Methods of detecting atherosclerosis in non-cardiac surgical patients; the role of biochemical markers

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Atherosclerosis is a common condition in both the developed and developing world and is now recognised to be an inflammatory condition leading to the development of ischaemic heart disease, cerebrovascular disease and peripheral vascular disease. Ischaemic heart disease is a major risk factor in the pathogenesis of perioperative adverse cardiovascular events which lead to significant morbidity and mortality within the high risk surgical patient population. Current methods of evaluating the likelihood of postoperative cardiovascular complications depend largely on risk scoring systems, and the preoperative assessment of the functional status of the cardiovascular system. However, the possible role of inflammation in the generation of atherosclerosis has led to the identification of several biochemical markers such as acute phase proteins, cellular adhesion molecules and cytokines. An alternative approach therefore is the measurement of preoperative levels of these biomarkers with the aim of assessing pre-existing disease activity. This review summarises the pathophysiology of atherosclerosis and perioperative myocardial infarction, and discusses the possible future role of biomarkers in the risk stratification of patients undergoing non-cardiac surgery.

Adverse cardiovascular events (including myocardial ischaemia and infarction) are a significant cause of major morbidity and mortality in the perioperative period and have considerable economic consequences to the health service. Data from the USA show that approximately 27 million patients undergo surgery of which 8 million have known coronary artery disease or risk factors for cardiovascular disease. There are 1 million perioperative cardiac complications, suggesting that the overall risk of a perioperative cardiovascular event is 1 in 27. Half a million of these are perioperative myocardial infarctions (MI). The rate of perioperative MI in males more than the age of 50 years is 0.7%, but this incidence increases to 3.1% after vascular surgery. Figures for the UK suggest a comparable incidence of approximately 8000 perioperative cardiovascular deaths per year from roughly 5 million general or regional anaesthetics performed. A major predisposing factor in the development of a perioperative cardiovascular event is the presence of ischaemic heart disease, whether diagnosed or previously unknown. Atherosclerosis is the main pathological disorder responsible for the development of ischaemic heart disease. Therefore, identifying patients at risk before operation is sensible, but only if the clinician can use the information to modify perioperative management and reduce complication rates. There are now therapies that are of possible benefit in reducing the incidence of cardiovascular events, and accurately predicting those at risk would allow these therapies to be appropriately targeted. These include beta-adrenoceptor blockade, \( \alpha_2 \)-adrenoceptor agonists and statin therapy. Although regional anaesthetic techniques attenuate the surgical stress response they do not completely abolish it, and have not been shown to alter the incidence of perioperative cardiovascular events. Further factors that may be affected by accurate risk stratification include the most appropriate location of surgery, possible modification of the original planned operation, sequenced or staged surgery and the location and level of post-surgical care.

Atherosclerosis

Atherosclerosis affects the endothelial lining of arterial blood vessels, resulting in atheromatous plaque formation. The consequences of atherosclerosis include increased...
stiffness and a loss of elasticity in the blood vessel, stenosis of the artery, plaque rupture and aneurysm formation. Atherosclerosis is now recognized as an inflammatory process, with many known risk factors (Fig. 1). There is some evidence that infection may have a role in the aetiology with herpes viruses (Cytomegalovirus, Epstein-Barr virus and Herpes simplex-1 virus) and certain bacterial (Chlamydia pneumoniae and Helicobacter pylori) DNA having been detected in atherosclerotic plaques.

Within an affected blood vessel, characteristic alterations of blood flow occur resulting in increased turbulence and decreased shear stress leading to endothelial changes. These early changes precede the formation of atherosclerotic lesions and are responsible for endothelial cell dysfunction. Many factors are involved in the atherogenic process (Fig. 2). Increased permeability to lipoproteins and other plasma constituents is mediated by increased concentrations of nitric oxide, prostacyclin, platelet-derived growth factor, angiotensin II and endothelin. A separate process (the attraction, rolling, adherence and migration of monocytes and T-cells to the arterial wall) is mediated by factors including the cell adhesion molecules, oxidized low-density lipoprotein (LDL), cytokines and chemokines. Migrated monocytes are transformed into tissue macrophages and ingest lipid deposits to form ‘foam cells’. The earliest visible evidence of the atherosclerotic lesion is a fatty streak consisting of foam cells and activated T lymphocytes. More advanced atherosclerotic lesions contain smooth muscle cells which form a fibrous cap walling the lesion off from the vessel lumen. This is a protective response covering the inflammatory core of leukocytes, lipid and debris beneath, which may be necrotic. Atherosclerotic lesions expand at their shoulders by continued leukocyte adhesion and entry. Platelets adhere to dysfunctional endothelium, exposed collagen and macrophages becoming activated and releasing cytokines and growth factors that together with thrombin contribute to the migration and proliferation of monocytes and smooth muscle cells. Erosion and rupture of the plaque with consequent exposure of thrombogenic material can lead to unstable coronary syndromes or MI. The different stages in the development of an atherosclerotic plaque can be seen in Figure 3.

