Performance evaluation of the prototype Abbott Alinity hq hematology analyzer

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Abstract

Introduction: The analytical and clinical performance as well as the workflow efficiency of the novel, prototype Alinity hq hematology analyzer was evaluated in the clinical laboratory of Universitair Ziekenhuis Brussel, Department of Hematology, Brussels, Belgium.

Methods: Within-run and within-laboratory imprecision, linearity, and carryover were assessed using clinical blood samples and commercial blood products. Four hundred and seventeen samples were selected for method comparison with Abbott CELL-DYN Sapphire, and for flagging performance analysis in comparison with smear review and manual microscopic white blood cell (WBC) differential.

Results: Within-run and within-laboratory imprecision verification demonstrated low %CV for complete blood count and WBC differential results within the normal ranges (0.1%-10.4%), except for basophil granulocytes. The linearity of the analytical measuring ranges was verified for WBCs, red blood cells, hemoglobin, and platelets. Alinity hq results showed strong agreement with those of CELL-DYN Sapphire. Good correlation was demonstrated with manual WBC differential results, with negative bias for neutrophil (NEU) granulocytes, and positive bias for lymphocytes and monocytes. Blasts were detected with 75% sensitivity and 96% specificity at 1% blast threshold, and 100% sensitivity at 5% blast threshold. Immature granulocyte detection was more sensitive (81% vs 76%, P = 0.086) and specific (88% vs 78%, P = 0.0002) than with CELL-DYN Sapphire. Nucleated red blood cell detection was more sensitive (89% vs 63%, P < 0.001) and just slightly less specific (96% vs 99%, P = 0.0067) than with CELL-DYN Sapphire. Re-run and reflex testing rates were lower with Alinity hq.

Conclusion: The Alinity hq hematology analyzer is suitable for clinical use.

Keywords
Abbott Alinity hq, automated hematology analyzer, complete blood count, performance evaluation

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Modern hematology analyzers provide complete blood cell counts (CBC), white blood cell (WBC) differential, reticulocyte (RETIC), and related measure concentrations with high sample throughput and reduced turnaround time, using electrical particle counting (impedance), optical and fluorescence flow cytometry, and most recently, digital imaging-based principles. Despite the increasing accuracy and precision of the results, manual microscopic review of blood smears remains necessary for certain blood samples, to confirm morphological abnormalities for WBC, red blood cells (RBC), and platelets (PLT), or to confirm abnormal numerical results. It is still considered the reference method for evaluating RBC, and PLT morphology and for WBC differentiation. In addition to reporting numerical results, analyzers provide various morphological flags to indicate the presence of certain immature or pathological cell types and other morphological findings. Accuracy of these flags may have significant impact on slide review rate.

The aim of this study was to evaluate the performance characteristics of a prototype Alinity hq hematology analyzer (Abbott Laboratories, Diagnostics Division, Hematology, Santa Clara, CA, USA) in a routine laboratory setting. A complete performance evaluation was conducted in accordance with professional recommendations including both analytical (within-run and within-laboratory imprecision, linearity, and carryover) and clinical performance characteristics. Results were compared to those obtained with CELL-DYN Sapphire. In addition, Alinity hq WBC differential was evaluated in comparison with the manual microscopic differential as the reference method, and flagging performance was analyzed in comparison with manual smear review. Workflow efficiency was assessed by comparing re-run and reflex testing rates on the two analyzers.

2 | MATERIALS AND METHODS

2.1 | Alinity hq

Alinity hq is a novel, high throughput hematology analyzer from Abbott Laboratories. Alinity hq uses optical principles for all measurements. Hemoglobin (HGB) is determined using spectrophotometry. Nucleated cells are measured using a proprietary, fluorescent nuclear dye-containing reagent that stains nuclei for subsequent multi-angle light scatter and fluorescence analysis. Nucleated cells are reported as a 6-part WBC differential (including immature granulocytes [IG]), and nucleated RBC (NRBC) count. IGs include promyelocytes, myelocytes, and metamyelocytes. RBC and PLT analysis is performed using multi-angle light scatter principles. The technology, referred to as Multi-Angle Polarized Scatter Separation (MAPSS™), has been used on previous Abbott hematology analyzers; however, three additional low angle scatter detectors were added on Alinity hq. The seven unique light detectors (axial light loss, polarized side scatter, depolarized side scatter, and four intermediate angles of light scatter), in combination with fluorescence (FL1), create a unique signature of each cell, allowing for the differentiation of WBC subpopulations, as well as reliable separation of PLT and RBC.

Tubes can be entered either in closed or in open mode. The analyzer has two incubation blocks to enable high throughput, which are calibrated separately. Calibration of the analyzer is performed by using commercial calibrators, but for the purpose of this comparison study, Alinity hq was cross-calibrated according to professional guidelines by running 10 samples (without flags) on CELL-DYN Sapphire, the reference analyzer, followed by testing them on Alinity hq and applying the appropriate calibration factors. CD29 Plus Tri-Level Controls (Streck Inc, La Vista, NE) were used for quality control purposes.

