Original Article

Platelet count; Automated Vs Manual Estimation on blood smear Prince Rashid Hospital, RMS

Asim Momani,MD, Rame Khasawneh,MD, Mansour Hyari,MD, Bayan Zgoul,MD, Hala Sokhnee,MD, Ngah Drous,MD, Mohamamd maaytah,MD

B-20, Saket, Meerut. Uttar Pradesh, India. 250003

ARTICLE INFO

ABSTRACT

Objective: The objective of this study is to evaluate the accuracy of manual platelet count estimates by comparing the results of platelet count using the automated counter with the manual method (the use the stained thin blood film) for the same sample at the same time in the Hematology Dept. at PRH Clinical Laboratory. Material and Methods: This is a cross-sectional study, which was conducted at PRH, where we randomly selected blood specimens of patients presenting to the Clinical Laboratory during August through October 2013. The specimens were processed by the automated method and the manual method simultaneously. Results: The manual method for performing platelet count estimates on blood smears gives estimates that are not significantly different from the counts by the automated method on the Sysmex KX21 automated counter at $p < 0.05$. Despite that platelet count estimates with the manual method in general are slightly higher than the automated method; it is a reliable technique and appears to provide platelet count estimates to use in quality assurance. Conclusion: The study concludes that the traditional method of estimating platelet counts from blood smears to evaluate automated results appears to provide adequate quality assurance.

1. Introduction

Platelets are small anucleate cell fragments adapted to adhere to damaged blood vessels, aggregate on one another and facilitate the generation of thrombin.

On films made from blood anticoagulated with the strong calcium chelating agent, EDTA and stained with Wright stain, platelets appear as small, bluish grey, oval to round bodies with several purple-red granules.(1)

In health, there is approximately 150-400x10^9 Platelets per liter of blood.

The counts are somewhat higher in women than in men. (2)

Since many laboratories use instruments that count platelets, red cells and leukocytes concurrently; a platelet count is a routinely reported result on complete or automated hemograms.

To get an accurate platelet count by the use of an automated hematology analyzer may be complicated by the presence of particles of similar size and/or light scatter properties (red cell fragments, microcytic red cells, apoptotic white blood cell fragments) and by giant platelets and platelet clumps .(3)

Platelet counts can be done manually using a hemocytometer, and a microscope.

Objective:

The objective of this study is to evaluate the accuracy of manual platelet count estimates by comparing the results of platelet count using the automated counter with the manual method (the use the stained thin blood film) for the same sample at the same time in the Hematology Dept. at PRH Clinical Laboratory.

Methods

This is a cross-sectional study, which was conducted at PRH, where we randomly selected blood specimens of patients attending the Clinical Laboratory during August through October 2013.

The randomly selected specimens were 220 blood specimens in EDTA anticoagulant Vacutainer tubes of patients more than 20 years old presenting to the clinical laboratory with any diagnosis during the study period.

Age and gender of the patients were taken from the lab requests without meeting the patients.

The study was dealing with blood samples from the lab.

All the obtained data were handled confidentially, instead of reporting the name of patients, each sample was given a unique numeric identifier.

The specimens were processed by trained hematology technicians.

* Corresponding Author : **Asim Momani**

Asim Momani et al. Int J Biol Med Res. 2015; 6(3): 5148-5150

5149

ü   Automated Method:

After thorough mixing of each blood specimen on an automated mixer for 10 min, a CBC including the platelet count was conducted using the automated hematology analyzer, Sysmex KX21 which was regularly maintained and calibrated as recommended by the manufacturer.

ü   Manual Method:

Thin air-dried blood smears made after thorough mixing of each specimen were stained manually with a May-Grünwald-Giemsa stain and examined under light microscopy with X100 oil-immersion lens.

The slides were scanned for platelet aggregates and/or giant platelets and, if any, the specimens were exempted.

If neither were found, platelets were estimated by counting the average number of platelets seen per 100x oil immersion field in the monolayer. In general, 10 oil immersion fields were counted and the results averaged (this accounts for uneven dispersal of platelets in the smear). Then the following formula was applied:

Estimated platelet count/μL = average count in 10 fields x 20,000.

The results were grouped as follows:

Number of patients	Platelet Count	Group
150	150-450	Normal
43	< 150	Low
27	> 450	High

The processing of the data was performed using the Statistical program (SPSS 18).

Simple regression analysis and coefficient of determination (r) for correlation analysis between the two methods was used. All tests were applied at a level of significance

(α=0.05). P-values of ≤ 0.05 were considered as statistically significant.

The mean for the automated platelet count was 275,000/μL, the manual platelet estimation gave a mean of 269,000/μL.

Results:

The manual method for performing platelet count estimates on blood smears gives estimates that are not significantly different from the counts by the automated method on the Sysmex KX21 automated counter at p < 0.05.

The platelet count estimates with the manual method in general are slightly higher than the automated count, but are accurate enough to provide platelet count estimates from peripheral blood smears to use in quality assurance.

