Integrated Cell Analysis Devices

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Introduction

The increased interest of biotechnologists to learn more about the functioning of cells has resulted in various research projects where microdevices and some times nanodevices are investigated to yield information from cells. The miniaturization of the analysis chambers and the integration of microsensors and -actuators allow new measurement techniques and new insights.

Two developments are visible; the analysis of single cells suspended in a buffer fluid, and the analysis of cell cultures that are, however, much smaller than the classical culture which contains millions of cells. Small cell cultures roughly contain less than one million cells and are grown on a sensor chip. The advantages of studying single cells and small cultures are technical; the origin and the initiation of diseases for example can be better examined. Also costs can play a significant role because of the small amount of cells and chemicals needed. For several applications it is investigated if physical and electronic analysis methods can be used to study cells and cell behavior. Optical, electrical, magnetic, and thermal properties are examples of physical effects that are being used.

Microfluidics has become an important field in on-chip analysis devices for the control of gases and liquids that contain the cells or chemicals to treat or nurture the cells.

Microfluidics for Single Cell Analysis

For on-chip manipulation and handling of particles and cells microfluidic elements can be designed. In passive hydrodynamic focusing, a channel is being narrowed down which causes a sample flow that is surrounded by a carrier flow to narrow down accordingly. In order to control the height and width of the sample flow, a non-coaxial fluidics chip was developed where the ratio of the flow rates of carrier flow and sample flow determines the height of the sample flow. By using fluidic sideports through which carrier flow can be injected or withdrawn from the main flow, the width of the sample flow can be adjusted and controlled. The position and size of the sample flow can thus be controlled allowing to steer the suspended particles or cells to certain locations for further handling, for analysis (sensors) and for sorting. Further, the chance of clogging is dramatically reduced [1]. An example where we benefit from this device is the optical projection cytometer where cells can flow very closely over integrated optical sensors so that near-field projection optics can be applied. A second example is an integrated Coulter counter where sensitivity can be greatly enlarged by controlling the diameter of the conductive sample flow containing the particles surrounded by a non-coaxial non-conductive carrier flow. The limits of further downscaling these (non-)coaxial fluidic devices have been investigated in [2].

Sensing

As mentioned in the introduction, cell parameters which are of interest to determine are identity, size, shape, quantity, viability, biochemical activity, and electrical activity. Outside electrochemical detection systems, the sensing systems applied for cell detection and characterization are mostly based on optical and electrical (impedance) principles.

The measurement of electrical impedance of cells also yields information on type, viability, membrane and cytoplasm. An example of a flow-through impedance characterization setup can be found in [3]. At lower frequencies, information on the cell membrane can be obtained, at higher frequencies, the conductivity of the cytoplasm can be determined. Instead of a setup where the cell flows between two electrodes, a Coulter counter can be used to determine the electrical impedance of particles. By applying conductive sample fluid and non-conductive carrier fluid in the non-coaxial sample sheath flow chip described in the previous section, the impedance of the sample can be monitored. When a particle or cell flows through, the impedance will change. For solid particles, successful measurements have been conducted [4]. For cells this has been demonstrated in [5].

Optical cytometry is one of the standard methods for cell characterization [6]. In a typical system scattered and reflected light – often fluorescent – is measured which gives information about cell size, viability and cell type. In a new approach an on-chip near-field optical projection cytometer has been developed. The shadow of a particle that flows through the near field of a strip photodiode is being recorded. The non-coaxial microfluidics setup described in the previous section is being applied to get the particles or cells in the near field of the detector, reducing interference and thus allowing the recording of a projection (shadow). Measurements with transparent and non-transparent particles showed the potential of the system to determine optical properties of particles and cells. (Semi)optical particles act as a lens and instead of a decrease in photocurrent output, an increase can be detected, depending on size and transparency of the particle. The use of this effect for identification of different cells has been shown for yeast and CHO cells [7]. The measurement response for those cells also shows a positive peak, which indicates that the light is indeed focused by the cell increasing the sensor signal – the cell acts like an optical lens.

Sorting

For selection and sorting of particles and cells, different principles have been applied. An important method is dielectrophoresis, where forces can be applied to cells by electric polarization. By switching electric fields in the microchannel single particles can be selected (e.g. [1]). This can be a very useful tool to accumulate particles of specific interest after analysis in a flow-through system. Due to the more complex structure of cells (wall, membrane, cytoplasm) the electric polarization is frequency dependent and so is the DEP force (can be positive of negative). This effect can be used to determine viability [8] or to separate cells from mixtures [9].

Conclusion

Different techniques for monitoring and analysis of single cells or small cultures are being investigated successfully, such as on-chip culturing, flow-cytometry and array analysis. In addition to electrochemical methods, physical detection methods are currently being developed, yielding extra information on cell properties (e.g. optical and mechanical) and cell behavior.

Acknowledgements

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