Plaque Rupture and Stabilization in Coronary Artery: Current Concepts and Therapeutic Implications

ALBERT WAI-SUEN LEUNG

From Division of Cardiology, Department of Medicine and Geriatrics, Princess Margaret Hospital, Lai Chi Kok, Kowloon, Hong Kong

LEUNG: ***Plaque Rupture and Stabilization in Coronary Artery: Current Concepts and Therapeutic Implications.*** *Rupture of an atherosclerotic plaque has become the most important mechanism leading to acute coronary syndrome. A plaque that is prone to rupture has certain characteristic structural, cellular and molecular features, all of which indicate a state of active inflammation within the plaque. The vulnerability of a plaque to rupturing is related to multiple factors including chronic inflammation or infection, oxidative stress, and mechanical and haemodynamic stresses. Once a plaque ruptures, thrombus formation occurs, which can be occlusive or non-occlusive, depending on the thrombogenic and haemodynamic factors. Plaque stabilization is a concept of how to prevent plaque rupture. Lipid-lowering drugs have been investigated and found to have various stabilizing effects including change of plaque composition, improvement in endothelial function, and benefit in thrombosis and fibrinolysis. Other therapies also play their own roles, but are less studied. Major clinical trials on lipid-lowering drugs, especially those published recently, have given important support to this concept. We are going to see in the near future further advances in the understanding, detection and treatment of plaque rupture. (J HK Coll Cardiol 2001;9:45-54)*

Atherosclerosis, acute coronary syndrome, endothelial dysfunction, lipid lowering, plaque rupture, plaque stabilization

摘要

動脈硬化斑塊的破裂是導致急性冠狀動脈綜合徵的最重要的機制。斑塊在破裂前有著其特定的結構上、細胞上乃至分子生物學上的性質，而這一切均預示著斑塊內急性炎症的發生。斑塊是否易於破裂與多種因素有關，包括慢性炎症或感染、氧化反應、以及機械和血流動力學的應激狀況。一旦斑塊破裂，血栓將逐步形成，依據血栓和血流動力學的特性可分為阻塞型與非阻塞型。斑塊穩定這一概念涉及如何防止斑塊的破裂。對降脂藥物的研究發現該類藥物有著多種穩定作用，如改變斑塊的構成，提高內皮細胞的作用，有助於溶栓和纖溶。其他的治療有著其各自的作用，但研究相對較少。圍繞降脂藥物的臨床試驗，尤其是近期頒佈的，對這概念提供了重要理論支援。我們期待著在不久的將來對斑塊破裂的理解、診察與治療有進一步的進展。

關鍵詞：動脈硬化　急性冠狀動脈綜合徵　內皮細胞失功能　降脂　斑塊破裂　斑塊穩定

Address for reprints: Dr. Albert Wai-Suen Leung
Department of Medicine and Geriatrics, Princess Margaret Hospital,
Lai Chi Kok, Kowloon, Hong Kong
Tel: (852) 2990 1111, Fax: (852) 2307 2270

Received December 21, 2000; revision accepted January 3, 2001

Introduction

Atherosclerosis is the most frequent cause underlying ischaemic heart disease. Atherosclerosis, by itself, however, is rarely fatal. It is the acute coronary syndrome, which is caused by acute thrombosis superimposed on an atherosclerotic plaque, that causes the potentially lethal consequences.[1,2] According to the traditional view, the pathogenesis of an acute coronary syndrome was related to the progression of the atherosclerosis in a coronary artery, until the lumen narrowed to the point where addition of a few platelets and a small thrombus clot could obstruct the artery. In other words, total obstruction was preceded by high-grade stenosis, and a stable plaque became unstable by being more obstructive. This concept had changed.

Several trials,[3-5] which studied the grade of stenosis of atherosclerotic plaques in coronary arteries in relation to subsequent myocardial infarctions, concluded that the severity of stenosis on angiography did not accurately predict the location of a subsequent coronary occlusion. For example, in 1988, Little and colleagues studied 42 consecutive patients who underwent coronary angiography before and up to a month after having an acute myocardial infarction.[4] They concluded that most of the infarctions resulted from coronary occlusions that had previously shown stenosis of less than 50% on angiography. Another support came from trials of lipid-lowering therapies aimed at regression of plaque studied angiographically.[6-8] The clinical benefit of these trials was disproportionately greater compared with the relatively trivial change in the degree of plaque stenosis. Thus, factors other than the reduction of the degree of stenosis may be more important in the process of stabilizing an otherwise active atherosclerotic plaque, whether it is of high grade stenosis or not.

With these studies emerged the concept of the vulnerable atherosclerotic plaque. In the majority of cases, coronary thrombosis takes place in a rupture of a fibrous cap overlaying a plaque with a lipid core. However, in up to 25% of cases, thrombosis may result from superficial erosion, in which a thrombus overlays on deendothelialized smooth muscle cell and proteoglycan-rich matrix, without a lipid core.[9-12] Although the prevalence of plaque erosion with superimposed thrombus is higher in victims of sudden death especially in smokers, the young, women, and diabetic populations,[11,12] the pathogenesis related to plaque erosion is still largely undetermined. The pathogenesis related to plaque rupture, on the other hand, is more established. Thus the discussion will be concentrated on how a plaque ruptures and why it becomes vulnerable. This process is complex and involves the interaction of many factors, which can be grouped into several headings, including plaque morphology, on-going inflammation, and triggering factors. This is depicted in a simplified flow-chart (Figure 1).

