DESIGN AND SIMULATION OF A NOVEL C-DEP MICROFLUIDICS FOR SINGLE CELL TRAPPING

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ABSTRACT

Dielectrophoresis (DEP), as a promising tool, have been used to separate, sort and deform bio-particles. In traditional method of DEP, the direct contact of electrodes with bio-particles leads to contamination and lysis of cells and joule heating in medium. In new DEP methods, such as isolated DEP (iDEP) or contactless DEP (cDEP), some of these rigors are overcame. In the method presented herein, these new techniques are used to provide a non-uniform electric field to trap a single cell in a desired area. We used the insulating structures to guide a flow as well as the manipulation of particles. Finite element analysis (FEA) is used to obtain an optimized microstructure. The joule heating and maximum DEP force is compared with traditional method and results prove the capability of these systems to trap a single cell efficiently.

Keywords: *Single Cell Trapping, DEP, CDep, Microfluidics, FEM, Joule Heating*

INTRODUCTION

Heterogeneity is a rudimentary property of a batch of cells in a tissue [1]. It is speculated that the tissue, for appropriate response to various environmental stimuli, needs the heterogeneity [2]. Sometimes, the built-in characteristic cannot be ignored and affluences further analyses [3]. Therefore, in innumerable new researches, various methods are used to study single cells [4-8].

Microfluidics with assistance of various tools, such as electric[9-12], magnetic [13, 14], hydrodynamics [15, 16] and ultrasonic techniques [17], have been utilized for cell trapping. Among these methods, Dielectrophoresis (DEP) shows more accurate and compatible response with regard to human cells. DEP has been used as an actuation force in Bio-MEMS devices such as the Lab On a Chips (LOC) or the micro total analysis systems (µTAS) [18-23]. In previous studies, DEP has been used for sorting, manipulation, separation and single cell trapping [24, 25]. If a dielectric particle locates in an electrical field (EF), it will experience a polarization and each dipole face a force due to the EF [26]. When there is a gradient of EF, a variable force acts on each part of an element; and this diversity of forces at each surface of elements leads to a net force on hole of the particle. Depend on electrical properties of the particle and media and the frequency that the system works in, the particles may pull toward high gradient of the field or low gradient of field, which are known as positive (pDEP) and negative DEP (nDEP), respectively.

The high rate improvement in micro-fabrication has provided opportunities to manipulate bio-particles with high resolution [27]. However, an EF has side effects such joule heating, gas evolution through electrochemistry, contamination of cell due to the contact of cells with electrodes and cell bursting and lysis due to high gradient of EF at sharp points [28-30]. A number of successful attempts has been done to reduce these side effects. Masuda et al. [31] used insulator obstacles, instead of electrodes at the bottom of the channel, to produce the required gradient of electric field; it so called as insulator Dielectrophoresis (iDEP). The innovation significantly reduced the formation of bubble and furthermore the fabrication of this system is much easier, especially in mass production. Afterward, Hadi Shafiee et al. [32, 33] presented a new method, called Contactless Dielectrophoresis (cDEP), that electric field will be provided along a barrier of PDMS which eliminates the side effects due to the contact between bio-particles and metal electrodes and reduces the joule heating .

Recently DEP has been utilized to trap a single cell at a desired location [34-36]. Cell patterning have been utilized to use of a cell itself as a sensor in Bio-sensors as well as to control the environment of cells [37]. A.Rosenthal and J. Voldman [12] introduced an innovative design to trap a single cell using the traditional DEP method. Nai-Chin Chen et al. [38] used the method to measure the impedance of a single cell. S.Bhattacharya et al. [39] used of a positive iDEP method to select and trap a single mammalian breast cancer cell. All the cells

experience the positive DEP only at low frequency and in a low-conductivity media which are not appropriate for all kind of cells and more importantly, due to a contact of the cells with isolator, it will limit the further analysis such as cell patterning.

On the basis of the aforementioned problems, herein we present a novel design of an isolator to manipulate and trap cells with assistance of a negative iDEP (N-iDEP). The N-iDEP contributes to keep the cells away from high gradient of electric field regions. Isolator obstacles are used to guide the fluid flow as well as electric field contributor. We examined our model with finite element analysis (FEA). Furthermore, considering the importance of increasing of temperature, the joule heating in the model is compared with traditional model. Results show that cells can be trapped firmly using the presented innovative N-cDEP.

