1

Gene Polymorphism and Coronary Heart Disease

Nduna Dzimiri
King Faisal Specialist Hospital and Research Centre
Saudi Arabia

1. Introduction

Like all other organs and tissues in a mammalian body, the heart muscle requires oxygen-rich blood in order to function, while oxygen-depleted blood needs to be transported away from organs. The first requirement is accomplished through the coronary artery networks which serve to furnish blood supply to the cardiac muscle, while the second is attained through a network of blood veins.

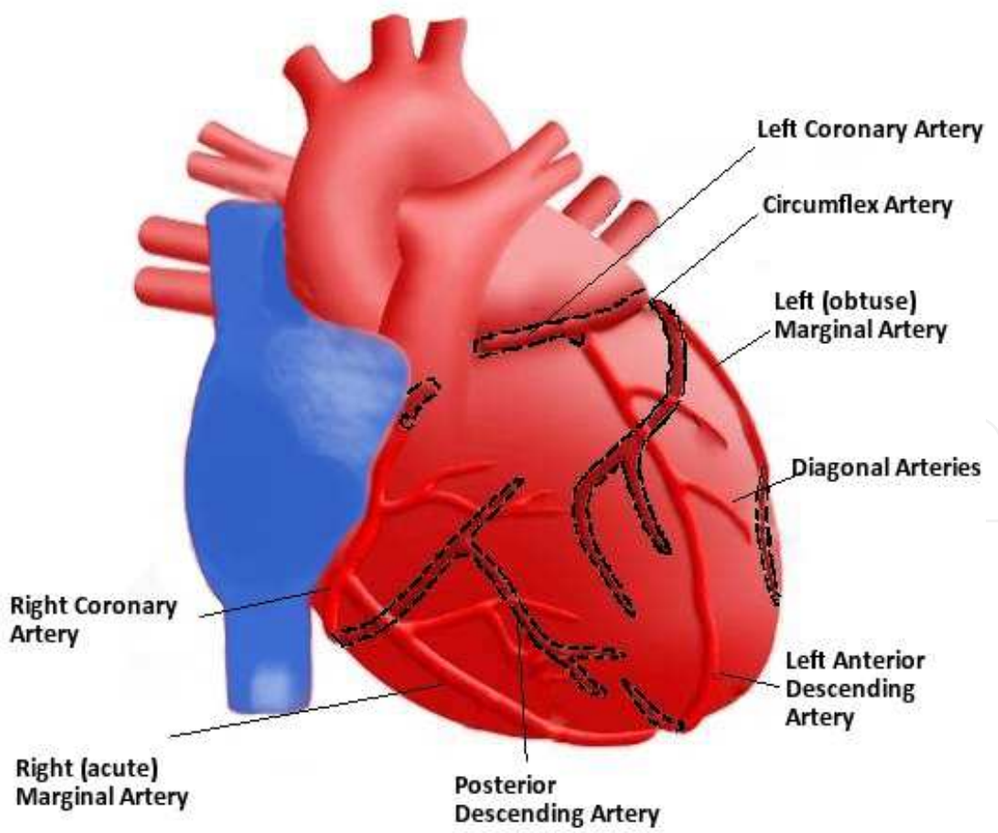


Fig. 1. The coronary artery network of the heart.

The coronary artery system consists of two main arteries, the left and right arteries, and their marginal arterial network. Anatomically, the left main coronary artery (LMCA) consists of the left anterior descending artery and the circumflex branch that supplies blood to the left ventricle and atrium (Figure 1). Additional arteries branching off the LMCA furnish the left heart side muscles with blood. These include the circumvent artery, which branches off the left coronary artery and encircles the muscle to supply blood to the lateral side and back of the heart, and the left anterior descending artery, which supplies the blood to the front of the left side of the heart. The right coronary artery, on the other hand, branches into the right posterior descending and acute marginal arteries to supply blood to the right ventricle, right atrium sinoatrial node (cluster of cells in the right atrial wall that regulate the heart's rhythmic rate) and the atrioventricular node. Other smaller branches of the coronary arteries include the acute marginal, posterior descending, obtuse marginal, septal perforator and the diagonals, which provide the surrounding network with blood supply.

2. What causes coronary artery disease?

Since coronary arteries deliver blood to the heart muscle, disease can have serious implications by reducing the flow of oxygen and nutrients to the heart, which in turn may lead to a heart attack and possibly death. Besides, the susceptibility and function of the vessel wall are dependent on the balance of several counteracting forces, i.e., vasoconstricting versus vasodilating, growth-promoting versus growth-inhibiting and proapoptotic versus antiapoptotic functions. While these factors are tightly balanced in a normal healthy vessel, under pathophysiologic conditions this balance is upset resulting in the development of vascular hypertrophy and the generation of proatherogenic vascular lesions, thereby triggering the initiation and progression of coronary artery disease (CAD) or atherosclerosis (as it is often called). CAD is a complex disorder, which culminates in the coronary bed system failing to furnish adequate blood supply to the heart. The disease has no clear etiology and manifests itself both in the young (early onset) as in familial cases as well as in adults (late onset).

One major culprit for CAD is a perturbation in lipid metabolism. A lipid is any fatty material or fat-like substance, e.g. normal body fat, cholesterol (i.e. high density and low density lipoproteins), other steroids, and substances containing fats, such as phospholipids, glycolipids and lipoproteins. Specifically, cholesterol (Chol) constitutes one of the most clinically important lipid substances, and alterations in the balance between its synthesis and the metabolism of its by-products is a major source of cardiovascular complications. Together with its metabolites, Chol plays a variety of essential roles in living systems. In fact, virtually all animal cells require Chol, which they acquire through synthesis or uptake, but only the liver can degrade it. Failure of metabolic pathways to break these substances down as required, or by the system to maintain the right balance, will lead to their increased levels, and therefore predispose an individual to acquiring CAD. Circulating lipoproteins are regulated at various levels, ranging from Chol synthesis, transport and metabolism. Chol biosynthesis comprises the condensation of acetyl-CoA and acetoacetyl-CoA into 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA), which in turn is reduced to mevalonate by the HMG-CoA reductase (HMGCR) followed by oxidoreduction and decarboxylation intermediate steps leading to the

conversion of lanosterol to Chol. It is regulated through feedback inhibition by sterols and non-sterol metabolites derived from mevalonate. The HMGCR, an integral glycoprotein of endoplasmic reticulum membranes that remains in the endoplasmic reticulum after synthesis and glycosylation, is the rate-limiting enzyme in Chol biosynthesis in vertebrates.

The maintenance of Chol homeostasis encompasses diverse metabolic pathways linked to its transportation, metabolism and its metabolic by-products. Since Chol is insoluble in blood, it is transported in the circulatory system within a large range of lipoproteins in blood. Thereby, its transportation to peripheral tissues is accomplished by the lipoproteins, chylomicrons, very low density lipoproteins (VLDLs) and low density lipoproteins (LDLs), while high-density lipoprotein (HDL) particles transport it back to the liver for excretion. Functionally, LDL is a metabolic end-product of the triglyceride (TG)-rich lipoproteins (i.e. VDLDs). Lipoprotein metabolism constitutes a major component in the regulation of Chol homeostasis. Systems regulating Chol levels include the LDL receptor (LDLR) pathway and its ligand the apolipoprotein B100 (Apo B), which are involved in the maintenance of its homeostasis in the body by regulating the hepatic catabolism of LDL-Cholesterol (LDL-C), and the proprotein convertase subtilisin/kexin type 9 (PCSK9). The serine protease PCSK9 is a member of the proteinase K subfamily of subtilases that also affects plasma LDL-C levels by altering LDLR levels via a post-transcriptional mechanism[1], and cellular Chol metabolism by regulating both LDLR and circulating Apo B-containing lipoprotein levels in LDLR-dependent and -independent fashions[2-4]. It regulates LDL-C levels apparently both intracellularly and by behaving as a secreted protein that can be internalized by binding the LDLR. In the liver, it controls the plasma LDL-C level by post-transcriptionally downregulating the LDLR through binding to the epidermal growth factor-like repeat A (EGF-A) domain of the LDLR[5], independently of its catalytic activity. The LDLR regulates the plasma LDL-C concentration by internalizing Apo B and Apo E-containing lipoproteins via receptor-mediated endocytosis. Apo B is a large, amphipathic glycoprotein that plays a central role in human lipoprotein metabolism. Two forms of Apo B are produced from the *Apo B* gene through a unique posttranscriptional editing process: Apo B-48, which is required for chylomicron production in the small intestine, and Apo B required for VLDL production in the liver. In addition to being the essential structural component of VLDL, Apo B is the ligand for LDLR-mediated endocytosis of LDL particles. Lipoprotein (a) [Lp(a)] contains an LDL-like moiety, in which the Apo B component is covalently linked to the unique glycoprotein apolipoprotein(a) (Apo A). Apo A-I is essential for the formation of HDL particles. Apo A-I-containing HDL particles play a primary role in Chol efflux from membranes, at least partly, through interactions with the adenosine triphosphate-binding cassette transporter A1 (ABCA1). The ABCA1 regulates the rate-controlling step in the removal of cellular Chol, i.e. the efflux of cellular Chol and phospholipids to an apolipoprotein acceptor. Apo A is composed of repeated loop-shaped units called kringles, the sequences of which are highly similar to a kringle motif present in the fibrinolytic proenzyme plasminogen. Variability in the number of repeated kringle units in the Apo A molecule gives rise to different-sized Lp(a) isoforms in the population. Based on the similarity of Lp(a) to both LDL and plasminogen, its function is thought to represent a link between atherosclerosis and thrombosis. However, determination of the function of Lp(a) in vivo remains elusive. Mechanistically, elevated Lp(a) levels may

either induce a prothrombotic/anti-fibrinolytic effect as Apo A resembles both plasminogen and plasmin but has no fibrinolytic activity, and/or may accelerate atherosclerosis because, like LDL, the Lp(a) particle is Chol-rich. Apolipoprotein E (apo E) is also a constituent of various lipoproteins and plays an important role in the transport of Chol and other lipids among cells of various tissues.

Apart from lipoproteins, circulating VLDLs serve as vehicles for transporting lipids to peripheral tissues for energy homeostasis. VLDLs are TG-rich particles synthesized in hepatocytes and secreted from the liver in a pathway that is tightly regulated by insulin. Hepatic VLDL production is stimulated in response to reduced insulin action, resulting in increased release of VLDL into the blood under fasting conditions, while it is suppressed in response to increased insulin release after meals. This effect is critical for preventing prolonged excursion of postprandial plasma lipid profiles in normal individuals. HDL particles are key players in the reverse Chol transport by shuttling it from peripheral cells (e.g. macrophages) to the liver or other tissues. This complex process is thought to represent the basis for the antiatherogenic properties of HDL particles. Accordingly, the particles mediate the uptake of peripheral Chol and return it to the liver for bile acid secretion through exchange of core lipids with other lipoproteins or selective uptake by specific receptors. These particles vary in size and density, mainly because of differences in the number of apolipoprotein (apo) particles and the amount of Chol ester in the core of HDL molecule.

The majority of lipoprotein disorders result from a combination of polygenic predisposition and poor lifestyle habits, including physical inactivity, increased visceral adipose tissue, increased caloric intake, and cigarette smoking. Plasma elevation of LDL-C, VLDL and Lp(a), as well as reduced HDL-C levels are predisposing factors for CAD. The amount of Chol transported is inversely correlated with the risk for CAD. Reduced circulating levels of HDL-C are a frequent lipoprotein disorder in CAD patients and can be caused by either genetic and/or environmental factors (sedentary lifestyle, diabetes mellitus, smoking, obesity or a diet enriched in carbohydrates. Lp(a) is a unique lipoprotein particle consisting of a moiety identical to LDL to which the glycoprotein Apo(a) that is homologous to plasminogen is covalently attached. These features have suggested that Lp(a) may contribute to both proatherogenic and prothrombotic/antifibrinolytic processes, which constitute a risk factor for premature cardiovascular disease. Among others, the association between elevated Lp(a) levels and increased CAD risk, indicates that elevated Lp(a), like elevated LDL-C, is causally related to premature CAD. However, although Lp(a) has been shown to accumulate in atherosclerotic lesions, its contribution to the development of atheromas is unclear. This uncertainty is related in part to the structural complexity of the Apo A component of Lp(a) (particularly apo A isoform size heterogeneity), which also poses a challenge for standardization of the measurement of Lp(a) in plasma. However, while LDL and Lp(a) are now believed to be atherogenic, the role of VLDL as an independent risk factor remains controversial. On the other hand, the HDL is the only lipoprotein that has been established as antiatherogenic, thought to reduce CAD risk by mediating Chol efflux from the periphery by way of transportation to the liver for excretion.

Hepatic lipase (HL), a member of the lipase superfamily, is a lipolytic enzyme that is produced primarily by hepatocytes, where it is secreted and bound to the hepatocyte

surface and readily released by heparin. It is homologous to LPL and pancreatic lipase and hydrolyzes TGs and phospholipids in all lipoproteins, but is predominant in the conversion of IDL to LDL and the conversion of post-prandial TG-rich HDL into the post absorptive TG-poor HDL. Thus, the enzyme contributes to the regulation of plasma TG levels by facilitating its exudation from the VLDL pool, in a fashion that is governed by the composition and quality of HDL particles. It is thought that HL directly couples HDL lipid metabolism to tissue/cellular lipid metabolism[43]. Accordingly, hepatic lipase HDL regulates the release of HL from the liver and HDL, controls HL transport and activation in the circulation in a fashion that is regulated by factors that release it from the liver and activate it in the bloodstream. Therefore, alterations in HDL-apolipoprotein composition can disturb HL function by inhibiting the release and activation of the enzyme, thereby affecting plasma TG levels and CAD risk. It has been suggested that the HL pathway potentially provides the hepatocyte with a mechanism for the uptake of a subset of phospholipids enriched in unsaturated fatty acids and may allow the uptake of cholesteryl ester, free Chol, and phospholipid without catabolism of HDL apolipoproteins[43]. HL plays a secondary role in the clearance of chylomicron remnants by the liver[43]. Consistent with IDL being a substrate for HL, the human post-heparin HL activity is inversely correlated with IDL-C concentration only in subjects with a hyperlipidaemia involving VLDL. HDL-C has been reported to be inversely correlated to HL activity, leading to the suggestion that lowering HL would increase its levels. Common hormonal factors, such as estrogen, have also been shown to upregulate Apo A and HDL-C and lower HL. Hence this relationship may not be specific. However, an increase in HDL-C, Apo A, or HDL –triglycerides has been observed in severe deficiency of HL. Apart from heparin, HL also binds o the LDLR-related protein. This has led to the notion that enzymatically inactive HL may play a role in hepatic lipoprotein uptake, forming a bridge by binding to the lipoprotein and to the cell surface, raising the possibility that production and secretion of mutant inactive HL could promote clearance of VLDL remnants.

One of the important proteins involved in lipoprotein metabolism is the CETP. Plasma CETP facilitates the transfer of cholesteryl ester from HDL to Apo B-containing lipoproteins by catalyzing the transfer of insoluble esters in the reverse transport of Chol. Thus, CETP is involved in maintaining the balance between the LDL-C, ("bad" Chol) and HDL-C ("good" Chol), which is a risk factor for hyperlipidaemia. Since CETP regulates the plasma levels of HDL-C and the size of HDL particles, it is considered to be a key protein in reverse Chol transport, a protective system against atherosclerosis. In mediating the transfer of cholesteryl esters from antiatherogenic HDL to proatherogenic apolipoprotein apo-B-containing lipoprotein particles (including VLDL, VLDL remnants, IDL, and LDL), the CETP plays a critical role not only in the RCT pathway but also in the intravascular remodeling and recycling of HDL particles[6,7].

In mammalian cells, Chol homeostasis is controlled primarily by regulated cleavage of membrane-bound transcription factors, the sterol regulatory element binding proteins (SREBPs)[8-10] and their cleavage-activating protein (SCAP)[11,12]. The SREBPs activate specific genes involved in Chol synthesis, LDL endocytosis, fatty acid synthesis and glucose metabolism, providing a link between lipid and carbohydrate metabolism[13]. All three SREBP isoforms SREBP-1a, SREBP-1c and SREBP-2 (encoded by two genes) that are

synthesized as 125 kDa precursor proteins localized to the endoplasmic reticulum[14], and play a central role in energy homeostasis by promoting glycolysis, lipogenesis and adipogenesis[15-19]. Functionally, SREBP-2 gene activation leads to enhanced Chol uptake and biosynthesis, while SREBP-1 is primarily involved in fatty acid and glucose metabolism[20-22]. Mechanistically, it is thought that when cells are deprived of Chol, SREBPs are cleaved by two proteolytic steps. First, the SREBP precursor is transported to the Golgi by the chaperone protein SCAP and cleaved via two proteases to release the mature, transcriptionally active 68 kDa amino terminal domain[11,12]. This domain is then released from the endoplasmic reticulum membrane and transported into the nucleus, where it binds to specific nucleotide sequences in the promoters of the LDLR and other genes regulating Chol and TG homeostasis[10].

While disorders of Chol metabolism and related lipoproteins occupy a pivotal position in events leading to CAD, other important contributors include those that regulate the viability of blood vessels affecting vascular function leading to the formation of plaques. This is particularly true for coronary arteries, for example, in which an imbalance in a number of these processes triggers a proatherogenic state initiating the progression of atherosclerosis. Several disease pathways associated with regulation of blood pressure and glucose metabolism are involved in intricate ways. The metabolic syndrome poses a major public health problem by predisposing individuals to CAD and stroke, the leading causes of mortality in developed countries. Its impact on the risk of atherosclerotic cardiovascular disease is greater than that of any of its individual components. Frequently, many of these metabolic manifestations, precede the development of overt diabetes by many years[23]. This syndrome is manifest clinically by such cardiovascular risk factors as hypertension, dyslipidaemic, and coagulation abnormalities. This abnormal metabolic milieu contributes to the high prevalence of macrovascular complications including CAD as well as more generalized atherosclerosis. It is frequently seen in obese individuals, and characterized by glucose intolerance, hyperinsulinemia, a characteristic dyslipidaemic (high TGs; low HDL-C, and small, dense LDL-C), obesity, upper-body fat distribution, hypertension, increased prothrombotic and antifibrinolytic factors, and therefore an increased risk of CAD[23]. However, while the conventional risk factors, insulin resistance parameters, and metabolic syndrome are important in predicting CAD risk, in some population environmental factors may be determinant in the ultimate manifestation of the disease.

