Christian Doppler Laboratory for Cardiac and Thoracic Diagnosis & Regeneration

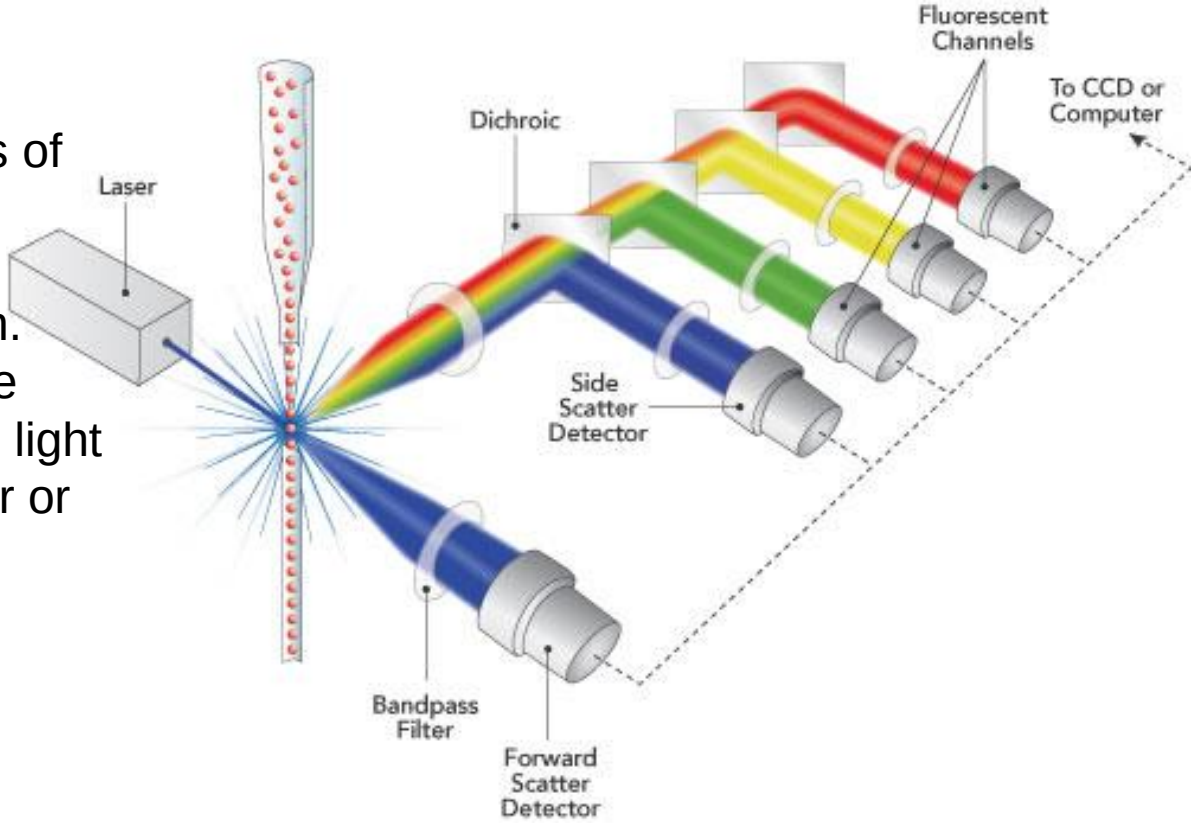
MEDIZINISCHE UNIVERSITÄT WIEN

Fluorescent Activated Cell Sorting (FACS)

