Cardiac Troponin: Clinical Role in the Diagnosis of Myocardial Infarction

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CASE REPORT

The patient, a 63-year-old Caucasian female, was hospitalized on 1 April 2002 through 10 April 2002 for a non-ST segment elevation myocardial infarction (non-Q-wave MI per chart documentation). She had a negative adenosine stress test after the initial event. Her serum cardiac-specific troponin I (cTnI) concentration 24 hours after her onset of chest pain was 1.4 ng/L (upper limit of normal is 0.3 ng/ml), and her creatine kinase (CK) MB level was 12.5 ng/L (upper limit of normal 6.0 ng/ml). Three days post-event her cTnI level was 0.5 ng/L and her CK-MB level was 4.5 ng/L (Fig. 5-1). MB refers to one of the isoenzyme forms of CK found in serum. The form of the enzyme that occurs in brain (BB) does not usually get past the blood-brain barrier and therefore is not normally present in the serum. The MM and MB forms account for almost all of the CK in serum. Skeletal muscle contains mainly MM, with less than 2% of its CK in the MB form. MM is also the predominant myocardial creatine kinase and MB accounts for 10%–20% of creatine kinase in heart muscle.

At 72 hours after presentation, the patient experienced new-onset chest pain, described as a burning pain in the left shoulder, arm, and epigastrum. The electrocardiogram (ECG) demonstrated only nonspecific T-wave abnormalities and was not different from the one obtained at the time of her initial presentation. Normal sinus rhythm was now present. Nitroglycerin provided some relief. Based on new symptoms, along with recurring T-wave abnormalities and an increasing cTnI, diagnosis of reinfarction (extension of her initial event) was made. The cTnI concentration on the day of her suspected reinfarction (day 4) was increased to 1.8 µg/L with a corresponding CK-MB value of 13.6 µg/L. Cardiac catheterization revealed a 95% distal left anterior descending stenosis, a 95% mid-right coronary artery narrowing, and a 90% occluded circumflex proximally. Stents were placed in both the distal and proximal right coronary artery. The rest of her hospital stay was uneventful, and she was discharged home on day 7. At 3-month follow-up, the patient was participating in a cardiac rehabilitation program and doing well.

DIAGNOSIS

The term acute myocardial infarction (AMI) is defined as an imbalance between myocardial oxygen supply and demand, resulting in injury to and the eventual death of myocytes. When the blood supply to the heart is interrupted, “gross necrosis” of the myocardium results. Abrupt and total loss of coronary blood flow usually results in a clinical syndrome known as ST segment elevation AMI (STEAMI or Q-wave MI; diagnostic by electrocardiogram) because of the characteristic electrocardiographic changes that occur. Partial loss of coronary perfusion, if severe, can also lead to necrosis, which is generally less extensive, and this type of infarction is usually termed non-ST elevation myocardial infarction (NSTEMI or...
non-Q-wave MI, not diagnostic by electrocardiogram). There is considerable overlap between the pathophysiology and the pattern of necrosis of the two entities. Other events of less severity may be missed entirely or if detected may be diagnosed as angina that can range from stable to unstable.

The term acute coronary syndrome (ACS) encompasses most of the patients defined so far in this chapter who present with unstable ischemic heart disease. Most of these syndromes occur in response to an acute event in the coronary artery when circulation to a region of the heart is obstructed. If the obstruction is high grade and persists, then necrosis usually ensues. Since necrosis takes some time to develop, it is apparent that therapy, including opening the blocked coronary artery in a timely fashion, often can prevent some of the death of myocardial tissue. These syndromes are usually, but not always, associated with chest discomfort.

Previously, the diagnosis of AMI established by the World Health Organization required at least two of the following criteria: a history of chest pain, evolutionary changes on the ECG, or elevations of serial cardiac biomarkers (initially defined as a twofold increase of total serum CK or CK-MB). However, it was rare for a diagnosis of AMI to be made in the absence of biochemical evidence of myocardial injury. A 2000 European Society of Cardiology/American College of Cardiology (ESC/ACC) consensus conference has codified the role of biomarkers, with specific focus on cardiac troponins, by advocating that the diagnosis be based on evidence of myocardial injury based on biomarkers of cardiac injury in the appropriate clinical situation.

For these guidelines, either of the following criteria satisfies the diagnosis for an acute, evolving, or recent MI. The first is a typical rise or gradual fall of cardiac troponin, or more rapid rise and fall of CK-MB, with at least one of the following: (1) ischemic symptoms; (2) development of pathologic Q waves on the ECG; (3) ECG changes indicative of ischemia (ST segment elevation or depression); or (4) coronary artery intervention (e.g., coronary angioplasty). For the second, there should be pathologic findings of an AMI as identified at autopsy. The guidelines recognized the reality that neither the clinical presentation nor the ECG had adequate sensitivity and specificity, but that the troponin markers, in particular, could provide both. These guidelines do not suggest that all elevations of these biomarkers should elicit a diagnosis of AMI, only those associated with the appropriate clinical (ischemic presentation) and ECG findings. When elevations of cardiac troponin are observed that are not due to acute ischemia, the clinician is obligated to search for another etiology for the cardiac injury.

