Study of Two-Dimensional Open EWOD System using Printed Circuit Board Technology

By Vandana Jain, Vasavi Devarasetty & Rajendra Patrikar

Visvesvaraya National Institute of technology

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Keywords: digital microfluidics· electro wetting-on-dielectric (EWOD)· polydimethylsiloxane (pdms)· printed circuit board (PCB)· open source computer vision (OPENCV).

I. Introduction

Many micro fluidics tools are developed to control fluidics on the micro scale level. The first generation is continuous micro fluidics and the second generation is digital micro fluidics [1]. In continuous micro fluidics for liquid flow, we need external components like micro pumps and micro valves and also very difficult to manage many operations at a time [2]. Whereas in digital micro fluidics on a single chip can perform parallel various fluidic operations like dispense, transport, splitting, and merging [3], which offers advantages of portability, automation, higher sensitivity and high throughput in diagnosis applications [4]. This chip can perform clinical diagnostics for human physiological fluid [5], polymer chain reaction [6], proteomics [7] and glucose detection [8].

The manipulation of droplets in digital micro fluidic has been achieved using various techniques like temperature gradient [9], acoustic wave [10],

dielectrophoretic (DEP) [11], Op to-electro-wetting (OEW) [12], electro wetting (EW) and electro wetting on dielectric (EWOD) [13]. EWOD outwits all the other methods because of recon figure ability, flexibility, dynamic nature and signal processing ability using optical and electrical techniques. EWOD is essentially the phenomenon where the wetting behavior of a conductive droplet placed on a dielectric surface can be modified by application of electric field across the dielectric below the droplet [14]. The contact angle change is predominantly because of accumulation of charge carriers at the solid and liquid interface.

We have seen that most reported EWOD chips use a series of electrode pads essentially in a one-dimensional line pattern, designed for a specific task by using highly sophisticated lithography for electrode patterning [13] and expensive dielectric materials like Teflon-AF or paralene-C [15]. In our previous works, we have demonstrated low-cost one- dimensional EWOD chip using PCB (Printed Circuit Board) technology [16]. But for desired universal chips allowing reconfigurable user paths would require the electrode pads in a two-dimensional pattern [14,17]. Compared to conventional lithography technique, PCB technology allows high recon figure ability and reusability at lower manufacturing cost [18,19]. However, the PCB-PDMS based inexpensive approach for two-dimensional EWOD system fabrication is not studied much. Also very less cost effective EWOD systems are available for the continuous monitoring of droplet parameter with accuracy [20], which is essential for precise control and accurate manipulation of droplet for enhancing the system performance.

In this work, the development of a PCB-PDMS based two-dimensional open EWOD system with continuous monitoring of droplet parameter using open source computer vision (OpenCV) is discussed. The effect of different ground wire configuration on droplet velocity is investigated. The detection of milk adulteration is demonstrated by the mixing of two droplets on the device.

The rest of the paper is organized as follows; in section 2 experimental aspects of device fabrication is discussed. The measurement setup is discussed in section 3. Results obtained are presented in section 4 followed by conclusions in section 5.

Author: Visvesvaraya National Institute of Technology.
e-mail: vandy087@gmail.com

