Evaluation of Antibodies to Troponin I
Executive Summary

The focus here was appraisal of five ‘Troponin I’ antibodies in ELISA format using other commercially available antibodies as controls. Two forms of Troponin I, recombinant and native human Troponin complex, were used as target analytes in the assays. The ELISA formats that were used were of a standard set-up and no directed optimisation was applied.
Main Findings

1. All 5 monoclonal antibodies recognise recombinant human troponin.
2. When using single clone antibodies for antigen capture, antibody DCAB-TJ201 was the most effective for capture of recombinant and native human troponin I, when using either HRP labelled DMAB4456MH or DMAB4452MH for antigen detection.
4. The most effective capture antibody mixtures contained DCAB-TJ201 as one of the components.
5. The 5 monoclonal antibodies show significantly lower recognition of native human troponin I in the form of a troponin complex relative to their recognition of the recombinant antigen.
6. Antibody DCAB-TJ203 may not be suitable for the detection of native human troponin I when the antigen is in the form of a troponin complex.
7. Antibody DMAB4456MH was found to be a more sensitive detecting antibody for human troponin I compared with antibody DMAB4452MH.
8. Between lots variability was indicated for antibody DMAB4452MH.
Results

The initial look at the pairing of the antibodies using the configurations specified showed that all the antibodies, with the exception of DCAB-TJ203, performed well when DMAB4456MH-HRP was used for detection (Figure 1). The best capture conditions were found when DCAB-TJ201 was used on its own or in combinations. There were no significant increases seen by using the combinations of antibodies as the capture in the assays used. The trends seen when DMAB4452MH-HRP (Figure 2) was used for detection was similar but the over signals generated were lower.
Correlation between troponin I concentration estimates for 26 human plasma samples determined using a clinical analyser and two in-house ELISA formats.

**Correlation between troponin I concentration estimates**

- **cTnI** estimate from DCAB-TJ201 capture / DMAB4456MH detection assay (ng/ml)
- **cTnI** estimate from 19C7 capture / 16A11 detection assay (ng/ml)

**Graphs:**

- Linear relationship between troponin I estimates:
  - **y = 0.1678x + 2.1607, R² = 0.867**
  - **y = 0.1509x + 4.3133, R² = 0.2895**
  - **y = 0.8371x + 2.7252, R² = 0.867**

**Legend:**
- [cTnI] (ng/ml) 19C7-16A11
- [cTnI] (ng/ml) DCAB-TJ201-DMAB-4456MH

**Analysis:**

- The correlation coefficient R² indicates the strength of the linear relationship. A value closer to 1 indicates a stronger correlation.
- The graphs show positive correlations between the troponin I estimates from different methods.
Conclusions

1. All 5 monoclonal antibodies recognise recombinant human troponin.
2. The 5 monoclonal antibodies show significantly lower recognition of native human troponin I in the form of a troponin complex relative to their recognition of the recombinant antigen.
3. Antibody DCAB-TJ203 may not be suitable for the detection of native human troponin I when the antigen is in the form of a troponin complex.
4. Antibody DMAB4456MH was found to be a more sensitive detecting antibody for human troponin I compared with antibody DMAB4452MH.
5. Between lots variability was indicated for antibody DMAB4452MH.
6. The recommended antibody pair for quantification of human troponin I is antibody DCAB-TJ201 for antigen capture and HRP labelled antibody DMAB4456MH for antigen detection.
7. It is recommended that native human troponin I is used as the standard when using assays to quantify troponin I in human plasma samples.