Recent Biomedical Applications of Dielectrophoresis

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Summary:
Examples are given of the application of dielectrophoresis for the isolation of single target cells, the manipulation of RNA at a sensor electrode, and the discrimination and selective separation of stem cells.

Introduction:
Dielectrophoresis (DEP) is a biomarker-free technique that utilizes the induced dynamic response of a particle to a non-uniform electric field. Recent reviews of DEP theory, technology and applications are available[1-3]. For a particular field frequency the DEP response of a particle depends on whether its dielectric polarizability is greater or less than the surrounding medium, with the particle moving up a field gradient towards an electrode (positive DEP) or down a field gradient away from an electrode (negative DEP), respectively. The results described here all concern aspects of this behavior.

DEP Pin Electrode for Single Cell Isolation:
The selection, isolation and accurate positioning of single cells in three dimensions are increasingly desirable operations in many areas of biological research and tissue engineering. Fig.1 shows the application of a simple and low cost DEP device for picking out and relocating target cells. It consists of a single electrode probe capable of selecting between live and dead cells[4].

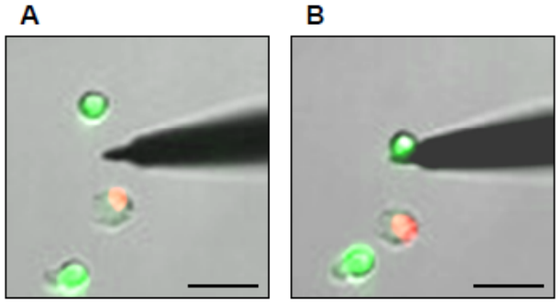

Figure 1: (A) CHO cells labeled with calcein-AM (green) and ethidium homodimer-1 (red), identifying living and dead cells, respectively. No voltage applied. (B) A 10 MHz signal attracts a viable cell and simultaneously repels the dead one[4]. Scale bar is 50 µm.

DEP Manipulation of Ribosomal RNA:
Fig. 2 shows the first demonstration of DEP for fast capture and release of rRNA at a sensor electrode[5].

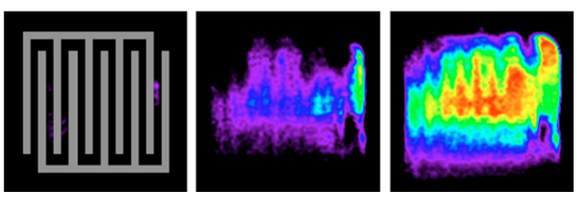

Figure 2: Total internal reflection fluorescence (TIRF) microscopy images of the DEP collection of rRNA at interdigitated electrodes (taken 4s, 10s and 30s after application of a 4 $V_{p\text{-}p}$, 3 MHz signal)[5]. The RNA was released by negative DEP with a 50 MHz signal.

This result opens up new opportunities for rRNA-based biosensors because the high copy numbers rRNA molecules present in individual cells can be used as a naturally amplified biomarker for the detection of bacteria.

DEP Sorting and Discrimination of Stem Cells:
Human embryonic stem cells (hESCs) isolated from early blastocyst stage embryos constitute a promising resource for disease modeling, drug screening and cell therapies for regenerative medicine. This is founded on their immortality and pluripotency, as compared to the restricted growth potential and repertoires of adult tissue-sourced stem cells. However, the breadth of this capacity also makes controlling cell behavior *in vitro* more difficult, potentially undermining their utility due to variable and heterogeneous cell production and the difficulty of distinguishing cells of a desired phenotype from other contaminating populations. There is a need for sensitive and non-invasive methods to discriminate and segregate live cell populations.

To achieve efficient DEP separation of stem cells from their differentiated progeny there must be no or little overlap of their distributions of DEP cross-over frequencies (the frequency marking the transition between negative and positive DEP). Fig.3 demonstrates that this requirement clearly holds for H9 hESCs (first derived at Wisconsin Regional Primate Research Center and licensed from WiCell) and those differentiated to a mesenchymal stem cell like phenotype (H9-MSC).

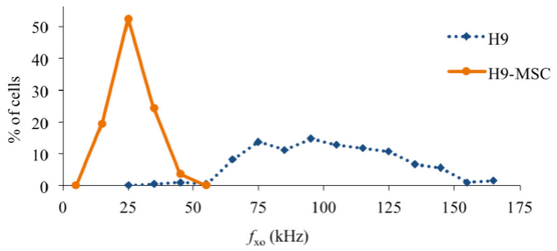

Figure 3: Percentage distributions of the DEP cross-over frequencies (f_{xo}) for H9 and differentiated H9-MSC.

Myoblasts are muscle derived mesenchymal stem cell progenitors that have great potential for use in regenerative medicine. DEP has been shown capable of discriminating cells between stages of differentiation in the C2C12 myoblast multipotent mouse model. Terminally differentiated myotubes were separated from C2C12 myoblasts to better than 96% purity, a result validated by flow cytometry and Western blotting[7].

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References:
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