

Kaiser Permanente Interlaboratory Abnormal Cell Study Comparing Slide Quality of the Sysmex SP-100 Automated Slide Preparation Unit to Manual Technique

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In a collaborative effort with 29 Kaiser-Berkeley clinical laboratories in Northern California, the Regional Laboratory and Hematology Supervisors' Peer Group developed and executed an interlaboratory test comparison to establish a defined relationship for the same tests performed in the Medical Centers' laboratories and Regional Laboratory. The test comparison supports the determination of test accuracy, quality and consistency of hematology slide preparation and evaluation. The abnormal cell case study described in this article compared the manually-prepared and conventionally-stained slides of a 14 year-old chronic myelogenous leukemia outpatient with slides prepared with the Sysmex SP-100 automated slide preparation unit utilized at Regional Laboratory. Medical technologists in each of the 29 participating laboratories then reviewed and evaluated both slides. It was the consensus of the laboratorians that the SP-100-prepared slides exhibited overall superior quality and consistency in comparison to the manually-prepared slides. The most notable difference was the distinct staining of cell granulation (i.e., basophils and immature cells) with the SP-100-prepared slides. The participants also commented regarding the excellent blood film quality and uniformity of the SP-100 "wedge-type" smear.

All laboratorians were able to readily identify abnormal cells consistent with myelogenous leukemia disorder on this patient when reviewing the SP-100-prepared slides. Based on the results of this test comparison and other subsequent comparisons, the Hematology Supervisors' Peer Group has selected the SP-100 as the "benchmark" for overall slide quality within the Kaiser Northern California Laboratory System.

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Key Words Complete Blood Count (CBC), Hematology Sample Transportation (HST) System, SP-100, Automated Slide Preparation, Unit, Stain Quality

INTRODUCTION AND BACKGROUND INFORMATION

The Kaiser Permanente Regional Laboratory, located in Berkeley, California, was founded in 1977. The Regional Laboratory's mission is to provide comprehensive specialized testing, non-urgent and selected urgent testing that achieves financial savings through economies of scale in utilization of technology, resources and specialization. The laboratory offers a wide range of clinical testing using state-of-the-art instrumentation and methodologies. Over 300 employees work at Kaiser Regional, providing staffing 24 hours per day, year round. There are approximately 10,000 specimens processed and tested on a normal work day. In addition to the Regional, Laboratory, there are twenty-nine Medical Center laboratories that comprise the Kaiser Permanente Laboratory System in Northern California.

The test menu in the Hematology section of Kaiser Regional includes Complete Blood Count (CBC), Autodifferential (Autodiff), Reticulocytes and Manual Differentials. The specimen volume is approximately 3,500 - 4,000 CBCs and 400 - 600 Autodiffs per day. The specimens are processed in a fully automated, robotic and integrated computerized system. Hemogram results with

abnormal flag(s), and/or those that have exceeded user-defined smear review criteria, require microscopic smear review by medical technologists. All specimens that require smear review are processed in an automated fashion using two Sysmex SP-100 automated slide preparation unit integrated into a Hematology Sample Transportation (HST) System. The automated slide preparation unit process approximately 400-500 slides a day. The SP-100 has a throughput of 120 samples per hour per instrument. The instrument generates wedge-type smears. The patient smears processed at Medical Centers' laboratories are manually prepared and conventionally stained with various commercial stainers.

In a cooperative effort among Medical Centers' laboratories and Regional Laboratory, an ongoing and comprehensive quality assurance program is in place to insure accurate test results, consistency and reliability. This abnormal case study provides only one example of many approaches which the Kaiser Permanente Laboratory network utilizes in their ongoing commitment to Quality Assurance. These types of interlaboratory correlation studies are now required by the 1999 College of American Pathologists (CAP) regulations.

The interlaboratory abnormal cell case study described in this article was conducted in March of 1999. The objec-

tives of the test comparison study on abnormal cells were as follows:

1. Determine the qualitative correlation of smear and staining quality of SP-100 automated slide preparation and staining instrument versus manual technique and conventionally-stained smears.
2. Determine technologists' smear evaluation and consistency in reporting abnormal cell identification.
3. Determine areas of weaknesses on abnormal smear evaluation for continuous quality improvements.

All slides were prepared using the same abnormal patient specimen collected from a 14 year-old male who was subsequently diagnosed with chronic myelogenous leukemia.

MATERIALS AND METHODS

As noted above, the patient sample chosen for this study was collected from a 14 year-old male who had been hospitalized for four days. Two weeks prior to admission to the hospital, he complained of tiring readily during physical education class, and he started to display flu-like symptoms. His health continued to deteriorate until admission to the hospital. The preliminary diagnosis was leukemia of unknown subset. His fourth day CBC results were as follows: White blood cell (WBC) of $74.0 \times 10^9/L$; hemoglobin (HGB) of 8.4 g/L; Platelet count of $487 \times 10^9/L$. The final diagnosis was chronic myelogenous leukemia (CML). After receiving interferon and hydroxyurea, the patient's WBC receded to $6.7 \times 10^9/L$, with some left shift observed on the smear.

