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Label-Free Bioparticles Detection Using Lab-on-Chip

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Abstract
Lab-on-Chip (LOC) is a miniaturized device that is able to perform single or multiple laboratory operations on a single chip. This paper intends to study, explore and transform the laboratory-based bioparticles detection technique to part of LOC device. The combination of MEMS capacitive sensor and microfluidics is expected to develop and use as a LOC device to detect the E.coli bacteria in a water-based medium. In this research, the interdigitated electrodes (IDEs) were applied as the capacitive sensor due to their low cost, ease of fabrication process and excellent sensitivity. The capacitive sensor LOC was designed and simulated using the MEMS module in COMSOL Multiphysics. The effect of the number of bacteria on capacitance change was also discussed. The simulation results showed that by increasing the number of bacteria, the capacitance decreased.

Keywords: Lab-on-Chip, label-free detection, interdigitated electrodes, sensor, bioparticle

Introduction
Tropical diseases which are more prevalent in the tropical regions are caused by the introduction of biological agents or bioparticles namely, viruses, bacteria and parasites into the human body [1, 2]. In the process of detecting infectious diseases, prompt results during clinical diagnosis are very important to prevent transmission and death. Unfortunately, diagnostics of a suspected infectious disease requires for the use of expensive equipment that may take days to complete due to the demand of strict procedures as well as other health issues. This situation often leads to delays in treatment and has been the cause of a high number of fatality rate in developing countries [3]. The solution to this problem is to develop a simpler, quicker and cheaper bioparticles detection mechanism that is suitable for use in clinical diagnostics.

In order to detect the bioparticles, various methods have been utilized such as conventional methods using culturing and biosensor techniques. The most reliable methods of detecting the presence of bacteria in samples are culturing techniques which are very reliable with low detection limits. Unfortunately, it takes too long to wait for a confirmed result when these conventional methods are used [4]. Therefore, numerous researches have been conducted to find a faster culturing technique that will still retain these desirable properties. As a result, scientists began to develop biosensors – analytical devices that combine biological components with detectors – to detect analytes, which is especially useful when detecting pathogen [5].

The microfluidics field has emerged as a new tool in clinical diagnostics that has potential in chemical and biological processing and analysis. As the name implies, the microfluidics concept involves the transportation of small volume (microlitres to attolitres) of fluid through micro-sized channels which have a dimension of tens to hundreds of micrometers [6].

The Lab-on-Chip (LOC) is a miniaturized device that is able to perform single or multiple laboratory operations on a single chip [7]. In the interest of developing new, faster, cheaper and easier methods for handling the detection of bioparticles as well as other biological and chemical samples, the integration of microfluidics field with the Lab-on-Chip technology is inevitable [8].

The two most common techniques used to detect bioparticles are by label detection and label-free detection. In the former technique, the labels (normally fluorescent tags) that bind with the target bioparticles will be used to indicate presence. Alternatively, the latter detection technique does the opposite where the bioparticles are not modified or labeled prior to the detection process and they are detected in their natural forms [9, 10].

A number of optical and electrical techniques have been used to detect the bioparticles that are present in water, foodstuff, and blood. Label detection using fluorescent tags is the preferred optical method. However, this method involves complex procedures using expensive diagnostics tools that take too long to yield any result. Additionally, there is also a problem with the target bioparticles being modified by the label which may disrupt the bioparticles' function [9]. Magnetophoresis is another detection technique based on utilizing the magnetic fields. This technique requires to label target cells which can change their structures as well as the detection result [11].

The label-free impedimetric techniques allow scientists to read the impedance value of target bioparticles by dipping the electrodes straight into biological mixtures [12]. In addition, bioparticles can cause changes in the dielectric properties (also known as capacitance change) when they come in adjacency of the sensing electrode. Therefore, another label-free technique uses a capacitive sensor that can detect the changes in capacitance, instead of the impedimetric sensors.