THEORY

Two major forces which affluence particles movement are dielectrophoresis (F_{DEP}) and hydrodynamic frictional (F_{Drag}) forces. The time-average DEP force has been calculated as:

$$F_{DEP} = 2\pi\varepsilon_m r^3 \operatorname{Re}\{K(\omega)\}\nabla\left(E_{rms}.E_{rms}\right) \tag{0.1}$$

where ε_m is the permittivity of medium, r is radius of particle, E_{rms} is root mean square of an electric filed and $\operatorname{Re}\{k(\omega)\}$ is the real part of the Clausius-Mossotti factor which is given by:

$$K(\omega) = \frac{\varepsilon^*_p - \varepsilon^*_m}{\varepsilon^*_p + 2\varepsilon^*_m} \tag{0.2}$$

where ε^*_p and ε^*_m are complex permittivities of particle and medium, respectively. The complex permittivity is related to conductivity and permittivity of substance as:

$$\varepsilon^* = \varepsilon - i\frac{\sigma}{\omega} \tag{0.3}$$

where i is imaginary unit. The hydrodynamic frictional force is determined by stokes equation as:

$$F_{Drag} = \frac{1}{t_p}m_p(u_m - u_p) \tag{0.4}$$

where m_p is the mass of the particle, u_m and u_p are the velocity of medium and particle, respectively and t_p can be obtained by:

$$t_p = \frac{\rho_p d_p^2}{18\mu} \tag{0.5}$$

where ρ_p is density of particle, μ is dynamic viscosity of fluid and d_p is diameter of the particle.

METHODS

The inlet and outlet width of channel are considered as 20 µm and 40 µm, respectively. Depth of the channel is 40 µm. The overall scheme of the design is presented in Figure 1 which is heart-shape.

AC electric potential is applied to the left side channel and the right side channel is connected to the ground. Cells pass through the main channel. PDMS barrier, with 20 µm thickness, acts as barrier between the side channel and the main channel.

The Velocity Field (VF), Electric Field (EF) and the heat transfer are solved by Finite Element Analysis (FEA) using COMSOL commercial Finite Element package. Modeling is done base on cDEP presented by Hadi Shafiee et al. [32] Electrical and thermodynamic properties of materials used in the modeling are given in Table 1.

Because of the uniform VF and EF along the z axis, 2D simulation is used. At micro-scale, the effect of viscosity forces is much considerable than the inertia forces; therefore, the regime of flow field is a creeping motion. In order to apply a negative DEP force, the electric field is applied using Electric Current physic at high frequency (1 MHz). Then gradient of EF and VF, along middle line (it is showed in Figure 4 (b) and Figure5 (b)), are exported to MATLAB mathematical software. Then the drag force, due to the VF, and the DEP force, due to the gradient of EF, are calculated. Afterward, trajectory of the particle is modeled using Velocity Verlet algorithm. For comparison between traditional DEP trapping method and the herein present c-DEP method, the traditional method is modeled using 3D simulation. One of the most important reasons of lysing cells is increasing of temperature in microfluidics; therefore, increased temperature due to the joule heating is studied. Creeping Flow, Heat transfer in Solids and Electric Currents physics are used simultaneously.
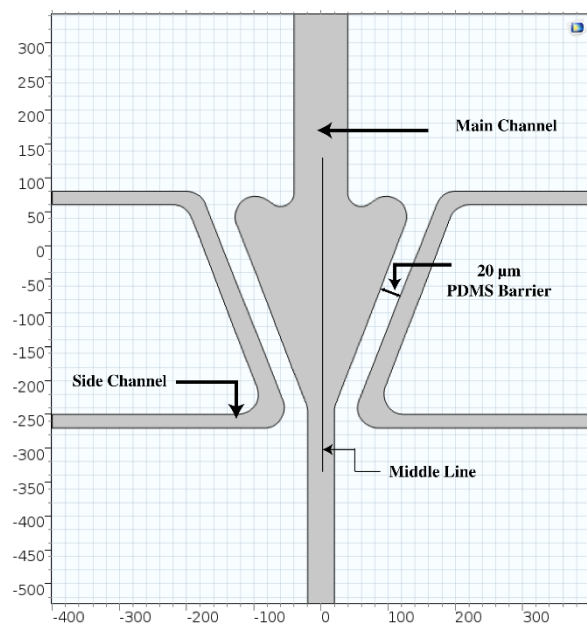

