

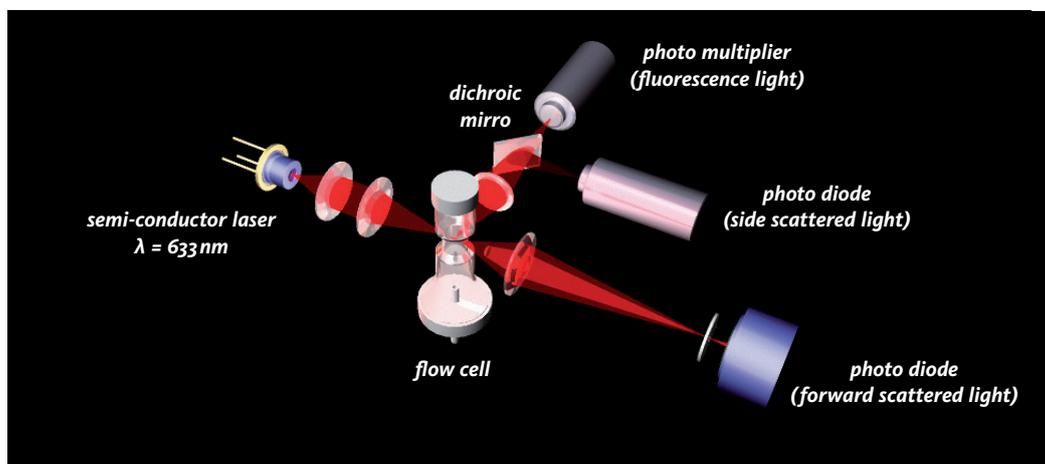
## Examples of body fluid analysis with the XE-5000

Sysmex Xtra Online | March 2011

If patients are to be treated rapidly and effectively, laboratory results must be both rapid and reliable – if at all possible, 24h a day. This applies particularly to some special body fluids, such as CSF, peritoneal fluid (in ascites or CAPD), pleural punctate and synovial fluid. An automated analytical procedure is needed in principle for this work.

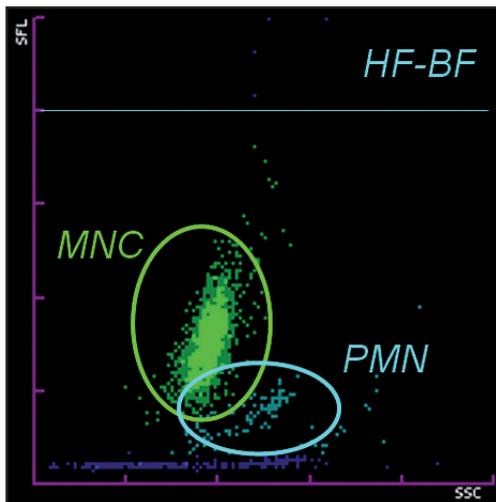
Using the established technology of fluorescence flow cytometry, Sysmex XE-5000 provides the so-called 'body fluid mode', for rapid and reliable cell counts in various body fluids with simple sample handling. About 1 minute after the sample has been aspirated, the special body fluid mode provides the leukocyte and erythrocyte counts, together with the differentiation of the leukocytes into mononuclear and polymorphonuclear cells.

It is particularly important to be able to detect low numbers of cells in CSF, as even low leukocyte concentrations may indicate an infection. Because of the low normal range of cells in CSF, the counting volume in the body fluid mode is increased, to ensure an improved imprecision. The fluorescence signal prevents potential interference from counting e.g. microscopic air bubbles. Using this approach, the XE-5000 can achieve the very low background count value of 0 to 1 leukocyte per  $\mu\text{L}$ .



**Fig. 1** Optical unit of the XE-5000. The fluorescence flow cytometer is very well suited for the analysis of body fluids. Each cell which contains RNA or DNA is stained with a polymethine dye, giving a fluorescence signal which is proportional to the nucleic acid content.

