Letter to the Editor

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Clinical implications of a recent adjustment to the high-sensitivity cardiac troponin T assay: some results

Keywords: cardiac disease; cardiac troponin T (cTnT); healthy subjects; high-sensitivity troponin assay; normal range.

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To the Editor,

Recently, some concerns have been reported about a technical bulletin by Roche Diagnostics GmbH (Mannheim, Germany) regarding the adjustment to the calibration curve of the Elecsys Troponin T high-sensitivity (hs) assay [1]. According to the bulletin, the goal of the manufacturer was to recalibrate the control materials of the Troponin T hs assays to return to the original assay specifications. The revised lots of calibrators (lots 167345 and 167650) should yield measurable cardiac troponin T (cTnT) concentrations in a greater proportion of patient samples for which the concentrations were undetectable with previous lots, including lot 163704 [1]. This adjustment regarding calibration materials may have major implication for patient care, including the recalculation of the 99th percentile value for cTnT assay [1], which is the cornerstone of the definition of myocardial infarction [2].

The aim of the present report is to compare the results obtained by measuring several plasma samples of healthy subjects and patients with acute or chronic cardiac diseases to give the users of Elecsys Troponin T hs assay some information on the difference in cTnT values measured using the previous and the new recalibrated lots.

From March to June 2012, cTnT was measured with the ECLIA method using lot 163704 on 520 heparinized plasma samples (collected from 367 healthy subjects and 153 patients with acute or chronic cardiac diseases) and stored in several aliquots at −80°C. From October to December 2012, we reassayed another aliquot stored at −80°C of the same plasma samples with lot 167345. The cTnT was assayed by the ECLIA Troponin T hs method on the Cobas e411 platform in accordance with the instructions provided by the manufacturer [3].

Patients with acute or chronic cardiac diseases were admitted to the clinical wards and the intensive coronary unit of the Fondazione Toscana G. Monasterio, Pisa, Italy. All healthy people were recruited from the laboratory staff, blood donors, or voluntary subjects included in screening programs for preventive medicine (also including adolescent subjects). The presence of cardiac or systemic acute or chronic diseases was ruled out in all subjects by checking the medical history, accurate clinical examination, electrocardiography, cardiac imaging (if needed), and laboratory tests.

We reported the linear regression analyses among the cTnT values measured in 520 samples of healthy subjects and patients with lot 163704 (x-axis) and those with lot 167345 (y-axis) in Figure 1A. Moreover, we reported the regression analysis including only the 367 healthy subjects in Figure 1B. All samples with measured values below 3 ng/L were reported in the figure and considered for the statistical analysis as containing 3 ng/L of cTnT. Although these data suggest a strict linear relationship between the values measured with the two lots, the linear regression equation reported in Figure 1A does not allow a correct estimation of the true difference between cTnT concentrations, obtained with the two lots, throughout all the working range (from 3 to approx. 25,000 ng/L), especially when considering the cTnT values in the normal range.

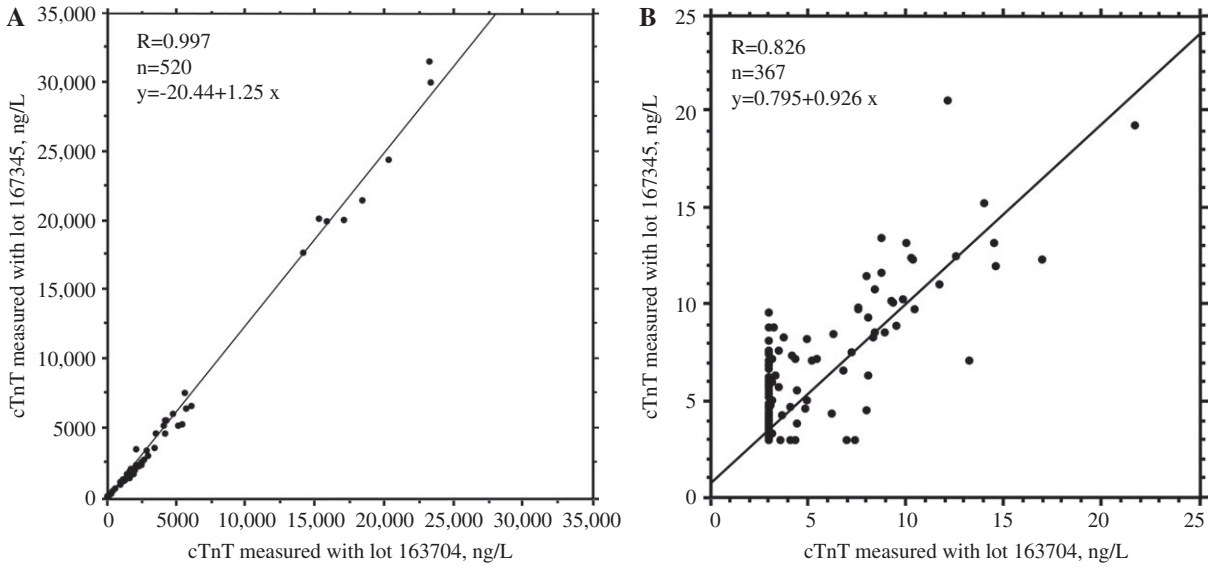

Figure 1 Linear regression analysis between cTnT values measured with lot 163704 (x-axis) and those with lot 167345 (y-axis). (A) Regression analysis including all plasma samples from healthy subjects and cardiac patients. (B) Regression analysis including only healthy subjects. All samples with measured values <3 ng/L were reported and considered for the statistical analysis as containing 3 ng/L of cTnT (considered as the blank of the method) [2].

