Review

Bertil Lindahl*

Acute coronary syndrome – the present and future role of biomarkers1)

Abstract: Over the past two decades there have been dramatic changes in the diagnosis, treatment and prognosis of acute coronary syndrome (ACS). Several new treatment modalities have been added and the prognosis has improved dramatically. Biomarkers play a crucial role in the management of ACS. At present, cardiac troponin is the biomarker of choice for diagnosis of acute myocardial infarction (AMI). Currently, there are no other biomarkers, which can compete, neither regarding specificity nor regarding early sensitivity. However, there is still a clinical need of a biomarker able to reliably rule-in or rule-out AMI immediately on admission. MicroRNAs seem to be promising new candidates for diagnostic purposes. The optimal combination of biomarkers and new imaging techniques is another important area for research. The list of biomarkers associated with an adverse prognosis in ACS is long. However, for most of them it has been very difficult to prove an added clinical value. Only cardiac troponin, and to some degree also B-type natriuretic peptides, is widely used in clinical practice for risk assessment. Among new markers, growth differentiation factor 15 and the mid-regional part of the prohormone of adrenomedullin, have shown some promising results. Since the renal function is assessed in clinical routine, also markers of the renal function have gained increasing interest. Cardiac troponin has been proven useful for selection of antithrombotic, antiplatelet and invasive treatment. Besides cardiac troponin, no other markers have consistently been shown to be useful for selection of specific treatments.

Keywords: acute coronary syndromes; biomarkers; diagnosis; prognosis.

Introduction

Patients presenting with symptoms suggestive of an acute coronary syndrome (ACS) encompass a heterogeneous group of patients with a variable clinical background, different severity of the underlying coronary artery disease, large variation in clinical course and variable risk of subsequent cardiac events. In a substantial proportion of patients with an initial suspicion of ACS, the diagnosis will eventually be ruled out, and the patients will be found to have other cardiac or non-cardiac diagnoses. Thus, patients presenting with symptoms suggestive of ACS constitute a diagnostic, prognostic and therapeutic challenge. Biomarkers, together with the ECG, play crucial roles for early diagnosis of ACS and assessment of the prognosis and are important for tailoring the treatment to the individual patient.

Acute coronary syndrome

Acute coronary syndrome is an umbrella term for acute myocardial infarction (AMI) and unstable angina (UA) and is characterized by abrupt and unpredictable reductions in coronary blood flow causing myocardial ischemia at rest or decreasing levels of exertion and in case of AMI, measurable amounts of myocardial necrosis. ACS is subdivided based on the ECG-changes at presentation in ST-segment elevation myocardial infarction (STEMI) and non-ST-segment elevation ACS (NSTEMI). NSTEMI is further divided in non-ST-segment elevation myocardial infarction (NSTEMI) and UA depending on whether myocardial necrosis is demonstrated or not.

Key events for the development of ACS are in most cases a disruption of an atherosclerotic plaque and thrombus formation causing partial or complete occlusion of the infarct-related artery or distal embolization in the coronary tree [1]. However, ACS (and so called type II/secondary AMI) may occur in the absence of plaque rupture.
and thrombosis in conditions causing supply-demand mismatch to the myocardium, e.g., hypotension, anemia, infection and tachyarrhythmia [2].

The incidence of ACS is declining in most developed countries [3, 4]. Lifestyle adjustments in the general population, e.g., reduction in smoking and of serum cholesterol levels, have contributed to the reduction in incidence [5]. Among patients with AMI, the relative occurrence of STEMI has decreased and the occurrence of NSTEMI has increased, and NSTEMI is now more common than STEMI [6]. The decrease in incidence has been paralleled by an impressive improvement in mortality, e.g., in Sweden the standardized 30-day mortality in STEMI went from 12.9% to 6.3% between 1996 and 2007 [7]. During the same period there has been an increasing use of evidence-based therapies in ACS, such as revascularization (PCI and CABG), anti-platelet and anti-coagulant agents, ACE-inhibitors and statins [7].

**Biomarkers in ACS**

Measurements of biomarkers in serum or plasma might be used for diagnosis, prognosis and selection of appropriate treatment. A multitude of biomarkers has been suggested and evaluated for these purposes [8, 9]. The biomarkers can be grouped in areas reflecting different pathophysiological mechanisms operating in ACS (Figure 1). Biomarkers available (or expected to soon be available) for use in clinical routine are summarized in Table 1. However, currently only a few of them have gained widespread use.