Sysmex's unique fluorescence dye only stains cells containing nucleic acid, so that erythrocytes do not interfere with the leukocyte count or differentiation. As a consequence, even individual leukocytes can be recognised in the DIFF scattergram of the XE-5000 body fluid mode.

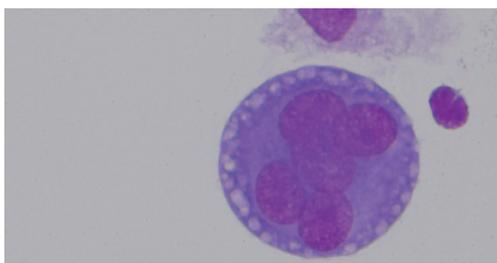
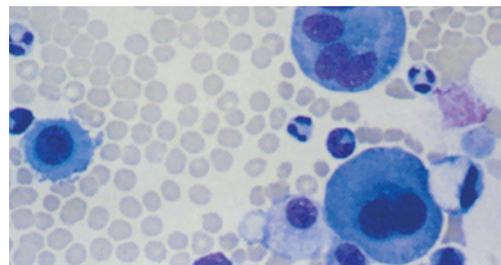
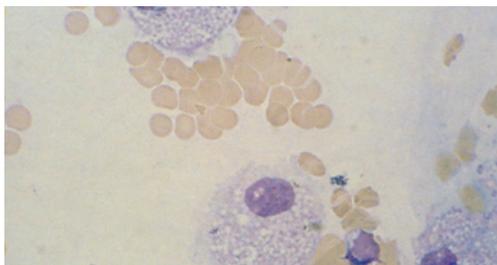


**Fig. 2** DIFF scattergram in the body fluid mode. On the y-axis, the intensity of lateral fluorescence (SFL) is plotted and on the x-axis, the intensity of lateral scattered light (SSC) is plotted. Each dot in the scattergram represents a counted cell. Mononuclear cells, polymorphonuclear cells and the 'ghost' population in each sample are individually and reliably separated from each other based on adaptive cluster analysis (ACAS).

In addition, so-called 'high fluorescent body fluid cells' (HF-BF) are excluded from the leukocyte count and differentiation, as these cells may be macrophages, mesothelial cells or malignant cells. This cell population (HF-BF) is reported separately as a research parameter (% and #) and a flag is displayed in case the user-definable trigger levels are exceeded.

#### Selected cases

The first two cases deal with the detection of malignant cells in CSF (1) and in pleural fluid (2) with the help of the XE-5000 body fluid mode. CSF is very often examined to confirm or exclude infection. For this reason, the third case deals with bacterial meningitis as an example.



