# Lab on a Chip



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#### Introduction

The utilization of electrowetting-on-dielectric (EWOD)<sup>1,2</sup> to modulate liquid-solid contact angles has been widely used in creating, transporting, sorting, manipulating and mixing liquid droplets in many chemical and biological applications.<sup>3-7</sup> The majority of studies focus on the manipulation of aqueous droplets because of the rapid accumulation of charges at the interface between the aqueous drops and the solid surface<sup>8-10</sup> and the polarization of the dielectric layer.<sup>11</sup> Non-aqueous droplets, such as viscous oil drops, organic solvents and gas bubbles, however, are challenging to modulate by eletrowetting.<sup>12</sup> Although it has been demonstrated that water-in-oil compound drops sandwiched between two plates of an electrode can be manipulated in air by changing the contact angle of the aqueous inner drop (core) on a solid surface,<sup>13</sup> the viscosity of the oil shell should be small enough, *i.e.*  $\leq$  1 cSt, to ensure a low viscous drag force. When the oil phase to manipulate has high viscosity or contains hydrophobic solid particles, the entire drop needs to overcome a much higher viscous drag force because the aqueous core is surrounded by the oil shell, which is also in contact with the solid surface.<sup>14</sup> In addition, although drops of some watermiscible organic solvents, such as ethanol and dimethylformamide (DMSO), have been manipulated by EWOD with

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compound drops† Jiang Li,<sup>a</sup> Yixuan Wang,<sup>a</sup> Haosheng Chen<sup>\*b</sup> and Jiandi Wan<sup>c</sup>

**Electrowetting-on-dielectrics for manipulation** 

of oil drops and gas bubbles in aqueous-shell

We present the manipulation of oil, organic and gaseous chemicals by electrowetting-on-dielectric (EWOD) technology using agueous-shell compound drops. We demonstrate that the transport and coalescence of viscous oil drops, the reaction of bromine with styrene in benzene solution, and the reaction of red blood cells with carbon monoxide bubbles can be accomplished using this method.

> AC potentials applied to the electrodes, drops of some other water-immiscible organic solvents, such as toluene and decane, could not be moved under any conditions tested.<sup>12</sup> Therefore, control of the dynamics of non-aqueous drops using EWOD is still of great interest.

> In this work, we develop the EWOD technology for compound drops with aqueous shells and control the movement and reaction of oil, organic and gas cores encapsulated in aqueous shells. We provide an approach of EWOD for modulation of non-aqueous drops and expand the application of EWOD from aqueous drops to more complex fluids.

### Schematics of EWOD for aqueous-shell compound drops

The oil-in-water compound drops were generated in a microfluidic device, the schematic of which is shown in Fig. 1(a). A cylindrical glass capillary with an inner diameter of 0.3 mm was tapered by a pipette puller (P-97, Sutter Instrument Company) and was then fed into another tapered capillary with an inner diameter of 0.7 mm and an outer diameter of 1 mm. The two tapered capillaries were then inserted into a square glass capillary with an inner size of 1.05 mm. Air was injected into the outermost capillary as the outer phase, while the water phase was injected into the middle capillary as the middle phase, and the oil or gas phase was injected into the innermost capillary as the inner phase. By adjusting the flow rate ratios of the three phases, oil-in-water or gas-in-water compound drops separated by air bubbles were generated, and the size of the oil/gas cores was controlled. The video of the generation of the compound drops is available as Videos S1 and S2 in the ESI.†

Manipulation and coalescence of the compound drops using EWOD were performed on a dielectric substrate fabricated based on a standard process of printed circuit board (PCB).<sup>15</sup> The copper layer was etched into electrodes with a designed pattern on the substrate using the PCB process and

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<sup>†</sup> Electronic supplementary information (ESI) available: Video S1 shows the generation of aqueous-shell compound drops. Video S2 shows the compound drop on the EWOD substrate. Video S3 is a movie for Fig. 1(c) and 3. Video S4 is a movie for Fig. 2(b). Video S5 shows the manipulation of a compound drop with a viscous oil core of 100 mPa s viscosity, where the aqueous shell is dyed in blue. See DOI: 10.1039/c4lc00977k.