II. Device Fabrication

The proposed open EWOD device in this work has dimensions of 3 X 3 cm2 PCB, consists of 22 copper electrodes (2mm X 2mm) separated by 155μm gap. Each electrode pad is connected by eight control signals through a 160μm wide line. The PCB is designed in such a way that when one electrode is activated it will not affect the adjacent electrode. The pictorial view of PCB is shown in Fig. 1, where the same color regions are representing electrodes are activated by the same control signal, and black dot is a PTH (Plated through hole) hole which allows the electrode to connect from the bottom side of PCB. The physical design of PCB is shown in Fig. 2 in which backside PTH hole (300μm) is filled with soldering paste to avoid leakage through the hole. Then we have coated PCB with PDMS, which acts as both a dielectric as well as a hydrophobic layer using spin coater system. Note that the PDMS is a biocompatible polymer which has an average static contact angle of 1100, which is capable of easing the droplet operation [21]. After spin coating and testing of this device, we have found that water hydrolysis is occurred, which is creating a problem for the droplet motion. We have observed that this is happening due to the improper dielectric coating on the device because of copper thickness (35μm) and the PTH hole. So avoiding this we have coated device two times with different speed. In first time device is coated with 1500rpm and allowed to cure for 45mins at 100oC and second time device is coated with 2500 rpm and then cured at 100oC for 45mins. The PDMS coatings are removed from contact pads by gentle scraping with a scalpel to facilitate electrical contact for droplet actuation. The electrode pad contact is provided through the female connector.
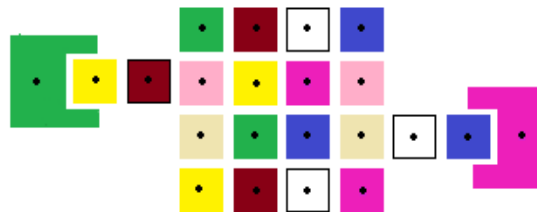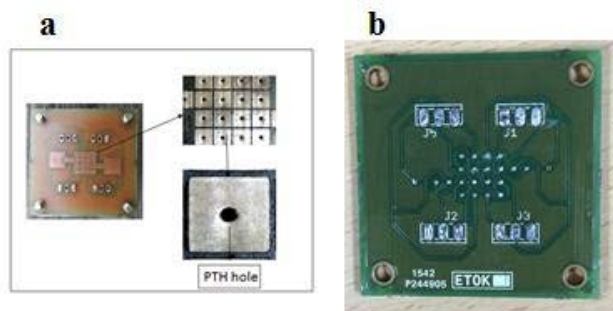

Fig. 1: Pictorial image of EWOD PCB

Fig. 2: Fabricated Device (a) Front view (b) Back view

For ground connection of droplet, we have used four aluminum wire catena (diameter 100μm) attached to separate PCB as shown in Fig. 3(a). The ground electrode wires are carefully arranged along the centerline of control electrode array on fabricated open EWOD device as shown in Fig. 3(b).
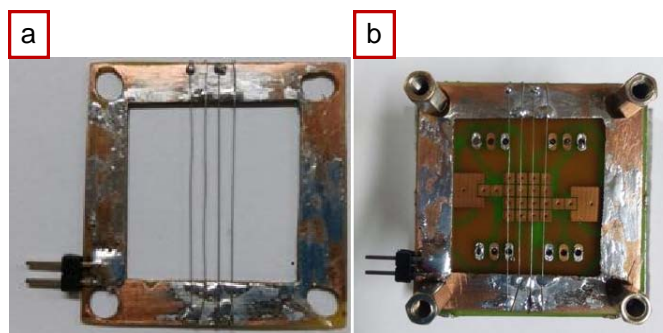

Fig. 3: (a) Catena arrangement on PCB (b) Final open EWOD device

III. System Description

A novel and precise droplet handling system is developed as shown in Fig. 4. A high voltage source is obtained from a DC boost converter. The application of high voltage is programmed by a microcontroller, which actuates a specific electrode through an electromagnetic relay. The complete setup consists of USB digital camera for capturing image/video in real time, XYZ stage for holding the sample, Arm7 based electronic system which has standard circuits for providing actuation of voltage and handling image processing along with HDMI display. The system is

designed to achieve precise real-time control over the movement of the droplet in real time embedded Linux platform. The C++ and python codes are developed in Qt software for precise control of droplet on the electrode. Ground and a positive voltage of power supply are connected respectively to the ground wire and the contact pads of the glass plate [16].

Fig. 4: Test setup

a) Image processing

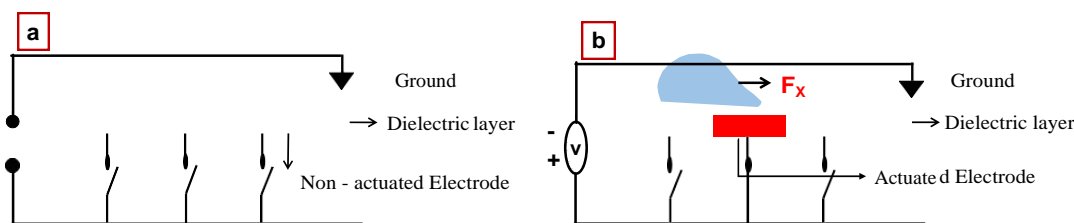The developed system is combined with integrated development environment (IDE) software with Open CV libraries for continuous mentoring of droplet velocity measurement and position detection. The functionalities of Open CV library are enhancing the image processing analysis. To characterize the developed DMF device, we have performed droplet transporting with velocity measurement in open EWOD system.