Patients with ACS can be categorized into four groups. First, there is the group of patients who present early to the emergency room, within 0 to 4 hours after the onset of chest pain, and who lack diagnostic ECG evidence of AMI. These patients require rapid laboratory testing for evidence of cardiac injury. Thus, useful laboratory makers of cardiac injury are those that are released rapidly from the heart and are highly specific for cardiac myocyte damage. These assays must be rapid and sensitive enough to detect even the small changes within the reference interval that can occur in blood early after the onset of symptoms.

The second patient group presents 4 to 48 hours after the onset of chest pain but without diagnostic evidence of AMI by ECG. This group of patients also requires serial monitoring of cardiac biomarkers and ECG changes.

The third group is patients who present still later, more than 48 hours after the onset of chest pain, and also lack diagnostic ECG changes. The ideal biomarker of myocardial injury for this group would have to be one that persists in the circulation for several days to provide a late diagnostic time window. The shortfall of such a marker might be its inability to distinguish recurrent injury from the prior, older injury.
The fourth group is those who present to the emergency department at any time after the onset of chest pain with clear ECG evidence of AMI. In this group, detection with serum biomarkers of myocardial injury is not necessary initially. Many of these patients may qualify for reperfusion therapy at a time before blood markers of cardiac injury have increased, and therapy should not be withheld if these criteria are met. Subsequently, specific and sensitive myocardial markers could be employed to monitor the success of reperfusion during the 60- to 90-minute period after therapy. Rapid assays providing early serial values followed by interpretation of the markers’ patterns of appearance are often helpful in determining subsequent management.

**BIOCHEMICAL PERSPECTIVES**

**Cardiac Troponin I and T**

The contractile proteins of the myofibril include three troponin regulatory proteins. The troponin complex includes three protein subunits, troponin C (the calcium-binding component), troponin I (the inhibitory component), and troponin T (the tropomyosin-binding component). The subunits exist in a number of isoforms. The distribution of these isoforms varies between cardiac muscle and slow- and fast-twitch skeletal muscle. Only two major isoforms of troponin C are found in human heart and skeletal muscle. These are characteristic of slow- and fast-twitch skeletal muscle. The heart isoform is identical with the slow-twitch skeletal muscle isoform. Isoforms of cardiac-specific troponin T (cTnT) and cTnI also have been identified and are the products of unique genes. All cardiac troponins are localized primarily in the myofibrils (94%–97%), with a smaller cytoplasmic fraction (3%–6%).

Cardiac troponin subunits I and T are encoded by different genes than the respective skeletal muscle isoforms and have different amino acid sequences, giving them unique cardiac specificity. cTnI has never been shown to be expressed in normal, regenerating, or diseased human or animal skeletal muscle. By contrast, small amounts of cTnT are expressed as one of four identified isoforms in skeletal muscle during human fetal development, in regenerating rat skeletal muscle, and in diseased human skeletal muscle. cTnT isoform expression has been demonstrated in skeletal muscle specimens obtained from patients with muscular dystrophy, polymyositis, dermatomyositis, and end-stage renal disease. Thus, care is necessary to choose antibody pairs for cardiac assay use that do not detect the isoforms reexpressed in noncardiac tissue. The commercial assay used in clinical practice only detects the heart cTnT form.

Cardiac troponin I exists as a part of the troponin T-I-C ternary complex as a structural and regulatory component of the myofibril. Following myocardial injury, multiple forms of cardiac troponins are elaborated both in tissue and in blood (Fig. 5-2). These include the T-I-C ternary complex, IC binary complex, free I, and multiple modifications of these three forms resulting from oxidation, reduction, phosphorylation, and dephosphorylation, as well as both C- and N-terminal degradation. What is elaborated likely reflects the nature of the injurious stimulus, blood flow that determines how long the protein remains in the tissue prior to reaching the circulation, the timing of the insult (i.e., forms may change as the tissue damage evolves), and perhaps genetics. Depending on which fragments are elaborated, the selection of antibodies used to detect cTnI (i.e., different antibody configurations) can lead to substantially different recognition patterns. It is now clear that assays need to be developed with the antibodies that recognize epitopes in the stable region of cTnI and ideally demonstrate an equinorm response to the different cTnI forms that may circulate in the blood.

