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Case Report

Evaluation of white blood cell count in peritoneal fluid with five different hemocytometers

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ABSTRACT

Objectives: Evaluation of automated flow cytometric analysis of white blood cell (WBC) count in peritoneal fluids.

Methods: One hundred peritoneal fluids were analyzed with manual microscopy, Sysmex XE-2100 and XE-5000, Siemens Advia 2120, Mindray BC-6800, Abbott Sapphire.

Results: High correlations (0.978 to 0.999) and modes biases (−132 to 80 WBC/mm^3) were found. Agreement at septic peritonitis cutoff ranged between 96% and 99%.

Conclusions: These hemocytometers display acceptable performance for WBC screening in peritoneal fluids.

Introduction

The term ascites is conventionally used for designating the accumulation of fluid in the peritoneal cavity, which may onset as a complication of several conditions such as cirrhosis, decompensated heart failure, hepatic veins occlusion, malnutrition and anorexia, renal disorders, cancers and carcinomatosis, infections and pancreatitis [1].

The accurate analysis of peritoneal fluid is of paramount importance in patients with ascites, since it helps troubleshoot the pathogenesis. In particular, assessment of albumin and/or protein concentration is necessary to differentiate between transudative and exudative ascites, whereas the presence of elevate numbers of white blood cells (WBCs) and polymorphonucleated leukocytes (PMNs) is pivotal to diagnose spontaneous bacterial peritonitis (SBP) [2]. According to recent evidence-based recommendations, ascitic WBC count of >1000 cells/mm^3 (positive and negative likelihood ratios of 9.1 and 0.25, respectively) and ascitic PMN count of >500 cells/mm^3 (positive and negative likelihood ratios of 10.6 and 0.16, respectively) provide the greatest accuracy for diagnosing SBP [3]. The microscopic analysis has been the gold standard for enumeration and classification of WBC in biological fluids, even if it carries several inherent drawbacks such as the need of skilled personnel, high imprecision and inter-observer variability, reduced throughput, along with the possibility of generating post-analytical errors due to manual transcription of data. Automated flow cytometry, using either urine [4] or hematology instrumentation [5], represents thereby an appealing perspective for screening or analysis of biological fluids, since it allows to overcome most of the previously mentioned shortcomings of manual analysis. Nevertheless, an accurate assessment of the analytical and diagnostic performance is needed before peritoneal fluid analysis can be routinely performed on automated hemocytometers. The aim of this study was to assess the diagnostic performance of five different hemocytometers for WBC enumeration of peritoneal fluids compared with manual microscopy analysis.

Materials and methods

Instrument characteristics

The following hematological analyzers were used in our study: Sysmex XE-2100 and XE-5000 (Dasit SpA, Cornaredo, Italy), Siemens Advia 2120 (Diagnostic Solutions, Milan, Italy), Mindray BC-6800 (Medical Systems S.p.A., Genova, Italy), and Abbott Cell-Dyn Sapphire (Abbott Diagnostics Division Italia, Roma, Italy).

The WBC count is determined on XE-2100 by flow cytometry, using forward-scattered and side-scattered light, whereas the WBC differential entails a specific nucleic acid dye to measure the cells by side-fluorescent light and side-scattered light [6]. The analysis of WBC is performed on XE-5000 with flow cytometry technique by means of a semiconductor laser and fluorescent measurement. The specific body fluid mode uses the 4-DIFF scattergram to assess and display WBC count and subpopulations [7]. In Advia 2120, light scatter, differential WBC lysis, and myeloperoxidase staining are used to

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analyzed WBC and their parameters. In particular, WBC is enumerated and classified in biological fluids with three optical measurements, including high-angle scatter, low-angle scatter and absorbance. The signals are then digitized and used to construct the cytogram. The primary count is derived from the lobularity/nuclear density channel, but WBC is also enumerated in the peroxidase channel, and a flag is triggered when there is significant difference between the two parameters [8]. The BC-6800 is an evolution of the smaller BC-3600, and uses scatter of laser light at two angles and fluorescence signals to perform complete blood cell count and analyze the WBC subpopulations by means of a 3D