**Fig. 3a-c** Macrophages, mesothelial cells and tumour cells

### Example case 1: Cerebrospinal Fluid – Tumour Cells

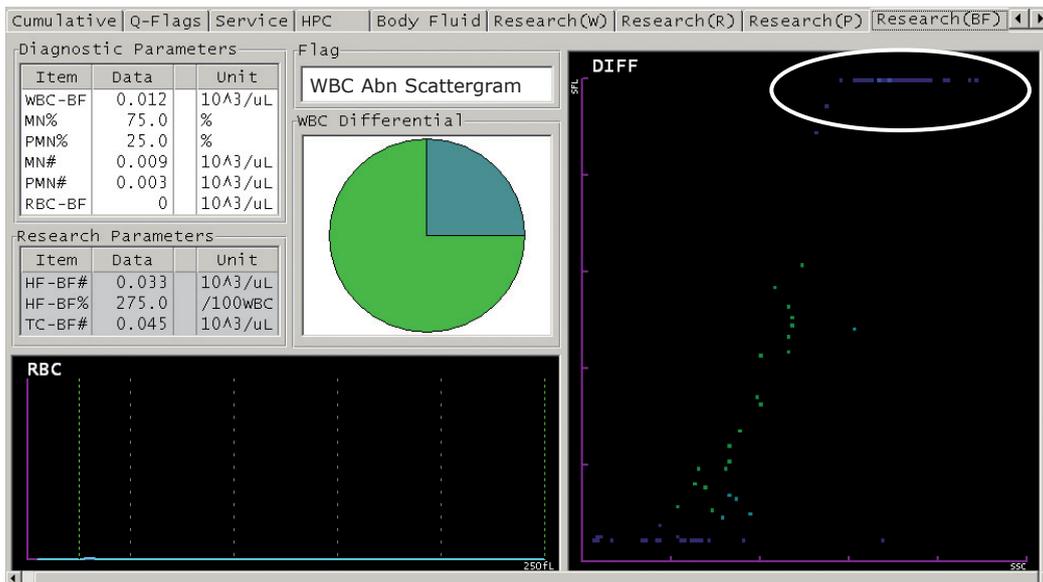


Fig. 4 Information from the XE-5000's Body Fluid Research display

The flag 'WBC Abn Scattergram' indicates an increased concentration of so-called 'high fluorescence body fluid cells' (HF-BF). In this sample, the amount of HF-BF is 275% (cells per 100 WBC) and they are visible at the upper border of the scattergram (see dark blue dots within the ellipse mark).

Although HF-BF cells are not included in the WBC count, it is important to review these kinds of samples under the microscope for abnormalities. Therefore a cytospin smear was prepared of this CSF sample. Microscopic inspection revealed large atypical cells. This patient suffered from a metastasis of an adenocarcinoma (bronchus carcinoma).

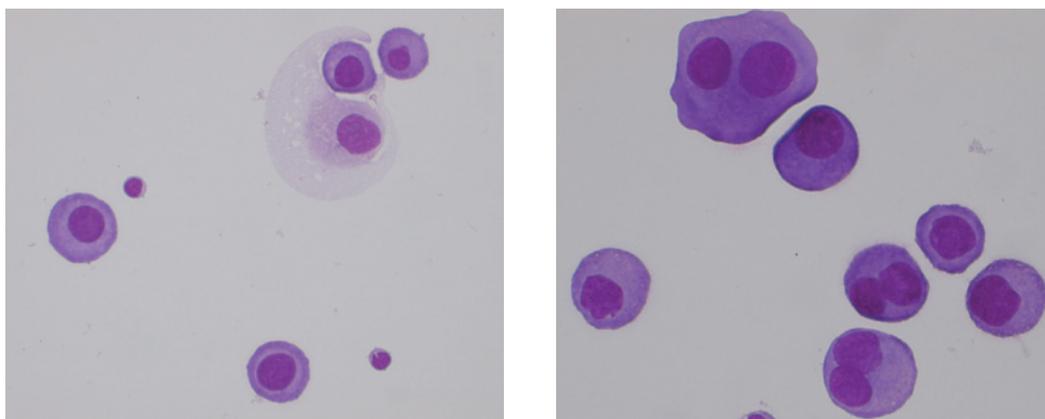


Fig. 5 Large, atypical cells are visible in this CSF sample under the microscope.