Apart from the PPARs, the Forkhead transcription factor (Foxo1) is also believed to play an essential role in controlling insulin-dependent regulation of microsomal TG transfer protein (MTP) and apolipoprotein C-III (Apo C-III), two key components that catalyze the rate-limiting steps in the production and clearance of triglyceride-rich lipoproteins[24,25]. Under physiological conditions, Foxo1 activity is inhibited by insulin, while under insulin resistant conditions Foxo1 becomes uncontrolled, contributing to its hyperactivity in the liver[24]. This effect contributes to hepatic overproduction of VLDL and impaired catabolism of triglyceride-rich particles, accounting for the pathogenesis of hypertriglyceridemia. Thus, augmented Foxo1 activity in insulin resistant livers promotes hepatic VLDL overproduction and predisposes to the development of hypertriglyceridemia[25], offering a possible route for this manifestation.

As such the contributions of the individual disease pathways such as diabetes or hypertension may manifest themselves independently or in combination with other disorders, leading to other complications with added risk as is the case with the metabolic syndrome. Among these, type 2 diabetes mellitus (T2DM) constitutes probably the most important risk disease for CAD. In contrast to type 1 diabetes mellitus, the type 2 endocrinopathy is clustered in minority populations and has both strong genetic and environmental components that influence its manifestation. This disease is a heterogeneous disorder and patients are often characterized by features of the insulin resistance syndrome, also referred to as the metabolic syndrome. This syndrome is defined as a cluster of interrelated common clinical disorders, including obesity, insulin resistance, glucose intolerance, hypertension, and dyslipidaemic (hypertriglyceridaemia and low HDL-C levels), exhibiting rising incidence to epidemic levels in the developed world. Among others, insulin has important vascular actions to stimulate production of nitric oxide from endothelium. This leads to capillary recruitment, vasodilatation, increased blood flow, and subsequent augmentation of glucose disposal in classical insulin target tissues, such as the skeletal muscle. Individuals displaying three of the features of insulin resistance (elevated plasma insulin and apolipoprotein B concentrations and small, dense LDL particles) show a remarkable increase in CAD risk and the increased risk associated with having small, dense LDL particles may be modulated to a significant extent by the presence/absence of insulin resistance, abdominal obesity and increased LDL particle concentration[26]. They influence risk for CAD through promoting endothelial dysfunction and enhanced production of procoagulants by endothelial cells[27].

Systems regulating the coagulation mechanisms also have their fair share of disease pathways leading to CAD. To begin with, blood platelets play a crucial role in physiological haemostasis and in the pathology of prothrombotic states, including atherosclerosis. Platelets are anucleate cells with no DNA. The differentiation of their precursor, the megakaryocyte, is characterized by nuclear polyploidization through a process called endomitosis. Changes in the megakaryocyte-platelet-haemostasis axis may precede acute thrombotic events. Thereby, the changes in megakaryocyte ploidy distribution may be associated with the production of large platelets. Large platelets are denser and more active haemostatically. Mean platelet volume, an important biological variable determinant of platelet reactivity, is increased in patients after MI and is a predictor of a further ischaemic event and death following MI[28]. Apart from platelets, changes also in the parental megakaryocyte are associated with chronic and acute vascular events[29]. The regulation of megakaryocytopoiesis depends on several haematopoietic factors such as thrombopoietin. In T2DM, platelet abnormalities, including altered adhesion and aggregation, render the cells hypersensitive to agonists. Besides, disturbed carbohydrate and lipid metabolism may lead to physicochemical changes in cell membrane dynamics, and consequently altered exposure of surface membrane receptors. These manifestations, together with increased fibrinogen binding, prostanoid metabolism, phosphoinositide turnover and calcium mobilization often present in diabetic patients, contribute to enhanced risk of small vessel occlusions and accelerated development of atherothrombotic disease of coronary, cerebral and other vessels in diabetes. The disease has emerged as an important condition of older patients in

which both microvascular and macrovascular complications are a common cause of morbidity and mortality. Microvascular complications have only been recently recognized as an important and frequent complication of T2DM[30].

Additionally, environmental factors, such as food consumption, constitute important components regulating pathways to complex diseases such as CAD and cancer. Dietary Chol absorption, endogenous Chol synthesis and biliary Chol excretion regulate whole body Chol balance as a result of biotransformation into bile acids or direct Chol excretion. Nuclear hormone receptors, such as the liver X, farnesoid X and retinoid X receptors, regulate the absorption of dietary sterols by modulating the transcription of several genes involved in Chol metabolism[31]. The ABC proteins transport dietary Chol from enterocytes back to the intestinal lumen, thus limiting the amount of absorbed Chol. By means of the same mechanism, ABC transporters also provide an efficient barrier against the absorption of plant sterols[31], which may vary among different ethnic populations. For example, it appears that some ethnic groups are at higher risk than others for the development of obesity and obesity-related non-communicable diseases, including insulin resistance, the metabolic syndrome, T2DM and CAD. Put together therefore, because of the various interactive factors regulating the functionality of coronary vascular beds, many risk factors may contribute to an individual acquiring the disease.

2.1 Coronary artery disease manifestation in the adult

These risk factors for CAD can be classified into those that regulate the levels of circulating lipoproteins, and those that regulate the susceptibility of arterial walls. Factors regulating lipoprotein levels include high circulating Chol and saturated fats in diet, obesity and emotional and neurogenic factors and those that regulate arterial susceptibility are insulin resistance, diabetes, hypertension, smoking and age (Figure 2). All these factors contribute to the pathways leading to atherosclerosis, oft in combination with environmental factors. Apart from the fact that most of the predisposing diseases are themselves complex ailments, combinations of some of these disorders can cluster to induce other forms of maladies, such as the metabolic syndrome, which in themselves constitute an added risk for coronary disease.

For almost all of these disease pathways, their impact on CAD manifestation can be classified as major or minor, depending on their assessed contribution (Figure 2). For example, in the regulation of blood pressure homeostasis insulin-resistant diabetes mellitus and essential hypertension are among the primary culprits, while angiotensin converting enzyme (ACE) and angiotensin 1 make up secondary causative factors. Furthermore, a great majority of these risk factors (e.g. hypertension, diabetes, hyperlipidaemia) are themselves complex disorders underlying a variety of etiologies. In essence therefore, CAD is shaped by a whole spectrum of phenotypic expression evolving through various events that lead to the formation and progression of atherosclerotic plaque in the inner lining of an artery causing it to narrow or become blocked and finally exude circulatory complications. The ultimate disease manifestation is an expression of the shift in the balance of interactions among factors influencing various aspects of coronary artery function and structure with prevalent genetic factors.

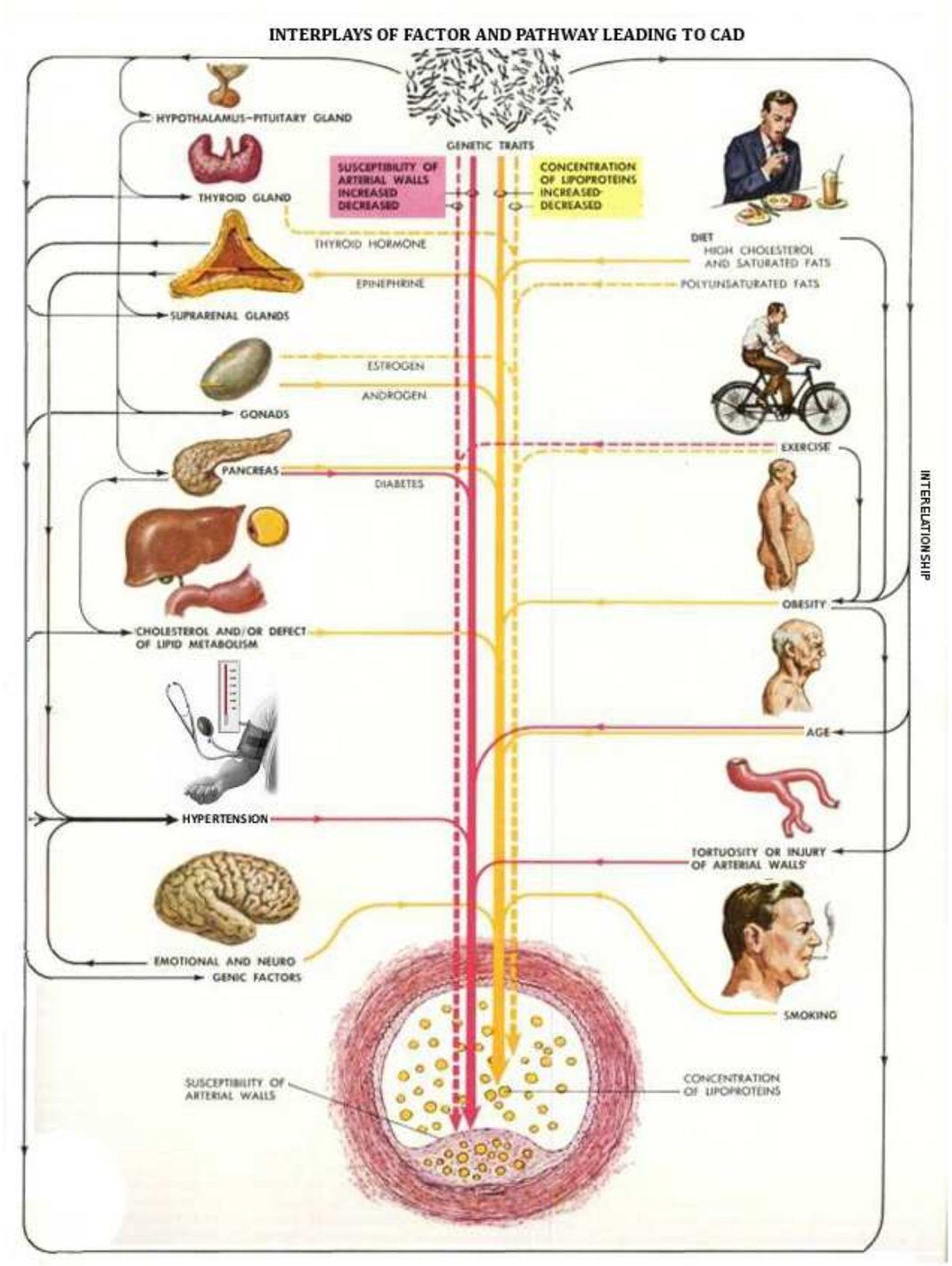


Fig. 2. The risk factors contributing to coronary artery disease pathways. The pathways may lead to either an increase in in circulating lipoproteins, influence susceptibility of the vessel walls or both components of atherosclerotic plaque formation.

2.1.1 Lipid metabolism and manifestation of coronary artery disease

As stated above, defects in lipid metabolic pathways, leading to increased circulating Chol levels constitutes the major underlying cause of CAD. Hyperlipidaemia, also sometimes referred to as hyperlipoproteinemia or dyslipidaemia, is the condition in which the blood lipid levels are too high, often leading to various cardiovascular disorders, particularly CAD. These disorders exist as hypercholesterolaemia, which is characterized by increased LDL levels, hypertriglyceridaemia characterized by increased chylomicrons or VLDL levels or combined disease manifest as increased LDL and VLDL, or VLDL and chylomicrons. Affected individuals may also have hypercholesterolaemia or chylomicron abnormalities of VLDL origin, with normal LDL-C levels, which is also defined as hyperlipoproteinemia.

Six subtypes of hyperlipoproteinemia types (I, II, III, IV, V and unclassified forms) have been characterized to date. Type I (also known as Buerger-Gruetz syndrome, primary hyperlipoproteinemia, or familial hyperchylomicronemia) is a very rare form due to a deficiency of lipoprotein lipase (LPL) or altered apolipoprotein C2. It results in elevated chylomicrons, the particles that transfer fatty acids from the digestive tract to the liver, exhibiting a prevalence of 0.1% of the population. Hyperlipoproteinemia type II, by far the most common form, is further classified into type IIa and type IIb, depending mainly on whether there is elevation in the TG level in addition to LDL-C. Thereby type IIa is known as familial hypercholesterolaemia. This form may be sporadic (due to dietary factors), polygenic, or truly familial as a result of a mutation either in the LDLR gene on chromosome 19 (0.2% of the population) or the Apo B gene (0.2%). In type IIb, the high VLDL levels are due to overproduction of substrates, including TGs, acetyl CoA, and an increase in Apo B synthesis. They may also be caused by the decreased clearance of LDL, with a prevalence of 10% in the population. These include (a) familial combined hyperlipoproteinemia (FCH) and (b) secondary combined hyperlipoproteinemia (usually in the context of metabolic syndrome, for which it is a diagnostic criterion). Hyperlipoproteinemia type III, also known as broad beta disease or dysbetalipoproteinemia, is due to high chylomicrons and intermediate density lipoprotein (IDL). It is due to the presence of elevated Chol-rich VLDL (β-VLDL) levels showing a prevalence of 0.02% in the general population.

The type IV form, also known as hypertriglyceridaemia (or pure hypertriglyceridaemia) is due to high TGs, with prevalence of 1%, while type V is very similar to type I, but with high VLDL in addition to chylomicrons. The rate of synthesis of TG-rich lipoproteins, LPL-mediated TG hydrolysis, and the hepatic capture of chylomicron remnants via the interaction of the lipoprotein receptor with Apo E and LPL, is key to the metabolism and modification of these lipoproteins. The modulation of such phenomena is influenced by both genetic and environmental factors, thus explaining their extraordinary individual variance[32]. Non-classified forms are extremely rare. They include hypo-alpha lipoproteinemia and hypo-beta lipoproteinemia with a prevalence of 0.01 - 0.1%. In subjects with and T2DM, the ability of insulin to regulate VLDL production becomes impaired due to insulin resistance in the liver, resulting in excessive VLDL secretion and accumulation of TG-rich particles in the blood. Such abnormality in lipid metabolism characterizes the pathogenesis of hypertriglyceridaemia and accounts for increased risk of CAD in obesity and T2DM. Accumulating evidence also points to hypertriglyceridaemia as a marker for increased risk for CAD, and in fact, several atherogenic factors, such as increased concentrations of TG-rich lipoproteins, the atherogenic lipoprotein phenotype, (or lipid triad) and the metabolic syndrome[33,34]. The lipid triad consists of elevated serum TGs, small LDL particles, and HDL-C. The metabolic syndrome includes the

coexistence of the lipid triad, elevated blood pressure, insulin resistance (plus glucose intolerance), and a prothrombotic state. However, the molecular basis that links insulin resistance to VLDL overproduction remains poorly understood.

Pathway	Primary risk determinants	Secondary risk determinants
Lipid metabolism	Familial dyslipidaemic disorders, including familial hypercholesterolaemia, familial defective apopliporptein B-100; Apopliporptein B-100; Sitosterolemia; Type III Hyperlipoproteinemia, Homocystinuria, Familial HDL deficiencies (apolipoprotein A1), Familial Combined Hyperlipidemia Apopliporptein B-100;.	Lipoprotein lipase; Hepatic lipase;, Lysosomal acid lipase; Sphingomyelinase; Lecithin-cholesterol acyltransferase; Apolipoproteins AII, AIV, CII, and CIII; proprotein convertase subtilisin/kexin type 9; ATP binding cassette protein subtype G5/8; Niemann-Pick type C1
Homeostasis of blood pressure	Insulin-resistant diabetes mellitus with acanthosis nigricans and hypertension Peroxisome proliferator-activated receptor-α; Angiotensinogen; ; Angiotensin converting enzyme; Primary hypertension;	Angiotensin II; Angiotensin II receptor, 11ß-ketoreductase; Cytochrome P450, family 1, subfamily B, polypeptide 1; Aldosterone synthase; Adducin; Nitric oxide synthase; Metafolin folate receptor; Thrombospondin
Glucose metabolism	Monogenic disorders leading to type 2 diabetes mellitus (e.g. insulin receptor, glucokinase); Peroxisome proliferator-activated receptor-α, -γ, -β;	Glucose transportersHuman leukocyte antigen locus DQ; Adiponectin; leptin receptor, melanocortin receptor 4; transcription factor 7-like 2; E-selectin
Hemostasis	Prothrombin; Factor V Leiden; Blood clotting disorders in conjunction with open foramen ovale; platelet-derived growth factor	Clotting factors II, V, VII; Fibrinogen; Plasminogen activator inhibitor-1; Thrombomodulin; transforming growth factor-β1
Inflammatory response genes	lInterleukin-6 and nuclear factor kappa-light-chain-enhancer of activated B cell; connexin 37;	Pyrin; Epithelial cell adhesion moleculeapolipoprotein *A*-I; Adenosine triphosphate-binding cassette transporter A1, and lecithin-cholesterol acyltransferase
Oxidative mediators	Homocystinuria; Paraoxone;, Methylenetetrahydrofolate reductase;	Cystathionine-beta-synthase; Nitric oxide synthase;
Adhesion mediators	Collagen IIIa; Glycoprotein IIIa, E-selectin; vascular cell adhesion molecule-1;	Epithelial cell adhesion molecule; Inter-cellular adhesion molecule 1
Gene regulators/ Transcritpion factors	Peroxisome proliferator-activated receptor-α,γ; Myocyte-specific enhancer factor 2A; Paraoxonase 1; GATA transcription factor-2	Sterol regulatory element binding protein; Hepatocyte nuclear factor 1-5; transcription factor 7-like 2

Table 1. Summary of the important risk genes and disease pathways leading to the manifestation of coronary artery disease.

Familial hypercholesterolemia (FH) is a clinical expression for the presence of elevated concentrations of LDL, deposition of LDL-derived Chol in tendons, skin xanthomas, and premature CAD, and an autosomal dominant trait of either increased serum Chol or premature CAD. It is also characterized by increased levels of total Chol and LDL-C, which result in excess deposition of Chol in tissues, leading to accelerated atherosclerosis and increased risk of premature CAD. Autosomal dominant FH is caused by LDLR deficiency and defective Apo B, respectively. Deficient LDLR activity results in elevated circulating LDL-C leading to its accumulation within blood vessel walls, which may result in arterial plaque formation, and therefore eventually occlude the arterial lumen. As such, HDL-C is often reduced in homozygous FH, while Lp(a) levels are high when corrected for Apo A isoforms[35]. While this phenomenon is likely to underlie genetic factors, the greater majority of these genes remain to be identified.

Autosomal recessive hypercholesterolemia (ARH) is a rare Mendelian dyslipidaemia characterized by markedly elevated plasma LDL levels, xanthomatosis, and premature CAD. LDLR function is normal, or only moderately impaired in fibroblasts from ARH patients, but their cultured lymphocytes show increased cell-surface LDL binding, and impaired LDL degradation, consistent with a defect in LDLR internalization[36]. Although the clinical phenotypes of ARH and homozygous FH are similar, autosomal recessive hypercholesterolaemia seems to be less severe, more variable within a single family, and more responsive to lipid-lowering drug therapy. In some individuals, the cardiovascular complications of premature atherosclerosis are delayed and involvement of the aortic root and valve is less common than in homozygous FH.