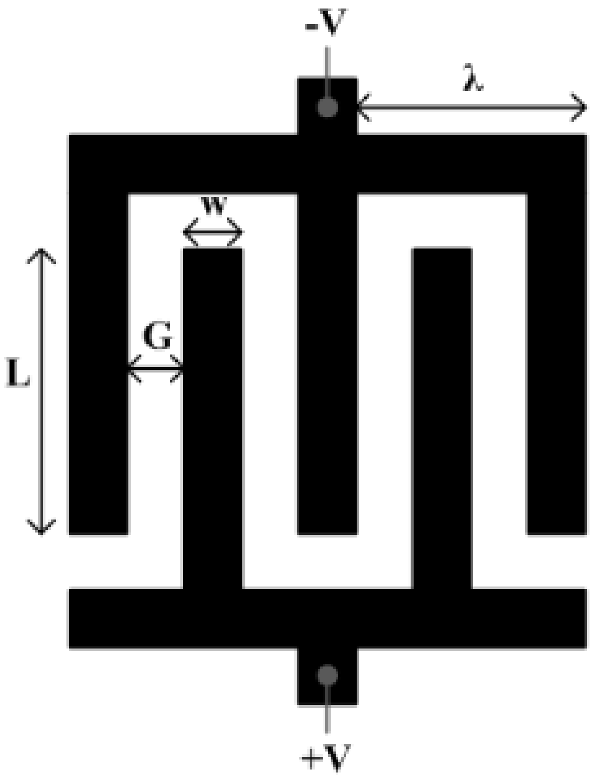
To date, interdigitated electrodes (IDEs) are used as the sensor in different devices such as MEMS biosensor. The IDEs have also been used in transducers for Lab-on-Chip devices [13]. The IDEs are the periodic grouping of two parallel electrodes that are designed by length (*L*), width (*W*) and gap between electrodes (*G*) [14]. The miniaturization of electrodes and their low cost have made the planar IDE arrays a popular framework for designing sensor device. Uncomplicated and economical mass-fabrication procedure, excellent sensitivity, and the potential of usage in a number of applications without substantial changes in the sensor design are the most significant advantages of these sensors [15, 16]. The applications of this sensor can be in the capacitance measurement and detection of conductivity and dielectric constant in biological solutions [17].

The capacitive sensor LOC can be applied in clinical diagnostics such as in bioparticles detection in a suspected case of tropical infectious disease. In Malaysia, the focus of researchers in the health field is on pathogenic bacteria such as Escherichia coli (E.coli) [18] and salmonella [19] as both bacteria can lead to food poisoning and illness in humans.

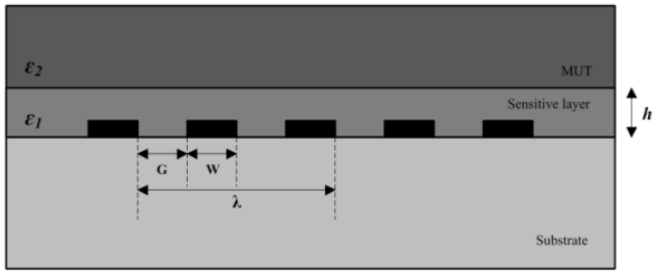
This research has been conducted with the objective of developing a convenient Lab-on-Chip device that can detect the presence of the E.coli bacteria and suitable for use in the tropical regions.

Physical Parameters of IDE Sensor

A schematic and cross-section of an IDE with two comb electrodes are illustrated in Figure 1; where *W* is the width of the fingers and *G* is the width of the gaps between electrodes. Each comb electrode is connected to a pre-set potential (+V/−V) and comprises of a number of fingers with length *L*.



(a)



(b)

Fig.1. (a) Schematic of an IDE (b) cross section of an IDE sensor

The spatial wavelength of electrodes (λ) is defined as [14]:

$$\lambda = 2(W+G) \quad (1)$$

The ratio of sensitive layer height (h) to electrodes spatial wavelength (λ) is defined as the layer adimensional thickness ($r = h/\lambda$). Another parameter is the metallization ratio (η) that is defined as [14]:

$$\eta = \frac{W}{W+G} = \frac{2W}{\lambda} \quad (2)$$

Sensor Optimization

In this section, we investigate the dependence of the capacitance of IDE on the geometric parameters including IDE finger spacing, thickness, sensor spatial wavelength and sensitive layer thickness. It is crucial to understand such features for evaluating and developing future IDE design.

A. Effect of IDE finger spacing

IDE electrodes with various spacing have been calculated and assessed. Figure 2 illustrates the capacitance changes over various IDE finger spacing. The experimental results reflect a strong influence of IDE finger spacing on the overall capacitance.

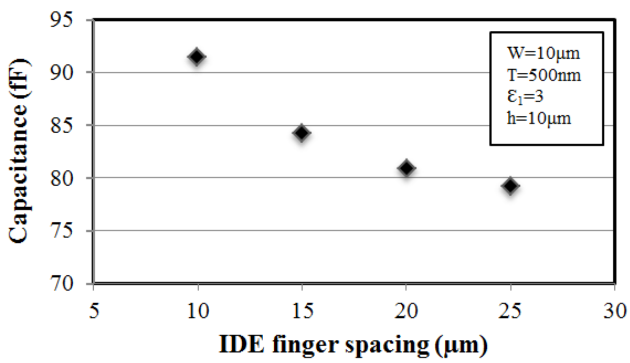


Fig.2. Capacitance change over various IDE finger spacing

B. Effect of IDE finger thickness

For the purpose of investigating the importance of finger thickness, it is essential to compute the capacitances of various thicknesses of IDE finger. As illustrated in Figure 3, the effect of the thickness of finger on the IDE capacitance is not significant, and there will be a minimal increase in the capacitance as a result of the increase in the fringing field capacitance.

