showed that antioxidants inhibited oxidised epitopes on human serum albumin and apo A-I as well as LDL particles.[227] This suggests that atherosclerosis is not only caused by oxidised LDL. More study is needed to determine the effects of oxidative modifications on atherogenesis *in vivo*, as the proposed effects remain largely hypothetical.

Although inconsistent, a number of antioxidants including probucol, tocopherol, ascorbic acid, β-carotene, selenium and others have been observed to inhibit diet-induced atherosclerosis in animal models and in humans.[228] Under normal circumstances, endogenous antioxidants are available in the plasma to prevent native LDL oxidation. Several observational data suggest that dietary and supplemental antioxidants may lower the risk for CHD.

Probucol, in addition to its lipid-lowering properties, prevents LDL oxidation and subsequent foam cell formation *in vitro*.[35] In animal models, probucol prevents atherogenic lesions, possibly because of its antioxidant properties.[229] One study demonstrated that probucol may reduce atherogenesis by mechanisms not shared by all antioxidants. The combination of 0.025% probucol plus tocopherol 1000 IU and 0.05% of the probucol analogue, BM-150639 was compared with varied dose probucol plus placebo in 10 LDL receptor-deficient rabbits. In the varied dose probucol group (average 0.091% probucol), there was a reduction of aortic atherosclerosis by 51.7% versus the untreated group ($p < 0.005$). However, both treatment groups had similar antioxidant prevention of plasma LDL. Lag-time measurement of the formation of conjugated dienes provides a measure of resistance to LDL oxidation and is an indicator for atherogenic risk. However, it seems that both tocopherol and probucol sufficiently prolonged the lag-time before LDL oxidation, but there was only a weak correlation with atherosclerotic activity in this study.[230,231] The investigational probucol analogue, MDL-2831, reduced triglycerides and nonesterified fatty acids without losing its antioxidant property in diabetic animals.[232] However, as shown by the PQRST trial, there is conflicting data for the potent antioxidant probucol. Although probucol reduces the development of experimental atherosclerosis and improves endothelial function, there was no decrease in progression of PVD.[83]

Tocopherol has demonstrated a strong and independent inverse association with CAD.[233] Tocopherol protects LDL from lipid peroxidation and heavy oxidative modification of LDL may occur if tocopherol is depleted. Increasing the plasma concentration of tocopherol may delay the oxidation of LDL.[234] Tocopherol 800 IU/day reduced thiobarbituric acid reactive substance (TBARS), a measure of oxidation and lipid peroxidation products, in patients with hypercholesterolaemia.[235] In addition, tocopherol 600 IU/day or ascorbic acid 1.5 g/day prevented an increase in plasma TBARS, native LDL and smooth muscle-containing LDL associated with cigarette smoking.[236] Among the vitamins, tocopherol shows the most consistent effects at lowering LDL oxidation. β-carotene and ascorbic acid have no effect to a mid to moderate effect on oxidative ability.[237] β-carotene inhibits LDL peroxidation and macrophage modification of LDL, thereby decreasing the uptake and degradation of oxidised LDL by the macrophage scavenger receptor *in vitro*.[234] In theory, it should have antiatherogenic effects. However, in the Alpha-Tocopherol, Beta Carotene Study that was not designed to primarily evaluate cardiovascular endpoints, β-carotene supplementation failed to show clinical benefit.[238] Ascorbic acid is a water-soluble antioxidant that inhibits LDL peroxidation by destroying aqueous

free radicals before they enter the lipid phase. When ascorbic acid is depleted and radicals enter the lipid phase, tocopherol is the primary free radical scavenger. Ascorbic acid also helps regenerate the active form of tocopherol by reducing the tocopheroxyl radical. Ascorbic acid has only had a weak correlation with the incidence of CAD.[234] The combination of tocopherol, ascorbic acid and β-carotene did reduce LDL oxidation, but may not be superior to tocopherol administration alone.[239,240]

In vitro selenium has also demonstrated an antioxidant effect on LDL. Selenium is an integral part of the antioxidant enzyme, glutathione peroxidase.[241] Levels of this naturally occurring intracellular antioxidant enzyme may be reduced in patients with atherosclerosis. One study in rabbits that were given selenium (glutathione) supplementation (22 μg/day) to the diet demonstrated an increase in glutathione peroxidase levels.[242] This may prevent atherosclerosis when combined with other antioxidants such as tocopherol and ascorbic acid. These results were significant compared with placebo (without selenium) or positive control (probucol, mean dose of 406 mg/day). After 22 weeks, tocopherol plus selenium equally inhibited aortic atherosclerosis compared with probucol via a mechanism that is probably independent of effects on lipoprotein levels.

Among the investigational antioxidants, agents such as pimagedine (aminoguanidine; pimagedine hydrochloride) or its derivatives may have a future role in reducing LDL glycation and oxidation.[8,243] In Europe, another antioxidant, silibinin (CAS 22888-70-6), is currently being evaluated for its effects on LDL oxidation and vascular smooth muscle proliferation *in vitro*.[244] Earlier laboratory experiments showed that the drug prolonged the lag-time of both LDL auto-oxidation and copper ions, as assessed by diene formation. Other agents like N-acetylcysteine (NAC) and thiazolidine-4-carboxylic acid have reduced the effects of oxygen radicals *in vivo*. One study showed a reduction of matrix-degrading capacity of macrophage-derived foam cells *in vivo* and *in situ* by NAC in cholesterol-fed rats.[245] More research is required to establish these utilities in the prevention of CAD.

Current drugs such as the statins may prevent LDL oxidation. Lovastatin (40 mg/day) and pravastatin (40 mg/day) reduced LDL oxidation induced by copper ions by 30 and 37%, respectively, after 24 months of treatment.[246]

With a better understanding of the process of LDL oxidation, pro-oxidants such as homocysteine and iron have been found to be associated with CAD. Intake or blood levels of folate are inversely related to levels of homocysteine. Therefore, a reduction of blood homocysteine levels may be achieved with supplemental folic acid and/or combination with pyridoxine and cyanocobalamin. Possible mechanisms for hyperhomocysteinaemia's negative effects in CAD are that homocysteine is toxic to the endothelium, is prothombotic, increases collagen production and decreases the availability of NO. Reduction of homocysteine levels by folate, pyridoxine and cyanocobalamin supplementation should therefore, prevent endothelial dysfunction and improve coronary outcomes. Clinical trials are ongoing to determine if these co-factors can prevent or cause the regression of atherosclerosis.[9,247]

There are substantial differences in the biological properties of various antioxidants mentioned in this section and data are limited. Few conclusions can be drawn about what role antioxidant therapy will ultimately have in the prevention of atherosclerosis and clinical outcomes. Hopefully, ongoing clinical trials in the secondary prevention of CAD will confirm earlier findings and show evidence of the benefit of reducing new cardiovascular events.

The Cambridge Heart Antioxidant Study (CHAOS) evaluated the effects of high dose tocopherol 400 IU to 800 IU compared with placebo, at reducing the risk of MI in 2002 patients with angiographically proven coronary atherosclerosis. The results demonstrated that tocopherol therapy lowered the risk of the combined end-point of death and nonfatal MI by 47%. However, there were more cardiovascular deaths in the tocopherol group compared with placebo. Therefore, the authors concluded that the benefit was due to a reduction in nonfatal MI.

They postulated that the reason for the increase in cardiovascular deaths may be related to differences in antioxidant effects on the biological processes leading to death and MI.[248] The Lyon Diet Heart Trial also showed benefit for a Mediterranean diet high in fruits and vegetables.[249] The adverse event reduction in this trial and in the CHAOS study was not associated with change in cholesterol levels. The Women's Antioxidant and Cardiovascular Study (WACS) is an ongoing randomised, double-blind, placebo-controlled secondary prevention trial to observe the benefits of tocopherol, ascorbic acid, β-carotene and/or placebo in 8000 females ≥40 years of age with CHD. The primary end-points include nonfatal MI, stroke, coronary interventions and overall mortality.[250] Angiotensin converting enzyme (ACE) inhibitors have demonstrated antiatherosclerotic and antioxidant activity unrelated to lipid-lowering effects.[251] The Heart Outcome Prevention Evaluation (HOPE) trial is currently evaluating the effectiveness of the ACE inhibitor ramipril and tocopherol 400 IU/day on the progression of CAD in patients with existing CHD. The 5-year trial has completed enrolling patients and is in the process of follow-up evaluations of the patients. The preliminary results appear promising; an additional 2-year follow-up has been initiated to the tocopherol-treated arm since there may be a trend toward lower risk of cancer. The ramipril-treated arm was prematurely discontinued because of positive results, but the final published results are eagerly anticipated.

4.12 Other Developing Modalities

Currently, there is much interest in additional, non-LDL related therapeutic strategies to prevent CAD. As previously stated in section 1, atherosclerosis is considered an inflammatory disorder. The formation of atherosclerotic lesions is a multicellular process in which lipids and the extracellular matrix accumulate in the intima of arteries and a local inflammatory response is elicited secondary to activation of macrophages, T-lymphocytes, SMCs and endothelial cells. Various researchers have targeted these strategies and they are briefly discussed here. The studies are limited to animal data but may carry clinical significance in the future for humans.

4.12.1 Peroxisome Proliferator-Activated Receptors Activators

PPARs are transcription factors belonging to the nuclear receptor gene family.[252,253] The three subtypes α, δ and γ function as ligand-dependent transcription factors by binding to specific response elements termed peroxisome proliferator-response elements and regulating the expression of target genes. PPAR-α and -γ are known to regulate the expression of genes involved in lipid metabolism.[254] They have also demonstrated inhibition of inflammatory cytokine production by stimulated monocytes/macrophages as well as production of MMPs, which are associated with atherosclerotic plaque rupture.[253,255] Thus, there is the hypothesis for the function of PPARs at the level of the vascular wall which, independent of their role in lipid metabolism, could modulate the pathogenesis of atherosclerosis.

Some natural prostaglandins and synthetic antidiabetic thiazolidinedones [ciglitazone, troglitazone, rosiglitazone (BRL-49653)] are ligands for PPAR-γ, while hypolipidaemic fibrates and certain eicosanoids (LTB_4) are ligands for PPAR-α.[252,253] Treatment with troglitazone inhibited SMC proliferation and decreased the intima and media thickness of carotid arteries in humans.[256]

Fibrates may inhibit apo C-III transcription by this process.[257] In cholesterol-fed rabbits, fenofibrate, a new fibrate, decreased atherosclerotic plaque formation in thoracic aorta, in the absence of any effect on lipoprotein levels.[258] Fenofibrate also lowered CRP, fibrinogen and IL-6 levels, all of which are markers of inflammation.[259] In the BECAIT and LOCAT intervention trials, the fibrates prevented the progression of coronary atherosclerosis, in the absence of marked lowering of plasma atherogenic lipoprotein levels.[55,56] Another PPAR-α selective activator, WY-14643, also inhibited inflammatory responses in human aortic SMCs.[259] These findings carry enormous clinical significance, particularly in patients with diabetes.