Another form of FH, heterozygous familial hypercholesterolemia (HFH) is an autosomal dominant disorder known to be associated with elevated Chol levels and increased risk of premature CAD. The estimated prevalence of HFH is 0.2% in most populations of the world. Apparently, the prevalence of peripheral arterial disease is increased dramatically in FH subjects compared with non-FH controls. In addition, the intima-media thickness of the carotid and/or femoral artery is increased in FH subjects.

Familial combined hyperlipidaemia (FCH) is a common heterozygous complex metabolic disorder characterized by (a) increase in cholesterolaemia and/or triglyceridaemia in at least two members of the same family, (b) intra-individual and intrafamilial variability of the lipid phenotype, and (c) increased risk of premature CAD. It is thought that in FCH elevated circulating levels of chylomicrons are broken down to produce atherogenic remnants through lipolysis of adipose cells, thereby contributing to the formation of atherosclerotic plaques. Thus, FCH is a common complex dominant disease condition associated with mixed hyperlipidaemia that accounts for up to 20% of premature CAD. The disease is sensitive to environmental effects. One of the roles of lipoprotein lipase is in the LDLR-like protein-mediated uptake of lipoprotein remnants in the liver and up to 20% of FCH patients show a genetic abnormality of this enzyme. FCH is very frequent (estimated prevalence: 0.5%-2.0%) and is one of the most common genetic hyperlipidaemias in the general population, being the most frequent in patients affected by CAD (10%) and among acute MI (AMI) survivors aged less than 60 (11.3%). The disease is subject to wide-scale environmental confounding traits such as obesity and the metabolic syndrome which remain to be elucidated.

Hypertriglyceridaemia represents one of the attributes of metabolic syndrome and is present in the most common genetic dyslipidaemia, the FCH. The disease also appears to underlie the phenomenon of small dense LDL in most instances. These particles are found in families with various disorders including premature CAD and hyperapobetalipoproteinemia, FCP, LDL subclass pattern B, familial dyslipidaemic hypertension, and syndrome X, as components of the metabolic syndrome[37]. Their presence is often accompanied by increased TGs and low HDL. While overproduction of VLDLs by the liver and increased secretion of large, Apo B-containing VLDL is the primary metabolic characteristic of most of these patients, this atherogenic syndrome seems to encompass also subclinical inflammation and elevated procoagulants. Their production occurs as a result of TG hydrolysis by LPL in VLDL, leading to the generation of IDL and in turn LDL. In LDL, the Chol esters are then exchanged for TG in VLDL by the Chol ester transfer proteins (CETPs), followed by hydrolysis of TG by HL to produce the particles. Apparently, CETP mediates a similar lipid exchange between VLDL and HDL, producing a Chol ester-poor HDL. In adipocytes, a primary defect in the incorporation of free fatty acids into TGs may trigger reduced fatty acid trapping and retention by adipose tissue, leading to increased levels of free fatty acids in plasma, increased flux of free fatty acids back to the liver, enhanced production of TGs, decreased proteolysis of Apo B, and increased VLDL production. Alternatively, insulin resistance may promote reduced retention of free fatty acids by adipocytes also leading to the same process. These metabolic disorders are often accompanied by decreased removal of postprandial TGs. Genes regulating the expression of the major players in this metabolic cascade, such as LPL, CETP, and HL, may also modulate the expression of the small, dense LDL[37]. Irrespective of the source, small, dense LDL particles have a prolonged retention time in plasma, are more susceptible to oxidation because of decreased interaction with the LDLR, and enter the arterial wall more easily, where they are retained more readily.

2.1.2 Glucose metabolism and coronary artery disease manifestation

Glucose is the primary source of energy required for normal organ function. Its metabolism involves the processing of simple sugars in foods to be utilized to produce energy in the form of adenosine triphosphate (ATP). Once consumed, glucose is absorbed by the intestines and into the blood. Extra glucose is stored in the muscle and liver as glycogen, which is hydrolyzed to glucose through gluconeogenesis and released into the bloodstream when needed. While tissues such as the brain and red blood cells can only utilize glucose, others can also use fats. Glucose can also be produced from non-carbohydrate precursors, such as pyruvate, amino acids and glycerol. Insulin and glucagon work synergistically to keep blood glucose concentrations normal. Insulin is produced in the pancreas, where its secretion is increased by elevated glucose concentrations, gastrointestinal hormones and beta(β)-adrenergic stimulation and can be inhibited by catecholamines and somatostatin. Accordingly, an elevated blood glucose concentration results in the secretion of insulin and glucose is transported into body cells, while glucagon has an opposite effect. Insulin exerts several functions that may influence the regulation of coronary vessel activity. Poor glucose metabolism as observed in insulin resistance is a primary cause for T2DM. Insulin resistance presents the impaired ability of either endogenous or exogenous insulin to lower blood glucose. It is typically characterized by decreased sensitivity and/or responsiveness to metabolic actions of insulin constituting a central feature of T2DM, obesity and dyslipidaemia, as well as a prominent component of hypertension and atherosclerosis that

are all characterized by endothelial dysfunction. In some insulin-resistant individuals, insulin secretion will begin to deteriorate under chronic stress (glucose toxicity) and overt diabetes will result. If not, individuals will remain hyperinsulinemic, with perhaps some degree of glucose intolerance, together with other hallmarks of the insulin resistance syndrome (IRS). Increased free fatty acids and lipid accumulation in certain organs are mediators of insulin resistance[38].

The insulin resistance syndrome (syndrome X, metabolic syndrome) is a multifactorial disorder with obesity, dyslipidaemia, atherosclerosis, hypertension, and T2DM acting together to shorten life spans[23]. While a growing number of single genetic diseases affecting energy metabolism have been found to produce the clinical phenotype, strong familial occurrences, especially in racially prone groups together with modern genetic approaches are beginning to unravel the polygenetic nature of the syndrome. However, the strong lifestyle factors of excessive carbohydrate and fat consumption and lack of exercise are important keys to its phenotypic expression. The natural history includes small foetal gestational age birth weight, excessive weight gains during childhood, premature puberty, an allergic diathesis, acanthosis nigricans, striate compounded by gynecomastia, hypertriglyceridaemia, hepatic steatosis, premature atherosclerosis, hypertension, polycystic ovarian syndrome, and focal glomerulonephritis appearing increasingly through adolescence into adulthood. T2DM, which develops because of an inherent and/or an acquired failure of an insulin compensatory response, is increasingly seen from early puberty onward, as is atheromatous disease leading to CAD and stroke[23]. A number of physiological alterations of glucose metabolism including hepatic overproduction of glucose, and reduced glucose utilization by peripheral tissues as a result of insulin resistance contribute to the development of the metabolic manifestations of this disease. Ultimately, pancreatic failure and reduced insulin secretion lead to hyperglycemia and the diabetic state.

Inherited metabolic disorders contribute importantly to adverse cardiovascular outcomes and affect all tissue types[39]. Primary defects in energy balance that produce obesity (and visceral adiposity in particular) are sufficient to drive all aspects of the metabolic syndrome. Common obesity is polygenic, involving complex gene-gene and gene-environment interactions, which explain the multi-factorial obese phenotypes. Obesity also leads to a proinflammatory and prothrombotic state that potentiates atherosclerosis. Fat-derived adipokines, including tumor necrosis factor-alpha (TNF-α) and adiponectin have also been recently implicated as pathogenic contributors or protective factors. Distinct MAPK-dependent insulin-signaling pathways (largely unrelated to metabolic actions of insulin) regulate secretion of the vasoconstrictor endothelin-1 from endothelium. Endothelial dysfunction is often characterized by pathway-specific impairment in phosphatidylinositol 3-kinase (PI3K)-dependent signaling that contributes to a reciprocal relationship between insulin resistance and endothelial dysfunction. Thus, for example, PI3K-dependent insulin-signaling pathways regulating endothelial production of nitric oxide share parallels with metabolic insulin-signaling pathways. These and other cardiovascular actions of insulin contribute to coupling metabolic and hemodynamic homeostasis under healthy conditions.

2.1.3 Blood pressure homeostasis and coronary heart disease manifestation

Blood pressure is the pressure exerted by circulating blood on vessel walls and presents one of the principal vital signs of life. In general, blood pressure refers to arterial pressure of the

systemic circulation. It can be influenced by various factors including the heart pumping rate, blood volume, vascular resistance and blood viscosity[40]. Although the endogenous regulation of arterial blood pressure is not yet completely understood, a number of mechanisms have been defined. These include the baroreceptor reflex, the renin-angiotensin (RAS) system and aldosterone release. Hence, the renin-angiotensin-aldosterone system (RAAS) constitutes a hormonal system primarily responsible for regulating blood pressure and fluid balance. Low blood volume induces secretion of renin in the juxtaglomerular cells in the kidneys. Renin in turn stimulates the production of angiotensin I from angiotensinogen, which is converted to angiotensin II (Ang II) by the ACE. Ang II induces vasoconstriction of the vessels resulting in increased blood pressure. It also stimulates the secretion of aldosterone from the adrenal cortex, which mediates an increase in the reabsorption of sodium and water from the kidney tubules into the blood. This in turn increases the fluid volume in the body and therefore the blood pressure. Increased RAAS activity leads to high blood pressure, and therefore to hypertension.

Hypertension is a vascular disease that may influence the viability of blood vessels. The disease is inheritable (primary) or may underlie environmental causes such as high salt consumption level. Evidence also suggests that Ang II plays an important role in the regulation of structure and function of both the heart and vessel wall. Under pathologic conditions, the ability of the heart and vessel wall to generate Ang II is increased, because of increased ACE expression. Hypertension is thought to correlate with impaired glucose tolerance. However, although the mechanism is not yet fully understood, this has been partly attributed to rapid nutrition and lifestyle transitions contributing to acceleration of obesity-related non-communicable diseases in some ethnic populations[41]. These include body phenotypes, such as high body fat, high truncal, subcutaneous and intra-abdominal fat, and low muscle mass, biochemical parameters such as hyperinsulinemia, hyperglycemia, dyslipidaemia, hyperleptinemia, low levels of adiponectin and high levels of C-reactive protein as well as procoagulant state and endothelial dysfunction[41]. Major atherogenic risk factors are likely mediators in the oxidative modification of DNA, and an increase in oxidative stress may derive from oxidatively damaged mitochondria[42].

2.1.4 Regulation of inflammatory processes and coronary artery disease

Inflammation is part of the complex biological response of vascular tissue to harmful stimuli, such as pathogens, damaged cells and irritants[44]. It is a protective mechanism the organism employs to remove injurious stimuli and to initiate the healing process. Inflammation can be classified as acute or chronic, whereby it presents the initial response of the body to harmful stimuli. This is achieved by the increased movement of plasma and leukocytes (especially granulocytes) from the blood into the injured tissues. A cascade of biochemical events propagates and matures the inflammatory response, involving the local vascular system, the immune system and various cells within the injured tissues. Prolonged inflammation, known as chronic inflammation, leads to a progressive shift in the type of cells present at the site of inflammation and is characterized by simultaneous destruction and healing of the tissue from the inflammatory process. Vascular endothelial dysfunction underlies the genesis and progression of numerous diseases. Among others, it has recently emerged as an early step in the development of atherosclerosis and is mainly characterized by a reduction in the bioavailability of nitric oxide. All of the traditional cardiovascular risk

factors, including dyslipidaemia, arterial hypertension, hyperglycemia and diabetes, are associated with endothelial dysfunction, and oxidized LDLs, the renin-angiotensin axis and insulin resistance play important roles in the pathogenesis of impaired endothelial function. The increased expression of adhesion molecules and pro-inflammatory cytokines leads to abnormal endothelium-dependent vasodilatation. Apart from perturbation in systems regulating vasoconstriction and vasodilatation, ailments such as diabetes mellitus contribute equally to the formation of the plaque in blood vessels. Also, the immune system has recently been implicated in the pathogenesis of endothelial dysfunction and atherosclerosis with a particular regard towards autoimmunity due to the high prevalence of the atherosclerotic process in systemic autoimmune diseases. Dysfunction of HDL and its APOs A-I, A-II, and C-III has been observed in general populations, whereby high concentrations of HDL or Apo A-I in individuals with diabetes or CAD were found to reveal dysfunction in some population-based studies. It has been suggested that dysfunctional HDL particles even become proinflammatory or lose atheroprotective properties. While HDL dysfunctionality has been closely linked to obesity and low-grade inflammation, it appears nonetheless to act partly independently of them[45].

Chemokines are important mediators of angiogenesis, hematopoiesis and leucocyte trafficking. Chemokine Ligand-18 (CCL18)/pulmonary and activation-regulated chemokine (PARC) is a circulating chemokine that plays a role in injury healing, physiological homing of mononuclear blood cells and inflammatory responses. CCL18/PARC is also expressed in atherosclerotic plaques, and its levels have been associated with decreased cardiac function, decreased exercise capacity and increased inflammatory parameters including interleukin-6 (IL-6) and high sensitive C-reactive protein[46]. More importantly high CCL18/PARC levels appear to present an independent predictor of future cardiovascular events[46]

2.1.5 Environment and coronary artery disease management

Like all other risk factors of CAD, the type of food one consumes and style of life an individual leads, are equally important components for pathways leading to the disease, as they affect directly some of the risk factors, such as increased circulating Chol and predisposition to obesity. This is a chronic disease with a multifactorial etiology including genetics, environment, metabolism, lifestyle, and behavioral components which have been shown to contribute to the development of obesity[45]. Elevated body mass index, particularly caused by abdominal or upper-body obesity, has been associated with a number of diseases and metabolic abnormalities, many of which have high morbidity and mortality. These include hyperinsulinemia, insulin resistance, T2DM, hypertension, dyslipidaemia, CAD, and certain other malignancies. Besides, it appears that sedentary subjects with upper-body and visceral obesity who have the metabolic syndrome tend to be at higher risk for hypertriglyceridaemia in response to high-sucrose and high-carbohydrate diets[47]. Thereby, moderate weight loss mitigates the effect. Hyperinsulinemia or insulin resistance may play a role in promoting higher rates of VLDL synthesis and hypertriglyceridaemia in obesity, but the mechanisms remain unclear[47]. Some of these effects may be gender-related. For example, high dietary glycemic load is associated with higher serum triacylglycerol concentrations and greater risk of CAD in women[47], and cigarette smoking in overweight women with low-grade inflammation appears to offer limited protection against cardiometabolic risk[45].

Postprandial lipemia is traditionally defined by the extent and duration of the increase in plasma TGs in response to a fat-enriched meal. Alimentary lipemia has been associated with CAD, in a fashion that is influenced by both genetic and environmental factors. Disease processes, such as dyslipidaemia, hypertension, glucose and insulin metabolism, lifestyle habits, such as eating and exercise patterns, as well as socioeconomic status aggregate in families with CAD. The degree of risk associated with a family history varies with the degree of relationship and the age at onset of disease. Postprandial lipoprotein metabolism is modulated by background dietary pattern as well as meal composition including fat amount and type, carbohydrate, protein, fibre, alcohol, and several lifestyle conditions such as physical activity, tobacco use, physiological factors including age, gender, menopausal status and pathological conditions such as obesity, insulin resistance, diabetes mellitus. Overall, the variability in postprandial response is important and complex, and the interactions between nutrients, dietary or meal compositions and gene variants need further investigation. Furthermore, extremely low fat dietary habits or major gene interactions may influence the lipid profile and the excess cardiovascular mortality observed in heterozygous FH, whereas minor gene determinants do not seem to play any significant role. All other factors put into consideration, the prevalence of the disease may vary among populations, due to differences in the awareness of the disease in a given population.

2.2 Early onset coronary artery disease

Acute coronary syndrome (ACS) is a manifestation of myocardial infarction (MI) at young age, usually described as being under 45 years of age. It is usually associated with coronary thrombosis, and chest pain can also be precipitated by anemia, bradycardias or tachycardias. AMI represents the main thrombotic complication of CAD, with a presence of approximately 9% of the new events of MI occurring in patients. The condition is produced by development of a thrombus at the site of an atherosclerotic plaque that initiates abrupt arterial occlusion, with ischemia and cell death. Evidence of significant disease at coronary angiography suggests the presence of a premature atherosclerotic process. Based on the appearance of the electrocardiogram (ECG), they are also defined as non-ST elevation myocardial infarction (NSTEMI) and ST segment elevation myocardial infarction (STEMI) or unstable angina[48], whereby STEMI constitutes about 30%, NSTEMI 25% and unstable angina 38% of the affected individuals. Unstable angina occurs suddenly often at rest with minimal exertion, or at lesser degrees of exertion than a previous angina. MI at a young age is commonly characterized by evidence of multiple cardiovascular risk factors and by a favorable prognosis in short- and medium-term follow-up. Thus, AMI reflects a degree of damage to the coronaries by atherosclerosis. Apparently, the type of ischemic event shows gender-specific differences. Thus, in women it appears to present more frequently with unstable angina and NSTEMI, whereas men will have ACS with STEMI[48-50]. However, it is still questionable whether this gender association has any genetic background to it.

Several modifiable factors such as hypertension, diabetes, smoking, obesity, and hypercholesterolemia are involved in AMI manifestation. However, in a large number of patients with AMI, modifiable risk factors are not present. Furthermore, apart from early onset of CAD, recurrent coronary events in the young also present an important problem. Recently, the role of Lp(a), homocysteine, inflammation and infection as prime culprits in the pathogenesis of CAD has become a subject of intense research and debate. Specifically, hyperhomocysteinemia is a risk factor of recurrent coronary event in the young and an

element of gene-environment interaction may also contribute significantly to its manifestation. Monogenic disorders are seen in approximately 5% of premature CAD cases, and this prevalence is higher in populations with the founder effect. Family history of MI is positively associated with the risk of early MI in women. While the association with parental history of MI is mediated through the clustering of other common risk factors, the association of sibling history of MI with early-onset MI in young women is only partially explained by the clustering of established and newly-identified risk factors[51].