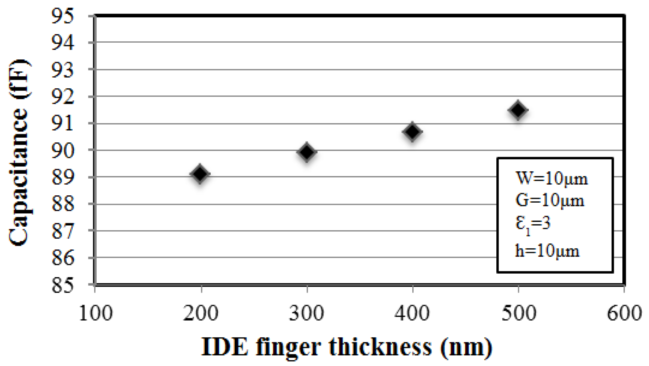


Fig.3. Capacitance change over various IDE finger thicknesses

C. Effect of IDE spatial wavelength (λ) and sensitive layer thickness (h)

The relevance among the overall IDE capacitance, sensitive layer thickness (*h*) and spatial wavelength (λ) is a significant factor in designing the IDE sensor. As described in Section 2, the layer adimensional thickness is $r=h/\lambda$. As depicted in Figure 4, the capacitance of IDE sensor has reached to an approximately fixed value for $r \geq 0.5$. This indicates that the IDE sensor is insensitive to the distance from the IDE plane larger than half of the spatial wavelength (λ).

According to Figure 4, the overall capacitance decreases by increasing the value of *r*. The reason behind this trend is that for material under test (MUT) relative permittivity larger than the sensitive layer relative permittivity, the difference of capacitance component which is in series with the capacitance of MUT has a more significant role.

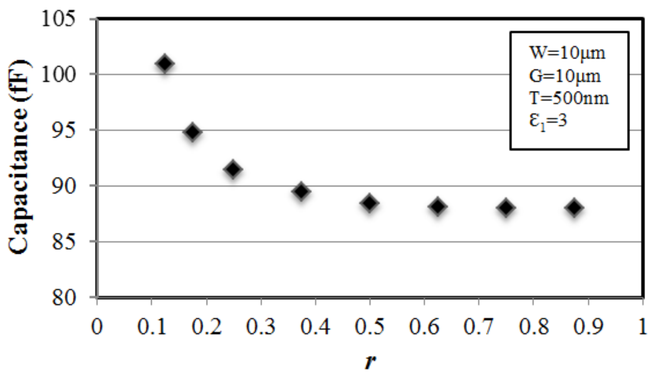


Fig.4. Capacitance as a function of the parameter *r*

Model Description

In this research, a 3D model of capacitive sensor LOC was designed and simulated using electrostatics mode of the MEMS module in COMSOL Multiphysics software to detect the E.coli bacteria in water. In each finite element, the following equations are used for determining the capacitance [20]:

$$-\nabla \varepsilon_0 \varepsilon_r \nabla V = \rho \quad (3)$$

$$\boldsymbol{D} = \varepsilon_0 \varepsilon_r \boldsymbol{E} \quad (4)$$

where ρ is the charge density, E is the electric field and D is the electric displacement. The energy needed for charging a capacitor is equal to that of the electrostatic field, and is demonstrated by:

$$W_e = \int_{\Omega} (\boldsymbol{D}.\boldsymbol{E}) d\Omega \quad (5)$$

The capacitance is calculated from the integral of the electric energy density by the following equation:

$$C = \frac{2}{V_p^2} \int_{\Omega} W_e \, d\Omega \quad (6)$$

where $V_p = 1V$ is the value of applied voltage in the port of the sensor.

In our model, the sensing electrodes fabricated from gold were formed on a silicon substrate with a relative permittivity of 11.9. A sensitive layer made of SU-8 with relative permittivity of 3 was deposited upon the electrodes. Then, the sensor was filled with water as the medium with relative permittivity of 80 [21, 22]. Figure 5 shows the capacitive sensor LOC simulated in COMSOL Multiphysics.