4.12.2 Matrix Metalloproteinases Inhibitors

Extracellular matrix turnover is integral to the pathology of atherosclerotic plaque development. The erosion of the fibrous cap, plaque rupture and MI are associated with attenuation of the extracellular matrix.[260] This has led to the hypothesis that an altered balance between matrix degrading enzymes and their endogenous inhibitors is a key factor in determining the stability of the plaque cap.[261] Matrix degrading metalloproteinases (MMP-2,-9) are believed to play a primary role in the turnover of the vascular extracellular matrix. Increased levels of MMPs show increased expression and/or activation in atherosclerotic plaques.[261,262] They may also be correlated with lesion severity.[263] These MMPs and their proteolytic enzymes are tightly controlled by endogenous tissue inhibitors of matrix metalloproteinases (TIMP-1, -2, -3).[262] However, it is not clear if sufficient amount of TIMPs are produced to inhibit the increased activity of MMPs in atherosclerosis. It is presumed that an excess of MMPs over TIMPs promotes extracellular matrix turnover. Studies have demonstrated this excess of MMPs over TIMPs in human atherosclerotic plaques and plaques from balloon-injured aortas and iliac arteries from cholesterol-fed rabbits.[261,264] This suggests a possible therapeutic role for synthetic MMP inhibitors or TIMP gene therapy.[265,266]

4.12.3 Scavenger Receptor Class B Type I Enhancers

The HDL receptor SR-B1 mediates the selective uptake of plasma HDL cholesterol by the liver and steroidogenic tissues. As a consequence, SR-BI can influence plasma HDL cholesterol, HDL structure, biliary cholesterol levels, and the uptake, storage and utilisation of cholesterol by steroid hormone-producing cells.[267] The finding that overexpression leads to significant increases in biliary cholesterol content is consistent with gene-targeting studies that suggest an important role for SR-BI in reverse cholesterol transport.[268-270] In addition to HDL, SR-BI can also bind to other ligands, including LDL, VLDL, apo's and can mediate efflux of unesterified cholesterol from cells to HDL.[271-273]

The theory is if the activation of SR-BI increases reverse cholesterol transport and increases HDL cholesterol, overexpression may promote antiatherogenesis. The antiatherosclerotic effect from adenovirus– or transgene–mediated hepatic overexpression of SR-BI has been shown in cholesterol and fat-fed mice.[267,274]

Pharmacological stimulation of endogenous SR-BI activity may also produce antiatherogenic effects, possibly because of its importance for reverse cholesterol transport. Some hypolipidaemic agents have shown that they can influence HDL concentrations and the process of reverse cholesterol transport. For example, probucol, a hydrophobic antioxidant drug, when administered to mice, showed a 2-fold increase in selective uptake of cholesteryl esters compared with control mice after being incubated with SR-BI–transfected ovary cells.[275] Although the study did not show overexpression of SR-BI, probucol or its metabolites increased selective cholesteryl ester uptake *in vivo* by modifying HDL in a way that caused enhanced interaction with SR-BI. This mechanism may be partly responsible for the effects seen of probucol on atherosclerosis and restenosis.

PUFA have also been shown to up-regulate SR-BI expression and HDL cholesteryl ester uptake in hamsters.[276] Substitution for saturated fatty acids with PUFA in the diet increased expression of SR-BI receptors and lowered plasma HDL cholesteryl ester levels but did not affect reverse cholesterol transport, although the plasma HDL was low.

Interestingly, there may be treatment available in the forseeable future for preventing loss of paroxonase, a fat-complex protein from HDL that prevents LDL triggering vascular inflammation. Exogenous supply of these enzymes may counteract the adverse effects of LDL.[277]

5. Nutritional Strategies

Lifestyle modifications including diet, exercise, smoking and decreasing alcohol intake are essential in the management of dyslipidaemia. Strict dietary intervention is important for prevention and treatment of CHD since it may lower cholesterol

by at least 6%.[278] A diet low in total and saturated fat and cholesterol, and high in complex carbohydrates is the cornerstone of treatment in hypercholesterolaemia. Dietary interventions have been specified for individuals with or without CHD according to their baseline LDL levels.[20] The American diet typically contains 36 to 37% of calories from fat. The NCEP guidelines advocate less than 30% of total calories from fat; including less than 10% as saturated fat with the remaining 20% equally distributed between polyunsaturated and mono-unsaturated fat (Step I diet).[20] The Step II diet further restricts saturated fat.

In terms of dietary fats, saturated fats, as in meat, containing long-chain stearic acid and short-chain fatty acids [caproic acid (10 : 0) and caprylic acid (8 : 0)] are less likely to increase LDL levels compared with palmitic, myristic and lauric fatty acids.[35,279] Therefore, the consumption of various dietary fats may or may not offer benefits in preventing atherosclerosis. For example, polyunsaturated fat compared with oleic acid (monounsaturated fat) is more likely to promote LDL oxidation.[280] Thereby, oleic acid may also directly interfere with the inflammatory response that characterises early atherogenesis and modulates gene expression for endothelial leukocyte adgesion molecules.[281] Thus, a diet rich in fish oil such as 3-PUFA may reduce VLDL and triglyceride levels. In addition, there is evidence that 3-PUFA may possess anti-atherosclerotic and antithrombogenic activity.[282] Epidemiological studies have shown a 44% reduction in all cardiac-related death and a 67% reduction in risk of sudden death from MI in patients consuming 35g of fish versus those who did not consume fish.[283] In addition, dietary intake of 3-PUFA 4 g/day for 12 weeks has reduced triglycerides (mostly VLDL) by 24%.[284] Thus, fish oil may potentially be combined with other lipid-lowering agents as a third-line agent for the treatment of type IV/V hypertriglyceridaemia in patients not able to tolerate nicotinic acid or fibric acid derivatives. However, consumption of diets rich in fish oil of any kind may slowly promote LDL oxidation because they are polyunsaturated fats. This was demonstrated in one study where tocopherol levels declined in response to diets containing large amounts of fish oil.[285] This may represent accelerated depletion of tocopherol in the environment of increased free-radical formation.

As described in earlier sections, dietary interventions such as soluble fibres, fish oils, synthetic or substituted fats, and natural dietary antioxidants may slow down the atherosclerotic process. One study showed that a 2oz serving (56g) of oat bran or oatmeal lowered LDL by 15%.[286] The dose-dependent reduction in LDL levels was due to the amount of oat bran, which contains the soluble fibre, β-glucan. Another study demonstrated a similar trend using the soluble fibre, psyllium hydrophilic mucilloid.[287]

Dietary flavonoids (as in red wine) may also have antioxidant properties, but their association with CAD in humans is uncertain. Dietary supplements, including medical foods (e.g. Heartbar), of arginine,[288] a NO precursor, enhanced endogenous NO formation *in vivo* to reduce monocyte accumulation and cell proliferation in cholesterol-fed rabbits.[288,289] In humans, intracoronary arginine showed improved endothelium-derived NO in dysfunctional coronary segments in patients with atherosclerosis[290] and syndrome X.[12,291]

In summary, regular exercise, alcohol restriction, smoking cessation, reducing dietary fats and supplementation with soluble fibres, fish oil or various antioxidants may prevent atherosclerotic progression. With regard to a low-fat diet, however, the benefits are inconclusive. Independent of cholesterol lowering, high-fat diet may also impair endothelial function and increase thrombogenicity, yet substitution of a low-fat diet may not fully negate these harmful effects. A diet high in polyunsaturated fats may enhance LDL oxidation, possibly by overwhelming mechanisms of oxidative metabolism. On the other hand, LDL oxidation may be prevented by various dietary interventions, which could affect the atherosclerotic process in a beneficial way.

6. Recommendations

With all the information available on the prevention of atherosclerosis and the reduction of cardiovascular events, there is still a need to consider the best treatment options for individual patients. It is important to follow NCEP guidelines, as these remain the minimum standard of practice for treating hypercholesterolaemia. It is imperative that patient compliance to diet, lifestyle modifications and drug therapy are strongly enforced. With drug therapy, at least 2 years of continuous lipid-lowering therapy is required before any reduction in the risk of CHD is seen.[292] In the case of statins, at least one year of drug adherence is required to produce clinical benefits. Many patients withdraw from drug therapy during the first year, leading to inadequate control. All clinicians can improve patient compliance to NCEP guidelines. The importance of lowering LDL is far less pervasive among US adults. Approximately only 18 to 46% achieve NCEP-defined target LDL goals. The importance of meeting LDL goals, especially in those patients with CAD (≤100 mg/dl) for secondary prevention, were demonstrated where aggressive therapy with statins was able to achieve LDL goals and favourable CHD outcomes.[21,293] The publication of NCEPIIb is forthcoming and eagerly anticipated. These updated recommendations may reflect the current emerging risk factors for CHD such as elevated homocysteine, Lp(a) and triglyceride levels, low HDL levels, and subgroups such as the elderly, postmenopausal women and people with diabetes.

Although current drugs, when added to low-fat diet and lifestyle changes, are effective, ongoing research on lipids is identifying new and better alternatives that may prove well tolerated and effective. LDL cholesterol is still the most used surrogate marker, but clinical outcomes are favoured. There is a movement towards more aggressive (<85 mg/dl) versus moderate lowering of serum lipids. Although this is not unequivocally established, it is supported by clinical trials. Indeed, LDL apheresis using dextran sulfate cellulose column has received approval in the US for patients with inadequate response to diet and drug therapy with LDL cholesterol levels ≥200 mg/dl (5.17 mmol/L) and CAD, or LDL cholesterol levels ≥300 mg/dl (7.76 mmol/L) without CAD.[294] The treatment of hypercholesterolaemia is being applied to the entire spectrum of patients – those with CHD and those at seemingly low-risk for CHD. Whenever possible, clinical trial data can and should be applied to individual treatment decisions. The new treatments discussed in this review may prove valuable as a substitute and/or in combination with current drugs. There is a great need to conduct well designed clinical trials to assess emerging agents and their combination with existing agents, in order to assess efficacy and, moreover, safety.

7. Conclusion

With improvements in the understanding of the pathophysiology of dyslipidaemia and what makes lipoproteins atherogenic, the discovery and development of agents in the future will focus on the specific causes of the disease. Since atherosclerotic lesions can occur even with marked plasma LDL reduction, other atherogenic or thrombogenic influences contribute greatly to atherogenesis and subsequent CAD. This is evident with the current agents, nicotinic acid, probucol, fibrates and statins, in that their multiple target effects have produced beneficial effects as demonstrated in clinical trials. Newer derivatives of current agents will hopefully be developed with improved safety profiles.

