

Determination of the Charge Attached to Micro-scale Devices Used in Fluidic Self-Assembly Processes

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ABSTRACT

Self-assembly of micron sized and smaller particles has previously been demonstrated using biologically inspired events such as DNA hybridization and interactions of ligands and receptors. In order to implement these techniques to create practical electronic devices, a quantitative measure of the amount of substance attached to the device surface just prior to the final assembly is essential. In the present investigation, this crucial quantity was investigated from the electrophoretic mobility of particles, which was ascertained by examining their motion under applied electric fields ranging from 0 to 1 V/mm. Sequential CCD camera images processed with custom software enabled calculation of particle velocities during their viscous motion in an inexpensive electrophoresis chamber filled with a low-conductivity buffer solution. A linear fit through the velocity vs. electric field data points yielded the electrophoretic mobility, which was utilized in the Stokes equation to calculate the net amount of charge present on each device. For 5.44 micron carboxyl-coated polystyrene beads, this method indicated a charge of 2.69×10^{-15} C per particle. The manufacturer of the beads, Spherotech corporation, quoted 6.37×10^{-11} C as the expected charge. The more than three orders of magnitude discrepancy is at least partially attributable to the electrophoretic retardation and relaxation effects of small electrolyte ions in the buffer solution. The method was also applied to silicon islands in the shape of a cone frustum with similar dimensions to the beads. A mercapto-ethane-sulfonate monolayer, attached via thiol bonds to the gold-coated surface of the islands, provided the charge. The amount of charge on an island was calculated to average 2.48×10^{-15} C, corresponding to a density of 3.82×10^{10} mercapto-ethane-sulfonate groups per square centimeter of Au surface.

INTRODUCTION

Understanding of self-assembly processes for nano-meter and micro-scale structures has been attributed the potential to revolutionize fields ranging from nanoelectronics and materials synthesis to biological and chemical sensing and medical diagnostics and therapeutics [1]. As a result, self-assembly techniques have enjoyed an explosion of research activity in recent years. The proposed techniques rely on any combination of biological [2,3], chemical [4], electrostatics [5], or fluidic [6] methods.

Nature has already perfected accurate and controlled nano-scale assembly of components using molecular recognition of highly specific biological molecules such as DNA and proteins. In the case of DNA, hydrogen bonding drives base-pairing of complementary single-stranded (ss) DNA

to hybridize into a double strand (ds) of helical DNA. Assuming that each base pair binds with 0.5 kCal/mol of energy [7], the binding energy of a 18mer oligonucleotide can be estimated around 9 kCal/mol, although the actual dsDNA binding energy will be influenced by numerous factors such as the base-pair sequence, salt concentration of surrounding media, and temperature. In the case of antibodies/antigens and ligands/receptors, binding takes place by a combination of electrostatic forces, chemical bonding, and shape-mediated effects. For example, the protein avidin binds to biotin forming a complex with an affinity of roughly 21kCal/mol [8]. Proteins and DNA can be covalently attached to the gold surface of a patterned substrate via the Au-S thiolate bond, which has an energy of 44kCal/mol [9]. Thus, these complexes are ideal for self-assembly applications since their binding, even though non-covalent, can be as strong as many covalent interactions.

In order to utilize such bio-inspired techniques to create practical electronic devices, it will be essential to develop a quantitative measure of the amount of DNA or protein attached to the device surface just prior to the final assembly. Dielectrophoresis (DEP) and electrohydrodynamic forces, which bring complementary biological entities into the proximity required for capture, have been studied for assembly of particles in fluids and electric fields [10]. We believe that the behavior of a particle under these forces can also be used to assess binding success of DNA to its surface. In the present investigation, we report on a method to determine this crucial quantity from the electrophoretic mobility of particles.

THEORY

Since we plan on using electrophoresis to study the charges on poly-styrene beads and silicon islands, it is important to briefly review the basic theory behind this phenomenon, as discussed below.