Each of the twenty-nine participating laboratories received two slides, with the exception of four laboratories that received one slide which was prepared and stained on the SP-100 instrument due to insufficient volume of sample. The two slides sent to the remainder of the participants consisted of one slide prepared and stained on the automated instrument (SP-100), and one unstained methanol-fixed slide. The unstained methanol-fixed slides were then stained using the various commercial staining devices available in each respective Medical Center laboratory. Medical technologists at each of the laboratories then proceeded to review, evaluate and compare their own conventionally-stained smear with Regional Laboratory's SP-100-stained smear.

RESULTS

Four weeks after the two slides were sent to each participating laboratory, the interlaboratory test comparison results were discussed in the Hematology Supervisors' Peer Group meeting. The results were both surprising and dramatic. The results of the abnormal cell test comparison study were as follows:

1. The participants were able to identify, with ease, the presence of significant Basophilia and other distinct granulation on immature cells (*Photo 1*). The presence of an abnormal number of a specific cell type, along with distinct granulation on immature cells, are valuable morphological criteria utilized by technologists to determine identification of abnormal cells on a

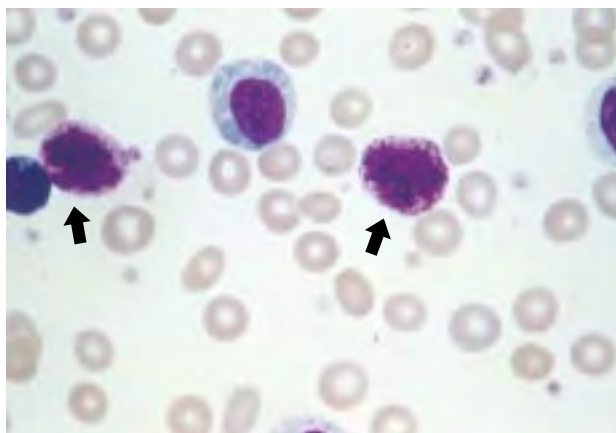


Photo 1 Peripheral blood smear on a 14 year-old male patient diagnosed with CML. The SP-100 smear above showed the presence of a significant number of Basophils. (Oil immersion field).

smear.

2. The majority of participants had observed a significant variation in slide quality between the automated instrument (SP-100) and slides stained by various commercial stainers. The SP-100 stain quality was found to be superior when compared to the other stainers used in Kaiser's Medical Center laboratories.
3. Consistency in cell distribution was observed on the smears prepared on the SP-100 when compared to the manually-prepared smears (*Photo 2 (a) and (b)*). The SP-100 instruments' consistency in cell distribution was attributed to the standard delivery of a precise volume of whole blood to the slides. Thus, it is possible to gain improved precision on manual differential counts when using slides prepared by automated instruments such as SP-100.

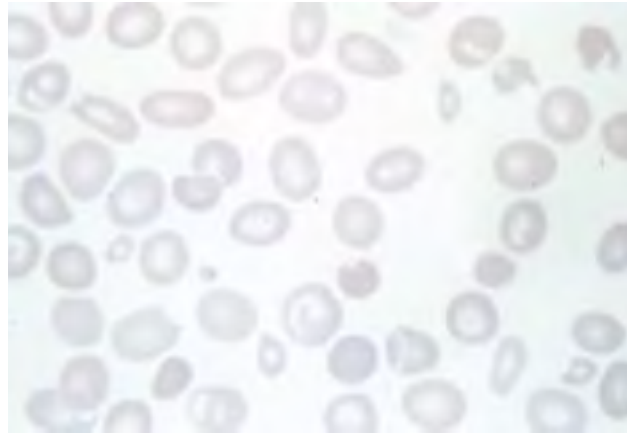
DISCUSSION

CML, a clonal myeloproliferative disorder, results from the neoplastic transformation of the hematopoietic stem cell. The disease is characterized by the presence of a balanced translocation between the long arms of chromosome 9 and 22, (t(9;22)(q34;q11), referred to as Philadelphia (Ph) chromosome. This abnormality is present in over 90% of patients with CML, and 30-50% of those without cytogenetic evidence of the Ph chromosome have molecular evidence of rearrangement. Although CML occurs in all age groups, less than 10% of cases occur in patients under 20 years of age, such as the case described in this article. Approximately 20% of patients are asymptomatic, with mild anemia, leucocytosis and adequate platelet counts. The natural history of CML is characterized by a biphasic, and sometimes a triphasic, course. The disease is usually diagnosed at the chronic phase, such as the sample used in this case. Following treatment, the disease may evolve into a blastic phase, which is sometimes preceded by an intermediate or accelerated phase.