The pathophysiology of perioperative myocardial infarction
There are two mechanisms involved in the causation of perioperative MI.  
(i) Coronary artery occlusion: Plaque erosion or rupture leading to thrombogenesis and consequent occlusion or thromboembolic occlusion of an already narrowed coronary lumen.
(ii) Prolonged ischaemia (usually silent) secondary to an imbalance between myocardial oxygen demand and supply. The first type resembles acute MI in the non-surgical setting and is related to the concept of the ‘vulnerable plaque’. These plaques tend to be the newer, less stable coronary atherosclerotic lesions that have undergone rapid progression and contain a substantial lipid core filled with a large mass of thrombogenic lipids and macrophages along with various cytokines covered by a relatively thin fibrous cap. This protective cap undergoes constant inflammation and repair, and the balance between these two processes enables the plaque to expand in size but during this process the plaque is liable to fissure and rupture. Figure 4 shows some of the histological features of a vulnerable plaque. The older, more stable plaques have uniformly dense protective fibrous caps with a small lipid core and are therefore less likely to rupture. In the non-surgical setting, there is evidence that small, non-occlusive plaques rather than the large plaques contribute more to cardiovascular morbidity and mortality. These vulnerable plaques can be difficult to diagnose as they are not highly occlusive with angiography being of limited value in identifying them.\textsuperscript{17}

The second type of perioperative MI occurs most commonly in patients with severe but stable coronary artery disease, and is usually associated with prolonged silent postoperative ischaemia.\textsuperscript{40} This is thought to be the result of an imbalance between a limited myocardial oxygen supply and an increased perioperative oxygen demand. As these patients may have severe coronary artery disease, preoperative angiography can be useful in identifying those with highly occlusive coronary stenosis. Unlike the plaque rupture type of MI, the higher the grade of occlusion that a coronary stenosis causes, the more likely a prolonged stress-induced ischaemia type of MI is to occur. Therefore, identification of patients with high grade coronary artery stenosis is useful in allowing targeted interventions to minimize individual risk.

Intensive monitoring of myocardial damage by biomarkers has added to the evidence that two distinct pathophysiological mechanisms of perioperative MI exist. The first type of infarction occurring in the postoperative period is not preceded by ischaemic myocardial damage, is associated with a sudden increase in the serum troponin concentration to a level diagnostic of MI, and is probably because of coronary occlusion secondary to plaque haemorrhage, rupture or thrombus formation. The later or delayed type of perioperative MI is preceded by a long period, >24 h, of ischaemic myocardial damage observed as a moderate increase in the troponin level, not initially in the range diagnostic of MI but above the upper reference limit of normal.\textsuperscript{42} Pathological studies examining the coronary vessels at autopsy of patients who have suffered fatal perioperative MI shows that the incidence of these two types of MI is roughly equal.\textsuperscript{14 17}

As the pathophysiological mechanisms involved in these two types of perioperative MI are quite different, it follows that the tests to identify high-risk patients and treatment options available may also be different.

Predicting perioperative cardiovascular events
The adverse cardiovascular events that occur in the perioperative period are not limited to MI; acute coronary syndromes, congestive cardiac failure, arrhythmias and cerebrovascular accidents can all cause major morbidity and mortality. There are guidelines published on the recommended preoperative risk stratification of patients,\textsuperscript{5 12 21 22} and there is extensive literature studying the various different preoperative investigations that are available. Table 1 lists these different preoperative tests.

