How to choose a point-of-care testing for troponin

Ya-hui Lin | Yang Zhang | Yu-tao Liu | Kai Cui | Jin-suo Kang | Zhou Zhou

Abstract

Background: Point-of-care (POC) cTn assays are needed when the central laboratory is unable to provide timely results to the emergency department. Many POC devices are available. The prospect of choosing them is daunting. In order to provide a quick decision-making reference for POC cTn device selection comparing them to the central laboratory, seven POC devices commonly employed by emergency department were evaluated.

Methods: Firstly, we reviewed all devices package inserts. Secondly, we evaluated several POC cTn assays for imprecision, linearity, and correlation with central laboratory assays according to CLSI EP protocols. The linear regression analyses were performed only for the detectable concentrations. Five cTnI devices (Alere Triage, BioMerieux Vidas, Mitsubishi Pathfast, ReLIA TZ-301, and Radiometer AQT90) were evaluated against a contemporary cTnI assay (Beckman Access II Accu TnI). Two cTnT assays (Radiometer AQT90 and Roche Cobas h232) were compared to a high-sensitivity (hs) cTnT method (Roche Cobas e601).

Results: For cTn levels around the 99th percentile upper reference limits (URLs) of the comparator assays, imprecision could not be assessed for the Alere, BioMerieux, and Cobas h232 as they gave undetectable readings due to a lack of assay sensitivity. Imprecision (CV) was unacceptably high for the ReLIA (33.3%). On account of this precision metric, these four assays were deemed unsuitable. Regression analyses showed acceptable linearity for all the POC devices. The correlation coefficients for ReLIA, BioMerieux, Cobas h232, and Radiometer cTnT were >0.95. Unlike the cTnT devices, the cTnI assays employ different capture and detection antibodies leading to non-commutable results. The POC cTn results were concordant with their comparator—Radiometer cTnT 90%, Pathfast cTnI 85%, and Radiometer cTnT 75%.

Conclusion: Our study provides the procedure and essential data to guide selection of a POC cTn device. Of the point-of-care devices, methods evaluated Radiometer AQT90 (cTnI and cTnT) and Pathfast might be considered.

Keywords
analytical evaluation, cardiac troponin, point-of-care testing
1 | INTRODUCTION

Measurements of cardiac troponin (cTn) are vital in the management and diagnosis of myocardial injury and are the preferred cardiac biomarker.1–2 This is especially critical in the emergency department (ED) where rapid decision-making in chest pain patients improves outcome and decreases mortality.3–5 The main focus in the ED is to discharge patients home as soon as possible or to admit them to the hospital ward. Laboratory tests are vital for making such decisions in the ED.6 In fact, laboratory tests are requested in up to half of the patients presenting at the ED.7 Clinical guidelines stipulate a maximum turnaround time of 60 minutes (preferably 30 minutes) from blood draw to results availability.8 The central laboratory is often challenged to deliver such rapid test results.9 This has given rise to the introduction of many point-of-care (POC) devices that can provide rapid troponin results.10–13 However, different POC cTn assays exhibit variable analytical sensitivity and accuracy14 and give results discordant with those of the central laboratory.15 Laboratorians and clinicians need a handy information resource to decide on which POC cTn to adopt. While a sensitive POC cTn is preferred for rapid diagnosis of acute myocardial infarction,16 expert committees of Cardiology societies concede that assays with imprecision of up to 20% may be used for diagnosis and risk stratification.17 Thus, we undertook to evaluate the analytical performance of seven POC cTn assays (5 cTnI and 2 cTnT) against our central laboratory cTnI assay and a high-sensitivity cTnT (hs-cTnT) method according to Clinical and Laboratory Standards Institute (CLSI) protocols.