Heredity has a significantly great share in AMI. Particularly, familial lipidaemic disorders play a pivotal role in early onset of CAD. Familial lipoprotein disorders are seen frequently in subjects with premature CAD. In FH, early CAD is a complex trait that results from a large monogenic component of susceptibility due to elevated LDL-C even in the absence of any other risk factors. Increased LDL and decreased HDL-C predict premature CAD, as do elevated levels of Apo B or reduced levels of Apo A. The metabolic abnormality in many FH-affected individuals is overproduction of Apo B-containing lipoproteins causing elevated levels of plasma Chol, TGs, or both. Low levels of HDL-C and an abundance of dense LDL particles are other features contributing to the high association of this disorder with premature CAD. Although for most children the process of atherosclerosis is subclinical, dramatically accelerated atherosclerosis occurs in some pediatric disease states, with clinical coronary events occurring in childhood and very early adult life. A striking example is found in children who have the homozygous form of FH with extremely high levels of LDL-C, whereby severe atherosclerosis and CAD often develop during the first decades of life.

Among patients with CAD onset before the age of 55, about 5% of cases are attributable to HFH, a monogenic disorder that affects about 1 in 500 people, with a higher prevalence in certain subpopulations. The disease manifests with severe hypercholesterolemia since birth (Chol levels >5-6 the upper normal limit), which, if untreated, leads to early onset accelerated atherosclerosis and premature coronary death, usually before the second or third decades of life. It occurs primarily as an autosomal dominant disorder with a gene-dosage effect. The clinical phenotype of HFH is characterized by increased plasma levels of total Chol and LDL-C, tendinous xanthomata, and premature symptoms of CAD. The disease is characterized by Chol deposits affecting the corneas, eyelids and extensive tendons, elevated plasma concentrations of LDL-C and accelerated vascular disease, especially CAD. It is inherited as an autosomal dominant disorder with homozygotes having a more severe phenotype than do heterozygotes. Dyslipidaemic states associated with premature atherosclerotic disease and high cardiovascular risk are characterized by a disequilibrium related to an excess of circulating concentrations of atherogenic lipoproteins relative to those of atheroprotective HDL, thereby favouring arterial Chol deposition and enhanced atherogenesis. In such states, CETP activity is elevated and contributes significantly to the Chol burden in atherogenic Apo B-containing lipoproteins[6,7].

3. The genetic basis of coronary artery disease

Variation in human genes is a result of structural alteration(s) in the sequence of the gene due to a modification in any one of the DNA bases, producing different forms of the gene known as polymorphisms. These polymorphic changes may or may not lead to changes in amino acids, consequently constituting an alteration in the functional expression of the

resultant protein. The types of gene variants are defined as missence, silent, nonsense or Frameshift mutations, depending on the resultant effect of the changes on the protein product. These sequence variations are classified as large-scale or small-scale mutations, and may influence any type of protein, including structural, signaling, or enzymes engaged in various functions. Large-scale mutations usually lead to a gain or loss of chromosomal regions or translocation of parts of a chromosome, whereas small-scale mutations are nucleotide-based substitutions, deletions or insertions leading to new gene products, as well as gene, protein and phenotypic expression. One of the most important findings in recent times is the recognition of the fact that sequence variations in the human genes are confined to single nucleotide polymorphism (SNPs). The number of SNPs varies within one gene from one population to another. It is also thought that more than 50% of all coding SNPs produce a predictable change in the protein sequence. This fact points to a high level of human protein diversity, with potentially great impact on disease manifestation and modes by which patients respond to drug therapy.

Sequence variations implicated in complex diseases such as CAD are often found in genes regulating diverse mechanisms (e.g. lipid metabolism, glucose handling, blood pressure regulation, or vascular contractility). Indeed, virtually all risk factors for the disease themselves underlie genetic background. Besides, many of these risk factors seem to share some genomic regions and/or gene variants among themselves as well as with CAD, pointing to common loci and possible gene-gene interactions as a mechanism for such complex diseases. An added intricacy to the situation with complex diseases, such as CAD, is the fact that changes in various genes may be related to clusters of dysfunctions that also contribute to the disease, rendering it oft difficult to decipher exactly which factors may be the primary or secondary culprits. Hence, ultimately manifestation of CAD is a product of interaction of environmental factors with alteration in genes that regulate numerous pathways. This section summarizes some of the important genes engaged in the different pathways leading to its manifestation. The catalogue of gene discussed is far from being exhaustive, but simply intended to reflect on some of the established pathways, as well as findings that have recently stimulated further search into their relevance for CAD.

3.1 Pathways regulating lipid metabolism

Understandably, alterations in genes regulating circulation Chol levels and its metabolites should occupy a key position among the disease pathways leading to CAD manifestation. As such, lipidaemic disorders leading to an increase in circulating Chol could theoretically result from mutations in genes involved in any of the various pathways regulating its homeostasis. Hence, perhaps by far the most well-studied risk pathways for CAD are those leading to FH, an autosomal defect related to mutations in the LDLR, the Apo B and the PCSK9 genes. However, mutations in several other genes, such as LDLR adapter protein 1 (LDLRAP1) Apo A-I, Apo A-IV, Apo A-V, Apo C-III, can also lead to a phenotype similar to the disease. In humans, *LDLR* mutations induce hypercholesterolemia and, subsequently, Chol deposition to a variable degree, not only confirming the pathogenetic role of *LDLR* but also highlighting the existence of additional factors in determining the phenotype. Apparently, receptor-negative mutations result in a more severe phenotype than do receptor-defective mutations[52]. At least five specific monogenic disorders, are known to be related to this phenomenon. These are familial hypercholesterolemia, familial ligand-

defective Apo B, autosomal recessive hypercholesterolemia, sitosterolemia and cholesterol 7-alpha-hydroxylase deficiency. The diseases may occur in the form of a Mendelian (caused by a defect in a single gene) or as a complex trait (resulting from an interaction of genetic predisposition and environmental factors). Several loci including LDLR-associated protein, PCSK9 and ATP-binding cassette transporters ABCG5 and ABCG8 have been identified for monogenic hypercholesterolemia. Functionally, ABCG5 and 8 pump sterols out of the hepatic and intestinal cells into bile and intestinal lumen, respectively. A number of PCSK9 variants have been identified, some of which are gain-of-function mutations causing hypercholesterolemia by a reduction of LDLR levels, while loss-of-function variants have been associated with a reduction of LDL-C levels and a decreased risk of CAD[53]. Furthermore, at least nine of the apolipoproteins known to play a major role in lipid metabolism have been implicated in influencing plasma levels of HDL, VLDL, LDL chylomicrons or TGs[54]. Mutations in the Apo B gene can result in a phenotype that is clinically indistinguishable from FH, and have also been shown to be associated with CAD. Although many studies indicate that the FH phenotype is influenced by other genetic and environmental factors, it remains unclear whether or not these are synergistic interactions or simply additive effects[52]. Furthermore, although HFH is genetically heterogeneous, it is most often caused by heterozygous mutations in the gene encoding the LDLR.

Another form of FH, the ARH has been associated with the insertion mutation in a phosphotyrosine binding domain of the so-called ARH protein[55], encoding a cellular adaptor protein required for LDL transport, and consequently for internalization of LDLs in the liver. This protein is thought to function as an adaptor protein that couples LDLR to the endocytotic machinery[36]. The phenotypic expression of this homozygous FH appears to be dominated by the consequences of the *LDLR* gene mutations. An autosomal recessive form of FH caused by loss-of-function mutations in *LDLRAP1a* has also been reported. In heterozygous FH, however, the underlying mutational *LDLR* type determines only to a much lesser extent, if any, the variable phenotypic expression. Conversely, some *PCSK9* loss-of-function mutations resulting in low levels of LDL-C appear to protect against CAD[56 57]. However, despite compelling evidence indicating that PCSK9 impairs the LDLR pathway, its role in Chol metabolism remains incompletely defined. Additional atherogenic risk factors of environmental, metabolic, and genetic origin are presumed to influence the clinical phenotype in FH. Other risk factors include the *LPL* gene which is thought to present a risk factor for dyslipidaemia, characterized by hypertriglyceridaemia and low HDL-C levels which increases with age and weight gain, and is associated with CAD and T2DM.

The HDL-C levels are under considerable genetic control with heritability estimates of up to 80%, and low HDL-C syndromes have generally been correlated with an increased risk of CAD. The cardioprotective effects of HDL-C have been attributed to its role in reverse Chol transport, its effects on endothelial cells, and its antioxidant activity[58]. On the other hand, relative HDL-C deficiency states have been associated with the ATP binding cassette protein (ABCA1), Apo A1 and lecithin cholesteryl acyl transferase defects. However, although numerous candidate genes contribute to the low HDL-C phenotype, their impact on CAD is heterogeneous, single gene abnormalities responsible for HDL-C deficiency states may have variable effects on atherothrombotic risk, reflective of diverse gene-gene interactions and gene-environmental relationships. Other potentially important candidates involved in low HDL-C syndromes in humans include *Apo C3*, *LP*L, sphingomyelin phosphodiesterase 1,

and glucocerebrosidase. Molecular variation in ABCAI and Apo AI and partly lecithin cholesteryl acyl transferase deficiency have been associated with increased CAD, while some of the *Apo A1* variants, such as Apo A1 Milano and Apo A1 Paris have been associated with reduced risk. Endothelial lipase gene polymorphisms have also been associated with HDL-C concentrations[59]. This lipase participates in HDL metabolism by promoting the turnover of HDL components and increasing the catabolism of Apo A-I[59], which distinguishes itself from other TG lipases in showing the highest activity on HDL[59]. Similarly, mutations in the *CETP* gene associated with CETP deficiency are characterized by high HDL-C levels and reduced cardiovascular risk.

In contrast to FH, which is caused by mutations in a number of affected genes, the genetics of FCH have remained obscure and very few definite candidate genes have been identified thus far. Of these, the strongest evidence links the lipid components of FCH to intronic variants in the upstream transcription factor 1 (*USF1)* gene on chromosome 1q21-23. Modifying genes, particularly those that influence the high TG trait, include apo C3 and apolipoprotein A5 (*apo A5*). *Apo A5* is a member of the apolipoprotein *Apo A1/C3/A4* gene cluster, and is important in regulating plasma TG levels. It also represents a downstream target of USF1, implicating a USF1-dependent pathway in the molecular pathogenesis of dyslipidemias[60]. However, the relationship of *USF1* polymorphisms with diabetes and the metabolic syndrome, which co-localize to this region and are also associated with mixed hyperlipidaemia, has yet to be defined. Additionally, Apo AV is an activator of LPL, linked to familial hypertriglyceridaemia and FCH to upstream stimulatory factor 1[61]. A case study of a patient with αβ-lipoproteinaemia suggested that the hypertriglyceridaemia seen in patients with FCH may result from an abnormality in microsomal TG transport protein function. A direct relationship between the post-prandial TG levels and LDL-C levels in the fasting state of patients with FCH and sporadic hypercholesterolaemia has also been postulated, which increases considerably when all the MI survivors are considered independent of age. The *USF1* has been linked with familial combined hyperlipidaemia in some ethnic groups. Currently, genetic and functional evidence is supportive of a role for the *USF1* in the etiology of FCP and its component traits, although the mechanism still remains largely unknown[62]. While the genetic defect remains unknown, linkage to the region of the Apo *AI- CIII- AIV* gene cluster on chromosome 11 has been implicated.

Other mutations, especially the truncation-causing mutations in Apo *B* and MTP can cause familial hypobetalipoproteinemia, characterized by hypercholesterolemia and resistance to atherosclerosis[63]. Therefore, in genetic dyslipidaemia elevated Apo B levels and reduced Apo A levels (or increased Apo B/AI ratio) differ and predict premature CAD[64]. One of the most extensively studied lipoprotein genes in this respect is the *apo E*, which has been linked with various forms of lipidaemic disorders. An example of an *apo E*-mediated, autosomal recessive, lipid disorder is familial dysbetalipoproteinemia (FD). The gene exists in three allelic forms (epsilon2 (ε2), ε3 and ε4 coding for isoforms E2, E3, and E4 and having different binding affinities for the apo E receptors. This genetic variation is associated with different plasma lipoprotein levels, different response to diet and lipid-lowering therapy, and a variable risk for cardiovascular disease and Alzheimer's disease. While the ε2 allele is associated with elevated TG levels, the ε4 allele is associated with increased Chol levels. The ε2/ε2 genotype is thought to present the most common cause for hyperlipoproteinemia type III. However, although several studies support the role of *apo E* polymorphism in CAD

either directly or indirectly via its influence on lipid and lipoprotein levels, there are some studies which show lack of such relationships.

3.2 Pathways regulating blood pressure

Given that hypertension, diabetes, dyslipidaemia, and obesity exhibit a substantial heritable component, it is to be expected that important genes encoding proteins related to these pathways may predispose individuals to this cluster of cardiovascular risk factors. Specifically, as the core regulator of systemic blood pressure, and being involved in the cardiovascular homeostasis, several polymorphisms in genes of the RAAS system have been found to have pleiotropic effects on cardiovascular disorders. A constituent of this system is angiotensinogen (*AGT*), a serum renin substrate glycoprotein synthesized in the liver, which is released as the precursor of angiotensin, the hormone that forms part of the systemic blood pressure regulatory system. Risk variants in this gene have been identified that lead not only to hypertension[65,66], but also to CAD[67-70]. Apart from the *AGT*, several other genes engaged in regulating components of the RAAS pathway have also been identified as constituting risk for primary hypertension. These include among others, the alpha(α-) adducin (*ADD1)*[71], *ACE*[72-75], nitric oxide synthase[76], metafolin folate (methyl tetrahydrofolate; MTHF) receptor (*MTFHR)*[77] and thrombospondin (*THBS)* genes[78,79]. The *THBS*s are a 5-member gene family that mediates cell-cell and cell-matrix interactions. These proteins are either trimers or pentamers, and their functions depend on their abilities to interact with numerous extracellular ligands and cell surface receptors through the multiple domains that compose each subunit. Thus, polymorphisms in 3 *THBS* genes encoding *THBS1*, *THBS2*, and *THBS4* were proposed to modulate the risk of premature CAD[78,79] and MI[80-82]. Interestingly, literally all of these genes have also been implicated in CAD manifestation. However, the important question remains as to whether their role in CAD is by virtue of their influence on the blood pressure control or some other signaling links may exist among the pathways involved in the disease process.

3.3 Pathways regulation glucose metabolism

The impact of T2DM as a primary cause for CAD has now become common knowledge. In fact, diabetic patients are thought to have a 3-fold higher risk of developing atherosclerosis and its clinical complications as compared to non-diabetic individuals. Part of the cardiovascular risk associated with this disease is probably due to genetic determinants influencing both glucose homeostasis and the development of atherosclerosis. Besides, T2DM frequently coexists with other cardiovascular risk factors like arterial hypertension, central obesity and dyslipidaemia in the manifestation of CAD. Several genes including *ADIPOQ* and peroxisome proliferator-activated receptor (*PPAR*) genes have been implicated in manifestation of metabolic syndrome (MS)[83,84]. Adiponectin (*ADIPOQ*) is an adipocyte-derived hormone and an essential modulator of insulin sensitivity. It is abundantly secreted by adipocytes, plays important roles in lipid and glucose metabolisms, and has direct anti-inflammatory and anti-atherogenic effects. Since it modulates several metabolic processes, it is thought to play an important role in the suppression of the metabolic derangements that may lead to MS. It may serve as an important biomarker for metabolic syndrome, and some of its common polymorphisms have been associated with the phenotypes related to body weight, glucose metabolism,

insulin sensitivity, and risk of T2DM and CAD[85]. While circulating ADIPOQ is involved in the atherosclerotic process and has been associated with cardiovascular disease as well as obesity, insulin resistance, metabolic syndrome, and T2DM[83,84,86,87], its relationship with the early onset of atherosclerosis in hyperlipidaemia is still not completely understood.

The PPAR-α, -γ, -β are members of the nuclear receptor superfamily of ligand-activated transcription factors that have central roles in the storage and catabolism of fatty acids. All three PPARs are activated by naturally occurring fatty acids and fatty acid metabolites, indicating that they function as the fatty acid sensors of the body. Hence, they constitute major regulators of energy homeostasis. They not only control lipid metabolism, but also regulate vascular diseases, such as atherosclerosis and hypertension, as well as the expression of genes involved in fatty acid β-oxidation. Specifically, PPAR-γ plays a critical role in adipocyte differentiation and serves as the receptor for the glitazone class of insulin-sensitizing drugs used in the treatment of T2DM. *PPARβ* polymorphisms are associated with plasma lipid levels, body mass index and the risk for diabetes and CAD[88]. *PPARγ2* variants have also been shown to influence insulin sensitivity in interaction with *ADIPOQ* or to influence plasma ADIPOQ levels. It has recently been suggested that PPARs may present a functional link between obesity, hypertension and diabetes. However, in contrast to PPARα and PPARγ, relatively little is known about the biology of PPARβ, although recent findings suggest that this subtype has a role in lipid homeostasis also.

Candidate gene variants for polygenic obesity appear to disrupt pathways involved in the regulation of energy intake and expenditure and include adrenergic receptors, uncoupling proteins, PPARG, POMC, MC4R and a set of variants in the FTO locus. Notably, the FTO gene is the most robust gene for common obesity characterized to date, and recent data shows that the FTO locus seems to confer risk for obesity through increasing energy intake and reduced satiety. Gene variants involved in pathways regulating addiction and reward behaviours may also play a role in predispositioning to obesity[89]. Other obesity-related genes including *ADIPOQ*, leptin receptor, and fat mass obesity-related genes have also been described with respect to manifestation of metabolic syndrome and CAD/MI[90]. While genes, such as the *PPARs*[91] have been discussed as independent risk for CAD[88], several of them may share common variants for metabolic syndrome with CAD/MI, among themselves and/or with others, including *Ang II*[92], AMP deaminase-1[93] and *ACE*[91] genes. For example, the risk of acquiring CAD in patients with T2DM appears to increase significantly in the presence of the several gene variants such as those of the transcription factor 7-like 2 (*TCF7L2*)[94] and E-selectin genes[95]. Other genes associated with CAD include the *C5L2,* a stimulator of TG synthesis or glucose transport and constitutes a functional receptor of acylation-stimulating protein[96]. Together, these data only reveal the complexity of mechanism involved in disease pathways regulating CAD and its risk factors.

A rare genetic form of insulin resistance is Dunnigan-type familial partial lipodystrophy (FPLD), which is characterized by loss of subcutaneous fat from extremities, trunk, and gluteal region. It is always accompanied by insulin resistance and hyperinsulinemia, often with hypertension, dyslipidaemia, T2DM and early endpoints of atherosclerosis. FPLD is thought to result from mutated *LMNA*, which encodes nuclear lamins A and C. Familial studies indicate that dyslipidaemia precedes the plasma glucose abnormalities in FPLD subjects with mutant *LMNA*, and that the hyperinsulinemia is present early in the course of

the disease. Plasma leptin is also markedly reduced in subjects with FPLD due to mutant *LMNA*. Thus, a defect in the structure and function of the nuclear envelope can result in a phenotype that shares many aspects with the common syndrome of insulin resistance.