## Example case 2: Pleural Fluid – Tumour Cells

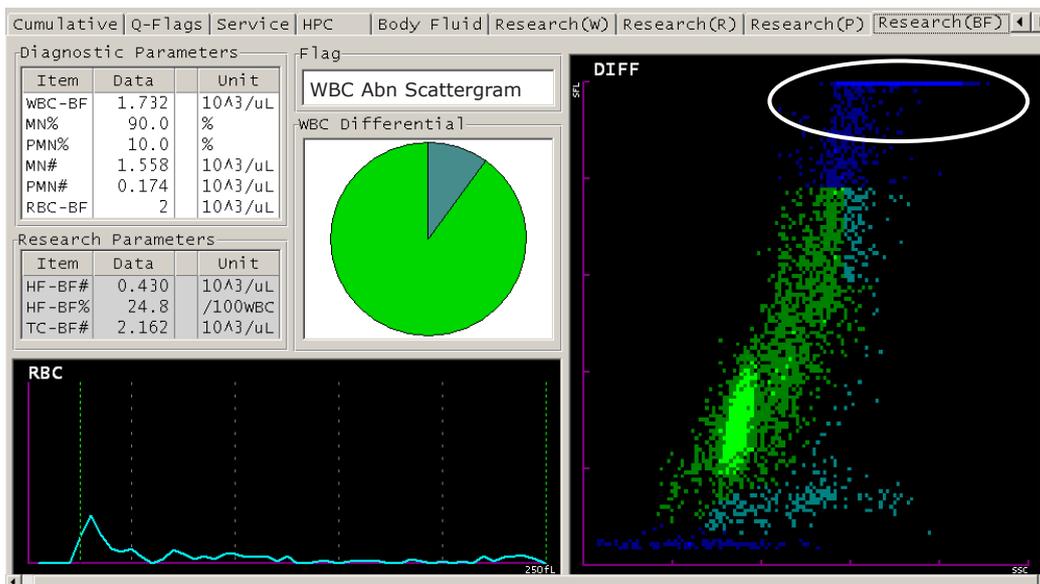


Fig. 5 Information from the XE-5000's Body Fluid Research display

The user-definable flag 'WBC Abn Scattergram' indicates in this pleural fluid sample the presence of macrophages, mesothelial cells or malignant cells. The amount of HF-BF is 24.8% and a high concentration is clearly visible at the upper border of the scattergram (see dark blue population within the ellipse mark).

Because of this increased amount of high fluorescence body fluid cells, a cytospin smear was prepared. Microscopic inspection revealed large, atypical cells. The nuclei are enlarged, hyperchromatic, and sometimes bi- and multinucleated. The N/C-ratio is disturbed and the cytoplasm contains vacuoles. This picture is consistent with an adenocarcinoma.

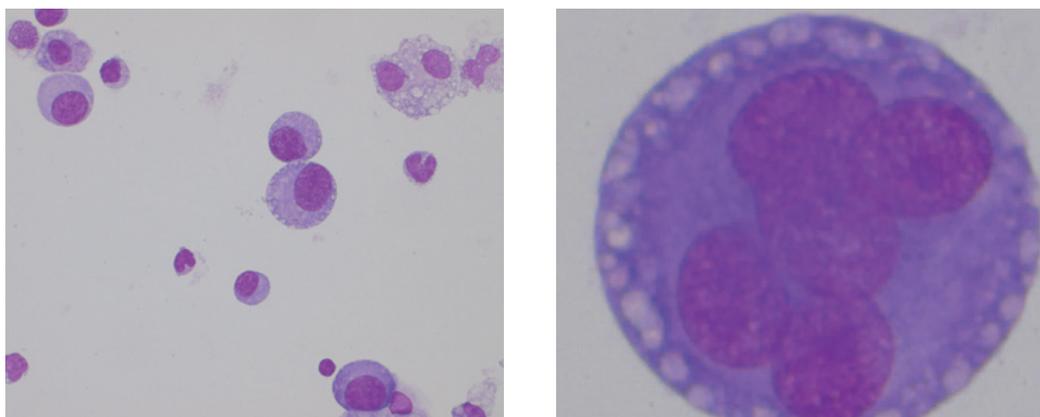


Fig. 7 In the cytospin smear of the pleural fluid sample, large atypical cells can be seen under the microscope.