2 | MATERIALS AND METHODS

2.1 | Analyzers

Five POC cTnI assays—Alere Triage Cardio3TnI (Alere International), BioMerieux Vidas Troponin I Ultra (Biomerieux), Mitsubishi Pathfast cTnI (LSI Medience Corporation), ReLIA TZ-301 cTnI (Ruihai Biological Engineering), and Radiometer AQT90 TnI (Radiometer Medical), were evaluated against the central laboratory cTnI assay (Access II Accu TnI, Beckman Coulter). Two POC cTnT assays—Radiometer AQT90 cTnT (Radiometer Medical) and Roche Cobas h232 Cardiac (Roche Diagnostics), were compared to a central laboratory hs-cTnT assay—Roche Cobas e601 (Roche Diagnostics).

2.2 | Sample preparation

Samples were collected into Greiner serum tubes (VACUETTE® 454204, Greiner Bio-One), centrifuged at 3000 g for 10 minutes, and immediately made into multiple aliquots. One aliquot was tested on the AccuTnI (Beckman Access II) and the results released to the ED, while the other aliquots were stored at −80°C until used. Before analysis, the samples were thawed at room temperature, centrifuged at 3000 g for 10 minutes, and the supernatant tested on the various cTn platforms.

| Company · platform · assay | LoD, (µg/L) | 99th percentile upper reference limit, µg/L | Time, (min) | Volume, (µL) | Sample type | Amino acid residues of epitopes recognized by Antibodies |
|-----------------------------|-------------|------------------------------------------|-------------|-------------|-------------|------------------------------------------------------|
| Beckman Access II Accu TnI  | 0.01        | 0.04                                     | 12          | 55          | Plasma/Serum | Capture antibodies: 41-49, Detection antibodies: 24-40 |
| Alere Triage cTnI           | 0.05        | 0.05                                     | 20          | 250         | Plasma/Whole blood | Capture antibodies: 27-39, Detection antibodies: 83-93, 190-196 |
| BioMerieux Vidas cTnI Ultra | 0.01        | 0.01                                     | 20          | 200         | Plasma/Whole blood | Capture antibodies: 41-49, 22-29, Detection antibodies: 87-91, 789 |
| Mitsubishi Pathfast cTnI   | 0.02        | 0.029                                    | 17          | 100         | Plasma/Serum | Capture antibodies: 41-49, Detection antibodies: 71-116, 163-209 |
| ReLIA TZ-301 cTnI           | 0.02        | 0.039                                    | 8           | 70          | Plasma/Serum/Whole blood | Capture antibodies: 41-49, 83-93, Detection antibodies: 24-40 |
| Radiometer AQT90 cTnI      | 0.01        | 0.023                                    | 18          | 1500        | Plasma/Serum/Whole blood | Capture antibodies: 41-49, 190-196, Detection antibodies: 137-149 |

| Company · platform · assay | LoD, (µg/L) | Time, (min) | Volume, (µL) | Sample type | Amino acid residues of epitopes recognized by Antibodies |
|-----------------------------|-------------|-------------|-------------|-------------|------------------------------------------------------|
| Roche Cobas e601 hs-cTnT    | 0.005       | 9           | 50          | Plasma/Serum | Capture antibodies: 125-131, Detection antibodies: 136-147 |
| Radiometer AQT90 cTnT      | 0.01        | 12          | 1500        | Plasma/Serum/Whole blood | Capture antibodies: 125-131, Detection antibodies: 136-147 |
| Roche Cobas h232 Cardiac cTnT | 0.05       | NA          | 12          | Whole blood | Capture antibodies: 125-131, Detection antibodies: 136-147 |

Abbreviations: cTnI, cardiac troponin I; cTnT, cardiac troponin T; LoD, limit of detection.
2.3 | Analytical performances

Imprecision studies were based on the CLSI EP15-A protocol (duplicate measurements twice per day on 3 levels for 5 consecutive days) using pooled human serum. The cTn concentration of the low serum pool was within the respective assay’s 99th percentile upper reference limits (URL), while the medium and high level pools were around fivefold and 10-fold of their respective URLs. Troponin values at the 99th percentile with good imprecision (CV ≤ 10%) are “guideline acceptable,” intermediate imprecision (CV 10%-20%) is “clinically usable,” and poor imprecision (CV > 20%) is not acceptable.