The study was performed with a prelaunch prototype instrument, equipped with software version 0.1.0.24203 and algorithm version 5.0.1 (Research Use Only).

2.2 | CELL-DYN Sapphire

CELL-DYN Sapphire (Abbott) uses MAPSS™ technology for WBC differential and PLT measurement, and impedance for RBC. It reports a standard 5-part WBC differential. Other differences between CELL-DYN Sapphire and Alinity hq include the lack of the RBC fragments flag on CELL-DYN Sapphire, and the presence of an IG flag, instead of reporting the concentration and %IG. In addition, CELL-DYN Sapphire features a band alert that warns the user about the potential presence of band NEUS/IGs. It also features a CD61 monoclonal antibody-based ImmunoPLT method, which has been reported to be equivalent with the CD41/CD61 reference method.

2.3 | Blood samples

All 486 samples were leftover, de-identified, K2-EDTA-anticoagulated (Sarstedt S-Monovette®, Nümbrecht, Germany) clinical samples used in a 2-month period of 2017 from the routine patient population of Universitair Ziekenhuis Brussel, Department of Hematology, Brussels, Belgium. Samples were selected to represent certain analytical ranges for various measurements for the analytical studies (n = 35 for within-run imprecision, n = 30 for carryover and n = 11 for linearity). In addition, 417 clinical samples were chosen for comparison purposes (between instruments and between Alinity hq and manual differential results). These samples represent 163 normal samples, 163 pathological samples as recommended by professional guidelines, including chronic and acute leukemia, myeloma, thalassemia, lymphoma, sickle cell disease, and cold agglutinins. Neonatal and cord blood samples were considered as pathological samples due to the large amount of NRBC and IG. Samples were processed on both analyzers within 4-8 hours of venesection. In addition, commercial CD29 Plus Tri-Level Controls and the CBC-LINE CL011 linearity kit (R&D Systems, Minneapolis, MN, USA) were analyzed. The study protocol was approved by the ethics committee of the hospital (BUN 143201630594).
2.4 | Imprecision

2.4.1 | Within-run

Thirty-five clinical samples were used to determine within-run imprecision for all primary CBC parameters and WBC differential, ensuring that each measurand is represented in normal, low, and high concentration ranges in at least two samples. Samples were tested in 10 replicates in the same run. Mean, standard deviation (SD), and coefficient of variation (%CV) were calculated for each measurand. %CV obtained on samples were compared to Ricos’ desirable specifications15 and to imprecision results published in the operator’s manual of CELL-DYN Sapphire.

2.4.2 | Within-laboratory

CD29 Plus Tri-Level Controls were analyzed at each level (low, normal, and high) in 10 replicates per day (five replicates on the left and five on the right block) for 5 days, per CLSI EP05-A3 guideline.16 Within-laboratory imprecision was calculated for WBC, RBC, HGB, hematocrit (HCT), MCV, red cell distribution width (RDW), PLT, neutrophil (NEU), eosinophil (EO) and basophil (BASO) granulocytes, lymphocytes (LYM), monocytes (MONO), IG, and NRBC and were compared to Ricos’ desirable specifications.15

2.5 | Sample carryover

Carryover was determined for WBC, RBC, HGB, and PLT, on each incubation block separately, per ICSH guidelines,14 by running a whole blood sample with elevated parameters in triplicate, immediately followed by one with low parameters in triplicate. The sequence was performed three times, with three different high/low sample pairs for each measurand. Some samples were concentrated to obtain the desirable concentration.

Alinity hq features a data flag (alert), which warns the user for the presence of potential carryover, and invalidates the affected analyte (WBC or PLT). This flag is triggered if a sample with less than 0.5 × 10⁹/L WBC is preceded by one with more than 40 × 10⁹/L WBC, or when a sample with less than 50 × 10⁹/L PLT is preceded by one with more than 1000 × 10⁹/L PLT. If the flag is triggered, the run needs to be repeated, which eliminates the opportunity for carryover impacting the result.

2.6 | Linearity

Linearity of the analytical measuring ranges (AMR) for WBC, RBC, HGB, and PLT was tested using the commercially available CBC-LINE CL011 kit containing separate full range and low range kits. Testing was performed according to the relevant package inserts. Additionally, 11 clinical samples were manually diluted in ratios of 3:4, 1:2, 1:4, 1:8, 1:16 with Alinity hq Diluent. Each dilution was tested in 4 replicates.

2.7 | Interinstrument comparison

Alinity hq absolute and percent NEU and IG results were added up, and the sum of the results was compared to Sapphire NEU concentration and %NEU, as CELL-DYN Sapphire does not report IG concentration separately, instead, includes that in the NEU count. In addition to the whole AMR, regression and correlation analysis was performed separately for NEU, HGB, and PLT data on a subset of clinical samples in the low range, to assess performance at medical decision points. Measurands with invalidating data flags either on CD-Sapphire or Alinity hq or both analyzers were excluded from the calculations on a measurand-by-measurand basis; therefore, the final number of samples included in the comparison is different for each parameter and is shown in the Results section (Table 2). In two samples with hypereosinophilic syndrome (with over 70% EOs), EOs were misclassified as NEUs by Alinity hq. Manual smear review revealed hypogranular EO granulocytes. These two samples were excluded from the WBC differential comparison calculations.

