

2

Analytical Issues for Clinical Use of Cardiac Troponin

Alan H. B. Wu, PhD

CONTENTS

INTRODUCTION
RELEASE OF TROPONIN AFTER MYOCARDIAL INJURY
QUALITY SPECIFICATIONS FOR ANALYTIC ASSAYS
AMI CUTOFF CONCENTRATIONS AND ASSAY IMPRECISION
FOR TROPONIN
CARDIAC TROPONIN CUTOFF FOR RISK STRATIFICATION
PRE- AND POSTANALYTIC VARIABLES
CONCLUSION
REFERENCES

SUMMARY

Guidelines jointly developed by the European Society of Cardiology and the American College of Cardiology have established cardiac troponin (T or I) as the biomarker of choice for the diagnosis of acute coronary syndromes (ACSs) and risk stratification of patients who present with ischemic symptoms suggestive of ACS. Despite these international guidelines, a number of analytic issues have slowed the acceptance and implementation of this test worldwide. Regarding precision, analytic sensitivity, and specificity, the performance of commercial assays is variable. Moreover, there is a lack of assay standardization for troponin I results among commercial assays. The release of a standard reference material should initiate the process of assay harmonization. It is important that clinicians be familiar with the analytic performance (i.e., level of 10% coefficient of variation) and appropriate cut points (99th percentile and/or evidence-based decision limits) for all troponin assays used (both laboratory-based and point of care) at their institution. In time, it is hoped that all troponin assays will be standardized and exhibit similar performance to each other, as is the case for the majority of other clinical laboratory analytes today.

Key Words: Troponin; imprecision; interference; harmonization; false positive.

From: *Contemporary Cardiology:
Cardiovascular Biomarkers: Pathophysiology and Disease Management*
Edited by: David A. Morrow © Humana Press Inc., Totowa, NJ

INTRODUCTION

Guidelines that were jointly developed by the European Society of Cardiology (ESC) and the American College of Cardiology (ACC) have established cardiac troponin T (cTnT) or cTnI as the biomarker of choice for the diagnosis of acute coronary syndromes (ACSs) and risk stratification of patients who present with ischemic symptoms suggestive of ACS (1). Despite the international guidelines on the use of troponin by cardiologists, emergency department (ED) physicians, and clinical laboratory scientists (1–3), there are a number of analytic issues that have slowed the acceptance and implementation of this test worldwide. The performance of commercial assays is quite variable regarding precision, analytic sensitivity, and specificity. Furthermore, there is a lack of assay standardization for troponin I results among commercial assays. Recently, the National Institute of Standards and Technology (NIST) released a standard reference material that should initiate the process of assay harmonization. These problems in cardiac troponin have led to confusion as to the proper cutoff concentrations that should be used in routine clinical practice and in clinical trials. In addition, the reagent costs for cardiac troponin are higher than for other older markers such as creatine kinase (CK) and the CK-MB isoenzyme. In this chapter, these important issues are discussed with reference to their impact on the clinical interpretation of test results.

RELEASE OF TROPONIN AFTER MYOCARDIAL INJURY

cTnT and cTnI are part of a complex of three regulatory proteins that includes troponin C. This ternary complex is bound to the thin filament of striated muscle and regulates the contraction of actin and myosin filaments. Following myocardial damage, the troponin T-I-C complex is gradually released into the blood, where it degrades into the binary I-C complex and free troponin T, the predominant forms of cardiac troponin in the circulation (Fig. 1) (4). In addition, a small amount of free troponin T and I exists within the cytoplasm and appears within the initial hours after the onset of cardiac damage (5). In Fig. 2, peak A illustrates the biphasic release pattern of cardiac troponin from damaged myocytes. Troponin I can also exist in phosphorylated forms and in oxidized/reduced forms. The degree of phosphorylation affects cardiac myocyte contractility in normal and failing hearts (6). Once in the blood, the various troponin forms undergo further degradation into smaller molecular weight fragments by serum proteolytic enzymes (*see next section*) (7).

An ongoing controversy is whether or not troponin can be released following reversible ischemia. Cardiac markers that are large in molecular weight, such as CK-MB (84 kDa) and lactate dehydrogenase (135 kDa), are released into the blood only after irreversible injury. However, some investigators have questioned whether cTnT and cTnI, at 37 and 24 kDa, respectively, can be released during ischemia (8,9). Although these proteins are themselves too large to traverse across viable cell membranes, studies have shown that troponin can undergo *in situ* degradation into smaller fragments during prolonged periods of myocardial ischemia (10). Using denaturing Western blot analysis, troponin fragments appear in the blood of patients with acute coronary syndromes within the first hour after the onset of chest pain, well before detection by commercial troponin assays (11). Others have suggested that the release of troponin following reversible injury is not likely (12). Although the debate is intellectually interesting, most agree that a definitive answer regarding this area of question will be difficult, if not impossible, to prove (13).