**Diagnosis**

Up to 1954, with the first report that the enzyme, glutamic oxaloacetic transaminase, could be used for diagnosis of AMI [10], the clinical diagnosis relied solely on the history and on development of diagnostic Q-waves in the ECG. Today, elevated levels of a marker of myocardial damage, particularly cardiac troponin I (cTnI) or cardiac troponin T (cTnT), is a prerequisite for the diagnosis of AMI [2]. An ideal biomarker for diagnosis of AMI should (adopted from [11]): 1) exist in high concentration in the myocardium; 2) not exist in any other tissue, neither under normal, nor under pathological conditions; 3) not be measurable in plasma under normal conditions; 4) be released only after irreversible damage to the myocardium; 5) be released in direct proportion to the extent of myocardial necrosis; 6) be rapidly released and persist in the plasma long enough to allow a convenient diagnostic time window; and 7) be suitable for development of rapid, reliable and inexpensive methods for measurement. Cardiac troponins fulfill most, but not all, of these requirements. The development of high sensitivity cardiac troponin assays has increased the analytical sensitivity with almost three orders of magnitude compared to the first generation assays, and has made it evident that cardiac troponins indeed are measurable in plasma also under normal conditions [12]. The previous belief that cardiac troponins are only released after irreversible myocardial damage is seriously challenged [13]. The vast majority of patients diagnosed with UA in the pre-troponin era have slight elevations.

![Figure 1: Different pathophysiological mechanisms and associated biomarkers. BNP, B-type natriuretic peptide; CRP, C-reactive protein; cTn, cardiac troponin I or T; IMA, ischemia modified albumin; LV, left ventricular.](image)

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AMI, acute myocardial infarction; BNP, B-type natriuretic peptide; cTn, cardiac troponin I or T; GDF-15, growth-differentiation factor-15; h-FABP, heart fatty acid-binding protein; MR-proADM, mid-regional part of the prohormone of adrenomedullin; nd, no data.
of cardiac troponin, and using the most sensitive assays available it can be questioned whether ‘true’ UA does exist, i.e., without any evidence of myocardial damage (=cardiac troponin elevation) [14, 15]. Furthermore, the cardiac troponin assays with high analytical sensitivity have also shown that cardiac troponins are released at least as early as the previously thought ‘earlier’ markers myoglobin and heart fatty acid binding protein [16, 17]. Therefore, it has been difficult to prove that adding another marker of myocardial damage to cardiac troponin, provide any clinically meaningful benefit over using cardiac troponin alone for diagnosis [16, 17]. However, some 10%–20% of patients with AMI still had non-elevated levels of cardiac troponin at presentation to the emergency room, necessitating serial measurements over at least a 3-h period [2] in order to be able to reliably rule out AMI. Hence, there is still an unmet clinical need to be able to diagnose ACS, and particularly AMI, with certainty already on admission. Therefore it seems logical to add a marker that reflects some other important pathophysiological aspect of ACS, such as ischemia, activation of the coagulation system or the plaque rupture. However, to find a marker of ischemia has been notoriously difficult, and studies of the suggested marker so far, ischemia modified albumin (IMA), have failed to convincingly and consistently show added clinically relevant diagnostic value although some initial studies showed positive results [18]. The IMA assay was approved by FDA but is no longer available for clinical use. In a study evaluating multiple markers for early diagnosis of AMI [19], neither of the three markers suggested to indicate plaque rupture, myeloperoxidase (MPO), matrix metalloproteinase 9 (MMP-9) and pregnancy-associated plasma protein-A (PAPP-A) added any clinically meaningful diagnostic information to that of cTnT alone, nor did CD40L and D-dimer. In a systematic review of the literature on novel biomarkers for diagnosing ACS [18], not a single marker thought to reflect inflammation in general, plaque rupture or activation of the coagulation system or platelets, demonstrated supportive evidence in diagnosing ACS alone or in combination with cardiac troponin (except for MMP-9 in one study [20]). For a few other markers, heart fatty acid-binding protein (marker of myocardial injury), B-type natriuretic peptide (a marker of ventricular wall stress) and Copeptin (the C-terminal end of the prohormone of vasopressin), the various studies have shown conflicting results [17, 18]. Thus, so far there is inadequate evidence to support routine use, either alone or in combination with cardiac troponin, of any of these novel biomarkers for diagnosing ACS.