**Creatine Kinase Isoenzymes and Isoforms**

Three cytosolic isoenzymes (CK-MM, CK-MB, CK-BB) and one mitochondrial isoenzyme (CK-Mt) of CK have been identified. Three different genes have been identified that encode for and are specific for CK-M, CK-B, and CK-Mt subunits. Although CK-MM is predominant in both heart and skeletal muscles, CK-MB has been shown to be more specific for the myocardium, which contains 10% to 20% of its total CK activity as CK-MB, compared to amounts varying from 0% to 7% in skeletal muscle.

Early studies involving animal hearts or specimens obtained at autopsy from human hearts suggested a uniform distribution of CK-MB ranging from 5% to 50% of the total CK activity. However, it has been shown by Ingwall and colleagues (1985) that the proportion of CK-MB was 6% to 15% lower in the surrounding normal areas of tissue than in infarcted myocardium in
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Figure 5-2. Schematic of cardiac troponin I and T release following myocardial cell necrosis into the circulation, demonstrating the multiple forms that exist in the blood. cTnI, cardiac-specific troponin I; cTnT, cardiac-specific troponin T.

When studied more completely in humans, CK-MB concentrations ranged from 15% to 24% of total CK in myocardial tissue obtained from patients with left ventricular hypertrophy (LVH) due to aortic stenosis, from patients with coronary artery disease (CAD) without LVH, and from patients with CAD and LVH due to aortic stenosis. In contrast, patients with normal left ventricular tissue had a low percentage of CK-MB (<2%). These data suggest that changes in the CK isoenzyme distribution are dynamic and occur in hypertrophied and diseased human myocardium. Diseased cells also have less total CK per cell.

Normal skeletal muscle usually contains very little CK-MB. Levels as high as 5%-7% have been reported in some muscles, but less than 2% is much more common. Severe skeletal muscle injury following trauma or surgery can lead to absolute elevations of CK-MB above the upper reference (normal) limit of CK-MB in serum. Increases in serum total CK and CK-MB in several patient groups often present a diagnostic challenge to the clinician. Persistent elevations of serum CK-MB resulting from chronic muscle disease occur in patients with muscular dystrophy, end-stage renal disease, and polymyositis (a degenerative disease of skeletal muscle) as well as in healthy subjects who undergo extreme exercise or physical activity. The increase in serum CK-MB in runners, for example, may be related to the adaptation by the skeletal muscle during regular training and after acute exercise, resulting in increased CK-MB tissue concentrations. The mechanism responsible for increased CK-MB in skeletal muscle following chronic muscle disease or injury is thought to be due to the regeneration of muscle, with reexpression of CK-B genes similar to those found in the heart, thus giving rise to increased CK-MB levels in skeletal muscle. Thus, skeletal muscle can become like heart muscle in its CK isoenzyme composition, with up to 50% CK-MB in some patients with severe polymyositis.

**IMMUNOASSAY PERSPECTIVES FOR MONITORING CARDIAC BIOMARKERS**

**Cardiac Troponins**

The first assay (radioimmunoassay) that measured cTnI used polyclonal anti-cTnI antibodies. The first monoclonal enzyme-linked immunosorbent assay, anti-cTnI antibody-based immunoassay, was described by Bodor and co-workers (1992). Numerous manufacturers have now developed monoclonal antibody-based diagnostic immunoassays for the measurement of cTnI in serum. Assay times range from 5 to 30 minutes.
As shown in Table 5-1, over a dozen assays have been cleared by the Food and Drug Administration (FDA) for patient testing within the United States on central laboratory and point-of-care (POC) or near-bedside testing platforms. In addition to these quantitative assays, several assays have been FDA-cleared for the qualitative determination of cTnI.

In practice, two obstacles limit the ease for switching from one cTnI or cTnT assay to another. First, there is currently no primary reference cTnI material available for manufacturers to use for standardizing their assays. Second, because of the different epitopes recognized by the different antibodies used, assay concentrations fail to agree. While standardization of assays remains elusive, harmonization of cTnI concentrations by different assays has been narrowed from a 20-fold difference to a 2- to 3-fold difference.

Several adaptations of the Roche Diagnostics cTnT immunoassay have been described, resulting in an FDA-cleared third-generation assay available worldwide. Two monoclonal anticardiac troponin T antibodies are used in the third-generation assay. Skeletal muscle TnI is no longer a potential interferent, as was found in the first-generation enzyme-linked immunosorbent cTnT assay. In contrast to cTnI, no standardization bias exists for cTnT because the same antibodies (M11, M7) are used in both the central laboratory and POC quantitative and POC qualitative assay systems.