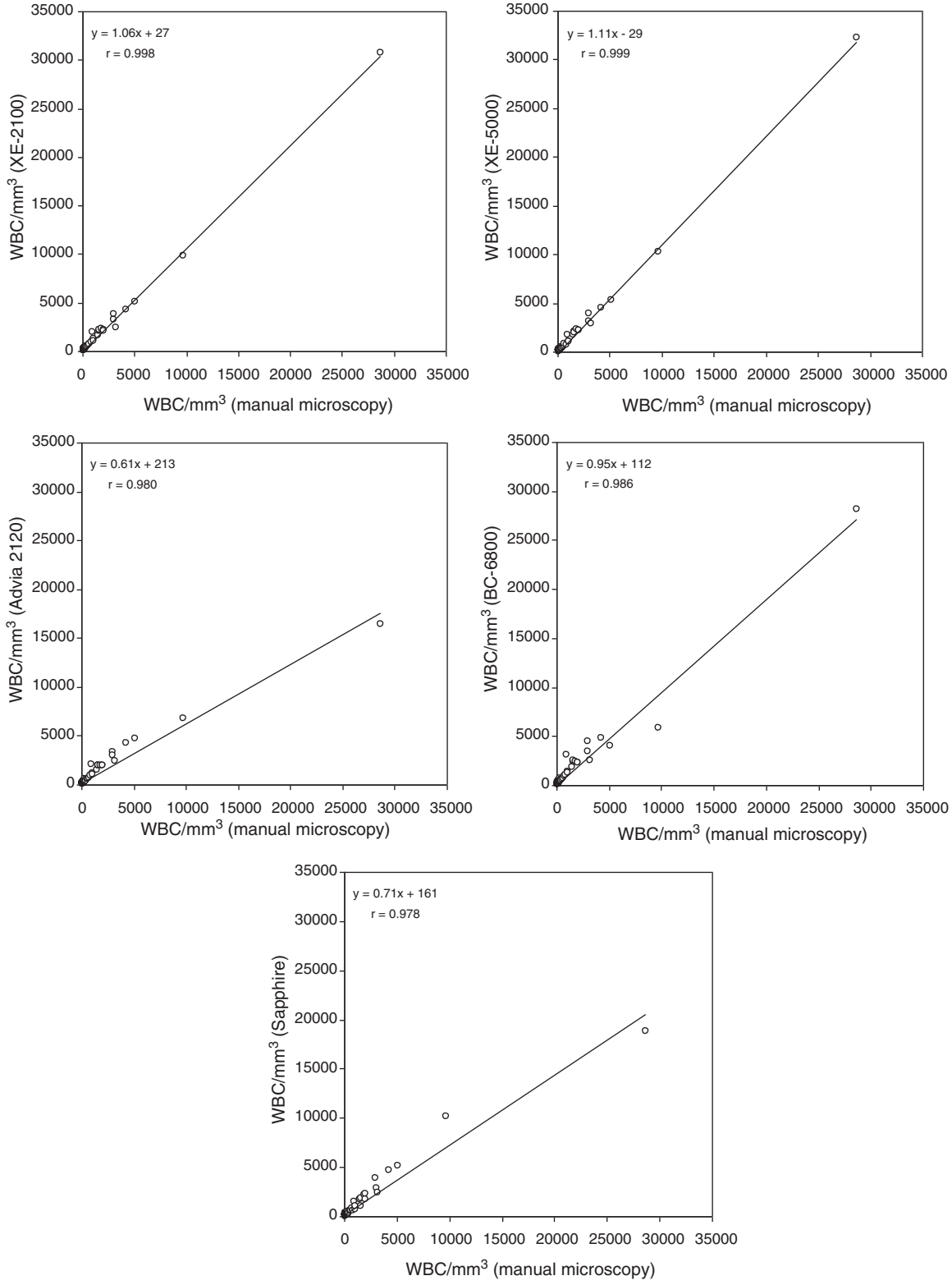

Fig. 1. Linear regression analysis and Pearson's correlation between manual microscopy and Sysmex XE-2100, Sysmex XE-5000, Siemens Advia 2120, Mindray BC-6800 and Abbott Sapphire for white blood cell (WBC) count in 100 inpatient specimens of peritoneal fluid referred to the laboratory for routine analysis.