

Path Bulletin

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FACS (FLUORESCENCE ACTIVATED CELL SORTING)

Flow cytometry is a technique for counting, examining, and sorting microscopic objects (cellular level objects mainly) suspended in a fluid based on their optical properties (scattering and fluorescence).

Why FACS?

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- FACS is a technique to purify specilc cell populations based on phenotypes detected by flow cytometry.
- FACS is used as a cell sorter and enriched for a subset of cells which is often then studied in further detail using flow cytometry or other analytical techniques.

It is used for cell analysis and is focused on measuring protein expression or co-expression within a mixed population of cells.

Apart from that the main advantages of the process includes-



Highly precise data. No photobleaching. Single cell analysis. Cell sorting in complete separate tubes. Non-biased nonmanipulated computer generated dot plot & histogram data

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Working Postulate:

Flow cytometer measures the physical and chemical characteristics of a cell. A flow cytometer is made up of three main systems: fluidics, optics, and electronics. Fluidics system transports cells to the laser beam for interrogation. The optics system consists of lasers to illuminate the cells in the sample (tagged with specific dye) stream and optical fibers to direct the resulting light signals to the appropriate detectors. The electronics system converts the detected light signals into electronic signals that can be processed by the computer.

Practical Implication!

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Fluorescence Activated Cell Sorting (FACS) is a major application of flow cytometry by which physical separation of cells of interest from a heterogeneous population is done. When a charged droplet passes through a high voltage electrostatic field, between the deflection plates, it is deflected and collected into the corresponding collection tubes. The deflection of the droplet is towards the oppositely charged plate, so that this droplet is separated from unchanged and oppositely charged droplets. Flow cytometric data can be represented as histograms or as dot plots automatically without any biasness. The traditional cytometers are the common cytometer using sheath fluid for focusing the sample stream. The most common lasers used in traditional flow cytometers are 488 nm (blue), 405nm (violet), 532nm (green), 552 nm (green), 561 nm (green-yellow), 640 nm (red) and 355 nm (ultraviolet). Some of the dyes used for sample tagging includes FITC, CD22, PE, BCMA etc.

UNBIASED AUTO ANALYSIS CHARTS



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