IV. RESULTS AND DISCUSSIONS

a) Droplet transport by EWOD

The droplet transport under electrostatic force is studied in the device. The pictorial view of droplet motion in the open configuration is shown in Fig. 5. The droplet motion occurs as a result of capillary force which sequels an apparent wettability gradient between actuated and non-actuated electrode. Using Lippmann-Young law, we can translate that electro wetting effect into a capillary effect. So the net capillary force or electro wetting force is rewritten in the following expression [22].

Where $\varepsilon 0$ is the permittivity of free space, εr is its relative dielectric constant, d is the thickness of the dielectric layer, V is the applied voltage, L is the effective contact line length. The contact line length L is determined by the boundary structure formation of the adjacent electrode.

Fig. 5: Pictorial representation of open EWOD operation (a) without voltage (yellow color indicate no actuation)

(b) With voltage (Red color indicate actuation)

The velocity of the droplet in the open EWOD device is derived for 1100 static contact angle of droplet by using [23].

$$U = \frac{\frac{\varepsilon_r \varepsilon_0 L}{d} \cdot V^2}{\pi K_1 K_2 K_c C_V \mu R \left(5.248 + \frac{\varepsilon_r \varepsilon_0 L}{d \gamma_{LG}} \cdot V^2\right)} \quad (2)$$

Where KC is the damp factor caused by the pinning effect in the triangle region, K1 is acceleration and deceleration time process factor, K2 is the considering dragging effect due to the ground wire, R is the radius of the droplet CV is an empirical consta the solid– liquid surface tension and μ is the viscosity of the fluid.

A 5μL DI water droplet with 0.1M KCL is placed on electrode pad with the top ground and transport is realized by sequentially energizing the adjacent electrode pad. But high pinning effects and sticky nature of PDMS, transportation of droplet from one pad to another pad is not observed. For getting a proper droplet motion very thin layer of silicone oil (350mPa.s) is spread on the PDMS coated device. We have noted that with and without oil film there is no change in initial contact angle of the droplet. On the other hand, oil film reduces the minimum electrical field required to move the droplet. Thus all the experiments reported in this paper are performed with PDMS layer covered with silicone oil film. The vertical and horizontal movement of droplet on different electrodes as shown in Fig. 6
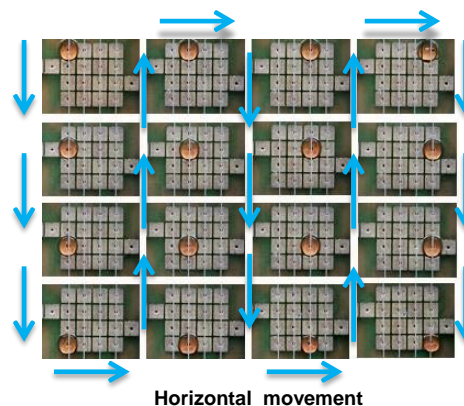

Horizontal movement

Fig. 6: Droplet transport vertically and horizontally on electrodes pads

b) Droplet position detection

The position of a droplet on electrode pads is monitored continuously using Open CV. Each frame of the live stream is correlated through image processing algorithm. Open CV libraries and IDE platform are combined using C++ codes. The written code, along with HSV values of the droplet tincture, plays a vital role in the detection of centroid pixel coordinate of a droplet through color thresholding as shown in Fig. 7(b). If the droplet centroid pixel coordinates lie within the respective limits of the coordinates of the electrode pads, the position of the droplet is printed on the terminal as shown in Fig. 7(c).
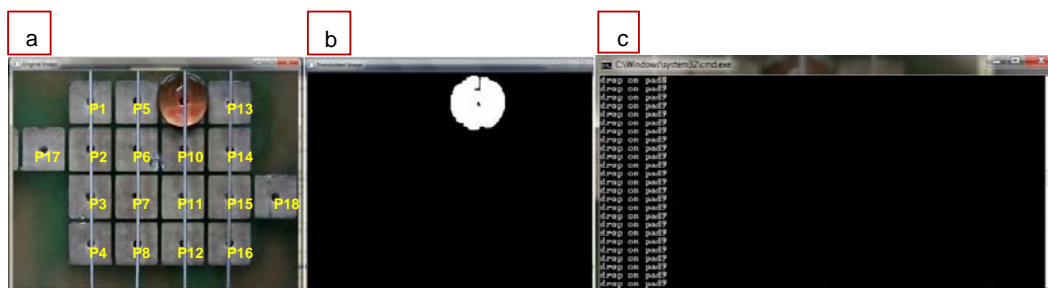
Fig. 7: Droplet position detection (a) Original image (b) Threshold image (c)) Terminal image with droplet position