November 2013

Flow Cytometry

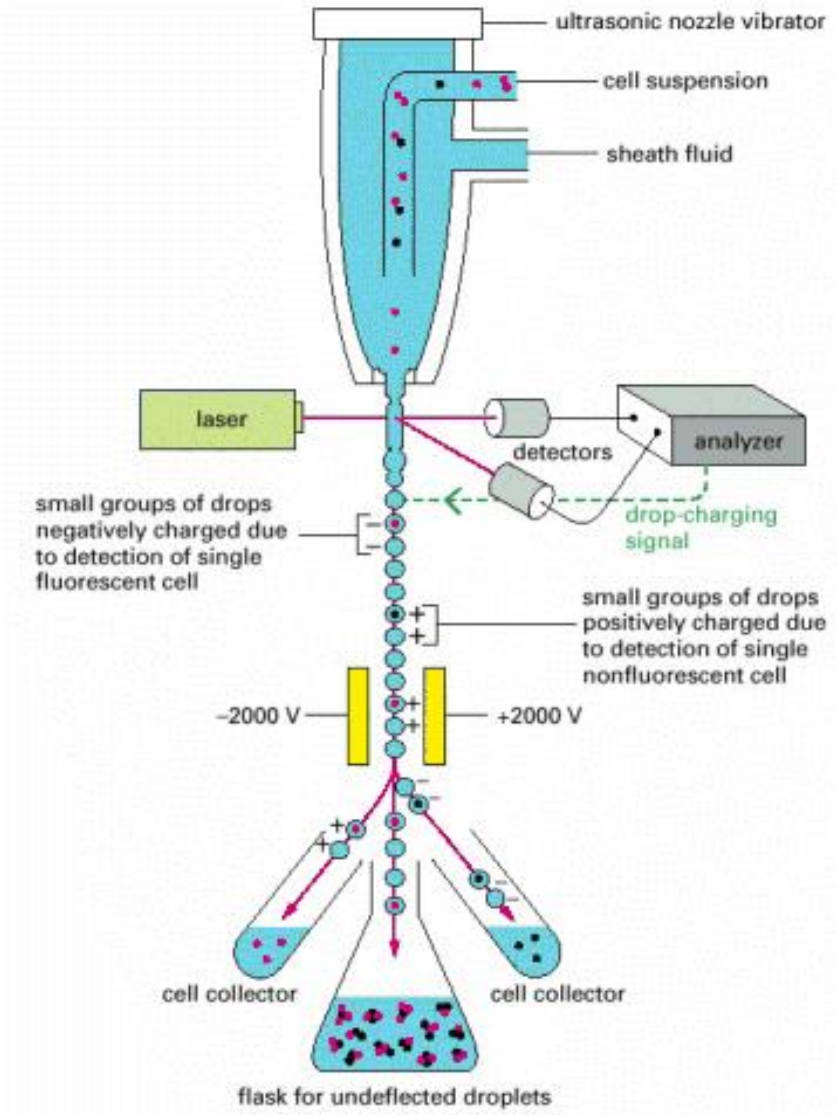
- Technique for the analysis of multiple parameters of individual cells within a heterogeneous population.
- Characteristics of cells are analyzed by the scattered light (Forward and Side Scatter or Fluorescent light)



FACS-Definition

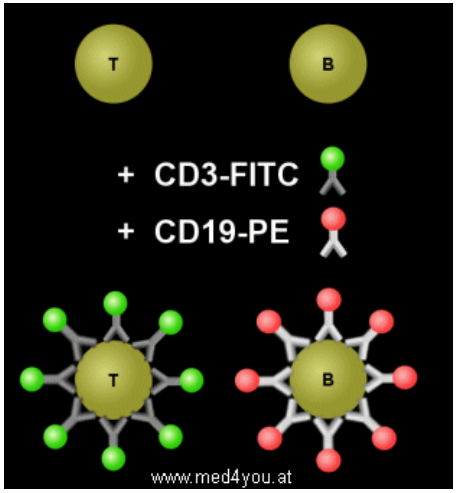
- FACS is a trademarked name by *Becton and Dickinson*, but is commonly used as a term for flow cytometry
- FACS is a special type of flow cytometry
- It provides a method for sorting a heterogeneous mixture of biological cells into two or more containers, one cell at a time, based upon the specific light scattering and fluorescent characteristics of each cell.
- Uses ultrasonic nozzle vibrator to create single cell droplets

FACS - Principle



Protocol (Whole Blood – extracellular staining)

