High-Sensitivity Troponin T Validation Study Data

Marianne Roser, Lan Nguyen, Graham RD Jones
Department of Chemical Pathology, St Vincent's Hospital, Sydney, Australia.
gjones@stvincents.com.au

Introduction

❖ When a new assay is introduced into use a validation study must be performed.

❖ As well as technical validation, laboratories should consider reporting issues, laboratory workflow, handling of interferences and clinical implications of introducing the test.

❖ We describe the validation and other processes undertaken as part of changing from Siemens Ultra TnI to Roche high-sensitivity Troponin T (TnT-hs) at our laboratory.

Aim

❖ The aim of this presentation is to describe a number of factors identified during the validation process for high-sensitivity troponin T in our laboratory.

Methods

The study included the following components:

❖ **Precision:**
CLSI-like protocol with triplicate measurement of QC over 6 days using BioRad QC material.

❖ **Method comparison**
With Siemens Centaur TnI Ultra and Roche 4th generation TnT (TnT 4th)

❖ **Interference assessment**
Haemolysis by addition of haemolysate in vitro
Lipaemia by addition of Intralipid in vitro

❖ **Clinical Implications**
Measurement of all TnI requests for TnT-hs for 1 week for all Emergency Department requests (TnT-hs results not released for clinical assessment).

❖ **Reporting issues:** Selection of units, reference intervals, testing protocol were discussed with Cardiology and Emergency departments.

Results (1)

❖ **Precision** within-run, between-run and total precision of TnT-hs showed a CV of below 10% down to at least 10 ng/L (see table 1).

Average (ng/L)	9.4	41.4	412
CVwr	2.3%	1.5%	0.9%
CVbr	3.2%	4.4%	3.9%
CVtot	4.0%	4.7%	4.0%

Table 1. Precision characteristics of TnT-hs.

❖ **Comparison** with TnT-4th
The study confirmed the previously described positive bias of TnT-hs at low concentrations. A 4th generation decision point of 0.035 ug/L is approximately equivalent to 55 ng/L with TnT-hs (see figure 1).

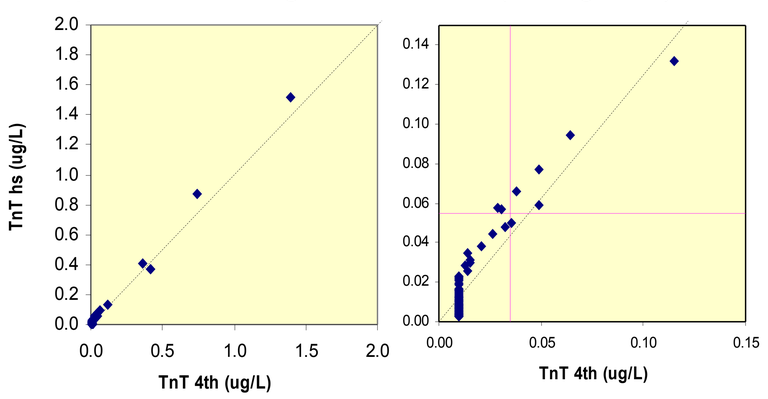

Figure 1. Comparison of TnT-hs with TnT-4th

Results (2)

❖ **Method comparison** with the Siemens Centaur Troponin assay (TnI Ultra).
A wide scatter of results was seen. In general the TnT-hs results were between 30 and 1000 x the TnI values with their respective units. By ROC analysis a TnT-hs value of 20 ng/L was equivalent to 0.055 ug/L TnI (SydPath upper limit of normal) with 85% sensitivity and specificity.

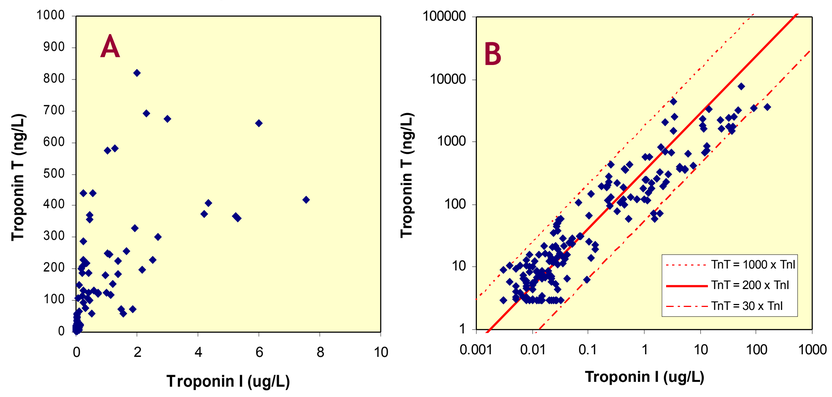

Figure 2. Comparison of TnT-hs with TnI Ultra. A – linear scales; B – log scales.

❖ **Interferences.**
Haemolysis caused a reduction in measured TnT–hs dependent on the amount of haemoglobin. Local data was similar to published data. A correction for all samples was developed to allow reporting in nearly all samples.

TnT-hs(cor.) = TnT-hs(meas.) x {1 + 0.0005 x H (mg/dL)}

Samples with H>600 not released.