To demonstrate the linearity of low cTn concentrations, we employed the CLSI EP06-A protocol by proportionally mixing high and low human serum pools. Six concentrations were measured twice at each level.

### TABLE 2 Imprecision summary

| Assay platform | Mean concentration (µg/L) | %CV Within run Total | Judgment              |
|----------------|---------------------------|----------------------|-----------------------|
| cTnI           |                           |                      |                       |
| Beckman Access II |                           |                      |                       |
| Low            | 0.033                     | 4.48                 | 8.59                  | Guideline Acceptable |
| Medium         | 0.382                     | 2.69                 | 5.26                  |                       |
| High           | 1.587                     | 5.03                 | 9.59                  |                       |
| AccuTnI        |                           |                      |                       |
| Low            | <0.05                     | NA                   | NA                    | Clinically Acceptable |
| Medium         | 0.074                     | 16.55                | 16.65                 |                       |
| High           | 0.472                     | 7.55                 | 11.08                 |                       |
| Alere Triage cTnI |                           |                      |                       |
| Low            | <0.01                     | NA                   | NA                    | Guideline Acceptable  |
| Medium         | 0.430                     | 2.77                 | 4.15                  |                       |
| High           | 1.017                     | 1.17                 | 2.18                  |                       |
| BioMerieux Vidas cTnI Ultra |           |                      |                       |
| Low            | <0.01                     | NA                   | NA                    | Guideline Acceptable  |
| Medium         | 0.361                     | 14.77                | 15.02                 |                       |
| High           | 1.400                     | 6.58                 | 8.89                  |                       |
| Mitsubishi Pathfast cTnI |           |                      |                       |
| Low            | 0.006                     | 12.55                | 13.37                 | Clinically Acceptable |
| Medium         | 0.104                     | 7.76                 | 8.58                  |                       |
| High           | 0.361                     | 14.77                | 15.02                 |                       |
| ReLIA TZ-301 cTnI |                           |                      |                       |
| Low            | 0.044                     | 33.24                | 32.32                 | Unacceptable          |
| Medium         | 0.407                     | 17.08                | 15.74                 |                       |
| High           | 1.400                     | 6.58                 | 8.89                  |                       |
| Radiometer AQT90 cTnI |                           |                      |                       |
| Low            | 0.011                     | 7.69                 | 7.96                  | Guideline Acceptable  |
| Medium         | 0.144                     | 3.71                 | 3.35                  |                       |
| High           | 0.304                     | 3.83                 | 4.40                  |                       |

| cTnT           |                           |                      |                       |
| Roche Cobas e601 hs-cTnT |           |                      |                       |
| Low            | 0.013                     | 3.44                 | 5.90                  | Guideline Acceptable  |
| Medium         | 0.148                     | 1.39                 | 2.73                  |                       |
| High           | 0.593                     | 1.21                 | 1.54                  |                       |
| Radiometer AQT90 cTnT |                           |                      |                       |
| Low            | 0.014                     | 6.76                 | 9.53                  | Guideline Acceptable  |
| Medium         | 0.104                     | 3.93                 | 4.99                  |                       |
| High           | 0.584                     | 2.78                 | 2.99                  |                       |
| Roche Cobas h232 Cardiac cTnT |     |                      |                       |
| Low            | <0.05                     | NA                   | NA                    | Unacceptable          |
| Medium         | 0.05-0.10                 | NA                   | NA                    |                       |
| High           | 0.415                     | 5.750                | 10.587                |                       |

Abbreviations: CV, coefficient of variation; cTnI, cardiac troponin I; cTnT, cardiac troponin T; NA: not applicable.
regression analysis was used for correlation studies. Values below the limit of detection were replaced with one half the limit of detection. A P value < .05 was considered statistically significant.

2.5 | Others

This study was conducted according to the Declaration of Helsinki and approved by the Institutional Review Board of Fuwai Hospital (IRB approval number 2016-809).

