

## Letter to the Editor

William A. Parsonage, Jillian R. Tate\*, Jaimi H. Greenslade, Christopher J. Hammett, Jacobus P.J. Ungerer, Carel J. Pretorius, Anthony F.T. Brown and Louise Cullen

# Effect of recalibration of the hs-TnT assay on diagnostic performance

**Keywords:** acute myocardial infarction; high sensitivity troponin T; traceability.

**\*Corresponding author: Jillian R. Tate**, Chemical Pathology, Pathology Queensland, Block 7, Level 3, Royal Brisbane and Women's Hospital, Herston Road, Herston, QLD 4029, Australia, Phone: +61 7 36460082, Fax: +61 7 36463417, E-mail: jill\_tate@health.qld.gov.au

**William A. Parsonage:** Department of Cardiology, Royal Brisbane and Women's Hospital, Brisbane, QLD, Australia; and School of Medicine, University of Queensland, Brisbane, QLD, Australia

**Jaimi H. Greenslade and Christopher J. Hammett:** Department of Emergency Medicine, Royal Brisbane and Women's Hospital, Brisbane, QLD, Australia; and School of Public Health, Queensland University of Technology, Brisbane, QLD, Australia

**Jacobus P.J. Ungerer and Carel J. Pretorius:** Pathology Queensland, Brisbane, QLD, Australia

**Anthony F.T. Brown:** School of Medicine, University of Queensland, Brisbane, QLD, Australia; and Department of Emergency Medicine, Royal Brisbane and Women's Hospital, Brisbane, QLD, Australia

**Louise Cullen:** School of Medicine, University of Queensland, Brisbane, QLD, Australia; Department of Emergency Medicine, Royal Brisbane and Women's Hospital, Brisbane, QLD, Australia; and School of Public Health, Queensland University of Technology, Brisbane, QLD, Australia

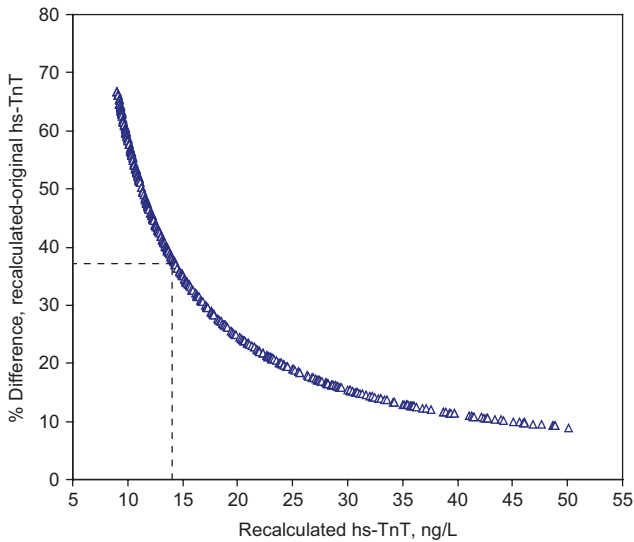
To the Editor,

Apple and Jaffe [1] recently reported on an adjustment to the high-sensitivity cardiac troponin T (hs-TnT) assay and questioned the consequent clinical implications. We report further on the effect of restandardisation of the assay on diagnostic performance. In our study between November 2008 and February 2011 (Trial registration ACTRN12610000053022) the diagnostic accuracy of the hs-TnT assay for acute myocardial infarction (AMI) was determined in a well-characterised but unselected cohort of patients presenting to the emergency department with chest pain suggestive of acute coronary syndrome (ACS) in whom blood was drawn for assay at presentation and after 2 h [2]. Serum was stored at  $-80^{\circ}\text{C}$  for later cTnT

analysis in 2012 using the Roche cobas e601 hs-TnT assay (Roche Diagnostics, Sydney, Australia; reagent lot no. 00163704; calibrator lot no. 00165094). Clinical endpoint adjudication was performed by one of two cardiologists using all clinical information available up to at least 30 days following presentation including results of the hospital's usual troponin assay performed at presentation and after 6–8 h (Beckman Coulter AccuTnI, decision cut-off 40 ng/L cTnI). The outcome for the analysis of diagnostic accuracy was AMI at presentation and was diagnosed according to the Universal Definition of Myocardial Infarction published at the time of adjudication [3]. Briefly, this was defined as evidence of myocardial necrosis together with clinical evidence of myocardial ischaemia (ischaemic symptoms, ECG changes, or imaging evidence). Necrosis was diagnosed on the basis of a rising or falling pattern of the laboratory cTnI concentrations, with at least one value above the 99th percentile.

Following completion of the hs-TnT analysis in 2012 a technical bulletin (No: 12–023) was published by Roche Diagnostics (Penzberg, Germany) calling for adjustment to the calibration of the hs-TnT assay when performed with certain reagent lots [1]. All study samples were found to require adjustment. This was performed according to the manufacturer's method in which a new value assignment of the calibrator set used in this study was required. The result was that following recalibration the number of values that were below the limit of detection (5 ng/L) decreased from 1178 (71.0%) to 560 (33.8%). Eighty-six of 1659 values were in the cTnT concentration range 8.71–13.94 ng/L, and when recalculated were on average 5 ng/L above the decision cut-off of 14 ng/L in the range 14.04–18.93 ng/L. The difference between recalculated and original values was +38% at the decision limit of 14 ng/L compared with +15.3% at 30 ng/L cTnT (Figure 1).