### Biochemical markers of cardiovascular disease
A more recent and alternative approach has been measurement of biomarkers of cardiovascular disease in the blood before operation and after operation, and correlating the levels with the incidence of cardiovascular adverse events.

#### (1) Traditional biomarkers
Traditionally biochemical markers of cardiovascular disease risk have measured myocardial cell damage and include

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<th>Table 1</th>
<th>The different preoperative tests useful for predicting perioperative cardiovascular events or the need for coronary revascularization before non-cardiac surgery. *AECG, ambulatory electrocardiogram. †MUGA, multiple gated acquisition scan; §LVEF, left ventricular ejection fraction</th>
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<tbody>
<tr>
<td>Preoperative investigation</td>
<td>At rest ECG&lt;br&gt;Holter (AECG)* monitoring&lt;br&gt;Exercise ECG&lt;br&gt;Radionucleotide ventriculography (MUGA)*&lt;br&gt;Stress myocardial perfusion imaging&lt;br&gt;Resting echo §LVEF&lt;br&gt;Pharmacological stress echo&lt;br&gt;Anaerobic threshold</td>
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Fig 4 Drawing of blood vessel containing unstable, complicated atheromatous plaque.
creatinine kinase, aspartate aminotransferase and lactate dehydrogenase. However, these tests have been largely superseded by measurement of troponin I or T concentrations, which are more specific for myocardial cell injury.

(a) Creatine kinase (CK)
This enzyme is expressed in many body tissues and catalyses the conversion of creatine to phosphocreatine. The enzyme has three different isoenzymes with different tissue distributions. The CK-MB isoenzyme is present in the highest concentrations within the myocardium, but it is not completely specific for myocardial tissue, as it is also found in skeletal muscle. To improve specificity, the ratio of plasma CK-MB to total CK can be used to assess whether the source is likely to be myocardial cell damage. In patients presenting with MI, concentrations of the enzyme begin to increase 4–6 h after the onset of chest pain, peak at 12–24 h and return to baseline within 48–72 h.

(b) Aspartate aminotransferase (AST)
This enzyme is distributed widely in the body, being found in red blood cells, liver, pancreas, myocardium, muscle and kidney, catalysing the reversible transfer of an amino group from aspartate to α-ketoglutarate to form glutamate and oxaloacetate. About 6–10 h after a MI, the serum concentration begins to increase, peaking at 12–48 h and returning to normal after 3–4 days. It has low specificity for cardiac disease and is therefore not used for this purpose.

(c) Lactate dehydrogenase (LDH)
This widely distributed enzyme catalyses the interconversion of lactate and pyruvate. Five isoenzyme forms exist including LDH-1, which is more common in heart muscle and red blood cells. Plasma concentrations start to increase 12–24 h after a MI, reaching a peak within 2–3 days, but taking as long as 14 days to return to baseline. Under normal conditions the concentration of isoenzyme LDH-2 is greater than that of LDH-1, but in MI this pattern is reversed.

(d) Troponins
These proteins are involved in the calcium interaction necessary for muscle contraction. Three subunits of the troponin protein can be found: -I, -T and -C, and a variety of tissue specific subtypes exist. However troponin-I and -T are structural components of cardiac muscle and are consequently highly specific for myocardial tissue. Elevated plasma concentrations are detected 3–12 h after myocardial cell damage (in a similar time frame as CK-MB) but the concentrations remain elevated for longer (5–9 days for troponin-I and up to 14 days for troponin-T).

An increase in cardiac troponin proteins perioperatively has been found to be useful in the prediction of cardiovascular events and is therefore a useful screening test in high-risk individuals perioperatively. Many studies have investigated the significance of early postoperative troponin increases above the upper reference limit of the test assay. These are summarized in Table 2. A meta-analysis of the 4910 patients included in the 18 studies listed show increased troponins to have a high sensitivity, specificity and negative predictive value. One problem in confirming results has been the difficulties in determining the upper reference limit of the normal range for different tests.

(2) Other biomarkers
There is increasing interest in other plasma biomarkers, especially those which are markers of inflammation and the atherosclerotic process rather than myocardial cell damage. Among these biomarkers, the majority of work has focused on investigating C-reactive protein, but there are several other plasma biomarkers that are postulated to have a role in the pathogenesis of atherosclerosis, and are discussed below.

