Original Article

Utility of Peripheral Film Findings and its Correlation with Automated Analyzer – An Audit from Tertiary Care Hospital

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ABSTRACT

Background and Objective: With the advent of automated hematology analyzer, the use of traditional microscopy of blood film has become limited. The objective of our study was to determine the percentage of peripheral blood smear review in our institution in the era of automation and to identify reasons of manual review.

Materials and Methods: This was a prospective audit from January 1, 2015, to January 15, 2015. Consecutive complete blood count (CBC) samples and peripheral smear requests made up the sample size. All age groups and genders were included. CBCs were performed on Sysmex XE-5000. The variables to be analyzed included inpatient and outpatient samples, frequency of peripheral film review, identifying reasons of smear review, and addition of information missed by the automated analyzer.

Results: We analyzed 1200 consecutive CBC samples. Peripheral smear was reviewed in 500 (42%) of the cases of which, 241 were inpatient, and 259 were outpatient samples. In 384/500, the findings of hematology analyzer correlated with peripheral smear review. Flags identified included nucleated red blood cells (NRBCs) in 155 (40%), immature white blood cell (WBC) 129 (34%), and atypical lymphocytes 100 (26%). In 23% of the cases, the analyzer missed important findings. The sensitivity of abnormal histogram in our study was 91.3%, while the sensitivity of abnormal parameters was 100%.

Conclusion: Peripheral smear review was performed in 42% of the cases. The analyzer identified NRBC, immature WBC precursors, and atypical lymphocytes as the most common abnormality. The information correlated in 77% of the cases.

Key words: Automated analyzer, complete blood count, peripheral film

INTRODUCTION

Examination of properly prepared peripheral blood film offers invaluable information about morphological changes which are not provided by automated instruments. It also provides quality assurance information of complete blood counts (CBCs) data generated by laboratory hematology analyzers.

It is a useful and economical diagnostic tool which can be used both in adults and children and despite the advent of automated blood cell analyzers, examination of peripheral smear by the experienced technologist, and qualified hematologists cannot be repudiated. Rapid, reliable access to information about a variety of hematologic disorders is provided; in some cases, review of peripheral smear along with clinical data may be sufficient enough to establish a diagnosis.[1]

The blood film reflects functional status of the bone marrow, the factory producing all blood elements and its examination is particularly

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important when assessing patients with cytopenias. Other conditions in which the peripheral film findings can be diagnostic include microangiopathic hemolytic anemia, hemoglobinopathy, myeloproliferative disorders, and parasitic infections (especially malaria).

With the advent hematology analyzers, microscopic review of peripheral blood film is declining. Sophistication of analyzers has increased to the point that they are able to provide cells counts, differentials, plots, and histograms.

Microscopy and manual differential counting, in most of the automated laboratories, is restricted to cases in which the instrument "flags" the potential presence of abnormal cells or in cases where findings may interfere with analysis (such as overlap in the distribution of different cell types or interference from matrix components). In cases of clinical suspicion of leukemia, review of the peripheral blood smear is mandatory to make the presumptive diagnosis. There are other morphological findings also which may have a clinical significance which cannot be reliably identified by the various automated analyzers. These other findings include the presence of giant platelets, platelet clumps, basophilic stippling, hypersegmented neutrophils, red cell fragments, and Howell-Jolly bodies.[2]

Since the inception of automated differential counting methods, manual blood smear review is recommended as a validation, rather than as a replacement of automated methods.[3]

Reviewing peripheral smear and performing manual differential counts need the expertise of well-trained laboratory staff and leads to under productivity and consumption of time. This has a much greater impact when the automated and manual results are similar leading to decreased working capacity of house staff. In this era of medical advancements and automation, it is important to reduce the workload and improve turnaround time to combat the continuing pressure on laboratory resources.[4] However for that purpose, important diagnostic information must not be missed by completely relying on morphological findings given by the analyzer as automation does not provide all the information that is potentially important to the physician.[5] The Colleges of American Pathologists (CAP) have conducted numerous studies though Q-probes program to determine performance benchmarks in Pathology. Novis *et al*. in 2006 have reported peripheral smear review frequency of 26.7%.[6]

With this background, our objective was to determine the frequency of peripheral smear review in our institution and compare it with CAP standards. We also wanted to correlate the findings of peripheral smear and analyzer flags and to identify information which was missed by the analyzer.

MATERIALS AND METHODS

This was a retrospective audit conducted in the section of Haematology, Department of Pathology and Laboratory Medicine of The Aga Khan University (AKU) located in Southern Pakistan. AKU is a tertiary care hospital with well-equipped Clinical Laboratory offering over 700 test menu in different sections and is considered as a national reference setup. In our section, approximately 1500 CBCs are reported every day. Peripheral blood film is made of only those blood samples which trigger laboratory policy of smear review defined as abnormal counts (set of criteria established by our expert opinion consensus) and flagging given by the analyzer.

For the study, data for 2 weeks (January 1, 2015–January 15, 2015) were collected from three traditional shifts with simple random sampling technique as the sampling frame. All age groups and genders were included. Specimens with clots, obvious hemolysis, insufficient amount of sample, incorrect addressograph, or wrong vacutainer were excluded.