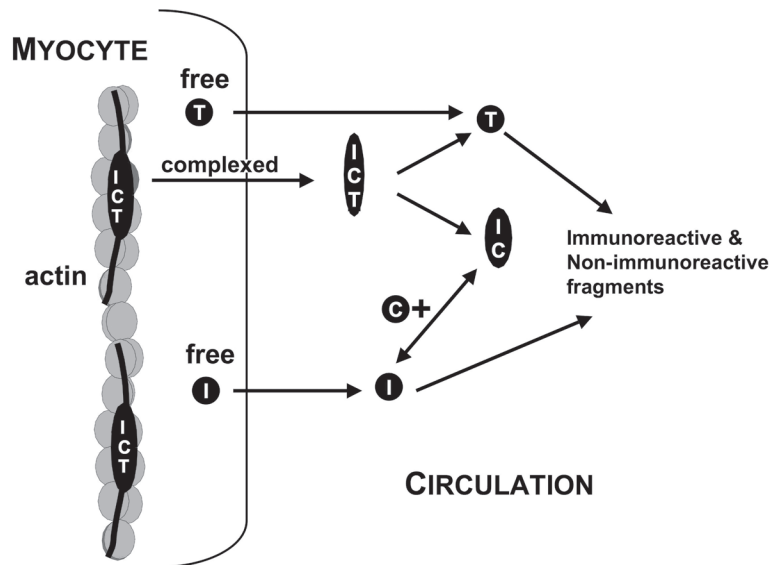


Fig. 1. The troponin T-I-C complex of the thin filament is released from damaged myocytes into various molecular forms. The T-I-C complex appears in the blood and degrades first into the I-C complex (the predominant form of cTnI in blood) and free TnT. The I-C complex further degrades into free troponin subunits and fragments of intact subunits (both immunoreactive and nonimmunoreactive). A small proportion of troponin I and T and fragments is also found in the cytoplasm. Troponin I can exist in either oxidized or reduced forms, or up to two phosphorylations.

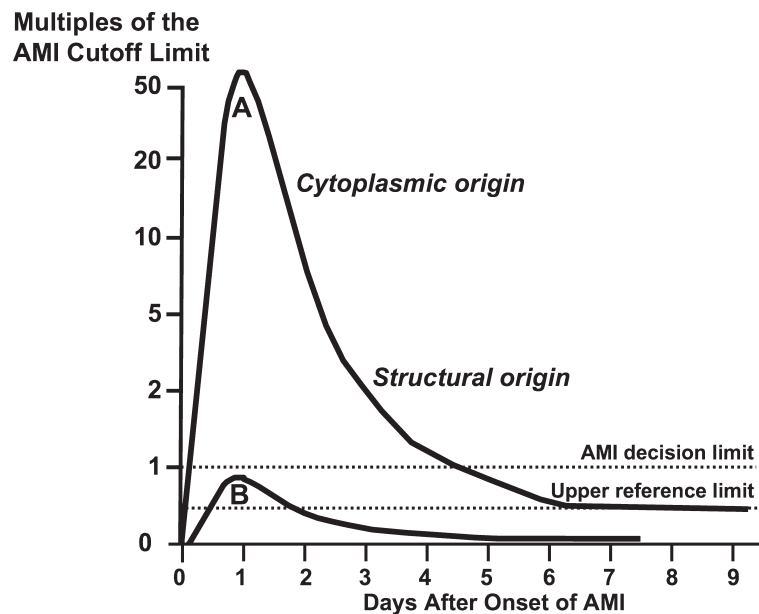


Fig. 2. Biphasic release pattern of cardiac troponin and relationship of cutoff concentrations. Peak A, myocardial infarction (MI); peak B, unstable angina. AMI, acute myocardial infarction. (Modified from ref. 3.)

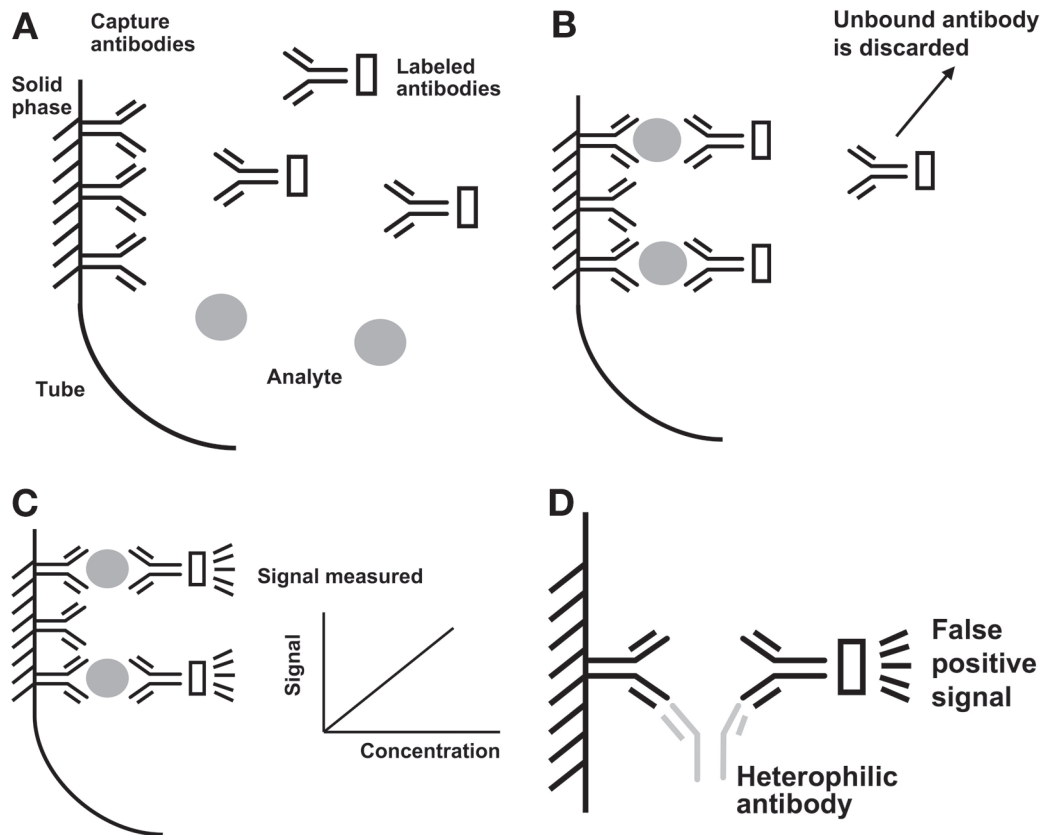


Fig. 3. (A) Two-site (“sandwich”) immunoassays. The capture antibody is immobilized to a solid surface (e.g., test tube wall). The detection antibody is labeled with a detecting enzyme, fluorophore, or chemiluminescent tag. (B) The presence of analytes (circles) enables capture. Unbound labeled antibody is removed. (C) The signal of the labeled antibody (left) is generated and measured. The concentration of the analyte is extrapolated from a calibration curve (right). (D) Mechanism for interferences owing to the presence of unusual antibodies. Heterophilic or human antimouse antibodies bind to both the capture and detection antibodies, producing an analytic signal in the absence of the analyte. (Modified from ref. 14).

