Detection of E.coli Cell Using Capacitance Modulation

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Abstract: Biosensors can be used to detect the microorganisms present in water [1], [2]. This paper presents a method to detect E.coli bacteria in water depending upon the capacitance modulation in the presence and absence of E.coli cell, which is simulated in Comsol Multiphysics (ver-3.5). Debye layer and cell wall effects [Figure 6, 7, 8] can also be observed using this method. It also presents the simulated results of change in capacitance with microorganism moving with fluid flow [Figure 9].Results show that capillary force is suffices for flow of E.Coli flow with water.

Keywords: Biosensors, Capacitance modulation, Cell wall, Capillary force, E.Coli, microfluidics.

1. Introduction

Water and food borne diseases caused by bacterias such as E.coli, Listeria, Salmonella etc. result in hundreds of thousands of hospitalizations each year. For food producers, water suppliers and water purifiers always the problem is to be able to detect pathogen presence as rapidly as possible and at the minimum possible concentration. Traditional methods for the detection of water borne bacteria or any other bacteria rely on culturing and plating, requiring from 24to 48 hours confirming the analysis and only when at least 10,000 colony forming units per milliliter (cfu/ml) have been cultured. After that PCR (polymerase chain reaction), a DNA based method was devised which requires 36 hours of processing time. A new process for detecting biomaterial has been developed using capacitance modulation that amplifies the sensitivity of the sensor [3].

Indian Health Services (IHS) fact sheet shows that While 1% of the U.S. general population lacks access to safe water, 12% of Indian homes lack access to safe water. There is a backlog of over 3,300 needed sanitation facilities construction projects. The cost to provide all American Indians and Alaska Natives with safe drinking water and adequate sewerage systems in their homes is estimated to be almost $2.9 billion. With inflation, new environmental requirements, and population growth, the current sanitation appropriations are not reducing the backlog. These all problems leads to deaths as shown in Figure1.although death date is reducing and percent of homes with potable water is increasing still there is backlog. It needs more accurate and precise technology to move out this problem. A better water purification and quality of water can reduce death rate.

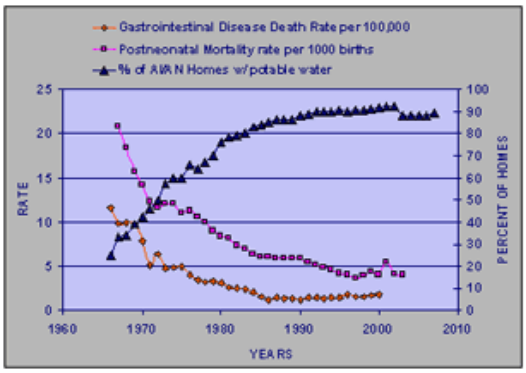

Figure1.Death rates due to microorganisms in water [4]

2. Principle

Bacteria inside water can be modeled as an ellipsoid capacitor in between two plates of a parallel plate capacitor as shown in Figure 2.equivalent capacitance without bacteria is capacitance of a parallel plate capacitor and equivalent capacitance can be obtained through series parallel combination of the capacitances in complete system.

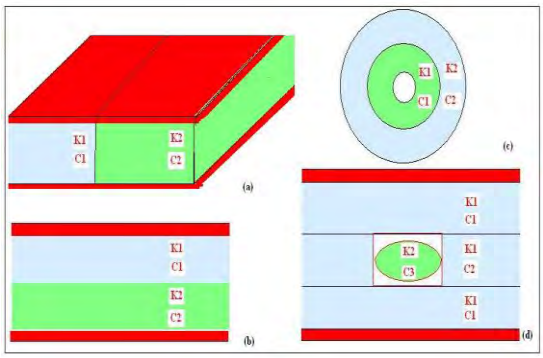

Figure2. Capacitance equivalent (a) lateral combination of dielectrics makes parallel combination of C1 and C2 (b) layered combination of two dielectrics makes series combination of C1 and C2 (c) concentric spherical combination also equivalent two series combination of C1 and C2. (d) represents cell between two electrodes that can be modeled from (a),(b) and (c).

Capacitance difference calculated by equation (1) and equation (2) gives us sense of bacteria.

$$C_{eq1}=C_0 \qquad (1)$$

$$\frac{1}{C_{eq2}}=\frac{1}{C_1}+\frac{1}{2C_2+C_3} \qquad (2)$$

Where C_0 is the capacitance of parallel plate capacitor without any bacteria and for our case C_1, $C_2$and C_3 are shown in Figure2d.