3.4 Pathways regulation smooth muscle cellular viability and function

Since inflammatory mechanisms play a central role in mediating all phases of atherosclerosis, genes encoding for inflammatory or anti-inflammatory molecules are candidates for the risk of developing atherosclerosis. Genetic variability affecting many functional areas such as lipid and energy metabolisms, hypertension and haemodynamic mechanisms, blood clotting homeostasis, inflammation, and matrix turnover in the vascular wall will have an impact on the development of macrovascular complications in diabetic patients. During atherogenesis intercellular communication via gap junctions as well as cell membrane channels linking the cytoplasmic compartments of adjacent cells play a critical role. The component protein subunits of these channels, called connexin (Cx), belong to a multigene family. Cx37 is involved in growth, regeneration after injury and ageing of the endothelial cells, pointing to a potential role in atherosclerosis. A variant of the Cx37 gene has been associated with thickening of the carotid intima and therefore CAD[97]. Recently a higher prevalence of pro-inflammatory polymorphisms of several genes including pyrin, epithelial cell adhesion molecule (*EPCAM)*, *Cx37* and *PCR* genes and a lower prevalence of anti-inflammatory polymorphisms, such as the *TLR4, IL10* and *CCR5Δ32*) has been documented[98,99], suggesting early MI could be associated with a genetic predisposition to an intense inflammatory response, linked also to an hyperviscosity syndrome[98,99]. Intima media thickness studies have provided evidence that hypoalphalipoproteinemia due to mutations in apo *A-I*, *ABCA1*, and *LCAT* is associated with increased progression of atherosclerosis[100]. In contrast, hyperalphalipoproteinemia as a result of loss of CETP function is associated with unaltered atherosclerosis progression.

The generation of oxidative stress is believed to be an important cause of DNA damage in atherosclerosis. Under oxidative stress, both blood monocytes and plasma lipoproteins invade the arterial wall, leading to their exposure to atherogenic modifications[101,102]. Hence, increased oxidative stress has been implicated in the pathogenesis of the atherothrombotic process. A recent addition to the list of genes thought to be involved in oxidative stress are the paraoxonases (*PONs*), a family of closely related antioxidants encoded by at least three clustered genes (*PON1,* PON2 and PON3) on chromosome 7q. The human PON1 is an HDL-associated arylesterase that hydrolyzes paraoxon[103]. It is thought to exert an antiatherogenic effect by protecting LDLs against oxidation[104,105] partly by slowing down the accumulation of lipid peroxides in LDLs under oxidizing conditions. Understandably therefore, alterations in the *PON1* gene are implicated in dyslipidaemic disorders. Furthermore, free radical production in the immediate postrecanalization phase after thrombotic occlusion of a major coronary artery in humans is associated with MI[106]. The NADPH oxidase system as a main source of reactive oxygen species in vascular cells has been implicated in development and progression of CAD. In contrast, some gene variants, such as c.-930A>G in the promoter region of p22-PHOX gene have been shown to have protective effects[107].

Apart from inflammatory processes, alterations in genes regulating blood cell formation (hematopoiesis) and blood coagulation mechanisms constitute important cell function-related risk for CAD. The fraction of immature platelets is increased in ACS, especially in

the acute phase of STEMI[108]. Immature platelets with an increased hemostatic potential may contribute to coronary thrombus formation. Recently a quantitative trait locus 12q24 associated with mean platelet volume and platelet count has been implicated as a risk locus for CAD[109]. Polymorphisms in genes regulating various aspects of hematopoiesis and blood coagulation processes including leptin[110], the GATA transcription factor *GATA-2* have also been implicated in CAD. GATA-2 is an important regulator of hematopoiesis. Among others, glycoprotein growth factors regulate the proliferation and maturation of the cells that enter the blood from the marrow, as well as causing cells in one or more committed cell lines to proliferate and mature. Other recently added culprits for CAD include the lipase A, lysosomal acid, cholesterol esterase (*LIP*A), platelet-derived growth factor (*PDGF*), *ADAMTS7-MORF4L1*, and KIAA1462[111] genes.

3.5 Gene and mRNA regulatory pathways

One of the most exciting findings in recent times is the discovery of several loci in DNA regions that regulate transcription rather than the coding regions for protein as risk sequences for disease. As such, several transcription factors (TFs) have been associated with CAD/MI, some of which have been discussed above. These include the *GATA2*[112], myocyte-specific enhancer factor 2A (*MEF2A)*[113-115], proteasome subunit alpha 6 (*PSMA6)*[116,117], transcription factor 7-like 2 (*TCF7L2*) genes, among others. GATA-2 constitutes a target gene ensemble consisting of genes encoding key determinants of endothelial cell identity and inflammation. These sites characteristically contained motifs that bind activator protein-1 (AP-1), a pivotal regulator of inflammatory genes[118]. It plays an essential role in the establishment and maintenance of adult hematopoiesis, and is present in hematopoietic stem cells, as well as cells that make up the aortic vasculature, such as the aortic endothelial and smooth muscle cells. Several of the *GATA2* gene variants have been associated with familial early onset of CAD[112]. The *MEF2* is a member of the MADS gene family (name for the yeast mating type-specific transcription factor MCM1, the plant homeotic genes 'agamous' and 'deficiens' and the human serum response factor SRF, a family that also includes several homeotic genes and other transcription factors, all of which share a conserved DNA-binding domain). This proteasome is a multicatalytic proteinase complex distributed throughout eukaryotic cells at a high concentration, and cleaves peptides in an ATP/ubiquitin-dependent process in a non-lysosomal pathway. An essential function of a modified proteasome, the immunoproteasome, is the processing of class I MHC peptides. Interestingly, a great majority of entities associated with disease reside in the untranslated regions (UTRs), particularly the 3 prime (3′-UTR) of these genes. Thus, for example, the *MEF2A* gene is highly polymorphic and harbours several deletions in the 3′-UTR that are implicated in CAD/MI[113]. The 3′-UTR regions contain sequences involved in gene regulatory and mRNA maturation processes. It also harbours binding sites for micro RNAs, which are involved in transcriptional and protein processing mechanisms. While the exact mechanisms are still to be elucidated, it appears nonetheless that changes in transcription factors, gene regulatory and mRNA maturation mechanisms exert important influence impacting disease pathways of complex diseases, such as CAD. Hence, the speculation that the disease-causing loci are more likely to be in DNA regions that regulate transcription rather than being in coding regions for protein[119]. Besides, novel genes also continue to be uncovered through genome-wide studies which do not belong to the canonical pathways of CAD, lipid metabolism or any of its other risk factors.

3.6 Gene-gene and gene-environmental interaction in manifestation of coronary artery disease

The fact that several pathways commonly lead to CAD points to the possibility of interactions of these risk factors at various levels in the path to disease manifestation. Besides, the exceedingly high prevalence of risk factors such as T2DM, dyslipidaemia and hypertension in CAD individuals raises the question as to which pathway(s) or mechanism may be the primary cause of disease in cases harbouring combinations of such factors. This is compounded by the fact that, for example, the set of metabolic and physiologic risk factors associated with elevated cardiovascular disease risk including hypertriglyceridaemia, low HDL-C levels, hypertension, abdominal obesity, and insulin resistance all contribute to similar ailments, each of which may underlie genetic factors. The expression of each one of the major factors is now known to be the result of complex interactions between genetic and environmental factors. For example, as discussed above, obesity may play a major role in triggering the metabolic syndrome by interacting with variants in candidate genes for dyslipidaemia, hypertension and insulin resistance.

Not surprisingly, in the last decade, a sizeable portion of research interest has focussed on trying to understand possible interactions among individual disease-causing variants or genes on complex disease manifestation. One of the most well studied systems is the RAAS pathway, which has been evaluated with respect to interactions among its risk variants/genes as well as with other genes in triggering CAD. Significant two-way and three-way gene-gene interactions between ACE I/D, AT1R A1166C polymorphisms and AGT gene haplotypes have been associated with CAD in a number of studies[120]. One study has described several synergistic effects between the studied polymorphisms and classical risk factors such as hypertension, obesity, diabetes and dyslipidaemia[121]. Thus, the presence of the DD genotype of ACE I/D (and also ACE11860 GG) increased the odds of developing CAD when related to each one of these classical risk factors, particularly in the male and early onset CAD subgroup analysis. Interactions have also been observed between ACE DD or ACE 8 GG with PON1 192RR in increasing the risk of CAD[122,123]. Concomitant presence of ACE DD and AT1R 1166 CC genotypes has also been reported to synergistically increase the predisposition to diastolic heart failure[126]. Significant interaction between APOE and LPL variants and HDL-C levels was also reported[130], furnishing support to the idea that several polymorphisms in apolipoprotein genes may by themselves and/or in interaction with other polymorphisms contribute to risk for CAD in men[130]. Interaction of the GNB3 825T allele with the ACE D allele was also reported in MI[131].

Apart from the RAAS system, interactions have also been evaluated involving multiple gene variants with respect to CAD manifestation. One such a study linked increased risk of the disease in association with changes in genes belonging to different enzymes compared to the isolated polymorphisms[122]. Another study suggested that several polymorphisms in apolipoprotein genes may by themselves and/or in interaction with for example the *PPARγ* C161-->T variant and apo ε4 genotype influence serum Chol level, in which the impact of the later on CAD was attenuated through the former genotype[125]. Epistatic, high-order, gene-gene interactions between RAS gene polymorphisms and CAD has also been discussed[127]. Similar interaction between variations in the ACE and ATR2 genes has been hypothesized in relation to the extent of coronary atherosclerosis[129].

Other studies have addressed the possible interactions of various gene variants with risk diseases, such as hypertension or T2DM in acquiring CAD. For example, the AGT235 TT increased the CAD risk in the presence of hypertension and dyslipidaemia, while AT1R1166 interacted positively with hypertension, smoking and obesity[121]. An interaction between RAAS predisposing genes and some biochemical/environmental risk factors was also reported in CAD onset, pointing to a significant enhancement of the effects of classical markers especially by ACE I/D and ACE11860[121].

One form of interaction currently attracting great attention is that of the haplotypes versus individual risk variants. Such an interaction would involve nucleotide changes occurring in the same genomic region, either within a single gene or in several genes at the same locus. Generally, it can be acknowledged that in many cases, haplotyping has been found to be more meaningful than individual variants. One study involving methylenetetrahydrofolate reductase C677T, plasminogen activator inhibitor 4G/5G, and endothelial nitric oxide synthase 3-27 base pair repeat polymorphism in patients with early-onset CAD suggested the coexistence of high-risk alleles as augmenting the severity of the disease[128]. These results are a manifestation of the relevance of the concept of multilocus and multi-gene effects in complex diseases, such as CAD.

Besides, a number of genes are linked with changes in environmental factors affecting disease outcome. Thus, variables such as the postprandial lipid response have been shown to be modified by polymorphisms within multiple genes for the apolipoproteins, *LPL*, *HL*, fatty acid binding and transport proteins, *MTP* and scavenger receptor class B type I[132], while several other genes including the a*po A1/C3/A4/A5* cluster, *ABCA1*, *CETP*, human glucokinase regulatory protein (*GCKR*), *HL,* IL-6, *LPL*, lipid-droplet associated protein, perilipin, and *TCF7L2* have all been implicated in the modulation of the postprandial lipid metabolism[133]. All these genetic changes in combination with other disease-causing elements are likely to exacerbate CAD. Furthermore, it has been suggested that the interplay of genetic and environmental factors places first-degree relatives of individuals with premature CAD at greater risk of developing the disease than the general population. However, the data on this subject is still very limited. Besides, reliability of available data on these interactions is not established, since replication studies are still lacking in the literature.

4. Genetics of acute coronary syndrome in the young

Acute myocardial infarction AMI in young individuals presents a typical pattern of risk factors, clinical, angiographic and prognostic characteristics that differ from those related to adult disease manifestation. An important feature of early onset is a positive family history, which has become a recognized cardiovascular risk factor. Cutaneous and tendinous xanthomata develop in childhood and are the most common reason for initial presentation. However, the pathogenic mechanisms are multifarious and complex. The frequency of FH is estimated to be 1 in 500. About 50% of individuals with FH die before the age of 60 due to MI.

Since CAD manifests itself both as an early and late onset event, different risk factors may determine the stages at which the disease becomes apparent. A most useful approach for identifying genes associated with the condition involves linkage studies, which provide

leads through potential genomic loci that can be mined for candidate genes. A wealth of data pointing to several genomic links for ACS at a young age has already been produced in different laboratories. Examples include linkage to the 2q36-q37.3, 3q26-q27 and 20q11-q13[134], for which a number of gene are currently being targetted for further investigation. Among the suspect genes described thus far is the insulin receptor substrate-1 gene, thought to present a locus for combined disease processes of atherosclerosis, plaque instability, and coronary thrombosis[134].

The natural link between familial dyslipidaemic disorders and early onset of CAD has placed genes involved in lipid metabolism at the focus of research interest. These studies have led to the discovery of several polymorphisms in the *Apo B*, *LDLR* and *PCSK9* genes among others. Missense mutations in the LDLR-binding domain of Apo B are believed to cause familial ligand-defective Apo B, characterized by hypercholesterolemia and premature CAD. Heterozygosity of the LDLR is relatively common, and gain-of-function mutations in PCSK9 also cause autosomal dominant hypercholesterolemia, through elevation of plasma Chol concentrations associated with low-density lipoproteins (LDLs). Other gene polymorphisms involved in the increase in LDL include those in the *Apo B*, *Apo E* and *LPL* genes. The *Apo* ε4 has also been linked to an increased risk of AMI as well as independent predictor of adverse events at young age[135]. Familial LPL deficiency is a rare inborn error of metabolism caused by mutational changes within the *LPL* gene, leading to massive hypertriglyceridemia and reduced HDL-C, both of which are risk factors for the development of CAD. Besides, human HL deficiency as a second causative factor for hyperlipidaemia is also strongly associated with premature CAD[43].

Apart from the regulation of lipid metabolism, several other genetic variants have surfaced as risk for AMI in the last decade. Cross-sectional studies endeavouring to identify variants in candidate genes for early development of CAD and AMI have focused primarily on polymorphisms influencing certain biological functions, such as coagulation and fibrinolysis, platelets, vascular function, lipid metabolism and inflammation. Indeed various genes have been identified encoding coagulation proteins, fibrinolytic proteins, platelet receptors, homocysteine metabolizer, and those related to endothelial dysfunction, abnormal blood flow and oxidative stress[136]. Examples include genes encoding the prothrombin, apo E, Factor V Leiden, Factor VII, and transforming growth factor-β1 (*TGF-β1*) genes[137]. Limited and controversial data also exists on the impact of the plasminogen activator inhibitor-1 (*PAI-1*) gene polymorphism in the pathogenesis of AMI. Thereby, some studies suggest that young patients <35 years possessing the 4G allele exhibit higher PAI-1 plasma levels but lower Lp(a) levels compared to 5G/5G homozygotes, indicating that the 4G allele of the *PAI*-1 4G/5G polymorphism is less frequent among survivors of MI at very young age compared with matched controls[138]. It has further been argued that AMI at young age could be also caused by a reduction of the fibrinolytic activity, in the presence of the *PAI* 4G allele.

Because ischemic stroke with arterial occlusion or undetermined etiology is more likely to be related to a genetic prothrombotic state, polymorphic changes in genes regulating clotting processes have attracted attention as potential culprits of ACS. Thus, the platelet membrane receptor glycoproteins (GPs) are essential for the platelet activation process, and the genetic polymorphisms in the encoding genes may influence the risk of ACS and atherosclerosis. Recently, a metaanalysis implicated platelet glycoprotein IIb/IIa (GPIIb-

IIIa), a membrane receptor for fibrinogen and von Willebrand factor and thrombopoetin variants in AMI[98]. The GP IIIa PI A2 allele has also been strongly associated with a previous history of MI, as well as the severity of disease, while in young smokers, the PAI-1 4G allele appears to present a mild risk factor for the development of MI[139]. Other gene variants including the GPIIb W86R mutation which produces a nonfunctional enzyme associated with elevated TG levels in the affected individual[140], and the prothrombin G20210A variant which displayed a modest but significant risk factor for AMI at young ages[141] have also been associated with the syndrome. An important aspect of the impact of these risk genes on ACS is the influence of the environmental components such as smoking, gender and socioeconomical factors. An example is the suggestion that the impact of the dominant mode for the thrombomodulin -33G/A polymorphism (GA+AA genotype) on a combination of CAD risk factors such as MI, hypertension and T2DM may be related to smoking in young individuals[142 143]. Thrombomodulin is an endothelial cell surface receptor for thrombin, which plays an important role in the regulation of blood coagulation by decreasing thrombin activity and activating protein C. A case study also reported an association of the Apo E (p.Arg136Cys) mutation and obese gene carriers, but with no severe dyslipidaemia in young smokers[144]. Also, synergistic effect of the *MMP-3* 5A/6A variant in the promoter region with smoking on the onset of AMI has been described in young patients with MI[145]. Furthermore, the contribution of the activated protein C resistance (APCR) as an increased risk for AMI has been described specifically in women smokers.

Other studies also point to the metafolin folate (methyl tetrahydrofolate; *MTHF*), *CD14*, E-selectin, *eNOS* and the *PECAM1* genes as potential candidates for ACS[98]. Apart from being implicated in the onset of CAD, the MTHF C677T variant has been associated with the risk for recurrent coronary events. Elevated plasma homocysteine level at admission is considered to be an independent risk factor for these events after the first episode of ACS in young patients, irrespective of the status of this variant[146]. Interestingly, while an increased risk of developing coronary artery lesions was described in the presence of a mannose-binding lectin (MBL2) variant in pediatric patients, in patients older than one year, an increased risk of the lesions was observed in wild-type genotype carriers, leading to the suggestion that MBL has an ambiguous role in Kawasaki disease and contributes differently to the pathophysiologic development CAD[147]. Another gene variant, the annexin V -1C/T seems to have a minor effect in bleeding disorders, but to play a protective role against AMI, reducing the risk of developing the disease[148].

Apart from genes regulating lipid metabolism and coagulation pathways, other gene families of importance in ACS include those encoding growth factors, proteins for proinflammatory processes, dyslipidaemia and obesity-related genes. These include two *TGF-β1* variants that have been linked AMI in young patients[149]. Associations between obesity-related variants, metabolic syndrome and AMI include those of *ADIPO Q*, *LEPR MC4R* and *FTO* polymorphisms. Furthermore, in a couple of case-control studies, Cx37 was implicated in AMI[97]. Also, a case-control study involving young patients with a first event of acute CAD or ischemic stroke, suggested that common variants in the VWF gene were associated with VWF levels and the risk for cardiovascular disease[150]. However, information on the contribution of common VWF gene variants to VWF levels and cardiovascular disease risk is still limited.