2.8 | Comparison of PLT count with ImmunoPLT results

Platelets counts on 98 samples (included in the 417 samples used for method comparison) were additionally compared with the results of the CD61 ImmunoPLT method on CELL-DYN Sapphire.12,13 Samples were selected to cover a wide range, however, enriched in samples with low PLT count (below the reference range and around medical decision levels). Statistical analysis was performed for all 98 samples and separately for the samples with PLT results of <100 × 10⁹/L.

2.9 | Comparison of Alinity hq WBC differential with manual microscopic differential

A smear was prepared from each sample that was run on the analyzers. Eventually, 398 samples were included in this evaluation, due to sample exclusions related to inappropriate test selection on Alinity hq (n = 4), WBC concentration of less than 0.20 × 10⁹/L (n = 12), or inadequate smear quality (n = 3). Measurands with invalidating data flags by Alinity hq were excluded from the calculations on a measurand-by-measurand basis; therefore, the final number of samples included in the comparison is different for each parameter and is shown in the Results section (Table 3). Two experienced morphologists performed a 200-cell microscopic WBC differential on each sample, and when results disagreed, a third morphologist was used to resolve discrepancies, as advised by the ICSH guidelines.14 The mean of the final counts was taken as gold standard. Results for %NEU, %LYM, %MONO, %EO, %BASO, %IG, and the number of NRBC per 100 WBC (NR/W) produced by Alinity hq were compared to the manual differential result. Manual %NEU count included both segmented and band NEUs, just as Alinity hq %NEU does. Alinity hq %IG was compared to the combined concentrations of promyelocytes, myelocytes, and metamyelocytes obtained with the manual differential.
TABLE 1  Within-laboratory imprecision at low, normal, and high concentration levels compared to Ricos’ desirable specifications:15

| Measurand       | Low control |         | Normal control |         | High control |                  | Desirable spec. 15 |
|-----------------|-------------|---------|----------------|---------|--------------|-------------------|--------------------|
|                 | Mean | SD    | %CV | Mean | SD    | %CV | Mean | SD    | %CV | %CV |
| WBC (10^9/L)    | 3.36 | 0.07  | 2.2 | 7.35 | 0.12  | 1.7 | 17.00 | 0.22  | 1.3 | 5.7 |
| RBC (10^{12}/L) | 2.80 | 0.01  | 0.5 | 4.17 | 0.03  | 0.8 | 5.23  | 0.11  | 2.2 | 1.6 |
| HGB (g/L)       | 80.10 | 0.05  | 0.6 | 123.00 | 0.08  | 0.6 | 171.00 | 0.12  | 0.7 | 1.4 |
| HCT (L/L)       | 27.20 | 0.21  | 0.8 | 41.20 | 0.53  | 1.3 | 56.20  | 1.41  | 2.5 | 1.4 |
| MCV (FL)        | 96.80 | 0.64  | 0.7 | 98.40 | 1.17  | 1.2 | 107.00 | 1.06  | 1.0 | 0.7 |
| RDW (%)         | 13.90 | 0.05  | 0.4 | 14.00 | 0.12  | 0.8 | 13.30 | 0.38  | 2.8 | 1.8 |
| PLT (10^9/L)    | 64.10 | 1.23  | 1.9 | 205.00 | 2.91  | 1.4 | 480.00 | 11.60 | 2.4 | 4.6 |
| NEU (10^9/L)    | 1.54  | 0.05  | 3.3 | 3.53  | 0.07  | 2.1 | 8.95  | 0.12  | 1.4 | 8.6 |
| LYM (10^9/L)    | 1.01  | 0.04  | 4.4 | 1.97  | 0.07  | 3.7 | 3.56  | 0.13  | 3.8 | 5.1 |
| MONO (10^9/L)   | 0.37  | 0.04  | 10.1 | 0.86 | 0.07  | 8.2 | 1.98  | 0.11  | 5.7 | 8.9 |
| EO (10^9/L)     | 0.08  | 0.01  | 14.7 | 0.17 | 0.02  | 10.4 | 0.49  | 0.03  | 5.4 | 10.5 |
| BASO (10^9/L)   | 0.04  | 0.03  | 74.2 | 0.07 | 0.05  | 66.5 | 0.11  | 0.08  | 76.8 | 14.0 |
| IG (10^9/L)     | 0.32  | 0.02  | 5.3 | 0.75  | 0.02  | 3.2 | 1.84  | 0.04  | 2.4 | N/A |
| NRBC (10^9/L)   | N/A   | N/A   | N/A | N/A   | N/A   | N/A | 2.38  | 0.08  | 3.3 | N/A |

BASO, basophil; EO, eosinophil; HCT, haematocrit; HGB, hemoglobin; IG, immature granulocyte; LYM, lymphocyte; MCV, mean corpuscular volume; MONO, monocyte; N/A, Not Available; NEU, neutrophil; NRBC, nucleated red blood; PLT, platelet; RBC, red blood cell; RDW, red cell distribution width; WBC, white blood cell.