Plaque Morphology

Features of rupture-prone plaques are shown in Table 1. More than 70% of the average atherosclerotic plaque is composed of extracelluar matrix, whereas lipid forms a much smaller component. Plaques prone to rupture, however, have a much larger lipid core in general.[13] Davies and coworkers found a relation between size of lipid core and plaque disruption in aortic plaques in human and identified a critical threshold: intact aortic plaques containing a core occupying more than 40% of the plaque were considered particularly vulnerable and at high risk of rupture and thrombosis.[14] The nature of the lipid present in a plaque may also be a factor. Lipid in the form of cholesteryl ester softens the plaque, whereas crystalline cholesterol may have the opposite effect.[15] The fibrous cap is a critical structure; when intact, it prevents the exposure of the thrombogenic components of the plaque to circulating blood. Fibrous caps vary considerably in thickness and tend to be

Table 1. Features of rupture-prone plaques

Morphological
Large lipid-rich core
Thin fibrous cap
Biochemical
Reduced collagen content
Cholesteryl ester
Local chronic inflammation
Increased macrophage density and activity
T-lymphocyte accumulation near sites of rupture
Increased neovascularization
Reduced density of smooth muscle cells
Increased number and activity of mast cells
Expression of markers of inflammatory activation
Matrix metalloproteinase secretion
Increased tissue-factor expression

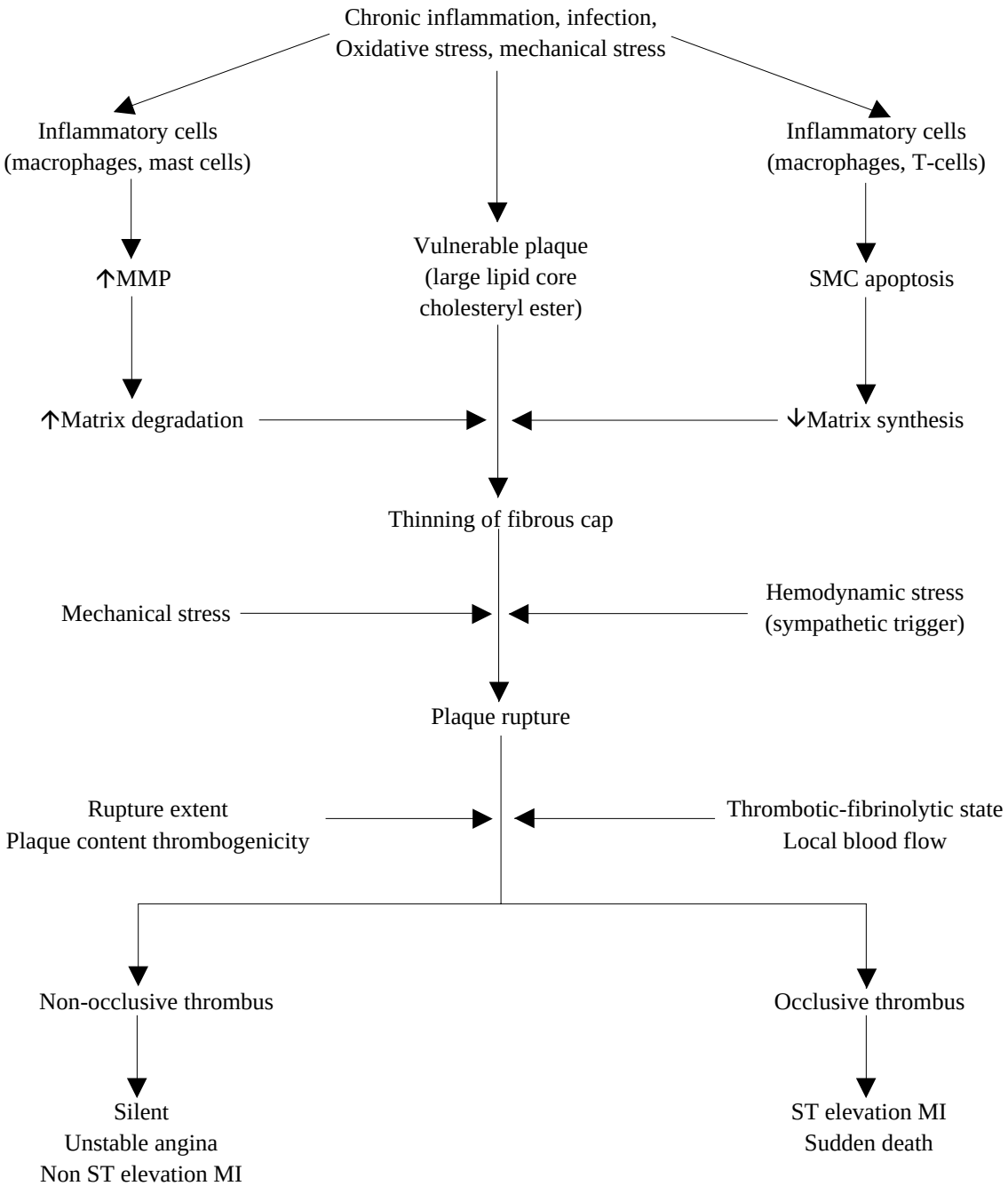

Figure 1. *A summary of the pathophysiololgy of plaque rupture and its consequences. MMP=matrix metalloproteinase; SMC=smooth muscle cell; MI=myocardial infarction*

thinnest at the shoulder regions, where most plaque ruptures tend to occur.[16]