Prognosis

Early prognostic evaluation is essential for the application of appropriate treatment and further management. However, when assessing the risk for new cardiac events it is important to define which event or events are of interest, since the biomarkers’ predictive ability might differ considerably between different endpoints. Generally, more biomarkers are predictive of mortality than of AMI. Therefore, information from studies only using composite endpoints may be difficult to interpret and even misleading. Furthermore, to be clinically useful the biomarker must add independent prognostic information to what is already available in routine practice, i.e., the patient history and the ECG. Another important issue to consider when comparing different studies on the prognostic value of risk markers is in which populations the study were performed. Patients enrolled in randomized clinical trials are often highly selected and with established ACS, whereas most observational studies have less restricted inclusion criteria. Generally, the relative risk or odds ratio for a clinical event associated with a positive marker is higher in observational studies. A large number of biomarkers have been shown to predict death or the combined endpoint death/AMI, while much fewer have been shown to be predictive of AMI (Table 1).

Troponin

It has been convincingly demonstrated, that patients with, compared to without, elevation of cTnl or cTnT have a higher mortality, both short- and long-term. The odds ratio for death for patients with elevated cTnT and cTnl was 3.0 (95% CI 1.6–5.5) and 2.6 (95% CI 1.8–3.6), respectively, in a meta-analysis of randomized clinical trials [21]. In a separate analysis of observational cohort studies the corresponding odds ratios were 5.1 (95% CI 3.2–8.4) and 8.5 (95% CI 3.5–21.1) [21]. Furthermore, there seems to be a dose-response relation, since mortality increases by increasing levels of troponin [22, 23]. In contrast to most other biomarkers, patients with elevated cardiac troponin also have an increased risk of suffering a new AMI. However, there seems to be more of a threshold effect for the risk of a new MI, every reliable elevation of cardiac troponin is associated with an increased risk of a new MI in non-STE ACS [23]. Information about the association between troponin level and risk of re-hospitalization and congestive heart failure, respectively, are limited.
C-reactive protein

C-reactive protein (CRP) is an acute phase protein, which is synthesized in the liver and increases within 4–6 h after tissue damage or in response to inflammation. It is commonly used in clinical routine for the diagnosis and monitoring of bacterial infection, tissue damage and inflammatory diseases. The currently used high-sensitivity CRP assays have high analytical sensitivity and assay precision, allowing reliable measurement of CRP also at levels found in healthy individuals. CRP has gained interest as a prognostic marker in ACS with the recognition that atherosclerosis is an inflammatory disease. The optimal decision limit as well as the optimal time point for risk stratification, however, is still unclear. Most studies so far have used 3 or 10 mg/L as the decision limit. In AMI, CRP peaks about 24–48 h after onset and the peak level is related to the infarct size [24]. In ACS patients the CRP level is independently associated with long-term risk of death [25, 26], however, for short-term risk the results are contradicting [25]. The ability of CRP to predict non-fatal AMI is questioned [27] and few studies have studied the incremental value of CRP in relation to usual risk prediction [25]. Measurement of CRP for risk stratification of patients with ACS has received a class IIa recommendation in the National Academy of Clinical Biochemistry Laboratory Medicine Practice Guidelines [28], but has not gained widespread use in clinical practice.

The B-type natriuretic peptide

The B-type natriuretic peptide (BNP) and the N-terminal part of its prohormone (NT-proBNP) are mainly released from myocytes in the cardiac ventricles in response to increased stretch and wall tension [29]. However, the natriuretic peptides might also rise in response to a number of other stimuli, among them ischemia per se and cytokines [30, 31]. BNP and NT-proBNP have received widespread use for diagnosis of heart failure and evaluation of acute dyspnea. However, BNP and NT-proBNP also rise early after the onset of symptoms in patients with ACS and the levels of natriuretic peptides have been shown to be strong and independent predictors of mortality in a number of studies [32, 33]. However, despite the convincing evidence as markers for risk prediction [25], the lack of diagnostic value of natriuretic peptides in ACS have so far prevented a more widespread use of BNP or NT-proBNP also in ACS.

Cystatin C

Measurements of renal function such as serum creatinine and estimation of creatinine clearance have been shown to carry independent prognostic information in ACS [34]. However, creatinine concentration is an unreliable estimate of the glomerular filtration rate (GFR). The level of creatinine is influenced by factors such as age, gender, muscle mass, physical activity, and diet. Because of the non-linear relationship between creatinine concentration and GFR, it is also too insensitive to detect small decreases in GFR and mild renal dysfunction. Cystatin C is an endogenous inhibitor of cathepsins, which are cysteine proteases. Cystatin C is produced in all nucleated cells at a constant rate and is freely filtered by the glomerulus without secretion or subsequent reabsorption to the blood flow and therefore, has been suggested as a better marker of GFR than serum creatinine, which has been verified in several, but not all comparative studies [35]. However, Cystatin C might also be a systemic marker of ongoing inflammatory processes since cathepsins are proinflammatory [36]. The predictive value of Cystatin C in ACS patients has been evaluated in several studies and Cystatin C has been shown to be an independent predictor of mortality and in some, but not all, a better marker than serum creatinine [36–39].