TABLE 2  Interinstrument comparison between CELL-DYN Sapphire and Alinity hq

| Measurand       | n   | Range tested on Alinity hq | Bland-Altman bias, (95% CI) | Passing-Bablok r (95% CI) | Intercept | Slope |
|-----------------|-----|---------------------------|----------------------------|---------------------------|-----------|-------|
|                 |     | Min. | Max. |                |                          |        |       |
| WBC (10^9/L)    | 373 | 0.16 | 177.00 | 0.02 (-0.06, 0.10) | 1.00 (1.00-1.00) | -0.04 | 1.00  |
| NEU < 1.5 (10^9/L) | 29  | 0.14 | 1.48 | -0.05 (-0.15, 0.05) | 0.81 (0.63-0.91) | -0.02 | 1.02  |
| NEU (10^9/L)    | 357 | 0.14 | 57.90 | 0.03 (0.00, 0.06) | 1.00 (1.00-1.00) | -0.02 | 1.01  |
| LYM (10^9/L)    | 367 | 0.07 | 20.00 | -0.04 (-0.10, 0.01) | 0.97 (0.96-0.98) | -0.04 | 1.00  |
| MONO (10^9/L)   | 351 | 0.00 | 29.50 | 0.12 (-0.01, 0.25) | 0.82 (0.78-0.85) | 0.00  | 1.02  |
| EO (10^9/L)     | 356 | 0.00 | 1.92 | -0.01 (-0.01, -0.01) | 0.98 (0.98-0.98) | -0.01 | 1.02  |
| BASO (10^9/L)   | 336 | 0.00 | 0.18 | -0.01 (-0.02, -0.01) | 0.48 (0.39-0.56) | 0.00  | 0.57  |
| RBC (10^{12}/L) | 409 | 1.51 | 6.65 | -0.11 (-0.12, -0.10) | 0.99 (0.99-0.99) | -0.28 | 1.05  |
| HGB < 80 (g/L)  | 26  | 5.25 | 7.96 | 0.08 (-0.01, 0.17) | 0.96 (0.92-0.98) | -0.15 | 1.03  |
| HGB (g/L)       | 407 | 5.25 | 18.80 | 0.14 (0.11, 0.17) | 0.99 (0.99-0.99) | -0.11 | 1.02  |
| HCT (L/L)       | 409 | 14.10 | 55.70 | -1.10 (-1.22, -0.97) | 0.99 (0.98-0.99) | -1.30 | 1.01  |
| MCV (FL)        | 409 | 57.70 | 124.00 | -0.79 (-0.92, -0.66) | 0.99 (0.99-0.99) | -4.71 | 1.04  |
| MCH (pg)        | 409 | 17.30 | 46.00 | 1.21 (1.13, 1.29) | 0.98 (0.97-0.98) | 0.22  | 1.03  |
| MCHC (g/L)      | 409 | 28.10 | 41.40 | 1.59 (1.49, 1.68) | 0.78 (0.73-0.81) | 1.32  | 1.01  |
| NRBC (10^9/L)^a | 386 | 0.00 | 27.10 | 0.01 (-0.04, 0.06) | 0.95 (0.94-0.96) | 0.02  | 0.93  |
| PLT < 100 (10^9/L) | 67  | 8.64 | 99.50 | 2.52 (1.45, 3.61) | 0.99 (0.98-0.99) | 2.80  | 1.00  |
| PLT (10^9/L)    | 379 | 8.64 | 1040.00 | 4.87 (3.64, 6.10) | 1.00 (1.00-1.00) | 1.07  | 1.01  |

BASO, basophil; EO, eosinophil; HCT, haematocrit; HGB, hemoglobin; LYM, lymphocyte; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; MCV, mean corpuscular volume; MONO, monocyte; NEU, neutrophil; NRBC, nucleated RBC; PLT, platelet; RBC, red blood cell; WBC, white blood cell.

*Deming regression.
2.10 | Flagging performance

The same samples that were included in the Alinity hq comparison with manual microscopic differential were used to assess the performance of the BLAST flag and IG and NRBC detection. The consensus guidelines of the International Society for Laboratory Hematology (ISLH) were followed to determine a positive smear finding: ≥1 blast per 100 WBC, ≥1 NRBC per 100 WBC, ≥5 atypical lymphocytes (AL) per 100 WBC, and the presence of ≥1 promyelocyte or myelocyte, or ≥2 metamyelocytes.17 The performance of Alinity hq to detect blast cells was determined at 1% and 5% blast thresholds (based on manual WBC differential), using the BLAST flag alone and also the combination of BLAST flag with WBC data flags. Alinity hq does not report an IG or NRBC flag; instead, it reports the exact concentrations of these cell types. An optimal cutoff on Alinity hq was determined for IG detection (see Statistical analysis2.8).

