[Full Paper]

Measurement of dielectrophoretic force by employing controllable gravitational force

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SUMMARY

Dielectrophoresis (DEP) is the motion of a matter caused by polarization effects in a non-uniform electric field. Recently, studies on DEP are promoted in various medical fields, including separation and characterization of biological cells. It is very important but not easy to measure this motive force, so-called dielectrophoretic force (DEP force). In general, the DEP force generated by the designed electrodes is analyzed by the computer simulation that employs the finite element analysis method. However, it does not always calculate the correct DEP force. Therefore, we propose the method of measuring the DEP force accurately based on the null method. The experiment is conducted with the dielectrophoretic device (DEP device) in which microfabricated electrodes were formed and which contains the polystyrene particles exhibiting the negative DEP in distilled water in a non-uniform electric field. The displacement of the particle reaches a steady state 15 min. after the change of applied voltage v , angle θ or frequency f . The equilibrium state of the particle in a non-uniform electric field can be reached at any place by adjusting both of angles θ and ϕ , where θ is the “Pitch” angle and ϕ the “Yaw” angle for the device. The proposed method can measure the incredibly minute DEP force ranging from 25 fN (femto-Newton) at $\theta=5^\circ$ to 298 fN at $\theta=90^\circ$, the accuracy of which is determined by the static friction and rolling friction between particles and the inner floor of the DEP device.

Key words: dielectrophoresis, DEP force, null method, controllable gravitational force, adjustment of 2D angle.

INTRODUCTION

Recent interests in the dielectrophoresis (DEP) has spurred a great deal of research into separation and characterization methods of biological cells in medical field. Li and Bashir separated alive and dead *Listeria innocua* cells on the interdigitated microelectrodes by utilizing difference of dielectric properties (positive and negative DEP, under the voltage of 1Vpp at 50kHz, of live and dead cells, respectively)¹. Gascoyne et al. separated malarially-infected erythrocytes from normal erythrocytes and nucleocytes by using a DEP-Field-flow fractionation (FFF)². In order to

develop applications of the dielectrophoretic force (DEP force) in the near future, it is necessary to understand the force. Li et al. discussed the distribution of DEP forces which was calculated by a finite-element approach³. The profile of DEP forces is very helpful to understand the behavior of biological species qualitatively. The profile is determined by conductive and dielectric properties of the particles and the medium in the chamber with electrodes of complicated shape under an appropriate voltage and frequency. The accurate measurement of the DEP forces may be required in potential applications. However, there is no report about the accurate measurement of the DEP forces.

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Abbreviations: DEP, dielectrophoresis; DEP force, dielectrophoretic force; FFF, Field-flow fractionation; AC voltage, alternative current voltage.

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In this paper, we propose the method to measure the DEP force accurately, design and fabricate the dielectrophoretic device (DEP device), and measure the DEP force generated on the polystyrene particles in a non-uniform electric field⁴. For accurate measurement of the DEP force, we employ “the null method (zero method or balance method)”. In the null method, the difference between the amount to be measured and that of reference is adjusted to be zero. If the difference and the reference are measured accurately, the null method guarantees accurate measurement.

In our null method system, the difference and the reference can be measured accurately as follows. The difference between the DEP force and the gravitational force can be accurately measured as immovability of particles. The reference can be measured as the gravitational force modified by the angle of the device to the horizontal. The modified gravitational force can be measured accurately as described in the section 2 of *THEORETICAL BACKGROUND*. Therefore our null method system can be regarded as an accurate measuring system.

In this work, the transient responses and static characteristics of the negative DEP force is measured. The DEP force is generated on the polystyrene particles (9.9 μm^ϕ) by the micro-fabricated electrodes in distilled water. This work suggests the possibility to realize a self-balancing instrument to measure the DEP force automatically.

THEORETICAL BACKGROUND

1. Dielectrophoresis

When the dielectric properties of particles are different from those of medium surrounding them, the motion of the particles (charged or neutral) in a non-uniform AC electric field, is termed dielectrophoresis by Pohl^{5, 6}.

The DEP force (F_{DEP}) generated on the particles in a non-uniform AC electric field (see Fig. 1) is given by the following equation^{6, 7}.

$$F_{DEP} = 2\pi\epsilon_0\epsilon_m r^3 \text{Re}(f_{CM}) V E_{rms}^2 \quad (1)$$

Fig. 1. Principle of dielectrophoresis in the case of $\epsilon_p < \epsilon_m$.