Seven hundred and thirty-seven patients were included in the clinical analysis and an adjudicated diagnosis of AMI at presentation was made in 50 (6.8%)



**Figure 1** Percent difference between recalculated and original hs-TnT values after a mathematical recalibration of measured cTnT values.

Dotted line indicates +38% bias at the decision limit of 14 ng/L cTnT.

individuals. Troponin T was elevated at either baseline or after 2 h in 168 (22.8%). The effect of recalibration on the diagnostic performance of the hs-TnT assay was to increase diagnostic sensitivity from 94.0% to 96.0% and negative-predictive value from 99.5% to 99.6%, and to decrease diagnostic specificity from 88.2% to 82.5% and positive-predictive value from 36.7% to 28.6%.

In order for laboratory troponin measurements to be reliable for patient management, measurement performance (i.e., bias, imprecision, total error) should fall within clinically relevant limits. The definition of acceptable performance for cTnI and cTnT measurements is still under discussion, but the biological variability data can help in objectively defining the allowable goals. A total CV <10% and an assay bias within  $\pm 15\%$  may represent a good compromise for minimum analytical requirements and is consistent with a minimum total error goal of  $\sim 33\%$  at the 99th percentile reference limit [4]. The observed bias in this study of  $-38\%$  at 14 ng/L cTnT for the hs-TnT assay exceeded this goal and a reassignment of hs-TnT

calibrators was required to regain traceability to the manufacturer's master calibrator.

In this study the assay recalibration had only a modest effect on diagnostic accuracy parameters with one patient reassigned a diagnosis of AMI. However, this may not be the case for other clinical studies [5]. Roche Diagnostics has implemented more stringent criteria in the quality control process, including the use of narrower intervals and additional control samples both at and below the clinical decision limit to detect drifts in calibration traceability [6]. Laboratories measuring troponin are well advised to monitor assay bias and any drifts in calibration traceability across different reagent lots by using not only the manufacturer's quality controls but also a patient pool with a concentration close to the decision limit for troponin [7].

### Conflict of interest statement

**Authors' conflict of interest disclosure:** The authors stated that there are no conflicts of interest regarding the publication of this article. Research funding played no role in the study design; in the collection, analysis, and interpretation of data; in the writing of the report; or in the decision to submit the report for publication. JRT, JPJU have no conflicts of interest.

**Research funding:** WAP: Queensland Emergency Medicine Research Foundation, Abbott, Roche, Alere, Beckman Coulter; LC: Queensland Emergency Medical Research Foundation, Abbott, Roche, Alere, Siemens, Radiometer Pacific; JHG: Queensland Emergency Medical Research Foundation, Roche, Siemens; AFTB: Queensland Emergency Medical Research Foundation.

**Employment or leadership:** None declared.

**Honorarium:** WAP: Abbott, Astra Zeneca, Hospira, Sanofi Aventis; LC: Alere, Boehringer Ingelheim, Pfizer, Astra Zeneca, Abbott, Radiometer Pacific; AFTB: Astra Zeneca, Boehringer Ingelheim, Aventis, Roche; CJH: Abbott, Astra, Bayer, Boehringer, Boston Scientific, Cordis, Lilly, Medtronic, Pfizer, Sanofi, Schering-Plough.

Received July 1, 2013; accepted July 12, 2013; previously published online August 8, 2013

## References

- Apple FS, Jaffe AS. Clinical implications of a recent adjustment to the high-sensitivity cardiac troponin T assay: user beware [letter]. *Clin Chem* 2012;58:1599–600.
- Than M, Cullen L, Aldous S, Parsonage WA, Reid CM, Greenslade J, et al. 2-hour accelerated diagnostic protocol to assess patients with chest pain symptoms using contemporary troponins

- as the only biomarker, The ADAPT Trial. *J Am Coll Cardiol* 2012;59:2091–8.
3. Thygesen K, Alpert JS, White HD, Jaffe AS, Apple FS, Galvani M, et al. Universal definition of myocardial infarction. *Circulation* 2007;116:2634–53.
  4. Panteghini M. Quality specifications for troponin – an overview. In: Tate J, Johnson R, Jaffe A, Panteghini M, editors. *Laboratory and clinical issues affecting the measurement and reporting of cardiac troponins: a guide for clinical laboratories*. Alexandria: AACB; 2012:53–61.
  5. Kavsak PA, Hill SA, McQueen MJ, Devereaux PJ. To the Editor. Author reply; Apple FS, Jaffe AS. 2012;58:1599–600. *Clin Chem* 2013;59:574–6.
  6. Hallermayer K, Jarausch J, Menassanch-Volker S, Zaugg C, Ziegler A. To the Editor. Author reply; Apple FS, Jaffe AS. 2012;58:1599–600. *Clin Chem* 2013;59:572–4.
  7. Tate JR. Troponin revisited 2008: assay performance. *Clin Chem Lab Med* 2008;46:1489–500.