QUALITY SPECIFICATIONS FOR ANALYTIC ASSAYS

All assays for cardiac troponin require the use of immunoassay techniques. Two-site “sandwich” immunoassays make use of a capture antibody to bind to the analyte of interest, and a labeled antibody that is used to determine the quantity that is bound to the capture antibody (Fig. 3A–C). The concentration is determined from a calibration curve (plot of the analytic signal vs concentration of calibrators). An immunoassay analyzer is used to measure these proteins. Point-of-care (POC) testing devices offer an alternative to a large analyzer/central laboratory testing approach (*see* Chapter 32). Because the analytic performance of assays for cardiac markers can have a major impact on how results are interpreted, the International Federation of Clinical Chemistry Committee on Standardization of Markers of Cardiac Damage (IFCC C-SMCD) developed quality specifications for cardiac troponin assays (15). Table 1 summarizes the major recommendations made by this committee.

Table 1
Quality Specification for Cardiac Troponin Assays

<i>Antibody specificity:</i> Antibodies in troponin assays should recognize the stable part of the molecule and should not be affected by complex formation or in vivo modifications.
<i>Calibration:</i> The natural and native troponin ternary complexes should be used.
<i>Sample dilution:</i> There should be no matrix effects for immunoassays, as determined by superimposable curves on dilution of high samples.
<i>Assay specificity:</i> There should be no crossreactivity with heterophilic, rheumatoid factor, or human antianimal antibodies.
<i>Documentation of preanalytic factors:</i> The types of blood collection tube and in vitro stability at different temperatures should be documented.

Adapted from ref. 36.

Epitope Specificity of Commercial Antibodies

The performance of immunoassays is greatly dependent on the reactivity of the antibodies used toward the epitopes of the targeted proteins. Proper selection of antibodies is particularly important for troponin assays, because blood from patients with ACS contains a variety of different forms of troponin (Fig. 1). Whereas some assays produced an equimolar response to these forms, others produced a higher signal to the binary and ternary troponin forms (16). Thus, a tight linear regression of troponin I results from human patients was not achieved with a comparison of early commercial troponin assays (e.g., $r = 0.811$ for Beckman Access vs Stratus) (17).

The primary sites of *in situ* and in vitro degradation for cTnT and cTnI are the C- and N-terminal sequences. These low-molecular-weight fragments are rapidly cleared from the circulation by glomerular filtration. As a consequence, assays that use antibodies directed toward the central stable portion of the molecule will exhibit greater relative increases in concentration over time and a longer duration of elevation (Fig. 4) (18). Antibodies directed toward the fragments will have a limited window of detectability. However, if troponin is released during reversible ischemia, development of assays directed to the N- and C-terminal fragments (the unstable parts of the molecule) may have added clinical utility.

Assay Standardization

A major issue for cTnI assays is the current lack of industry standardization among commercial assays. Although this is not a problem for troponin T assay, because only one manufacturer (Roche) has the intellectual property rights for use of this test, it is an issue for other cardiac markers such as myoglobin and CK-MB (19).

CLINICAL IMPLICATIONS OF LACK OF STANDARDIZATION

Figure 5 illustrates the extent of the problem for cTnI regarding the lack of standardization. The data are from the College of American Pathologists 2004 Proficiency Survey (20). Although each of the commercial assays demonstrates linearity between individual concentrations and the zero point (demonstrating minimal *offset bias*), there is substantial *proportional bias* between assays, as recognized by the slope of each line against an arbitrarily selected predicate assay. Using results of assays that produce the lowest and highest cTnI results (Triage and AxSYM, respectively), the slopes differ by a factor of nearly 100 to 1, resulting in very different reported values for the same sample. For

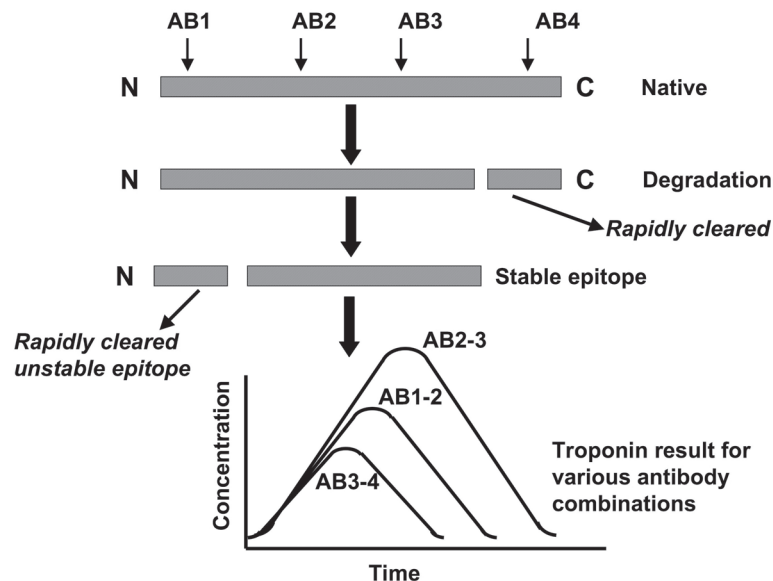


Fig. 4. Effect of antibody selection on measurement of cardiac troponin. (**Top**) Epitopes to four antibodies to the intact primary amino acid sequence of cTnI are shown. After troponin is released from the blood, it undergoes degradation at the C-terminal and then N-terminal sequences. These smaller peptides are rapidly removed from the circulation. (**Bottom**) Dual-site troponin assay using various combinations of antibodies. Assay AB3–4 shows a rapid return to baseline levels, owing to removal of the C-terminal fragment; assay AB1–2 shows an intermediate return to baseline, owing to removal of the N-terminal fragment; and assay AB2–3 shows a prolonged return to baseline, because the central portion of the peptide is the most stable.

example, for a sample reported as 1.0 ng/mL with the Dimension assay, results with other assays will vary from <0.2 with the Triage to >2.0 with the AxSYM. Table 2 provides the reasons for the lack of concordance between assays. The most influential reason is the fact that different manufacturers have used different calibrator materials and value assignment of calibrator concentrations.