Inflammation and Apoptosis

Ruptured plaques contain a greater number of inflammatory cells, often in the fibrous cap and around the lipid core, with a preferential concentration at the rupture-prone shoulder regions.[14,17-19] The inflammatory cells, such as activated macrophages, mast cells and T lymphocytes, will trigger production of cytokines and matrix-degrading proteins. The cytokines, such as interferon-γ, tumor necrosis factor and interleukin-1, are themselves pro-inflammatory and can trigger more cellular and molecular activities.[20] The matrix-degrading proteins are a family of structurally related proteases involved in the regulation of matrix turnover and are collectively termed as matrix metalloproteinases (MMPs).[21,22] Besides macrophages and T lymphocytes, other cells like smooth muscle cells can also trigger MMPs. The ultimate result is a weakening of the connective-tissue framework of the plaque, including the fibrous cap which is therefore predisposed to rupture. The MMP production is tightly regulated in part by co-secretion of tissue inhibitors of MMPs by smooth muscle cells, which can neutralize the effects of MMPs.[20]

Apart from matrix degradation, the thinning of fibrous cap is also related to paucity of matrix synthesis, a process performed by smooth muscle cells. This is supported by data showing a reduced density of smooth muscle cells in the ruptured fibrous cap.[23] The precise mechanism is unknown, but one possibility could be programmed cell death or apoptosis.[24,25] Apoptosis may be mediated by cytokines as produced by inflammatory cells.[20,26] In one example, interferon-γ produced by activated T-lymphocytes has been shown to inhibit smooth muscle cell collagen gene expression, which leads to a reduction of smooth muscle cell growth and proliferation.[26]

Triggers of Inflammation or Rupture

The process that leads to the initiation, growth, composition and rupture of an atherosclerotic plaque is a continuous and on-going complexity. Many of the factors that are associated with the trigger of rupture are also factors responsible for producing inflammation within an atherosclerotic plaque. A brief but integrated description is hereby given to introduce the possible factors that may either cause inflammation or trigger rupture, or both.

Chronic Inflammation and Infections

The precise triggers for inflammation in atherosclerosis remain to be defined. A number of inflammatory mediators, such as plasma fibrinogen, C-reactive protein (CRP), amyloid A protein, and interleukin-6, have been associated with a higher risk of acute coronary events.[27] Data from the Physicians Health Study indicate that baseline plasma CRP level can predict future coronary events.[28] Whether these acute-phase reactants have a direct vascular effect or are simply markers of systemic inflammation is not yet settled,[29] but there are more and more evidences towards the direct pro-inflammatory effect of CRP in the pathogenesis of atherosclerosis.[30-32]

Several infectious agents have gained increasing interest as potential provocateurs of plaque destabilization. Examples are Chlamydia pneumoniae, Helicobacter pylori, Herpes simplex virus and Cytomegalovirus.[33] Some of the associations may be invalidated because of a high prevalence of these infections in the general population. Of particular interest is C. pneumoniae infection, because the pathogen has been identified in atherosclerotic plaques,[34] and two small intervention studies with macrolide antibiotics reported reduced coronary events in treated patients.[35,36] Results from the big ACADEMIC trial, which studied the use of azithromycin in patients with coronary artery disease and positive titres to C. pneumoniae, however, were not convincing.[37]

Oxidative Stress

Endothelial dysfunction is an important concept that triggers the formation of atherosclerosis as well as the inflammation of the existing atherosclerotic plaques. A number of insults may be responsible for endothelial injury. One mechanism is by increasing oxidative stress, which relates to the appearance of reactive oxygen species or oxygen free radicals that fail to be neurtalized by nitric oxide or scavenged by antioxidants.[38] Hypercholesterolaemia, hypertension, hyperglycaemia, smoking and hyperhomocysteinaemia can all increase oxidative stress.[39-43] One method by which reactive

oxygen species exert their toxicity is by oxidation of low-density lipoprotein (LDL) to form oxidized LDL (oxLDL), a process called lipid peroxidation. OxLDL is taken up by macrophages to form lipid-laden macrophages, or foam cells, that contribute to plaque formation.[44] OxLDL (or oxidative stress) may also promote plaque inflammation and rupture by producing cytokines, enhancing thrombus formation, impairing fibrinolysis and promoting smooth muscle cell proliferation.[45] OxLDL has also been shown to be cytotoxic for and to promote apoptosis of smooth muscle cells.[46,47]

Mechanical and Haemodynamic Stresses

A variety of local mechanical and haemodynamic forces subject coronary plaques to different stresses. These include compressive pressure forces, tensile stretch forces, and tangential frictional (or shear) stresses. In particular, alterations of shear stress on arterial wall have a great impact on endothelial function and atherosclerosis. Shear stress of physiological magnitude is atheroprotective. Regions of disturbed flow with low or reversed shear stress lead to plaque progression.[48] High shear stress contributes significantly to plaque rupture.[49] The shear stress is largely determined by arterial geometry (branches and curvature), blood flow pulsatility and blood flow rates.[50] Once a plaque exists, it can alter the local shear stress and this perpetuates plaque growth or triggers plaque rupture in a self-amplifying manner.[50]

Shear stress may trigger plaque rupture spontaneously due to cap fatigue, a phenomenon analogous to metal fatigue, after plaques are subjected to repetitive bending, compression, stretching, flexion, or fluctuating pressure.[51,52] Rupture takes place at the point of greatest weakness (or greatest stress), which is usually the shoulder region of the fibrous cap.[53]

Plaque rupture may also be triggered by certain events. Half of patients with myocardial infarction report a trigger event, most often emotional stress or physical activity.[54] This may be due to a sudden rise in sympathetic activity with an increase in blood pressure, heart rate, force of cardiac contraction, or coronary blood flow, all of which can suddenly increase local mechanical stresses.[55] Burke and his colleagues studied 25 men with severe coronary artery disease who died during strenous activity or emotional stress as compared with 116 whose deaths occurred at rest.[11] Plaque rupture was found significantly more in men dying during exertion against those dying at rest.