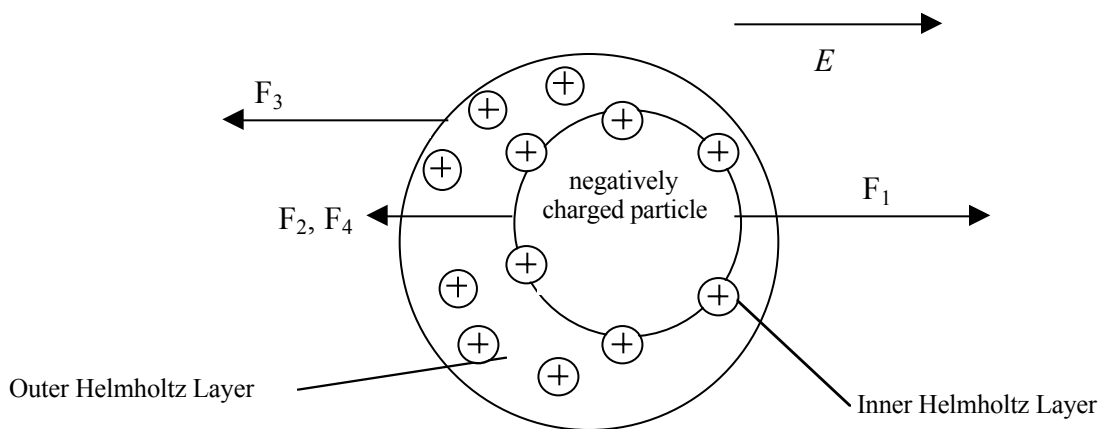


Figure 1: Forces in electrophoresis

The motion of a charged particle in a dc field is the result of four different forces acting on the particle (Figure 1). The force exerted by the electric field on the attached charge can be expressed as

$$F_1 = qE, \quad (1)$$

where q is the charge on the particle and E is the electric field. A second force, known as the Stokes friction, quantifies the drag, which opposes the direction of motion:

$$F_2 = -f_c V, \quad (2)$$

where V is the velocity of the particle, and f_c is the friction coefficient of the particle. For a spherical particle that is large compared to the molecules of surrounding liquid,

$$f_c = 6\pi\eta a \quad (3)$$

where η is the viscosity of the solution and a is the radius of the particle [11].

The two remaining forces, F_3 and F_4 , are attributed to the Helmholtz electric double layer that forms around each charged particle. Oppositely charged ions in the solution collect around the particle, forming two layers. The inner layer is held closely to the particle and may “shield” the charge of the particle, and an outer layer of more loosely held ions slow down the movement. F_3 is known as “electrophoretic retardation”, and reflects the flow of the molecules of solvent induced by the force the electric dc field exerts on the ionic atmosphere. According to the Debye-Huckel theory, the electrophoretic retardation force acting on a spherical particle is given by

$$F_3 = (\hat{\epsilon} a - q)E, \quad (4)$$

where $\hat{\epsilon}$ is the dielectric constant, E is the electric field, and the zeta potential ζ is the potential at the surface of shear between the two layers in the double layer. F_4 is known as the “relaxation effect” and results from the constant reformation of the double layer around the moving particle. As the center of the ionic atmosphere constantly lags behind the center of the particle, F_4 is an electrostatic force always opposing the direction of motion. The net force acting on the particle is the sum of all four forces described above [12].

When the Reynolds’ number is low (i.e. when size of the particle is relatively large compared to the size of the double layer and the particle moves slowly) the relaxation effect F_4 may be neglected. Thus, we are able to extract the zeta potential:

$$\frac{V}{E} = \frac{\epsilon\zeta}{6\pi\eta} \quad (5)$$

Although more advanced mathematical treatment will be required to correlate zeta potential to charge, as an initial estimate we neglect F_3 . Accordingly, we substitute $\hat{\epsilon} a = q$ (4), and (5) becomes:

$$q = 6\pi\eta a \hat{\epsilon} \zeta, \quad (6)$$

where $\zeta = V/E$, the electrophoretic mobility.

EXPERIMENTS

To conduct the experiments, an electrophoresis chamber was constructed on a glass microscope slide (Figure 2). A square well of silicone rubber 1/8 inch deep was fixed on the slide using epoxy glue. Metal wires were placed in two slits made in the rubber $\frac{1}{8}$ inch apart and half as deep as the

chamber. Under known applied DC voltage, movement of the particles was observed in the regions of constant electric field directly between these electrodes.

Polystyrene Beads

The method was validated with commercial polystyrene beads (Spherotech Inc., catalog number CP-50-10), which carry a negative charge imparted by a coating of carboxyl groups. These beads were convenient due to their spherical shape, low density, and high concentration. Manufacturer's data for the beads is listed in Table 1.

The beads were received suspended in a buffer solution of 0.01% of a neutral detergent, Nondidet P-40, in DI water. 5 μ l of the beads were diluted with 1 ml of Tris-Glycine solution (low-conductivity buffer with pH=8.5) and then pipetted into the electrophoresis chamber before superposition of the cover slip. In order to dampen Brownian currents, the well had to be completely full with no air gap between the fluid and the cover slip.