In addition, some plasma biomarkers appear to be risk factors for the atherosclerotic process (e.g. an unfavourable lipid profile). Other less routinely measured risk factors include uric acid, homocysteine and leptin. None of these has been investigated for their ability to predict perioperative cardiovascular events.

(a) Acute phase reactants

(i) C-reactive protein (CRP)
CRP was discovered in the 1930s by Tillet and Francis, who observed that a non-type-specific polysaccharide fraction was precipitated from the sera of acutely ill patients with pneumococcal pneumonia. It was later discovered that, in the presence of calcium ions, CRP binds a range of ligands most of which contain phosphoryl choline.

CRP is produced by hepatocytes in response to some pro-inflammatory cytokines, mainly interleukin-6 (IL-6), but also IL-1β in the presence of IL-6. CRP is found as a trace constituent of normal plasma with the median level being 0.8 mg litre$^{-1}$ and the interquartile range 0.3–1.7 mg litre$^{-1}$. Most apparently healthy subjects have serum levels less than 3 mg litre$^{-1}$ with levels greater than this not considered normal. However, an individual’s baseline CRP level is fairly constant, substantially genetically predetermined, and there is no gender or age determined variation.

CRP was the first protein to be discovered that acted as a positive acute phase reactant. With the onset of the acute phase reaction, CRP levels increase rapidly and reach peak levels, which may be as high as 300 mg litre$^{-1}$, within 24–48 h. The half time of CRP in the plasma is 19 h and is constant in all conditions; hence levels decrease rapidly on resolution of the inflammatory stimulus. It is pro-inflammatory and pro-atherogenic, with the level reflecting the extent and activity of the disease process.

Recent studies hypothesize that CRP is not only an inflammatory marker of atherosclerosis, but also actively participates in the process of atherogenesis, and is found...
within atherosclerotic plaques in both coronary and peripheral arterial vessels. In vitro, CRP binds to LDL and may become trapped in the vessel intima attached to the deposited lipids. It is well known that CRP can activate the complement system, and identification of activated complement system components along with CRP in early atherosclerotic lesions has led to the concept of CRP being involved in sustaining chronic inflammation within the arterial wall. Foam cells also stain positively for CRP, and it is hypothesized that CRP has a role in the formation of these cells by opsonization of lipid particles. However, CRP levels have not been shown to consistently correlate with the extent or burden of atherosclerotic disease when quantified by Doppler ultrasound of the carotid arteries and electron beam computerized tomography for coronary calcium.

The many inflammatory processes known to raise CRP are shown in Table 3. In general, drugs or other treatments do not affect CRP production unless they also affect the disease process involved, while liver impairment will affect production of CRP.

At present, most hospital biochemistry laboratories can measure CRP concentrations, but will not quantify low concentrations, reporting those under a certain threshold limit as 'normal'. However, the accurate measurement of serum CRP concentrations up to 3 mg litre\(^{-1}\) is now possible with the advent of new high sensitivity assays (hs-CRP). Measurements at these low concentrations, which were previously considered to be within the normal range, have been shown to be a useful screening tool for the risk of coronary artery disease providing prognostic information in patients with known atherosclerotic disease. All individuals will have trace levels of CRP detectable on admission, with the hs-CRP level of greater than 1 mg litre\(^{-1}\) should be considered as a risk factor for future complications in patients with stable coronary disease, stroke and peripheral artery disease, while levels above 10 mg litre\(^{-1}\) have greater prognostic value in those suffering from acute coronary syndromes.

The current utility of CRP. The median level of hs-CRP, within apparently healthy individuals, has been found to be...
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Table 3 Conditions known to significantly raise CRP