The lack of assay standardization poses a major problem when interpreting results generated from different assays, such as when a patient is transferred from another hospital whose laboratory uses a different cTnI assay. Confusion may also occur if the same hospital uses different testing platforms, such as a POC assay when the patient is in the ED followed by a central laboratory assay when the patient is admitted to the coronary care unit. For risk stratification of patients with ACS, differences in cTnI cutoff concentrations make it very difficult to determine the proper cutoff concentrations for one assay when the published data from clinical studies use a different cTnI assay.

DEVELOPMENT OF REFERENCE MATERIAL FOR cTnI

To address the standardization issue, the American Association for Clinical Chemistry (AACC) established The Troponin I Standardization Committee (21). This committee obtained recombinant and heart-purified cTnI materials in the T-I-C and I-C complex forms, and free cTnI. Candidate reference materials (cRMs) were characterized for purity by liquid chromatography/mass spectrometry (LC/MS), and the cTnI concentrations were assigned by a combination of amino acid analysis and LC/MS. cRMs were evaluated to

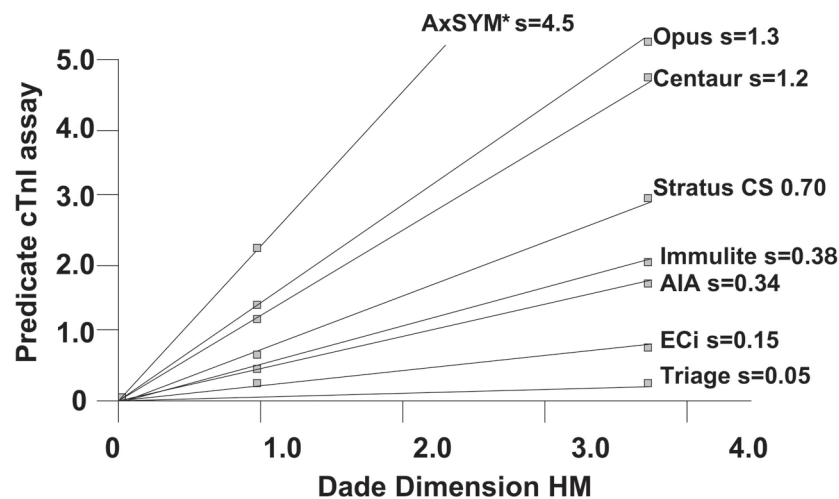


Fig. 5. Lack of standardization for cTnI assays. Data are from the 2004 College of American Pathologists CAR-A Survey for Cardiac Markers (*see ref. 20*). Although each commercial assay (y-axis) demonstrates linearity for the two survey materials, they differ in the slope (“s”) of the line relative to the Dimension HM assay (arbitrarily selected as the predicate x-axis). *First-generation assay, no longer available.

Table 2
Reasons for Lack of Concordance Among Cardiac Troponin Assays

- Lack of standardization of the calibrating materials
- Differences in the specificity of the antibodies used in the assays
- Variability in the various forms of troponin found in blood and the reactivity of antibodies to these forms
- Differences in the analytic performance of assays with particular reference to analytic sensitivity and assay imprecision

determine whether they had acceptably low matrix-associated variations (“commutability”) when diluted in human serum or a suitable diluent selected by the manufacturer, and whether they could be used as calibrators to produce identical results for different cTnI assays (“harmonization”). To select the best material, each participating manufacturer of troponin I assays was given cRMs and instructed to calibrate their analyzers using these materials, and then measure serum pools prepared by the committee containing varying concentrations of cTnI. Based on three round-robin cycles of testing, the AACC standardization committee determined that the CIT ternary complex was most commutable. Using manufacturer-specified calibrations, results of the serum pools produced a variability of 90% among results of different assays. This variability was reduced by seven- to fivefold to 12% for these same assays when the cRM troponin CIT complex was used as a calibrator (22). The NIST has certified a Standard Reference Material based on the CIT complex (SRM #2921), and it is available at www.nist.gov. One manufacturer (Abbott) has released an improved cTnI assay that is calibrated to the new NIST standard. Although other manufacturers have no obligation to use this material, many are in the process of reformulating their assays. The availability of a standard reference material will not enable complete standardization among assays, because of the variability of

epitopes targeted by antibodies used in the assays. However, results from individual patients using standardized assays will differ only by a small percentage, instead of orders of magnitude, as is the case today.

Assay Interferences

All antibody-based assays are subjected to interferences owing to the presence of unusual antibodies such as heterophiles and human antianimal antibodies. As shown in Fig. 3D, these antibodies recognize and bind to antitroponin antibodies, thereby mimicking the analyte itself. Several case reports have documented this problem (23,24). There has been increasing attention by manufacturers of troponin I assays to reformulate their assays, but the incidence of false positive results has not been eliminated (25). Although less commonly encountered, false-negative results can also occur (26).

Repeated testing showing the absence of a rise and fall characteristic of acute myocardial injury can be very helpful in suggesting the presence of an interfering substance. Reversible interference such as that from fibrin strands resulting from incomplete isolation of serum ought to resolve completely with repeat testing using appropriate sample handling (e.g., centrifugation). Testing for heterophile antibodies may be conducted by a laboratory when an interfering antibody is strongly suspected.