3 | RESULTS

3.1 | Review package inserts

Firstly, the analytical characteristics of the seven POC cTn assays and two central laboratory assays according to the manufacturer’s claims were summarized in Table 1.

3.2 | Evaluate the possible candidates

The imprecision of all devices assessed is shown in Table 2. For the low-level serum pool, the central laboratory instruments showed a CV of 5.90% for the Roche hs-cTnT and 8.59% for the Beckman Accu TnI, respectively. At this cTn concentration, the imprecision of the Radiometer AQT90 cTnI and AQT90 cTnT (7.96% and 9.53%, respectively) was quite acceptable while that for the Mitsubishi Pathfast cTnI (13.37%) was clinically acceptable. For the low-level serum pool, the Alere cTnI, BioMerieux cTnI, and Roche Cobas h232 cTnT gave undetectable readings, while the imprecision of the ReLIA cTnI was guideline unacceptable (>20%). Imprecision for the medium and high serum pools was less than 10% for most of the methods except the Alere cTnI and ReLIA cTnI (CV between 15% and 20%).

Regression analyses showed that all devices have acceptable linearity. The average recovery at lower cTn concentrations (close to the 99th percentile URL) was uneven among different devices ranging from 71.2% to 128.7%. The recovery at higher cTn concentrations was satisfactory ranging from 80.9% to 102.3% (See Table 3). The recovery of Beckman Access II · Accu TnI and Roche · Cobas e601 · hs-cTnT was 98.8% and 99.2% at lower cTn concentrations, and 97.6% and 99.4% at higher cTn concentrations, respectively.

As per CLSI document EP09-A3, the correlation between devices and the comparator assays using Passing-Bablok regression is summarized in Table 4. The correlation coefficients for ReLIA T301 and BioMerieux Vidas versus Beckman Accu TnI were surprisingly good (>0.95) while that between Radiometer AQT90 cTnI and Beckman Accu cTnI were slightly lower at 0.86.

The Mitsubishi Pathfast, BioMerieux Vidas, Alere Triage, and Radiometer AQT90 cTnT values were consistently lower than the Beckman Accu TnI (see Figure 1). For cTnT, the correlation coefficients for Roche Cobas h232 and Radiometer AQT90 results versus Roche Cobas e601 were 0.969 and 0.982, respectively.

3.3 | Analyze candidate POC cTn assays

For the three suitable candidate POC cTn assays, we performed a concordance analysis according to their cTn result categories—<LoD, LoD to 99th percentile URL (99P), and >99P (Table 5). Concordance between Roche Cobas hs-cTnT and Radiometer cTnT was 90% (36/40). For the Beckman Accu TnI, the concordance was 85% (34/40) and 75% (30/40) with Mitsubishi Pathfast and Radiometer AQT90, respectively.
4 | DISCUSSION

The primary goal of cTn assays is to aid in the early assessment of acute myocardial infarction especially in hospital emergency departments. Therefore, the analytical imprecision at the decision limit (at the 99th percentile URL) of different cTn assays is vital for clinical decision-making. For this imprecision metric, the optimal imprecision (CV ≤ 10%) is considered “guideline acceptable.” Intermediate imprecision (10%–20%) is “clinically usable,” but imprecision >20% is not acceptable. Information on POC cTn assay imprecision is frequently obtained from manufacturer’s package inserts or published literature under ideal, non-routine laboratory conditions. In the current study, the total imprecision of POC cTn assays is compared to central laboratory assays by simultaneously measuring common pooled human sera containing cTn concentrations covering the clinical decision range—99th percentile URL, fivefold URL, and 10-fold URL. Although POCT devices are optimized for whole blood samples, we used serum in our investigations since no stable whole blood materials are available. Besides, manufacturers employ serum-based liquid controls and report precision data in their package inserts based on serum or plasma. It is noteworthy that two of the seven POC cTn assays (Radiometer AQT90 cTnI and cTnT) achieved precision that is guideline acceptable, while one other assay (Mitsubishi Pathfast cTnI) met the clinically usable designation. The Cobas h232 cTnT assay is semi-quantitative; cTnT values < 0.050 µg/L are reported as negative, while those between 0.050 and 0.100 µg/L are reported as positive; actual cTnT results are only reported for values above 0.100 µg/L. Roche has since improved the Cobas h232 to render it fully quantitative with a measuring range of 0.040-2.000 µg/L.