In 2001, quality specifications for cardiac troponin assays were published. These specifications were intended for use by the manufacturers of commercial assays and by clinical laboratories utilizing troponin assays. The overall goal was to attempt to establish uniform criteria so all assays could objectively be evaluated for their analytical qualities and clinical performance. Both analytical and preanalytical factors were addressed. The following recommendations have been proposed:

1. The antibody specificity (which epitope locations are identified) needs to be delineated. Epitopes located on the stable part of the cTnI molecule should be a priority.
2. Assays need to clarify whether different cTnI forms (i.e., binary vs ternary complex) are recognized in an equimolar fashion by the antibodies used in the assay. Specific relative responses need to be described for the following cTnI forms: free cTnI, the T-C binary complex; the T-M ternary complex; and oxidized, reduced, and phosphorylated isoforms of the three cTnI forms.
3. The effects of different anticoagulants on binding of cTnI need to be addressed.
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The source of material used to calibrate cTn assays, specifically for cTnI, should be traceable.

While clinicians and laboratorians continue to publish guidelines supporting turnaround times (TATs), defined as the time from blood draw to reporting of the result to a health care provider), of less than 60 minutes for cardiac biomarkers, the largest TAT study published to date demonstrated that TAT expectations are not being met in a large proportion of hospitals. A survey study of 7020 cardiac troponin and 4368 CK-MB determinations in 159 hospitals demonstrated that the median and 99th percentile TATs for troponin and CK-MB were as follows, respectively: 7.5 min, 129 min; 82 min, 131 min. Less than 25% of hospitals were able to meet the TAT in less than 60 minutes. Preliminary data has shown that implementation of POC cardiac troponin testing can decrease TATs to less than 30 minutes in cardiology critical care and short-stay units. These data highlight the continued need for laboratory services and health care providers to work together to develop better processes to meet a TAT that is less than 60 minutes as requested by physicians.

Reference Intervals for Cardiac Troponins and Creatine Kinase Isoenzymes

If possible, each laboratory should determine a 99th percentile of a reference group for cardiac troponin assays using the specific assay used in clinical practice or validate the assay based on findings in the literature. Further, acceptable imprecision (coefficient of variation, %CV) of each cardiac troponin assay (as well as for CK-MB mass assay) has been defined as 10% or lower CV at the 99th percentile reference limit. Unfortunately, the majority of laboratories do not have the resources to perform adequately powered reference interval studies. Therefore, clinical laboratories have to rely on the peer-reviewed published literature to establish reference intervals.

When reviewing reference studies, caution must be taken when comparing the findings reported in the manufacturer's approved package inserts with the findings reported in journals because of differences in total sample size, distributions by gender and ethnicity, age ranges, and the statistic used to calculate the 99th percentile given.

There is no established guideline set to mandate a consistent evaluation of the 99th percentile reference limit for cardiac troponins. The largest and most diverse reported reference interval study to date showed plasma (heparin; used for anticoagulation of blood) 99th percentile reference limits for eight cardiac troponin assays (seven cTnI, one cTnT) and seven CK-MB mass assays (Table 5-2). These studies were performed in 696 healthy adults (age range 18 to 84 years) stratified by gender and ethnicity. The data demonstrate several issues. First, two cTnI assays showed a 1.2- to 2.5-fold higher 99th percentile for males versus females. Second, two cTnI assays demonstrated a 1.1- to 2.8-fold higher 99th percentile for African Americans versus Caucasians. Third, there was a 13-fold difference between the lowest versus the highest measured cTnI 99th percentile limit. The lack of cardiac troponin assay standardization (there is no primary reference material available) and the differences in antibody epitope recognition between assays (different assays use different antibodies) give rise to substantially discrepant results.

Although many studies have addressed the total imprecision of cTn assays, the manufacturers' package inserts prefer to publish imprecision data primarily based on within-run or within-day precision. Again, there is no consistent specification regarding the precision value that should be reported in the package insert. Published findings demonstrating the total imprecision for 13 commercial assays have indicated none of the assays were able experimentally to achieve a 10% CV (total imprecision) at their 99th percentile cutoff.

Therefore, to avoid the potential for false-positive diagnostic criteria based on cTn monitoring at the 99th percentile, a group of experts in both the laboratory medicine and cardiology communities has endorsed the concept that until cardiac troponin assay imprecision improves at the low concentrations, the lowest concentrations to attain a 10% CV (CV is the same as precision) should be used as a modified ESC/ACC diagnostic cutoff for detection of myocardial injury. This concept has been endorsed by several cardiology and laboratory medicine groups. The ultimate goal will be to have all cTnI assays attain a 10% CV at the 99th percentile reference limit. This approach should reduce false-positive analytic results from lack of imprecision values between the 10% CV cutoff and the 99th percentile. However, all biomarker increases above the 99th percentile should be interpreted cautiously, especially in the high-risk patient, and
Table 5-2. Heparin-Plasma 99th Percentile Reference Limits (μg/L) by Gender and Race for Cardiac Troponin Assays Cleared by the Food and Drug Administration