<table>
<thead>
<tr>
<th>Infections</th>
<th>Inflammatory disease</th>
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<tr>
<td>Allergic complications of infection</td>
<td>Rheumatic fever</td>
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<tr>
<td>Erythema nodosum leprosum</td>
<td>Erythematoid arthritis</td>
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<tr>
<td>Rheumatoid arthritis</td>
<td>Juvenile chronic arthritis</td>
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<td>Ankylosing spondilitis</td>
<td>Ankylosing spondilitis</td>
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<td>Psoriatic arthritis</td>
<td>Spondylitis</td>
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<tr>
<td>Systemic vasculitis</td>
<td>Polymyalgia rheumatica</td>
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<td>Polyarteritis rheumatica</td>
<td>Reiter’s disease</td>
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<td>Crohn’s disease</td>
<td>Crohn’s disease</td>
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<td>Familial Mediterranean fever</td>
<td>Surgery</td>
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<tr>
<td>Burns</td>
<td>Burns</td>
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<tr>
<td>Fractures</td>
<td>Burns</td>
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<td>Necrosis</td>
<td>Burns</td>
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<td>Myocardial infarction</td>
<td>Burns</td>
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<td>Trauma</td>
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<td>Trauma</td>
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<td>Necrosis</td>
<td>Burns</td>
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<td>Malignant neoplasia</td>
<td>Burns</td>
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<tr>
<td>Inflammatory disease</td>
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<td>Conditions known to significantly raise CRP</td>
<td>Burns</td>
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Table 4 Patient characteristics known to alter hsCRP levels

<table>
<thead>
<tr>
<th>Increased levels</th>
<th>Decreased levels</th>
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<tr>
<td>Raised BMI</td>
<td>Moderate alcohol consumption</td>
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<tr>
<td>Cigarette smoking</td>
<td>Increased activity or endurance exercise</td>
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<tr>
<td>Metabolic syndrome and diabetes mellitus</td>
<td>Weight loss</td>
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<tr>
<td>Low HDL and high triglyceride</td>
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<tr>
<td>Oestrogen/progesterone hormone use</td>
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significantly higher in those who later develop cardiovascular events than in those who do not, and these individual’s risk of future cardiovascular disease can be stratified according to their hs-CRP level. The Framingham Risk Score was originally developed in 1991, and is a widely used scoring system designed to estimate 10 yr future risk of coronary heart disease in subjects without known disease. Recent work has shown that inclusion of the hs-CRP measurement can add additional prognostic information to this risk score especially in intermediate-risk men and high-risk women. CRP retains its independent association with incident coronary events even after adjusting for many of the confounding factors known to affect levels including age, total cholesterol, high-density lipoprotein (HDL) cholesterol, cigarette smoking, BMI, history of diabetes mellitus, history of hypertension, exercise level and family history of coronary heart disease.

In patients with stable or unstable angina, CRP concentrations can act as a predictor of the patients at high risk of suffering a coronary event in the future. Similarly, CRP concentrations are an independent predictor of ischaemic stroke, and in patients with peripheral vascular disease (PVD), the CRP concentration acts as a predictor of the severity PVD and the risk of subsequent cardiovascular events.

CRP and outcome. A preoperative CRP value of greater than 2 mg litre\(^{-1}\) was shown to be associated with postoperative complications in small group of patients undergoing cardiac surgery, and an uneventful recovery occurred in all patients with a concentration less than 2 mg litre\(^{-1}\). In patients undergoing non-cardiac surgery, there are few studies investigating preoperative CRP concentration and cardiovascular outcome. One study of 51 surgical patients undergoing revascularization procedures for PVD found that a CRP concentration greater than 9 mg litre\(^{-1}\) was predictive of perioperative MI (it should be noted that all patients with an ejection fraction of <40% were excluded).

(ii) Serum amyloid A (SAA) SAA belongs to a family of proteins that form a major component of the acute-phase inflammatory response. It is synthesized in the liver in response to a pro-inflammatory stimulus. It is thought to alter HDL-cholesterol delivery to cells, which is how it may be involved in the pathogenesis of atherosclerosis. The WISE study showed a strong independent relationship between SAA concentrations and future cardiovascular events in women referred for angiography.

(iii) Fibrinogen Fibrinogen has many functions—playing an essential part in thrombogenesis, acting as an acute phase reactant, and as a mediator of both inflammation and coagulation. It is therefore a key molecule in the processes involved in atherogenesis. Epidemiological studies have shown that patients with peripheral arterial disease have an increased plasma fibrinogen concentration. It has also been shown that in high-risk patients with PVD an elevated fibrinogen concentration is predictive of fatal cardiovascular complications over a 10 yr follow-up period.