### TABLE 4 Regression analyses between POC cTn devices against Beckman Access II or Roche Cobas e601

|            | Passing-Bablok Regression | Correlation | % of undetectable values |
|------------|----------------------------|-------------|--------------------------|
|            | Linear model | Bias at 99th URL, µg/L | R       | P value |
| cTnI       |              |                          |         |         |
| ReLiA TZ-301<sup>a</sup> | y = −0.021 + 0.931x | 40%   | 0.969 | <.0001  | 0 |
| Mitsubishi Pathfast<sup>a</sup> | y = −0.001 + 4.033x | 290%  | 0.934 | <.0001 | 17.5 |
| Alere Triage<sup>a</sup> | y = 0.001 + 1.734x | 220%  | 0.875 | <.0001 | 25 |
| BioMerieux Vidas<sup>a</sup> | y = −0.009 + 2.033x | 28%   | 0.959 | <.0001 | 25 |
| Radiometer AQT90<sup>a</sup> | y = −0.040 + 9.805x | 465%  | 0.864 | <.0001 | 30 |
| cTnT       |              |                          |         |         |
| Roche Cobas h232<sup>b</sup> | y = −0.021 + 0.456x | 14%   | 0.969 | <.0001 | 35 |
| Radiometer AQT90<sup>b</sup> | y = 0.011 + 0.940x | 193%  | 0.982 | <.0001 | 25 |

Abbreviations: cTnT, cardiac troponin T; hs-cTnT, cTnI, cardiac troponin I.
<sup>a</sup>Linear regression analysis against Beckman Access II cTnI.
<sup>b</sup>Linear regression analysis against Roche Cobas e601.

### TABLE 5 Concordance between result categories of POC devices against Beckman Access II or Roche Cobas e601

| Radiometer cTn (µg/L) | Mitsubishi Pathfast (µg/L) |
|----------------------|---------------------------|
| Beckman · Access II · AccuTnI |           |
| <LoD (4)             | 0.001                     |
| LoD–99P (1)          | 0.005                     |
| >LoD (35)            | 0.026                     |
| Sub-total (40)       | 0.026                     |
| Roche Cobas e601 hs-cTnT |                      |
| <LoD (2)             | 0.002                     |
| LoD–99P (6)          | 0.006                     |
| >LoD (32)            | 0.010                     |
| Sub-total (40)       | 0.007                     |

Abbreviations: 99P, 99th percentile URL; cTnI, cardiac troponin I; cTnT, cardiac troponin T; LoD, limit of detection.
FIGURE 1  Passing-Bablok regression analysis of POC cTn assays. A, ReLIA TZ-301, (B) BioMerieux Vidas, (C) Alere Triage, (D) Mitsubishi Pathfast, (E)Radiometer AQT90 cTnI, (F) Radiometer AQT90 cTnT, and (G) Roche Cobas h232. Legend: Solid line is Passing and Bablok regression; dot line is identity; dash line is 95% CI bands
At the time, our evaluation was performed, the improved Cobas h232 was not available to us as it had not yet obtained regulatory approval for clinical use.