Failure of the analyzer to provide results for one or more WBC differential cell types was regarded as a morphological flag, according to the recommendations of CLSI H20-A2.18

2.11 | Workflow efficiency

Workflow efficiency was assessed by comparing re-run and reflex testing rates for the same samples that were included in the inter-instrument comparison on both analyzers. The number of blood samples that required re-run (re-analysis of the sample in the same or different test selection), or reflex testing (manual differential or slide review based on WBC flags, PLT and RBC flags, and numerical abnormalities), was assessed for both analyzers. The laboratory’s routine procedures, based on ICSH guidelines and good laboratory practice, were applied.19,20

2.12 | Statistical analysis

Statistical analysis was performed with the Analyse-it® for Excel software, version 2.2.6 (Leeds, UK).

TABLE 3 | Comparison of Alinity hq WBC differential with manual WBC differential

| Measurand | n | Range tested on Alinity hq | Bland-Altman bias, (95% CI) | Passing-Bablok |
|-----------|---|---------------------------|-----------------------------|---------------|
|           |   | Min. | Max. | r (95% CI) | Intercept | Slope |
| %NEU      | 377 | 3.4 | 97.3 | −7.67 (−8.23, −7.10) | 0.96 (0.95-0.97) | −7.05 | 1.00 |
| %LYM      | 380 | 0.8 | 92.6 | 4.13 (3.69, 4.57) | 0.97 (0.96-0.97) | 2.81 | 1.05 |
| %MONO     | 360 | 0.1 | 45.8 | 3.15 (2.83, 3.49) | 0.83 (0.79-0.86) | 1.12 | 1.34 |
| %EO       | 376 | 0.0 | 27.7 | 0.49 (0.37, 0.60) | 0.93 (0.92-0.94) | 0.04 | 1.23 |
| %BASO     | 346 | 0.0 | 2.7 | −0.14 (−0.19, −0.09) | 0.38 (0.29-0.47) | 0.02 | 0.54 |
| %IG       | 382 | 0.0 | 26.5 | 0.19 (0.04, 0.35) | 0.83 (0.79-0.86) | 0.24 | 0.94 |
| NR/W      | 388 | 0.0 | 313.0 | −1.06 (−3.06, 0.93) | 0.99 (0.99-0.99) | 0.33 | 0.45 |

BASO, basophil; EO, eosinophil; IG, immature granulocyte; LYM, lymphocyte; MONO, monocyte; NEU, neutrophil; NR/W, nucleated red blood cells per 100 white blood cells

Deming regression.

3 | RESULTS

3.1 | Imprecision

3.1.1 | Within-run

Within-run imprecision results were within desirable specifications for measurand values within and above the normal ranges, except for the BASO count (20.2%–41.3%) due to the low concentrations. The imprecision of WBC, PLT, LYM, MONO, EO, and BASO counts with very low concentrations exceeded desirable specifications, but the SD values were small compared to the actual concentration (data not shown).

3.1.2 | Within-laboratory

Results for the normal-level control were within Ricos’ desirable specifications, except for MCV, which had slightly higher imprecision (1.2% vs 0.7%; Table 1). Imprecision for the low- and high-level controls was also within the desirable imprecision for most parameters.

Linearity was assessed with correlation analysis by plotting obtained results (mean of four replicates) against expected values. In addition, % recovery (obtained results divided by expected results) was calculated at each level.

Bland-Altman analysis, Passing-Bablok, and Deming regressions (as appropriate) were used to evaluate interinstrument agreement, as well as Alinity hq and manual WBC differential comparison. Correlation coefficients ($r$) were determined by Pearson correlation, and $r$ values of ≥0.95, 0.80–0.94, 0.60–0.79, and ≤0.59 were considered as very strong, strong, moderate, and weak correlation.

Receiver operating characteristics analysis was applied to determine the best threshold for IG positivity by Alinity hq, using manually counted promyelocytes, myelocytes, and metamyelocytes as reference. The optimal cutoff for differentiating nonpathological from pathological samples was determined as 0.94% IG (specificity 87.5%, sensitivity 80.0%). For flagging performance and workflow efficiency, a chi-square test was used to compare proportions.
The RBC (and consequently, HCT) %CV results, however, were somewhat higher than the desirable specifications in the high-level control. Imprecision for the low-level EO concentration also exceeded the desirable specification (14.7% vs 10.5%). BASO concentrations showed a high level of imprecision due to the low concentrations; however, SD values were small at all three levels (0.03-0.08 × 10^9/L).

### 3.2 Sample carryover

There was no significant carryover for any tested parameter. The highest carryover observed was 0.1% for WBC (range: 0.21-214 × 10^9/L) and 0.2% for RBC (range: 1.2-8.7 10^12/L), HGB (range: 39-232 g/L), and PLT (range: 14-1223 × 10^9/L). No difference in carryover was observed between incubation blocks.

### 3.3 Linearity

A perfect correlation (r = 1.00) was observed between obtained and expected values for WBC, RBC, HGB, and PLT with both the commercial linearity materials and the diluted clinical samples, across the full AMR and also in the low concentration range (data not shown). In addition, % recovery ranged from 95.7% (PLT, CBC-Line Low Range) to 113.3% (WBC, clinical sample Low Range).