ϵ_p : relative permittivity of particles.
 ϵ_m : relative permittivity of medium.

where ϵ_0 is the permittivity of vacuum, ϵ_m the relative permittivity of medium, r the particle radius, E_{rms} the root-mean-square value of electric field. And the factor f_{CM} is so-called Clausius-Mosotti factor which is derived from the well-known Clausius-Mosotti relation, first derived by Mosotti⁸ in 1850 and independently by Clausius⁹ in 1879.

$$f_{CM} = \frac{\epsilon_p^* - \epsilon_m^*}{\epsilon_p^* + 2\epsilon_m^*} \quad (2)$$

where ϵ_p^* and ϵ_m^* are the relative complex permittivities of the particles and the medium, respectively. Both of relative complex permittivities are written by $\epsilon^* = \epsilon + \sigma/(j\omega)$, where ϵ and σ are the permittivity and the conductivity of the particles or the medium, and j is $\sqrt{-1}$. Eqs. (1) and (2) imply that the strength and the direction of the DEP force depend upon the magnitude relation between ϵ_p^* and ϵ_m^* in a non-uniform electric field. For instance, when a particle is more polarizable than its surrounding medium, the particle is pulled towards higher field regions and it is termed positive dielectrophoresis. The opposite phenomenon is negative dielectrophoresis such as the DEP force of the polystyrene particles in distilled water.

2. Accurate measurement of the DEP force by the null method

In the inclined floor environment where the DEP force F_{DEP} is upward along the floor inclined at a degree of θ , the parallel component of gravitational force to the floor $F_G(\theta)$ acts oppositely to the DEP force as shown in Fig. 2. The normal component of gravitational force $F_{GN}(\theta)$ causes as static friction or dynamic friction depending upon whether the particle is standing still or moving. A spherical particle of radius r moving at a velocity $\dot{x}$ is affected by viscous drag force $6\pi\eta r\dot{x}$ in the fluid of viscosity η , where x is the sloping coordinate.

The particle is affected by the force from external environment, and the force is given by the following equation

$$\frac{4}{3}\pi\rho_p r^3 \ddot{x} = F_{DEP} - F_G(\theta) - \mu_s F_{GN}(\theta) - \mu_r F_{GN}(\theta) - 6\pi\eta r \dot{x} \quad (3)$$

Fig. 2. The DEP force cancelled by the controllable gravitational force as reference force.

where ρ_p is the mass density of the particle, μ_s the coefficient of static friction, μ_r the coefficient of rolling friction. Let us consider Eq. (3) under the condition of the negligibly small frictional terms $\mu_s F_{GN}(\theta)$ and $\mu_r F_{GN}(\theta)$. If the particle is standing still ($\dot{x}=0$), the acceleration is also zero ($\ddot{x}=0$), and thus $\frac{4}{3}\pi\rho_p r^3 \ddot{x}=0$ and $6\pi r\eta \dot{x}=0$. Therefore, Eq. (3) is reduced to the following simple equation.

$$F_{DEP}-F_G(\theta)=0 \quad (4)$$

In the null method, the difference between the DEP force F_{DEP} (to be measured) and the reference force $F_G(\theta)$ is adjusted to be zero. If the difference and the reference are measured accurately, the null method guarantees accurate measurement of F_{DEP} . In our null method system, the difference between the DEP force and the gravitational force can be accurately measured as immovability of particles ($\dot{x}=0$). The reference can be calculated by;

$$F_G(\theta)=\frac{4}{3}\pi(\rho_p-\rho_m)r^3g\sin\theta=F_{G0}\sin\theta \quad (5)$$

where ρ_m is the mass density of the medium, g the gravitational acceleration and thus $F_{G0}=\frac{4}{3}\pi(\rho_p-\rho_m)r^3g$, the effective gravitational force applied to the particle considering the buoyant force. From Eqs. (1), (4) and (5) the following equations are obtained.

$$\frac{F_{DEF}}{F_G(\theta)} = \frac{3\varepsilon_0\varepsilon_m RE(f_{CM})^2 VE_{RMS}^2}{2(\rho_p-\rho_m)g\sin\theta} = 1 \quad (6)$$

$$F_{DEP}=F_G(\theta)=\frac{4}{3}\pi(\rho_p-\rho_m)r^3g\sin\theta=F_{G0}\sin\theta \quad (7)$$

Eq. (7) implies that F_{DEP} is equal to $F_G(\theta)$ which is measurable. In other words, $F_G(\theta)$ can be obtained by measuring the angle θ , if ρ_p and ρ_m are known. Eq. (6) represents that Eq. (7) is valid independently of the size of the particle. Eq. (7) holds even in case of a chain of some particles as described in the section 1 of MEASUREMENT AND RESULTS.