<table>
<thead>
<tr>
<th></th>
<th>Abbott</th>
<th>Beckman</th>
<th>Dade</th>
<th>OCD</th>
<th>Roche</th>
<th>Tosoh</th>
<th>Bayer</th>
<th>DPC</th>
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<tbody>
<tr>
<td>All</td>
<td>696</td>
<td>0.8</td>
<td>0.08</td>
<td>0.06</td>
<td>0.10</td>
<td>&lt;0.01</td>
<td>475</td>
<td>0.07</td>
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<tr>
<td>Male</td>
<td>315</td>
<td>0.8</td>
<td>0.10</td>
<td>0.06</td>
<td>0.11</td>
<td>&lt;0.01</td>
<td>223</td>
<td>0.07</td>
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<tr>
<td>Female</td>
<td>381</td>
<td>0.7</td>
<td>0.04</td>
<td>0.06</td>
<td>0.09</td>
<td>&lt;0.01</td>
<td>250</td>
<td>&lt;0.06</td>
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<tr>
<td>P</td>
<td>.739</td>
<td>.034</td>
<td>.985</td>
<td>.017</td>
<td>534</td>
<td>.521</td>
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<td>Caucasian</td>
<td>400</td>
<td>0.8</td>
<td>0.07</td>
<td>0.04</td>
<td>0.11</td>
<td>&lt;0.01</td>
<td>215</td>
<td>&lt;0.06</td>
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<tr>
<td>African-American</td>
<td>218</td>
<td>0.5</td>
<td>0.08*</td>
<td>0.03</td>
<td>0.10</td>
<td>&lt;0.01</td>
<td>196</td>
<td>0.17</td>
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*Number of samples tested in the Abbott, Beckman, Dade-Behring, OCD, and Roche assays.

1 Number of samples tested in the Tosoh assay.

2 Number of samples tested in the Bayer assay.

3 Number of samples tested in the DPC assay.

4 Significantly different (P = .05) from Caucasians based on mean concentrations.

DPC = Diagnostics Products Corporation
OCD = Ortho-Clinical Diagnostics

followed with serial samples over a 6- to 12-hour period after presentation.

Myocardial Infarction Detection Rates and Cardiac Troponins

Advances in diagnostic technology for the development of improved low-end analytical detection of cardiac troponins have impacted the prevalence of AMI detection. Accumulating data suggest that the more sensitive cardiac troponin tests result in greater rates of MI diagnosis and greater rates of cardiac troponin positivity, compared to CK-MB. Smaller MIs will be detected. Clinical cases that were earlier classified as unstable angina will be given a diagnosis of MI (due to an increased cTn), and now procedure-related troponin increases (i.e., following angioplasty) will be labeled MI.

The importance of small troponin increases has been confirmed by their association with a poor prognosis. Based on several studies that compared CK-MB and cardiac troponin assays in patients with ACS, a substantial increase in rate of MIs ranging from 12% to 127% was detected. In one study of 1719 patients with ACS presenting to rule in/rule out MI, a subset (5%) of cTnI-negative but CK-MB-positive patients revealed the potentially underlying false-positive MI rate when using CK-MB as a standard for MI detection. This was likely due to release of CK-MB from skeletal muscle, in the absence of myocardial injury. Further, a subset (12%) of cTnI-positive, CK-MB-negative patients demonstrated a subset of MIs that would not have been detected without cardiac troponin monitoring. All these data taken together support the implementation of cardiac troponin in place of, not in combination with, CK-MB. This fact will then have an impact on the prognosis of AMI overall.

Creatine Kinase MB

CK-MB can be measured in numerous ways. Immunoassays developed in recent years have improved on the analytical and clinical sensitivity and specificity of the earlier immunoinhibition and immunoprecipitation assays. These assays now: (1) measure CK-MB directly and provide mass measurements, (2) are easily automated, and (3) provide rapid results (<30 minutes). Mass assays reliably measure low CK-MB concentrations in both samples with low total enzyme activity (<100 U/L) and with high total enzyme activity (>10,000 U/L). Furthermore, no interferences from other proteins have been documented. The majority of commercially available immunoassays that use monoclonal anti-CK-MB antibodies are the same as those listed in Table 5-2 for cardiac troponin assays. Excellent concordance has been shown between mass concentration and activity assays. A primary reference material is commercially available to assist in harmonization. If used for assay standardization, then this material allows
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concentrations to be reported within 20% of each other.

As has been recognized for years for total CK, all CK-MB assays demonstrate a significant 1.2- to 2.6-fold higher 99th percentile for males versus females. Several assays showed higher, up to 2.7-fold, concentrations for African-Americans versus Caucasians. These data demonstrate that clinical laboratories must consider establishing different CK-MB reference cutoffs for at least men versus women.