The capacitance of an ellipsoid is given by [5]

$$C=4\pi\varepsilon_0 R_{eq} \qquad (3)$$

Here Req is the equivalent radius of the ellipsoid. It is calculated from

$$R_{eq}=\frac{2\lambda}{F(\varphi/m)} \qquad (4)$$

$$F(\varphi/m)=\int_0^{\varphi}(1-m\sin^2\theta)^{(-1/2)}d\theta \qquad (5)$$

Here λ is

$$\lambda=\frac{c}{2}\sqrt{1-(\frac{a}{c})^2} \qquad (6)$$

Ellipsoid is shown in Figure3.c is the length and a and b are other sides of ellipsoid.

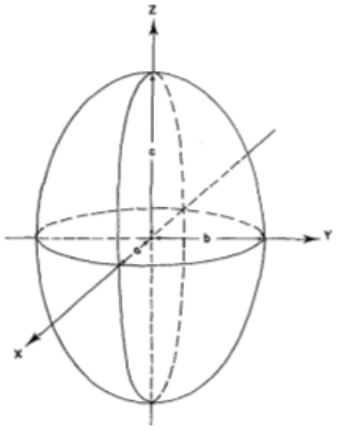

Figure3. Ellipsoid $(\frac{x^2}{a^2}+\frac{y^2}{b^2}+\frac{z^2}{c^2})=1$, [5]

These formulae are valid only when c>b>=a; and Φ and m are given by

$$\varphi=\arccos(\frac{a}{c}) \qquad (6)$$

$$m=\frac{1-(\frac{b}{c})^2}{1-(\frac{a}{c})^2} \qquad \text{for } m<1 \qquad (7)$$

The capacitance of capacitor changes if the dielectric constant of the material inside the capacitor also changes or its having combination of dielectric materials.

The bacteria size and shapes [6] are given below and shown in Figure4.

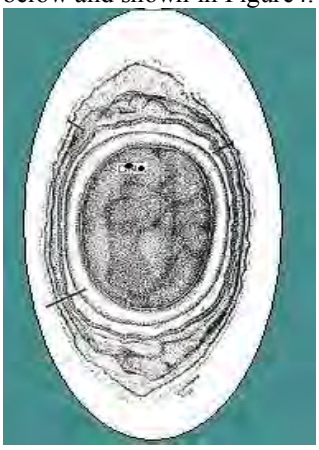

Figure4. A typical cell structure like an ellipsoid [6]

Institute of bimolecular design provides a lot information[7] useful for design from there we have length and diameter of the cell equal to 2μ

and 0.8μ respectively. Other Parameters like conductivity and dielectric constant are modeled by the data used by a group working on impedance based detection at university of Arkansas. These values are 0.5 S/m and 60 respectively. The parameters are for cell cytoplasm. In this study the parameters are calculated based on impedance variations due to presence of E.coli in the water. As impedance is sensitive to ionic secretions from the cell so it cannot be considered constant with time. Apart from this it is difficult to consider complete cell as Homogenous media because Cell wall and cytoplasm have different electrical properties. On the other hand in capacitive modeling changes can be observed due to presence of both cell wall and cytoplasm of the bacteria. Although dielectric constant of the cell wall itself is to be still found but for simplicity we can assume it little lesser than the cytoplasm and see the changes.

Simulation results using Comsol multiphysics confirms this principle and shows a reasonable capacitive changes.

For processing the signal of capacitive change we can have normalized capacitive change which improves sensitivity

$$\frac{\Delta C}{C} = \frac{C_{wc} - C_{woc}}{C_{woc}} \qquad (8)$$

Here C_{woc} refers to capacitance without cell and C_{wc} is capacitance with Cell. We can calculate equivalent capacitance using formulae (1),(2) .

