Technology Offer

Advanced Flow Cytometry System for Precise Image Refocusing and Cellular Mass Analysis

Ref.-No.: 1629-6741-WT

Abstract

This invention introduces a novel method for image refocusing and mass density measurement in imaging flow cytometry. It combines phase imaging with numerical post-processing to overcome common challenges of focus instability and measurement limitations in current systems. By capturing both bright field and optical phase retardation images of cells in microfluidic channels, the system enables accurate, high-throughput quantification of cellular dry mass and mass density. This technological advancement significantly improves the reliability and analytical capabilities of flow cytometry, enhancing its utility in cellular diagnostics, disease monitoring, and pharmaceutical screening. The integration of numerical focusing techniques compensates for focal shifts caused by sample movement or optical drift, thereby providing consistently sharp and analyzable images for complex cellular assessments.

Background

Conventional imaging flow cytometry enables high-throughput imaging of cells in flow but suffers from critical limitations. Focus stability is difficult to maintain due to mechanical vibration, thermal expansion, and the inherent axial distribution of cells in microfluidic channels. As a result, many images are acquired out of focus, leading to inaccuracies in morphological analysis and reliance on operator expertise. Moreover, current systems typically assess only external features such as shape and brightness, lacking the capacity to measure intrinsic physical parameters like dry mass and mass density. The need for a more robust and informative cytometry platform has become urgent, particularly for applications in diagnostics and drug development where subtle cellular differences must be precisely identified.

Technology

The presented invention combines optical phase imaging and numerical processing to enhance imaging flow cytometry with high-resolution refocusing and biophysical cell analysis capabilities. The core of the system is an imaging setup that captures both bright field and phase retardation images of single cells as they flow through a microfluidic channel. Phase images are acquired using interferometric or non-interferometric holography and encode the optical path length differences caused by variations in cellular refractive index. These variations are directly linked to the dry mass and mass density of each cell, which are quantitatively derived using established physical models.

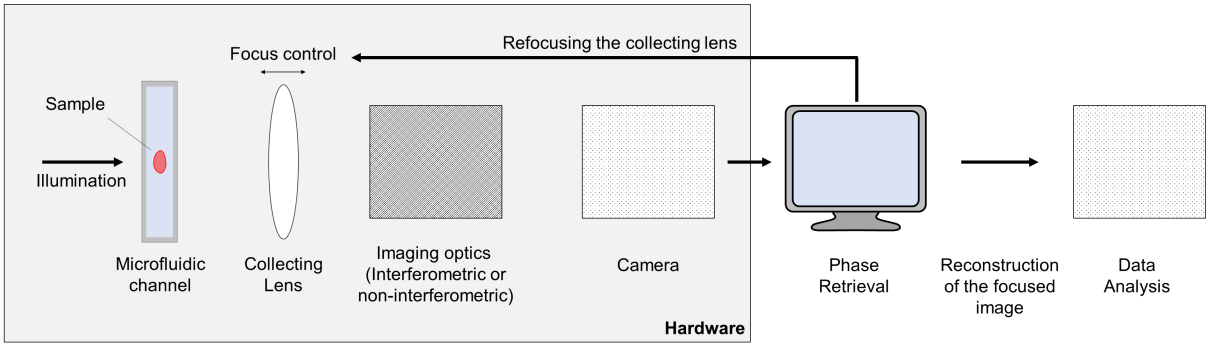

Figure 1: Schematic of the present invention. The work-flow of the present invention. The phase images of samples flowing in a microfluidic channel are measured by an optical setup (hardware) employing either interferometric or non-interferometric microscopy, and the retrieved phase images are used to refocus the hardware or reconstruct images with correct optical focus for data analysis.

Max-Planck-Innovation GmbH
Arnulfstr. 58
80335 München
www.max-planck-innovation.de

Dry mass is computed from the integrated optical phase shift, while mass density is determined by correlating refractive index with measured cell volume and area. Importantly, the system supports numerical refocusing using methods such as Rayleigh-Sommerfeld back-propagation or machine learning algorithms. These techniques reconstruct images at the optimal focal plane, compensating for axial distribution and focal drift during acquisition. This process greatly reduces errors due to mechanical instability or uneven sample flow.

The combined optical and computational platform not only enhances morphological feature extraction (e.g., shape, deformation) but also reveals intrinsic biophysical parameters that are otherwise inaccessible. This dual-level analysis significantly improves the accuracy and diagnostic value of flow cytometry for biomedical and pharmaceutical applications.

Advantages

- **Direct Measurement of Cell Mass and Density**: Extracts dry mass and mass density from phase images, providing deeper insight into cellular composition than morphology alone.
- **Numerical Refocusing for Image Accuracy**: Corrects for defocused images due to axial drift or flow irregularities, improving image reliability without requiring mechanical autofocus.
- **Enhanced Cell Differentiation**: Identifies differences between cell populations that appear morphologically similar but differ in internal properties - crucial for accurate diagnosis or classification.
- **Scalable to High-Throughput Systems**: Works efficiently in microfluidic environments and is compatible with existing flow cytometry systems, enabling large-scale analysis.
- **Automation-Friendly and Reproducible**: Reduces operator variability and minimizes manual intervention, allowing for consistent and automated data acquisition.

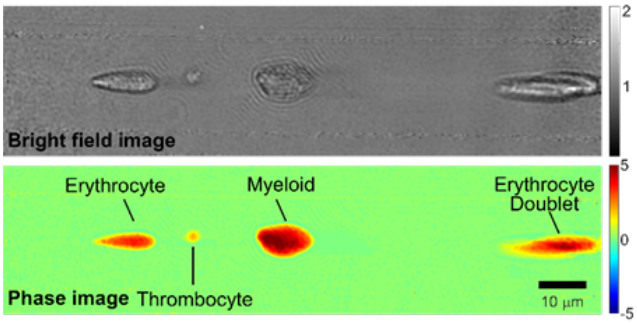

Figure 2: The bright field and phase images of blood cells flowing in a microfluidic channel.

Potential applications

- **Clinical Diagnostics**: Detects subtle differences in cell mass and density between healthy and diseased cells, useful in oncology, hematology, and infectious diseases.
- **Drug Development and Screening**: Quantifies how therapeutic compounds affect cell physiology at the single-cell level, enhancing pharmacological profiling.
- **Immune Cell Analysis**: Differentiates immune subtypes based on combined physical and morphological data, improving immunophenotyping accuracy.
- **Cell Biology Research**: Monitors dynamic changes during cell growth, stress, or differentiation, aiding studies in stem cells and developmental biology.
- **Cell Manufacturing and QA**: Verifies consistency in cultured cells for therapies (e.g., CAR-T cells), supporting quality control in biomanufacturing.

Contact

PD Dr. Wolfgang Tröger Senior Patent- & License Manager, Physicist
Phone: +49 (0)89 / 29 09 19 – 27
eMail: troeger@max-planck-innovation.de

Max-Planck-Innovation GmbH
Arnulfstr. 58
80335 München
www.max-planck-innovation.de