c) Droplet velocity measurement

The velocity of the droplet is a vital parameter for various applications of LOC device and observing and controlling this key parameter in real time is one of the challenging tasks. In this work, we have successfully measured the droplet velocity between electrode pads in real time using the Open CV libraries. The timer count 1 starts when the centroid of droplet acquiesces with pad1 centroid and the timer count 2 starts when the centroid of the droplet coincides with pad 2 centroid. The difference between the two timer's times is measured. The difference between two centroid pads is calculated and multiplied by calibration factor for getting the actual distance in mm units. The droplet velocity is given as the ratio of distance to time; droplet velocity result is shown in Fig. 8b. Using this technique, we can track efficiently droplet parameter for multiple droplets. In our system image processing tools and high voltage control unit functionality works parallel, this makes EWOD system a smart system to perform various tasks on single platform simultaneously.

i. Droplet velocity measurement by varying the ground configuration

By using the developed velocity measurement system as described above, it becomes easy to analyze the velocity of the droplet at different voltages. In this section, we have investigated the effect of ground wire configuration on droplet velocity with respect to different voltages. In this paper we have taken three ground wire configurations namely; meshed, single line and diagonal which is shown in fig. 8.
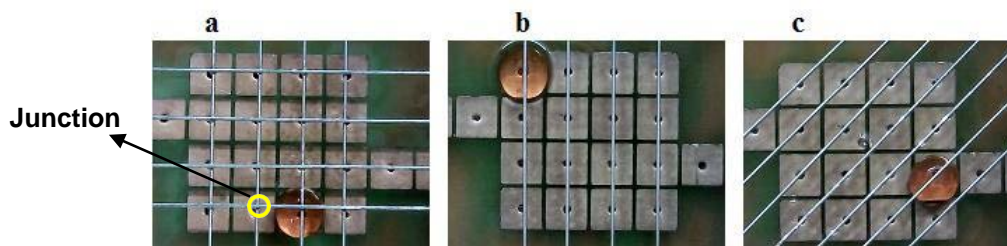

Fig. 8: Different ground configuration (a) Meshed (b) single line (c) Diagonal

The horizontal and vertical velocity of the droplet for the voltage range of 200V to 400 V at 155μm gap for all configurations is extensively studied in this work and summarized in Fig. 9. We have found that up to threshold voltage Vth, no droplet movement is observed. In these experiments, it is noted that up to 200V for all configuration, droplets are not moving. After that, any increment in the voltage beyond Vth, the significant movement of the drop, proportional to the applied voltage is noticed. To move the drop from its initial rest position sufficient electric field has to be built within the drop to reduce the interfacial energy [25,26]. The horizontal droplet velocity at 400V for meshed, single line and diagonal cases are 5.92mm/sec, 5.43mm/sec, and 5.18mm/sec respectively. The vertically droplet velocity at 400V for meshed, single line and diagonal cases are 5.1mm/sec, 5.43mm/sec, and 4.84mm/sec respectively.
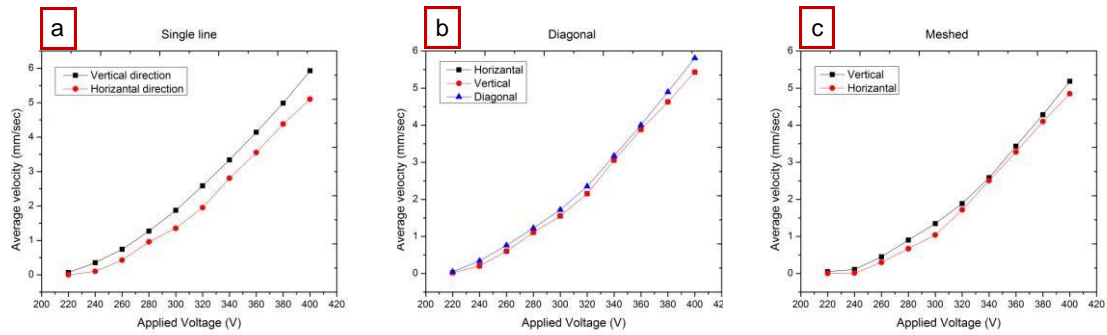
Fig. 9: Velocity of droplet (a) Meshed (b) Single line (c) Diagonal