In the near future, new pharmacological agents will reduce serum cholesterol levels and slow or reverse the progression of atherogenesis. ACAT inhibitors reduce fat and cholesterol absorption. The development of eldacimide is promising since the drug possesses multiple target effects such as lowering LDL, increasing HDL and protecting against LDL oxidation. Agents such as the CETP inhibitors that increase HDL levels could enhance reverse cholesterol transport. Various dietary and exogenous antioxidants prevent LDL modification. Moreover, alterations of macrophage formation may also halt atherogenesis. In addition to the statins, the squalene synthetase inhibitors reduce the synthesis of cholesterol. Gene therapy and cholesterol vaccina-

tion may be valuable in patients with diminished, defective or missing hepatic LDL receptors. Parenteral recombinant proteins such as apo E and A-I may increase HDL levels and enhance the reverse cholesterol transport process. In addition, fully understanding the role of non-LDL–related targets such as PPARs, MMPs and SR-BI may bring about new treatment modalities.

Currently, aggressive therapy with agents possessing pleiotropic effects or combination therapy is favoured, particularly in patients with recent coronary events. Early aggressive treatment stabilises plaques and subsequently improves clinical outcomes. New drugs will be developed to specifically control Lp(a) or homocysteine, or to correct defects in CETP and LPL activity of lipoprotein metabolism.

Overall, research is identifying agents to correct the underlying disease and moreover, which patients will derive the most benefit from these new pharmacological interventions. In the future, the influence of genetic variation will determine a patients' response to specific drug therapy, thereby improving the recognition, treatment and prevention of CAD.

Acknowledgements

The authors wish to thank Bob Kezele, R.Ph. for his critical review in the preparation of this manuscript.

References

1. Kannel WB, Castelli WP, Gordon T. Cholesterol in the prediction of atherosclerotic disease: new perspective based on the Framingham Heart Study. Ann Intern Med 1979; 90: 85-91
2. National Heart, Lung and Blood Institute. Morbidity and mortality 1996 chartbook of cardiovascular, lung, and blood diseases. Washington (DC): US Department of Health and Human Services: Public Health Service: National Institute of Health, 1996
3. American Heart Association. 1998 heart and stroke statistical update. Dallas (TX): American Heart Association, 1997
4. Farmer JA, Gotto Jr AM. Dyslipidemia and other risk factors for coronary artery disease. In: Braunwald E, editor. Heart disease: a textbook of cardiovascular medicine. 4th ed. Philadelphia (PA): W.B. Saunders Co., 1992: 1126-60
5. Talbert RL. Hyperlipidemia. In: DiPiro JT, Talbert RL, Yee GC, et al, editors. Pharmacotherapy: a pathophysiologic approach. Norwalk (CT): Appleton & Lange, 1997: 459-66
6. Shepard J. Lipoprotein metabolism: an overview. Drugs 1994; 47 Suppl. 2: 1-10
7. Brown MS, Goldstein JL. A receptor-mediated pathway for cholesterol hemostasis. Science 1986; 232: 34-47
8. Lyons TJ. Glycation and oxidation: a role in the pathogenesis of atherosclerosis. Am J Cardiol 1993; 71: 26B-31
9. Ross R. Atherosclerosis: an inflammatory disease. N Engl J Med 1999; 340: 115-26
10. Esper RJ. The role of lipid-lowering therapy in multiple risk factor management. Drugs 1998; 56 Suppl. 1: 1-7
11. Kinlay S, Selwyn AP, Libby P, et al. Inflammation, the endothelium, and the acute coronary syndromes. J Cardiovasc Pharm 1998; 32 Suppl. 3: S62-6
12. Vogel RA, Corretti MC, Gellman J. Cholesterol, cholesterol lowering, and endothelial function. Prog Cardiovasc Dis 1998; 41: 117-36
13. Henry PD, Chen CH. Inflammatory mechanisms of atheroma formation: influence of fluid mechanics and lipid-derived inflammatory mediators. Am J Hypertens 1993; 6 (11 Pt 2): 328S-34
14. Mehta JL, Saldeen TGP, Rand K. Interactive role of infection, inflammation and traditional risk factors in atherosclerosis and coronary artery disease. J Am Coll Cardiol 1998; 31: 1217-25
15. Castelli WP. The new pathophysiology of coronary artery disease. Am J Cardiol 1998; 82: 60T-5
16. Muhlestein JB. Bacterial infections and atherosclerosis. J Investig Med 1998; 46: 396-402
17. Kwiterovich Jr PO. State-of-the-art update and review: clinical trials of lipid-lowering agents. Am J Cardiol 1998; 82 (12A): 3U-17; discussion 39U-41
18. Arroyo LH, Lee RT. The unstable atheromatous plaque. Can J Cardiol 1998; 14 Suppl. B: 11B-13
19. Davignon J. Prospects for drug therapy for hyperlipoproteinemia. Diabetes Metab 1995; 21: 139-46
20. National Cholesterol Education Program Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel II). Summary of the second report of the National Cholesterol Education Program (NCEP) expert panel on detection, evaluation, and treatment of high blood cholesterol in adults (adult treatment panel II). JAMA 1993; 269: 3015-23
21. West of Scotland Coronary Prevention Study Group. Influence of pravastatin and plasma lipids on clinical events in the West of Scotland Coronary Prevention Study (WOSCOPS). Circulation 1998; 97: 1440-5
22. The LIPID Study Group. Prevention of cardiovascular events and death with pravastatin in patients with coronary heart disease and a broad range of initial cholesterol levels. N Engl J Med 1998; 339: 1349-57
23. Down JR, Clearfield M, Weis S, et al. Primary prevention of acute coronary events with lovastatin in men and women with average cholesterol levels: results of AFCAPS/TexCAPS. JAMA 1998; 279: 1615-22
24. Sacks FM, Pfeffer MA, Maye LA, et al. The effect of pravastatin on coronary events after myocardial infarction in patients with average cholesterol. N Engl J Med 1996; 335: 1001-9
25. Scandinavian Simvastatin Survival Study Group. Baseline serum cholesterol and treatment effect in the Scandinavian Simvastatin Survival Study (4S). Lancet 1995; 345: 1274-5
26. Cullen P, Von Eckardstein A, Assman G. Diagnosis and management of new cardiovascular risk factors. Eur Heart J 1998; 19 Suppl. O: O13-9
27. Grundy S, Balady GJ, Criqui MH, et al. Primary prevention of coronary heart disease: guidance from Framingham: a statement for health care professionals from the AHA Task Force

on Risk Reduction. American Heart Association. Circulation 1998; 97: 1876-87
28. Marcovina SM, Koschinsky ML. Lipoprotein(a) as a risk factor for coronary artery disease. Am J Cardiol 1998; 82: 57U-66
29. Durrington PN. Triglycerides are more important in atherosclerosis than epidemiology has suggested. Atherosclerosis 1998; 141 Suppl. 1: S57-62
30. Sprecher DL. Triglycerides as a risk factor for coronary artery disease. Am J Cardiol 1998; 82: 49U-56
31. Assmann G, Schulte H, Funke H, et al. The emergence of triglycerides as a significant independent risk factor in coronary artery disease. Eur Heart J 1998; 19 Suppl. M: M8-14
32. Stein JH, Rosenson RS. Treatment of severe hypertriglyceridemia lowers plasma viscosity. Atherosclerosis 1998; 137: 401-5
33. Sirtori CR. New targets for lipid lowering and atherosclerosis prevention. Pharmacol Ther 1995; 67: 433-47
34. Badimon JJ, Fuster V, Badimon L. Role of high density lipoproteins in the regression of atherosclerosis. Circulation 1992; 86 (6 Suppl.): III-86-94
35. Davidson, MH. Implications for the present and direction for the future. Am J Cardiol 1993; 71: 32B-6
36. Welsh GM, Loscalzo J. Homocysteine and atherosclerosis. N Engl J Med 1998; 338: 1042-50
37. Hoogerbrugge N, Kerkhofs LG, Jansen H. Gemfibrozil decreases autoantibodies against oxidized low-density lipoprotein in men with combined hyperlipidemia. J Intern Med 1998; 243: 355-9
38. Broijersen A, Eriksson M, Angelin B, et al. Gemfibrozil enhances platelet activity in patients with combined hyperlipoproteinemia. Artheroscler Thromb 1995; 15: 121-7
39. Fujii S, Sobel BE. Direct effects of gemfibrozil on the fibrinolytic system: diminution of synthesis of plasminogen activator inhibitor type 1. Circulation 1992; 85: 1888-93
40. Keber I, Lavre J, Suc S, et al. The decrease of plasminogen activator inhibitor after normalization of triglycerides during treatment with fibrates. Fibrinolysis 1994; 8 Suppl. 2: 57-8
41. Ernest G, Resch KL. Fibrinogen as a cardiovascular risk factor: a meta-analysis and review of literature. Ann Intern Med 1993; 118: 956-63
42. Foger B, Drexel H, Hopferwieser T, et al. Fenofibrate improves postprandial chylomicron clearance in II B hyperlipoproteinemia. Clin Investig 1994; 72: 294-301
43. Bimmermann A, Boerschmann C, Schwartzkopff W, et al. Effective therapeutic measures for reducing lipoprotein (a) in patients with dyslipidemia: lipoprotein (a) reduction with sustained-release bezafibrate. Curr Ther Res 1991; 49: 635-43
44. Maggi FM, Biasi GM, Catapano AL. Reduction of Lp(a) plasma levels by bezafibrate. Atherosclerosis 1993; 100: 127-8
45. Perez-Jimenez F, Hilgado L, Zambrana JL, et al. Comparison of lovastatin and bezafibrate on lipoprotein(a) plasma levels in cardiac transplant recipients. Am J Cardiol 1995; 75: 648-50
46. Madej A, Okopien B, Kowalski J, et al. Effects of fenofibrate on plasma cytokine concentrations in patients with atherosclerosis and hyperlipoproteinemia IIb. Int J Clin Pharmacol Ther 1998; 36: 345-9
47. Steinmetz A, Schwartz T, Hehnke U, et al. Multicenter comparison of micronized fenofibrate and simvastatin in patients with primary type IIa or IIb hyperlipoproteinemia. J Cardiovasc Pharmacol 1996; 27: 563-8
48. Wierzbicki AS, Lumb PJ, Cheung J, et al. Fenofibrate plus simvastatin therapy versus simvastatin plus cholestyramine therapy for familial hypercholesterolaemia. QJM 1997; 90: 631-4
49. Ooi TC, Heinonen T, Alaupovic P, et al. Efficacy and safety of a new hydroxymethylglutaryl-coenzyme A reductase inhibitor, atorvastatin, in patients with combined hyperlipidemia: comparison with fenofibrate. Arterioscler Thromb Vasc Biol 1997; 17: 1793-9
50. Gholami K, Tavakoli N, Maleki M, et al. Comparison of the efficacy and safety of fenofibrate and lovastatin in patients with primary type IIa or IIb hyperlipidemia. J Clin Pharm Ther 1998; 23: 213-21
51. Avogaro L, Beltramello P, Marin R, et al. Insulin action and glucose metabolism are improved by gemfibrozil treatment in hypertriglyceridemic patients. Atherosclerosis 1995; 113: 117-24
52. Frock MH, Leo O, Hap K, et al. Helsinki Heart Study: primary-prevention trial with gemfibrozil in middle-aged men with dyslipidemia. Safety of treatment, changes in risk factors and incidence of coronary heart disease. N Engl J Med 1987; 317: 1237-45
53. Harman HW, Bunt T, Ah HN, et al. Progression and regression of minor arterial narrowing by quantitative angiography after fenofibrate therapy. Am J Cardiol 1991; 67: 957-61
54. Bunte T, Hahmann HW, Hellwig N, et al. Effects of fenofibrate on angiographically examined coronary atherosclerosis and left ventricular function in hypercholesterolemic patients. Atherosclerosis 1993; 98: 127-38
55. Ericsson C-G, Hamsten A, Nilsson J, et al. Angiographic assessment of effects of bezafibrate on progression of coronary artery disease in young male postinfarction patients. Lancet 1996; 347: 849-53
56. Frick MH, Syvanne M, Nieminen MS, et al. Prevention of the angiographic progression of coronary and vein-graft atherosclerosis by gemfibrozil after coronary bypass surgery in men with low levels of HDL cholesterol. Circulation 1997; 96: 2137-43
57. Goldbourt U, Behar S, Reicher-Reiss H, et al. Rationale and design of a secondary prevention trial of increasing serum high-density lipoprotein cholesterol and reducing triglycerides in patients with clinically manifest atherosclerotic heart disease (the bezafibrate infarction prevention trial). Am J Cardiol 1993; 71: 909-15
58. Rubins HB, Robins SJ, Collins D, et al. Gemfibrozil for the secondary prevention of heart disease in men with low levels of high-density lipoprotein cholesterol: Veterans Affairs High-Density Lipoprotein Cholesterol Intervention Trial Study Group. N Engl J Med 1999; 341: 410-8
59. Steiner G. The diabetes atherosclerosis intervention study (DAIS): a study conducted in cooperation with the World Health Organization. Diabetologia 1996; 39: 1655-61
60. Fruchart JC, Brewer HB, Leitersdorf E. Consensus for the use of fibrates in the treatment of dyslipoproteinemia and coronary heart disease. Am J Cardiol 1998; 81: 912-7
61. Adkins JC, Faulds D. Micronised fenofibrate: a review of its pharmacodynamic properties and clinical efficacy in the management of dyslipidemia. Drugs 1997; 54 (4): 615-33
62. Committee of Principal Investigators. A co-operative trial in the primary prevention of ischemic heart disease using clofibrate: report from the Committee of Principal Investigators. Br Heart J 1978; 40: 1069-118
63. Committee of Principal Investigators. WHO cooperative trial on primary prevention of ischemic heart disease with clofibrate to lower serum cholesterol: mortality follow-up. Lancet 1980; II: 379-85
64. Kashyap ML. Mechanistic studies of high-density lipoproteins [discussion 85U-6]. Am J Cardiol 1998; 82: 42U-8