Plaque rupture is a well established critical factor in the pathogenesis of AMI. Recently, interest has also focussed on the relationship between inflammation and manifestation of atherosclerosis, leading to the identification of stromelysin-1 (i.e. matrix metalloproteinase-3; MMP-3) as a risk gene. MMP-3 can degrade extracellular matrix and has been identified extensively in human coronary atherosclerotic plaques. It may contribute to the weakening of the cap and subsequent plaque rupture. Also, the aldosterone synthase (*CYP11B2*) -344 C/T gene variant, which may influence plasma aldosterone levels, has been reported to strongly influence left ventricular diameters and mass in young adults and arterial stiffness in essential hypertensives.

However, since CAD is a multifactorial disease, single mutations are likely to provide a small or modest contribution to risk, which may depend on interaction with other genes and/or a particular environment. Besides, there is still some controversy or uncertainties with regards to the role of some individual pathways or gene variants in ACS, such as those encoding some of the obese-related or RAAS genes[90]. For example, a borderline association for AMI with the ACE D/I polymorphism was described as a risk only in the occurrence and the long-term prognosis of AMI at young age in one study[151], leading to the conclusion that polymorphisms in RAAS genes may be important in the onset of a first AMI in young patients, but not in the disease progression after a long follow-up period[151]. Besides, the role of some of the genes/ gene variants has been refuted by a number of studies. This includes the role of Factor V Leiden, where some studies reported protective effects of certain variants initially thought to be causative. A metaanalysis also refuted the association between the PlA1/A2 of the *GP IIIa* gene and young AMI, which had been implicated in a number of studies. The prothrombotic gene polymorphisms did not appear to have a significant influence on the prognosis in young ischemic stroke due to arterial occlusion of undetermined causes in a Taiwanese study[152]. However, these facts only stress the importance of replication of studies especially those that involve small numbers. More importantly, they reveal the importance of environmental factors especially for complex diseases such as CAD. Furthermore, this scenario points to the fact that not everything is known yet about the gene/gene variant responsible for CAD and its risk factors. Identifying such genes should greatly enhance our understanding of the intricacy of CAD manifestation, to identify individuals at risk and more importantly facilitate the development of more efficacious treatment strategies and introduction of early preventive measures. Thus, research efforts continue to address the identification of acquired and inherited risk factors of this complex disease.

5. Genetics and drug therapy of coronary artery disease

Treatment of coronary artery disease entails among others, lowering of circulating cholesterol, antiplatelet therapy representing the basis of treatment for the short- and long-term prevention of atherothrombotic disease processes, as well as measures to prevent or treat hypertension, heart failure and T2DM. Factors involved in inflammation (cytokines, TNF), proliferation of smooth muscle cells and vasoactivation are also important. In all these incidences however, it is well established that patients often respond differently to drug therapy. For example, in the treatment of lipidaemic disorders, not all patients respond equally well to therapy with statins. There are several reasons why patients respond in different fashions to drug therapy. One major explanation is the mode by which the drugs

are metabolized by the various enzymes. It is now well recognized that genetic variations can contribute to these differences in drug disposition and, consequently, clinical efficacy at the population level. Thus, the metabolism may occur too rapidly, thereby leading to accelerated elimination from the body before it has had enough time to exert its effect. The opposite may equally be true that the drug is broken down too slowly resulting in its accumulation and therefore toxicity. Furthermore, the ability of the drug to bind to its receptor site may be hampered by changes in functional motifs of the target protein. Moreover, the level of the signalling message may be influenced by changes in the amount of target protein produced in different patients. All these factors could be due to sequence variations in human genes.

With respect to cardiovascular disease per se, statins are probably the most frequently prescribed class of drug, and understandably great effort has been invested in trying to understand the genetics of variations in patient response as well as toxicity of these drugs. To begin with, the fact that considerable interindividual variation exists in response to statin therapy, in terms of both lipid-lowering and adverse drug reactions has been demonstrated in several candidate gene studies as well as meta-analyses from several primary and secondary intervention studies. Notably, most statins are the substrates of several cytochrome P450s (CYPs), and polymorphisms in these enzymes appear to be responsible for variations in their hypolipidemic activity. Strong association of sequence variants of several genes including *HMGCR, CETP, SREBF1 and ABCG8* genes with the alteration in LDL-C reduction capability of different statins appears to be well-established[153]. Besides, drugs such as pravastatin and rotuvastatin are not susceptible to CYP inhibition, but are substrates of the a hepatic transporter and solute organic anion-transporting polypeptide (OATP) 1B1 (encoded by the *SLCO1B1* gene) which is responsible for liver transportation of the statins[154]. Variants of the apolipoprotein, particularly the apo E and apo A5 also have their share in these processes[155-158]. Similarly, genetically impaired ABCG2 transporter efflux activity results in a marked increase in systemic exposure to various statins[159]. Importantly, the effects of these genetic polymorphisms differ depending on the specific statin that is used. With respect to reduction of MI though the use of statin, defective interaction may occur with a number of variants in several genes including the scavenger receptor class B member 1 (*SCARB1*) *HMGCR, SREBF1*, *ABCG5/8*, *PCSK9,* hepatic triglyceride lipase *(LIPC*), *ABCA1*, *PPAR*, LDLR-related protein 1 (*LRP1*) and *SOAT1*[160 161 162]. Other important features of statin pharmacokinetics include calmin gene polymorphism, which may exhibit differences on the actions of simvastatin, pravastatin and artovastatin on total and LDL-C reduction[163] and ABCG2 which is associated with significantly greater LDL-C reduction with rosuvastatin compared to other statins [164].

Furthermore, despite the high efficacy and almost universal use of statins in cardiovascular disease prevention, their adverse actions appear to be their greatest disadvantage. In this regard, the use of high dose of statins in combination with other drugs is also not without problems, since their pharmacokinetics may be altered, leading to increasing blood levels with consequent risk of liver or muscle toxicity as demonstrated most commonly for agents metabolized by the CYP450 3A4 enzyme. Specifically, simvastatin has been associated with an increased risk of rhabdomyolysis when taken in combination with multiple inhibitors of CYP3A4[161]. Moreover, while the safety data for the statins suggest a very low incidence of severe adverse reactions, such as rhabdomyolysis and myopathy, myalgias without serum

creatine kinase elevation remains a common side effect and the important reason for discontinuation of therapy. Myopathy has been reported in a considerable number of patients, but the mechanisms underlying muscle injury have yet to be fully characterized. These mechanisms may include statin-induced differences in Chol:phospholipid ratio, isoprenoid levels, small GTP binding proteins and apoptosis[165]. Furthermore, while depletion of Chol within the myocyte cell wall and/or the depletion of key intermediates within its synthesis pathway are hypothesized as possible mechanisms of statin-associated adverse drug actions (ADRs), pharmacogenetic variability may also contribute as a risk for ADRs[155]. These may include, for example, enzymes, transporters, cell membrane receptors, intracellular receptors or components of ion channels that contribute to the pharmacokinetics or pharmacodynamics of response to a particular drug. One of the genes identified thus far as likely to contribute to the development of simvastatin-induced myopathy and myalgia is the *SLCO1B1* gene[161,166]. Thereby, the *SLCO1B* variant increases the risk of statin-induced myopathy by reducing the hepatic uptake of the statins. Essentially all statins are, in fact, substrates of membrane transporters, whereby *SLCO1B1* polymorphisms can decrease the liver uptake, as well as the therapeutic potential of these agents, and may be linked to their muscular side-effects. Elevated levels of simvastatin metabolites (but not pravastatin) have been observed among carriers of *SLCO1B1*5* allele, pointing to a genetic susceptibility to both myopathy and statin-induced myalgias in the absence of elevated serum creatinine kinase[167]. The risk of myalgia among *SLCO1B1* carriers appears to be unique for artovastatin. Also, polymorphic changes in the CYPs, *CYP2D6, CYP3A4* and *CYP3A5* are linked with statin-induced myopathy and show variable effects on altering the pharmacokinetic profile of statin metabolism. Furthermore, although myalgia appears to be associated with the whole group of drugs, there is a significant amount of variability within the class[168 169].

The major site of statin action is within hepatocytes and recent interest has also focussed on seeking the explanation for the differences in the impact of genetic variations in hepatic influx and efflux transporters[170]. Among the most promising candidate genes for pharmacogenomic analysis of statin therapy is the *HMGCR* as a direct target gene and other genes modulating lipid and lipoprotein homeostasis such as *PPARs*. A synergist effect has also been reported in which attenuation in lipid-lowering response to simvastatin caused by some *LDLR* haplotypes was enhanced in the presence of *HMGCR* haplotypes[171], pointing to modulation of effects through gene-gene interactions.

Apart from statin therapy, antiplatelet therapy represents the basis of treatment for the short- and long-term prevention of atherothrombotic disease processes, in particular in high-risk settings such as in patients with ACS and those undergoing percutaneous coronary intervention. Currently, the most common treatment involves dual antiplatelet therapy with aspirin and clopidogrel. Specifically, clopidogrel is a prodrug that undergoes hepatic biotransformation by CYP2C19 into its active metabolite. However, a considerable number of patients continue to experience adverse outcomes, including both bleeding and recurrent ischemic events, possibly attributable, in part at least, to the broad variability in individual response profiles to this standard antiplatelet treatment regimen[172]. Gene polymorphisms affecting clopidogrel metabolic bioactivation and platelet function may be responsible. These include CYP2C19 *2, *3 and *17, CYP2C9 *2 and *3, MDR1*2, and functional variants in the genes encoding platelet membrane receptors and intracellular

signaling proteins[173,174]. Moreover, gene polymorphisms may also have an important role in determining levels of platelet inhibition and provide a tool for identifying patients at risk of adverse events.

In ischemic heart disease, the most commonly suspected genes are those related to lipid metabolism, coagulation and fibrinolytic systems as well as the RAAS pathway. Drugs that modulate the RAAS play an important role in advanced cardiovascular disease prevention strategies. Inhibitors of the RAAS, in particular ACE inhibitors are beneficial in specific patient groups, including those with hypertension, heart failure, diabetes mellitus and stable CAD. Hence ACE inhibitors are among the most commonly used drugs in stable CAD since they have been proven to be effective for reducing the risk of cardiovascular morbidity and mortality. However, while clinical trials demonstrated some consistent beneficial effect of ACE inhibitors across groups of patients based on clinical characteristics, the variability in treatment response on the individual patient level is high. Just as in the treatment of lipidaemic disorders, a primary cause seems to be genetic variations. Furthermore, treatment of diabetes constitutes an integral part of regimens to control coronary vascular disorders, which is similarly subject to issues of differences in patient responses to therapy. Examples include the uncoupling protein variant which has been correlated with β-blocker response among ACS patients with diabetes[175], and the *SLCO1B1* gene variants also thought to be responsible for intolerance in diabetic patients[176]. Variability within the *SLCO1B1* and *ABCB1* genes are also linked with the modification of the effectiveness of statins in the prevention of the clinical outcome of MI[177].

However, while a great deal has been written about the potential of pharmacogenetic testing to informed therapy based on an individual's genetic makeup, and to decide the most effective choice of available drugs, or to avoid dangerous side effects, currently, there is little hard data for either in the field of cardiovascular disease. The usual approach has been the opportunistic use of drug trials in unrelated patients, and to look for differences in response or outcome by candidate gene genotype, for example, in those encoding drug metabolizing enzymes (activators and metabolizers), as well as enzymes and receptors involved in lipid metabolism, adrenergic response, among others. As with all association studies, initially promising results have often failed the test of replication in larger studies. An example is the attempt to employ relationship between the CETPTaq-I variant and response to statins, which has been ultimately withdrawn[178]. Nonetheless, ongoing exploration of genetic polymorphisms that influence response to drug therapy may one day allow the clinician to customize treatment strategies for patients in order to improve their success rate. In the treatment of CAD or ACS, greatest chance appears to have been accomplished in the area of regulating circulating lipids. The hope is that pharmacogenomic testing in future will allow risk stratification of patients to avoid serious side effects and enable clinicians to select lipid-lowering drugs with the highest efficacy resulting in the best response to therapy. The compounds include drugs indicated for dyslipidaemia, such as statins, fibrates, niacin and cholestyramine, as well as those used for other purposes, including calcium channel blockers, angiotensin receptor blockers and glitazones. Furthermore, currently available data from pharmacogenetic trials, a combined analysis of multiple genetic variants in several genes is more likely to give significant results than single gene studies in small cohorts. Thus, larger studies or combination analyses involving more than two different polymorphisms would enable us to find clinically or biologically more meaningful differences, and genes influencing Chol biosynthesis in the liver, such as ABCG5/G8,

CYP7A1, HMGCR, would be good candidates. In this regard, plasma TG concentration is also reemerging as an important cardiovascular disease risk factor. More complete understanding of the genes and variants that modulate plasma TG should enable development of markers for risk prediction, diagnosis, prognosis, and response to therapies and might help specify new directions for therapeutic interventions.

6. Summary

Atherosclerosis is a multifactorial, multistep disease that involves chronic inflammation at every step, from initiation to progression, and all the risk factors contribute to pathogenesis by aggravating the underlying inflammatory process. Despite well-invested efforts to minimize attributable risk from known contributors to the disease such as hypertension, dyslipidaemia, and smoking, CAD remains the number one cause of death in industrialized countries. Clinical trials have consistently demonstrated a family history of coronary disease to be predictive for future cardiovascular events beyond that which would be explained by traditional risk factors. These findings do not only support but also have prompted great interest to study the genomic basis of CAD and MI. Recent advances in genotyping technology have allowed for easier identification and confirmation of susceptibility genes for complex traits across different cohorts in the pathways leading to CAD. This has been facilitated through increased power of studies enhanced by faster accrual of cases and control subjects and more precise genetic mapping, and have allowed us define the genes contributing to a possibly great majority of population-attributable risk for T2DM, hypertension and ACS. Similar progress in replicating novel susceptibility genes for CAD and specifically MI is now advancing rapidly. Several genes have already been identified and more are in the pipeline. With improved resequencing technology and better phenotypic characterization of the CAD cases and control subjects, comprehensive identification and confirmation of genes associated with CAD risk appear to be around the corner, in order to allow us to better quantify CAD risk early enough in life and institute more effective therapy reducing the burden of an individual developing CAD. However, being a complex disease, the disease pathways to CAD are interwound in an intricate network, which together with environmental factors require complex tools to decipher. Such an in-depth knowledge of the various pathogenic mechanisms involved in the disease will significantly enhance our substantiation of the existing knowledge about the disease.

Furthermore, while substantial progress has been made in the identification of common DNA sequence variations in genes influencing the pharmacokinetics and pharmacodynamics of related drugs and in disease-modifying genes relevant for CAD, our present understanding of pathophysiological mechanisms does not offer a reliable approach to address the same at preclinical level. Pharmacogenomics can provide important insights into the therapy of CAD and related risk factors through elucidation of the genetic (or genomic) contribution to variable response to different drugs. The search for genetic polymorphisms may enable us to identify novel determinants of drug responsiveness by studying candidate gene belonging to the gene families that encode proteins involved in pharmacokinetics, the genes encoding proteins engaged pharmacodynamics and genes encoding proteins involved in the underlying disease condition or intermediate phenotype. A better understanding of the pathogenesis of CAD will enhance the advancement of pharmaceutical and lifestyle modifications for reducing mortality resulting from the disease.

7. References

[1] Park SW, Moon YA, Horton JD. Post-transcriptional regulation of low density lipoprotein receptor protein by proprotein convertase subtilisin/kexin type 9a in mouse liver. J Biol Chem 2004;279:50630-8.

[2] Maxwell KN, Breslow JL. Proprotein convertase subtilisin kexin 9: the third locus implicated in autosomal dominant hypercholesterolemia. Curr Opin Lipidol 2005;16:167-72.

[3] Cohen J, Pertsemlidis A, Kotowski IK, Graham R, Garcia CK, Hobbs HH. Low LDL cholesterol in individuals of African descent resulting from frequent nonsense mutations in PCSK9. Nat Genet 2005;37:161-5.

[4] Benjannet S, Rhainds D, Essalmani R, et al. NARC-1/PCSK9 and its natural mutants: zymogen cleavage and effects on the low density lipoprotein (LDL) receptor and LDL cholesterol. J Biol Chem 2004;279:48865-75.

[5] Li H, Li H, Ziegler N, Cui R, Liu J. Recent patents on PCSK9: a new target for treating hypercholesterolemia. Recent Pat DNA Gene Seq 2009;3:201-12.

[6] Le Goff W, Guerin M, Chapman MJ. Pharmacological modulation of cholesteryl ester transfer protein, a new therapeutic target in atherogenic dyslipidemia. Pharmacol Ther 2004;101:17-38.

[7] Le Goff W, Guerin M, Nicaud V, et al. A novel cholesteryl ester transfer protein promoter polymorphism (-971G/A) associated with plasma high-density lipoprotein cholesterol levels. Interaction with the TaqIB and -629C/A polymorphisms. Atherosclerosis 2002; 161:269-79.

[8] Sakai J, Rawson RB. The sterol regulatory element-binding protein pathway: control of lipid homeostasis through regulated intracellular transport. Curr Opin Lipidol 2001;12:261-6.

[9] Horton JD, Goldstein JL, Brown MS. SREBPs: transcriptional mediators of lipid homeostasis. Cold Spring Harb Symp Quant Biol 2002;67:491-8.

[10] Horton JD, Goldstein JL, Brown MS. SREBPs: activators of the complete program of cholesterol and fatty acid synthesis in the liver. J Clin Invest 2002;109:1125-31.

[11] DeBose-Boyd RA, Brown MS, Li WP, Nohturfft A, Goldstein JL, Espenshade PJ. Transport-dependent proteolysis of SREBP: relocation of site-1 protease from Golgi to ER obviates the need for SREBP transport to Golgi. Cell 1999;99:703-12.

[12] Nohturfft A, DeBose-Boyd RA, Scheek S, Goldstein JL, Brown MS. Sterols regulate cycling of SREBP cleavage-activating protein (SCAP) between endoplasmic reticulum and Golgi. Proc Natl Acad Sci U S A 1999;96:11235-40.

[13] Eberle D, Hegarty B, Bossard P, Ferre P, Foufelle F. SREBP transcription factors: master regulators of lipid homeostasis. Biochimie 2004;86:839-48.

[14] Edwards PA, Tabor D, Kast HR, Venkateswaran A. Regulation of gene expression by SREBP and SCAP. Biochim Biophys Acta 2000;1529:103-13.