We have observed that in meshed configuration velocity of the droplet is more in the vertical direction as compared with the horizontal direction; because vertical catena wires have been placed first then over that horizontal catena which makes the junction, and it is affecting the horizontal movement of the droplet shown in Fig. 9(a)

In the single ground line configuration, ground lines are arranged vertically as shown in Fig. 8(b). In this configuration, we have observed that velocity of the droplet is more in the vertical direction as compared with the horizontal direction because droplet gets proper grounding along the direction of the catena.

In the diagonal ground configuration, the ground lines are arranged diagonally as shown in Fig. 8 (c). In this configuration, we have observed the droplet velocity is same in both horizontal and vertical direction and more in a diagonal direction. In this configuration, we can transport the droplet in all direction.

We have compared the droplet velocity in all configurations for both horizontal and vertical directions as shown in Fig. 10. We have found that the horizontal droplet velocity is more in diagonal configuration and droplet vertical velocity is more in single line configuration.
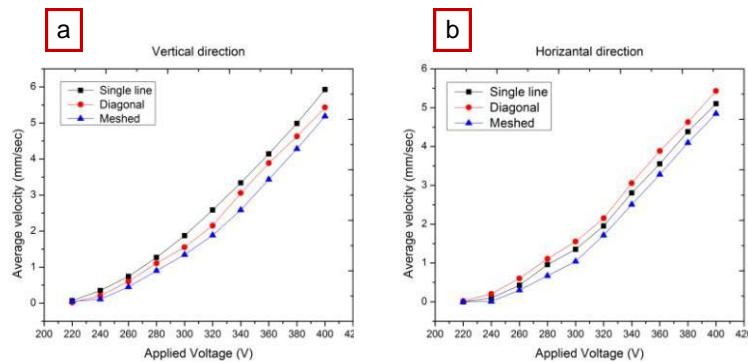

Fig. 10: Velocity of droplet (a) Horizontal direction (b) Vertical direction

To validate the experiment results with analytical results, we have plotted droplet velocity for vertical single line ground configuration with analytical value as shown in Fig. 11. We have calculated the analytical droplet velocity using Eq. 2 with adjusting the empirical parameter and taking K2 (4-6) into account. It is observed that the average droplet velocity is proportional to the square of the applied voltage (with R^2 is 0.99) which is in good agreement with the analytically calculated droplet velocity. The obtained relationship between droplet velocity and applied voltage is in good agreement with the analytical EWOD model [2,27] which relates the average velocity using Eq. 3
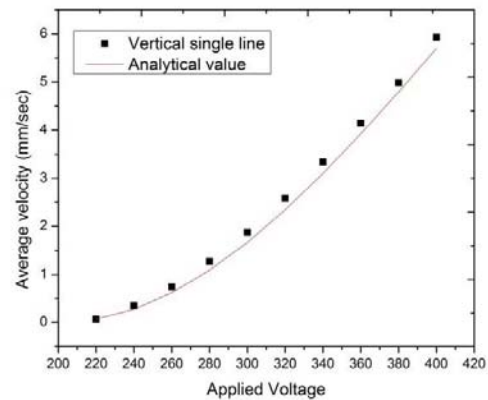

Fig .11: Comparison between experimental vertical single line and analytical value of droplet velocity

Also to validate the effect of ground wire configuration on droplet velocity; we have measured the maximum electric field in all configurations for both horizontal and vertical directions using COMSOL Multi physics. The estimation of the electric field is related to electro wetting force acting on the droplet. The electro wetting forces directly influence the droplet velocity.