AMI CUTOFF CONCENTRATIONS AND ASSAY IMPRECISION FOR TROPONIN

For the majority of clinical chemistry analytes, a reference range can be established, because the analyte is present in measurable concentrations in healthy individuals. The reference range is determined by measuring the analyte in a cohort of subjects who are free from the disease in question. Ideally, the healthy subjects should be matched to the targeted disease group regarding age, gender, race, and other factors. The reference range for two-tailed tests (i.e., clinical significance for both low and high results) is calculated by the mean \pm 2 SDs if the distribution of results is parametric, or by the central 95% of results if the distribution is nonparametric. Because there is no significance for low cardiac marker results, the reference range can be computed as the upper 97.5% of healthy individuals using a one-tailed test.

In contrast to the majority of clinical chemistry analytes, cutoff concentrations for cardiac markers such as CK-MB and myoglobin were established on the basis of clinical criteria for the diagnosis of MI and not from a cohort of healthy individuals. Using receiver operating characteristic (ROC) curve analysis, which plots clinical sensitivity vs 1 – specificity, the optimum cutoff concentrations were determined at an analyte value that best discriminated between subjects with disease (i.e., AMI) and those without disease (Fig. 6). The latter category consisted of patients who presented to the ED with chest pain owing to a noncardiac etiology or who had cardiac disease that was not ruled out for AMI. Patients with unstable angina were considered to be in the non-AMI group, despite a minor increase in cardiac biomarker results. The cutoff concentrations were therefore higher than the 97.5% of a cardiac-healthy population.

The notion that biomarkers were only useful for diagnostic purposes changed with the demonstration that troponin was more sensitive than CK-MB for the detection of MI. Clinical trials showed that troponin could also be used for risk stratification of patients who were previously ruled out for AMI by the older biomarkers. The term *minor myocardial damage* was initially coined to refer to troponin concentrations that were between

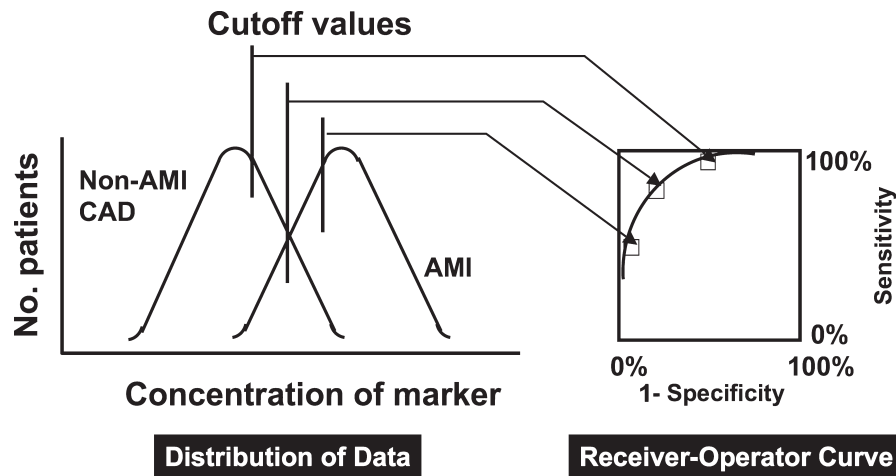


Fig. 6. ROC curve analysis plotted from distribution of cardiac marker results in disease vs a healthy population. CAD, coronary artery disease.

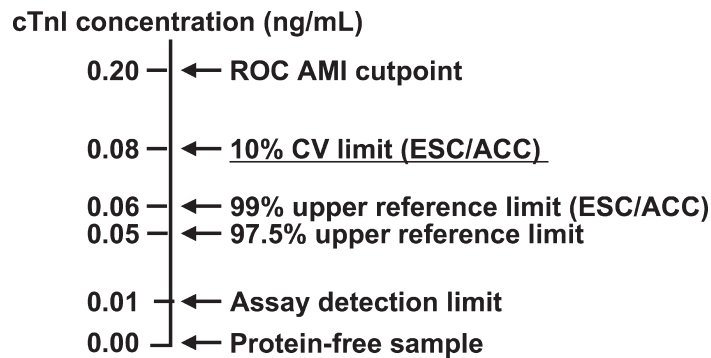


Fig. 7. Comparison of cutoff concentrations for cardiac troponin according to various criteria. (Adapted from ref. 27.)

the upper reference limit and the ROC-determined AMI cutoff concentration. Because cardiac patients with troponin concentration in this region were also shown to have a high incidence of poor outcomes, a joint committee of the ESC and ACC recommended lowering the cutoffs for cardiac markers to the upper 99% of healthy individuals (Fig. 7). Peak B in Fig. 2 shows that a patient with unstable angina may have a troponin concentration that is below the AMI decision limit but exceeds the upper reference limit of a healthy population. Use of this lower cutoff concentration posed a problem because most assays do not have the sensitivity to consistently detect troponin in the blood of healthy individuals and, therefore, the 99% cutoff could not be computed with any acceptable degree of analytic precision. Previously, the National Academy of Clinical Biochemistry had recommended that the assay imprecision be $\leq 10\%$ at the cutoff concentration (2). A subcommittee of the ESC/ACC suggested that the AMI cutoff should be the marker concentration that first produces a 10% imprecision (coefficient of variation [CV]) (9) until new assays for troponin are developed with the necessary sensitivity. Figure 8 illustrates data for several commercial troponin I assays and the value and the concentration at the 10% CV level (28). Figure 9 compares the 99th percentile (obtained from the manufacturer's