Consequences of Rupture

Plaque rupture leads to various degrees of thrombus formation. Thrombosis may result in unstable angina, myocardial infarction, or sudden death. However, if the plaque rupture is minor, thrombus formation may be minimal, making the rupture clinically silent. Indeed, plaque disruption is a common event that often occurs during the development of atherosclerotic lesions and it is in the majority of cases clinically silent. In up to 8% of patients with coronary atherosclerosis who died of non-cardiovascular causes, a small, recent plaque disruption was found at autopsy.[56] The numbers increase to 22% in patients with diabetes or hypertension. The magnitude of thrombotic response also depends on the thrombogenicity of the exposed plaque components, local flow condition, and the systemic thrombotic-thrombolytic milieu at the time of plaque rupture.[57-59] Asymptomatic thrombus formation on a ruptured plaque may seal off the rupture and become part of the underlying plaque. This may be an important mechanism of plaque growth and may contribute to the evolution of mature plaques and the symptoms in chronic stable angina.[60]

Stabilization by Statins

From the angiographic trials using lipid-lowering agents,[6-8] the discordance between the substantial reduction in clinical events and the minor reduction in coronary stenosis warranted an explanation. This has led to the concept of plaque stabilization: by changing the biology of a plaque through risk-factor modification or drug intervention, the plaque is stabilized and its rupture can be avoided, and the process can reduce the incidence of clinical events. Among drugs with possible plaque-stabilizing effects, the statins are probably the most studied. Several mechanisms have been postulated, including change of plaque composition, improvement in endothelial function, and a favorable effect on thrombosis and fibrinolysis. The latter two mechanisms produce initial benefits, while effects on plaque composition is operative later.[61] These mechanisms are

discussed below. Most, but not all, of the beneficial effects of statins are probably related to their lipid lowering effect.[62]

Shift of Plaque Composition

The lipid content of atherosclerotic plaques can be altered by lipid lowering, probably at physical, cellular and biochemical levels. In the experimental model of atherosclerosis, lipid lowering results in disappearance of macrophages and foam cells from plaques, depletion of cholesterol ester with consequent reduction in the lipid core, and an increase in the content of mature collagen.[63-65] In primates with diet-induced atherosclerosis, the lipid content within plaques begins to decrease 6 months after the normalization of cholesterol values, and at 2 years, 60% of plaque cholesterol is depleted.[63] At the molecular level, a decrease in MMP activity is observed.[66] Lipid lowering therapy may thus decrease the formation of lipid-rich plaques and promote reduction of lipid content of pre-existing lipid-rich plaques.

Endothelial Function Improvement

As mentioned, high cholesterol is associated with endothelial dysfunction and atherosclerosis, which is attributed to oxidative stress and oxLDL. Statins have been shown to improve endothelium-dependent vasomotor function in experimental models, in uncontrolled clinical studies, and in randomized controlled trials.[67-70] Dupuis and colleagues demonstrated in a randomized controlled study that statins could improve endothelial function after 6 weeks of therapy.[70] Andrews and colleagues objectively demonstrated a decrease in myocardial ischemia during daily life (by ambulatory ECG monitoring of ST-segment depression episodes) as a result of cholesterol lowering with a statin compared with placebo.[71] The benefit appeared only after 4 to 6 months of therapy, antedating any structural evidence of atherosclerosis regression.

Thrombogenicity and Fibrinolysis

High cholesterol is associated with enhanced platelet reactivity as a result of several mechanisms, including lipid peroxidation, enhanced thromboxane production, and alterations of platelet cell membrane and cytosolic calcium.[72] The statins have been shown to decrease ADP-induced platelet aggregation, thromboxane B2 production, and cytosolic calcium in platelets.[73-75] Statins also reduce blood viscosity, and this may enhance more rapid blood flow across the plaque-laden arterial lumen.[76] Finally, statins may decrease plasminogen activator inhibitor type-1 production, which has anti-fibrinolytic effect.[77]

Stabilization by Other Therapies

Besides lipid-lowering agents, other interventions may have similar effects. Angiotensin-converting enzyme inhibitors, through their local tissue-specific effects, may reduce oxidative stress to the endothelium by increasing bradykinin and by up-regulation of expression of nitric oxide synthase.[78] Trials on quinapril, for example, have shown significant improvement in endothelial function.[79,80] Anti-oxidants, oestrogen replacement therapy, smoking cessation, avoidance of psychosocial stress, and blood pressure lowering have all been suggested.[81-84] Avoidance of trigger events such as strenuous exercise may prevent plaque rupture. Nevertheless, only a small fraction of all myocardial infarctions (about 5%) is related to vigorous exertion.[85] Moreover, regular exercise may retard plaque progression and seems to provide protection against myocardial infarction and coronary deaths, at least in part by eliminating the triggering effect of sudden vigorous exertion (exercise pre-conditioning).[86,87] With a better understanding of the molecular bases of vulnerable plaque, gene therapy to stabilize the plaque may be an exciting possibility. Possible strategies include over-expression of tissue inhibitors of MMP, anti-sense therapy to block pro-inflammatory molecules such as nuclear factor κB, and over-expression of nitric oxide synthase.[88,89]

Support by Major Clinical Trials

For many years, the management of coronary artery disease has been guided by the severity of stenosis, as assessed by coronary angiography. Severe stenoses are managed by balloon angioplasty or bypass surgery. This approach, however, cannot detect and treat those plaques which are not severely stenotic but which are

prone to rupture. The concept of plaque stabilization, on the other hand, is an attractive alternative in reducing the consequences of acute coronary events. The evidence mentioned so far can serve to provide supporting data only from laboratory or from small-scaled clinical studies with surrogate endpoints. What we need is rigorous validation of this concept in big clinical trials with major endpoints.