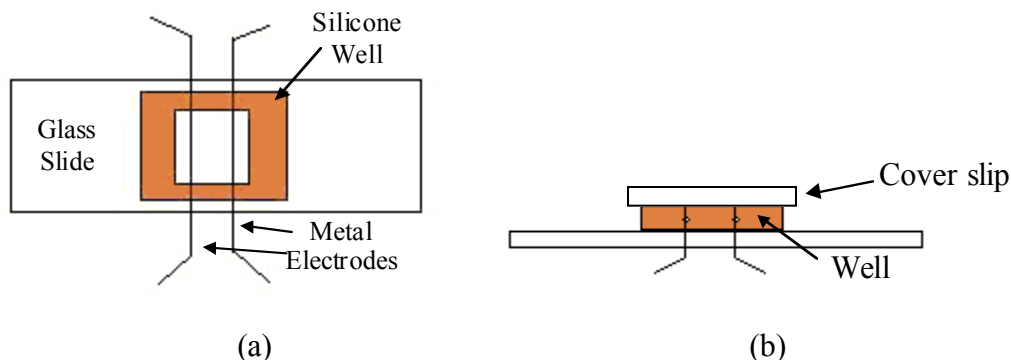


Figure 2: (a) Top view of the electrophoresis chamber. (b) Side view of the chamber.

Table 1

Diameter (μ m)	Volume (cm^3)	Mass (gm)	Surface Area (cm^2)	Concentration (#/ml)	# of Carboxyl group/bead	Charge/bead (Coulombs)
5.48	8.62×10^{-11}	9.05×10^{-11}	9.43×10^{-7}	5.53×10^8	4×10^8	6.37×10^{-11}

Voltage from a 20V power supply measured with a digital multimeter was applied to the electrodes. The resulting electrophoretic motion in the plane of the electrodes was observed and captured in sequential CCD camera images for later processing. Due to hardware limitations, the images could only be captured every four seconds, which proved adequate. The experiments were repeated several times with different applied voltages in the -5 to 5V range.

Silicon Islands

Our motivation using silicon islands is derived from the fact that there are many practical applications where single crystal silicon needs to be heterogeneously integrated onto different substrates such as plastics, glass, and other semiconductors. The use of bio-inspired assembly

techniques, such as the ones described [1,5,10] and also form the basis of the work in this paper, can provide solutions to these practical applications. We used silicon islands, as shown in Figure 3 and described earlier, which were fabricated using lithography, e-beam evaporation, and etching techniques on a silicon-on-insulator (SOI) substrate with an Au/Cr layer.

Mercaptoethyl sulfate groups were attached to the islands via a thiol bond to the Au surface to provide a negative charge in solution. The islands were initially suspended in sterilized water but transferred to the same low conductivity buffer as the beads for experimentation. The solution of islands was transferred to the electrophoretic chamber, and sequential images were captured under a range of applied voltages. The tendency of the islands to rapidly sink to the bottom of the well required periodic agitation of the solution with a pipette between data points. Due to the time for the resulting eddy currents to subside, this left only a short window of opportunity to acquire the data.

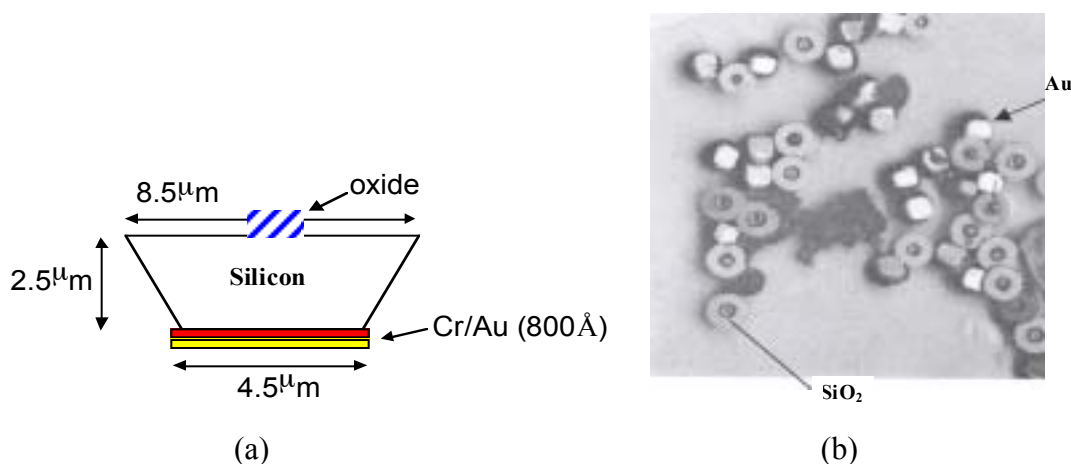


Figure 3: (a) Cross-section of a silicon particle. (b) Scanning electron micrograph of islands following release from the handle substrate [1,5,10].