65. Philipp CS, Cisar LA, Saidi P, et al. Effect of niacin supplementation on fibrinogen levels in patients with peripheral vascular disease. Am J Cardiol 1998; 82: 697-9
66. McKenney JM, Proctor JD, Harris S, et al. A comparison of the efficacy and toxic effects of sustained vs. immediate-release niacin in hypercholesterolemic patients. JAMA 1994; 271: 672-7
67. Capuzzi DM, Guyton JR, Morgan JM, et al. Efficacy and safety of an extended-release niacin (niaspan): a long-term study. Am J Cardiol 1998; 82: 74U-81
68. Goldberg AC. Clinical trial experience with extended-release niacin (niaspan): dose-escalation study. Am J Cardiol 1998; 82: 35U-8
69. Morgan JM, Capuzzi DM, Guyton JR. A new extended-release niacin (niaspan): efficacy, tolerability, and safety in hypercholesterolemic patients. Am J Cardiol 1998; 82: 29U-34
70. Knopp RH, Alagona P, Davidson M, et al. Equivalent efficacy of a time-release form of niacin (niaspan) given once-a-night versus plain niacin in the management of hyperlipidemia. Metabolism 1998; 47: 1097-104
71. Gardner SF, Marx MA, White LM, et al. Combination of low-dose niacin and pravastatin improves the lipid profile in diabetic patients without compromising glycemic control. Ann Pharmacother 1997; 31: 677-82
72. Britton ML, Bradberry JC, Letassy NA, et al. ASHP therapeutic position statement on the safe use of niacin in the management of dyslipidemias. Am J Health Syst Pharm 1997; 54: 2815-9
73. Guyton JR. Effect of niacin on atherosclerotic cardiovascular disease. Am J Cardiol 1998; 82: 18U-23
74. The Coronary Drug Project Research Group. Clofibrate and niacin in coronary heart disease. JAMA 1975; 231: 360-81
75. Carlson LA, Rosenhamer G. Reduction of mortality in the Stockholm Ischaemic Heart Disease Secondary Prevention Study by combined treatment with clofibrate and nicotinic acid. Acta Med Scand 1988; 223: 405-18
76. Blankenhorn DH, Nessim SA, Johnson RL, et al. Beneficial effects of combined colestipol-niacin therapy on coronary atherosclerosis and coronary venous bypass grafts. JAMA 1987; 257: 3233-40
77. Cashin-Hemphill L, Mack WJ, Pogoda JM, et al. Beneficial effects of colestipol-niacin on coronary atherosclerosis: a 4-year follow-up. JAMA 1990; 264: 3013-7
78. Brown G, Albers JJ, Fisher LD, et al. Regression of coronary artery disease as a result of intensive lipid-lowering therapy in men with high levels of apolipoprotein B. N Engl J Med 1990; 323: 1289-98
79. Kane JP, Malloy MJ, Ports TA, et al. Regression of coronary atherosclerosis during treatment of familial hypercholesterolemia with combined drug regimens. JAMA 1990; 264: 3007-12
80. Sacks FM, Pasternak RC, Gibson CM, et al and the HARP Group. Effect on coronary atherosclerosis of decrease in plasma cholesterol concentrations in normocholesterolemic patients. Lancet 1994; 344: 1182-6
81. Baker SG, Jaffe BI, Mendelsohn D, et al. Treatment of homozygous familial hypercholesterolemia with probucol. S Afr Med J 1982; 62: 7-11
82. Yamamoto A, Matsuzawa Y, Yokoyama S, et al. Effects of probucol on xanthomata regression in familial hypercholesterolemia. Am J Cardiol 1986; 57: 29H-5
83. Walldius G, Erikson U, Olsson AG, et al. The effects of probucol on femoral atherosclerosis: the Probucol Quantitative Regression Swedish Trial (PQRST). Am J Cardiol 1994; 74: 875-83
84. Carew TE, Schwenke DC, Steinberg D. Antiatherogenic effect of probucol unrelated to its hypocholesterolemic effect: evidence that antioxidants in vivo can selectively inhibit low density lipoprotein degradation in macrophage-rich fatty streaks and slow the progression of atherosclerosis in the Watanabe heritable hyperlipidemic rabbit. Proc Natl Acad Sci U S A 1987; 84: 7725-9
85. Setsuda M, Inden M, Hiraoka N, et al. Probucol therapy in the prevention of restenosis after successful percutaneous transluminal coronary angioplasty. Clin Ther 1993; 15: 374-82
86. Reinoehl J, Frankovich D, Machado C, et al. Probucol-associated tachyarrythmic events and QT prolongation: importance of gender. Am Heart J 1996; 131: 1184-91
87. Maron DJ, Fazio S, Linton MF, et al. Current perspectives on statins. Circulation 2000; 101 (2): 207-13
88. Jones P, Kofonek S, Laurora I, et al. for the CURVES Investigators. Comparative dose efficacy study of atorvastatin versus simvastatin, pravastatin, lovastatin, and fluvastatin in patients with hypercholesterolemia (the CURVES study). Am J Cardiol 1998; 81: 582-7
89. Brown AS, Bakker-Arkema RG, Yellen L, et al. Treating patients with documented atherosclerosis of National Cholesterol Education Program recommended low-density-lipoprotein cholesterol goals with atorvastatin, fluvastatin, lovastatin, and simvastatin. J Am Coll Cardiol 1998; 32: 665-72
90. The Post Coronary Artery Bypass Graft trial Investigators. The effect of aggressive lowering of low-density lipoprotein cholesterol levels and low-dose anticoagulation on obstructive changes in saphenous-vein coronary-artery bypass graft. N Engl J Med 1997; 336: 153-62
91. Pitt B, Waters D, Brown WV, et al. Results of the atorvastatin versus revascularization treatment (AVERT) study: 18 month study of aggressive lipid-lowering in patients with coronary artery disease indicated for catherization based revascularization (CR) [abstract no. 3342]. 71st Scientific Sessions of the American Heart Association; 1998 Nov 8-11; Dallas (TX)
92. Grundy SM. Statin trials and goals of cholesterol-lowering therapy. Circulation 1998; 97: 1436-9
93. Data on file, Parke-Davis, 1999
94. Egashira K, Hirooka Y, Kai H, et al. Reduction in serum cholesterol with pravastatin improves endothelium-dependent coronary vasomotion in patients with hypercholesterolemia. Circulation 1994; 89: 2519-24
95. Treasure CB, Klein JL, Weintraub WS, et al. Beneficial effects of cholesterol-lowering therapy on the coronary endothelium in patients with coronary artery disease. N Engl J Med 1995; 332: 481-7
96. Eichstadt HW, Eskotter H, Hoffmann I, et al. Improvement of myocardial perfusion by short-term fluvastatin therapy in coronary artery disease. Am J Cardiol 1995; 76 Suppl.: 122A-5
97. Soma MR, Donetti E, Parolini C, et al. HMG-CoA reductase inhibitors: in vivo effects on carotid intimal thickening in normocholesterolemic rabbits. Arterioscler Thromb 1993; 13: 571-8
98. Corsini A, Raiteri M, Soma MR, et al. Pathogenesis of atherosclerosis and the role of 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitors. Am J Cardiol 1995; 76 Suppl.: 21A-8A
99. Hidaka Y, Eda T, Yonemoto M, et al. Inhibition of cultured vascular smooth muscle cells migration by simvastatin (MK-733). Atherosclerosis 1992; 95: 87-94
100. Rosenson RS, Tangney CC. Antithrombotic properties of statins: implications for cardiovascular event reduction. JAMA 1998; 279: 1643-50

