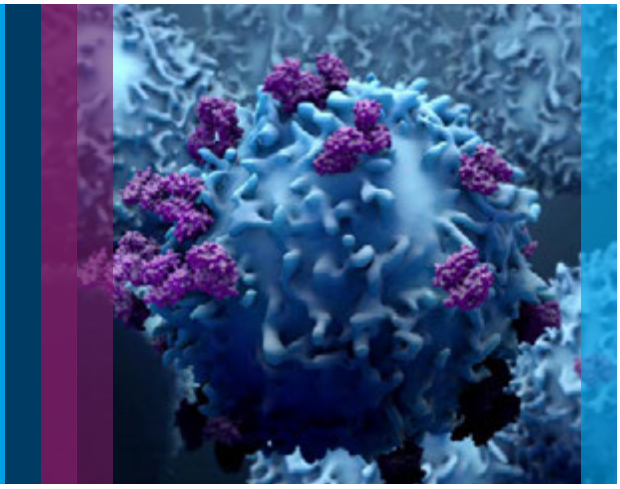
QIFIKIT® for Quantitative Flow Cytometry

Unlock the full potential of your cellular phenotyping analysis



Why Quantitative Flow Cytometry?

Surface antigens are useful for identifying cell types and functions, benefiting early-stage research and pharmaceutical product development. Flow cytometry allows simultaneous analysis of multiple antigens and cells. However, traditional methods are labor-intensive and provide limited information about antigen presence or absence on cell membranes. The advancement of cell and gene therapy highlights the need to quantify antigens per cell for a comprehensive understanding of cellular phenotyping. Quantitative Flow Cytometry Measurements (QFCM)[1] have become important for precise, conclusive, and rapid analyses in the biopharmaceutical industry.

Standardization with Indirect Immunofluorescence Staining

Standardization and reproducibility are crucial for labeling and quantifying targets when characterizing and quantifying surface proteins. Using an indirect staining protocol, the QIFIKIT® delivers a consistent and robust assay by employing a standardized fluorochrome (FITC) conjugated secondary antibody $F(ab')_2$ fragment.

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QIFIKIT® in research applications

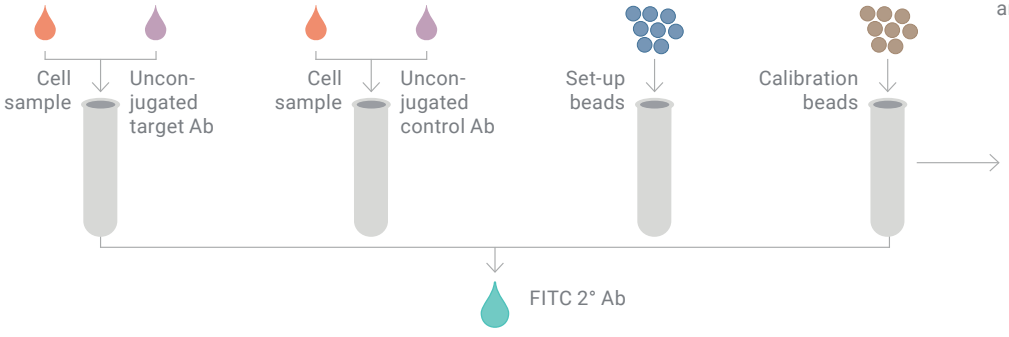
- Cellular biology, biotherapeutics, and immunotherapy development
- Hematology for identification of hematopoietic cell populations
- Infections, looking at the expression of virus receptors
- Immunophenotyping, oncogene products, steroid receptors, drug receptors

Sample and control labeling | **Immunoflurorescence staining** | **Flow cytometric analysis**

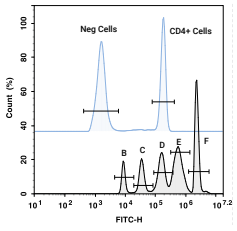
Sample
Label cells with primary mouse mAb against antigen of interest

Control
Label cells with irrelevant mouse mAb

Sample, control, set-up beads and calibration beads
Label, in parallel, with conjugated secondary anti-mouse Ab labelled with FITC



1. Establish window of analysis (set-up beads)
2. Build calibration curve MFI against ABC (calibration beads)
3. Analyze cells and calculate ABC (sample and control)



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Precision and Efficiency with QIFIKIT®

Intended use

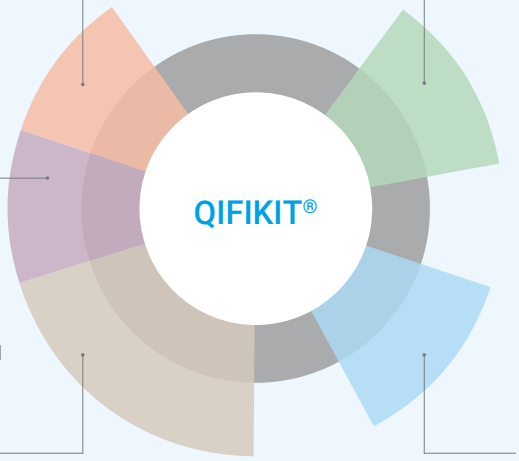
- Quantitative determination of antibody-binding and antigen density per cell using indirect immunofluorescence staining
- Applicable to any cell line, isolated cells, clones, and whole blood samples

Capacity

- 10 calibrations using calibration beads stained with FITC conjugate
- 80 samples stained with FITC conjugate
- Dynamic range: 2,000 - 800,000 sites per cell

Components

- **Set-up beads: (1 mL)**
  Two populations of beads (10 µm in diameter)
  - Blank beads (A) and
  - Beads with a high number of mouse mAb molecules
- **Calibration beads: (1 mL)**
  Five populations of beads (B, C, D, E, and F) bearing different numbers of mouse mAb IgG molecules
- **FITC conjugate: (0.2 mL)**
  $F(ab')_2$ fragment of FITC-conjugated goat anti-mouse immunoglobulins (affinity isolated)



Enhanced flexibility

- Any primary unlabeled mouse mAb of IgG isotype can be used
- Standardized $F(ab')_2$ fragment labeled with FITC for detection, removing the need for custom labeling and reducing variability in Ab : FI Ratio
- Flexible options for instrument setup and calibration

Standardized quantification

- Values based on absolute quantities
- Baseline antigen expression levels as standard reference
- Comparisons between different laboratories and time periods
- Quantifiable definitions of "positive" vs "negative" and "bright" vs "dim" cells

Scan the code for online ordering and product information

Ordering information

Product name	Part Number
QIFIKIT®	K007811-8

References

1. H.M. Shapiro, Practical Flow Cytometry, Wiley-Liss, New York, 2003.

For more information, please visit:
www.agilent.com

Please contact us for more details:
rpsupport@agilent.com

This information is subject to change without notice.

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