Table 1: Maximum electric field for all configurations

S. No.	Configuration type	E_x (V/m)	E_y (V/m)
1	Meshed Ground	7.201×10^5	7.235×10^5
2	Single line Ground	7.24×10^5	7.327×10^5
3	Diagonal Ground	7.268×10^5	7.268×10^5

The resultant maximum electric field in horizontal and vertical directions for all configurations is given in Table 1. We have observed that single ground configuration is offering a more electric field in the vertical direction as compared to other configurations and a diagonal ground configuration having a more electric field in the horizontal direction as compared to other, which is well satisfied with our experimental results.

ii. *Droplet velocity with different liquids*

In this section, we have analyzed the effect of droplet viscosity on velocity. We have taken three different liquid sample; 0.1M KCL solution (1 Cps), 0.1M Potassium buffer (1.4-1.6 Cps) [28] and milk (3 Cps). The velocity comparison for different liquids is shown in Fig.12. It is observed that velocity of the water droplet is more compared to all. The experimental results are well in agreement with an analytical expression of droplet velocity given in Eq. 3, which shows droplet velocity is inversely proportional to the viscosity of the droplet.
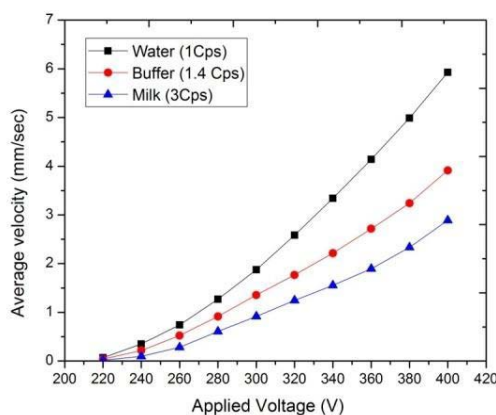

Fig. 12: The droplet velocity with different viscosity

d) *Detection of Milk adulteration*

One of the significant problems in the lab on chip area is merging and mixing of the two droplets dynamically using EWOD [29]. In this work we have demonstrated, mixing of two different droplets for one of milk adulteration application using two-dimensional open EWOD device which detects the starch existence in the milk. Starch is one such component that is added to adulterate milk for making milk fat [30]. We have used iodine solution for detection of starch in milk [31]. If starch content present in the milk sample, milk color becomes dark blue due to the formation of starch- iodo complex otherwise it turns into pale yellow color [32].

The 5μL milk droplet is first allowed to merge with a 1μl droplet of iodine solution on the open EWOD device by actuating the middle electrode between these droplets. Then coalesced droplet is repeatedly moved along the electrode pattern for proper mixing. After some movement on the different electrodes pads, it will turn into pale yellow color if there is no starch content present as shown in Fig. 13b. Otherwise, it turns into blue color as shown in Fig. 13c.
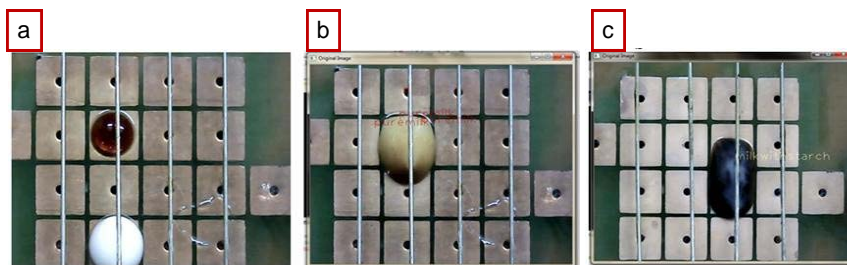
Fig. 13: Identification of pure milk a. before mixing b. after mixing along with result in Open CV

V. CONCLUSION

A low-cost reconfigurable open two-dimensional EWOD device has been demonstrated. A droplet transport is performed in both horizontal and vertical directions on the proposed device. The droplet position detection and droplet velocity measurement by varying the ground configuration are obtained, by using the open source computer vision (Open CV) platform. The electromagnetic simulations are done to measure the effect of electric field distribution by varying the ground configuration, which is well matching with experimental results. The testing of milk adulterations is successfully demonstrated. This work shows the potential for low-cost, rapid DMF device design and fabrication for future lab on chip applications.

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