101. Bustos C, Hernandez-Presa A, Ortego M, et al. HMG-CoA reductase inhibition by atorvastatin reduces neointimal inflammation in a rabbit model of atherosclerosis. J Am Coll Cardiol 1998; 2057-64
102. Jukema JW, Bruschke AVG, van Boven AJ, et al. Effects of lipid lowering by pravastatin on progression and regression of coronary artery disease in symptomatic men with normal to moderately elevated serum cholesterol levels: the Regression Growth evaluation Statin Study (REGRESS). Circulation 1995; 91: 2528-40
103. Pitt B, Mancini GBJ, Ellis SG, et al. Pravastatin Limitation of Atherosclerosis in the Coronary Arteries (PLAC-I): reduction in atherosclerosis progression and clinical events. J Am Coll Cardiol 1995: 26: 1133-9
104. MAAS Investigators. Effects of simvastatin on coronary atheroma: the Multicentre Anti-Atheroma Study (MAAS). Lancet 1994; 344: 633-8
105. Waters D, Higginson L, Gladstone P, et al. Effects of monotherapy with an HMG-CoA reductase inhibition on the progression of coronary atherosclerosis as assessed by serial quantitative atherography: the Canadian Coronary Atherosclerosis Intervention trial. Circulation 1994; 89: 959-68
106. Lipid Research Clinics Program. The Lipid Research Clinics Coronary Primary Prevention Trial Results: I. Reduction in the incidence of coronary heart disease. JAMA 1984; 251: 351-4
107. Sirtori CR, Franceschini G. Hypoalphalipoproteinemia: from a mutation to drug development. In: Woodford FP, Davignon J, Sniderman A, editors. Proceedings Xth International Congress of Atherosclerosis; 1994 Oct 9-14: Montreal. New York (NY): Elsevier Science Publisher BV, 1995: 1083
108. Sperber AD, Kenkin Y, Zuil I, et al. The hypocholesterolemic effect of an antacid containing aluminum hydroxide. Am J Med 1991: 91: 597-604
109. The Writing Group for the PEPI Trial. Effects of estrogen or estrogen/progestin regimens on heart disease risk factors in postmenopausal women: the Postmenopausal Estrogen/Progestin Interventions (PEPI) trial. JAMA 1995; 273: 199-208
110. Brown L, Rosner B, Willett WW, et al. Cholesterol-lowering effects of dietary fiber: a meta-analysis. Am J Clin Nutr 1999; 69: 30-42
111. Ma P, Yamamoto T, Goldstein J, et al. Increased mRNA for low density lipoprotein receptor in livers of rabbits treated with 17 alpha-ethinyl estradiol. Proc Natl Acad Sci U S A 1986; 83: 792-6
112. Darling GM, Johns JA, McCLoud PI, et al. Estrogen and progestin compared with simvastatin for hypercholesterolemia in postmenopausal women. N Engl J Med 1997; 337: 595-601
113. Cauley JA, LaPorte RE, Kuller LH, et al. Menopausal estrogen use, high density lipoprotein cholesterol subfraction and liver function. Atherosclerosis 1983; 49: 31-9
114. Walsh BW, Schiff I, Rosner B, et al. Effects of postmenopausal estrogen replacement on the concentrations and metabolism of plasma lipoproteins. N Engl J Med 1991; 325: 1196-204
115. Tuck CH, Holleran S, Berglund L. Hormonal regulation of lipoprotein(a) levels: effects of estrogen replacement therapy on lipoprotein(a) and acute phase reactants in postmenopausal women. Arterioscler Thromb Vasc Biol 1997; 17: 1822-9
116. Barrett-Connor E, Bush TL. Estrogen and coronary artery disease. JAMA 1992; 265: 1861-7
117. Kon K, Cardillo C, Bue MN, et al. Vascular effects of estrogen and cholesterol-lowering therapies in hypercholesterolemic postmenopausal women. Circulation 1999; 99: 354-60
118. Hulley S, Grady D, Bush T, et al. Randomized trial of estrogen plus progestin for secondary prevention of coronary heart disease in postmenopausal women: Heart and Estrogen/Progestin Replacement Study (HERS) research group. JAMA 1998; 280: 605-13
119. Josefson D. Women with heart disease cautioned about HRT. BMJ 1999; 318 (7186): 753
120. Anonymous. Design of the Women's health initiative clinical trial and observational study: The Women's Health Initiative Study Group. Control Clin Trials 1998; 19: 61-109
121. McDonald CC, Alexander FE, Whyte BW, et al. Cardiac and vascular morbidity in women receiving adjuvant tamoxifen for breast cancer in a randomized trial: the Scottish Cancer Trials Breast Group. BMJ 1995; 311: 977-80
122. Walsh BW, Kuller LH, Wild RA, et al. Effects of raloifene on serum lipids and coagulation factors in healthy postmenopausal women. JAMA 1998; 279: 1445-51
123. Hasibeder H, Staab HJ, Seibel K, et al. Clinical pharmacology of the hypocholesterolemic agent K 12.148 (lifibrol) in healthy volunteers. Eur J Clin Pharmacol 1991: 40 Suppl. 1: S91-4
124. Schwandt P, Elsasser R, Schmidt C, et al. Safety and efficacy of lifibrol upon four-week administration to patients with primary hypercholesterolemia. Eur J Clin Pharmacol 1994; 47: 133-8
125. Locker PK, Jungbluth GL, Francom SF, et al. Lifibrol: a novel lipid-lowering drug for therapy of hypercholesterolemia. Clin Pharmacol Ther 1995; 57: 73-88
126. Stedronsky ER. Interaction of bile acids and cholesterol with nonsystemic agents having hypocholesterolemic properties. Biochim Biophys Acta 1994; 1210: 255-87
127. Davidson MH, Dillon MA, Gordon B, et al. Colesevelam hydrochloride (cholestagel): a new, potent bile acid sequestrant associated with a low incidence of gastrointestinal side effects. Arch Intern Med 1999; 159 (16): 1893-900
128. Ormrod DJ, Holmes CC, Miller TE. Dietary chitosan inhibits hypercholesterolaemia and atherogenesis in the apolipoprotein E-deficient mouse model of atherosclerosis. Atherosclerosis 1998; 138: 329-34
129. Morehouse LA, Bangerter FW, DeNinno MP, et al. Comparison of synthetic saponin cholesterol absorption inhibitors in rabbits: evidence for a non-stoichiometric, intestinal mechanism of action. J Lipid Res 1999; 40: 464-74
130. Harris WS, Dujovne CA, Windsor SL, et al. Inhibiting cholesterol absorption with CP-88,818 (beta-tigogenin cellobioside; tiqueside): studies in normal and hyperlipidemic subjects. J Cardiovasc Pharmacol 1997; 30: 55-60
131. Coats AJ. The potential role of soluble fiber in the treatment of hypercholesterolemia. Postgrad Med J 1998; 74: 391-4
132. Higaki I, Hara S, Takasu N, et al. Inhibition of ilieal Na+/bile acid cotransporter by S-8921 reduces serum cholesterol and prevents atherosclerosis in rabbits. Arterioscler Thromb Vasc Biol 1998: 18: 1304-11
133. Huettinger M, Hermann M, Goldenberg H, et al. Hypolipidemic activity of HOE-402 is mediated by stimulation of the LDL receptor pathway. Arterioscler Thromb 1993; 13: 1005-12
134. Hoffmann A, Schmalz M, Leineweber M. Cholesterol lowering action of HOE 402 in the normolipidemic and hypercholesterolemic Golden Syrian hamster. Biochim Biophys Acta 1996; 1299: 95-102
135. Krause BR, Pape ME, Kieft K, et al. ACAT inhibition decreases LDL cholesterol in rabbits fed a cholesterol-free diet: marked changes in LDL cholesterol without changes in LDL receptor mRNA abundance. Arterioscler Thromb 1994; 14: 598-604