**Fig. 1** Schematics of EWOD for aqueous-shell compound drops. (a) Compound drops are generated in a microfluidic device. The collected drops can be moved on the EWOD substrate. (b) The fabrication process of the EWOD substrate with patterned electrodes. (c) The motion and the reaction of the oil-in-water compound drops controlled by EWOD (see Video S3 in the ESI†), where the cores of the compound drops can be oil, organic and gaseous chemicals and the shells are aqueous solutions.

was then coated with a thin layer of polydimethylsiloxane (PDMS, SYLGARD® 184 Silicone Elastomer Kit, Dow Corning) using a spin coater, where the weight ratio of the base to the crosslinker is 10:1. After heating at 70 °C for 72 hours, a 10  $\mu$ m thick PDMS layer was formed, and then it was further coated with a 100 nm layer of Teflon AF (DuPont Co.). The whole processing procedure of the EWOD substrate is shown in Fig. 1(b).

The electrodes formed a T-shaped rail for manipulating the aqueous-shell compound drops, as shown in Fig. 1(c). The two compound drops were first placed at each end of the lateral rail and then were driven to move toward each other under the software-controlled pulse sequences of the voltage applied to the electrodes. When they collided at the junction of the T-shaped rail, the two compound drops merged into one drop with two inner cores. The compound drop with two cores was then moved to the vertical rail. Under the pulse sequences applied to the diagonal electrodes occupied by the drop, the merged drop was stretched and deformed to cause the coalescence of the two cores.

#### Manipulation of viscous oil cores

The contact angles of an oil-in-water compound drop were measured under different voltages between 0 V and 500 V. The inner oil drop is liquid paraffin (Sinopharm Chemical Reagent Beijing Co.) with 0.1% ABIL® EM90 (Evonik Industries), which is dyed red, and the aqueous shell is 8% polyvinyl alcohol (PVA, Sigma-Aldrich) solution. We found that the contact angles of the compound drop decreased from 93° to 57° with the increase in the applied voltage from 0 V to 500 V, and the contact angle returned to 78° when the voltage was decreased to 0 V, as shown in Fig. 2(a1). The returned contact angle was smaller than its initial contact angle, which is attributed to the occurrence of trapping of charge in or on the insulating layer under such a high voltage.<sup>10</sup> The oil core in the compound drop did not affect the contact angles of the aqueous shell under different voltages, because the oil core had a lower density, such that it floats in the aqueous shell without contacting the solid surface, as shown in Fig. 2(a2)-(a3). Thus, the aqueous shell can be used as the carrier to move viscous oil cores on the electrodes, as shown in Fig. 2(b1)–(b3), where the viscosities of the oil phase and the water phase are 29.0 mPa s and 16.6 mPa s, respectively. The oil cores moved with the compound drops and finally coalesced on the EWOD substrate under the controlled voltage pulses on the patterned electrodes.

In addition, oil cores with higher viscosity were also successfully manipulated by EWOD of the compound drops, for example, the oil core in the compound drop was silicone oil with a viscosity of 100 mPa s (Sigma-Aldrich), as shown in Video S5 in the ESI.<sup>†</sup> The viscosity of the oil cores will not increase the friction force between the drop and the substrate of the EWOD board. In contrast, an individual oil drop and the water-in-oil compound drops were not moved by EWOD. The case of an oil drop of liquid paraffin, dyed in



**Fig. 2** Manipulation of aqueous-shell compound drops. (a1) Variation of the contact angles of an oil-in-water compound drop. The square (blue) and the circle (red) symbols represent the measured data when the applied voltage is increased and decreased, respectively. (a2) and (a3) Images of the contact angle of the compound drop at voltages of 0 V and 500 V, respectively. (b1)–(b3) show the movement and coalescence of two oil-in-water compound drops driven by EWOD; the corresponding video is available in the ESI† (Video S4). (c1) and (c2) show a single oil drop of liquid paraffin that was not moved by EWOD; (d1) and (d2) show a water-in-oil compound drop that was not moved by EWOD, where the oil shell is silicone oil with a viscosity of 100 mPa s and the aqueous core is dyed in blue. All of the scale bars are 2 mm.

red, is shown in Fig. 2(c1)–(c2), and the case of a water-in-oil compound drop is shown in Fig. 2(d1)–(d2), where the inner drop is 8% PVA solution, dyed in blue, and the outer oil phase is silicone oil with a viscosity of 100 mPa s.

It should be noted that the core-shell structure of the compound drop may break when the applied voltage to the control electrodes is higher than a specific value. For example, the oil-in-water compound drop in Fig. 2(a) broke when the voltage was increased to 600 V, and the oil core flew out of the aqueous shell and became a thin layer coating outside the aqueous drop.