### 3.4 Interinstrument comparison

Very strong correlation was observed between results obtained with Alinity hq and CELL-DYN Sapphire (Table 2) for most parameters. Strong correlation was obtained for MONO count (r = 0.82), moderate correlation for MCHC (r = 0.78), and weak correlation for BASO concentration (r = 0.48). Regression and correlation analysis performed for NEU, HGB, and PLT on a subset of samples in the low range, yielded strong or very strong correlation. Small negative bias was observed for MCV and HCT based on the intercept of the regression line and by Bland-Altman analysis, and small positive bias for PLT results, compared to CELL-DYN Sapphire results (Table 2).

### 3.5 Comparison of Alinity hq PLT count with ImmunoPLT results

The comparison of Alinity hq PLT results to those of the CD61-based ImmunoPLT results in the range of 9.64-927 × 10^9/L showed a correlation coefficient of 1.00. Very strong correlation was also demonstrated in the low range (PLT < 100 × 10^9/L, n = 28; r = 0.99). The intercept of the regression line indicated small positive constant bias in the low range.

#### TABLE 4 Performance of the Alinity hq to detect blasts, IG, and NRBC on samples analyzed by manual microscopy (n = 398)

| Blast detection ≥1% | TP | FP | TN | FN | Sens, % (95%CI) | Spec, % (95%CI) | Overall efficiency% |
|---------------------|----|----|----|----|----------------|-----------------|---------------------|
| BLAST flag          |    |    |    |    |                |                 |                     |
| CELL-DYN Sapphire   | 8  | 16 | 366| 8  | 50 (25-75)     | 96 (93-98)      | 94                  |
| Alinity hq          | 6  | 15 | 367| 10 | 38 (15-65)     | 96 (94-98)      | 94                  |
| BLAST flag + invalidating WBC differential flags | | | | | | | |
| CELL-DYN Sapphire   | 15 | 16 | 366| 4  | 75 (48-93)     | 96 (94-98)      | 95                  |
| Alinity hq          | 12 | 15 | 367| 10 |                |                 |                     |

| Blast detection ≥5% | TP | FP | TN | FN | Sens, % (95%CI) | Spec, % (95%CI) | Overall efficiency% |
|---------------------|----|----|----|----|----------------|-----------------|---------------------|
| BLAST flag          |    |    |    |    |                |                 |                     |
| CELL-DYN Sapphire   | 6  | 16 | 375| 1  | 86 (42-100)    | 96 (93-98)      | 96                  |
| Alinity hq          | 5  | 16 | 375| 2  | 71 (29-96)     | 96 (93-98)      | 95                  |
| BLAST flag + invalidating WBC differential flags | | | | | | | |
| CELL-DYN Sapphire   | 7  | 16 | 375| 0  | 100 (59-100)   | 96 (93-98)      | 96                  |
| Alinity hq          | 7  | 16 | 375| 0  |                |                 |                     |

| IG detection        | TP | FP | TN | FN | Sens, % (95%CI) | Spec, % (95%CI) | Overall efficiency% |
|---------------------|----|----|----|----|----------------|-----------------|---------------------|
| IG flag (Sapphire)  |    |    |    |    |                |                 |                     |
| CELL-DYN Sapphire   | 47 | 75 | 261| 15 | 76 (63-86)     | 78 (73-82)      | 77                  |
| Alinity hq          | 50 | 41 | 295| 12 | 81 (69-90)     | 88 (84-91)      | 87                  |

| NRBC detection      | TP | FP | TN | FN | Sens, % (95%CI) | Spec, % (95%CI) | Overall efficiency% |
|---------------------|----|----|----|----|----------------|-----------------|---------------------|
| Alinity NR/W and CELL-DYN Sapphire |    |    |    |    |                |                 |                     |
| CELL-DYN Sapphire   | 22 | 5  | 358| 13 | 63 (45-79)     | 99 (97-100)     | 95                  |
| Alinity hq          | 31 | 16 | 347| 4  | 89 (73-97)     | 96 (93-97)      | 95                  |

FN, false negative; FP, false positive; NR/W, NRBC/100 WBC; Sens, sensitivity; Spec, specificity; TN, true negative; TP, true positive.

*Blast concentration in some of these samples is 1%-5%.*
3.6 | Comparison of Alinity hq WBC differential with manual differential

Results showed strong or very strong correlation for all parameters, except for %BASO (Table 3). Bland-Altman analysis revealed an average negative bias of −7.67% for %NEU, and a positive bias of 4.13% and 3.15% for %LYM and %MONO. Comparison of manual and automated %IG and NR/W showed an overall low bias of 0.19% and −1.06%, respectively (Table 3).

3.7 | Flagging performance

When using 1% as threshold, the combined presence of BLAST flag and WBC differential invalidating data flags resulted in a blast detection sensitivity of 75% on Alinity hq and 94% on CELL-DYN Sapphire (P = 0.0007), with the same specificity (96%). At 5% blast level, both analyzers identified 100% of samples with blasts (Table 4).