3.Bacterial Model in Comsol multiphysics™

The model for biosensor has been defined in the 3D analysis application mode of the electrostatics of MEMS module in COMSOL Multiphysics™. .where the unknown field quantity is the voltage V = V(x,y,z).The modeling domain consists of cuboid with dimensions 10μm X 5μm X5μm.The cube is filled with a buffer of a relative dielectric constant of 80. The sides of the cuboid confine the buffer volume. The electrode plates confine the block. Electrode plates are defined like metallic plates. The boundary condition on four sides of cuboid was set to electric insulation. The electrodes are defined on two boundaries of the sensor. They form a parallel plate capacitor. The electrodes were defined to be 10μm long and 5μm wide with an inter-electrode spacing of 10μm. The electrodes have been modeled as perfect conductors and are treated as equipotential surfaces with finite thickness. The E .coli cells were defined as ellipsoids of dimension 1 μm by 0.5 μm by 0.5 μm. The conductivity and relative dielectric constant of cytoplasm of the E .coli cell was set 60 and cell wall was set to dielectric constant of 40 respectively. Meshing is done using free triangular method. Results of simulation showed potential variation. In the postprocessing we can get capacitance value. Voltage difference applied between electrodes was 10V.

Figure6 represents the geometry of the model Electrodes are shown in blue color and the material in between without cell in green color. Figure 6 represents the geometry with bacteria modeled

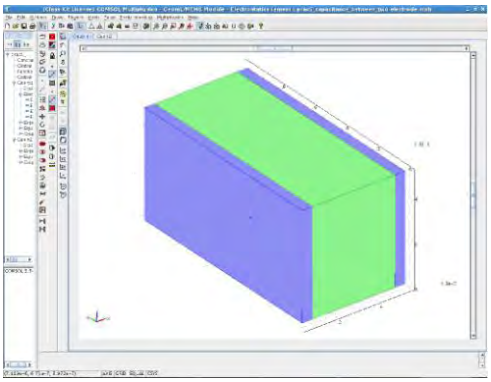

Figure5: cuboid capacitor in Comsol Multiphysics™

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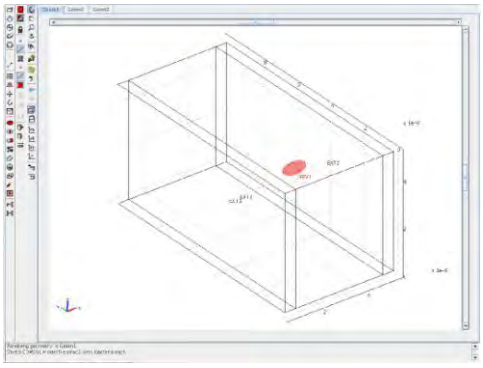

Figure6: one bacterial cell between two electrodes 15μm apart

4. Analysis of Model

When model is simulated using Comsol multiphysics in electrostatics module. some parameters observed are listed in the table1.

Table 1.

Model	Value(F)
Capacitance between two electrode Without Any cell	2.29426*10-16
Capacitance between two electrode with a cell of 1um * 0.5 um * 1um. Here cell wall was not defined	2.3184 * 10-16
Capacitance with five cell, cell wall not defind	2.3196 * 10-16

As values of Capacitance, are very less and difficult to measure with accuracy. To improve resolution and accuracy and readable capacitance value we have designed capacitive sensor array using comb drive as shown in figure 7.

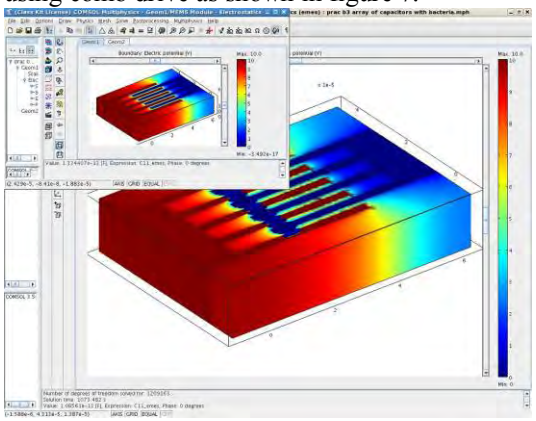

Figure7: array of 8 sensors using interdigitated comb drive structure with bacteria present and without bacteria [in inset].

After using comb drive we got results that have values around 0.01 pF which is measurable. The difference we have got change of 25 fF without bacteria and with bacteria. Results obtained are shown in Table 2

Table 2

Model	value(F)
Equivalent Capacitance of electrode array without any cell	$1.33*10^{-13}$
Equivalent capacitance with 16 bacterial cell over electrode array	$1.08*10^{-13}$
Resolution in the capacitance measurement required	$25*10^{-15}$

5. Micro fluidic flow of bacteria in Capillary

Figure 8 represents the model of the sensor as a capillary .electrodes is defined between two points in the upper boundary and lower boundary. Bacteria moves along the capillary then capacitance change can be observed using time dependent analysis.