On the contrary, the linear regression calculated using log-transformed cTnT values allowed a better definition and estimation of differences between lots throughout all the working range (Figure 2). For example, the linear regression of Figure 2 gives a value of 15.7 ng/L (y-axis) measured with lot 167345 at the suggested decision level of 14 ng/L [3] measured with lot 163704 (x-axis), whereas the corresponding cTnT value (y-axis) calculated from the equation reported in Figure 1B, which only takes into account values ranging from 3 to 22 ng/L, is 13.8 ng/L.

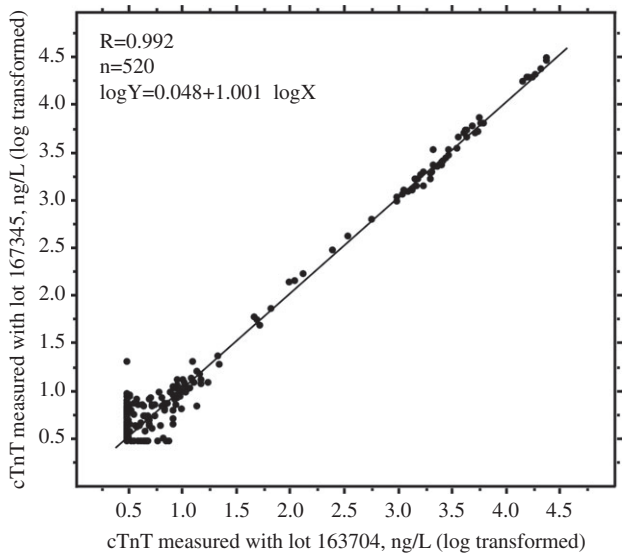

Figure 2 Linear regression analysis between the log-transformed cTnT values measured with lot 163704 (x-axis) and those with lot 167345 (y-axis).
All samples with measured values <3 ng/L were reported and considered for the statistical analysis as containing 3 ng/L of cTnT (considered as the blank of the method) [2].

The measured 99th percentile values of cTnT concentrations of 367 healthy people (also including adolescent subjects) were 14.3 ng/L for lot 163704 and 14.9 ng/L for lot 167345, respectively. Considering only the 173 apparently healthy adults (age range, 43–76 years; mean±SD, 59.4±8.0 years; median, 60.0 years; 25th–75th percentile, 52.0–65.0 years), the measured 99th percentile value was 16.4 ng/L for lot 163704 and 18.4 ng/L for lot 167345. Moreover, in the 194 adolescent subjects (age range, 10–14 years), all the cTnT values measured with lot 163704 were <3 ng/L, whereas 26 samples showed cTnT values >3 ng/L (corresponding to 13% of total) when measured with lot 167345. In the 173 apparently healthy adult subjects, 114 samples showed cTnT values <3 ng/L (i.e., 66% of total) with lot 163704, whereas only 59 with lot 167345 (i.e., 34% of total; $p<0.0001$ by χ^2-test).

The assay reproducibility for the two considered reagent lots was similar as shown by internal quality control data on the low-level control serum (PC TN1 REF 05095107 190 PreciControl Troponin by Roche Diagnostics; lot 163704: mean cTnT value, 26.9 ng/L; CV, 8.1%; n=35; lot 167345: mean cTnT value, 28.5 ng/L; CV, 7.2 %; n=120). These data also confirm that the recalibrated lot 167345 gives values higher than the previous one of approximately

1.5 ng/L at cTnT concentrations around the decision level. Given that lot 163704 is no longer available, we were not able to perform a complete imprecision profile for the two considered reagent lots; thus, we could not demonstrate if the recalibration also had an effect on the LoD of method.

Our data indicate that the difference between the measured values with the two lots [percent difference (mean±SD), 17.0%±43.9%, calculated as (lot 167345-lot 163704)/lot 163704×100], although statistically significant, is probably not clinically relevant for the diagnosis of acute myocardial infarction (AMI). From a clinical point of view, it is important to note that to diagnosis AMI, two or more samples are needed to confirm the presence of the increase/decrease trend of cTnT values [2]. As a result, the typical pattern of cTnT values in patients with AMI should not be substantially affected.

If one is interested in the recalculation of some plasma samples previously measured with lot 163704, the linear regression reported in Figure 2 can be used for this purpose. Considering that the linear relationship is very strict (R=0.992) and the number of samples used for the statistical analysis is very high (i.e., n=520), the absolute difference between the values measured with the two lots can be accurately estimated using the equation reported in Figure 2. However, our data indicate that at very low cTnT concentrations, the recalibration made by the manufacturer may have a more relevant clinical effect. Indeed, using the recalibrated lot 167345, cTnT can be measured with the ECLIA method above the limit of blank (i.e., 3 ng/L) [3] in approximately 66% of apparently healthy adults subjects compared with only 34% with lot 163704. According to the scorecard designation proposed by Apple [4], these data indicate that the ECLIA method, which uses the recalibrated lot 167345, should be considered to be a first-generation high-sensitivity method (level 2) for the measurement of cTnT.

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.
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