MetaSystems White Paper

Innovations in FISH Scoring: Rapid Scoring

A methodology to dramatically reduce the time to score and result FISH cases

Introduction

Automated fluorescence *in-situ* hybridization analysis (FISH signal counting), in conjunction with the MetaSystems slide scanning platform Metafer, generates results in a reliable, standardized fashion. The automated scanning system provides unbiased interpretation, scoring many cells that are often ignored via manual spot counting. Human scorers, however, regularly pick cells that they can easily interpret to be included in their analysis, while tending to ignore the difficult-to-interpret cells. Therefore, many labs are used to working with very low abnormal cut off values for positivity during manual observation. As a result of these discrepancies between automated and manual scoring strategies, people spend significant time to correct automated results, or to reject cells they do not like.
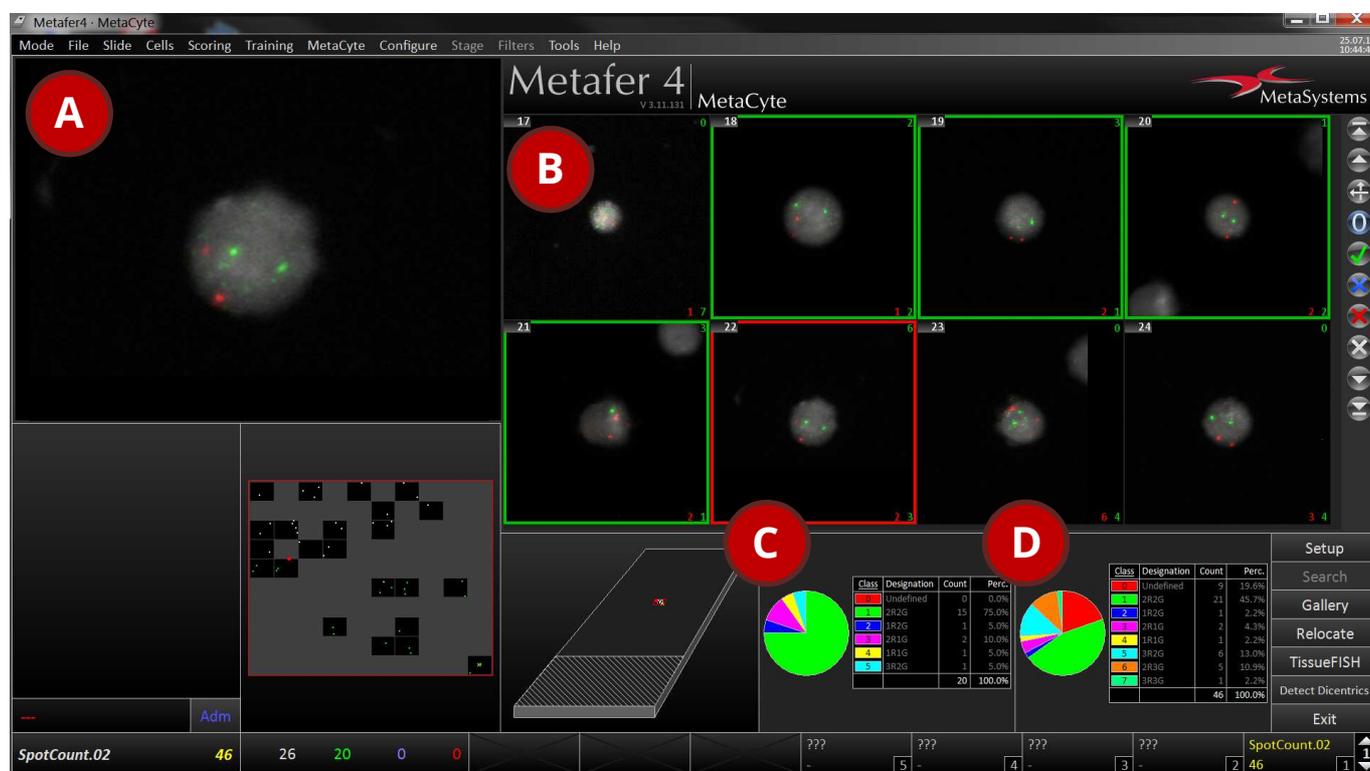

Fig. 1: Rapid Scoring screen in Metafer. For details on the display components please see text.