136. Musanti R, Giorgini L, Lovisolo PP, et al. Inhibition of acyl-CoA: cholesterol acyltransferase decrease apolipoprotein B-100-containing lipoprotein secretion from HepG2 cells. J Lipid Res 1996; 37: 1-14
137. Chiari A, Lovisolo P, Fogliatto G, et al. Correction of dyslipoproteinemia of casein-fed rabbit by FCE 27677, a potent novel ACAT inhibitor. Pharmacol Res 1996; 33: 181-9
138. Wilcox LJ, Barrett PH, Newton RS, et al. The ACAT inhibitor CI-1011 decreases ApoB100 secretion from hepG2 cells: an effect associated with enhanced intracellular degradation of ApoB. Arterioscler Thromb Vasc Biol 1999; 19: 939-49
139. Murakami S, Yamagishi I, Asami Y, et al. Effect of the ACAT inhibition, HL-004, on cholesterol metabolism in macrophage. Cell Mol Biol 1996; 42: 865-72
140. Asami Y, Kondo Y, Murakami S, et al. The ACAT inhibitor HL-004 inhibits cholesterol absorption and lowers serum cholesterol in rats. Gen Pharmacol 1998; 31: 593-6
141. Hainer JW, Terry JG, Connell KM, et al. Effects of the acyl-CoA: cholesterol acyltransferase inhibitor DUP128 on cholesterol absorption-and serum cholesterol in humans. Clin Pharmacol Ther 1994; 56 (1): 65-74
142. Harris NS, Dujovne C, von Bergmamn K, et al. Effects of the ACAT inhibitor CL277,082 on cholesterol metabolism in humans. Clin Pharmacol 1990; 48: 189-94
143. Bocan YMA, Bak Mueller S, Uhlendorf PD, et al. Comparison of CI-976, and ACAT inhibitor, and selected lipid-lowering agents for antiatherosclerotic activity in iliac-femoral and thoracic aortic lesions. Arterioscler Thromb 1991; 11: 1830-43
144. Tanaka H, Ohtsuka I, Kogushi M, et al. Effect of the acyl-CoA: cholesterol acyltransferase inhibitor E5324, on experimental atherosclerosis in rabbits. Atherosclerosis 1994; 107: 187-201
145. Nakaya N, Nakamichi N, Sekino H, et al. Effect of a novel ACAT inhibitor, E5324, on serum lipids and lipoproteins in healthy volunteers [abstract]. Atherosclerosis 1994; 109: 253
146. Tanaka A, Terasawa T, Hagihara H, et al. Inhibitors of acyl-CoA: cholesterol O-acyltransferase: 2. Identification and structure activity relationships of a novel series of N-alkyl-N-(heteroalkyl-substituted benzyl)-N-arylureas. J Med Chem 1998; 41: 2390-410
147. Nicolosi RJ, Wilson TA, Krause BR. The ACAT inhibitor, CI-1011 is effective in the prevention and regression of aortic fatty streak area in hamsters. Atherosclerosis 1998; 137: 77-85
148. Uchida T, Aoyama K, Watanabe T, et al. Relationship between bioavailability and hypercholesterolemic activity of YM17E, an inhibitor of ACAT, in cholesterol-fed rabbits. Atherosclerosis 1998; 137: 97-106
149. Asami Y, Yamagishi I, Murakami S, et al. HL-004, the ACAT inhibitor, prevents the progression of atherosclerosis in cholesterol-fed rabbits. Life Sci 1998; 62: 1055-63
150. Davis HR, Watkins RW, Salisbury BG, et al. Effects of the cholesterol absorption inhibitor SCH 48461 in combination with the HMG-CoA reductase inhibitor lovastatin in rabbits, dogs, and rhesus monkeys [abstract]. Atherosclerosis 1994; 109: 162
151. Adelman SJ, McKean M, Prozialeck DH, et al. ACA-147: an inhibitor of ACAT and of the oxidative modification of LDL. In: Woodford FP, Davignon J, Sniderman A, editors. Proceedings Xth International Congress of Atherosclerosis; 1994 Oct 9-14: Montreal (PQ). New York (NY): Elsevier Science Publisher BV, 1995: 316-20
152. Yasuhara M, Saito K, Kubota H, et al. Inhibitory effect of a new ureidophenol derivative T-2591 on LDL oxidation and ACAT activity. Biol Pharm Bull 1997; 20: 1056-60
153. Bocan TM, Mueller SB, Brown EQ, et al. HMG-CoA reductase and ACAT inhibitors act synergistically to lower plasma cholesterol and limit atherosclerotic lesion development in the cholesterol-fed rabbits. Atherosclerosis 1998; 139: 231-30
154. Picard IA, Sliskovic DR, Anderson MK, et al. Structure-activity relationships of a series of oxysulfonyl carbamate inhibitors of acylCoA: cholesterol O-acyltransferase (ACAT): identification of PD 138142-15, a water soluble ACAT inhibitor displays excellent lipid regulation in rats [abstract]. Atherosclerosis 1994; 109: 162
155. Franceschini G, Maderna P, Sirtori CR. Reverse cholesterol transport: physiology and pharmacology. Atherosclerosis 1991; 88: 99-107
156. Inazu A, Brown ML, Hesler CB, et al. Increased high-density lipoprotein levels caused by a common cholesteryl-ester transfer protein gene mutation. N Engl J Med 1990; 323: 1234-8
157. Abbey M, Calbert GD. Effects of blocking plasma lipid transfer protein activity in the rabbit. Biochim Biophys Acta 1989; 1003: 29-30
158. Ishigami M, Yamashita S, Sakai N, et al. Large and cholesteryl ester-rich high-density lipoprotein in cholesteryl ester transfer protein (CETP) deficiency can not protect macrophages from cholesterol accumulation induced by acetylated low-density lipoproteins. J Biochem 1994; 116: 257-62
159. Sakai N, Yamashita S, Hirano K-I, et al. Decreased affinity of low density lipoprotein (LDL) particles for LDL receptors in patents with cholesteryl ester transfer protein deficiency. Eur J Clin Invest 1995; 25: 332-9
160. Chiesa G, Michelagnoli S, Cassinotti M, et al. Mechanisms of high-density lipoprotein reduction after probucol treatment: changes in plasma cholesterol esterification/transfer and lipase activities. Metabolism 1993; 42: 229-35
161. Kothari HV, Poirier KJ, Lee WH, et al. Inhibition of cholesterol ester transfer protein CGS 25159 and changes in lipoproteins in hamsters. Atherosclerosis 1997; 128: 59-66
162. Morton RE, Greene DJ. Suppression of lipid transfer inhibition protein activity by oleate: a novel mechanism of cholesterol ester transfer protein regulation by plasma free fatty acids. Arterioscler Thromb Vasc Biol 1997; 17: 3041-8
163. Guerin M, Dolphin PJ, Talussot C, et al. Pravastatin modulates cholesteryl ester transfer from HDL to apoB-containing lipoproteins and lipoprotein subspecies profile in familial hypercholesterolemia. Arterioscler Thromb Vasc Biol 1995; 15: 1359-68
164. Cho KH, Lee JY, Choi MS, et al. A peptide from hog plasma that inhibits human cholesteryl ester transfer protein. Biochim Biophys Acta 1998; 1391: 133-44
165. Krause BR, Sliskovic QR, Anderson M, et al. Lipid-lowering effects of WAY-121,898, an inhibitor of pancreatic cholesteryl ester hydrolase. Lipids 1998; 33: 489-98
166. Saxena U, Klein MG, Vanni TM, et al. Lipoprotein lipase increases low density lipoprotein retention by subendothelial cell matrix. J Clin Invest 1992; 89: 373-80
167. Sirtori CR, Colli S. Influences of lipid-modifying agents on hemostasis. Cardiovasc Drugs Ther 1993; 7: 817-23
168. Tsutsumi K, Inone Y, Shjima A, et al. The novel compound NO-1886 increases lipoprotein lipase activity with resulting elevation of high-density lipoprotein cholesterol, and long-term administration inhibits atherogenesis in the coronary arteries of rats with experimental atherosclerosis. J Clin Invest 1993; 92: 411-7
169. Chiba T, Miura S, Sawamura F, et al. Antiatherogenic effects of a novel lipoprotein lipase-enhancing agent in cholesterol-fed

New Zealand white rabbits. Arterioscler Thromb Vasc Biol 1997; 17: 2601-8
170. Gotto AM. Overview of current issues in management of dyslipidemia. Am J Cardiol 1993; 71: 3B-8
171. Bergstrom JD, Kurtz MM, Rew DJ, et al. Zaragozic acids: a family of fungal metabolites that are picomolar competitive inhibitors of squalene synthetase. Proc Natl Acad Sci U S A 1993; 90: 80-4
172. Vaidya S, Bostedor R, Kurtz MM, et al. Massive production of farnesol-derived dicarboxylic acids in mice treated with the squalene synthetase inhibitor zaragozic acid A. Arch Biochem Biophys 1998; 355: 84-92
173. Baxter A, Fitzgerald BJ, Hutson JL, et al. Squalenestatin 1, a potent inhibitor of squalene synthetase, which lowers serum cholesterol in vivo. J Biol Chem 1992; 267: 11705-8
174. Amin D, Rutledge RZ, Needle SN, et al. RPR 107393, a potent squalene synthetase inhibitor and orally effective cholesterol-lowering agent: comparison with inhibitors of HMG-CoA reductase. J Pharmacol Exp Ther 1997; 281: 746-52
175. Hidaka Y, Hotta H, Nogata Y, et al. Effects of a novel squalene epoxidase inhibitor, NB-598, on the regulation of cholesterol metabolism in Hep G2 cells. J Biol Chem 1991; 266: 13171-7
176. Matzno S, Yamauchi T, Gohda M, et al. Inhibition of cholesterol biosynthesis by squalene epoxidase inhibitor avoids apoptotic cell death in L6 myoblasts. J Lipid Res 1997; 38: 1639-48
177. Gotto AM. Lipid-regulating and antiatherosclerotic therapy: current options and future approaches. Cleve Clin J Med 1996; 63: 31-41
178. Gotto Jr AM. Dyslipidemia and atherosclerosis: a forecast of pharmaceutical approaches. Circulation 1993; 87 Suppl. III: III-54-9
179. Glomset J, Gelb M, Farnsworth C. The prenylation of proteins. Curr Opin Lipidol 1991; 2: 118-24
180. Lopez D, Chambers CM, Keller RK, et al. Compensatory responses to inhibition of hepatic squalene synthetase. Arch Biochem Biophys 1998; 351: 159-66
181. Zhu B, Parmely WW. Modification of experimental and clinical atherosclerosis by dietary fish oil. Am Heart J 1990; 119: 168-70
182. Bocan TM, Rosebury WS, Mueller SB, et al. A specific 15-lipoxygenase inhibitor limits the progression and monocyte-macrophage enrichment of hypercholesterolemia-induced atherosclerosis in the rabbit. Atherosclerosis 1998; 13: 203-16
183. Maziere C, Barbu V, Anclair M, et al. Interleukin 1 stimulates cholesterol esterification and cholesterol deposition in J774 monocytes-macrophages. Biochim Biophys Acta 1996; 1300: 30-4
184. The Coronary Drug Project Research Group. The coronary drug project: findings leading to future modifications of its protocol with respect to dextrothyroxine. JAMA 1972; 220: 996-1008
185. Stephan ZF, Yurachek EC, Sharif R, et al. Demonstration of potent lipid-lowering activity by a thyromimetic agent devoid of cardiovascular and thermogenic effects. Atherosclerosis 1996; 126: 53-63
186. Steele RE, Wasvary JM, Dardik BN, et al. CGS 26214, the thyroxine connection revisited. In: Woodford FP, Davignon J, Sniderman A, editors. Proceedings Xth International Congress of Atherosclerosis; 1994 Oct 9-14: Montreal (PQ). New York (NY): Elsevier Science Publisher BV, 1995: 321-4
187. DeBruin TWA, van Barlingen H, van Linde-Sibenius, et al. Lipoprotein(a) and apolipoprotein B plasma concentrations in hypothyroid, euthyroid, and hyperthyroid subjects. J Clin Endocrinol Metab 1993; 76: 121-6
188. Engler H, Riesen WF. Effect of thyroid function on concentrations of lipoprotein(a). Clin Chem 1993; 39: 2466-9
189. Taylor AH, Stephan ZF, Steele RE, et al. Beneficial effects of a novel thyromimetic on lipoprotein metabolism. Mol Pharmacol 1997; 52: 542-7
190. Bailey JM, Bright R, Tomar R, et al. Anti-atherogenic effects of cholesterol vaccination. Biochem Soc Trans 1994; 22: 433S
191. Alving CR, Swartz Jr GM, Wassef NM, et al. Vaccination against cholesterol immunologic modulation of diet-induced hypercholesterolemia and atherosclerosis. In: Woodford FP, Davignon J, Sniderman A, editors. Proceedings Xth International Congress of Atherosclerosis; 1994 Oct 9-14: Montreal (PQ). New York (NY): Elsevier Science Publisher BV, 1995: 944-52
192. Travis J. Army targets a potential vaccine against cholesterol. Science 1993; 262: 1974-5
193. Nilsson A, Calara F, Regnstrom J, et al. Immunization with homologous oxidized low density lipoprotein reduces neointimal formation after balloon injury in hypercholesterolemia rabbits. J Am Coll Cardiol 1997; 30: 1886-91
194. Rader DJ. Gene therapy for atherosclerosis. Int J Clin Lab Res 1997; 27: 35-43
195. Grossman M, Raper SE, Kozarsky K, et al. Successful ex vivo gene therapy directed to liver in a patient with familial hypercholesterolemia. Nat Genet 1994; 6: 335-41
196. Kozarsky KF, McKinley DR, Austin LL, et al. In vivo correction of low density lipoprotein receptor deficiency in the Watanabe heritable hyperlipidemic rabbits with recombinant adenoviruses. J Biol Chem 1994; 269: 13695-702
197. Leclerc G, Gal D, Takeshita S, et al. Percutaneous arterial gene transfer in a rabbit model. J Clin Invest 1992; 90: 936-44
198. Steg PG, Feldman LJ, Scoazec JY, et al. Arterial gene transfer to rabbit endothelial and smooth muscle cells using percutaneous delivery of an adenoviral vector. Circulation 1994; 90: 1648-56
199. Laitinen M, Yla-Herttuala S. Vascular gene transfer for the treatment of restenosis and atherosclerosis. Curr Opin Lipidiol 1998; 9: 465-9
200. French BA. Gene therapy and cardiovascular disease. Curr Opin Cardiol 1998; 13: 205-13
201. Ueno H, Li JJ, Masuda S, et al. Adenovirus-mediated expression of the secreted form of basic fibroblast growth factor (FGF-2) induces cellular proliferation and angiogenesis in vivo. Arterioscler Thromb Vasc Biol 1997; 17: 2453-60
202. Rutherford C, Martin W, Salame M, et al. Substantial inhibition of neo-intimal response to balloon injury in the rat carotid artery using a combination of antibodies to platelet-derived growth factor-BB and basic fibroblast growth factor. Atherosclerosis 1997; 130: 45-51
203. Zsigmond E, Kobayashi K, Tzung KW, et al. Adenovirus-mediated gene transfer of human lipoprotein lipase ameliorates the hyperlipidemia associated with apolipoprotein E and LDL receptor deficiencies in mice. Hum Gene Ther 1997; 8: 1921-33
204. Shimano H, Yamada N, Katsuki M, et al. Overexpression of apolipoprotein E in transgenic mice: marked reduction in plasma lipoproteins except high density lipoprotein and resistance against diet-induced hypercholesterolemia. Proc Natl Acad Sci U S A 1992; 89: 1750-4
205. Excoffon KJ, Liu G, Miao L, et al. Correction of hypertriglyceridemia and impaired fat tolerance in lipoprotein lipase-deficient mice by adenovirus-mediated expression of human lipoprotein lipase. Arterioscler Thromb Vasc Biol 1997; 17: 2532-9