(iv) Complement proteins The complement proteins form a biochemical cascade involved in inflammation. There are three pathways by which the cascade can be activated; the classical pathway (by antigen–antibody complexes and CRP), the alternative pathway (by microbial surfaces) and the lectin-binding pathway (by acute phase proteins). Complement proteins are synthesized mainly by the liver, but other cells including monocytes and macrophages can produce them. Increased plasma concentrations of complement proteins indicate that an acute phase reaction or inflammatory process is occurring. Studies have shown that patients with atherosclerosis have elevated plasma complement concentrations and within atheromatous plaque-activated complement components have been found. Complement activation, as indicated by a raised C5a, in patients with advanced peripheral artery disease has been shown to predict an increased cardiovascular risk.

(b) Cell adhesion molecules

(i) Intercellular adhesion molecule-1 (ICAM-1) ICAM-1 plays a critical role in the formation of early atherosclerotic
plasmas by facilitating the adherence of monocytes to the vascular endothelium and deficiency of ICAM-1 has been shown to be associated with protection against atherosclerosis in mice. The soluble form of ICAM-1 (sICAM-1) can be measured in the plasma. The Bezafibrate Infarction Prevention study showed that an increased baseline serum concentration of sICAM-1 was associated with a higher incidence of future coronary events in patients with chronic coronary heart disease.28 The same study also showed that concentrations of sICAM-1 were significantly associated with the risk of ischaemic stroke.81 Another study showed that the concentration of sICAM-1 is related to the estimated risk of coronary heart disease in apparently healthy individuals. Concentrations in the upper quintile were associated with a 4.15% risk of coronary heart disease in the next 10 yr compared with 1.5% for those with a concentration in the lowest quintile.83 The Physicians Health Study also measured sICAM-1 concentrations for a proportion of case-controlled subjects who developed MI and found a significant association between baseline sICAM-1 concentrations and the risk of future MI independent of other risk factors.65

(ii) Vascular cell adhesion molecule-1 (VCAM-1) VCAM-1 is a cell adhesion molecule which is not expressed under baseline conditions, but is rapidly induced by proatherosclerotic conditions. There is highly suggestive, but not conclusive, evidence of a pathogenic role for VCAM-1 in the development of atherosclerotic plaques. The soluble form of VCAM-1 (sVCAM-1) can be measured in the plasma and is increased in patients with hyperglycaemia. It has been postulated to have a role in the excessive atherosclerotic plaque formation seen in these patients.30 A study looking at patients with previously documented coronary artery disease, found that sVCAM-1 was a stronger predictor of risk than sICAM-1,8 but both the Physicians Health study and the Atherosclerosis Risk in Communities study did not find that sVCAM-1 levels were predictive of future cardiovascular risk.

(iii) Selectins P-selectin is an adhesion molecule produced mainly by platelets that mediates initial monocyte rolling before adherence to the endothelium in the early stages of atherosclerotic plaque formation. Deficiency of P-selectin has been shown to be protective against atherosclerosis in mice and increased levels of P-selectin have been demonstrated in a variety of cardiovascular disorders including coronary artery disease, hypertension and atrial fibrillation.19 The concentration of a different selectin, E-selectin, has been shown to be raised in the plasma of hyperglycaemic men and is postulated to have a role in athogenesis.39

(c) Cytokines and chemokines

(i) Interleukins The interleukins (IL) make up a group of cytokines produced by a wide variety of cells. Certain pro-inflammatory IL are involved in the acute phase reaction, mainly IL-1, -6 and -8, and are termed acute phase proteins. In response to these cytokines the liver produces a number of acute phase reactants including CRP, SAA, complement and fibrinogen. IL-6 appears particularly important. It is secreted by T-lymphocytes and macrophages, and receptors for IL-6 are found on the surface of many cells. The Physicians Health study showed that baseline IL-6 concentrations can predict future cardiovascular events,68 and a further study showed that baseline IL-6 is predictive of peripheral atherosclerotic disease progression within 5 yr independent of other risk factors.85

(ii) Tumour necrosis factor-α (TNF-α) TNF-α is a multifunctional, pro-inflammatory cytokine with effects on many different tissues including the endothelium. It is involved in the acute phase reaction along with some of the IL. In the Cholesterol and Recurrent Events (CARE) trial, elevations of TNF-α in patients after MI were associated with an increased risk of recurrent coronary events.67