There were two samples in the study cohort with >5% AL. None of them was flagged by CELL-DYN Sapphire, but both samples triggered the VAR LYM flag on Alinity hq. No other WBC flags were present in both samples. The sensitivity of Alinity hq for IG detection was compared to that of the CELL-DYN Sapphire IG flag at a threshold that has provided the best discrimination between pathological and nonpathological samples based on manual differential results (as described in the Materials And Methods section). Alinity hq was found to be more sensitive (81% vs 76%, P = 0.0862), although not statistically significant, and specific (88% vs 78%, P = 0.0002) than CELL-DYN Sapphire.

Using the 1 NR/W threshold based on manual differential, the sensitivity of Alinity hq for detecting NRBC was 89%, compared to 63% with CELL-DYN Sapphire (P = 0.0010), with just slightly lower specificity (96% vs 99%, P = 0.0067; Table 4).

3.8 | Workflow efficiency

A significantly lower re-run rate was observed on Alinity hq (1.0% vs 4.3%, P = 0.0037), compared to CELL-DYN Sapphire. The percentage of samples that needed manual WBC differential was somewhat lower for Alinity hq (41.0% vs 47.0%; P = 0.0876). Smear review rate was also in favor of Alinity hq with 7.8% compared to 12.5% (P = 0.0260) for CELL-DYN Sapphire.

4 | DISCUSSION

The analytical and clinical performance evaluation of the prototype Alinity hq instrument showed acceptable performance for routine use.

Within-run imprecision and within-laboratory imprecision for all parameters were low, except for BASO granulocytes. A few imprecision results exceeded Ricos’ desirable imprecision. However, these specifications are based on biological variation, determined on apparently normal subjects with measurand values within the reference range; therefore, they are not necessarily suitable for measurand values at the extremes of the measuring range. The larger within-laboratory imprecision for RBC and MCV in the high-level control was unexpected, as within-run imprecision results did not show the same tendency.

No clinically relevant sample carryover was detected. Linearity testing showed excellent correlation between obtained and expected results, including low concentration ranges.

Interinstrument comparison with CELL-DYN Sapphire demonstrated high level of correlation and agreement for most of the parameters, except for BASO counts. It is well known that interinstrument correlation between BASO results is usually low, partly due to the low concentrations, but also as a result of different detection technologies. The strong correlation between Alinity hq PLT and CELL-DYN Sapphire ImmunoPLT results demonstrates the accuracy of the multidimensional optical technology.

The observed negative bias for Alinity %NEU compared to the manual differential, and the positive bias for %MONO and %LYM is attributed to the uneven distribution of cells on the smear, that is, larger cells being accumulated toward the feather edge of the smear. No bias was seen when these results were compared to those obtained with CELL-DYN Sapphire.

Blast detection on CELL-DYN Sapphire was better at 1% blast level compared to Alinity hq. At 5% level; however, all samples containing blasts were identified on both analyzers. For example, an acute myeloid leukemia with over 20% blasts did not show morphological flags on Alinity hq, but invalid results were obtained for WBC differential; therefore, the sample was efficiently identified for smear review.

Alinity hq showed significantly higher sensitivity for NRBC detection compared to CELL-DYN Sapphire (89% vs 63%, P < 0.0010) and was also found to be slightly more sensitive (81% vs 76%, not significant) and more specific (88% vs 78%, P = 0.0002) than CELL-DYN Sapphire for detecting IG. In addition, Alinity hq accurately reports the absolute concentration of IG, as confirmed by the strong correlation and low bias between manual and Alinity hq %IG results.

The lower re-run rate on Alinity hq can be attributed to the different technologies. When RBC resistant to lysis are detected by CELL-DYN Sapphire, the run needs to be repeated in an extended lyse mode, while no repeat is necessary with Alinity hq. Moreover, samples with WBC concentrations between 0.20 and 2.00 × 10^9/L are repeated with extended differential counts with CELL-DYN Sapphire, but not with Alinity hq. On the other hand, WBC or PLT carryover alerts were the main reason for re-running samples with Alinity hq, which, however, can be configured as an automatic process.

The percentage of samples that required manual differential due to morphological and numerical abnormalities was high with both instruments, although somewhat lower with Alinity hq. This is attributed to the composition of the sample cohort, the majority of which was selected to be pathological.
Limitations of the study include the use of a prototype instrument and software. The most significant difference between the launch software and the prototype software was the lack of the left-shift flag, which indicates the presence of increased number of band NEUs. In addition, RETIC results were not evaluated due to technical issues. Further evaluation is needed for assessing accuracy of results around medical decision limits, such as samples with PLT concentration of <20 × 10^9/L.

In conclusion, good analytical and clinical results were obtained during this performance evaluation of the prototype Alinity hq hematology analyzer. The ability of Alinity hq to report IG count and %, the higher sensitivity of NRBC and IG detection along with the lower number of samples requiring re-run or reflex testing are important steps toward the reduction of manual differentials.

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CONFLICT OF INTEREST

Dr Hoffmann is former employee of Abbott Diagnostics. Dr Lakos is an employee of Abbott Diagnostics.