### Example case 3: Cerebrospinal Fluid – Bacterial Infection

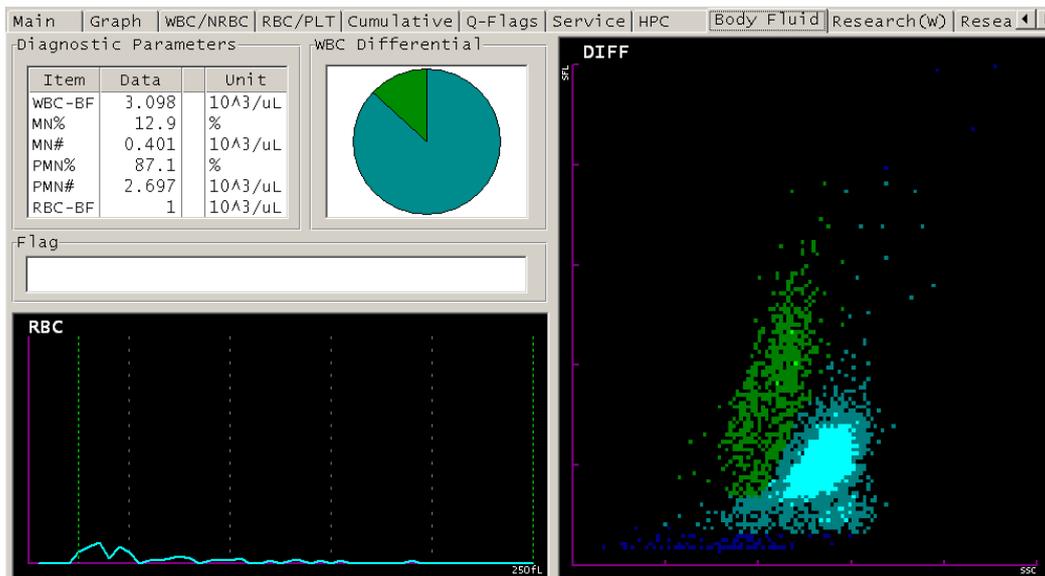
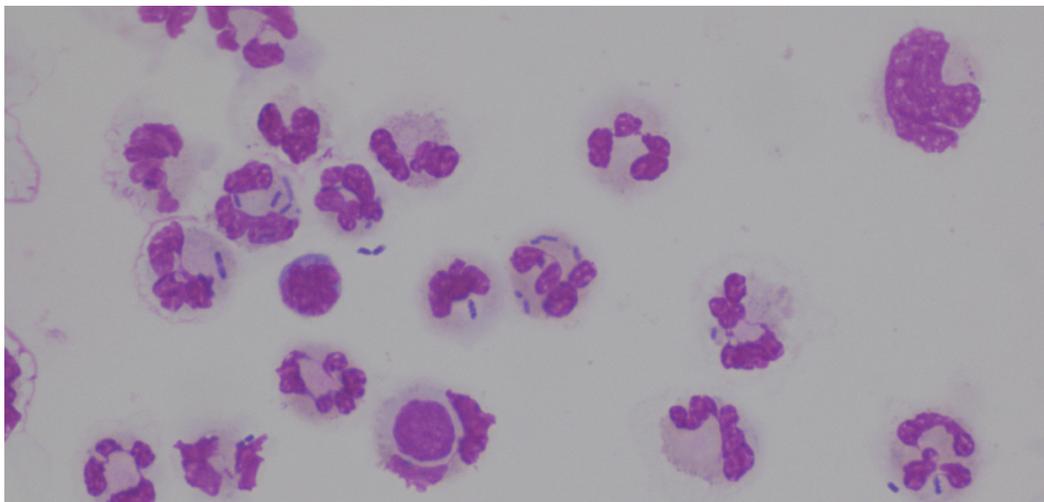


Fig. 8 Information from the XE-5000's 'Body Fluid' display

With a concentration of 3,101 white blood cells per  $\mu\text{L}$ , this CSF sample immediately attracted the attention of the operator. Since the RBC concentration was low, the high WBC count could not have been caused due to a traumatic tap. The WBC Differential pie chart indicates a high concentration of polymorphonuclear cells (PMN). A bacterial infection was suspected and a cytospin smear was prepared. Microscopic inspection revealed the presence of bacteria in the cerebrospinal fluid and also intracellular within the white blood cells. White blood cell clumps were also present in this sample. This patient was diagnosed with bacterial meningitis.



**Fig. 9** The microscopy view shows bacteria inside the polymorphonuclear leukocytes.