[15] Eberle D, Clement K, Meyre D, et al. SREBF-1 gene polymorphisms are associated with obesity and type 2 diabetes in French obese and diabetic cohorts. Diabetes 2004;53:2153-7.

[16] Chirieac DV, Collins HL, Cianci J, Sparks JD, Sparks CE. Altered triglyceride-rich lipoprotein production in Zucker diabetic fatty rats. Am J Physiol Endocrinol Metab 2004;287:E42-9.

[17] Wang Z, Jiang T, Li J, et al. Regulation of renal lipid metabolism, lipid accumulation, and glomerulosclerosis in FVBdb/db mice with type 2 diabetes. Diabetes 2005;54:2328-35.

[18] Biddinger SB, Almind K, Miyazaki M, Kokkotou E, Ntambi JM, Kahn CR. Effects of diet and genetic background on sterol regulatory element-binding protein-1c, stearoyl-CoA desaturase 1, and the development of the metabolic syndrome. Diabetes 2005;54:1314-23.

[19] Lin J, Yang R, Tarr PT, et al. Hyperlipidemic effects of dietary saturated fats mediated through PGC-1beta coactivation of SREBP. Cell 2005;120:261-73.

[20] Amemiya-Kudo M, Shimano H, Hasty AH, et al. Transcriptional activities of nuclear SREBP-1a, -1c, and -2 to different target promoters of lipogenic and cholesterogenic genes. Journal of lipid research 2002;43:1220-35.

[21] Shimano H, Yahagi N, Amemiya-Kudo M, et al. Sterol regulatory element-binding protein-1 as a key transcription factor for nutritional induction of lipogenic enzyme genes. J Biol Chem 1999;274:35832-9.

[22] Yahagi N, Shimano H, Hasty AH, et al. A crucial role of sterol regulatory element-binding protein-1 in the regulation of lipogenic gene expression by polyunsaturated fatty acids. J Biol Chem 1999;274:35840-4.

[23] Ten S, Maclaren N. Insulin resistance syndrome in children. J Clin Endocrinol Metab 2004;89:2526-39.

[24] Kim DH, Zhang T, Ringquist S, Dong HH. Targeting FoxO1 for Hypertriglyceridemia. Current drug targets 2011;12:1245-55.

[25] Kamagate A, Dong HH. FoxO1 integrates insulin signaling to VLDL production. Cell Cycle 2008;7:3162-70.

[26] Lamarche B, Lemieux I, Despres JP. The small, dense LDL phenotype and the risk of coronary heart disease: epidemiology, patho-physiology and therapeutic aspects. Diabetes Metab 1999;25:199-211.

[27] Sitia S, Tomasoni L, Atzeni F, et al. From endothelial dysfunction to atherosclerosis. Autoimmun Rev;9:830-4.

[28] Kutti J, Weinfeld A. Platelet survival and platelet production in acute myocardial infarction. Acta Med Scand 1979;205:501-4.

[29] van der Loo B, Martin JF. Megakaryocytes and platelets in vascular disease. Baillieres Clin Haematol 1997;10:109-23.

[30] Khan MA, Collins AJ, Keane WF. Diabetes in the elderly population. Adv Ren Replace Ther 2000;7:32-51.

[31] Norata GD, Catapano AL. Lipid lowering activity of drugs affecting cholesterol absorption. Nutr Metab Cardiovasc Dis 2004;14:42-51.

[32] Perez-Martinez P, Lopez-Miranda J, Perez-Jimenez F, Ordovas JM. Influence of genetic factors in the modulation of postprandial lipemia. Atheroscler Suppl 2008;9:49-55.

[33] Grundy SM. Hypertriglyceridemia, atherogenic dyslipidemia, and the metabolic syndrome. The American journal of cardiology 1998;81:18B-25B.

[34] Grundy SM, Balady GJ, Criqui MH, et al. Primary prevention of coronary heart disease: guidance from Framingham: a statement for healthcare professionals from the AHA Task Force on Risk Reduction. American Heart Association. Circulation 1998;97:1876-87.

[35] Boes E, Coassin S, Kollerits B, Heid IM, Kronenberg F. Genetic-epidemiological evidence on genes associated with HDL cholesterol levels: a systematic in-depth review. Exp Gerontol 2009;44:136-60.

[36] Cohen JC, Kimmel M, Polanski A, Hobbs HH. Molecular mechanisms of autosomal recessive hypercholesterolemia. Curr Opin Lipidol 2003;14:121-7.

[37] Kwiterovich PO, Jr. Clinical relevance of the biochemical, metabolic, and genetic factors that influence low-density lipoprotein heterogeneity. The American journal of cardiology 2002;90:30i-47i.

[38] Moller DE, Kaufman KD. Metabolic syndrome: a clinical and molecular perspective. Annu Rev Med 2005;56:45-62.

[39] Wilcken DE. Overview of inherited metabolic disorders causing cardiovascular disease. J Inherit Metab Dis 2003;26:245-57.

[40] Richter WF, Whitby BR, Chou RC. Distribution of remikiren, a potent orally active inhibitor of human renin, in laboratory animals. Xenobiotica 1996;26:243-54.

[41] Misra A, Khurana L. Obesity-related non-communicable diseases: South Asians vs White Caucasians. Int J Obes (Lond);35:167-87.

[42] Andreassi MG. Coronary atherosclerosis and somatic mutations: an overview of the contributive factors for oxidative DNA damage. Mutat Res 2003;543:67-86.

[43] Connelly PW, Hegele RA. Hepatic lipase deficiency. Crit Rev Clin Lab Sci 1998;35:547-72.

[44] Ferrero-Miliani L, Nielsen OH, Andersen PS, Girardin SE. Chronic inflammation: importance of NOD2 and NALP3 in interleukin-1β generation. Clinical & Experimental Immunology 2007;147:227-35.

[45] Onat A, Hergenc G. Low-grade inflammation, and dysfunction of high-density lipoprotein and its apolipoproteins as a major driver of cardiometabolic risk. Metabolism;60:499-512.

[46] De Sutter J, Struyf S, Van de Veire NR, Philippe J, De Buyzere M, Van Damme J. Cardiovascular determinants and prognostic significance of CC Chemokine Ligand-18 (CCL18/PARC) in patients with stable coronary artery disease. J Mol Cell Cardiol 2010;49:894-6.

[47] Fried SK, Rao SP. Sugars, hypertriglyceridemia, and cardiovascular disease. Am J Clin Nutr 2003;78:873S-80S.

[48] Hochman JS, Tamis JE, Thompson TD, et al. Sex, clinical presentation, and outcome in patients with acute coronary syndromes. Global Use of Strategies to Open Occluded Coronary Arteries in Acute Coronary Syndromes IIb Investigators. N Engl J Med 1999;341:226-32.

[49] Elsaesser A, Hamm CW. Acute coronary syndrome: the risk of being female. Circulation 2004;109:565-7.

[50] Hasdai D, Porter A, Rosengren A, Behar S, Boyko V, Battler A. Effect of gender on outcomes of acute coronary syndromes. Am J Cardiol 2003;91:1466-9, A6.

[51] Friedlander Y, Arbogast P, Schwartz SM, et al. Family history as a risk factor for early onset myocardial infarction in young women. Atherosclerosis 2001;156:201-7.
[52] Austin MA, Hutter CM, Zimmern RL, Humphries SE. Genetic causes of monogenic heterozygous familial hypercholesterolemia: a HuGE prevalence review. Am J Epidemiol 2004;160:407-20.
[53] Abifadel M, Rabes JP, Devillers M, et al. Mutations and polymorphisms in the proprotein convertase subtilisin kexin 9 (PCSK9) gene in cholesterol metabolism and disease. Hum Mutat 2009;30:520-9.
[54] Damani SB, Topol EJ. Future use of genomics in coronary artery disease. J Am Coll Cardiol 2007;50:1933-40.
[55] Barbagallo CM, Emmanuele G, Cefalu AB, et al. Autosomal recessive hypercholesterolemia in a Sicilian kindred harboring the 432insA mutation of the ARH gene. Atherosclerosis 2003;166:395-400.
[56] Costet P, Krempf M, Cariou B. PCSK9 and LDL cholesterol: unravelling the target to design the bullet. Trends Biochem Sci 2008;33:426-34.
[57] Humphries SE, Norbury G, Leigh S, Hadfield SG, Nair D. What is the clinical utility of DNA testing in patients with familial hypercholesterolaemia? Curr Opin Lipidol 2008;19:362-8.
[58] Assmann G, Gotto AM, Jr. HDL cholesterol and protective factors in atherosclerosis. Circulation 2004;109:III8-14.
[59] Jaye M, Krawiec J. Endothelial lipase and HDL metabolism. Curr Opin Lipidol 2004;15:183-9.
[60] Naukkarinen J, Ehnholm C, Peltonen L. Genetics of familial combined hyperlipidemia. Curr Opin Lipidol 2006;17:285-90.
[61] Garg A, Simha V. Update on dyslipidemia. J Clin Endocrinol Metab 2007;92:1581-9.
[62] Enas EA, Chacko V, Senthilkumar A, Puthumana N, Mohan V. Elevated lipoprotein(a)--a genetic risk factor for premature vascular disease in people with and without standard risk factors: a review. Dis Mon 2006;52:5-50.
[63] Hooper AJ, van Bockxmeer FM, Burnett JR. Monogenic hypocholesterolaemic lipid disorders and apolipoprotein B metabolism. Crit Rev Clin Lab Sci 2005;42:515-45.
[64] Zambon A, Brown BG, Deeb SS, Brunzell JD. Genetics of apolipoprotein B and apolipoprotein AI and premature coronary artery disease. Journal of internal medicine 2006;259:473-80.
[65] Konopka A, Szperl M, Piotrowski W, Roszczynko M, Stepinska J. Influence of Renin-Angiotensin System Gene Polymorphisms on the Risk of ST-Segment-Elevation Myocardial Infarction and Association with Coronary Artery Disease Risk Factors. Mol Diagn Ther 2011;15:167-76.
[66] Dzimiri N, Muiya P, Mohamed G, et al. Haplotypes encompassing the PSMA6 and KIAA0391 Gene Cluster confer risk for Myocardial Infarction. FASEB J 2009; 23:573.1-.
[67] Inoue N, Kawashima S, Kanazawa K, Yamada S, Akita H, Yokoyama M. Polymorphism of the NADH/NADPH oxidase p22 phox gene in patients with coronary artery disease. Circulation 1998;97:135-7.

[68] Sethi AA, Nordestgaard BG, Tybjaerg-Hansen A. Angiotensinogen gene polymorphism, plasma angiotensinogen, and risk of hypertension and ischemic heart disease: a meta-analysis. Arteriosclerosis, thrombosis, and vascular biology 2003;23:1269-75.

[69] Sethi AA, Nordestgaard BG, Gronholdt ML, Steffensen R, Jensen G, Tybjaerg-Hansen A. Angiotensinogen single nucleotide polymorphisms, elevated blood pressure, and risk of cardiovascular disease. Hypertension 2003;41:1202-11.

[70] Sethi AA, Tybjaerg-Hansen A, Gronholdt ML, Steffensen R, Schnohr P, Nordestgaard BG. Angiotensinogen mutations and risk for ischemic heart disease, myocardial infarction, and ischemic cerebrovascular disease. Six case-control studies from the Copenhagen City Heart Study. Ann Intern Med 2001;134:941-54.

[71] Cha SH, Kim HT, Jang Y, et al. Association of alpha-adducin Gly460Trp polymorphism with coronary artery disease in a Korean population. J Hypertens 2007;25:2413-20.

[72] Zee RY, Cook NR, Cheng S, Erlich HA, Lindpaintner K, Ridker PM. Multi-locus candidate gene polymorphisms and risk of myocardial infarction: a population-based, prospective genetic analysis. Journal of thrombosis and haemostasis : JTH 2006;4:341-8.

[73] O'Donnell CJ, Lindpaintner K, Larson MG, et al. Evidence for association and genetic linkage of the angiotensin-converting enzyme locus with hypertension and blood pressure in men but not women in the Framingham Heart Study. Circulation 1998;97:1766-72.

[74] Raman VK, Lee YA, Lindpaintner K. The cardiac renin-angiotensin-aldosterone system and hypertensive cardiac hypertrophy. The American journal of cardiology 1995;76:18D-23D.

[75] Lindpaintner K. Genes, hypertension, and cardiac hypertrophy. N Engl J Med 1994;330:1678-9.

[76] Wang XL, Sim AS, Badenhop RF, McCredie RM, Wilcken DE. A smoking-dependent risk of coronary artery disease associated with a polymorphism of the endothelial nitric oxide synthase gene. Nat Med 1996;2:41-5.

[77] Klerk M, Verhoef P, Clarke R, Blom HJ, Kok FJ, Schouten EG. MTHFR 677C-->T polymorphism and risk of coronary heart disease: a meta-analysis. Jama 2002;288:2023-31.

[78] Stenina OI, Byzova TV, Adams JC, McCarthy JJ, Topol EJ, Plow EF. Coronary artery disease and the thrombospondin single nucleotide polymorphisms. Int J Biochem Cell Biol 2004;36:1013-30.

[79] Stenina OI, Desai SY, Krukovets I, et al. Thrombospondin-4 and its variants: expression and differential effects on endothelial cells. Circulation 2003;108:1514-9.

[80] Cui J, Randell E, Renouf J, et al. Thrombospondin-4 1186G>C (A387P) is a sex-dependent risk factor for myocardial infarction: a large replication study with increased sample size from the same population. American heart journal 2006;152:543 e1-5.

[81] Zwicker JI, Peyvandi F, Palla R, et al. The thrombospondin-1 N700S polymorphism is associated with early myocardial infarction without altering von Willebrand factor multimer size. Blood 2006;108:1280-3.

[82] Gao L, He GP, Dai J, et al. Association of thrombospondin-1 gene polymorphisms with myocardial infarction in a Chinese Han population. Chin Med J (Engl) 2008;121:78-81.

[83] Suriyaprom K, Phonrat B, Namjuntra P, Harnroongroj T, Tungtrongchitr R. The -11377C > G adiponectin gene polymorphism alters the adiponectin concentration and the susceptibility to type 2 diabetes in Thais. Int J Vitam Nutr Res;80:216-24.

[84] Siitonen N, Pulkkinen L, Lindstrom J, et al. Association of ADIPOQ gene variants with body weight, type 2 diabetes and serum adiponectin concentrations: the Finnish Diabetes Prevention Study. BMC Med Genet;12:5.

[85] Yang WS, Chuang LM. Human genetics of adiponectin in the metabolic syndrome. J Mol Med (Berl) 2006;84:112-21.

[86] Du W, Li Q, Lu Y, et al. Genetic variants in ADIPOQ gene and the risk of type 2 diabetes: a case-control study of Chinese Han population. Endocrine.

[87] Ikeda Y, Hama S, Kajimoto K, Okuno T, Tsuchiya H, Kogure K. Quantitative Comparison of Adipocytokine Gene Expression during Adipocyte Maturation in Non-obese and Obese Rats. Biol Pharm Bull;34:865-70.

[88] Galgani A, Valdes A, Erlich HA, et al. Homozygosity for the Ala allele of the PPARgamma2 Pro12Ala polymorphism is associated with reduced risk of coronary artery disease. Dis Markers 2010;29:259-64.

[89] Hetherington MM, Cecil JE. Gene-environment interactions in obesity. Forum of nutrition 2010;63:195-203.

[90] Ranjith N, Pegoraro RJ, Shanmugam R. Obesity-associated genetic variants in young Asian Indians with the metabolic syndrome and myocardial infarction. Cardiovasc J Afr;22:25-30.

[91] Nagi DK, Foy CA, Mohamed-Ali V, Yudkin JS, Grant PJ, Knowler WC. Angiotensin-1-converting enzyme (ACE) gene polymorphism, plasma ACE levels, and their association with the metabolic syndrome and electrocardiographic coronary artery disease in Pima Indians. Metabolism 1998;47:622-6.

[92] Assali A, Ghayour-Mobarhan M, Sahebkar A, et al. Association of angiotensin II type 1 receptor gene A1166C polymorphism with the presence of diabetes mellitus and metabolic syndrome in patients with documented coronary artery disease. Eur J Intern Med;22:254-61.

[93] Safranow K, Czyzycka E, Binczak-Kuleta A, et al. Association of C34T AMPD1 gene polymorphism with features of metabolic syndrome in patients with coronary artery disease or heart failure. Scand J Clin Lab Invest 2009;69:102-12.

[94] Muendlein A, Saely CH, Geller-Rhomberg S, et al. Single nucleotide polymorphisms of TCF7L2 are linked to diabetic coronary atherosclerosis. PLoS One 2011;6:e17978.

[95] Abu-Amero KK, Al-Mohanna F, Alboudairi O, Mohamed GH, Dzimiri N. The interactive role of diabetes mellitus type 2 and E-selectin S128R mutation on coronary heart disease manifestation. BMC Med Genet 2007;8:35.

[96] Zheng YY, Xie X, Ma YT, et al. Relationship between a Novel Polymorphism of the C5L2 Gene and Coronary Artery Disease. PLoS One 2011;6:e20984.

[97] Listi F, Candore G, Lio D, et al. Association between C1019T polymorphism of connexin37 and acute myocardial infarction: a study in patients from Sicily. Int J Cardiol 2005;102:269-71.

[98] Incalcaterra E, Caruso M, Balistreri CR, et al. Role of genetic polymorphisms in myocardial infarction at young age. Clin Hemorheol Microcirc;46:291-8.

[99] Incalcaterra E, Hoffmann E, Averna MR, Caimi G. Genetic risk factors in myocardial infarction at young age. Minerva Cardioangiol 2004;52:287-312.

[100] Frikke-Schmidt R, Nordestgaard BG, Stene MC, et al. Association of loss-of-function mutations in the ABCA1 gene with high-density lipoprotein cholesterol levels and risk of ischemic heart disease. Jama 2008;299:2524-32.

[101] Barlic J, Murphy PM. An oxidized lipid-peroxisome proliferator-activated receptor gamma-chemokine pathway in the regulation of macrophage-vascular smooth muscle cell adhesion. Trends Cardiovasc Med 2007;17:269-74.

[102] Hansson GK. Atherosclerosis--an immune disease: The Anitschkov Lecture 2007. Atherosclerosis 2009;202:2-10.

[103] Mackness MI, Mackness B, Durrington PN, Connelly PW, Hegele RA. Paraoxonase: biochemistry, genetics and relationship to plasma lipoproteins. Curr Opin Lipidol 1996;7:69-76.