(iii) Endothelins Endothelins are powerful vasoconstrictor peptides that are produced by a variety of tissues including the endothelial lining of blood vessels. Endothelin-1 is expressed by endothelial cells, macrophages and smooth muscle cells. It is produced as an inactive precursor called ‘big endothelin-1’. Plasma endothelin concentrations are elevated in patients with traditional atherosclerotic risk factors and in those with early atherosclerosis and coronary endothelial dysfunction. In advanced atherosclerotic disease, plasma and tissue endothelin concentrations have been found to be raised in proportion to the extent of atherosclerosis. Endothelin is known to be a chemo-attractant for monocytes and macrophages and is also thought to have a role in neovascularization. The presence of receptors for endothelin on neovessels within the atherosclerotic plaque implies an angiogenic role of endothelin-1 in atherosclerosis.73 Levels of endothelin-1 and big endothelin-1 have been shown to be significant prognostic factors in patients with congestive cardiac failure and there is some evidence that endothelin concentrations are better at predicting survival than brain natriuretic peptide levels, but this has not been applied to the perioperative period.87

(d) Other

(i) Matrix metalloproteinases (MMPs) The MMPs are endopeptidases with the capacity to cleave components of the extracellular matrix, such as collagen and elastin. They are secreted as a pro-form and require activation for proteolytic activity. The activity of MMPs is normally low in healthy tissue, but it is postulated that they may play a role in the pathophysiology and progression of cardiovascular disease. Depletion of matrix components from the fibrous cap of atherosclerotic plaques causes an imbalance between
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(e) Risk factors

(i) Uric acid  Serum uric acid is the major product of purine metabolism and is formed from xanthine. Epidemiological studies have shown that elevated uric acid levels predict an increased risk of cardiovascular events with a recent population-based study of initially healthy men showing that serum uric acid concentrations are a strong predictor of cardiovascular disease mortality independent of other variables.\(^{55}\) However, the Framingham Heart study did not find a causal role for uric acid and concluded that any apparent association was probably because of the association between uric acid concentration and other risk factors.

(ii) Homocysteine  Homocysteine is an amino acid that is known to be a risk factor for cardiovascular disease. Patients suffering from homocysteinuria develop premature vascular disease, and there are many studies showing a correlation between plasma homocysteine concentration and coronary artery disease, peripheral arterial disease, stroke and venous thrombosis. Homocysteine is thought to affect coagulation by acting as a prothrombotic factor, and reduce the resistance of the endothelium to thrombosis.\(^{57}\) Elevated homocysteine concentrations on admission to hospital in patients with acute coronary syndrome are predictive of late but not early (within 28 days) cardiac events,\(^{79}\) and elevated concentrations before coronary angioplasty are predictive of late mortality and adverse outcome.\(^{74}\) However, a meta-analysis performed in 2002 found that an elevated homocysteine concentration is at best a modest predictor of ischaemic heart disease and stroke risk in the healthy population.\(^{13}\)

(iii) Leptin  This peptide hormone is produced by white adipose tissue. Plasma leptin concentrations are proportional to body adiposity, and are markedly increased in obese individuals. Leptin exhibits potentially atherogenic effects such as endothelial dysfunction, stimulation of inflammatory reaction, oxidative stress, decrease in paraoxonase activity, platelet aggregation, migration, hypertrophy and proliferation of vascular smooth muscle cells. Leptin deficient and leptin-receptor deficient mice are protected from arterial thrombosis and neointimal hyperplasia in response to arterial wall injury. Epidemiological studies show that a raised plasma concentration of leptin is predictive of acute cardiovascular events even after adjustment for BMI, plasma lipids, glucose and CRP.\(^{89}\) In patients with known coronary atherosclerosis, the plasma leptin level has been found to be predictive of future cardiovascular events over a 4 yr follow-up period.\(^{94}\)

A future role for plasma biomarkers?

There are still a number of patients who suffer perioperative cardiovascular events in whom the traditional preoperative tests for stress-induced myocardial ischaemia do not identify a highly increased risk. We postulate that this is because the traditional preoperative tests do not identify patients at risk of the plaque rupture type of perioperative MI. None of the preoperative tests we currently use can identify vulnerable plaques, and consequently identifying patients at risk of plaque rupture and haemorrhage, is not possible.

