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Peripheral Smear Validity Along With Automated Analyzer Regarding Platelet Count

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Abstract: Automated counts are being widely accepted in diagnostic field due to various advantages. Despite the sophistication of present day instruments there is still need to depend on manual techniques for primary calibration especially in case of platelet count. Automated analyzer had pitfalls due to mimickers of platelet like particles which validated the manual count in peripheral smear regarding platelet count by leishman's stain. A cross sectional study was conducted in a tertiary care hospital for 3 months. It includes 1200 venous samples with EDTA as the anticoagulant in vacutainers. Among the routine 100 to 120 samples/day, with platelets < 1,00,000 lakhs /cumm of 500 samples were subjected for study. Samples were processed by Symex 5000 autoanalyzer and compared with manual platelet count by leishman's stain. About 500 samples with platelets < 100,000 lakh/cu.mm were taken for study. In 384/500(76.8 %)haematology analyzed correlated with peripheral smear review. About 116(23%) were not correlating with peripheral smear findings because it revealed RBC's and large platelets in peripheral smear. Our study concludes there was significant correlation between automated and manual counts. But this was not applicable for very high or low platelet counts. As platelet count estimation is very important element of the diagnostic and treatment disorder.Peripheral smear examination along with automated analyzer valid in case of high or low value platelet counts. Keywords: Autoanalyzer, Dengue fever, Platelets, peripheral smear.

INTRODUCTION

Automation in laboratory diagnostics has revolutionized the patient care practices in the past few years. Automated counts are being widely accepted in diagnostic field. The abilities of automatic haematology analyzers have improved considerably, despite its disadvantages detection morphological in of abnormalities [1]. Automation reduces man power, minimizes turnaround time, and also provides quick and accurate results within the stipulated time. Hematology Autoanalyzers (HA) use various principles for counting platelets namely electrical impedence, light scattering, fluorescent detection and immunoplatelet counts. Despite the sophistication of present day instruments there is still a growing need to depend on manual techniques for primary calibration; especially in case of platelet count. There are several studies which have proven that HAs show spurious counts which can be lower or higher than the actual value [2]. Moreover, Automated analyzer had pitfalls due to mimickers of platelet like particles which validated the manual count in peripheral smear regarding platelet count by leishman's stain.

Evaluation of platelet counts is crucial for certain conditions like dengue fever. Platelet counts are considered the most valuable parameter to assess the intensity of the illness and also to monitor the progression of the illness and response to treatment. In such critical situations, manual counting of platelets are considered more reliable compared to automated analyzers. For many years Brecher and Cronkite method was considered the ideal method due to its reliability, accuracy, time and cost [3]. In situations involving daily clinical practice, the need for accuracy and reliability of platelet counts are far more higher. Therefore, this study was done to evaluate the validity of manual counting and automated autoanalyzer counts in peripheral smear for platelet counts.

MATERIALS AND METHODS Study setting

This study was carried out as a cross sectional study in our tertiary care hospital for a period of three months from April to June 2018.

Study samples

All the samples requested for peripheral smear during the study period were included. Overall, 1200

venous samples were selected with EDTA as the anticoagulant in vacutainers. Among the routine 100 to 120 samples/day, platelet counts < 100,000 lakh/cu.mm were taken up for the study. A total of 500 samples were subjected for study.

Data collection tools

Samples were processed by Symex 5000 autoanalyzer and compared with manual platelet count by leishman's stain. Data regarding the background characteristics like age, sex of the patients were noted from the laboratory records. All manual platelet counts were analyzed using standard hematological method described by Dacie and Lewis, while the automated analyzers were done following the manfacturers guidelines. All samples were analyzed within 30minutes.

Ethical approval

Approval was obtained from the Institutional Ethics Committee prior to the commencement of the study.

Data analysis

Data was entered and analyzed using Microsoft Excel spreadsheet for Windows 10. The results were expressed as percentages.

RESULTS

About 500 samples with platelet counts < 100,000 lakh/cu.mm were analyzed. Amog the study samples, 241 were inpatients and 259 were out patient samples. It was observed that in 384/500 (76.8 %) haematology analyzed correlated with peripheral smear review. In about 116(23%) there was no correlation between the manual counting and automated counting. Moreover, there were RBCs and large platelets in peripheral smear counted as platelets by autoanalyzer (Table-1).

Table-1: Comparison between manual and automated autoanalyzer among the study participants

S. No	Characteristics	Frequency (N=500)	Percentage (%)
1	Type of patients		
	In patients	241	48.2
	Out patients	259	51.8
2	Correlation between both the methods		
	Correlation present	384	76.8
	Correlation absent	116	23.2

DISCUSSION

In the study we did not obtain significant variation in results related with age and sex, male or female, younger or older individuals by counting platelets manually and in automatic analyzer. When platelets are activated they become spherical with hypogranular cytoplasm and release small particles. These particles mostly give erroneous results regarding automation but can be rectified with such suspected cases using manual smear examination method. Limitation of technology, automated platelet count can be inaccurate even at normal or high platelet ranges owing to the presence of substantial amount of interfering particles including WBC's fragments, RBC fragments, bacteria, lipid droplets and bacteria etc.,. But in case of neonatal thrombocytopenia, i.,e..due to placental insufficiency, fetal hypoxia, sepsis, Necrotizing enterocolitis, viral infections includes cytomegalovirus, Rubella virus, staphylococcus infection, Escherichia Coli, etc., platelet count has to be done manually for accurate diagnosis. Limitation of technology, automated platelet count can be inaccurate even at normal or high platelet ranges owing to the presence of substantial amount of interfering particles including WBC's

fragments,RBC'sfragments,bacteria,lipid droplets and bacteria etc.,

In a study done by De la Salle BJ *et al.*, around 67% of the automated analyzer results were found to be overestimated. Moreover, statistically significant differences in platelet counts were observed in 16.5% of the cases [4]. There are however newer modalities using two dimensional counting systems with the automated autoanalyzer, employed by the ADVIA counters which check for accuracy in differentiating platelets with non platelet particles, better compared to one dimensional counting systems [5]. In a study done by Bakhubaira *et al.*, there was a positive correlation observed between manual and automated autoanalyzers. This correlation was found to be statistically significant. This study also observed a difference in the mean platelet counts between both the techniques [3].

CONCLUSION

Our study concludes there was significant correlation between automated and manual counts. But this was not applicable for very high or low platelet counts. As platelet count estimation is very important element of the diagnostic and treatment disorder. The accuracy depends on the instrument bias and activated platelet components whereas in manual count this was eliminated. The examination of peripheral blood smear using leishman's stain did not reveal any interference with non platelet particles. Peripheral smear

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examination along with automated analyzer valid in case of high or low value platelet counts.

Declaration

Conflict of interest – nil Funding – nil Ethical approval – obtained

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