Letter to the Editor
Diagnostic Hematology

The Sysmex XN-2000 Hematology Autoanalyzer Provides a Highly Accurate Platelet Count than the Former Sysmex XE-2100 System Based on Comparison with the CD41/CD61 Immunoplatelet Reference Method of Flow Cytometry

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Dear Editor
The recently launched hematology autoanalyzer Sysmex XN-2000 (Sysmex, Kobe, Japan) provides a new method (PLT-F method) for platelet counting based on a fluorescent RNA staining dye named oxazine [1, 2]. The main advantage of the XN-2000's PLT-F method over the former instrument XE-2100's optical method (PLT-O method) is the ability to identify apoptotic white cells and not count them as platelets [2]. The XN-2000's PLT-F method was reported to estimate platelets in thrombocytopenic samples more accurately than does the PLT-O method [1]. However, the performance of XN-2000's PLT-F method still needs to be evaluated, including samples with broad range of platelet counts. We performed correlation and bias analyses comparing the platelet counts measured by the XN-2000 and XE-2100 autoanalyzers with those calculated by the immunoplatelet reference method (IRM) of flow cytometry.

In total, 195 peripheral blood samples, which had been submitted for complete blood counts (CBC) at the authors' institution between July and September 2013, were randomly selected for this study. All samples were analyzed using the XE-2100 (PLT-O method) and XN-2000 (PLT-F method) autoanalyzers within 4 hr of sample collection. In addition, the platelet count was acquired by measuring CD41/CD61 IRM of flow cytometry using a FACSCanto II (Becton-Dickinson, Sunnyvale, CA, USA) [3]. This method uses the red blood cell (RBC): platelet ratio, which is calculated from RBC and platelet events measured by flow cytometry (Fig. 1A). Linear regression and Bland-Altman analysis were applied to compare the platelet counts measured by the two hematology autoanalyzers and those produced by CD41/CD61 IRM of flow cytometry.

The platelet count-related parameters of the 195 samples obtained from the hematology autoanalyzers and by flow cytometry...
are summarized in Table 1. The correlation between the platelet counts measured using the XE-2100 (XE PLT, PLT-O method) and those obtained from CD41/CD61 IRM of flow cytometry using the XE-2100’s RBC counts (XE flow-based PLT) yielded a correlation coefficient of \( \gamma = 0.986 \) (Fig. 1B). In the XN-2000 system, the correlation between XN PLT (PLT-F method) and XN flow-based PLT produced a comparable correlation coefficient of \( \gamma = 0.984 \) (Fig. 1C). Subsequent bias analysis between the XE PLT and XE flow-based PLT showed a mean bias of \(-37.6 \times 10^9/L\), indicating that the platelet count measured by the XE-2100 was underestimated by a mean of \(37.6 \times 10^9/L\) in comparison with the platelet count measured by CD41/CD61 IRM of flow cytometry (Fig. 1D). The same bias analysis between the XN PLT and XN flow-based PLT demonstrated a mean bias of \(-24.2 \times 10^9/L\) (Fig. 1E). These results suggest a significant reduction in bias \((13.4 \times 10^9/L)\) in comparison with the reference method when the XN-2000 is used instead of the XE-2100.

The present study included an additional bias analysis focused on the 48 thrombocytopenic samples with platelet counts less than \(20.0 \times 10^9/L\), which is the critical decision-making level for platelet transfusion and is an indicator of increased spontaneous bleeding risk [4]. The bias analysis performed with the data from the XE-2100 and the XN-2000 analyzers exhibited a mean bias of \(1.0 \times 10^9/L\) and \(0.3 \times 10^9/L\), respectively (Fig. 1F, G). These results indicate that a more accurate estimation of platelet counts can be achieved using the XN-2000 PLT-F method, with a reduction of bias \((0.7 \times 10^9/L)\) than using the XE-2100 PLT-O method, as measured by comparison with the CD41/CD61 IRM of flow cytometry. These results support the conclusion of a previous study which reported that the XN-2000 PLT-F method could be more useful than the XN-2000 PLT-O method for accurate platelet counting in thrombocytopenic samples \(N=37,\) platelet counts \(<50.0 \times 10^9/L\) [1]. In the present study, platelet counts measured using the XN-2000 PLT-F method were com-