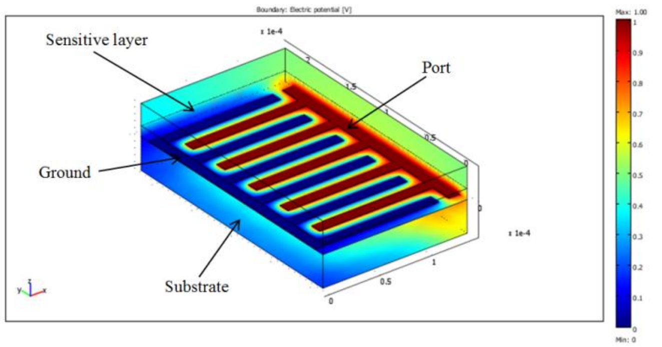


Fig.5. Model of capacitive sensor LOC

The E.coli bacteria were modeled as ellipsoids with a dimension of 2 µm long and 1 µm diameter [23]. Other parameters employed in the simulation of E.coli bacteria are listed in Table 1.

Table 1. Parameter values of E.coli [24]

Parameter	Value
Cell cytoplasm relative permittivity	60
Cell wall relative permittivity	60
Cell wall thickness	20 nm
Cell membrane relative permittivity	10
Cell membrane thickness	5 nm

Results of Capacitive Sensor LOC in the Presence of Bacteria

In this section, to indicate the presence of E.coli bacteria in water, the capacitance of water without bacteria was first measured as the reference value and then, the E.coli bacteria were introduced into the medium. In this case, a capacitance change occurred due to the change in dielectric property of the medium in proximity of sensing electrode.

The sensitivity of the model can be calculated by the following equation:

$$Sensitivity = \frac{C_2 - C_1}{C_1} \times 100 \qquad (7)$$

where C_1 is the capacitance of water without bacteria and C_2 is the capacitance of water with bacteria.

The simulation results showed that by increasing the number of bacteria in water, the capacitance decreased. This result is demonstrated in Figure 6.

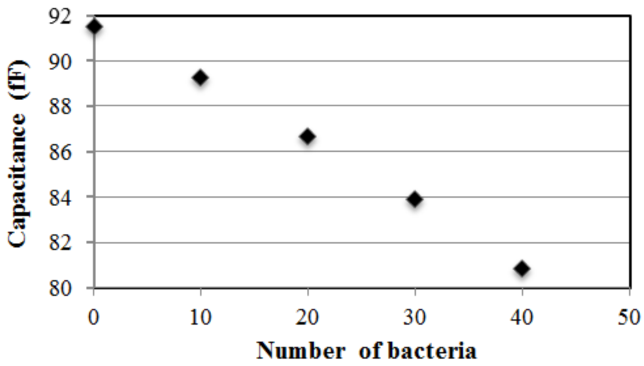


Fig.6. Relationship between the number of bacteria and capacitance change

The results in Figure 7 show the effect of increasing the number of bacteria on the sensor sensitivity. It can be clearly seen that the sensor was sensitive to the presence of bacteria (increase in the absolute value of sensitivity).

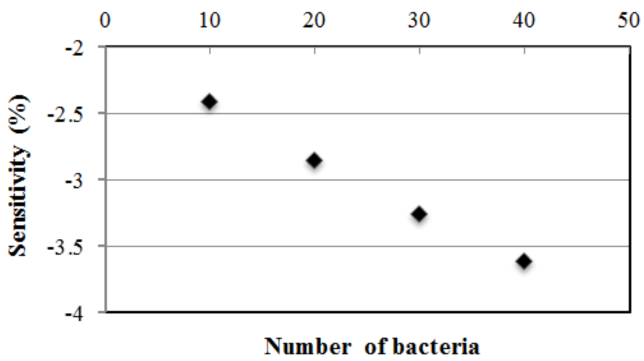


Fig.7. Effect of the number of bacteria on the sensor sensitivity

When the target bioparticles or bacteria were presented in the water-based medium, the capacitance change occurred. Since capacitance was directly proportional to relative permittivity, it was logical that when a sample with lower level of permittivity was introduced into the IDEs, which contained a suspension of higher dielectric permittivity, the capacitance would be reduced.

Conclusion

In the field of clinical diagnostics, fast, accurate, and low cost devices such as the LOC are the ideal alternative to conventional detection techniques especially for detecting bioparticles. In this paper, a capacitive sensor LOC was designed and simulated using COMSOL Multiphysics software. As shown, the electrode geometry is an important criterion for the IDE sensor design. The simulation results showed the capacitance changes in the absence and presence of bacteria in water and the relationship between the number of bacteria and capacitance change was also investigated. The application of the results obtained from this research is useful in commercializing the capacitive sensor LOC device as a future diagnostic tool to detect the E.coli bacteria.

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