CLINICAL UTILIZATION OF CARDIAC BIOMARKERS

Use in Patients with Acute Coronary Syndrome

The ideal marker of myocardial injury should: (1) provide early detection of injury, (2) allow rapid diagnosis of cardiac injury, (3) serve as a risk stratification tool in patients with ACS, (4) assess the success of reperfusion after thrombolytic therapy, (5) detect reocclusion and reinfarction, (6) determine the timing of an infarction as well as infarct size, and (7) detect procedural-related perioperative MI during cardiac or noncardiac surgery. At present, the perfect biomarker to satisfy all these needs does not exist. It is the function of the laboratory to provide advice to physicians about cardiac biomarker characteristics.

Patients present to emergency departments or other primary care providers with a multitude of clinical signs and symptoms for which the differential diagnosis of AMI (heart attack) is considered. Figure 5-3 demonstrates the complete spectrum of clinical presentations has been designated ACS. The cornerstone of the redefinition of MI is predicated on cardiac biomarkers, specifically cTnI or cTnT. The following are designated as biochemical indicators for detecting myocardial necrosis:

1. A maximal concentration of cTnI or cTnT exceeding the decision limit, defined as the 99th percentile of values for a reference control group, on at least one occasion during the first 24 hours after the index clinical event.

2. A maximal value of CK-MB (preferably mass) exceeding the 99th percentile of values for a reference control group on two successive samples or a maximal value exceeding twice the upper reference limit during the first hours after the index clinical event. Although the consensus document states values for troponin and CK-MB should rise and fall, either a rising or a falling pattern should be considered diagnostic. Values that remain elevated without change are rarely due to MI.

3. In the absence of availability of a cardiac troponin or CK-MB assay, total CK greater than two times the upper reference limit may be employed.

In addition to the ESC/ACC consensus document for redefining MI, the ACC/American Heart Association guidelines for management of unstable angina recommend monitoring cardiac troponin in patients with ACS as a way of differentiating unstable angina (defined as when cardiac troponin is within the 99th percentile reference limit) and non-ST segment-elevation MI (defined as when cardiac troponin is increased above the 99th percentile reference limit).

Several markers should no longer be used to evaluate cardiac disease, including aspartate aminotransferase, total CK, total lactate dehydrogenase (LDH), and LDH isoenzymes. Due to their wide tissue distribution, these markers have poor specificity for the detection of cardiac injury. Because total CK and CK-MB have served as standards for so many years, some laboratories may continue to measure them to allow for comparisons to cardiac troponin over time, before discontinuing use of CK and CK-MB. In addition, the use of total CK in developing countries may be the preferred or only alternative for financial reasons. However, it should be clear that, for monitoring ACS patients to assist in clinical classification, cardiac troponin is the preferred biomarker.

For the majority of patients, blood should be obtained for testing at hospital admission (0 hours), at 6 to 9 hours, and again at 12 to 24 hours if the earlier specimens are normal and the clinical index of suspicion is high. For patients in need of an early diagnosis that would parallel a rapid triage protocol, a rapidly appearing biomarker such as myoglobin has been suggested to be added to serial cardiac troponin monitoring. In practice, it appears that the majority of hospitals throughout the world do not use these markers.

Several general clinical impressions can be made regarding cTnI and cTnT. First, the early
release kinetics of both cTnI and cTnT are similar to those of CK-MB after AMI, with increases above the upper reference limit seen at 2 to 6 hours. The initial increase is due to the 3% to 6% cytoplasmic fraction of troponin (CK-MB is 100% cytoplasmic). Second, cTnI and cTnT can remain increased up to 4 to 14 days after AMI. The mechanism is likely the ongoing release of troponin from the 94% to 97% myofibril-bound fraction since the half-life in clearance studies of either the native protein or of complexes is in the range of 2 hours. The long interval of cardiac troponin increase has resulted in its utilization in place of the LDH isoenzyme assay in the detection of late-presenting AMI patients. Third, the very low to undetectable cardiac troponin values in serum from patients without cardiac disease (normal, healthy reference population) permits use of lower discriminator concentrations compared to CK-MB for the determination of myocardial injury. Finally, cardiac tissue specificity of cTnI and cTnT should eliminate false clinical impression of AMI in patients with increased CK-MB concentrations following skeletal muscle injuries.

Clinical use of the percentage relative index [%RI; %RI = (CK-MB mass/Total CK activity × 100)] or %CK-MB (CK-MB activity/Total CK × 100) aids in the interpretation of CK-MB concentrations for the detection of AMI. While not absolute, an increased %CK-MB or %RI points to the heart as the source of CK-MB in serum. However, the %RI and %CK-MB should not be used for interpretation when the total CK activity remains within the reference interval. Any concomitant skeletal muscle injury will decrease the sensitivity of the relative index for the detection of cardiac events.