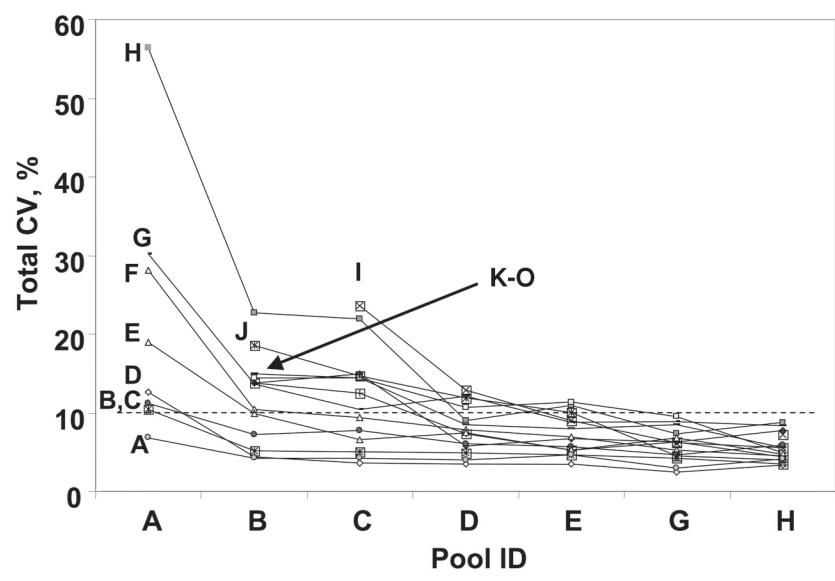


Fig. 8. Imprecision vs analyte concentration profiles for cTnI. A: AIA21; B, C: Access and Access 2; D: Stratus CS; E: Centaur; F: Immulite; G: Dimension; H: ACS:180; I: Immuno 1; J: ECI; K: Liaison; L: Opus Plus; M: Vidas; N: AxSym; O: Alpha Dx. (Reproduced from ref. 28, with permission from the American Association for Clinical Chemistry.)

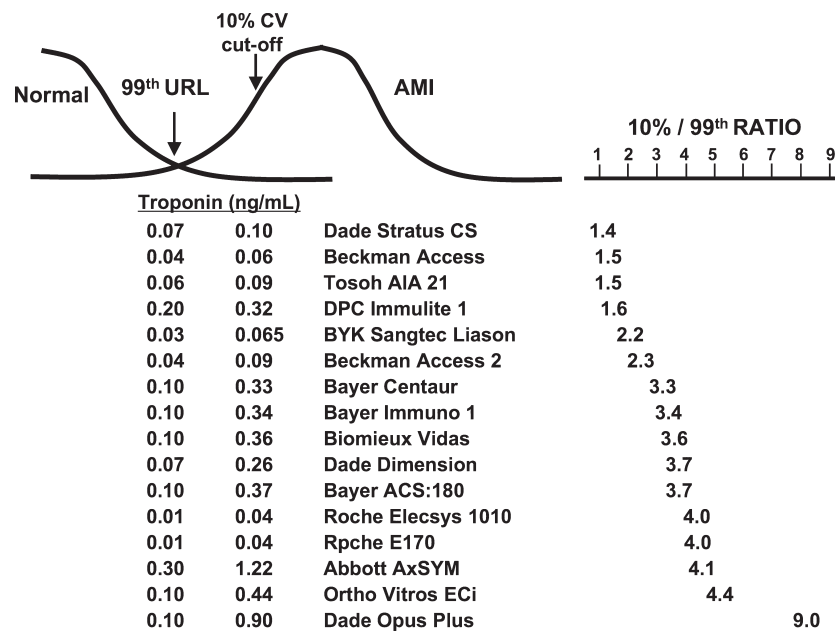


Fig. 9. Comparison of 99th percentile and 10% CV cutoffs for commercial cTnI assays. Data are taken from ref. 28. URL, upper reference limit.

package insert or personal communication from the manufacturer) to the 10% imprecision cut point (determined by the IFCC C-SMCD), and the ratio of the values for commercial cTnI assays. The smaller this ratio, the closer a particular assay is to achieving the goal of a CV $\leq 10\%$ at the 99th percentile.

	A		B		C	
	ACS		ACS		ACS	
	Yes	No	Yes	No	Yes	No
cTn NEG	<i>TN</i> 450	<i>FN</i> 50	<i>TN</i> 810	<i>FN</i> 10	<i>TN</i> 890	<i>FN</i> 1
cTn POS	<i>FP</i> 50	<i>TP</i> 450	<i>FP</i> 90	<i>TP</i> 90	<i>FP</i> 10	<i>TP</i> 99
	900	100	500	500	900	100
	High Prevalence		Low Prevalence		High Accuracy	

Fig. 10. Bayesian statistics for cardiac troponin for 1000 hypothetical patients as function of disease prevalence and assay accuracy. (A) Prevalence of 50%, accuracy of 90%; (B) prevalence of 10%, accuracy of 90%; (C) prevalence of 10%; accuracy of 99%. TP, true positive; TN, true negative; FP, false positive; FN, false negative.

Regarding the definition of MI, the British Cardiac Society Working Group has recommended the use of two cutoff concentrations for cardiac troponin (29). The first, higher cutoff corresponds to “typical ACS clinical myocardial infarction,” using concentrations for cTnT (Roche) of >1.0 ng/mL and cTnI (Beckman Access 2) of >5 ng/mL. These troponin concentrations were selected because they approximate the traditional definition of AMI, including the use of total CK. The second cutoff concentrations are set at cTnT and cTnI concentrations below 1.0 and 0.5 ng/mL, respectively, and correspond to “ACS with myocyte necrosis.” These recommendations are in contrast to the ECS/ACC recommendations and illustrate and contribute to the ongoing debate over the redefinition of AMI guidelines (30–32).

CARDIAC TROPONIN CUTOFF FOR RISK STRATIFICATION

Data from the TACTIC-TIMI 18 and FRISC-II trials have fueled debate regarding the optimum cutoff concentrations for cardiac troponin (33,34). These trials compared aggressive therapy such as with percutaneous intravascular intervention and dalteparin, respectively, against conservative treatment for patients with unstable angina. As predicted from previous studies, patients who had cardiac troponin levels above the 10% CV cut point had a higher 30-d death and MI rate than patients who had values below the upper limit of normal (99th percentile). However, patients who had troponin concentrations *between* the 99th percentile and the 10% CV cut point were also at increased risk for 30-d adverse events. For example, in the TACTICS-TIMI II Trial, the cTnI odds ratios for the 30-d death and AMI for unstable angina patients were 3.0 (95% confidence interval [CI]: 1.5–6.1) using the 10% CV cutoff, and 3.6 (95% CI: 1.8–7.3) using the 99th percentile cutoff (33). Similar findings were observed from the FRISC II Trial (34).

