A Study of *Carteria* sp. Cell Electrical Lysis in Straight and Tapered Microfluidic Systems

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Abstract. Electrical cell lysis is one of the most efficient techniques in cell analysis in extracting intracellular components for further use or examination. Cells are exposed to high electric field strength that can rupture cell membrane due to nano-size pores creation. Thanks to microfabrication technology, electrodes can be materialized in close proximity to each other leading to high electric field strength with low applied voltage. In this study, two microfluidic systems were designed: straight and tapered systems. The straight channel was chosen due to the ease of fabrication, while the tapered channel was picked since it intensifies electric field strength in the tapered area. Finite element method was used to simulate electric field distribution inside the microfluidic systems and transmembrane potential (TMP) across cell membrane. In the experiment, green algae *Carteria* sp. which contains large amount of lipid was used in performing microfluidic electrical cell lysis. For 30 nm thick gold planar electrodes with 110 µm spacing fabrication, the simulated TMPs produced in both microfluidic systems exceed 0.2 V which is a threshold potential for pore generation in cell membranes when applying 34 V<sub>p-p</sub>, 1000 Hz AC voltage. Only *Carteria* sp. cells were successfully lysed in straight microfluidic system after being exposed to high electric field strength for several minutes. Although electric field in the tapered region was higher than that of the straight microfluidic system, the cells were rushed out of the tapered region due to electrohydrodynamic effects.

1. Introduction
Cell lysis is a cell membrane rupturing process that is normally used to release the intracellular components for further analysis. Lysis processes can be categorized into mechanical, chemical, and electrical methods. The mechanical method uses mechanical forces to directly damage cell structure, while the chemical method relies on lysis surfactants to solubilize the membrane, and the electrical method employs high electric fields for lysis via electroporation. Normally, electrical lysis is performed in large devices that need high voltage for a sufficiently high electric field to disrupt the membrane [1]. In recent years, the development of microfluidics has made miniaturized lysis devices possible. Due to micro-dimension of the channels and electrodes, very
high electric field strength can be generated to perform cell lysis by using a much lower voltage. Moreover, geometric modification of microchannel could be exploited to amplify local electric field strength to a level that could lyse cells. AC voltages have also been used to alleviate electrode destruction due to water electrolysis. *Carteria* sp. cells were used in lysis study so that the extracted intracellular components could be further analyzed for lipid content.

2. Theory

For pores to be created on the membrane of the cell being in presence of an external electric field, the cell’s transmembrane potential (TMP) must be greater than the threshold value of 0.2 V [2]. If the TMP reaches a critical value of 1.0 V, the cell will undergo irreversible pore creation, leading to cell lysis [1].

The electric field in microfluidic systems and TMP across cell membranes were simulated using the AC/DC module of COMSOL Multiphysics simulation software using the following equations:

\[
\nabla \cdot J = Q_j
\]

\[
J = \sigma E + j\omega(\varepsilon_0\varepsilon_r E + D_r) + J_e
\]

\[
E = -\nabla V,
\]

where \(J\) is the current density per unit volume, \(Q_j\) is the current source, \(\sigma\) is the electrical conductivity, \(E\) is the electric field, \(\varepsilon_0\) is the permittivity of the vacuum, \(\varepsilon_r\) is the relative permittivity of material, \(D_r\) is the electric displacement when no electric field is present, \(J_e\) is the externally generated current density, and \(V\) is the electric potential.

3. Materials and Methods

Microfluidic devices for cell lysis in this work were fabricated using a printed circuit board technique and sputtering technique on polydimethylsiloxane (PDMS) (Sylgard 184, Dow Corning) microchannels and gold planar electrodes on glass slide fabrication respectively. High resolution transparency masks were used to fabricate microchannel molds and electrodes patterns by exposing 30 µm thick negative photoresist film (Riston Special FX Series - FX515, DuPont) to 400 nm wavelength light and were then developed. The electrodes were designed to be 100 µm apart. The designed width of straight microchannel was 60 µm while the tapered channel was designed to taper down from 600 µm to 20 µm where the thinnest part was 50 µm in length. The *Carteria* sp. cell lysis experiment was performed using fabricated microfluidic device. The electric field distributions inside microchannel and peak TMPs of *Carteria* sp. cells when applying AC voltage of 1000 Hz were simulated to find the appropriate TMP for cell lysis. The values of electrical properties of materials used in COMSOL simulation are listed in Table 1.