### Table 1. Descriptive statistics of platelet count-related parameters from 195 patients, measured using the XN-2000 and XE-2100 hematology autoanalyzers and the CD41/CD61 immunoplatelet reference method using flow cytometry

| Items | Results, median (range) |
|-------|-------------------------|
| Age of patients, years | 57.0 (10.0-84.0) |
| RBC/PLT ratio (R) obtained from flow cytometry | 23.0 (1.8-2,829.0) |
| RBC counts obtained using the XE-2100 (XE RBC), \(\times 10^12/L\) | 3.33 (1.56-6.74) |
| PLT counts obtained using the XE-2100 (XE PLT), \(\times 10^9/L\) | 136.0 (2.0-1,461.0) |
| Flow-based PLT counts calculated using the XE-2100 (XE flow-based PLT*), \(\times 10^9/L\) | 165.6 (1.0-1,551.1) |
| RBC counts obtained using the XN-2000 (XN RBC), \(\times 10^12/L\) | 3.25 (1.50-6.76) |
| PLT counts obtained using the XN-2000 (XN PLT), \(\times 10^9/L\) | 142.0 (1.0-1,458.0) |
| Flow-based PLT counts calculated using the XN-2000 (XN flow-based PLT*), \(\times 10^9/L\) | 162.3 (1.0-1,488.6) |

*XE or XN flow-based PLTs were calculated from RBC counts obtained from XE-2100 or XN-2000 hematology autoanalyzers (XE RBC or XN RBC) divided by the RBC/PLT ratio (R) obtained from flow cytometry.

Abbreviations: RBC, red blood cell; PLT, platelet.

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\[\text{Flow cytometry based PLT count (L)} = \frac{\text{RBC N by XE-2100 or XN-2000}}{\text{RBC/Platelet ratio by flow cytometry}}\]

**Fig. 1.** Results of the CD41/CD61 immunoplatelet reference method using flow cytometry (A). (Continued to the next page)
Fig. 1. (Continued) Plots of the correlation analysis between the platelet count measured by the Sysmex XE-2100 hematology autoanalyzer (Sysmex, Kobe, Japan) and the platelet count estimated by the CD41/CD61 immunoplatelet reference method (flow-based platelet counts) (B), and between the Sysmex XN-2000 hematology autoanalyzer (Sysmex) and the platelet count estimated by the CD41/CD61 immunoplatelet reference method (C). The regression equations, correlation coefficients, and coefficients of determination calculated from the results with the two analyzers are included in the figure. Bland–Altman plots comparing the platelet counts and the flow-based platelet counts measured by the two analyzers as a reference method (D-G). Plots of all 195 samples (D for the Sysmex XE-2100, E for the Sysmex XN-2000), and plots of 48 thrombocytopenic samples with platelet counts less than 20.0×10$^9$/L (F for the Sysmex XE-2100, G for the Sysmex XN-2000).

Abbreviations: RBC, red blood cell; SSC-A, side scattering-area; FSC-H, forward scattering-height; PLT, platelet; XE PLT, platelet count measured directly using the XE-2100; XN PLT, platelet count measured using the XN-2000.
pared with those measured using the XE-2100 PLT-O method in 48 samples with platelet counts less than $20.0 \times 10^{9}$/L. We identified significantly higher accuracy in platelet counts when using the XN-2000 PLT-F method than that by the XE-2100 PLT-O method, both in overall and thrombocytopenic samples. Our results suggest that the XN-2000 PLT-F method can be more useful than the XE-2100 PLT-O method in determining platelet transfusions in thrombocytopenic patients.

To summarize, we showed that the Sysmex XN-2000 PLT-F method can more accurately assess platelet counts than the Sysmex XE-2100 PLT-O method in both overall and low-platelet samples. The Sysmex XN-2000 PLT-F method can be more helpful than the XE-2100 PLT-O method in making appropriate clinical decisions regarding platelet transfusion in thrombocytopenic patients.

Authors’ Disclosures of Potential Conflicts of Interest

No potential conflicts of interest relevant to this article were reported.

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