Automated CBCs were performed on Sysmex XE-5000 hematology analyzers. Inpatient samples were received from all hospital locations (i.e., emergency department, wards, special care units, etc.). Outpatient stations included clinics and outside referrals by physicians.

Peripheral films were prepared by Sysmex SP-1000i automated hematology slide preparation unit. Each "positive smear" was reviewed by an experienced technologist and verified by a hematology resident/consultant. For each specimen on which a manual review was performed, we documented the hemoglobin value, white blood cell (WBC) count, and platelet count the primary reason for the review. The frequency of manual peripheral blood smear review, the manual scan review, and the manual differential count was also determined. Data were collected on a predesigned questionnaire and IBM SPSS Statistics for Windows, Version 19.0. Armonk, NY: IBM Corp., was used for data entry and analysis.

RESULTS

During the review period, we analyzed 1200 consecutive CBC samples. Peripheral smear was reviewed in $n = 500$ (42%) of the cases using random sampling technique. Of these

500 samples, 241 were inpatient and 259 were outpatient samples. In 384/500, the findings of hematology analyzer correlated with peripheral smear review. Flags which were identified included nucleated red blood cells in 155 (40%), immature WBC 129 (34%), and atypical lymphocytes 100 (26%). In 23% (116) of the cases, the analyzer missed important findings. These included abnormalities in hemoglobin indices in 54 (47%), WBCs differential counts in 36 (31%) and large platelets in 26 (22%). Furthermore, the sensitivity of abnormal histogram in our study was 91.3%, while the specificity of the same was 8.17%. Accordingly, the sensitivity of abnormal parameters was 100%, and the specificity of the same was 0%. Further details are given in Table 1.

DISCUSSION

Microscopic examination and morphological assessment are an essential part of CBC reporting that provides crucial information apart from the cell counts. Review of peripheral blood smear serves to ensure that no clinically significant finding is missed, besides providing a clue to the diagnosis, when interpreted by a physician.[7] This information is now provided by automated analyzers with more sensitivity reducing the need of manual examination. The purpose of automation is to provide faster reportable results, to reduce the technologist hands-on time, in addition to providing high quality and precision.[8]

Defining acceptable and safe rates for microscopic examination of the blood smear is crucial to ensure the quality of the results, but reported rates are highly variable.[9]

Comar *et al*. verified the review criteria for automated blood counts suggested by the International Society for Laboratory Haematology and their results showed microscopic review rate of 46.03% with false negatives of 6.73%, false positives of 23.27%, and efficiency equivalent to 70.0%. After adapting the review criteria, the microscopic review rate dropped to 37.3% with false negatives reaching 15.5%, false positives of 10.5%, and efficiency of 73.8%.[10]

Pratumvinit and his colleagues evaluated their criteria for manual smear review and after optimization, their review rate was found to be 24.2% with an efficiency of 87.13% and false negative rate of 2.98%.[11]

Another study by Xing *et al*. established review criteria for their analyzer. After modifying the criteria, the review rate was 34.2% versus 50.2%, false negative rate was 5.5% versus 4.2%, and false positive rate was 28.1% versus 18.7%.[12] In our study, the sensitivity of abnormal histogram was 91.3%, abnormal parameter was 100%, and the sensitivity for flagging was 97.9%. Our manual smear review rate was 42%, which is approximately twice as compared to CAP standards. Since we used random sampling technique, the possibility of missing important diagnostic findings cannot be entirely excluded which is the limitation of the study.

The clinical laboratory of AKU is a reference laboratory where CBCs are received not only from admitted patients but also from collection points located all over the country and outside Pakistan. Our goal of performing this audit was to reduce the workload of microscopy as far as possible to increase laboratory efficiency in terms of time, workforce, and resources by relying on automated analyzer but without missing vital information, i.e., false negative results. Our laboratory is in the process of obtaining CAP accreditation. The processes and standard operating procedures have been modified accordingly. With the accreditation underway and based on the results of this audit, our way forward will be to redefine our triggers for peripheral smear review which will not only decrease workload in laboratory but also shorten the turnaround time of CBC reporting. Once the changes have been implemented, we will perform a re-audit to complete the cycle.

CONCLUSION

It is concluded that the frequency of peripheral blood smear review in our setup was 42% with our less stringent criteria. In 77% of cases, the findings of peripheral blood smears review correlated with that of the analyzer.

Ethical approval

The study was given exemption from ethical approval by the Ethical Review Committee of The Aga Khan University (3573-Pat-ERC-15).

Table 1: Diagnostic accuracy

Variable	Sensitivity (%)	Specificity (%)	Positive predictive value (%)	Negative predictive value (%)	Positive likelihood ratio	Negative likelihood ratio	Accuracy (%)
Abnormal histogram	91.3	8.17	91.3	8.17	0.99	1.06	48.6
Abnormal parameter	100	0	-	-	-	-	-
Flag	97.9	4.2	95.5	4.2	1.01	0.59	49.8

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Conflicts of interest

There are no conflicts of interest.

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