These data suggested that the lower (99%) cutoff concentration for cardiac troponin is the most appropriate for risk assessment in patients with a high clinical probability of ACS. Figure 10A shows that a test with a 90% accuracy produces a high ratio of true-positive to false-positive results when the prevalence of the tested population is high (e.g., 50%). However, when the prevalence for ACS is low (e.g., 10% for a general ED chest pain population), use of this same 90% accurate troponin assay will result in an unacceptably high false-positive rate (equal numbers of false positives and true positives) (Fig. 10B).

Thus, for a low disease prevalence population, the accuracy of the assay must be higher (e.g., to 99%, as shown in Fig. 10C) (35). Precision is an important attribute to *analytic* accuracy, because assays that are imprecise cannot be accurate. However, for use of troponin in ACS, if there is sufficient separation in troponin concentrations between low- and high-risk groups, the imprecision may have little effect on the *clinical* accuracy of the test.

PRE- AND POSTANALYTIC VARIABLES

In addition to the analytic variables of assay performance, the quality of any laboratory data is also greatly dependent on pre- and postanalytic variables. Preanalytic variables include the types of blood collection tubes used and the time and manner by which specimens are transported to the laboratory. An important preanalytic variable is the stability of the analyte.

Collection of Samples

Most commercial assays can accommodate either serum or plasma (anticoagulated with heparin or EDTA), but there are exceptions (e.g., only serum for Roche Elecsys cTnT, only heparinized plasma for Ortho ECi cTnI, and whole blood or EDTA plasma for Biosite Triage cTnI). The user should consult the clinical laboratory for specific blood collection details. For sample stability, troponin begins to degrade soon after blood is collected. Antibodies of most commercial assays today are directed to the stable or conserved portion of the protein and, thus, troponin instability is not a major issue with contemporary assays, especially because testing is usually conducted immediately after collection.

Turnaround Time

Postanalytic variables include the overall turnaround time between ordering the test and reporting the results. Both cardiology and laboratory medicine groups recommend a turnaround time of 60 min for cardiac markers (3,33), although the ACC/American Heart Association state that a 30-min turnaround time is preferable. Meeting this lower figure would be very difficult for testing conducted in a central laboratory given the time required for sample delivery, centrifugation, and on-instrument analysis (typically 10–20 min). Increasingly, therapeutic management decisions are relying on the results of troponin tests. Unnecessary delays in either the pre- or postanalytic process reduce the usefulness of these tests.

The necessary turnaround time for cardiac marker results will be institution dependent. EDs with aggressive triage protocols (“chest pain centers”) are designed to make rapid therapeutic and/or management decisions and will benefit from rapid testing (e.g., turnaround time < 30 min) that can be delivered by the use of POC or near-patient (e.g., satellite) testing (*see* Chapter 32). EDs that have a more conservative triaging strategy may be able to justify testing from the central laboratory (turnaround time ~ 1 h), because patients are not triaged as quickly. The laboratory must work closely with ED physicians and cardiologists to deliver the needed turnaround times while maintaining cost-effectiveness.

CONCLUSION

As with any laboratory test, the quality of the clinical information provided by that test is only as good as the analytic performance of the test itself. In the case of troponin, improvements in sensitivity and specificity are warranted. It is incorrect to assume that

all commercial troponin assays are the same. Under ideal conditions, a laboratory should select the best troponin assay that meets its clinical needs. However, clinical laboratories are often bound to contracts from specific manufacturers or vendors of laboratory equipment. Therefore, the assay most likely used in that institution is the one that is available on the instrument. It is important that the clinician be familiar with the analytic performance (i.e., level of 10% CV) and appropriate cut points (99th percentile and/or evidence-based decision limits) for all troponin assays used (both laboratory-based and POC) at his or her institution. It is hoped that in time all troponin assays will be standardized and exhibit similar performance to each other, as is the case for the majority of other clinical laboratory analytes today.

