MEASUREMENT OF THE IMAGINARY PART OF THE CLAUSIUS-MOSSOTTI FACTOR

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ABSTRACT

A method is proposed for measuring the imaginary part of the Clausius-Mossotti factor (ICMF) of particles, which is crucial for particle manipulation and characterization using travelling wave dielectrophoresis (twDEP) and electrorotation. The method is based on the force balance of particle motion in a horizontal straight micro channel with an electrode array built on its bottom wall for generating twDEP. The experimental validity was checked using sephadex particles and liposomes. The ICMF spectrums of two lung cancer cells with different metastatic stages were also measured, and their difference indicates that ICMF can be served as a phenotype for cell characterization.

KEYWORDS

Imaginary part of Clausius-Mossotti factor, Cell characterization, Travelling wave dielectrophoresis, Microfluidics.

INTRODUCTION

The imaginary part of the Clausius-Mossotti factor (ICMF), K_i, is a crucial parameter in electrorotation and travelling wave dielectrophoresis (twDEP), which were employed successfully for many particle/cell characterization and manipulation [1] [2]. However, K_i cannot be determined through its dielectric definition for many particles, such as biological cells. The goal of this study is to propose a method and fabricate the associate device to measure K_i, which are absent in the literature.

THEORY

The measurement principle is based on the force balance in a horizontal straight micro channel with an electrode array built on its bottom wall as shown in Fig. 1(a), with gravity along the negative *z*-direction. A travelling wave electric field is generated when the electrodes are powered by ac voltages with 90° phase shift between neighboring electrodes. A particle suspended in a medium in the region above the electrodes is subject to both conventional dielectrophoresis (cDEP) and twDEP forces associated with the applied electric field. The cDEP force alternates its direction at regions above neighboring electrodes, while the twDEP is unidirectional [3] along the electrode array. For the present two-dimensional electric field in the *xz*-plane, the particle translates essentially with a constant speed (*U*) relative to its surrounding fluid along the *x*-direction for positive K_i (or vice versa if K_i < 0) at a given height under the force balance. The equilibrium settling height, *z**, is determined by the force balance along the *z*-direction, involving the weight, the buoyancy, and the *z*-component of the cDEP force. At such a settling height, the *x*-component of the cDEP force is negligible in comparison with that of the twDEP force, which is balanced by the viscous (Stokes) drag from the surrounding fluid under the low Reynolds number condition, $2\pi\varepsilon_m R^3 K_i E_x^2 \partial\varphi_x / \partial x = 6\pi\mu URC$, where ε_m is the fluid permittivity, μ is the fluid viscosity, *R* is the particle radius, *C* is a factor accounting for the wall effect on viscous drag [4], and E_x and φ_x are the *x*-component magnitude and phase of the electric field, respectively. In the present experiment, ε_m and μ are known for a given fluid, *R* and *U* are measured, and $E_x^2 \partial\varphi_x / \partial x$ can be evaluated if both the electric field and the location of the particle center are given. Here we determine the position of the particle center using a microscope. In particular, we measure *z** through the differences of the scales of the focus screw on the microscope when it is focused at the elevated particle and at the bottom wall of the chamber, respectively. Two ways were employed for calculating the electric field quantities. One use the numerical solution of the complex potential of Fig. 1(a) using the method of [5] with the aid of COMSOL software; and the other applies the theoretical result of Morgan et al. [6], who provided analytical expressions for the electric field and the DEP force for $z > 3\lambda/8 = 3\,s$ (see Fig. 1(a)). K_i is thus determined through the force balance relationship along the *x*-direction.

EXPERIMENT

The test section of the device (Fig. 1(a)) was employed in [3] as the central unit of a twDEP pump. Obstacles were built on both sides of the test section for flow stabilization (Fig. 1(b)), and the whole device is shown in Fig. 1(c). The device was fabricated using standard MEMS technique similar to those in [3], including photolithography, wet etching and molding using PDMS. The electrode array was fabricated with gold (150 nm thickness) on a glass substrate. A chrome layer (30 nm) was inserted between the gold layer and the glass substrate to improve the adhesion. The top and side walls and the obstacles are molded with PDMS, and bonded to the glass substrate. There are totally 24 electrodes in the array, and are actuated by a four channel functional generator. Several particles in selected solutions were employed for performing experiments. They are sephadex particles (g-25 super fine, GE

Healthcare Life Science), liposomes, CL1-0 or CL1-5 cells in 280 mM mannitol ($C_6H_{14}O_6$) solution with its conductivity adjusted using KCL solution, and CL1-0 and CL1-5 cells in RPMI solution. CL1-0 and CL1-5 are two lung cancer cells, with CL1-5 more invasive. The movement of the particles during the experiment was observed and recorded through a CCD camera mounted on a microscope along the negative *z*-direction of Fig. 1(a), and the image data (thirty frames per second) were stored in a computer and employed for deriving the particle velocity, *U*.

