SUMMARY

Automation is the replacement of human manipulative effort by mechanical and instrumental devices that are regulated by feedback of information so that an apparatus is self-monitoring or self-adjusting.

Advantages of automation are providing assays at shorter turn-over times, greater accuracy and better reproducibility, smaller sample and reagent volumes, decreased personnel involvement, reduced assay costs, and the capability of data storage and analysis.

Types of automation include microautomation and macroautomation, semi-automation and full automation and partial and total laboratory automation (TLA).

Automated hematology counters can be classified into aperture impedence counters, optical (light scattering) counters and combined aperture impedence and optical counters.

Advantages of automated hematology analyzers include performing variety of hematological measurements as hemoglobin, hematocrit, mean corpuscular volume (MCV) and leucocyte differential from one sample of blood with high speed and accuracy with the CV (*Coefficient of Variation*) for most of the parameters measured is in the range of 1 to 2%. This level of reproducibility is not achievable with the use of most manual techniques.

Other advantages of hematology analyzers include providing parameters couldn't be obtained in the past as

mean corpuscular volume (MCV), mean platelet volume (MPV) and red cell distribution width (RDW). Moreover, is the incorporation of argon laser technology, allowing integration of some flow cytometric data using specific fluorochrome stains, such as T-cell subsets (CD4:CD8) with routine hematologic analyses allowing flowcytometers to be redirected to more in-depth analyses of cell structure and function.

Recent hematology analyzers e.g. ***Sysmex XE-2100*** can enumerate, rather than just flag for, immature granulocytes. In addition, a new blast-detecting program has been developed through a combination of multiple parameters that were originally equipped in XE-2100 to improve the performance of finding blasts of acute myeloid leukemia (AML). This system will come within the range of the detection of MRD in the routine hematologic examination.

Also, automated techniques for blood smear preparation and staining have been developed e.g. ***Beckman Coulter™ Gen•S™ Cell*** that produce very uniform blood smears for accurate morphological examination, as the manual blood film differential remains the definitive tool for complete hematological analysis. In general, smears made by automated techniques are usually inferior to those made by an experienced technician.

More recently, automated methods for 3D visualization of peripheral blood smears have been developed e.g. ***DiffMasterTM Octavia***, with the ability to perform differential counting and sensitivity to detect blast cells slightly higher than manual microscope, markedly reducing effort, time and cost, together with the ability for data storage for future follow up and re-examination.

Flowcytometry is now a routine procedure used in hematology, immunology and oncology. It is regarded as the method of choice for immunophenotyping because it is fast, objective, quantitative and amenable for standardization. Flow cytometric immunophenotyping is an essential part of the diagnostic procedure in the ***acute lymphoblastic*** and ***myeloid leukaemias*** (ALL and AML) and ***chronic lymphoproliferative disorders***, and its results constitute useful information for therapeutic decision making in these diseases. Also, flow cytometric monitoring during residual disease may have diagnostic and therapeutic utility in patients with acute leukaemia

Automation in immunohistochemistry using ***top-down capillarity***, ***ascendant capillarity***, or ***flat immunohistolabelling*** allows standardization of slide preparation, a walkaway operation of IHC, an improved traceability, increase output, improve reproducibility and quality and selfchecking of critical steps.

Automated karyotyping systems, relying on computerized image analysis, have become available, greatly increasing the efficiency of karyotyping in laboratories that handle large numbers of samples. The precision is presently comparable to that of conventional methods. The number of steps and the amount of time required for cytogenetic analysis are thereby significantly decreased, allowing more rapid analysis and reduction of labor costs.

Also in the field of cytogenetics, automated methods for pretreatment of specimens, before Fluorescent *in situ* hybridization (FISH), have been developed with the end result of increased flexibility, greater consistency, reduced labor, and improved efficiency.

The new multicolor karyotyping techniques, "multiplex FISH" (M-FISH) and "spectral karyotyping" (SKY), allowed the visualization of the entire chromosome complement in 24 different colours. The power of M-FISH and SKY is in their ability to discriminate the components of highly re-arranged chromosomes such as those present in complex karyotypes. However, both techniques are unable to detect small intrachromosomal rearrangements (inversions, deletions, duplications).

With the introduction of microarray-based comparative genomic hybridization (array CGH), the main limitation of conventional CGH, a ***low resolution***, is overcame. Array CGH does not require dividing cells, as does karyotyping. Moreover, it enables the analysis of the whole genome in a single experiment, which can be compared with thousands of independent FISH hybridizations. In addition to the high specificity, the sensitivity of array CGH is also very high. Another advantage is that array CGH is a ***fast technique***, because part of the procedure is ***(semi)automated***.

Both ***comparative genomic hybridization*** **(CGH)** and ***interphase fluorescence in situ hybridization*** **(interphase FISH)** avoid the use of metaphase chromosomes altogether and have allowed the genetic analysis of previously intractable targets.

Polymerase chain reaction (PCR) has filled the gap in the respect of hematological malignancies diagnosis where the abnormal clone of cells needed to be studied are present only in a very small amounts, for example detection of ***minimal residual disease (MRD)***. The specificity of PCR can be further increased by the use of ***nested PCR*** which involves re-amplification of a small amount of the amplified product.

The use of automated PCR\OLA procedure for analysis of DNA polymorphisms and variants has many advantages e.g. small numbers of cells (cheek scraping) or DNA samples (10 ng) are sufficient for analysis, reagents are stable and easily obtained.

The ***GeneXpert***, instrument automates all aspects of RT-PCR analysis, including RNA isolation, reverse transcription, quantitative PCR, and data analysis. In addition to full automation, the GeneXpert is also capable of very fast RNA isolation (6 min) and PCR cycling times. The automation, standardization, and reproducibility provided by the GeneXpert could facilitate the testing, acceptance, and routine use of quantitative PCR and RT-PCR assays in clinical diagnostics.

The newly introduced automated coagulation analyzers, e.g. CA-7000 and ACL 10000 provide assays for hemostatic disorders at a short turnaround time, using small volumes of the sample, suitable for emergency room and intensive care, as well as pediatric samples. They provide coagulometric, immunoturbidimetric and chromogenic assays, including tests for D-Dimer, von Willebrand antigen and free protein S.

The new global assay of coagulation and fibrinolysis, the Clot Formation and Lysis (CloFAL) assay, is reproducible, sensitive, inexpensive, rapid assay and possesses the potential for automation.

Recently developed automated platelet function analyzers, e.g. **PFA- 100TM**, were found to be very sensitive to disturbances in VWF, and to the presence of VWD, also, they were found to be a superior VWD screening tool compared to skin bleeding times. However, neither of these analyzers could be described as being specific for vWD. Thus, an abnormal result may suggest the presence of a platelet-like or vWD-like disorder, but neither instrument can diagnose the presence of VWD.

The review of various automated techniques used in laboratory hematology revealed that application of these techniques greatly save effort, time and cost, together with increase efficiency to cope with the high work load.

However, application of these automated methods, especially the most recent of them, costs a lot of money owing to the advanced technology used. Example of these is total laboratory automation (TLA) which possesses a pay-back period of 5 to 7 years and more. This reason makes them difficult to be applied in our country especially at a large scale, leading to marked increase in workload on centralized laboratories and hospitals.

At last, our review of these recent automated techniques revealed that, in spite of their great and rapid advance they can never replace the expert hematologist, and the human interpretation of data and findings which remains the definitive, goldstandard tool to make any diagnosis.