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Radio Frequency Electrical Impedance Spectroscopy of Chinese Hamster Ovary Cells

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Measuring the frequency-dependent electrical impedance of biological cells, electrical impedance spectroscopy (EIS), has shown promising potential in differentiating cells of different types or at different states. The label-free and non-invasive technique is integratable with microfluidics enabling rapid single-cell analysis. Example applications of EIS incorporated into microfluidics include analysis of sub-populations of leukocytes, differentiation of diseased red blood cells from healthy ones, and stem cell differentiation studies [1], [2]. In this paper we investigate the application of radio frequency EIS for differentiating viable and apoptotic Chinese hamster ovary (CHO) cells in pharmaceutical bioreactors.

In single-cell electrical impedance spectroscopy, the multi-frequency impedance of individual cells is measured in a microfluidic channel as they flow between/over two sets of parallel-facing/coplanar electrodes embedded in the channel. In the β-dispersion frequency region and under the assumption that the cell's volume is small compared to the electrodes measurement volume (small volume fraction), the measured differential impedance (with one set of electrodes as reference) is related to the complex permittivity of the cell, $\tilde{\varepsilon}_{cell}$, as [3]

$$\Delta Z \approx \frac{\kappa}{j\omega\tilde{\varepsilon}_e}(-3\varphi K_{CM}(\omega)) \ , \ K_{CM} = \frac{\tilde{\varepsilon}_{cell}-\tilde{\varepsilon}_e}{\tilde{\varepsilon}_{cell}+2\tilde{\varepsilon}_e}. \quad (1)$$

Here $\tilde{\varepsilon}_e = \varepsilon_e - j\,\sigma_e/\omega$ is the complex permittivity of the external medium with ω the angular frequency, φ is the volume fraction, K_{CM} is the Clausius-Mossotti factor, and κ is the electrodes geometrical constant. $\tilde{\varepsilon}_{cell}$ is related to the dielectric properties of the cell's compartments through the single or double shell models [4].

We employed the double-shell model for CHO cells, with parameters reported in literature [4], and performed sensitivity analysis to determine the optimum measurement frequencies and external medium conductivity for differentiating viable and apoptotic CHO cells. We employed Ampha Z32 Impedance Flow Cytometer (Amphasys, Switzerland) to measure changes in the impedance amplitude and phase of CHO cells over the frequency range 0.3 to 30 MHz as they gradually became apoptotic under nutrient deprivation condition. Amphasys Biochips with channel cross-section 30 µm × 30 µm and parallel-facing electrodes at the top and bottom of the microfluidic channel were used. Approximately 12,000 individual cells were measured in the beginning and after 48 hours of starvation. The measured impedance at higher frequencies demonstrated one population of viable cells at time 0 and two distinct populations of viable and apoptotic cells after 48 hours of nutrient deprivation. The result is consistent with available reports in the literature using a different dielectric measurement technique, dielectrophoresis [5], [6]. Compared with dielectrophoresis, EIS has the advantage of multi-frequency measurement at the same time which provides broader information about the cells physiological states. The method can be employed in biopharmaceutical industry for continues bioprocess monitoring.

References

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