Of the tests described above, CRP appears the most promising for measurement in the perioperative period. CRP does not correlate with atherosclerotic burden, but may act as a marker of other atherosclerotic characteristics, possibly the activity of lymphocyte and macrophage populations within the plaque or the degree of plaque destabilization and ongoing ulceration or thrombosis.\(^{7}\) We question whether CRP could be useful in identifying patients with vulnerable plaques. If used as a preoperative test for unstable atherosclerotic plaques, the result would only be interpretable in those patients without other co-existing inflammatory conditions. This might limit its use. However, vascular surgical patients have the highest incidence of perioperative cardiovascular events and this test would be applicable in the majority of these patients.

Possible perioperative medical treatment for vulnerable plaques

It would be extremely useful to identify patients with vulnerable plaques before operation if an intervention could then be initiated with the aim of reducing the risk of perioperative plaque rupture.

Possible drugs of importance include the 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase inhibitors (statins), which modify lipid levels; lowering LDL and total cholesterol levels, whilst increasing HDL levels. They have been shown to be highly effective drugs in reducing the risk of cardiovascular events in the setting of both primary and secondary prevention. The magnitude of this risk reduction is much greater than can be predicted on the basis of lowering LDL cholesterol alone,
and it is postulated that some of this risk reduction is because of pleiotropic, non-lipid properties including the improvement of endothelial function, plaque stabilization and the reduction of oxidative stress in vascular inflammation. The anti-inflammatory effects appear to be mediated via interference with the synthesis of isoprenoid intermediates (mevalonate metabolites) and limitation of the nuclear factor-κB dependent transcriptional regulation in response to an inflammatory stimulus.

The question of whether or not inflammatory markers such as CRP are clinically useful in selecting patients who may benefit from statin therapy despite having normal LDL cholesterol levels has yet to be answered, and the JUPITER trial has been set up to address this question. Although statins lower CRP levels, it has yet to be proven that this represents a true reduction in inflammation. A recent study showed that CRP expression in human hepatocytes after statin therapy was blocked even in the presence of cytokines known to induce CRP, suggesting that statins block CRP expression at the level of transcription. Work has shown that CRP in itself may be a cardiovascular risk factor, by quenching the production of nitric oxide which in turn inhibits angiogenesis, an important compensatory mechanism in chronic ischaemia.

Statin therapy has been shown to reduce the incidence of perioperative cardiovascular complications in patients undergoing major non-cardiac surgery in a large retrospective cohort study, and a prospective double-blind randomized controlled trial. After abdominal aortic aneurysm repair, long-term statin therapy has been shown to be associated with a 3-fold reduction in cardiovascular mortality. Concerns about an increased incidence of statin-associated myopathy within the surgical population are unfounded.

Studies have shown that improvements in endothelial function, and reductions in serum inflammatory markers occur within 2–16 weeks after beginning statin therapy but the minimum period of preoperative and postoperative therapy has not yet been determined. The most efficacious dose of statin therapy in the perioperative period is another area lacking research. In acute coronary syndromes, high dose statin therapy is now advocated showing a reduction in future events over placebo or a standard dose regimen. The question of whether patients at increased risk of perioperative cardiovascular events with raised inflammatory markers would benefit from this sort of high dose statin regimen during the perioperative period has yet to be answered.

Another class of drug known to reduce the concentrations of inflammatory mediators including CRP is the thiazolidinedione group, which are used in the treatment of diabetes mellitus type 2. These drugs have been found to have a beneficial effect on the cardiovascular system independent of their anti-diabetic effect but any potential protective role of these drugs in the perioperative period has not been studied.

**Conclusion**

Most work to date has focused on the identification of a subgroup of surgical patients at high risk of PMI. Whilst traditional preoperative tests of myocardial reserve and coronary stenosis can be helpful in predicting those at risk of ischaemia-induced PMI, there is no currently available test that can reliably identify surgical patients with ‘vulnerable plaques’.

Of those molecules and mediators of the atherosclerotic process that can be measured in the plasma, CRP has been investigated more extensively than others with regard to prognostic significance. However, its role in identifying surgical patients at risk of perioperative plaque rupture and haemorrhage has yet to be studied. Future studies are needed to evaluate the perioperative potential of this and other biochemical markers.

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