3. Accurate measurement of the DEP force on the inclining floor by employing the null method

Two electrodes generate the electric field lines and the equipotential lines which cross at right angles as shown in Fig. 3(a). The electric field in the upper part of the gap between two electrodes is very small. And the differential electric field in the lower part of the gap is also very small, because two electrodes are designed so that the gap may be constant at the lower part. For these two reasons the profile of the DEP force exhibits almost zero at upper and lower parts of the gap as shown in Fig. 3(b).

The two forces which are canceled in the null method should be exactly equal to each other ($F_{DEP}=F_G(\theta)=F_{G0}\sin\theta$) as shown in Fig. 3(c). In this figure the plane β is rotated

on y axis by θ from the plane α . The upward DEP force can be canceled by adjusting the angle θ (see Fig. 3(d)). Since the gravitational force is directly downward, the DEP force should be assigned directly upward on the central line between two electrodes. However, the strength and direction of the DEP force varies depending upon the position of the particle and the electric field distribution as shown in Fig. 3(e). Therefore the DEP force can not be always directly upward, and thus gravitational force $F_G(\theta)=F_{G0}\sin\theta$ can not cancel the DEP force. The strength and direction of the canceling force $F_G(\theta)=F_{G0}\sin\theta$ should be adjusted in accordance with the DEP force. In order to achieve this cancellation properly in the null method, we rotate the device by φ as shown in Fig. 3(f). Thus the upward left DEP force is possibly cancelled by this downward right force $F_G(\theta)$ as shown in Fig. 3(g). In the similar manner the upward right DEP force can be cancelled by the downward left force by adjusting the opposite angle φ . Accordingly the accurate measurement of the DEP force can be realized by inclining the plane of the DEP device and rotating the device on z axis.

EXPERIMENTAL METHODS

1. Material and electrical characteristics of particles

Although major applications of DEP force employ biological cells, the polystyrene micro-particles ($9.9\mu\text{m}^\phi$, mass density of 1.06, relative permittivity of 2.4–2.65) are used to achieve the preliminary experiment of the DEP force in this paper. Since the relative permittivity of the particles is lower than that of distilled water (78.30 at 25°C), the DEP force is negative. The polystyrene micro-particle suspension employed in this experiment is obtained by sixtyfold dilution of the aqueous suspension (including a surfactant) of 1% solids by weight (Duke Scientific Corporation).

2. Fabrication of the device

The experiments are achieved in the DEP device shown in Fig. 4(a), (b), which was fabricated by micro-photolithography. Fig. 4(a) shows schematic view of the DEP device, where only the partial view of two electrodes is magnified and shown as well as electric terminals. Fig. 4(b) is a photograph of the whole shape of electrodes. Although Au or Pt electrodes are usually adopted in these kinds of experiments, we adopted Al electrodes of sputtered film (about $1\mu\text{m}$ in thickness) which easily adheres to a slide glass tightly. And they are covered with photosensitive polyimide (photoneece®, about $1\mu\text{m}$ in thickness, Tokyo Ohka Kogyo Co., Ltd.) as insulating film. A silicon rubber spacer ($100\mu\text{m}$ in thickness) is put between a cover glass and a slide glass to construct a thin chamber which is filled with the micro-particle suspension and where the DEP force is generated. The DEP device possesses two electric terminals for AC voltage application. The surface density of the polystyrene micro-particles in the chamber is about

Fig. 3. Principle of null method.

(a) Electric field lines (dashed line) and equipotential lines (solid line) generated between two electrodes, (b) Schematic profile of the DEP force, (c) The DEP force cancelled by adjusting angle θ , (d) The DEP force generated on the polystyrene particle can be cancelled by the controllable gravitational force on the central line which can not be cancelled by the controllable gravitational force, (e) The DEP force on the polystyrene particle out of the central line which can not be cancelled by the controllable gravitational force, (f) The device rotation on z axis by angle ϕ enables the direction to change without the change of amount of the controllable gravitational force, (g) The DEP force on the polystyrene particle out of the central line cancelled by the controllable gravitational force with the change of direction.

Fig. 4. The DEP device.
(a) The schematic view of the DEP device.
(b) The photograph of the whole shape of electrodes.

Fig. 5. The injected polystyrene particles in the chamber of DEP device in the initial state of the experiment.

62 particles/mm² as shown in Fig. 5.

The electrode spacing should be spread upward in order to realize the directly upward DEP force. The DEP force becomes weak upward in accordance with the distribution of electric field. Therefore the DEP force of various intensities can be obtained in the cove between electrodes.