Growth-differentiation factor-15

Growth-differentiation factor-15 (GDF-15) is a distant member of the transforming growth factor-β cytokine superfamily that is induced in the myocardium following pathological stress associated with inflammation or tissue injury [40]. An increasing number of studies have shown that the level of GDF-15 is a strong and independent predictor of mortality in patients with ACS [41–43].

Mid-regional part of the prohormone of adrenomedullin

Adrenomedullin is a 52-amino-acid peptide that is expressed by various tissues including the vessels and the myocardium. Adrenomedullin exhibits protective effects on the heart and the vasculature [44]. The stable mid-regional part of its prohormone, MR-proADM, is elevated in various cardiovascular pathologies and has been shown to be strongly predictive of mortality in populations with
cardiovascular disease, especially heart failure [45] but also AMI [46].

**Selection of therapy**

A large number of therapeutic options are available in the management of patients with ACS. However, some of these are rather costly and have potential serious side effects. Therefore, identification of those who benefit most from a particular therapy has become important. The beneficial effects of antithrombotic treatment with low molecular weight heparins, antiplatelet therapy with glucoprotein IIb/IIIa receptor inhibitors, and an invasive approach with early revascularization in NSTE-ACS have all been shown to be predominantly present in patients with elevated troponin [47–49]. Hence, the cardiac troponin level has been incorporated in the treatment algorithms in recent guidelines [50].

For other biomarkers there are limited data in the literature, GDF-15 has been shown to identify those who benefit from revascularization in one study [50], likewise Il-6 [51]. For NT-proBNP the results are conflicting regarding revascularization [52, 53].

**Future directions**

**Search for new markers**

There is an intense search for new biomarkers in the cardiovascular field. There has been a rapid development of technologies for proteomic studies and currently many proteomic studies observe 1000–5000 proteins [54]. In analogy with genome-wide association studies, hypotheses free association studies (‘whole proteome scanning’) are underway to identify completely new biomarkers in the cardiovascular field [54, 55].

MicroRNA constitutes a whole new class of biomarkers. MicroRNAs are short, non-coding RNAs that regulate gene expression at the post-transcriptional level and play a role in normal development and physiology, as well as in disease development, including in the cardiovascular system [56]. MicroRNAs are measurable also in circulating blood and are relatively stable. There are some promising results indicating the potential use of some microRNAs (e.g., miR-1, miR-133, miR-208, miR-328 and miR-499) for (early) diagnosis of AMI [56]. However, the studies are so far very small, and the suggested microRNAs need to be evaluated in comparison with cardiac troponin in large multicenter studies.

**Combination of biomarkers with cardiac imaging**

Rapid and non-invasive imaging techniques, i.e., computerized tomography (CT) coronary angiography and cardiac magnetic resonance imaging, giving detailed anatomical, but also functional, information of the heart are rapidly evolving. Therefore, the combination of imaging with biomarkers seems logical for both diagnostic and prognostic purposes. The optimal combination of measurements of biomarkers and CT coronary angiography for early diagnosis and risk assessment of patients with suspicion of ACS are evaluated in ongoing clinical studies, e.g., the ROMICAT II study [57].

**New applications for biomarkers**

Studies using very sensitive cardiac troponin assays have shown that chronically elevated levels above the 99th percentile level of healthy individuals are common among patients with structural heart disease [58, 59]. It is sometimes difficult in clinical practice to differentiate these chronic elevations from acute elevations. Therefore, a marker capable of separating acute from chronic myocardial injury in a single blood sample would be clinically very useful. Likewise, a biomarker capable of separating primary atherothrombotic AMI (AMI type 1) from secondary AMI due to supply/demand imbalance (AMI type 2) would be clinically helpful [2], since the treatment and management are different between the two forms of AMI. The distinction between the two forms is sometimes difficult, e.g., in the patient with preexisting coronary artery disease with chest pain, rapid atrial fibrillation, unspecific ST-segment depression in the ECG and rising levels of troponin.

**Conflict of interest statement**

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**References**


Bertil Lindahl is Professor of Cardiology, at the Department of Medical Sciences and Uppsala Clinical Research Center, Uppsala University, Sweden. He has published more than 180 original and review articles.