MetaSystems is pleased to introduce Rapid Scoring (RS), a new method of analyzing FISH signals which combines manual scoring strategies with the advantages of automated scoring. RS uses MetaCyte to automatically score cell signal patterns. All analyzed cells are displayed in a gallery (*Fig. 1 B*). Signal patterns are summarized in a convenient graph and a table (*Fig. 1 D*). Immediately after the scan technicians can begin categorizing cells with doubtful results. Categories are assigned to each cell using an external keypad, and new categories (groups of cells) can be added any time. An empty scoring sheet displayed left of the automated results (*Fig. 1 C*) dynamically displays the manual results while the categorization is proceeding. An enlarged image of

WHITE PAPER

MetaSystems

info@metasystems-international.com
www.metasystems-international.com

the unprocessed cell in question facilitates evaluation of signal patterns (*Fig. 1 A*). In order to test the performance of the RS workflow, we conducted a study with various probe panels at a cancer research institute in USA.

Materials and Methods

Three probes panels (MetaSystems XL probes) for CLL, MDS, and NHL, respectively, were included. 12 cases with multiple probe panels per case were investigated. Samples were hybridized following the standard protocol for MetaSystems probes. 200 cells per sample have been scored manually and automatically using the RS workflow. The manual workflow includes (a) loading slides on manual microscope, (b) preparing the microscope (set filters, apply immersion oil), (c) manual scoring of 200 cells, (d) completing the worksheet with the result, and (e) manual image acquisition of 2-3 images for documentation. The automated workflow includes (a) loading slides on Metafer, (b) RS analysis of 200 cells, (c) exporting of all 200 images and results data to XML files. Hands-on time for each analysis has been recorded in both workflows.

Results

Fig. 2 shows the direct comparison of analysis times for manual (*gray columns*) and automated Rapid Scoring (RS) analysis (*red columns*). Each pair of columns represents a different probe or set of probe (for details on the probes used see caption of *Table 1*). *Table 1* summarizes relative time saving for the analysis of 200 samples. Our results show that, in all experiments, manual analysis takes considerably longer than RS. On average a time saving of more than 40 % could be achieved if RS is used. The results of this study indicate that RS is an excellent method to combine the advantages of automated slide scanning with established manual scoring strategies. Additionally, all cells images are immediately available for long term archiving.
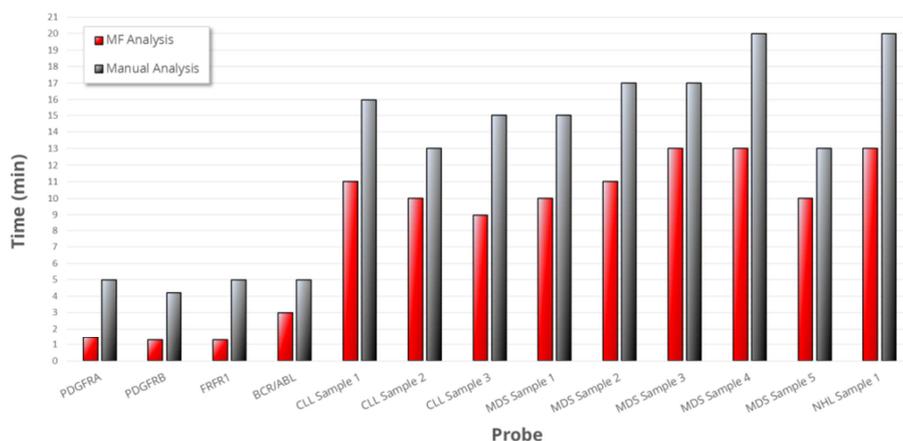
Sample	Time Saved (%)
PDGFRA	71,00 %
PDGFRB	69,05 %
FRFR1	74,00 %
BCR/ABL	40,00 %
CLL sample 1 [*]	31,25 %
CLL sample 2 [*]	23,08 %
CLL sample 3 [*]	40,00 %
MDS Sample 1 [†]	33,33 %
MDS Sample 2 [†]	35,29 %
MDS Sample 3 [†]	23,53 %
MDS Sample 4 [†]	35,00 %
MDS Sample 5 [†]	23,08 %
NHL Sample 1 [‡]	35,00 %
All (avg.)	**41,05 %**

Table 1: Relative time saving using Rapid Score with Metafer, compared to manual analysis.

Probe Panels:
[*]   ATM/P53, CEN12 and 13q/q(3 colors), CCND1/IGH(DFDC)
[†]   5p/q/q (3colors), CEN7/7q, CEN8 and 20q/q (3 colors), MLL+(BA)
[‡]   BCL6+(BA), MYC+(BA), CCND1/IGH (DCDF), IGH+(BA), IGH/BCL2 (DCDF), MALT1+(BA)

Fig 2: Comparison of scoring times using Rapid Scoring (red columns) and manual scoring (gray columns) for all tested probes or probes panels.

WHITE PAPER

MetaSystems

info@metasystems-international.com
www.metasystems-international.com