3. Experimental equipments

The experimental set-up to measure the DEP force generated on the polystyrene particles in distilled water is shown in Fig. 6. The microscope-holder is made of wood, where the angle of inclination " θ " is easily adjusted and fixed by the stopper as shown in Fig. 6. The DEP device "f" is placed under a microscope "c". The behavior of particles in the device is observed on the display "d" of the digital microscope (DIGITAL MICROSCOPE VHX-200, KEYENCE Co.,Ltd.). The AC voltage, which is applied to the device by the function generator "a", is viewed through the oscilloscope "b".

4. Preparation for measurement

Polystyrene micro-particles of about 62 particles/mm²

Fig. 6. Set up of the experimental equipments.

a, function generator; b, oscilloscope; c, digital microscope; d, display of digital microscope; e, microscope-holder; f, DEP device.

are suspended in distilled water in the chamber of the DEP device as shown in Fig. 5. Only one particle or several ones (in case of the pearl chain formation) should be located where the DEP force is measured and the residual particles should be removed from the view. Therefore the following preparation is needed before the measurement.

The device in which the polystyrene micro-particles are suspended in distilled water is set on the rotary table (see Fig. 3(c), (f)) of the digital microscope and vertically stood ($\theta=90$ degrees in Fig. 6). In the early stage, the voltage is not applied and falling particles are viewed through the digital microscope for about 1 hour. When almost all the particles have vanished from the view, a voltage (20 Vpp, 1 MHz) is applied to the device in order to trap only one particle or occasionally several particles. If there is no particle on the central line between electrodes, the rotary table is rotated by 90 degrees ($\varphi=+90$ degrees or $\varphi=-90$ degrees) so that the trapped particle(s) may be dragged to the central line by the gravitational force. When the trapped particle(s) is close to the central line, the rotary table

should be returned to the original state. Subsequently, the angle θ in Fig. 6 is adjusted to the initial state for measurement.

MEASUREMENT AND RESULTS

1. Inclining angle response

a. Transient response

The step response of the DEP force to inclining angle is measured by changing the angle θ rapidly from 60 degrees to 15 degrees and vice versa. The applied voltage v and the frequency f are 20 Vpp and 1 MHz, respectively. The experimental result is shown in Fig. 7. The exponential solution of the first-order linear differential equation is calculated and drawn by solid lines in the figure, where the time constants are 3 min. and 1.9 min. for decreased angle and increased angle, respectively. The measured displacement of particle Δ is indicated in black square. Since the data spread is less than the diameter of a particle ($9.9 \mu\text{m}^\phi$), the measured displacement conforms to the calculated line. Fig. 7 shows that the time to steady state of the displacement Δ is regarded to be about 15 min.

b. Static characteristics

The dependency of the particle displacement on the angle θ is examined. The position of the particles is measured for the angle every 5 degrees from 5 degrees to 60 degrees. It is shown in Fig. 8, where polystyrene particles form a chain along the electric flux. These pictures were taken 15 min. after angle assignment. A small division of the scale in the figure is $20 \mu\text{m}$ (the diameter of a particle is $9.9 \mu\text{m}^\phi$). The equilibrium position of a pearl chain is the same to that of one particle, which is described in the section 2 of THEORETICAL BACKGROUND.

From Eq. (5) the assignment of angle θ implies the assignment of gravitational force $F_G(\theta)$ downward along the inclining floor, which cancels the DEP force F_{DEP} upward along the floor to stop the movement of particles. For example, $\theta=5^\circ$ implies 25 fN ($=25 \times 10^{-15}$ N, femto-Newton) of the DEP force, $\theta=10^\circ$ does 52 fN, $\theta=15^\circ$ does 77 fN, and so on. The DEP forces at some positions on the central line between electrodes were obtained as indicated in Fig. 8. The very minute DEP force of the order of 10^{-15} N (femto-Newton) could be successfully measured.

Fig. 8 is rewritten to Fig. 9, where the displacement Δ is monotonically decreasing with angle θ . As the angle is increased, $F_G(\theta)$ is increased and the displacement by the DEP force is suppressed.

2. Applied voltage response

a. Transient response

The step response to applied voltage is also measured by changing the applied voltage rapidly from 8 Vpp to 20 Vpp and vice versa. The angle θ and the frequency f are 30 degrees and 1 MHz, respectively. The result is shown in Fig. 10. The measured displacement of particle Δ is indi-

cated in black square. Since the data spread is less than the diameter of a particle ($9.9 \mu\text{m}^\phi$), as in the case of Fig. 7, Δ conforms to the calculated lines. The time constants for the rising voltage and the falling voltage are 1 min. and 5 min., respectively.