Addition of Intralipid showed no significant effect to an L index over 1000 mg/dL (data not shown).

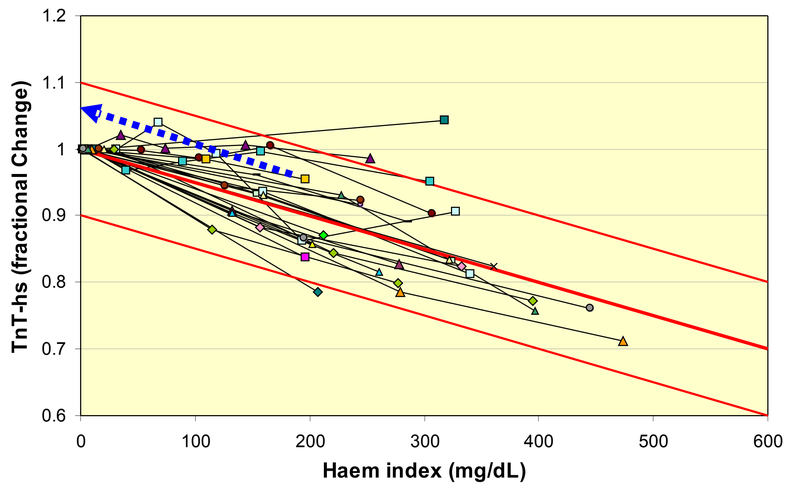

Figure 3. Effect of added haemolysate on TnT-hs. The fractional measured ThT-hs after addition of haemolysate is shown. The blue dashed line shows an example of correction for haemolysis. The red lines show a +/- 10% range. N=26, TnT-hs between 4 and 1205 ng/L.

After consultation within clinical users the following reporting criteria were determined:

❖ **Units**: ng/L (in place of ug/L)

❖ **Reference interval:** 0 – 14 ng/L (99th centile of normal)

❖ **Time to rule out ACS:** > 6 hours from onset of chest pain

❖ **Minimum time for delta troponin**: 3 hours.

❖ **Significant delta change**: +/- 30%

❖ **A footnote** is inserted calculating the delta value for all pairs of samples between 1 and 10 hours apart with at least one sample > 14 ng/L and a delta >+/- 30%

Results (3)

❖ **Clinical comparison** showed an increase in the number of "troponin positive" patients in the ED by more than double. Review of the notes indicated that 16 of the 20 "new positives" were admitted to the hospital despite unawareness of the TnT-hs result.

❖ The separation of stable from "delta positive" patients was clear in most cases. Clinical acumen is required with all results.

		Troponin I	
		<0.06	>=0.06
TnT	<14	72	0
	>=14	20	13

Table 2. Evaluation of ED patients according to TnT and TnI status.

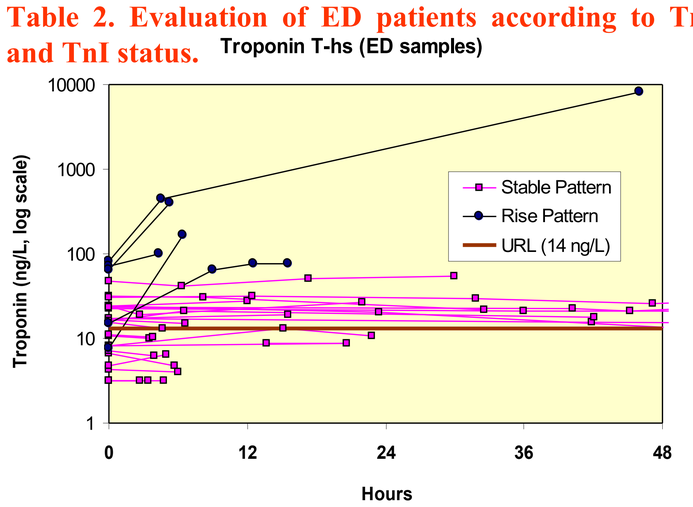

Figure 4. Changes in serum troponin in ED patients divided into increasing (blue circles) and no-change (pink squares).

Discussion

❖ The TnT-hs assay showed excellent within-run precision down to within the reference interval. No "flyers" were seen which might indicate a need for routine testing in duplicate.

❖ The previously described positive bias of TnT-hs compared to TnT-4th Gen was confirmed. This is disappointing as it breaks the previous "universal standardisation" of troponin T. Good comparison was seen above about 0.1 ug/L. Direct conversion between TnT-hs and TnI ultra was not possible.

❖ With-holding results with haemolysis > 100 mg/dL would have affected about 5% of samples. Our protocol allows release of over 99% of results. A footnote warns of increased uncertainty where this may be relevant for calculation of a delta troponin.

❖ While the number of "troponin positive" patients in the ED increased substantially, the majority of these patients were admitted to the hospital on the basis of other factors.

❖ In general there was a clear distinction between patients with a rise/fall pattern and those with stable troponin.

Conclusion

❖ The high Sensitivity troponin T assay has analytical and clinical performance characteristics markedly different to both TnT-4th and TnI Ultra.

❖ These factors need to be taken into account when introducing this assay and should be carefully discussed with relevant physicians.