RESULT AND DICSUSSION

Figure 1(d) shows the numerical values of the *x*-component total DEP force (cDEP plus twDEP forces, [7]) at different heights (*z*). It indicates that the cDEP force (which varies around zero along the *x*-axis) dominates at small *z*, while the twDEP force (positive) dominates at large *z*. Such a finding agrees with the analysis of Morgan et al. [6]. The agreement between the total DEP and the twDEP force in Figures 1(e) and 1(f) indicates that cDEP force is negligible when $z > s = 15$ µm. In the present study, the particles are under negative DEP, and move essentially on a horizontal plane at heights between $z^* = 30 - 40$ µm according to both calculation and measurement. Thus the force balance along the *x*-direction involves only the twDEP force and the viscous fluid (Stokes) drag, which provide the basis for measuring K_i, as discussed in the last section.

The method is validated by measuring particles with known K_i. Figure 2(a) and 2(b) show that the measured values of sephadex and liposome particles agree nicely and fairly, respectively, with the theory. The agreement between the experimental values using the numerical and the theoretical electric field indicates that the analytical expressions in [6] can be employed for the present design. Two lung cancer cells, CL1-0 and CL1-5, were also measured in RPMI solution (natural environment) and other medium with different conductivities. The results in Figures 2(c) to 2(f) show that the variations of K_i with frequency are different for different cells, which implies that K_i may be served as phenotypes for different cells with similar origin, and can be employed for cell characterization and separation. Figure 2(e) shows that K_i is negative near 10 MHz, which indicates that the particle might migrate along different directions in a twDEP device, or rotates in opposite directions in an electrorotation chamber, by changing the applied electric frequency if the medium conductivity is properly selected.

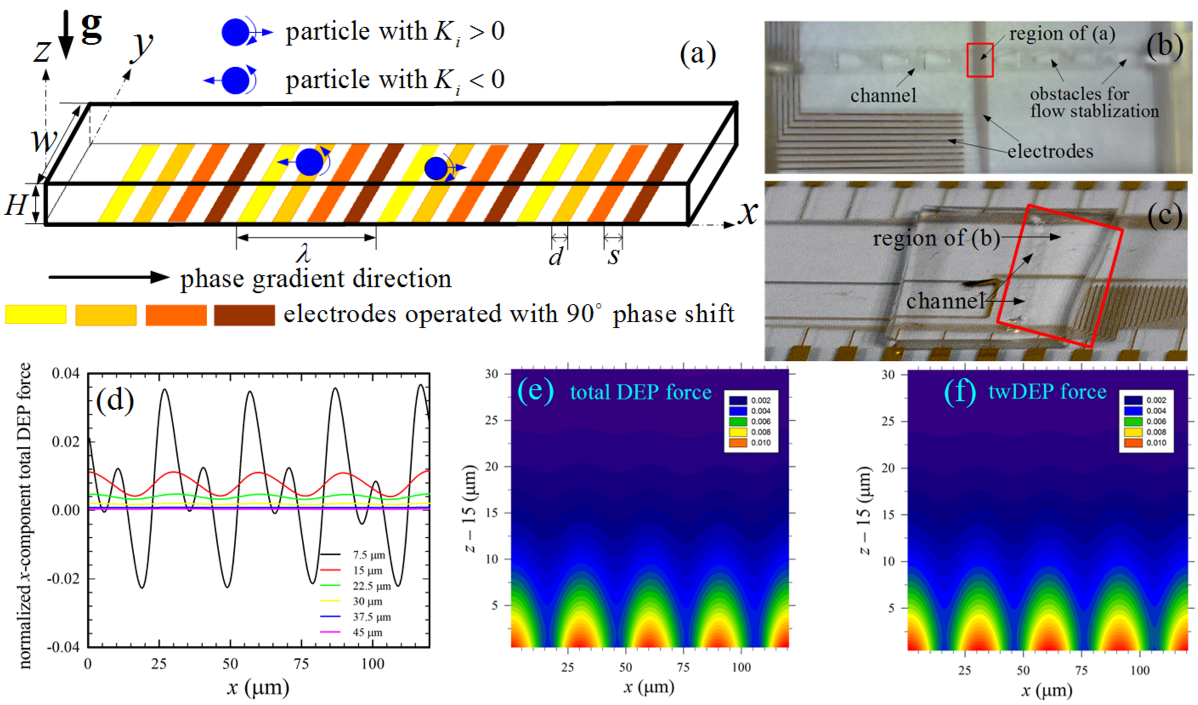

Figure 1: (a) Sketch for the test section of the device. A particle rotates as it translates downward the phase gradient if $K_i > 0$, or vice versa. In the experiment, $d = s = 15$ µm, $H = 107$ µm and $w = 1000$ µm. The electrodes are powered by a 5V peak-to-peak voltage with different frequencies. (b) Enlarged top view of the channel region containing the test section of the device. (c) The device. (d) The normalized *x*-component total DEP force at different heights from half (7.5 µm) to three times (45 µm) of the electrode gap. (e) and (f) The distribution of the normalized *x*-component total DEP and twDEP force, respectively. Numerical results in (d)-(f) are for a case using sephadex particle with conductivity 6.5 mS/m and relative permittivity 40, in a medium with conductivity 0.01 S/m and relative permittivity 78, operated at 5 V (peak-to-peak) and 4 MHz. The force scale for normalization is $2\pi\varepsilon_m R^3 V_0^2 s^{-3}$, with V_0 half the value of the applied peak-to-peak voltage.

