DIELECTROPHORETIC CHARACTERIZATION OF *CANDIDA* SPECIES WITH 3D CARBON ELECTRODES TOWARDS SEPARATION AND SPECIES-SPECIFIC DIAGNOSIS

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ABSTRACT

Here, we present preliminary results towards characterization of dielectrophoretic spectra of different *Candida* strains. 3D carbon electrodes were used to induce positive dielectrophoresis (pDEP) force on cells. We have characterized the pDEP response of *C. albicans*, *C. tropicalis* and *C. parapsilosis*. For all cells, stronger DEP response was observed between 50 kHz to 500 kHz. At 750 kHz, only *C. tropicalis* were trapped, whereas results below 10 kHz suggests only *C. albicans* trapping. Future work includes species-specific *Candida* separation via DEP.

KEYWORDS: Dielectrophoresis (DEP), *Candida*, Cell separation, Carbon electrodes

INTRODUCTION

Candidiasis (*Candida* infection) is the fourth most common cause of morbidity and mortality in hospital patients in the United States [1]. Timely detection of candidiasis is an urgent need in the clinical setting [2]. Species-specific detection is necessary as candidiasis caused by differing *Candida* strains may require different treatment strategies. For example, *C. glabrata* is resistant to commonly used –azole-based antifungals, therefore, initial treatment of candidiasis via *C. glabrata* with –azoles may present an increased risk of infection and death [3]. DEP separation of *Candida* species will reduce diagnosis time, improper treatment strategies, and fatalities. Several groups demonstrated DEP of *C. albicans* only for cell trapping [4], [5], while other strategies (*i.e.* imaging, PCR, or Raman scattering) were used for separation. However, there is no report of the DEP manipulation of emerging pathogens such as *C. tropicalis* and *C. parapsilosis* or separation of *Candida* by species via DEP. The DEP response of *C. albicans, C. glabrata, C. tropicalis, and C. parapsilosis* is determined here. This work is a fundamental step in enabling DEP for rapid separation of *Candida* species.

EXPERIMENTAL

Different strains of *Candida* were re-suspended in a DEP buffer media having conductivity of 12.62 µS/cm with a cell concentration in the order of 10^7 cells/ml. Experimental protocol was 1) flowing 52 µl of sample through a 3D carbon electrode array (see Ref. [6]) polarized by sinusoidal signal having a amplitude of 20 $V_{peak\text{-}peak}$ and frequency ranging from 10 kHz to 1 MHz; 2) washing the trapped cells (Fig. 1), if any, with 100 µl of clean buffer; and 3) releasing them by turning the field off. We recorded such release at the end of the electrode arrays and analyzed by ImageJ, an image processing software, to obtain the average intensity of the images.

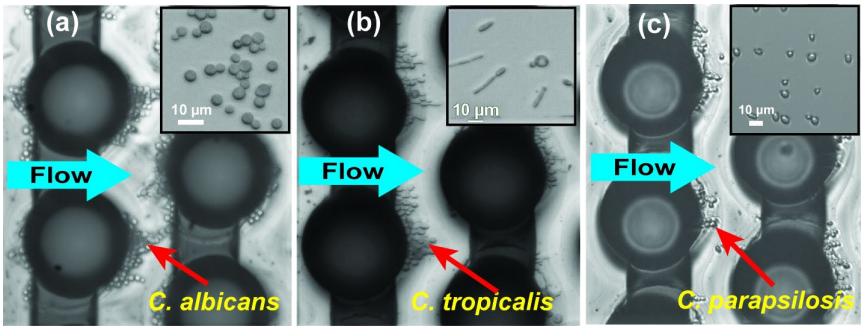

***Figure 1**: (a) Candida albicans, (b) Candida tropicalis, and (c) Candida parapsilosis cells trapped on carbon electrodes (dark circles and connecting vertical lines) by pDEP. Insets show the shape and the morphology of the corresponding cells.*

RESULTS AND DISCUSSION

The intensities obtained from image analysis represents the pDEP response of the cells (Fig. 1) with respect to the sinusoidal signal. The results were plotted in Fig. 2. If no cells were trapped, the intensity would be zero. High value of the intensity represented higher number of trapped cells. The majority of cell trapping focused between 50 kHz and 500 kHz, with significant decreases after 250 kHz. Several potential opportunities for separation emerge from these results. Results at 750 kHz suggest the ability to trap only *C. tropicalis,* potentially due to the shape differences of the strains. *C. albicans* and *C. parapsilosis* possess spherical morphology, whereas *C. tropicalis* exhibits both spherical and tubular morphology (insets of Fig. 1). Additionally, 10 kHz is the only instance where *C. albicans* demonstrates the strongest trapping. Frequencies below 10 kHz may offer isolated trapping of *C. albicans.*

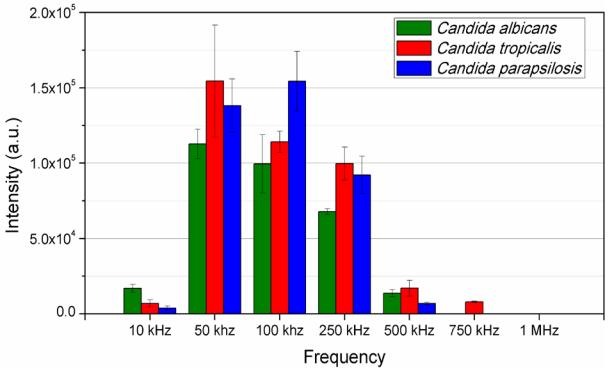

***Figure 2**: Positive DEP response of C. albicans, C. tropicalis and C. parapsilosis. At least 3 experiments were performed for each frequency.*

Ongoing work includes characterizing *C. glabrata,* where a similar result is expected. The immediate goal is to separate these cell types from a heterogeneous *Candida* population. While challenging, we anticipate that modifying the conductivity of the DEP buffer will generate shifts in the DEP behavior (due to variations in the Clausius-Mossotti factor) which may allow for more separation opportunities. We will also be able to capitalize on changes in cell permittivity based on cell morphology, however, several cell types (*i.e. C. glabrata* and *C. parapsilosis)* share similar shape and size.

CONCLUSION

Positive DEP response of *C. albicans*, *C. tropicalis* and *C. parapsilosis* was characterized using 3D carbon electrodes. Although the pDEP response was strong for all the three strains in the frequency range 50 kHz-500 kHz, only *C. tropicalis* was trapped at 750 kHz. This suggests separation of *C. tropicalis* at 750 kHz. Further, frequency below 10 kHz may offer separation of only *C. albicans*.

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