Figure 1. Scheme view of the channel and the designed model

Table 1. Electrical and thermal properties of the materials and fluids

	Properties	PDMS	Buffer DEP
Thermal properties	Heat capacity [$\frac{J}{kg \times K}$]	1460	4200
	Thermal Conductivity [$\frac{W}{m \times K}$]	1600	0.6
Electrical Properties	Relative Permittivity	2.75	80
	Electrical Conductivity [$\frac{S}{m}$]	0.85e-12	0.01

RESULTS

Trajectory of a particle for a number of voltages is shown in Figure 2. After a critical voltage, the particle is trapped in a limited area successfully. According to this figure, when the applied voltage to the side channel is more than 35V, the forces that act on particles are at equilibrium and the particle is trapped between -15 and 0 μm along the axis y. Therefore, above the specific voltage, particles will be trapped at the desired place.

Temperature of the medium in microfluidics increases because of two main reasons, conductive heat transfer between electrodes and fluid, and joule heating due to the existence of electrical field. One of the most important benefits of contactless DEP method is to prevent the increasing the temperature in microfluidics. The PDMS barrier acts as a thermal insulator, which decreases the effect of conductive heat transfer between electrodes and the buffer DEP. Secondly, it is theorized [32] that joule heating is negligible in cDEP method. For assessing this issue, increasing of temperature in the new model and the tradition DEP trapping method are simulated, as they are shown in Figure 3.a and 3.b respectively. Ambient temperature is considered to be 293 K. Then electric field is applied to the models and the increased temperature is calculated. The results show, as it is expected, in the range of trapping voltage, increasing of medium temperature in microfluidic is negligible (less than 1 ℃); however, in the traditional method, temperature of medium is increased 3-4 ℃ after 100 s (which is very significant in bio-MEMS systems). This amount of change in temperature has adverse impacts on cells.
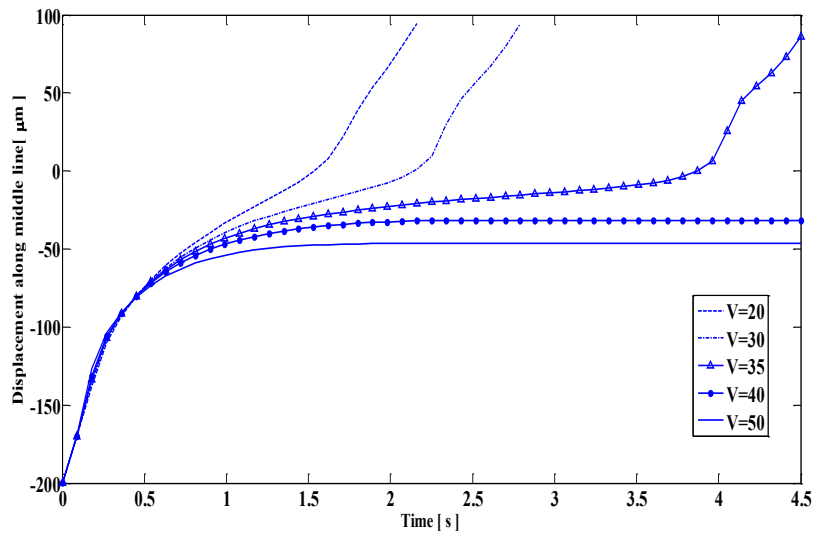

