Technology that counts

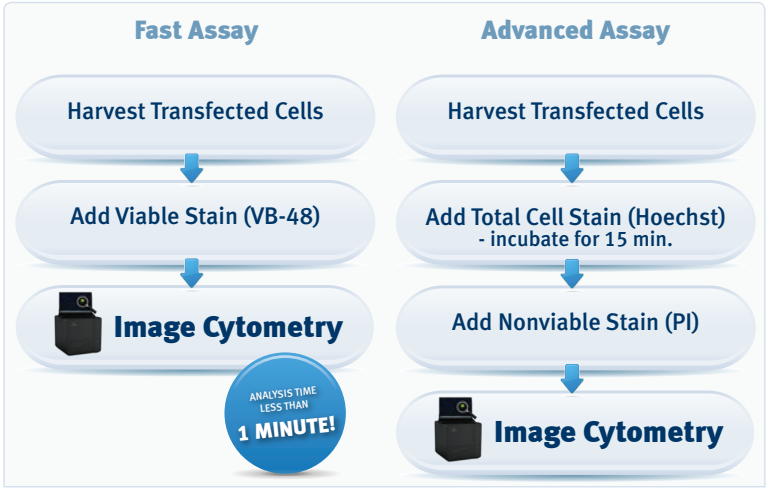
chemometec

NC-3000™ GFP Transfection Efficiency Assay

– For easy, fast and objective measurement of transfection efficiency

The introduction and expression of foreign genes in eukaryotic cells is a technique widely used for investigating gene regulation and function. It is often required that the efficiency of introducing the new gene in to the cells - transfection efficiency - is determined for either process optimization or for the accurate interpretation of results in down-stream results. An easy method for determining transfection efficiency is by the use Green Fluorescent Protein (GFP) as a reporter gene. Using an appropriate promoter, GFP can be expressed in the cells by itself or attached to the protein of interest as a fusion protein. Using GFP as a reporter gene, the transfection efficiency of a population of cells can then easily be determined as the percentage of cells expressing GFP in the entire population.

The NucleoCounter® NC-3000™ enables two assays for measuring GFP transfection efficiency; a rapid method taking less than a minute and an advanced assay with increased sensitivity for the detection of cells with low levels of GFP expression. The advanced assay also provides further information regarding the viability of both transfected and untransfected cell populations.



Key Benefits

of the NC-3000™ GFP Transfection Efficiency Assay

Total assay time less than one minute!

- ✓ Fast and easy-to-use assay for measuring transfection efficiency
- ✓ User friendly protocol with predefined settings
- ✓ Fully automated analysis procedure
- ✓ GFP intensity histogram displayed
- ✓ Objective analysis – standardized results
- ✓ No calibration required
- ✓ PlotManager for superior data presentation
- ✓ Automated PDF reports
- ✓ Export of data in FCS/ACS formats

The NucleoCounter® NC-3000™

- Next generation cell analysis

Viability and Cell Count | Cell Cycle Assays | Caspase 3/7, 8 & 9 | Annexin V | Mitochondrial Potential | Cell Vitality | DNA Fragmentation | GFP Transfection Efficiency | FlexiCyte™ (user adaptable protocols)

Principle: NC-3000™ GFP Transfection Efficiency Assay

In order to determine the transfection ratio, a suspension of cells transfected with GFP is stained with either VB-48 (the fast method) or Hoechst and PI (the advanced method). After staining cells are loaded into either of two types of ChemoMetec slides: the 2-chamber NC-Slide A2™ or the 8-chamber NC-Slide A8™.

Samples are analyzed using the NucleoCounter® NC-3000™ system.

For both methods the transfection ratio and a histogram showing the GFP intensity is shown, moreover, for the advanced method the transfection ratio of nonviable cells is also determined.