206. Simons M, Edelman ER, Dekeyser J-L, et al. Antisene c-myb oligonucleotide inhibit intimal arterial smooth muscle cell accumulation in vivo. Nature 1992; 359: 67-70
207. Abe J-I, Zhou W, Taguchi J-I, et al. Suppression of neointimal smooth muscle cell accumulation in vivo by antisene CDC2 and CDK2 oligonucleotides in rats carotid artery. Biochem Biophys Res Commun 1994; 198: 16-24
208. Rosengart TK, Patel SR, Crystal RG. Therapeutic angiogenesis: protein and gene therapeutic delivery strategies. J Cardiovasc Risk 1999; 6 (1): 29-40
209. Qian H, Neplioueva V, Shetty GA, et al. Nitric oxide synthase gene therapy rapidly reduces adhesion molecule expression and inflammatory cell infiltration in carotid arteries of cholesterol-fed rabbit. Circulation 1999; 99 (23): 2979-82
210. Hussain MM, Mahley RW, Boyles JK, et al. Chylomicron-chylomicron remnant clearance by liver and bone marrow in rabbits: factors that modify tissue-specific uptake. J Biol Chem 1989; 264: 9571-82
211. Yamada N, Shimano H, Mokuno H, et al. Increased clearance of plasma cholesterol after injection of apolipoprotein E into Watanabe heritable hypercholesterolemic rabbits. Proc Natl Acad Sci U S A 1989; 86: 665-9
212. Mahley RW, Weisgraber KH, Hussain MM, et al. Intravenous infusion of apolipoprotein E accelerates clearance of plasma lipoproteins in rabbits. J Clin Invest 1989; 83: 2125-30
213. Yamada N, Inoue E, Kawamura M, et al. Apolipoproteins E prevents the progression of atherosclerosis in Watanabe heritable hyperlipidemia rabbits. J Clin Invest 1992; 89: 706-11
214. Badimon JJ, Budimon L, Galvez A, et al. High density lipoprotein plasma fractions inhibit aortic streaks in cholesterol-fed rabbits. Lab Invest 1989; 89: 455-61
215. Aoyama T, Yui Y, Morishita H, et al. Prostaglandin I_2 half-life regulates by high density lipoprotein is decreased in acute myocardial infarction and unstable angina pectoris. Circulation 1990; 81: 1784-91
216. Saku K, Ahmada M, Glas-Greenwalt P, et al. Activation of fibrinogen by apolipoproteins of high density lipoproteins in men. Thromb Res 1985; 39: 1-8
217. Beitz I, Beitz A, Antonov IV, et al. Does a HDL injection reduce the development of serum hyperlipidemia and progression of fatty streaks in cholesterol fed rabbits. Prostaglandins Leukot Essent Fatty Acids 1992; 47: 145-52
218. Trachtenberg JD, Cochrane H, Sun S, et al. Apolipoprotein A-I inhibits atherosclerotic lesions progression [abstract]. Circulation 1993; 88: I-522
219. Franceschini G, Vecchio G, Gianfranceschi G, et al. Apolipoprotein A-IMilano: accelerated binding and dissociation from lipids from a human apolipoprotein variant. J Biol Chem 1985; 260: 16321-35
220. Franceschini G, Weber JP, D'Acquarica AL, et al. Microsomal enzyme inducers raise plasma high-density lipoprotein cholesterol levels in healthy controls but not in patients with primary *hypoalphalipoproteinemia*. Clin Pharmacol Ther 1995; 57: 434-40
221. Ameli S, Hultgardh-Nilsson A, Cercek B, et al. Recombinant apolipoprotein A-IMilano reduces initimal thickening after balloon injury in hypercholesterolemic rabbits. Circulation 1994; 90: 1935-41
222. Booth RGF, Martin JF, Honey AC, et al. Rapid development of atherosclerosis lesions in the rabbit carotid artery induced by perivascular manipulation. Atherosclerosis 1989; 76: 257-68
223. Soma M, Donetti E, Parolini C, et al. Recombinant apolipoprotein A-IMilano dimer inhibits carotid intimal thickening induced by perivascular manipulation in rabbits. Circ Res 1995; 76: 405-11
224. Moran CS, Campbell JH, Simmons DL, et al. Human leukemia inhibitory factor inhibits development of experimental atherosclerosis. Arterioscler Thromb 1994; 14: 1356-63
225. Morel DW, Hessler JR, Chisolm GM. Low density lipoprotein cytotoxicity induced by free radical peroxidation of lipid. J Lipid Res 1983; 24: 1020-6
226. Steinbrecher UP, Parthasarathy S, Leake DS, et al. Modification of low density lipoprotein by endothelial cells involves lipid peroxidation and degradation of low density lipoprotein phospholipids. Proc Natl Acad Sci U S A 1984; 81: 3883-7
227. O'Brien KD, Alpers CE, Hokanson JE, et al. Oxidation-specific epitopes in human coronary atherosclerosis are not limited to oxidized low-density lipoprotein. Circulation 1996; 94: 1216-25
228. Jha P, Flather M, Lonn E, et al. The antioxidant vitamins and cardiovascular disease: a critical review of epidemiologic and clinical trial data. Ann Intern Med 1995; 123: 860-72
229. Finckh B, Niendorf A, Rath M, et al. Antiatherosclerotic effect of probucol in WHHL rabbits: are there plasma parameters to evaluate this effect? Eur J Clin Pharmacol 1991; 40: S77-80
230. Suzukawa M, Ayaori M, Shige H, et al. Effect of supplementation with vitamin E on LDL oxidizability and prevention of atherosclerosis. Biofactors 1998; 7: 51-4
231. Fruebis J, Bird DA, Pattison J, et al. Extent of antioxidant protection of plasma LDL is not a predictor of the antiatherogenic effect of antioxidants. J Lipid Res 1997; 38: 2455-64
232. Johnson MB, Heineke EW, Rhinehart BL, et al. MDL 29311: antioxidant with marked lipid- and glucose-lowering activity in diabetic rats and mice. Diabetes 1993; 42: 1179-86
233. Kwiterovich Jr PO. The effects of dietary fat, antioxidants, and pro-oxidants on blood lipids, lipoproteins, and atherosclerosis. J Am Diet Assoc 1997; 97 (7 Suppl.): S31-41
234. Odeh RM, Cornish LA. Natural antioxidants for the prevention of atherosclerosis. Pharmacotherapy 1995; 15: 648-59
235. Watkins TR, Bryan DL, Bierenbaum ML, et al. Modification of CHD risk by vitamin E and flax seed supplementation [abstract no. 268]. 9th International Symposium on Atherosclerosis; 1991 Oct 6-11: Rosemont (IL)
236. Harats D, Ben-Naim M, Dabach Y, et al. Effects of vitamin C and E on susceptibility of plasma lipoproteins to peroxidation induced by acute smoking. Atherosclerosis 1990; 85: 47-54
237. Jialal I, Fuller CJ. Effect of vitamin E, Vitamin C and beta-carotene on LDL oxidation and atherosclerosis. Can J Cardiol 1995; 11 Suppl.: 97G-103
238. The Alpha-Tocopheral, Beta Carotene Cancer Prevention Study Group. The effect of vitamin E and beta carotene on the incidence of lung cancer and other cancers in male smokers. N Engl J Med 1994; 330: 1029-35
239. Jialal I, Grundy SM. Effect of combined supplementation with α-tocopherol, ascorbate, and beta-carotene on low-density lipoprotein oxidation. Circulation 1993; 88: 2780-6
240. Mosca L, Rubenfire, Mandel C, et al. Antioxidant nutrient supplementation reduces the susceptibility of low density lipoprotein to oxidation in patients with coronary artery disease. J Am Coll Cardiol 1997; 30: 392-9
241. Van de Vijer LP, Kardinaal AF, Grobbee DE, et al. Lipoprotein oxidation, antioxidants and cardiovascular risk: epidemiologic evidence. Prostaglandins Leukot Essent Fatty Acids 1997; 57: 479-87
242. Schwenke DC, Behr SR. Vitamin E combined with selenium inhibits atherosclerosis in hypercholesterolemic rabbits independently of effects on plasma cholesterol concentrations. Circ Res 1998; 83: 366-77