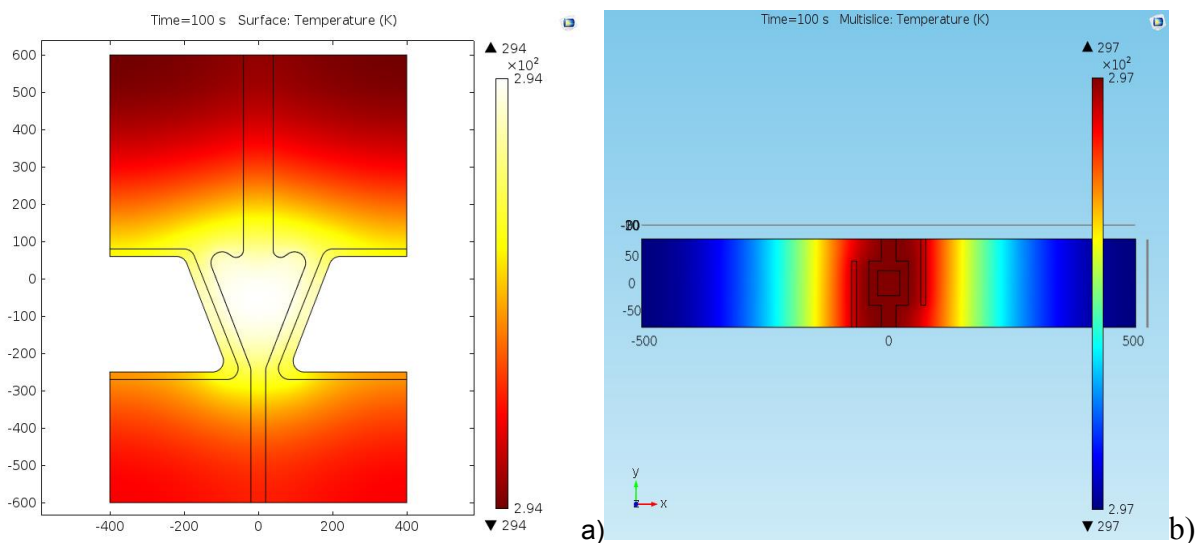**Figure 2.** Time history of particle trajectory for various voltages

Figure 3. Increased temperature in a) The new designed method and b) The traditional method

Discussion

Recently, dielectrophoresis has been used to trap a single cell at a desired place. In traditional DEP method there are some problems such as direct contact between cells and electrodes which contaminates the cells. The new methods of DEP such as insulator DEP and contactless DEP, surmount these problems. In insulator DEP method, insulator obstacles are used to create gradient of EF. It is possible to control direction and velocity of fluid flow using these obstacles too. We presented a new design to trap a single cell in a limited area. Gradient of Electric field and Velocity field are shown in Figure 4 and Figure 5, respectively.

In this paragraph, the way that the channel geometry controls the fluid flow, will be present. The geometry of the channel controls the velocity and direction of fluid flow. As it is shown in Figure 4, when a particle reaches the gorge of desired place, it will face low gradient of field which throws the particle at the desired place. When the particle locates in, the fluid velocity will be dropped abruptly; consequently, the thrust force due to the fluid flow will be reduced. It should be mentioned, eddy motion at two sides of heart-shape part of channel keeps the particles along the middle line. Finally, the channel outlet is wider than its inlet. This issue reduces the possibility of bio-particles clotting at the neck of the channel outlet as well as fluid resistance in the channel which is a big challenge in microfluidics.
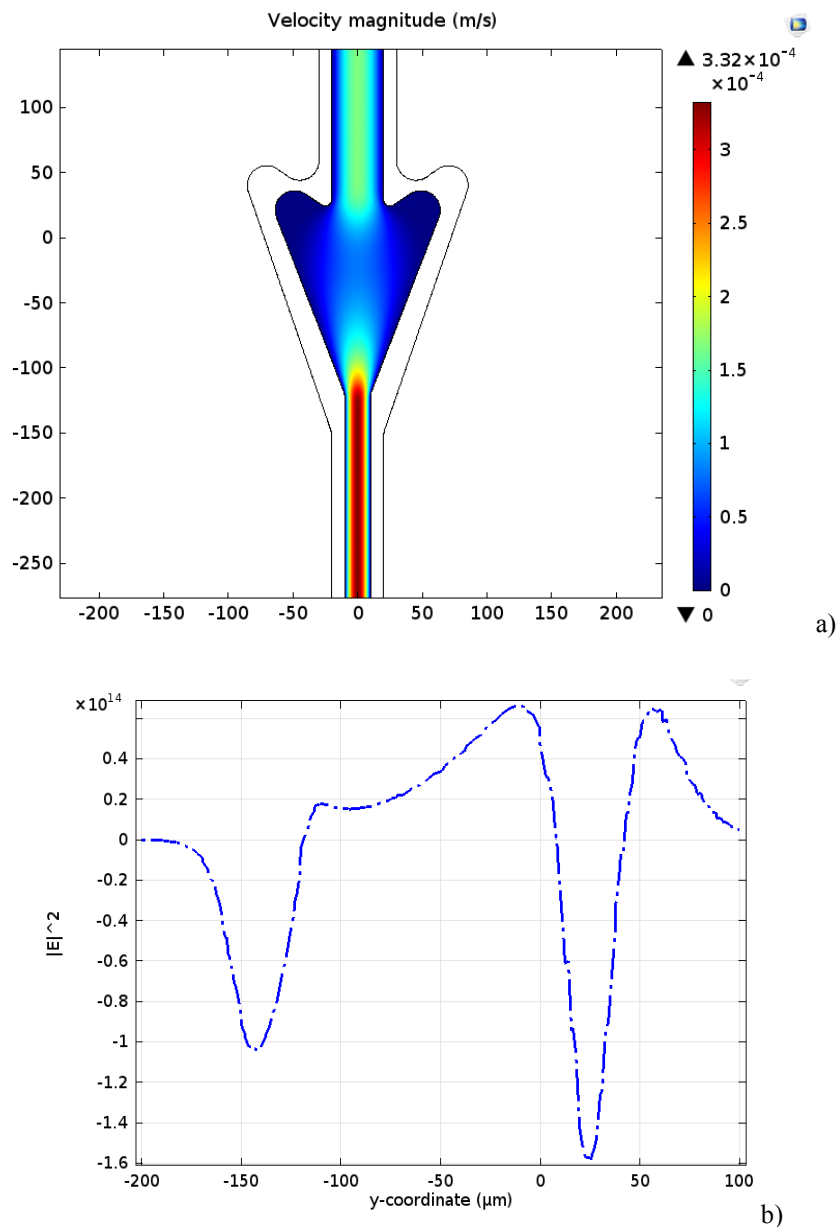