Fig. 7. Transient response of particle displacement to rapid change of angle from 60 degrees to 15 degrees and vice versa. Each solid line represents the equations inset in each figure. The time constants are estimated to be 3 min. and 1.9 min. for decrease and increase of the angle θ , respectively.

Fig. 8. The particle movement by changing the angle θ .

Fig. 9. The dependency of the particle displacement on the angle θ i.e. the controllable gravitational force.

b. Static characteristics

The dependency of the DEP force on the applied voltage is examined. The position of the particles is measured for the applied voltage every 1 volt from 9 volts to 20 volts. It is shown in Fig. 11 and it is rewritten to Fig. 12. As the applied voltage is increased, F_{DEP} is increased and the dis-

Fig. 10. Transient response of particle displacement to rapid change of the applied voltage from 8 Vpp to 20 Vpp and vice versa. Each solid line represents the equations inset in each figure. The time constants are estimated to be 1.0 min. and 5.0 min. for decrease and increase of the applied voltage v , respectively.

Fig. 11. The particle movement by changing the applied voltage.

Fig. 12. The dependency of the particle displacement on the applied voltage.

placement by the DEP force is increased. Therefore the displacement Δ is monotonically increased with applied voltage v as shown in Fig. 12. These pictures were taken 15 min. after applied voltage assignment.

3. Frequency response

a. Transient response

The step response to applied frequency is also measured by changing the frequency rapidly from 300 KHz to 5 MHz and vice versa. The angle θ and the applied voltage v are 30 degrees and 15 Vpp, respectively. The result is shown in Fig. 13. The measured displacement of particle Δ is indicated in black square. Since the data spread is less than the diameter of a particle ($9.9 \mu\text{m}$), as in the case of Fig. 7 and Fig. 10, Δ conforms to the calculated lines. The time constants for the increased frequency and the decreased frequency are 0.7 min. and 2.8 min., respectively.

b. Static characteristics

The dependency of the DEP force on the frequency is examined. The position of the particles is measured for the frequency every 0.5 MHz from 0.5 MHz to 5.0 MHz. It is shown in Fig. 14 and it is rewritten to Fig. 15. As the frequency is increased, F_{DEP} is increased and the displacement by the DEP force is increased. Therefore the displacement Δ is monotonically increased with frequency f as shown in Fig. 15. These pictures were taken 15min. after frequency assignment.

DISCUSSION

From the transient responses shown in Fig. 7, Fig. 10 and Fig. 13, the measurement error seems to be almost less than the diameter of the particle. These figures imply that the displacement of the particle reaches steady state after 15 min. In Fig. 8, Fig. 11 and Fig. 14, the equilibrium state of the pearl chains is centered by adjusting the angle ϕ . The equilibrium state of the pearl chains in the vertical direction is controlled by the angle θ . Therefore the equilibrium state of the particle in a non-uniform electric field can be reached at any place by adjusting both of angles θ and ϕ . The maximum DEP force measurable in our proposed method is obtained to be 298 fN at $\theta=90^\circ$ from Eq. (5). The minimum value of DEP force in our experiment is 25 fN at $\theta=5^\circ$. The gravitational force around $\theta=0^\circ$, which cancels the DEP force, is comparable with the static friction between particles and inner floor of the chamber. Thus the static friction and rolling friction determines the accuracy of the DEP force measurement.

CONCLUSION

We proposed the method to measure the DEP force accurately based on the null method. The experiment is conducted with the DEP device in which microfabricated electrodes were formed. The device contains the polystyrene particles exhibiting the negative DEP in distilled

Fig. 13. Transient response of particle displacement to rapid change of the frequency from 300 KHz to 5 MHz and vice versa. Each solid line represents the equations inset in each figure. The time constants are estimated to be 0.7 min. and 2.8 min. for decrease and increase of the applied frequency f , respectively.

Fig. 14. The particle movement by changing the applied frequency.

Fig. 15. The dependency of the particle displacement on the applied frequency.

water in a non-uniform electric field. The displacement of the particle reaches steady state 15 min. after change of applied voltage v , angle θ or frequency f . The equilibrium state of the particle in a non-uniform electric field can be reached at any place by adjusting both of angles θ and φ . The proposed method can measure the incredibly minute DEP force ranging from 25 fN ($\theta=5^\circ$) to 298 fN ($\theta=90^\circ$), the accuracy of which is determined by the static friction and rolling friction between particles and inner floor of the chamber.

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