- Collect blood sample
- Staining:
  - mix 90µl whole blood and 10µl antibodies(e.g. CD19-PE and CD3-FITC) and incubate for 20 minutes on ice
  - add 900µl (1:10) with Aqua bidest diluted Erylyse and incubate for 10 minutes
  - Centrifuge cells and remove supernatant
  - Add 400µl FACS-Buffer and vortex
- Analyse with FACS



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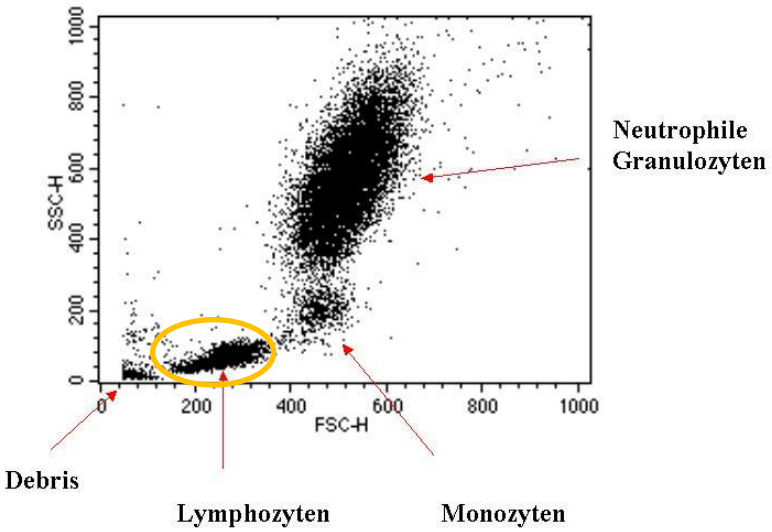
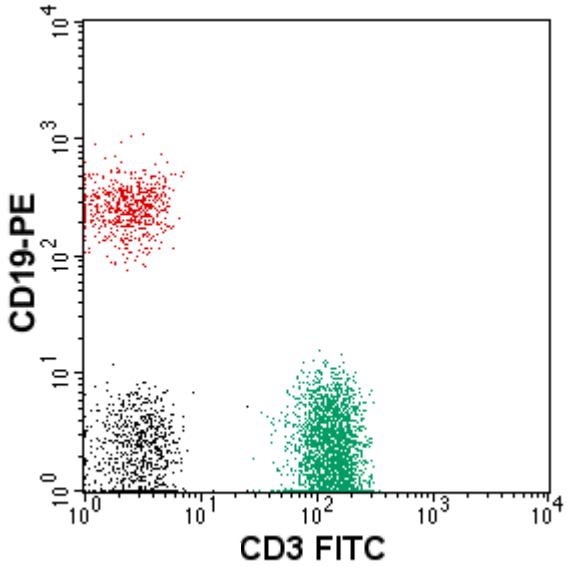
Data Analysis





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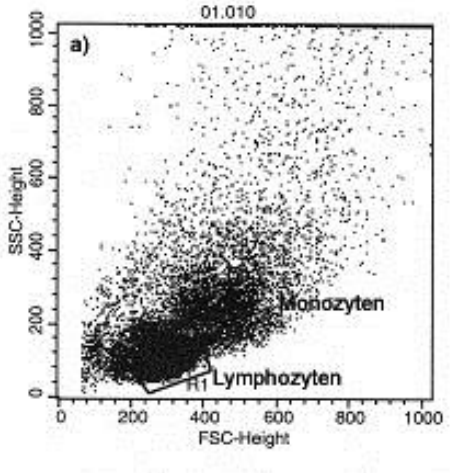

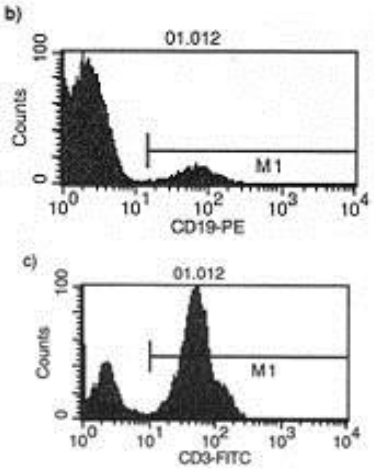

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