As increases in cardiac troponin detect any form of myocardial injury, nonischemic mechanisms of injury are also responsible for cardiac troponin release from the heart, causing increases in circulating troponin. Table 5-3 shows a list of potential etiologies that have been responsible for increases in non-ischemic damage to the heart. Thus, whenever cardiac troponin is monitored, it is important to follow the serial pattern of a rising or a falling pattern of the biomarker. An increased cTn pattern that remains relatively unchanged and is not indicative of this serial trend is likely not an MI.

Strategies for the Role of Cardiac Troponin for Risk Assessment

Numerous prospective and retrospective clinical studies have evaluated and compared the utility of measurements of cTnI and cTnT for risk stratification or clinical outcomes assessment of patients with ACS with possible myocardial
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Table 5-3. Elevations of Troponins without Overt Ischemic Heart Disease

<table>
<thead>
<tr>
<th>Condition</th>
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<tr>
<td>Trauma (including contusion, ablation, pacing, cardioversion)</td>
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<tr>
<td>Congestive heart failure, acute and chronic*</td>
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<tr>
<td>Aortic valve disease and hypertrophic obstructive cardiomyopathy with significant left ventricular hypertrophy*</td>
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<tr>
<td>Hypertension</td>
</tr>
<tr>
<td>Hypotension, often with arrhythmias</td>
</tr>
<tr>
<td>Postoperative noncardiac surgery patients who seem to do well*</td>
</tr>
<tr>
<td>Renal failure*</td>
</tr>
<tr>
<td>Critically ill patients, especially with diabetes, respiratory failure*</td>
</tr>
<tr>
<td>Drug toxicity, such as adriamycin, 5-fluorouracil, herceptin, snake venom*</td>
</tr>
<tr>
<td>Hypothyroidism</td>
</tr>
<tr>
<td>Coronary vasospasm, including apical ballooning syndrome</td>
</tr>
<tr>
<td>Inflammatory diseases such as myocarditis (e.g., with parvovirus B19, Kawasaki disease, sarcoidosis, smallpox vaccination, or myocardial extension of bacterial endocarditis)</td>
</tr>
<tr>
<td>Postpercutaneous coronary intervention patients who appear without complication*</td>
</tr>
<tr>
<td>Pulmonary embolism, severe pulmonary hypertension*</td>
</tr>
<tr>
<td>Sepsis*</td>
</tr>
<tr>
<td>Burns, especially if total burn surface area &gt;30%*</td>
</tr>
<tr>
<td>Infiltrative diseases, including amyloidosis, hemochromatosis, sarcoidosis, and scleroderma*</td>
</tr>
<tr>
<td>Acute neurological disease, including cerebrovascular accident, subarachnoid bleeds*</td>
</tr>
<tr>
<td>Rhabdomyolysis with cardiac injury</td>
</tr>
<tr>
<td>Transplant vasculopathy</td>
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<tr>
<td>Vital exhaustion</td>
</tr>
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*Designations imply prognostic information has been reported.

ischemia in the emergency department. Patients presenting with a complaint of chest pain or other symptoms suggesting ACS have been assigned to blood-sampling protocols including only a single draw at presentation as well as to several serial blood samplings over a 12- to 24-hour period following presentation. Overall, in the approximately 18,000 patients studied, at 30 days the odds ratio for an adverse outcome was 3.4 for increased troponin. As both cTnT and cTnI offer powerful risk assessment, cTn monitoring needs to be included in current practice guidelines not only regarding diagnosis and management of ACS patients, but also as useful risk stratification tools. It is recommended to draw two samples on patients with ACS who do not rule in for AMI: one at presentation and one at 6 to 9 hours following presentation. This will allow for an increase in either cardiac troponin to occur above baseline in a patient presenting with a very recent acute coronary lesion. However, it should be noted that a normal cardiac troponin does not remove all risk.

Several studies have now documented that assays with lower limits of detection are able to identify more patients with ACS with poor prognosis who may be candidates for early invasive procedures. In one representative study (Venge et al., 2000), two assays were compared to assess clinical performance in unstable patients with CAD. While both assays showed patients with normal cTnI levels had a significantly better prognosis than patients with increased levels, a cohort of 11% of patients (n = 98) with a poor prognosis was identified only by the second-generation assay with a lower limit of detection. Invasive treatment only reduced clinical events in the group of patients with increased cTnI. Thus, each troponin assay, I or T, needs to evaluate the stratification of patients at low-end concentrations to avoid the potential of analytical inaccuracies leading to inappropriate management decisions and therapy. These high-risk patients have been shown to benefit from aggressive therapies, including low molecular weight heparin, IIb/IIIa glycoprotein platelet inhibitors, and an early interventional strategy.