[104] Kaplan M, Aviram M. Oxidized low density lipoprotein: atherogenic and proinflammatory characteristics during macrophage foam cell formation. An inhibitory role for nutritional antioxidants and serum paraoxonase. Clin Chem Lab Med 1999;37:777-87.

[105] Li HL, Liu DP, Liang CC. Paraoxonase gene polymorphisms, oxidative stress, and diseases. J Mol Med 2003;81:766-79.

[106] Grech ED, Dodd NJ, Jackson MJ, Morrison WL, Faragher EB, Ramsdale DR. Evidence for free radical generation after primary percutaneous transluminal coronary angioplasty recanalization in acute myocardial infarction. Am J Cardiol 1996;77:122-7.

[107] Goliasch G, Wiesbauer F, Grafl A, et al. The effect of p22-PHOX (CYBA) polymorphisms on premature coronary artery disease (</= 40 years of age). Thromb Haemost;105:529-34.

[108] Grove EL, Hvas AM, Kristensen SD. Immature platelets in patients with acute coronary syndromes. Thromb Haemost 2009;101:151-6.

[109] Kunicki TJ, Nugent DJ. The genetics of normal platelet reactivity. Blood 2010;116:2627-34.

[110] Karakas M, Zierer A, Herder C, et al. Leptin, adiponectin, their ratio and risk of Coronary Heart Disease: results from the MONICA/KORA Augsburg Study 1984-2002. Atherosclerosis 2010;209:220-5.

[111] A genome-wide association study in Europeans and South Asians identifies five new loci for coronary artery disease. Nat Genet 2011;43:339-44.

[112] Connelly JJ, Wang T, Cox JE, et al. GATA2 is associated with familial early-onset coronary artery disease. PLoS Genet 2006;2:e139.

[113] Elhawari S, Al-Boudari O, Muiya P, et al. A study of the role of the Myocyte-specific Enhancer Factor-2A gene in coronary artery disease. Atherosclerosis;209:152-4.

[114] Yuan H, Lu HW, Hu J, Chen SH, Yang GP, Huang ZJ. MEF2A gene and susceptibility to coronary artery disease in the Chinese people. Zhong Nan Da Xue Xue Bao Yi Xue Ban 2006;31:453-7.

[115] Bhagavatula MR, Fan C, Shen GQ, et al. Transcription factor MEF2A mutations in patients with coronary artery disease. Hum Mol Genet 2004;13:3181-8.

[116] Dzimiri N, Muiya P, Alsmadi O, et al. The KIAA0391 and PSMA6 gene cluster confers a risk factor for coronary artery disease. FASEB J 2009;23:573.2-.

[117] Hinohara K, Nakajima T, Sasaoka T, et al. Replication studies for the association of PSMA6 polymorphism with coronary artery disease in East Asian populations. J Hum Genet 2009;54:248-51.

[118] Linnemann AK, O'Geen H, Keles S, Farnham PJ, Bresnick EH. Genetic framework for GATA factor function in vascular biology. Proc Natl Acad Sci U S A 2011.

[119] Roberts R, Chen L, Wells GA, Stewart AF. Recent success in the discovery of coronary artery disease genes. Can J Physiol Pharmacol 2011.

[120] Tsai CT, Hwang JJ, Wu CK, et al. Polygenetic regression model of renin-angiotensin system genes and the risk of coronary artery disease in a large angiographic population. Clinica chimica acta; international journal of clinical chemistry 2011;412:619-24.

[121] Freitas AI, Mendonca I, Brion M, et al. RAS gene polymorphisms, classical risk factors and the advent of coronary artery disease in the Portuguese population. BMC Cardiovasc Disord 2008;8:15.

[122] Mendonca MI, Dos Reis RP, Freitas AI, et al. Interaction of paraoxonase-192 polymorphism with low HDL-cholesterol in coronary artery disease risk. Rev Port Cardiol;29:571-80.

[123] Mendonca MI, Dos Reis RP, Freitas AI, et al. Gene-gene interaction affects coronary artery disease risk. Rev Port Cardiol 2009;28:397-415.

[124] Benes P, Muzik J, Benedik J, et al. Single effects of apolipoprotein B, (a), and E polymorphisms and interaction between plasminogen activator inhibitor-1 and apolipoprotein(a) genotypes and the risk of coronary artery disease in Czech male caucasians. Molecular genetics and metabolism 2000;69:137-43.

[125] Peng DQ, Zhao SP, Nie S, Li J. Gene-gene interaction of PPARgamma and ApoE affects coronary heart disease risk. International journal of cardiology 2003;92:257-63.

[126] Wu CK, Luo JL, Wu XM, et al. A propensity score-based case-control study of renin-angiotensin system gene polymorphisms and diastolic heart failure. Atherosclerosis 2009;205:497-502.

[127] Tsai CT, Hwang JJ, Ritchie MD, et al. Renin-angiotensin system gene polymorphisms and coronary artery disease in a large angiographic cohort: detection of high order gene-gene interaction. Atherosclerosis 2007;195:172-80.

[128] Agirbasli D, Agirbasli M, Williams SM, Phillips JA, 3rd. Interaction among 5,10 methylenetetrahydrofolate reductase, plasminogen activator inhibitor and endothelial nitric oxide synthase gene polymorphisms predicts the severity of coronary artery disease in Turkish patients. Coronary artery disease 2006;17:413-7.

[129] Ye S, Dhillon S, Seear R, et al. Epistatic interaction between variations in the angiotensin I converting enzyme and angiotensin II type 1 receptor genes in relation to extent of coronary atherosclerosis. Heart 2003;89:1195-9.
[130] Corella D, Guillen M, Saiz C, et al. Associations of LPL and APOC3 gene polymorphisms on plasma lipids in a Mediterranean population: interaction with tobacco smoking and the APOE locus. Journal of lipid research 2002;43:416-27.
[131] Naber CK, Husing J, Wolfhard U, Erbel R, Siffert W. Interaction of the ACE D allele and the GNB3 825T allele in myocardial infarction. Hypertension 2000;36:986-9.
[132] Lopez-Miranda J, Williams C, Lairon D. Dietary, physiological, genetic and pathological influences on postprandial lipid metabolism. Br J Nutr 2007;98:458-73.
[133] Perez-Martinez P, Delgado-Lista J, Perez-Jimenez F, Lopez-Miranda J. Update on genetics of postprandial lipemia. Atheroscler Suppl;11:39-43.
[134] Harrap SB, Zammit KS, Wong ZY, et al. Genome-wide linkage analysis of the acute coronary syndrome suggests a locus on chromosome 2. Arteriosclerosis, thrombosis, and vascular biology 2002;22:874-8.
[135] Brscic E, Bergerone S, Gagnor A, et al. Acute myocardial infarction in young adults: prognostic role of angiotensin-converting enzyme, angiotensin II type I receptor, apolipoprotein E, endothelial constitutive nitric oxide synthase, and glycoprotein IIIa genetic polymorphisms at medium-term follow-up. American heart journal 2000;139:979-84.
[136] Isordia-Salas I, Mendoza-Valdez AL, Almeida-Gutierrez E, Borrayo-Sanchez G. Genetic factors of the hemostatic system in young patients with myocardial infarction. Cir Cir;78:93-7.
[137] Mannucci PM, Asselta R, Duga S, et al. The association of factor V Leiden with myocardial infarction is replicated in 1880 patients with premature disease. J Thromb Haemost;8:2116-21.
[138] Rallidis LS, Gialeraki A, Merkouri E, et al. Reduced carriership of 4G allele of plasminogen activator inhibitor-1 4G/5G polymorphism in very young survivors of myocardial infarction. J Thromb Thrombolysis;29:497-502.
[139] Pegoraro RJ, Ranjith N. Plasminogen activator inhibitor type 1 (PAI-1) and platelet glycoprotein IIIa (PGIIIa) polymorphisms in young Asian Indians with acute myocardial infarction. Cardiovasc J S Afr 2005;16:266-70.
[140] Pasalic D, Jurcic Z, Stipancic G, et al. Missense mutation W86R in exon 3 of the lipoprotein lipase gene in a boy with chylomicronemia. Clin Chim Acta 2004;343:179-84.
[141] Burzotta F, Paciaroni K, De Stefano V, et al. G20210A prothrombin gene polymorphism and coronary ischaemic syndromes: a phenotype-specific meta-analysis of 12 034 subjects. Heart 2004;90:82-6.
[142] Li YH, Chen JH, Tsai WC, et al. Synergistic effect of thrombomodulin promoter -33G/A polymorphism and smoking on the onset of acute myocardial infarction. Thromb Haemost 2002;87:86-91.
[143] Ranjith N, Pegoraro RJ, Rom L. Haemostatic gene polymorphisms in young indian asian subjects with acute myocardial infarction. Med Sci Monit 2003;9:CR417-21.

[144] Hubacek JA, Poledne R, Pitha J, Aschermann M, Skalicka H, Stanek V. Apolipoprotein E Arg136 --> Cys in individuals with premature myocardial infarction. Folia Biol (Praha) 2009;55:116-8.

[145] Liu PY, Chen JH, Li YH, Wu HL, Shi GY. Synergistic effect of stromelysin-1 (matrix metallo-proteinase-3) promoter 5A/6A polymorphism with smoking on the onset of young acute myocardial infarction. Thromb Haemost 2003;90:132-9.

[146] Gonzalez-Porras JR, Martin-Herrero F, Garcia-Sanz R, et al. Hyperhomocysteinemia is a risk factor of recurrent coronary event in young patients irrespective to the MTHFR C677T polymorphism. Thromb Res 2007;119:691-8.

[147] Biezeveld MH, Geissler J, Weverling GJ, et al. Polymorphisms in the mannose-binding lectin gene as determinants of age-defined risk of coronary artery lesions in Kawasaki disease. Arthritis Rheum 2006;54:369-76.

[148] Gonzalez-Conejero R, Corral J, Roldan V, et al. A common polymorphism in the annexin V Kozak sequence (-1C>T) increases translation efficiency and plasma levels of annexin V, and decreases the risk of myocardial infarction in young patients. Blood 2002;100:2081-6.

[149] Crobu F, Palumbo L, Franco E, et al. Role of TGF-beta1 haplotypes in the occurrence of myocardial infarction in young Italian patients. BMC Med Genet 2008;9:13.

[150] van Schie MC, de Maat MP, Isaacs A, et al. Variation in the von Willebrand factor gene is associated with von Willebrand factor levels and with the risk for cardiovascular disease. Blood;117:1393-9.

[151] Franco E, Palumbo L, Crobu F, et al. Renin-angiotensin-aldosterone system polymorphisms: a role or a hole in occurrence and long-term prognosis of acute myocardial infarction at young age. BMC Med Genet 2007;8:27.

[152] Yeh PS, Lin HJ, Li YH, et al. Prognosis of young ischemic stroke in Taiwan: impact of prothrombotic genetic polymorphisms. Thromb Haemost 2004;92:583-9.

[153] Neuvonen PJ. Drug interactions with HMG-CoA reductase inhibitors (statins): the importance of CYP enzymes, transporters and pharmacogenetics. Curr Opin Investig Drugs 2010;11:323-32.

[154] Sirtori CR, Mombelli G, Triolo M, Laaksonen R. Clinical response to statins: Mechanism(s) of variable activity and adverse effects. Ann Med 2011.

[155] Maggo SD, Kennedy MA, Clark DW. Clinical implications of pharmacogenetic variation on the effects of statins. Drug Saf 2011;34:1-19.

[156] Hubacek JA, Adamkova V, Prusikova M, et al. Impact of apolipoprotein A5 variants on statin treatment efficacy. Pharmacogenomics 2009;10:945-50.

[157] Regieli JJ, Jukema JW, Grobbee DE, et al. CETP genotype predicts increased mortality in statin-treated men with proven cardiovascular disease: an adverse pharmacogenetic interaction. Eur Heart J 2008;29:2792-9.

[158] Nieminen T, Kahonen M, Viiri LE, Gronroos P, Lehtimaki T. Pharmacogenetics of apolipoprotein E gene during lipid-lowering therapy: lipid levels and prevention of coronary heart disease. Pharmacogenomics 2008;9:1475-86.

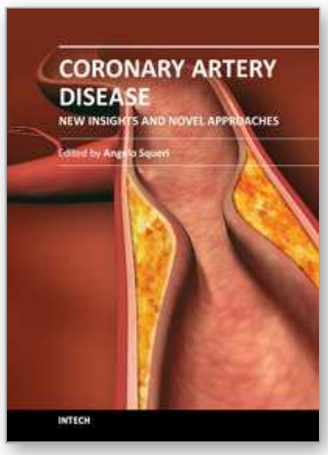
[159] Niemi M. Transporter pharmacogenetics and statin toxicity. Clin Pharmacol Ther 2010;87:130-3.

[160] Peters BJ, Pett H, Klungel OH, et al. Genetic variability within the cholesterol lowering pathway and the effectiveness of statins in reducing the risk of MI. Atherosclerosis 2011;217:458-64.
[161] Abbasi F, Chu JW, McLaughlin T, Lamendola C, Leary ET, Reaven GM. Effect of metformin treatment on multiple cardiovascular disease risk factors in patients with type 2 diabetes mellitus. Metabolism 2004;53:159-64.
[162] Chien KL, Wang KC, Chen YC, et al. Common sequence variants in pharmacodynamic and pharmacokinetic pathway-related genes conferring LDL cholesterol response to statins. Pharmacogenomics 2010;11:309-17.
[163] Barber MJ, Mangravite LM, Hyde CL, et al. Genome-wide association of lipid-lowering response to statins in combined study populations. PLoS One 2010;5:e9763.
[164] Hu M, To KK, Mak VW, Tomlinson B. The ABCG2 transporter and its relations with the pharmacokinetics, drug interaction and lipid-lowering effects of statins. Expert Opin Drug Metab Toxicol 2011;7:49-62.
[165] Tiwari A, Bansal V, Chugh A, Mookhtiar K. Statins and myotoxicity: a therapeutic limitation. Expert Opin Drug Saf 2006;5:651-66.
[166] Voora D, Shah SH, Spasojevic I, et al. The SLCO1B1*5 genetic variant is associated with statin-induced side effects. J Am Coll Cardiol 2009;54:1609-16.
[167] Rowan C, Brinker AD, Nourjah P, et al. Rhabdomyolysis reports show interaction between simvastatin and CYP3A4 inhibitors. Pharmacoepidemiol Drug Saf 2009;18:301-9.
[168] Niemi M, Pasanen MK, Neuvonen PJ. Organic anion transporting polypeptide 1B1: a genetically polymorphic transporter of major importance for hepatic drug uptake. Pharmacol Rev 2011;63:157-81.
[169] Peters B, Maitland-van der Zee AH. Pharmacogenomic importance of pravastatin. Pharmacogenomics 2008;9:1207-10.
[170] Romaine SP, Bailey KM, Hall AS, Balmforth AJ. The influence of SLCO1B1 (OATP1B1) gene polymorphisms on response to statin therapy. Pharmacogenomics J;10:1-11.
[171] Mangravite LM, Medina MW, Cui J, et al. Combined influence of LDLR and HMGCR sequence variation on lipid-lowering response to simvastatin. Arteriosclerosis, thrombosis, and vascular biology 2010;30:1485-92.
[172] Shuldiner AR, O'Connell JR, Bliden KP, et al. Association of cytochrome P450 2C19 genotype with the antiplatelet effect and clinical efficacy of clopidogrel therapy. JAMA 2009;302:849-57.
[173] Xie HG, Zou JJ, Hu ZY, Zhang JJ, Ye F, Chen SL. Individual variability in the disposition of and response to clopidogrel: pharmacogenomics and beyond. Pharmacol Ther 2011;129:267-89.
[174] Gladding P, Webster M, Zeng I, et al. The pharmacogenetics and pharmacodynamics of clopidogrel response: an analysis from the PRINC (Plavix Response in Coronary Intervention) trial. JACC Cardiovasc Interv 2008;1:620-7.
[175] Beitelshees AL, Finck BN, Leone TC, et al. Interaction between the UCP2 -866 G>A polymorphism, diabetes, and beta-blocker use among patients with acute coronary syndromes. Pharmacogenet Genomics 2010;20:231-8.

[176] Donnelly LA, Doney AS, Tavendale R, et al. Common nonsynonymous substitutions in SLCO1B1 predispose to statin intolerance in routinely treated individuals with type 2 diabetes: a go-DARTS study. Clin Pharmacol Ther 2011;89:210-6.

[177] Peters BJ, Rodin AS, Klungel OH, et al. Pharmacogenetic interactions between ABCB1 and SLCO1B1 tagging SNPs and the effectiveness of statins in the prevention of myocardial infarction. Pharmacogenomics 2010;11:1065-76.

[178] Chasman DI, Posada D, Subrahmanyan L, Cook NR, Stanton VP, Jr., Ridker PM. Pharmacogenetic study of statin therapy and cholesterol reduction. Jama 2004;291:2821-7.



Coronary Artery Disease - New Insights and Novel Approaches
Edited by Dr. Angelo Squeri

ISBN 978-953-51-0344-8
Hard cover, 260 pages
Publisher InTech
Published online 16, March, 2012
Published in print edition March, 2012

Coronary Artery disease is one of the leading causes of death in industrialized countries and is responsible for one out of every six deaths in the United States. Remarkably, coronary artery disease is also largely preventable. The biggest challenge in the next years is to reduce the incidence of coronary artery disease worldwide. A complete knowledge of the mechanisms responsible for the development of ischaemic heart disease is an essential prerequisite to a better management of this pathology improving prevention and therapy. This book has been written with the intention of providing new concepts about coronary artery disease pathogenesis that may link various aspects of the disease, going beyond the traditional risk factors.

How to reference
In order to correctly reference this scholarly work, feel free to copy and paste the following:

Nduna Dzimiri (2012). Gene Polymorphism and Coronary Heart Disease, Coronary Artery Disease - New Insights and Novel Approaches, Dr. Angelo Squeri (Ed.), ISBN: 978-953-51-0344-8, InTech, Available from: http://www.intechopen.com/books/coronary-artery-disease-new-insights-and-novel-approaches/gene-polymorphism-in-coronary-heart-disease-

INTECH
open science | open minds

InTech Europe
University Campus STeP Ri
Slavka Krautzeka 83/A
51000 Rijeka, Croatia
Phone: +385 (51) 770 447
Fax: +385 (51) 686 166
www.intechopen.com

InTech China
Unit 405, Office Block, Hotel Equatorial Shanghai
No.65, Yan An Road (West), Shanghai, 200040, China
中国上海市延安西路65号上海国际贵都大饭店办公楼405单元
Phone: +86-21-62489820
Fax: +86-21-62489821

IntechOpen