Figure 4. a) 2D plot of velocity field b) Velocity field along middle line
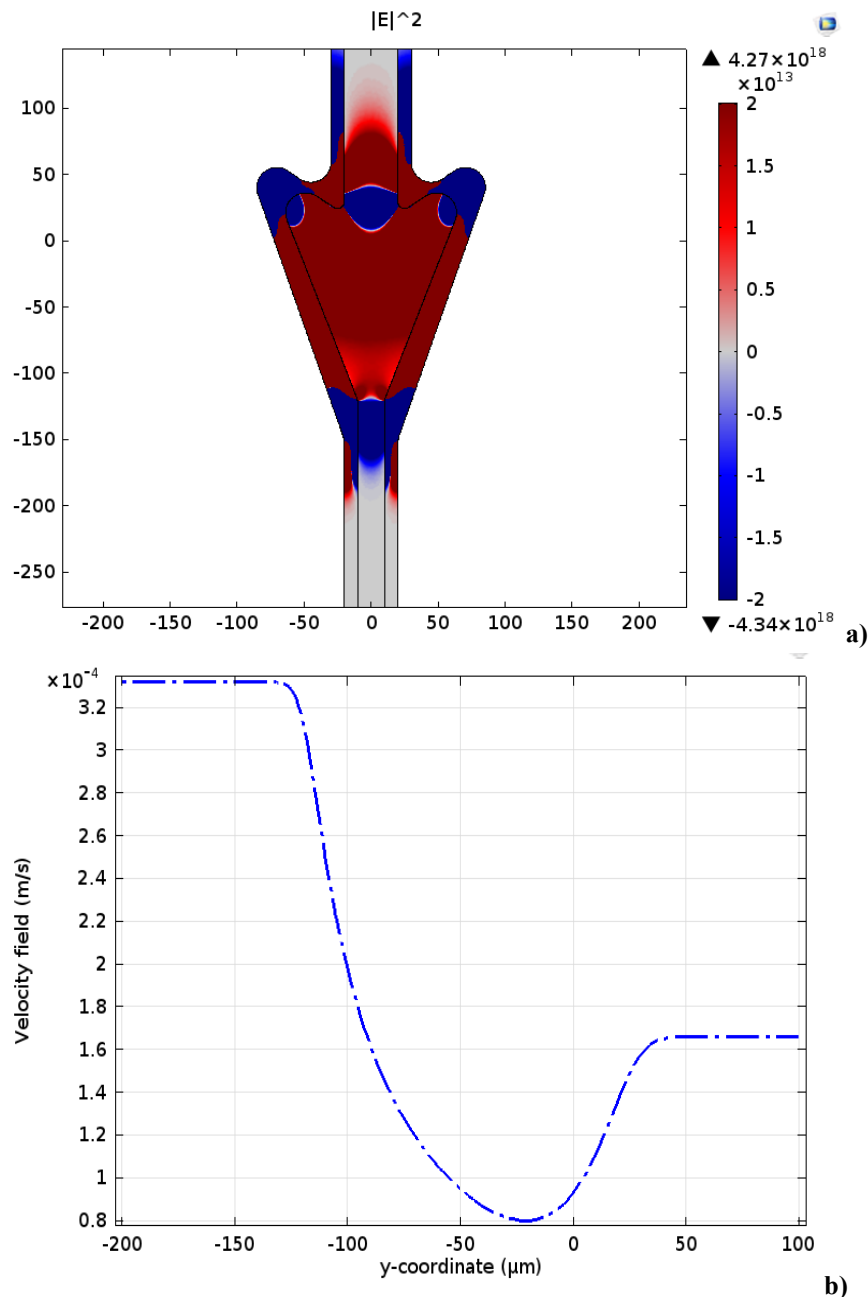
Figure 5. a) 2D plot of norm of electric field gradient b) Norm of electric field gradient along middle line