Clinicians are often confronted with a clinical history of a patient without overt CAD and a low probability of myocardial ischemia. However, as a
precautionary reflex, serial cardiac biomarkers, specifically cTn, are ordered. The 20% of suspected ACS patients who clinically do not rule in for MI but display an increased cTn represent those nonischemic pathologies such as myocarditis, blunt chest trauma, or chemotherapeutic agents for which the mechanisms of injury are well defined (Table 5-3) as well as the unexpected finding of myocardial injury, for which patients have been shown to have increased cTn, but the mechanism of release is not clear. These observations have led to important and novel investigations involving patients with nonischemic heart disease and the role for cardiac troponin as a diagnostic and prognostic tool. The conditions shown in Table 5-3 that are indicated by an asterisk demonstrate that, in addition to cardiac troponin being indicative of cardiac injury, the data have also indicated that increased cardiac troponin is useful as a prognostic tool for assessing risk of death and MI.

Monitoring Reperfusion Following Thrombolytic Therapy

Release of cTnl or cTnI from myocardium into the blood following AMI and after the washout that accompanies successful reperfusion generates an excellent signal compared to no detectable baseline levels prior to myocardial damage. The initial rapid release of cardiac troponin subunits I and T following successful reperfusion is most likely derived from the soluble cytosolic myocardial fraction (6% cTnI, 3% cTnI). The clinical utility of cardiac biomarkers for monitoring reperfusion following thrombolytic therapy has not gained favor as a routine form of testing for determining the success or failure of reperfusion therapy because it cannot distinguish TIMI 2 from TIMI 3 flow, which is a critical issue in regard to prognosis. (TIMI is the timed intervention in myocardial infarction. TIMI 2 and 3 refer to the extent of flow through coronary vessels, with TIMI 2 referring to partial flow and TIMI 3 to complete flow.)

It is accepted that the kinetics of myocardial protein appearance in the circulation depends on infarct perfusion. Early reperfusion causes an earlier increase above the upper reference limit and an earlier and greater enzyme peak after reperfusion. However, once the peak has occurred, there is no difference in the time of clearance of enzymes. In addition, enhanced washout identifies whether an artery is patent or closed but cannot distinguish between normal and abnormal coronary perfusion, which is another key prognostic parameter. Further, it is difficult to assess the amount of irreversible myocardial injury by infarct sizing because of the large variability in the amount of enzyme washout that appears after reperfusion.

Strategies Using Multimarkers

There is a growing body of evidence suggesting that different cardiac biomarkers provide independent and complementary information about pathophysiology, diagnostics, prognostics, and response to therapy in patients with ACS. Thus, it is probable that multimarker strategies or biochemical profiling may be used to characterize individual patients presenting with ACS. For example, in one multicenter study, cTnl, CK-MB, and myoglobin multimarker analysis identified positive patients earlier and provided a better risk stratification for 30-day mortality than central laboratory analysis of CK-MB alone (20% vs. 3%, respectively).

Estimation of Infarct Size

Older studies have shown that one can use the integrated values for total or CK-MB to estimate the biochemical extent of infarction. Further studies verified that the amount of cardiac damage is the primary determinant of prognosis. Such determinations have been correlated with morphological infarct size. Some have used the peak CK-MB value as a surrogate for the integrated data. Reperfusion changes the release ratio (the percentage of marker that appears in the blood relative to the amount depleted from myocardium), making infarct sizing problematic in the modern era. Additional data from both experimental and patient-related data have suggested that the 72-hour troponin measurement correlates with scintigraphically-determined infarct size. The data are stronger for troponin T than for I, although the principles are probably similar for both analytes. At present, it is not recommended that serial monitoring of cardiac troponin or CK-MB be used for infarct sizing.

Reinfarction

Figure 5-1 demonstrates the serial patterns for cardiac troponin I and CK-MB both during the initial infarct and reinfarction, as described for the initial case presentation. Although the number of reinfarction cases presented in the
Cardiac Troponin in MI Diagnosis

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QUESTIONS

1. Diagram the rising and falling serial biomarker
profiles for cardiac troponins compared to CK-MB.

2. Define the ESC/ACC consensus guidelines
for the redefinition of AMI.

3. Review the role of cardiac biomarkers for
infarct sizing and monitoring the success of
reperfusion following therapy.

4. List the multiple forms of cardiac troponin
I that may exist in the circulation.

5. Define the two major concerns that have
been responsible for substantial concentration
differences when measuring cardiac troponin I by different commercial
immunoassays.

6. Explain how the determination of reference
intervals can have an impact on the number
of MI cases that are defined in a
given population of patients with chest
pain.

7. Explain the role of cardiac troponin deter-
minations in risk-stratifying patients who
present with ACS.

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