| Material                  | Conductivity (S/m) | relative permittivity |
|---------------------------|--------------------|-----------------------|
| Intracellular fluid       | 0.3                | 60                    |
| Membrane (8 nm thick)     | \(1 \times 10^{-6}\)| 15                    |
| Mucilage (0.75 µm thick)  | 0.214              | 23.1                  |
| Extracellular medium      | \(2.13 \times 10^{-2}\)| 80                    |
4. Results and Discussion

The fabricated microfluidic chip for cell lysis is shown in Figure 1. The dimension of the straight system microchannel that fabricated was 60 µm in width and 30 µm in height with the actual spacing between gold planar electrodes being 110 µm. For tapered system, the tapered channel had actual dimension of width and length 17 µm and 40 µm, respectively.

![Figure 1](image)

**Figure 1.** (a) Fabricated microfluidic chip with magnified electrode areas of the (b) straight and (c) tapered systems.

The simulated electric fields from applying 34 V\_p−p\_ were strongest at the inner electrode edges for the straight channel but were strongest at the narrowed part for tapered channel as shown in Figure 2 (a) and (b), respectively. The strongest electric field values at were the middle of the straight channel and the narrowed area of the tapered channel at approximately 125 kV/m and 217 kV/m, respectively.

![Figure 2](image)

**Figure 2.** COMSOL simulations of electric field distributions between electrodes separated at a distance of 110 µm in (a) straight and (b) tapered microchannels.

The *Carteria* sp. cell was modeled as an ellipsoid with 11 µm and 14 µm in the minor and major axis, respectively. The TMPs of *Carteria* sp. cell sitting on top of the inner edge of an electrode and on the channel floor between electrodes are shown in Figure 3. Although the simulated TMPs of cells from applying AC of 34 V\_p−p\_ did not reach the critical 1.0 V, exceeding 0.2 V threshold is enough for generating pores and could lyse cells that were exposed to the electric field for a sufficiently long time. A number of cells could be lysed at the electrode edge for the straight system and the region between electrodes for the tapered system due to higher the TMPs of cells than elsewhere as can be seen from the graphs in Figure 3. The TMP simulation result in Figure 3 (b) predicted that the tapered system would be more efficient in performing lysis of cells because of its ability to cause the highest TMP.

For the experimental part in the straight microfluidic system, the *Carteria* sp. cells moved due to convection of the medium and was stuck at the edge of the electrodes due to high electric field interaction. After several minutes, intracellular components were observed to leak out of the cell as shown in Figure 4. For the tapered microfluidic system, the medium rapidly flowed when entering the narrowed part. Hence, most cells swiftly passed through tapered region and
Figure 3. COMSOL simulation of the TMPs along the major axis of an ellipsoid *Carteria* sp. cell orienting in longitudinal direction of the channel within (a) straight and (b) tapered microfluidic systems when applying AC voltage of 34 $V_{p-p}$.

remained intact. Some cells occasionally became stranded at the edge of the electrodes but were often blown away due to medium stream. The cell ‘s speed when passing narrowed area was approximately 6 mm/s.

Figure 4. (a) Microscope image of *Carteria* sp. cell responses when applying AC of 1000 Hz 34 $V_{p-p}$ in straight microfluidic system and (b) electron microscope image of lysed cell.

5. Conclusion
In the microfluidic *Carteria* sp. cell lysis study, the TMPs of the cells were simulated using COMSOL Multiphysics AC/DC module to ensure the high enough applied voltage to cause cell lysis in actual experiment. The experimental results showed successful cell lysis only in straight channel configuration. The tapered channel configuration created the hydrodynamic force liquid convection, providing unfavorable condition for cell lysis, even though the simulated TMP was high enough to cause electrical lysis in the tapered area.

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