In the landmark studies of the statin drugs in primary and secondary preventions of coronary artery disease, a dramatic decrease in overall and cardiovascular mortality were shown.[90-92] However, analyses on coronary bypass surgery and percutaneous revascularization did not show the same benefit.[93,94] This is an indirect evidence of the benefit of lipid-lowering as compared to revascularization. The AVERT trial, although small in scale (341 patients), is the first to compare aggressive lipid-lowering therapy with mechanical revascularization in the management of chronic coronary artery disease.[95] The time to first ischaemic event was significantly delayed in the drug treatment arm and the separation was noted after 6 months of treatment. The exact mechanism causing such a difference is unknown. The effect of lipid-lowering on plaque stabilization, however, may play a major role.

Another trial of aggressive lipid-lowering therapy, the MIRACL trial, was designed for acute rather than chronic cases.[96] The goal is to determine whether early, rapid, and profound cholesterol lowering therapy can reduce early recurrent ischaemic events in patients with acute coronary syndrome. Results were presented recently.[97] 3,086 patients received either atorvastatin 80mg daily or placebo for 16 weeks, initiated between 24 to 96 hours of an acute coronary syndrome. Those treated with atorvastatin experienced a significant reduction in the risk of the primary combined endpoint. This is the first trial to demonstrate that the clinical benefit of aggressive lipid-lowering therapy can be achieved within 4 months following an acute coronary event. This is a major step to solidify the concept of plaque stabilization into clinical benefit. The rapidity of this benefit means that the stabilizing effect on the rupture-prone plaques may have taken place through the improvements in endothelial function and blood coagulability.

The Coming Future

Our understanding of plaque biology and the triggering of plaque rupture has increased dramatically in the past decade. This knowledge is now being tested in clinical practice and the results are promising. The use of aggressive medical management to stabilize an unstable plaque may become an established alternative way of treating acute coronary syndrome, or it may lessen the need for invasive procedures in some patients. More data, however, is necessary to establish the direct relationship between plaque stabilization and improvement in clinical outcome. There are also studies directed at developing new strategies to stabilize vulnerable plaques. The promise of molecular and genetic therapies in this area is considerable.[88-89] The combination of plaque stabilization and other treatment modalities (such as new anti-thrombotics and anti-platelet agents) may be another possible goal. Finally, a search for new methods to easily and reliably identify vulnerable plaques is necessary.[98-101] Once these plaques are identified, pharmacologic and molecular therapies may allow their stabilization.

References

1. Falk E. Coronary thrombosis: Pathogenesis and clinical manifestations. Am J Cardiol 1991;68(Suppl B):28B-35B.
2. Fuster V, Badimon L, Badimon J, et al. The pathogenesis of coronary artery disease and the acute coronary syndromes. N Eng J Med 1992;326:242-50.
3. Brown BG, Gallery CA, Badger RS, et al. Incomplete lysis of thrombus in the moderate underlying atherosclerotic lesion during intracoronary infusion of streptokinase for acute myocardial infarction: quantitative angiographic observations. Circulation 1986;73:653-61.
4. Little WC, Constantinescu M, Applegate RJ, et al. Can coronary angiography predict the site of a subsequent myocardial infarction in patients with mild-to-moderate coronary artery disease? Circulation 1988;78:1157-66.
5. Ambrose JA, Tannenbaum MA, Alexopoulos D, et al. Angiographic progression of coronary artery disease and the development of myocardial infarction. J Am Coll Cardiol 1988; 12:56-62.
6. Brown BG, Zhao X-Q, Sacco DE, et al. Lipid lowering and plaque regression: New insights into prevention of plaque disruption and clinical events in coronary disease. Circulation 1993;87:1781-91.