The clinical courses of patients with CML are heterogeneous (*Photo 3 (a), (b) and (c)*). In the particular sample

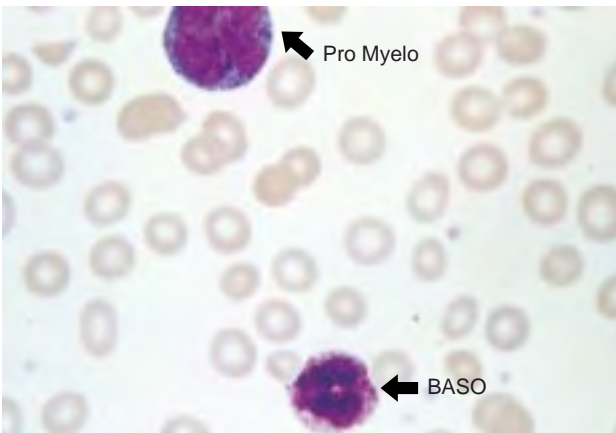


2 (a) Low Power Field

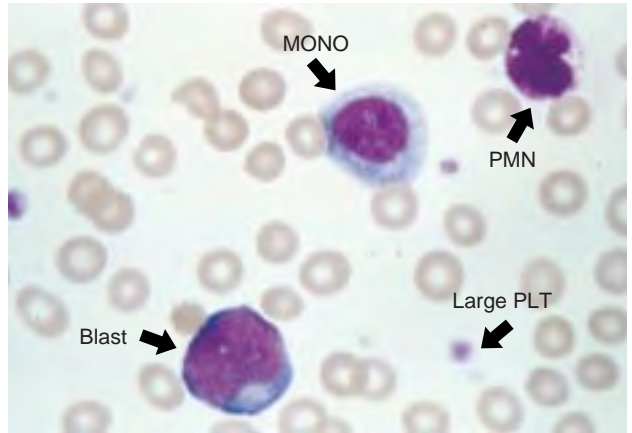


2 (b) Oil Immersion Field

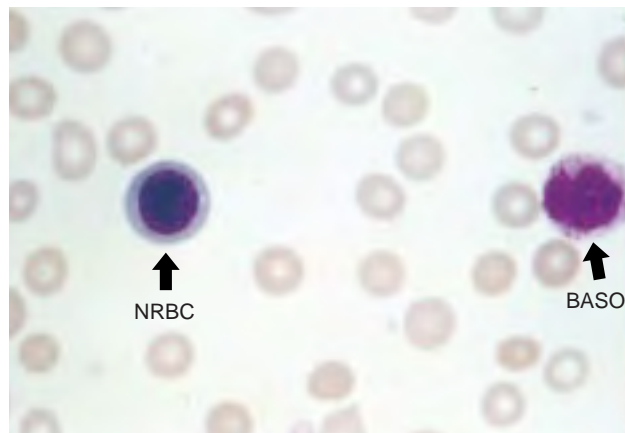
Photo 2 (a) and 2 (b) Low Power Field (a) and Oil Immersion Field (b) showing peripheral blood cell distribution of SP-100 prepared and stained smears on a 14 year-old patient diagnosed with CML.



3 (a) Pro Myelo and Basophil



3 (b) Blast, Monocyte, PMN, and large platelets



3 (c) NRBC and Basophil

Photo 3 (a), 3 (b) and 3 (c) Oil Immersion Fields showing heterogeneous cells characteristic of CML. Peripheral blood smears above were prepared and stained on SP-100.

utilized in this test comparison study, the SP-100 automated slide preparation unit demonstrated good smearing and staining quality. In addition, distinct granulation on abnormal cells was evident when compared to granulation on normal cells. The observer's capability to find and identify abnormal cells begins with a properly made and stained smear that preserves both cytoplasmic and nuclear characteristics of clinically significant cells. The SP-100 instrument has demonstrated its capability to consistently produce quality smears that meet the desired outcomes of our laboratory.

CONCLUSION

It was the consensus of participants that the SP-100 automated slide preparation unit produces slides of superior quality and consistency when compared to the manual technique. The above results demonstrate the enhancements and benefits gained when an automated slide preparation unit such as the SP-100 is utilized. The total automation process of slide making and staining with the SP-100 has achieved the following departmental objectives:

1. Improved precision on manual differential counts
2. Improved workflow efficiency and effective personnel time utilization
3. Improved quality on technologists' smear review and

evaluation

4. Shorter turnaround time on abnormal smear reviews
5. Improved employee safety resulting from decreased biohazard exposure
6. Increased test capacity
7. Affordability due to economies of scale

Based on the results of the test comparison conducted, as well as subsequent studies, the Hematology Supervisors' Peer Group has selected the SP-100 instrument as the quality "benchmark" for overall smear/stain quality within the Kaiser Permanente Laboratory System in Northern California.

References

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