AUTHOR CONTRIBUTIONS

All the authors have accepted responsibility for the entire content of this submitted manuscript and approved submission.

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REFERENCES

1. Bruegel M, George TI, Feng Bo, et al. Multicenter evaluation of the cobas m 511 integrated hematology analyzer. Int J Lab Hematol. 2018;40:672-682.
2. Pierre RV. Peripheral blood film review. The demise of the eyecount leukocyte differential. Clin Lab Med. 2002;22:279-297.
3. Buttarello M, Gadotti M, Lorenz C, et al. Evaluation of four automated hematology analyzers. A comparative study of differential counts (imprecision and inaccuracy). Am J Clin Pathol. 1992;97:345-352.
4. Clinical Laboratory Standards Institute. Validation and Quality Assurance of Automated Hematology Analyzer. CLSI document H26-A2. Wayne, PA: Clinical Laboratory Standards Institute; 2010.
5. Müller R, Mellors J, Johannessen B, et al. European multi-center evaluation of the Abbott Cell-Dyn Sapphire hematology analyzer. Lab Hematol. 2006;12:15-31.
6. Bourner G, Dhaliwal J, Sumner J. Performance evaluation of the latest fully automated hematology analyzers in a large, commercial laboratory setting: a 4-way, side-by-side study. Lab Hematol. 2005;11(4):285-297.
7. de Grooth BG, Terstappen L, Puppies GJ, Greve J. Light-scattering polarization measurements as a new parameter in flow cytometry. Cytometry. 1987;8:539-544.
8. Terstappen LW, de Grooth BG, Visscher K, van Kouterik FA, Greve J. Four-parameter white blood cell differential counting based on light scattering measurements. Cytometry. 1988;9:39-43.
9. Kim YR, Ornstein L. Isovolumetric sphere of erythrocytes for more accurate and precise cell volume measurement by flow cytometry. Cytometry. 1983;3:419-427.
10. Mohandas N, Kim YR, Tycko DH, Orlik J, Wyatt J, Groner W. Accurate and independent measurement of volume and hemoglobin concentration of individual red cells by laser light scattering. Blood. 1986;68:506-513.
11. CELL-DYN Sapphire Hematology Analyzer. https://www.corelaboratory.abbott/int/en/offers/brands/cell-dyn/cell-dyn-sapphire Accessed January 08, 2019.
12. Arroyo JL, Garcia-Marcos MA, Lopez A, et al. Evaluation of a CD61 MoAb method for enumeration of platelets in thrombocytopenic patients and its impact on the transfusion decision-making process. Transfusion. 2001;41:1212-1216.
13. Kunz D, Kunz WS, Scott CS, Gressner AM. Automated CD61 immunoplatelet analysis of thrombocytopenic samples. Brit J Haematol. 2001;112:584-592.
14. Briggs C, Culp N, Davis B, d'Onofrio G, Zini G, Machin SJ. ICSH guidelines for the evaluation of blood cell analysers including those used for differential leucocyte and reticulocyte. Int J Lab Hematol. 2014;36:613-627.
15. Ricos C, Alvarez V, Cava F, Garcia-Lario JV, Hernandez A, Jimenez CV, et al. Desirable specifications for total error, imprecision, and bias, derived from intra- and interindividual biologic variation. https://www.westgard.com/biodatabasel.htm Accessed January 08, 2019.
16. Clinical Laboratory Standards Institute. Evaluation of Precision Performance of Quantitative Measurement Methods. CLSI document EP05-A3. Wayne, PA: Clinical Laboratory Standards Institute; 2014.
17. International Society for Laboratory Hematology (ISLH). Consensus Guidelines: positive Smear Findings. http://www.islh.org/web/consensus_rules.php Accessed January 08, 2019.
18. Clinical Laboratory Standards Institute. Reference Leucocyte (WBC) Differential Count (Proportional) and Evaluation of Instrumental Methods; Approved. Standard—Second Edition. CLSI document H20-A2. Wayne, PA: Clinical Laboratory Standards Institute; 2007.
19. Palmer L, Briggs C, McFadden S, et al. ICSH recommendations for the standardization of nomenclature and grading of peripheral blood cell morphological features. Int J Lab Hematol. 2015;37:287-303.
20. Zini G, d’Onofrio G, Briggs C, et al. ICSH recommendations for identification, diagnostic value, and quantification of schistocytes. Int J Lab Hematol. 2012;34:107-116.
21. Amundsen EK, Henriksen CE, Holthe MR, Urdal P. Is the blood bosophil count sufficiently precise, accurate, and specific?: Three automated hematology instruments and flow cytometry compared. Am J Clin Pathol. 2012;137:86-92.
22. Béné M, Lacombe F. Differential leucocyte analysis. In: Kottke-Marchant K, Davis B, eds. Laboratory Hematology Practice. Oxford: Wiley-Blackwell; 2012:33-47.
23. Estcourt LJ, Birchall J, Allard S, et al. Guidelines for the use of platelet transfusions. Brit J Haematol. 2017;176:365-394.

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