7. MAAS Investigators. Effect of simvastatin on coronary atheroma: The Multicentre Anti-Atheroma Study (MAAS). Lancet 1994;344:633-8.
8. Superko HR, Krauss RM. Coronary artery disease regression: Convincing evidence for the benefit of aggressive lipoprotein management. Circulation 1994;90:1056-69.
9. Davies MJ. A macro and micro view of coronary vascular insult in ischemic heart disease. Circulation 1990;82(Suppl 3):1138-46.
10. Arbustini E, Burke A, dal Bello B, et al. Plaque erosion is a major substrate for coronary thrombosis in acute myocardial infarction (abstract). J Am Coll Cardiol 1998;31:379A.
11. Burke AP, Farb A, Malcom GT, et al. Coronary risk factors and plaque morphology in men with coronary disease who died suddenly. N Engl J Med 1997;336: 1276-82.
12. Burke AP, Farb A, Malcom GT, et al. Effect of risk factors on the mechanism of acute thrombosis and sudden coronary death in women. Circulation 1998;97:2110-6.
13. Gertz SD, Roberts WC. Hemodynamic shear force in rupture of coronary arterial atherosclerotic plaques. Am J Cardiol 1990; 66:1368-72.
14. Davies MJ, Richardson PD, Woolf N, et al. Risk of thrombosis in human atherosclerotic plaques: role of extracellular lipid, macrophage, and smooth muscle cell content. Br Heart J 1993; 69:377-81.
15. Loree HM, Toblas BJ, Gibson LJ, et al. Mechanical properties of model atherosclerotic lesion lipid pools. Arterioscler Thromb 1994;14:230-4.
16. Richardson PD, Davies MJ, Born GVR. Influence of plaque configuration and stress distribution on fissuring of coronary atherosclerotic plaques. Lancet 1989;2:941-4.
17. Kovanen PT, Kaartinen M, Paavonen T. Infiltrates of activated mast cells at the site of coronary atheromatous erosion or rupture in myocardial infarction. Circulation 1995;92:1084-8.
18. Moreno PR, Falk E, Palacios IF, et al. Macrophages infiltration in acute coronary syndromes: implications for plaque rupture. Circulation 1994;90:775-8.
19. Van der Wal AC, Becker AE, van der Loos CM, et al. Site of intimal rupture or erosion of thrombosed coronary atherosclerotic plaques is characterized by an inflammatory process irrespective of the dominant plaque morphology. Circulation 1994;89:36-44.
20. Libby P. Molecular bases of the acute coronary syndromes. Circulation 1995;91:2844-50.
21. Galis ZS, Sukhova GK, Lark MW, et al. Increased expression of matrix metalloproteinases and matrix degrading activity in vulnerable regions of human atherosclerotic plaques. J Clin Invest 1994;94:2493-503.
22. Dollery CM, McEwan JR, Henney AM. Matrix metalloproteinases and cardiovascular disease. Circ Res 1995; 77:863-8.
23. Burleigh MC, Briggs AD, Lendon CL, et al. Collagen type I and III, collalgen content, GAGs and mechanical strength of human atherosclerotic plaque caps: Span-wise variations. Atherosclerosis 1992;96:71-81.
24. Majno G, Joris I. Apoptosis, oncosis and necrosis: An overview of cell death. Am J Pathol 1995;146:3-15.
25. Geng YJ, Libby P. Evidence for apoptosis in advanced human atheroma. Am J Pathol 1995;147:251-66.
26. Wallner K, Shah PK, Fishbein MC, et al. Expression of tenascin-C in human coronary atherosclerotic plaque. Circulation 1999;99:1284-9.
27. Maseri A, Biasucci LM, Liuzzo G. Inflammation in ischaemic heart disease. BMJ 1996;312:1049.
28. Ridker PM, Cushman M, Stampfer MJ, et al. Inflammation, aspirin, and the risk of cardiovascular disease in apparently healthy men. N Engl J Med 1997;336:973-9.
29. Ridker PM. Inflammation, infection, and cardiovascular risk: How good is the clinical evidence? [editorial]. Circulation 1998; 97:1671-4.
30. Shah PK. Circulating markers of inflammation for vascular risk prediction: Are they ready for prime time? Circulation 2000;101:1758-9.
31. Pasceri V, Willerson JT, Yeh ET. Direct proinflammatory effect of C-reactive protein on human endothelial cells. Circulation 2000;102:2165-8.
32. Lagrand WK, Visser CA, Hermens WT, et al. C-reactive protein as a cardiovascular risk factor: more than an epiphenomenon? Circulation 1999;100:96-102.
33. Kol A, Libby P. The mechanisms by which infectious agents may contribute to atherosclerosis and its clinical manifestations. Trends Cardiovasc Med 1998;8:191-8.
34. Muhlestein JB, Hammond EH, Carlquiest JF, et al. Increased incidence of Chlamydia species within the coronary arteries of patients with symptomatic atherosclerotic versus other forms of cardiovascular disease. J Am Coll Cardiol 1996;27:1555-61.
35. Gupta S, Leatham EW, Carrington D. Elevated chlamydia pneumoniae antibodies, cardiovascular events, and azithromycin in male survivors of myocardial infarction. Circulation 1997;96:404-7.
36. Gurfinkel E, Bozovich G, Daroca A, ROXIS Study Group. Randomized trial of roxithromycin in non-Q wave coronary syndrome: ROXIS pilot study. Lancet 1997;350:404-7.
37. Muhlestein JB, Anderson JL, Carlquist JF, et al. Randomized secondary prevention trial of azithromycin in patients with coronary artery disease: Primary clinical results of the ACADEMIC study. Circulation 2000;102:1755-60.
38. Rubbo H, Radi R, Trujillo M, et al. Nitric oxide regulation of superoxide and peroxynitrite-dependent lipid peroxidation. Formation of novel nitrogen-containing oxidized lipid derivatives. J Biol Chem 1994;269:26066-75.
39. Davi G, Alessandrini P, Mezzetti A, et al. In vivo formation of 8-Epi-prostaglandin F2 alpha is increased in hypercholesterolemia. Arterioscler Thromb Vasc Biol 1997; 17:3232-5.
40. Lacy F, O'Connor DT, Schmid-Schonbein GW. Plasma hydrogen peroxide production in hypertensives and normotensive subjects at genetic risk of hypertension. J Hypertens 1998;16:291-303.
41. McCance DR, Dyer DG, Dunn JA, et al. Mallard reaction products and their relation to complications in insulin-dependent diabetes mellitus. J Clin Invest 1993;91:2470-8.
42. Reilly M, Delanty N, Lawson JA, et al. Modulation of oxidant stress in vivo in chronic cigarette smokers. Circulation 1997; 96:69-75.
43. Voutilainen S, Morrow JD, Roberts LJ, et al. Enhanced in vivo lipid peroxidation at elevated plasma total homocysteine levels.