REFERENCES

1. Joint European Society of Cardiology/American College of Cardiology Committee. Myocardial infarction redefined—a consensus document of the joint European Society of Cardiology/American College of Cardiology Committee for the redefinition of myocardial infarction. *J Am Coll Cardiol* 2000;36:959–969.
2. Fesmire FM, Decker WW, Howell JM, Kline JA. Clinical policy: critical issues in the evaluation and management of adult patients presenting with suspected acute myocardial infarction or unstable angina. *Ann Emerg Med* 2000;35:521–544.
3. Wu AHB, Apple FS, Gibler WB, Jesse RL, Warshaw MM, Valdes R Jr. National Academy of Clinical Biochemistry Standards of Laboratory Practice: recommendations for use of cardiac markers in coronary artery diseases. *Clin Chem* 1999;45:1104–1121.
4. Wu AHB, Feng YJ. Biochemical differences between cTnT and cTnI and its significance for the diagnosis of acute coronary syndromes. *Eur Heart J* 1998;19(Suppl N):25–29.
5. Katrukha AG, Bereznikova AV, Esakova TV, et al. Troponin I is released in bloodstream of patients with acute myocardial infarction not in free form but as complex. *Clin Chem* 1997;43:1379–1385.
6. Bodor GS, Oakeley AE, Allen PD, Crimmins DL, Ladenson JH, Anderson PA. Troponin I phosphorylation in the normal and failing adult human heart. *Circulation* 1997;96:1495–1500.
7. McDonough JL, Arrell DK, Van Eyk JE. Troponin I degradation and covalent complex formation accompanies myocardial ischemia/reperfusion injury. *Circ Res* 1999;84:9–20.
8. Wu AHB, Ford L. Release of biochemical markers in acute coronary syndromes: ischemia or only necrosis? *Clin Chim Acta* 1999;284:161–171.
9. Sobel BE, LeWinter MM. Ingenuous interpretation of elevated blood levels of macromolecular markers of myocardial injury: a recipe for confusion. *J Am Coll Cardiol* 2000;35:1355–1358.
10. Murphy AM, Kogler H, Georgakopoulos D, et al. Transgenic mouse model of stunned myocardium. *Science* 2000;287:488–491.
11. Labugger R, Organ L, Collier C, Atar D, Van Eyk JE. Extensive troponin I and T modification detected in serum from patients with acute myocardial infarction. *Circulation* 2000;102:1221–1226.
12. Carlson RJ, Navone AN, McConnell JP, et al. Effect of myocardial ischemia on cardiac troponin I and T. *Am J Cardiol* 2002;89:224–226.
13. Jaffe AS, Ravkilde J, Roberts R, et al. It's time for a change to a troponin standard. *Circulation* 2000;102:1216–1220.
14. Wu AHB. Analytical issues affecting the clinical performance of cardiac troponin assays. In: Adams JE, Apple FS, Jaffe AS, Wu AHB, eds. *Markers in Cardiology: Current and Future Applications*. Future Publishing Co., Armonk, NY, 2001.
15. Panteghini M, Gerhardt W, Apple FS, Dati F, Ravkilde J, Wu AH. Quality specifications for cardiac troponin assays. *Clin Chem Lab Med* 2001;39:174–178.
16. Wu AHB, Feng YJ, Moore R, et al. Characterization of cardiac troponin subunit release into serum following acute myocardial infarction, and comparison of assays for troponin T and I. *Clin Chem* 1998;44:1198–1208.
17. Christenson RH, Apple FS, Morgan DL, et al. Cardiac troponin I measurement with the ACCESS immunoassay system: analytical and clinical performance characteristics. *Clin Chem* 1998;44:52–60.
18. Katrukha AG, Bereznikova AV, Filatov VL, et al. Degradation of cardiac troponin I: implication for reliable immunodetection. *Clin Chem* 1998;44:2433–2440.
19. Christenson RH, Duh SH, Apple FS, et al. Standardization of cardiac troponin I assays: round robin of ten candidate reference materials. *Clin Chem* 2001;47:431–437.

20. College of American Pathologists. Cardiac marker CAR-A survey participant summary report. CAP, Northfield, IL, 2004; pp. 1–30.
21. Christenson RH, Duh SH, Apple FS, et al. Towards standardization of cardiac troponin I assays: assessing commutability of candidate reference materials and assay harmonization. *Clin Chem* 2005, submitted.
22. Fitzmaurice TF, Brown C, Rifai N, Wu AHB, Jeo KTJ. False increase of cardiac troponin I with heterophilic antibodies. *Clin Chem* 1008;44:2212–2214.
23. White GH, Tideman PA. Heterophilic antibody interference with CARDIAC T Quantitative Rapid Assay. *Clin Chem* 2002;48:201–203.
24. Yeo KT, Storm CA, Li Y, et al. Performance of the enhanced Abbott AxSYM cardiac troponin I reagent in patients with heterophilic antibodies. *Clin Chim Acta* 2000;292:13–23.
25. Mahalingam M, Ottlinger ME. False-negative qualitative cardiac troponin T in a 79-year-old man with myocardial infarction. *JAMA* 1997;278:2143, 2144.
26. Braunwald E, Antman EM, Beasley JW, et al. ACC/AHA guidelines for the management of patients with unstable angina and non-ST-segment elevation myocardial infarction: executive summary and recommendations. *Circulation* 2000;102:1193–1209.
27. Wu AHB. Analytical issues and the evolution of cutoff concentrations for cardiac markers. In: Wu AHB, ed. *Cardiac Markers*, Second Edition. Humana, Totowa, NJ: 2003.
28. Panteghini M, Pagani F, Yeo KT, et al. Evaluation of imprecision for cardiac troponin assays at low-range concentrations. *Clin Chem* 2004;50:327–332.
29. Fox KAA, Birkhead J, Wilcox R, Knight C, Barth J. British Cardiac Society Working Group on the definition of myocardial infarction. *Heart* 2004;90:603–609.
30. Richards AM, Lainchbury JG, Nicholls MG. Unsatisfactory redefinition of myocardial infarction. *Lancet* 2001;357:1635, 1636.
31. Jolobe OM, Tormey W, Birkhead JS, Norris RM. Redefinition of myocardial infarction. *Lancet* 2001; 358:764.
32. Norris RM. Dissent from the consensus on the redefinition of myocardial infarction [comment]. *Eur Heart J* 2001;22:1626, 1627.
33. Morrow DA, Cannon CP, Rifai N, et al. TACTICS-TIMI 18 Investigators. Ability of minor elevations of troponins I and T to predict benefit from an early invasive strategy in patients with unstable angina and non-ST elevation myocardial infarction: results from a randomized trial. *JAMA* 2001;286:2405–2412.
34. Venge P, Lagerqvist B, Diderholm E, Lindahl B, Wallentin L. Clinical performance of three cardiac troponin assays in patients with unstable coronary artery disease (a FRISC II Substudy). *Am J Cardiol* 2002; 89:1035–1041.
35. Wu AHB, Apple FS. Reporting of cardiac troponin concentration [letter]. *Clin Chem* 2002; 48:2077–2082.
36. Panteghini M, Gerhardt W, Apple FS, Dati F, Raukilde J, Wu AH. Quality specifications for cardiac troponin assays. *Clin Chem Lab Med* 2001;39:175–179.



<http://www.springer.com/978-1-58829-526-2>

Cardiovascular Biomarkers

Pathophysiology and Disease Management

Morrow, D.A. (Ed.)

2006, 640 p. 227 illus., Hardcover

ISBN: 978-1-58829-526-2

A product of Humana Press