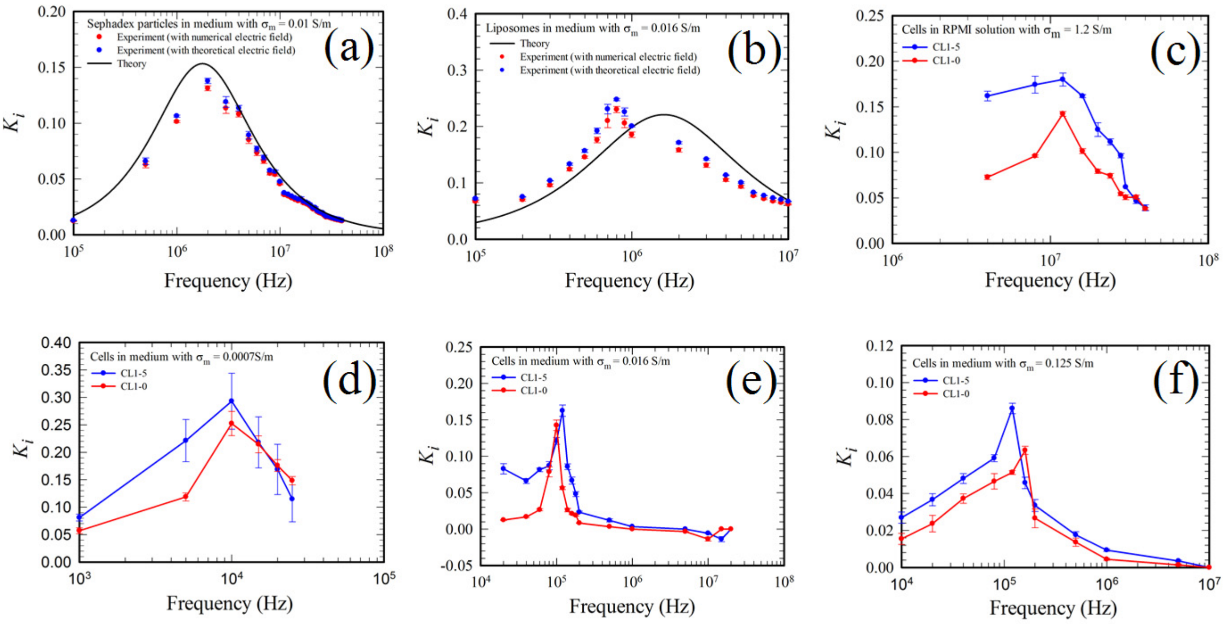

Figure 2: Variations of the imaginary part of the Clausius-Mossotti factor, K_i, with electric frequency for (a) sephadex particles, (b) liposomes (membrane thickness equals 4 nm, conductivity and relative permittivity are 10 μS/m and 2.2 for the membrane and 0.7 mS/m and 78 for the interior material, respectively), and (c)-(f) lung cancer cells in mannitol or RPMI solution with different conductivites (σ_m). CL1-5 cells are more invasive than the CL1-1 cells.

CONCLUSION

A method is proposed, and the associated device is fabricated and validated, for measuring the imaginary part of the Clausius-Mossotti factors (ICMF) of particles and cells, which is important for particle/cell manipulation and characterization using travelling wave dielectrophoresis and electrorotation. The difference of the experimental spectrums of ICMF for different lung cancer cells of similar origin indicates that the ICMF spectrum could be served as a phenotype for those cells, which may be regarded as an alternative for the bio-markers of many detection methods based on immune-labeling. Such a finding is important because markers are not always available and often recognize cells of similar origin, for example, the multiple members of a stem cell lineage [8].

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ACKNOWLEDGEMENTS

The work is supported partially by the National Science Council through the grant NSC 99-2221-E-002-082-MY3 of Taiwan, Republic of China. The authors also would like to express their gratitude to Professor P. C. Yang of the Department of Internal medicine, National Taiwan University, for providing the CL1-0 and CL1-5 cells for the experiment, and to Professor H. R. Jiang of the Institute of Applied Mechanics, National Taiwan University, for suggesting a way to measure the settling height of the particle.

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