Arterioscler Thromb Vasc Biol 1999;19:1263-6.
44. Henriksen T, Mahoney EM, Steinberg D. Interactions of plasma lipoproteins with endothelial cells. Ann N Y Acad Sci 1982; 401:102-16.
45. Forgione MA, Leopold JA, Loscalzo J. Roles of endothelial dysfunction in coronary artery disease. Curr Opin Cardiol 2000; 15:409-15.
46. Okura Y, Brink M, Itabe H, et al. Oxidized low-density lipoprotein is associated with apoptosis of vascular smooth muscle cells in human atherosclerotic plaques. Circulation 2000;102:2680-6.
47. Bjorkerud B, Bjorkerud S. Contrary effects of lightly and strongly oxidized LDL with potent promotion of growth versus apoptosis on arterial smooth muscle cells, macrophages, and fibroblasts. Arterioscler Thromb Vasc Biol 1996;16:416-24.
48. Malek AM, Alper SL, Izumo S. Hemodynamic shear stress and its role in atherosclerosis. JAMA 1999;282:2035-42.
49. Gertz SD, Roberts WC. Hemodynamic shear force in rupture of coronary arterial atherosclerotic plaques. Am J Cardiol 1990; 66:1368-72.
50. Feldman CL, Stone PH. Intravascular hemodynamic factors responsible for progression of coronary atherosclerosis and development of vulnerable plaque. Curr Opin Cardiol 2000; 15:430-40.
51. MacIsaac AI, Thomas JD, Topol EJ. Toward the quiescent coronary plaque [review article]. J Am Coll Cardiol 1993;22: 1228-41.
52. Lee RT, Kamm RD. Vascular mechanics for the cardiologist. J Am Coll Cardiol 1994;23:1289-95.
53. Cheng GC, Loree HM, Kamm RD, et al. Distribution of circumferential stress in ruptured and stable atherosclerotic lesions: A structural analysis with histo-pathological correlation. Circulation 1993;87:1179-87.
54. Tofler GH, Stone PH, Maclure M, et al. Analysis of possible triggers of acute myocardial infarction (the MILIS study). Am J Cardiol 1990;66:22-7.
55. Muller JE, Tofler GH, Stone PH. Circadian variation and triggers of onset of acute cardiovascular disease. Circulation 1989;79:733-43.
56. Davies M, Bland JM, Hangartner JR, et al. Factors influencing the presence or absence of acute coronary artery thrombi in sudden ischaemic death. Eur Heart J 1989;10:203-8.
57. Fernandez-Ortiz A, Badimon J, Falk E, et al. Characterization of the relative thrombogenicity of atherosclerotic plaque components: Implications for consequences of plaque rupture. J Am Coll Cardiol 1994;23:1562-9.
58. Fuster V, Lewis A. Conner Memorial Lecture: Mechanisms leading to myocardial infarction: Insights from studies of vascular biology. Circulation 1994:90:2126-46.
59. Meade TW, Ruddock V, Stirling Y, et al. Fibrinolytic activity, clotting factors, and long-term incidence of ischaemic heart disease in the Northwick Park Heart Study. Lancet 1993;342: 1076-9.
60. Edwards WD. Atherosclerotic plaques: Natural and unnatural history. Monogr Pathol 1995;37:12-46.
61. Vaughan CJ, Murphy MB, Buckley BM. Statins do more than just lower cholesterol. Lancet 1996;348:1079-82.
62. Rosenson RS, Tangney CC. Antiatherothrombotic properties of statins: Implications for cardiovascular event reduction. JAMA 1998;279:1643-50.
63. Amstrong ML, Megan MB. Lipid depletion in atheromatous coronary arteries in rhesus monkeys after regression diets. Circ Res 1972;30:675-80.
64. DePalma RG, Klein L, Bellon EM, et al. Regression of atherosclerotic plaques in rhesus monkeys: Angiographic, morphologic, and angiochemical changes. Arch Surg 1980: 115: 1268-78.
65. Kaplan JR, Manuck SB, Adams MR, et al. Plaque changes and arterial enlargement in atherosclerotic monkeys after manipulation of diet and social environment. Arterioscler Thromb 1993;13:254-63.
66. Aikawa M, Rabkin E, Okada Y, et al. Lipid lowering by diet reduces matrix metalloproteinase activity and increases collagen content of rabbit atheroma: a potential mechanism of lesion stabilization. Circulation 1998;97:2433-44.
67. Anderson T, Meredith I, Yeung A, et al. The effect of cholesterol lowering and antioxidant therapy on endothelial-dependent coronary vasomotion. N Engl J Med 1995;332:488-93.
68. Leung WH, Lau CP, Wong CK. Beneficial effect of cholesterol lowering therapy on coronary endothelium dependent relaxation in hypercholesterolemic patients. Lancet 1993;341: 1496-500.
69. Treasure C, Klein J, Weintraub W, et al. Beneficial effects of cholesterol lowering therapy on the coronary endothelium in patients with coronary artery disease. N Engl J Med 1995;332: 481-7.
70. Dupuis J, Tardif JC, Cernacek P, et al. Cholesterol reduction rapidly improves endothelial function after acute coronary syndromes (the RECIFE trial). Circulation 1999;99: 3227-33.
71. Andrews TC, Raby K, Barry J, et al. Effect of cholesterol reduction on myocardial ischemia in patients with coronary disease. Circulation 1997;95:324-8.
72. Badimon JJ, Badinon L, Turitto VT, et al. Platelet deposition at high shear rates is enhanced by high plasma cholesterol levels. In vivo study in the rabbit model. Arterioscler Thromb 1991;11:395-402.
73. Mayer J, Eller T, Brauer P, et al. Effects of long-term treatment with lovastatin on the clotting system and blood platelets. Ann Hematol 1992;64:196-201.
74. Notarbartolo A, Davi G, Averna M, et al. Inhibition of thromboxane biosynthesis and platelet function by simvastatin in type IIa hypercholesterolemia. Arterioscler Thromb Vasc Biol 1995;15:247-51.
75. Le Quan Sang KH, Levenson J, Megnien JL, et al. Platelet cytosolic Ca2+ and membrane dynamics in patients with primary hypercholesterolemia. Effects of pravastatin. Arterioscler Thromb Vasc Biol 1995;15:759-64.
76. Jay RH, Rampling MW, Betteridge DJ. Abnormalities of blood rheology in familial hypercholesterolaemia. Effects of treatment. Atherosclerosis 1990;85:249-56.
77. Wada H, Mori Y, Kaneko T, et al. Hypercoagulable state in patients with hypercholesterolemia: effects of pravastatin. Clin Ther 1992;14:829-34.
78. Koh KK, Bui MN, Hathaway L, et al. Mechanism by which quinapril improves vascular function in coronary artery disease. Am J Cardiol 1999;83:327-31.
79. Mancini GB, Henry GC, Macaya C, et al. Angiotensin-