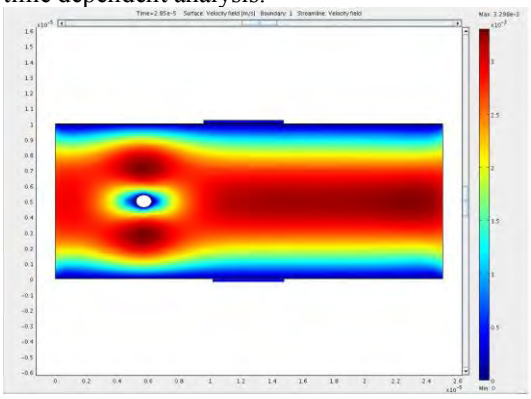

Figure8: bacterial cell flowing with fluid in capillary capacitive sensor

For better resolution array of electrodes is placed in between two boundaries and significant results can be observed as shown in Figure 9

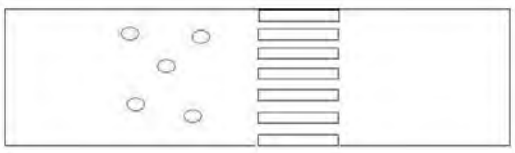

Figure9: bacterial cell flowing with fluid in array of 6 sensors

Height of meniscus is given by

$$h = \frac{2.\gamma.\cos\theta}{\rho g r} \quad (9)$$

Where γ is liquid air surface tension (Energy/Area), θ is contact angle density of liquid (mass/volume) and r is the radius of the capillary.

For a water filled glass tube in air at sea level at room temperature using S.I. units. Values of constant are as follows

$\gamma = 0.078 J/m^2$ at 20^0 C
$\theta = 0.35$ rad or 20^0
$\rho = 1000\ kg/m^3$
$g = 9.8\ m/s^2$

Using these values for 15 μm capillary heights is 0.993m. It means viscous force is sufficient for water flow in 15 μm tube

On spherical meniscus the pressure drop is given by equation [8]

$$P = \frac{\alpha.\gamma.\cos\theta}{r} \quad (10)$$

Where γ being the interfacial surface tension between the two fluids, θ the wetting angle and α numerical prefactor.α equal to 2 for cylindrical pores; more generally, $\boldsymbol{\alpha}$ /r can be replaced by the ratio of the perimeter to the area of the pore .The upper bound for this pressure represents our estimate of P_b, and it occurs for $\theta=\theta a$ which is called as advancing contact angle

$$P_b = \frac{\alpha.\gamma.\cos\theta_a}{r} \quad (11)$$

As long as differential pressure across the capillary is maintained below this threshold (Pb), the meniscus advances along the capillary and the fluid permeates through the capillary channel. Several factors can affect the prefactor α: two-dimensional confinement of the fluids (e.g. in the case of microfabricated capillaries connecting microfluidic channels of same depth), non-cylindrical cross-section of the capillaries (e.g. in the case of rectangular microfabricated channels or of asymmetric pores for certain membranes) etc

with $\gamma \approx 50$mN/m, $r \approx 15\mu$m, $\alpha \approx 1$ and $\boldsymbol{\theta}=0$ yields Pb≈3.3kPa (0.5psi), which is indeed quite low and difficult to reliably control. That is why radius of the capillary should not be less than 15 um.

Other boundary condition for channel diameter is decided by the length of E.Coli(Cells are like rods or long prolate spheroids having average length of 1.2 to 5.2 μm and diameter of 0.6 to 0.7 μm.[6]).

Using these boundary conditions the model was simulated in the fluid structure interaction (MEMS module) application mode we have observed following variation. it shows capacitance variation as bacteria enters into the cell. Minimum pressure difference needed for flow using equation (11) is also verified.

6. Conclusion and future work

Capacitive sensor woks without any incubation. It is proposed that after fabrication it will maximally take two-three hours in the total detection time from sample preparation to result display. The sensor is portable and can be used on site, and the same technology will work to detect non biological materials as well as biological pathogens.

7. References

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