Study report from a multicenter evaluation of the new cobas b 101 system for the measurement of HbA1c and lipid panel

Introduction
The new cobas b 101 system provides a point-of-care system for the measurement of HbA1c and lipid panel (total cholesterol (TC), triglycerides (TG), high density lipoprotein cholesterol (HDL), and calculated values for low density lipoprotein cholesterol (LDL), non-HDL, and a TC/HDL ratio). The main objective of the performance evaluation was to confirm the analytical performance of the system in the hands of healthcare professionals in a point-of-care environment.

Method overview
The evaluation was carried out at two ISO-certified clinical laboratory sites: Barcelona, Spain and Zurich, Switzerland. The HbA1c evaluation was performed from March to August 2012, and the lipid panel from June to December, 2012. In this evaluation, the cobas b 101 system was evaluated using both the HbA1c disc and the lipid panel disc for lot-to-lot reproducibility, precision according to CLSI EP5-A2 and method comparison. The cobas c 501 as a module of cobas® 6000 analyzer series served as a reference system for the method comparison. Measurements were performed on the cobas b 101 system by healthcare professionals and on the cobas c 501 module by professional laboratory technicians. Sample: The measurements were performed with various blood samples: for HbA1c, capillary whole blood, venous EDTA whole blood and Li-heparin whole blood were used; for the lipid panel, capillary whole blood, venous EDTA whole blood and EDTA plasma were used. Samples were collected prospectively, and informed consent was obtained from all patients enrolled in the study. In total, 135 patients (71 male, 64 female) were recruited with a HbA1c range from 4.1 % to 13.6 % HbA1c. 160 patients (84 male, 76 female) were recruited with lipid panel ranges: for TC from 1.9 – 12.63 mmol/L, for TG from 0.52 – 6.7 mmol/L and for HDL from 0.47 – 2.44 mmol/L. Quantitative assessment. All the data were analyzed using the WinCAEv tool. Statistical analysis of lot-to-lot reproducibility and method comparison data were evaluated using Passing-Bablok regression analysis. Precision experiments were analyzed by calculating the standard deviation (SD) and coefficient of variation (CV) between measurements. Qualitative assessment. Practicability and usability were qualitatively assessed by each site during the evaluation period.

Key conclusions
All measurements evaluated with both the HbA1c disc and the lipid panel disc on the cobas b 101 system met the pre-defined acceptance criteria for lot-to-lot reproducibility, precision and method comparison to the reference system (see Performance Evaluation Results below). The acceptance criteria for HbA1c were defined according to the National Glycohemoglobin Standardization Program (NGSP) guidelines as written prior to September 2012, as per the dates when the evaluation was carried out. Additional acceptance criteria were defined according to the Clinical and Laboratory Standards Institute (CLSI) guidelines and the National Cholesterol Education Program (NCEP) guidelines. All sites confirmed the usability and practicability of the cobas b 101 system and rated the system as convenient for use in a point-of-care environment.
Evaluation results
cobas b 101 and HbA1c disc
Lot-to-lot reproducibility
Guidelines from Roche internal development were used to define the acceptance criteria for lot-to-lot reproducibility.
For the concentration range of 4% to 9% HbA1c, a maximum 95% confidence interval (CI) of 0.5% HbA1c was considered acceptable.

<table>
<thead>
<tr>
<th>Sample type</th>
<th>N</th>
<th>Mean deviation</th>
<th>Lower 95% CI (%)</th>
<th>Upper 95% CI (%)</th>
<th>Acceptance criterion (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Capillary WB</td>
<td>60</td>
<td>0.01</td>
<td>-0.27</td>
<td>0.30</td>
<td>0.50</td>
</tr>
<tr>
<td>EDTA WB</td>
<td>70</td>
<td>0.00</td>
<td>-0.36</td>
<td>0.36</td>
<td>0.50</td>
</tr>
<tr>
<td>Li-heparin WB</td>
<td>41</td>
<td>-0.02</td>
<td>-0.30</td>
<td>0.28</td>
<td>0.50</td>
</tr>
</tbody>
</table>

Table 1: Lot-to-lot comparison of HbA1c disc. Measurements were made on the cobas b 101 system at the site in Barcelona. For analysis of all sample types, results obtained with Lot 1 were used as the reference values and results from Lot 2 as the comparator values. "WB" is whole blood.

Table 2 demonstrates that the precision measurements for all control samples are within the SD and CV ranges defined in the acceptance criteria.