243. Brownlee M, Vlassara H, Kooney T, et al. Aminoguanidine prevents diabetes-induced arterial wall protein cross-linking. Science 1986; 232: 1629-32
244. Skottova N, Krecman V, Simanek V. Activities of silymarin and its flavonolignans upon low density lipoprotein oxidizability in vitro. Phytother Res 1999; 13 (6): 535-7
245. Galis ZS, Asanuma K, Godin D, et al. N-acetyl-cysteine decreases the matrix-degrading capacity of macrophage-derived from cells: new target for antioxidant therapy. Circulation 1998; 97: 2445-53
246. Aviram M, Hussein O, Rosenblat M, et al. Interactions of platelets, macrophage, and lipoproteins in hypercholesterolemia: antiatherogenic effects of HMG-CoA reductase inhibitor therapy. J Cardiovasc Pharmacol 1998; 31: 39-45
247. Verhoef P, Stampfer MJ, Rimm EB. Folate and coronary heart disease. Curr Opin Lipidol 1998; 9: 17-22
248. Davey PJ, Schulz M, Gilksman M, et al. Cost-effectiveness of vitamin E therapy in the treatment of patients with angiographically proven coronary narrowing (CHAOS trial): Cambridge Heart Antioxidant Study. Am J Cardiol 1998; 82: 414-7
249. de Lorgeril M, Salen P, Martin J-L, et al. Effect of a Mediterranean type of diet on the rate of cardiac complications in patients with coronary artery disease: insights into the cardioprotective effects of certain nutrients. J Am Coll Cardiol 1996; 29: 1103-8
250. Manson JE, Gaziano JM, Spelsberg A, et al. A secondary prevention trial of antioxidant vitamin and cardiovascular disease in women: rational, design, and methods. The WACS research group. Ann Epidemiol 1995; 5: 261-9
251. Hayek T, Attias J, Smith J, et al. Antiatherosclerotic and antioxidative effects of captopril in apolipoprotein E-deficient mice. J Cardiovasc Pharmacol 1998; 31: 540-4
252. Jackson SM, Parhami F, Xi X-P, et al. Peroxisome proliferator-activated receptor activators target human endothelial cells to inhibit leukocyte-endothelial cell interaction. Arterioscler Thromb Vasc Biol 1999; 19: 2094-104
253. Chinetti G, Griglio S, Antonucci M, et al. Activation of proliferator-activated receptors α and γ induces apoptosis of human monocyte-derived macrophages. J Biol Chem 1998; 273: 25573-80
254. Willson TM, Wahli W. Peroxisome proliferator-activated receptor agonists. Curr Opin Chem Biol 1997; 1: 235-41
255. Marx N, Sukhova G, Murphy C, et al. Macrophages in human atheroma contain PPAR-γ: differentiation-dependent peroxisomal proliferator-activated receptor-γ (PPAR-γ) expression and reduction of MMP-9 activity through PPAR-γactivation in mononuclear phagocytes in vitro. Am J Pathol 1998; 153: 17-23
256. Minamikawa J, Yamauchi M, Inoue D, et al. Another potential use of troglitazone in noninsulin-dependent diabetes mellitus. J Clin Endocrinol Metab 1998; 83: 1041-2
257. Hertz R, Bishara-Shieban J, Bar-Tana J. Mode of action of peroxisome peroliferators as hypolipidemic drugs: suppression of apolipoprotein C-III. J Biol Chem 1995: 270; 13470-5
258. Saitoh K, Mori T, Kasai H, et al. Anti-atheromatous effects of fenofibrate, a hypolipidemic drug: I: Anti-atheromatous effects are independent of its hypolipidemic effect in cholesterol-fed rabbits. Folia Pharmacol Jpn 1995; 106: 41-50
259. Staels B, Koeing W, Habib A, et al. Activation of human aortic smooth-muscle cells is inhibited by PPAR-α but not by PPAR-γactivators. Nature 1998; 393: 790-3
260. Lendon CL, Davies MJ, Born GVR, et al. Atherosclerotic plaque caps are locally weakened when macrophage density is increased. Atherosclerosis 1991; 87: 87-91
261. Galis ZS, Sukhova GK, Lark MW, et al. Increased expression of matrix metalloproteinases and matrix degrading activity in vulnerable regions of human atherosclerotic plaques. J Clin Invest 1994; 94: 2493-503
262. Zaltsman AB, George SJ, Newby AC. Increased secretion of tissue inhibitors of metalloproteinases 1 and 2 from the aortas of cholesterol fed rabbits partially counterbalances increased metalloproteinase activity. Arterioscler Thromb Vasc Biol 1999; 19: 1700-7
263. Zaltsman AB, Newby AC. Increased secretion of gelatinases A and B from the aortas of cholesterol fed rabbits: relationship to lesion severity. Atherosclerosis 1997; 130: 61-70
264. Galis ZS, Sukhova GK, Libby P. Microscopic localization of active proteases by in situ zymography: detection of matrix metalloproteinase activity in vascular tissue. FASEB J 1995; 9: 974-80
265. Cawston TE. Metalloproteinase inhibitors and the prevention of connective tissue breakdown. Pharmacol Ther 1996; 70: 163-82
266. George SJ, Johnson J, Angelini GD, et al. Adenovirus mediated gene transfer of the human TIMP-1 gene inhibits smooth muscle cell migration and neointima formation in human saphenous vein. Hum Gene Ther 1998; 9: 867-77
267. Trigatti B, Rayburn H, Vinals M, et al. Influence of the high density lipoprotein receptor SR-BI on reproductive and cardiovascular pathophysiology. Proc Natl Acad Sci U S A 1999; 96: 9322-7
268. Kozarsky KF, Donahee MH, Rigotti A, et al. Overexpression of the HDL receptor SR-BI alters plasma HDL and bile cholesterol levels. Nature 1997; 387: 414-7
269. Rigotti A, Trigatti BL, Penman M, et al. A targeted mutation in the murine gene encoding the high density lipoprotein (HDL) receptor scavenger receptor class B type I reveals its key role in HDL metabolism. Proc Natl Acad Sci U S A 1997; 94: 12610-5
270. Varban ML, Rinninger F, Wanf N, et al. Targeted mutation reveals a central role for SR-BI in hepatic selective uptake of high density lipoprotein cholesterol. Proc Natl Acad Sci U S A 1998; 95: 4619-24
271. Xu S, Laccotripe M, Huang X, et al. Apolipoproteins of HDL can directly mediate binding to the scavenger receptor SR-BI, an HDL receptor that mediates selective lipid uptake. J Lipid Res 1997; 38: 1289-98
272. Ji Y, Jian B, Wang N, et al. Scavenger receptor BI promotes high density lipoprotein-mediated cellular cholesterol efflux. J Biol Chem 1997; 272: 20982-5
273. Stangl H, Cao G, Wyne KL, et al. Scavenger receptor, class B, type I-dependent stimulation of cholesterol esterification by high density lipoproteins, low lipoproteins, and nonlipoprotein cholesterol. J Biol Chem 1998; 273: 31002-8
274. Arai T, Wang N, Bezouevski M, et al. Decreased atherosclerosis in heterozygous low density lipoprotein receptor-deficient mice expressing the scavenger receptor BI transgene. J Biol Chem 1999; 274: 2366-71
275. Rinninger F, Wang N, Ramakrishnan R, et al. Probucol enhances selective uptake of HDL-associated cholesteryl esters in vitro by a scavenger receptor B-I-dependent mechanism. Arterioscler Thromb Vasc Biol 1999; 19: 1325-32
276. Spady DK, Kearney DM, Hobbs HH. Polyunsaturated fatty acids up-regulate hepatic scavenger receptor B1 (SR-BI) expression and HDL cholesteryl ester uptake in the hamster. J Lipid Res 1999; 40: 1384-94
277. Nevin DN, Zambon A, Furlong CE, et al. Paroxonase genotypes lipoprotein lipase activity, and HDL. Arterioscler Thromb Vasc Biol 1996; 16 (10): 1243-9

278. Temple NJ. Dietary fats and coronary heart disease. Biomed Pharmacother 1996; 50: 261-8
279. Bonanome A, Grundy SM. Effects of dietary stearic acid on plasma cholesterol and lipoprotein levels. N Engl J Med 1988; 318: 1244-8
280. Pathasarathy S, Khoo JC, Miller E, et al. Low density lipoprotein rich in oleic acid is protected against oxidative modification: implication for dietary prevention of atherosclerosis. Proc Natl Acad Sci U S A 1990; 87: 3894-8
281. Massaro M, Carluccio MA, De Caterina R. Direct vascular atherogenic effects of oleic acid: a clue to the cardioprotective effects of the Mediterranean diet. Cardiologia 1999; 44 (6): 507-13
282. Davidson MH, Burns JH, Subbaiah PV, et al. Marine oil capsule therapy for the treatment of hyperlipidemia. Arch Intern Med 1991; 151: 1732-40
283. Daviglus M, Stamler J, Greenland P, et al. Fish consumption and the 30-year risk of fatal myocardial infarction. N Engl J Med 1997; 336: 1046-53
284. Swahn E, van Schenk H, Olsson AG. Omega-3 ethyl ester concentration decrease total apolipoproteins CIII and increase antithrombin III in postmyocardial infarction patients. Clin Drug Invest 1998; 15: 473-82
285. Scaccini C, Hardini M, D'Aquina M, et al. Effects of fish oil in lipid peroxidation and antioxidant defense system of plasma and lipoprotein fraction in rats (abstract no. 193). 9th International Symposium on Atherosclerosis; 1991 Oct 6-11: Rosemont (IL), 144
286. Davidson MH, Daugan LD, Burns JH, et al. The hypocholesterolemic effects of beta-glucan in oatmeal and oatbran: a dose-controlled study. JAMA 1991; 265: 1833-9
287. Hunninghake DB. Cholesterol-lowering effects of psyllium hydrophilic mucilloid. JAMA 1989; 261: 3419-23
288. Muller C. The regulatory status of medical food and dietary supplements in the United States. Nutrition 1999; 15: 249-51
289. Boger RH, Bode-Boger SM, Kienke S, et al. Dietary L-arginine decreases myoinitimal cell proliferation and vascular monocyte accumulation in cholesterol-fed rabbits. Atherosclerosis 1998; 136: 67-77
290. Quyyumi AA, Mulcahy D, Andrews NP, et al. Coronary vascular nitric oxide activity in hypertension and hypercholesterolemia. Circulation 1997; 95: 104-10
291. Egashira K, Hirooka Y, Kuga T, et al. Effects of L-arginine supplementation on endothelium-dependent coronary vasodilation in patients with angina pectoris and normal coronary arteries. Circulation 1996; 94: 130-4
292. McKenney JM. New guidelines for managing hypercholesterolemia. Am Pharm 1993; NS33: 3-11
293. Pedersen TR, Olsson AG, Faergeman O, et al. Lipoprotein changes and reduction in the incidence of major coronary heart disease events in the Scandinavian Simvastatin Survival Study (4S). Circulation 1998; 97 (15): 1453-60
294. Gordon BR, Saal SD. Clinical experience and future directions for low-density lipoprotein apheresis in the United States. Ther Apher 1997; 1 (3): 249-52

Correspondence and offprints: Dr *Pang H. Chong*, Cook County Hospital, 1900 W. Polk St, Suite #552, Chicago, IL 60612-3785 USA.
E-mail: cpang@tigger.cc.uic.edu