Our results confirm the discrepant cTn concentrations obtained between different POC devices and the central laboratory analyzers. Even on an aggregate level by categories (below LoD, LoD to 99P, and above 99P), the discordance is not insignificant—10% with the Radiometer cTnT, but 15%-25% with the suitable cTn devices (Mitsubishi Pathfast and Radiometer). This discrepancy may be caused by a variety of reasons including the use of various capture and detection antibodies/labels, reacting conditions, detection principles, and calculation methods.24 In addition, the available and unique cTn standard reference material, SRM 2921, is a trimeric I-T-C complex which is difficult to simulate accurate cTn levels in vivo. Besides, SRM 2921 appears to have poor stability due to factors such as time and temperature.25 Besides, ALP-dependent and chemiluminescent immunoassays have been reported to show better total imprecision at the URL than cTn assays employing fluorophores or gold particles.26 The difference in antibody specificities has been a key factor hindering the harmonization of POCT measurements, especially for the cTnI assays. It is noteworthy that the epitopes recognized by the capture and detection antibodies employed by the Beckman Access II as well as the ReLIA TZ-301 are located at the stable mid-region of cTnI.27 However, the epitopes recognized by the detection antibodies of the Mitsubishi Pathfast, Alere Triage, and Radiometer AQT90 are close to the less stable C-terminus of cTnI at amino acids 163-209, 190-196, and 137-149, respectively.28 Our results show that the measured values of these POC assays were significantly lower than Beckman Access II when the same samples were tested (see Figure 1). With regard to cTnT, the results of Radiometer AQT90 and Roche Cobas e601 are similar and with good correlation since identical capture and detection antibodies are employed. However, the correlation coefficients were only marginally dissimilar for Cobas h232 and AQT90-TnT versus Cobas e601. Thus even when antibodies employed recognize the same antigenic epitopes, cTnT concentrations from POCT devices are unlikely to equate to those of the central laboratory.29

Harmonization of POCT cTn testing is sorely needed because cTn may be tested on both POC and central laboratory immunochemistry analyzers in the same patient with chest pain presenting to the emergency department. It is important that results from the POC cTn performed at the emergency department are comparable to those from the central laboratory. When choosing POC cTn assays, the analytical characteristics of the various POC devices especially their precision at around the 99th percentile URL should be carefully examined first. Instruments with high imprecision should be excluded. The package insert data (Table 1) reveal that the Cobas h232 has no statement on the 99th percentile URL and is unlikely to meet acceptability criteria. Moreover, identical limits of detection and the 99th percentile URLs are stated for the Alere cTnI and the Biomerieux cTnI; thus, a high imprecision at the URL can be expected. This view is confirmed by the poor CV at the low serum pool (Table 2) for Cobas h232, Alere cTnI, Biomerieux cTnI, and ReLia cTnI rendering them “not acceptable.” Our study provides laboratories and emergency department personnel some objective data for choosing a suitable POC cTn assay as well as not to pursue unlikely candidates.

POC cTn has decreased time in the emergency department (ED) for patients with chest pain,5 and the degree of benefit also depends on the setting in which it is used especially acceptance by ED personnel. In the pre-hospital phase, POC cTn has helped in the triage of chest pain patients to the appropriate hospital facility.30 Beyond hospitals, POC cTn has reduced costs from unnecessary referrals in primary care settings but at the expense of some missed cases of small or early myocardial infarction due to the limitation of assay sensitivity.31 However, higher sensitivity POC cTn platforms are looming on the horizon and promises to change the ED diagnostic landscape.32-34

Our study provided the step-by-step guide to choose POC for cTn and a head-head comparison of 7 POC troponin devices. It is aimed at providing a quick decision-making reference for POC cTn device selection comparing them to the central laboratory cTnI and hs-cTnT. This is the first POC cTn study conducted in the real-world conditions prevailing in the Asia-Pacific. Besides, there is paucity of peer-reviewed literature verifying manufacturers’ analytical claims. The study has some limitations in that we did not conduct reference interval studies or clinical performance studies of the different POC cTn platforms. This information is already available in part in the published literature.34,29,35-38 Further studies on larger population samples are needed.39 In another hand, POC cTn assays were compared against a contemporary assay, whereas cTnT assays were compared against a high-sensitivity assay. As we known, the imprecision or sensitivity of high-sensitivity troponin is better than contemporary troponin. It means that the standards of POC for troponin T were higher troponin I.

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CONFLICT OF INTEREST
We have no conflicts of interest to declare.

AUTHOR CONTRIBUTIONS
All the authors have accepted responsibility for the entire content of this submitted the study and approved submission.

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