converting enzyme inhibition with quinapril improves endothelial vasomotor dysfunction in patients with coronary artery disease. The TREND (Trial on Reversing Endothelial Dysfunction) Study. Circulation 1996;94:258-65.
80. Anderson TJ, Elstein E, Haber H, et al. Comparative study of ACE-inhibition, angiotensin II antagonism, and calcium channel blockade on flow-mediated vasodilation in patients with coronary disease (BANFF study). J Am Coll Cardiol 2000; 35:60-6.
81. Simon BC, Haudenschild CC, Cohen RA. Preservation of endothelium dependent relaxation in atherosclerotic rabbit aorta by probucol. J Cardiovasc Pharmacol 1993;21:893-901.
82. Chobanian AV, Haudenschild CC, Nickerson C, et al. Antiatherogenic effect of captopril in the Watanabe heritable hyperlipidemic rabbit. Hypertension 1990;15:327-31.
83. Williams JK, Adams MR, Herrington DM, et al. Short-term administration of estrogen and vascular responses of atherosclerotic coronary arteries. J Am Coll Cardiol 1992;20: 452-7.
84. Shively CA, Clarkson TB. Social status and coronary artery atherosclerosis in female monkeys. Arterioscler Thromb 1994; 14:721-6.
85. Hambrecht R, Niebauer J, Marburger C, et al. Various intensities of leisure time physical activity in patients with coronary artery disease: Effects on cardiorespiratory fitness and progression of coronary atherosclerotic lesions. J Am Coll Cardiol 1993; 22:468-77.
86. Mittleman MA, Maclure M, Tofler GH, et al. Triggering of acute myocardial infarction by heavy physical exertion: Protection against triggering by regular exertion. N Engl J Med 1993;329:1677-83.
87. Paggenbarger RS Jr, Hyde RT, Wing AL, et al. The association of changes in physical-activity level and other lifestyle characteristics with mortality among men. N Engl J Med 1993; 328:538-45.
88. Kullo IJ, Mozes G, Schwartz RS, et al. Adventitial gene transfer of recombinant endothelial nitric oxide synthase to rabbit carotid arteries alters vascular reactivity. Circulation 1997;96: 2254-61.
89. Kullo IJ, Mozes G, Schwartz RS, et al. Enhanced endothelium-dependent relaxations after gene transfer of recombinant endothelial nitric oxide synthase to rabbit carotid arteries. Hypertension 1997;30:314-20.
90. Shepherd J, Cobbe SM, Ford I, et al. Prevention of coronary heart disease with pravastatin in men with hypercholesterolemia. West of Scotland Coronary Prevention Study Group. N Engl J Med 1995;333:1301-7.
91. The 4S Investigators. Randomised trial of cholesterol lowering in 4444 patients with coronary heart disease: The Scandinavian Simvastatin Survival Study (4S). Lancet 1994;344:1383-9.
92. Sacks FM, Pfeffer MA, Moye LA, et al. The effect of pravastatin on coronary events after myocardial infarction in patients with average cholesterol levels. Cholesterol and Recurrent Events Trial investigators. N Engl J Med 1996;335: 1001-9.
93. Yusuf S, Zucker D, Peduzzi P, et al. Effect of coronary artery bypass graft surgery on survival: Overview of 10-year results from randomized trials by the Coronary Artery Bypass Graft Surgery Trialists Collaboration. Lancet 1994;344:563-70.
94. Coronary angioplasty versus medical therapy for angina: the second Randomized intervention Treatment of Angina (RITA-2) trial. RITA-2 trial participants. Lancet 1997;350: 461-8.
95. Pitt B, Waters D, Brown WV, et al. Aggressive lipid-lowering therapy compared with angioplasty in stable coronary artery disease. Atorvastatin Versus Revascularization Treatment investigators. N Engl J Med 1999;341:70-6.
96. Schwartz GG, Oliver MF, Ezekowitz MD, et al. Rationale and design of the Myocardial Ischemia Reduction with Aggressive Cholesterol Lowering (MIRACL) Study that evaluates atorvastatin in unstable angina pectoris and in non-Q-wave acute myocardial infarction. Am J Cardiol 1998;81:578-81.
97. Schwartz GG, Olsson AG, Ezekowitz MD, et al. The Myocardial Ischemia Reduction with Aggressive Cholesterol Lowering (MIRACL) trial: Effects of intensive atorvastatin treatment on early recurrent events after an acute coronary syndrome. Circulation 2000;102:-f2672.
98. Ilegbusi OJ, Hu Z, Nesto R, et al. Determination of blood flow and endothelial shear stress in human coronary artery in vivo. J Invasive Cardiol 1999;11:667-74.
99. Casscells W, Hathorn B, David M, et al. Thermal detection of cellular infiltrates in living atherosclerotic plaques: possible implication for plaque rupture and thrombosis. Lancet 1996; 347:1447-51.
100. Fujimoto JG, Boppart SA, Terrney GJ, et al. High resolution in vivo intra-arterial image with optical coherence tomography. Heart 1999;82:128-33.
101. Charash WE, Lodder RA, Moreno PR, et al. Detection of simulated vulnerable plaque using a novel near infrared spectroscopy catheter. J Am Coll Cardiol 2000;35(Suppl A):38A.