Precision
Intermediate precision and repeatability of the cobas b 101 system were measured according to CLSI EP5-A2 guidelines using four patient sample pools (A, B, C and D) with HbA1c in four concentrations: below medical decision level (5%), at medical decision level (5.5%), above medical decision level (6%), and significantly above medical decision level (12%). In addition, cobas b 101 system controls covering two concentration ranges were used to assess the precision. Control level 1 contains the concentration range 4.3% to 6.5% HbA1c, and control level 2 contains the concentration range 7.4% to 11.2% HbA1c. Acceptance criteria were specified according to the CLSI guidelines for two ranges of HbA1c: for HbA1c ≤5.5%, the SD should be used to assess precision, and an SD of ≤0.22% is specified as acceptable; for HbA1c >5.5%, the CV should be used to assess precision, and a CV of ≤4% is specified as acceptable.

Table 3: Method comparison with HbA1c disc. Measurements made on the cobas b 101 system using the HbA1c disc were compared with measurements made on the cobas c 501 module (reference system) at the site in Barcelona. For all cobas c 501 measurements, the sample type used was EDTA whole blood. "WB" is whole blood.
Figure 2 a, b, c: Linear regression analysis of method comparison with HbA1c disc. Passing-Bablok regression analysis was performed for comparison of the cobas b 101 system against the cobas c 501 module as reference with each sample type: capillary whole blood (a), EDTA whole blood (b) and Li-heparin whole blood (c). Only EDTA whole blood was used in the cobas c 501 module for all comparisons.

The acceptance criteria based on pre-September 2012 NGSP guidelines specify a 95 % CI of less than 0.75 % HbA1c in the concentration range 4 % to 10 % HbA1c. Table 3 and Figures 2a, b and c demonstrate that the method comparison 95 % CI values for the cobas b 101 system compared to the reference cobas c 501 module fully satisfy these criteria.

**Table 4: Lot-to-lot comparison of lipid panel disc.** Measurements were made on the cobas b 101 system at the site in Zurich. For analysis of all sample types, results obtained with Lot 1 were used as the reference values and results from Lot 2 as the comparator values. Results from Passing-Bablok regression analysis are reported (slope, intercept, Pearson’s r).

In Figure 3a, b and c, regression graphs are shown for the capillary blood samples only. Refer to Table 4 for results from EDTA whole blood and EDTA plasma (graphs available upon request). All mean biases for the lot-to-lot measurements fall within the specified criteria for each parameter: TC, ≤3 %; TG, ≤5 %; HDL, ≤5 %. For this reason, the two lots can be considered equivalent and only data from one lot will be shown for the precision and method comparison results below.
Precision
Intermediate precision and repeatability of the cobas b 101 system were evaluated according to CLSI EP5-A2 guidelines using two control samples covering both normal and pathological concentration ranges for all three lipid parameters. Acceptance criteria were specified according to the CLSI guidelines for two ranges of each parameter. For TC with a concentration of \( \leq 3.1 \text{ mmol/L} \), the criteria is an SD \( \leq 3\% \). For TG with a concentration of \( \leq 1.37 \text{ mmol/L} \), the criteria is CV \( \leq 5\% \). For HDL with a concentration of \( \leq 1.37 \text{ mmol/L} \), the criteria is CV \( \leq 3\% \). For HDL with a concentration of \( > 1.09 \text{ mmol/L} \), the criteria is an SD \( \leq 5\% \).

Table 5 demonstrates that the intermediate precision and repeatability measurements for both of the control sample levels fall within the SD or CV ranges defined, satisfying the acceptance criteria.

Method comparison
The performance of the cobas b 101 system with the lipid panel test was compared to the cobas c 501 module for TC, TG, and HDL measurement. As a control, measurements were performed with NIST (National Institute of Standards and Technology) standards (two different concentration levels for TC and TG, no material for HDL was available) before and after the method comparison experiments. Both prospective and residual patient samples were used in the experiments. The acceptance criteria were defined according to the NCEP and CDC (Cholesterol Disease Control) guidelines and are summarized in Table 6.

Table 6: Summary of acceptance criteria for method comparison of lipid panel disc.

Table 7: Method comparison with lipid panel disc.

(a) --- Identity (x = y); Regression; \( y = 1.029x - 0.175; r = 0.9906; N = 68 \)
(b) --- Identity (x = y); Regression; \( y = 1.014x + 0.018; r = 0.9937; N = 61 \)
(c) --- Identity (x = y); Regression; \( y = 0.913x + 0.115; r = 0.9748; N = 67 \)