And here, the effect of the channel geometry on DEP force will be presented. Geometric design of the channel provides a suitable electric field and consequently an accurate DEP force will be applied. As it is shown in Figure 5, when a particle reaches the entrance to heart-shape part, encounter a negative gradient EF which drive particles into the desired place; however, as soon as entering the area, the particle experiences a high positive gradient of EF which reduces its velocity (see Figure 4 (a)). If the voltage in side channels is more than a critical value (in this case $35V_{rms}$), DEP force will be more that the drag force; therefore, the particle comes to a halt in the limited area (Figure 5 (b)). And again, when the particle reaches the channel outlet, experiences a high negative gradient of electric field, which trapes the particle at the area.

In conclusion, affinity between EF and VF provides a suitable circumstance which a particle can preserve in the middle of the channel. In other word this model synchronizes the variation of EF with VF. For more clarification, when a particle receives the limited area, variations of EF and VF simultaneously stop the particle

and the variations of EF and VF at the output throat of the region act as barrier. Hence the bio-particle will be arrested in the place and the particle will be ready for further analysis.

CONCLUSION

In this paper, we have presented a new design of a microfluidic channel for trapping a single cell in a desired place. Our model relies on negative cDEP. Hydrodynamic frictional force acts as a thrust force and DEP force is used to manipulate particles. Geometry and width of the channel control these forces. Therefore, we used these features to position a single particle at a desired place. Finite Element Analysis software (COMSOL) showed that the novel channel can trap a single cell at a limited area successfully. This model overcomes some rigors of previous methods. In traditional methods, cells had been absorbed to the bottom of channel, which was an impediment to further analysis; however, in our model, cells remain wholly suspended due to the uniform electric field along the height of the channel. Due to the negative dielectrophoresis which preserves the bio-particles from high gradient of electric field, we suggest that cells are less vulnerable. Another challenge of DEP method is joule heating. Since bio-particles are very sensitive to the temperature deviations, this issue is examined and we compared our model with the previous method that has been used for trapping cells. Heat transfer simulation indicates less gradient of electric field due to a PDMS barrier and smooth curvatures, declines the temperature variations, and its side effects, effectively. In previous method, temperature of medium increases about 3-4℃ close to electrodes; however, in our model it is less than 1℃.

NOMENCLATURE

F_{DEP}	Dielectrophoresis Force
F_{Drag}	Drag Force
ε	Permittivity
σ	Conductivity
ε^*_m	Complex Permittivity of medium
ε^*_p	Complex Permittivity of Particle
$K(\omega)$	Clausius-Massotti
r	Radius of Cell
E_{rms}	Root mean square of Electric Field
m_p	Mass of Cell
u_p	The Velocity of Cell
μ	Dynamic Viscosity of medium
ρ_p	Density of particle

Abbreviations

DEP	Dielectrophoresis
cDEP	contactless-Dielectrophoresis
LoC	Lab on a Chip

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