ANALYSIS & DISCUSSION

The images collected from the experiments were processed using custom Matlab scripts, which overlaid sequential images and tracked the motion of individual islands. To eliminate confusion from stationary particles or dirt, information that was identical to all images within a set was subtracted from the cumulative image. The cumulative image displayed the chronological positions of each tracked island as colored dots, which in most cases appeared in a generally linear pattern. The net movement of selected particles over known intervals of time was calculated, yielding the velocity in pixels. A reference image enabled pixel to micron conversion. The velocities of the particles were then plotted against the applied electric fields. As expected, the resulting plots (Figure 4) fit a linear regression line with R^2 values of 0.947 or greater. The slope of the line was then substituted as $\dot{\Gamma}$ into equation (6), along with the values $\hat{A} = 1$ and $\ddot{E} = 1.00 \times 10^3 \text{ N}\cdot\text{m}/\text{s}^2$ associated with the buffer to yield the values of the charge.

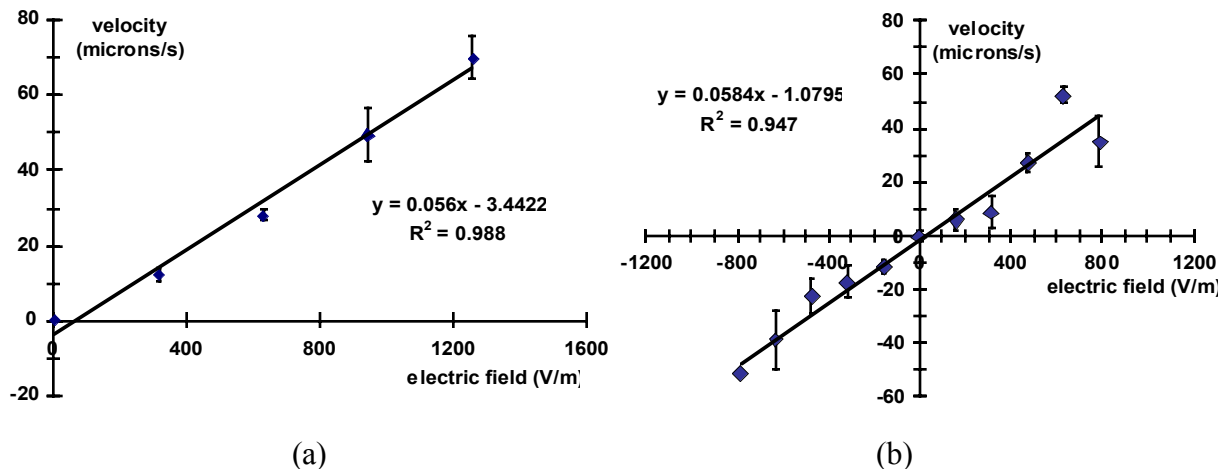


Figure 4: (a) Velocity of charged polystyrene beads vs. applied electric field, (b) Velocity of silicon islands with attached charge vs. applied electric field

Polystyrene Beads

The calculated charge for the beads was found to be 2.69×10^{-15} C/particle, which clearly falls short of the manufacturer's listed charge of 6.3708×10^{-11} C/particle. The most important factor in the several orders of magnitude difference is the shielding effect of the inner layer of the double layer. The oppositely charged ions in the fluid effectively neutralize some of the negative charge. The calculated zeta potential was 9.88×10^{-10} V.

As this phenomenon will occur in similar experiments conducted with other charged particles, it will be difficult to accurately estimate the charge on such particles. However, we postulate that it is possible to derive useful information from knowledge of the zeta potential alone, and further work is in progress.

Silicon Islands

Due to the shape, size, and density of the silicon islands, the capture and analysis of data posed several challenges. Their tendency to quickly sink to the bottom of the well required occasional agitation with vigorous pipetting, which resulted in remnant currents that caused difficulties with many experiments. Also islands were observed traveling in different orientations, suggesting a tumbling motion. Since the charge was localized to one surface, it was therefore difficult to determine where and how the electric force acted as. However, due to the low Reynolds number of the flow, it was assumed that the behavior of the island could be approximated to that of a uniformly charged sphere. The total effective charge was found to be 2.48×10^{-15} C per island, corresponding to a coating density of 3.82×10^{10} sulfate groups / cm^2 of the island's gold surface. The zeta potential for the island's gold surface was calculated to be 1.10×10^{-9} V.

A control experiment was also run on islands with no attached charged groups. The uncharged islands did not move with the electric field, confirming that the movement observed previously was indeed due to electrophoretic forces on the attached functional group.

CONCLUSION

The determination of surface charge attached to micro-scale particles using self-assembly processes was studied using electrophoresis. For the case of polystyrene beads, the values of charges obtained from this technique were different from the ones provided by the manufacturers. The difference can be partially attributed to shielding effects of the charges on the beads by the ions in the solution. The analysis can help compute zeta potential, which is an important quantity in fluidic studies of charged particles. The silicon islands with the charged self-assembled-monolayer attached to the gold surface also demonstrated clear electrophoretic movement and this technique can be useful in determining the amount of charged molecules attached to devices for the purposes of fluidic self-assembly.

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