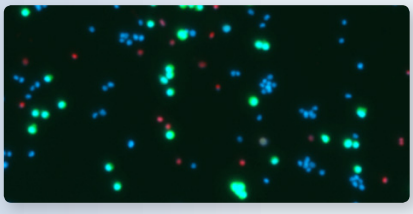
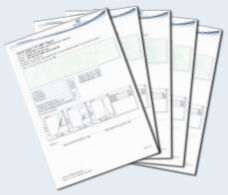


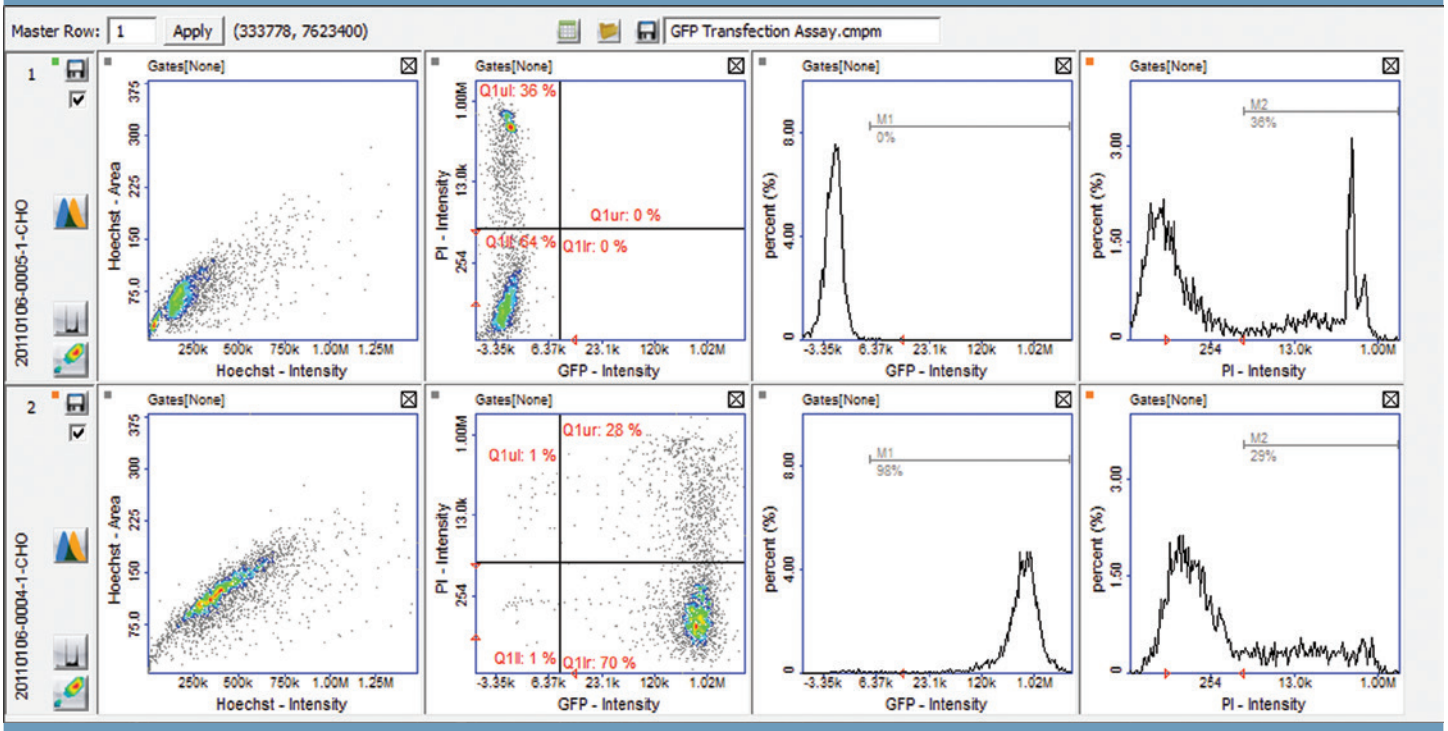
Image acquired with the NucleoCounter® NC-3000™ for the GFP Transfection Efficiency Assay



Automated PDF reports

The NucleoCounter® NC-3000™
- Next generation cell analysis

Results: Presented in PlotManager



CHO cells were transfected with, respectively, a GFP harbouring plasmid (lower row) and a plasmid without GFP (upper row). 24 hours after transfection cells were stained with Hoechst-33342 and Propidium Iodide (PI) and analysed using the GFP Transfection Assay – Hoechst PI and a NucleoCounter® NC-3000™. Scatter plots and histograms were obtained from the NucleoView™ NC-3000 software. Quadrants and markers in the displayed plots were used to demarcate the various cell populations.

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For more information, please visit **www.chemometec.com/NC-3000**

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