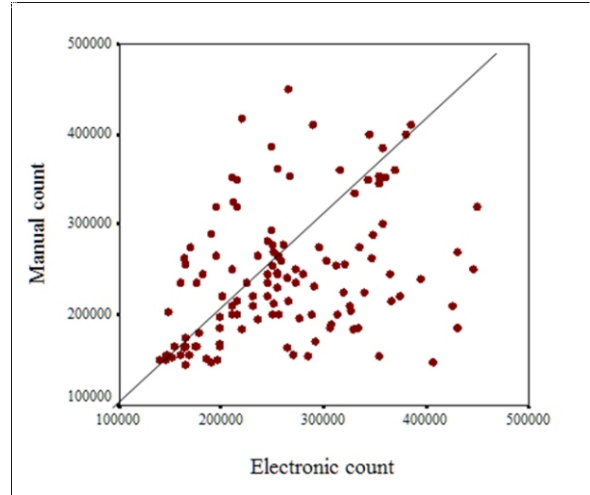**Fig 1:Samples of normal platelet count by the manual method Vs the automated method (n=43)**

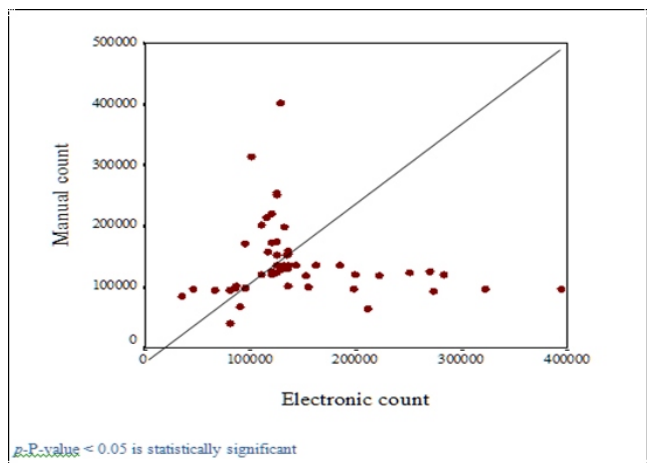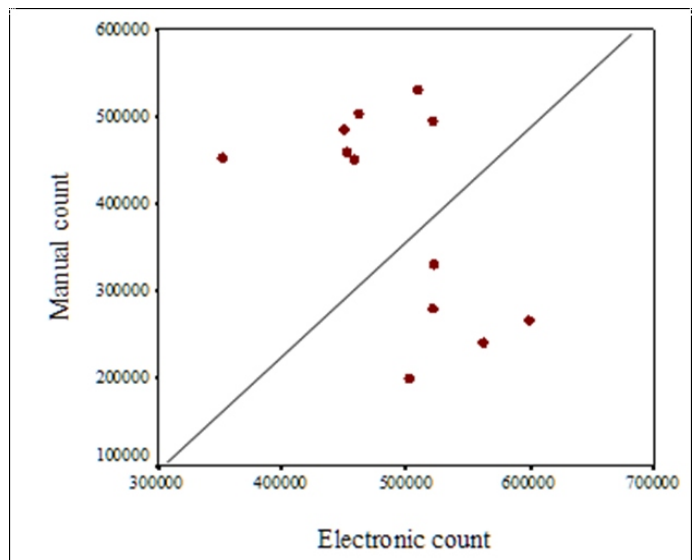**Fig 1:Samples of low platelet count by the manual method Vs the automated method (n=150)**

a.P-value < 0.05 is statistically significant

Fig 1:Samples of high platelet count by the manual method Vs the automated method (n=27)

Asim Momani et al. Int J Biol Med Res. 2015; 6(3): 5148-5150

5150

Discussion

To get an accurate platelet count by the use of an automated hematology analyzer may be complicated by the presence of particles of similar size and/or light scatter properties (microcytic red cells, white blood cell fragments) and by giant platelets and platelet clumps or aggregates.(6)

Even the most expensive and accurate hematology analyzers are not designed to eliminate peripheral blood film evaluation, and microscopic validation of platelet counts is an important component of the blood smear review.

Using the manual method of platelet count estimation on thin air dried blood smears appears to have enough accuracy to provide quality assurance,

Yet, it is worth remembering the important risk of error estimated up to 10-20% by some authors . (7)

Still, this method is fast, taking only few minutes on average per patient, while demonstrating good precision.

References:

1. Williams Hematology, McGraw-Hill Professional; 8 edition (July 9, 2010), ISBN-10: 0071621512, ISBN-13: 978-0071621519

2. Dacie and Lewis Practical Haematology, 11th Edition

3. Charie LA, Harrison P, Smith CU, Cobb JR, Briggs C, Machin S.Accuracy in the low platelet count range: a comparison of automatedplatelet counts on Beckman Coulter high-volume hematology analyzers with the ISLH/ICSH platelet reference method. Lab Hematol 2001; 7:236-44.

4. Brecher G, Cronkite EP Morphology and enumeration of human blood platelets. J Appl Physiol 1950; 3:165-77.

5. Torres SL, Velez EL. Platelet verification under microscope calculated by the patient's hemoglobin factor. Lab Med 2004; 7:430-3.

6. Rowan RM. Platelet counting and the assessment of platelet function.In: Koepke JA, editor. Practical Laboratory Hematology. New York, NY: Churchill Livingston, 1991: 157-70.

7. Jobin F. L'Hémostase. Paris, France: Edition Maloine, 1995. Brahimi et al.