#### Reaction of organic cores

The bromination of an alkene<sup>16</sup> is demonstrated here to show the application of EWOD to compound drops with organic cores. Two compound drops with different organic



**Fig. 3** Manipulation of two compound drops with organic cores. (a1)–(a3) The two drops were driven to collide and merged into one drop with two inner cores. The yellow core in the left drop is the solution of 0.2 M bromine in benzene, while the grey core in the right drop is the solution of 0.85 M styrene in benzene. (a4)–(a6) The stretch of the compound drop under the voltage pulses applied to the electrodes, and the two inner cores that have finally coalesced. Scale bars in the images are 2 mm. The video of the coalescence process is available in the ESI† (Video S3).

cores in the aqueous shells of 8% PVA solution were manipulated in the experiment. One has a yellow core of the solution of 0.2 M bromine in benzene, while the other has a grey core of the solution of 0.85 M styrene in benzene. The two compound drops were placed on each end of the T-shaped rail and then manipulated to collide. The aqueous shells fused first with the two individual cores inside, as shown in Fig. 3(a1)-(a3). To induce the coalescence of the two organic cores, the fused compound drop was moved to the four electrodes on the vertical rail, and then the voltage pulses were applied periodically to the two pairs of the diagonal electrodes to stretch and relax the compound drop in the orthogonal directions. The stretch-to-relax motion of the compound drop causes a compression-to-separation motion of the two organic cores, which helps the coalescence of the two cores.<sup>17,18</sup> After the periodical stretches, the two cores inside the compound drop coalesced, as shown in Fig. 3(a4)–(a6). The colour of the coalesced core changed from yellow to clear, indicating the completion of the reaction. In addition, the two toxic organic cores and the products of the reaction were all encapsulated in the aqueous shells, which reduced the cross contamination between the products and the environment.

## Reaction of gas cores

Gaseous chemicals in the cores of compound drops with aqueous shells can also be manipulated by EWOD and react with other chemicals. Here we demonstrate the reaction between red blood cells (RBCs) and carbon monoxide (CO) bubbles and then evaluate the CO poisoning of the RBCs using the sodium hydroxide (NaOH) test.<sup>19</sup>

A drop of whole blood from a healthy adult was washed with phosphate buffered saline (PBS) and then centrifuged for the separation of RBCs.<sup>20</sup> The resulting RBCs were diluted to 10% in physiological salt solution (PSS). For comparison, two drops of the diluted RBCs were driven to coalesce with a simple drop of 8% PVA aqueous solution (Fig. 4(a1)-(a2))



**Fig. 4** Reaction of RBCs with a CO bubble and its evaluation using EWOD. Sodium hydroxide tests of RBCs (a1)-(a7) without exposure to CO bubbles and (b1)-(b8) after reaction with a CO bubble. The red drops on the left in (a1) and (b1) are the diluted RBC drops; the transparent drop on the right in (a1) is 8% PVA aqueous solution; the gas-in-water compound drop on the right in (b1) is a CO bubble in 8% PVA aqueous solution; the transparent drops in (a3) and (b3) are 1 M NaOH solution. Microscopic images of (c) normal RBCs with red color and (d) with straw color after the reaction with NaOH. The scale bars for (a1)-(a7) and (b1)-(b8) are 2 mm, while those for (c) and (d) are 20 μm.

and a gas-in-water compound drop with a CO bubble in 8% PVA aqueous solution (Fig. 4(b1)–(b2)). The compound drop was generated using the same method presented in Fig. 1. The two coalesced drops were left on the substrate for 150 s before another drop of 1 M NaOH was added (Fig. 4(a3)–(a4) and (b3)–(b4)). Consequently, in the compound drop (Fig. 4(b4)), CO bound to haemoglobin (Hb) in the RBCs, instead of oxygen, and formed carboxyhemoglobin (COHb), which caused the CO poisoning of the RBCs.<sup>21</sup>

After the drop of normal RBCs was mixed with the drop of NaOH solution (Fig. 4(a5)–(a7)), the initial red color (Fig. 4(a4) and (c)) turned to straw color quickly (Fig. 4(a5)–(a7) and (d)). For example, the color change was obviously observed within 4 s (Fig. 4(a5)) and the color of the entire drop changed within 76 s (Fig. 4(a6)). In contrast, for the CO-poisoned RBCs, the compound drop maintained the red color with a slight color change at the edge within 76 s (Fig. 4(b6)) and the color change of the entire drop (Fig. 4(b6)) and the color change of the entire drop (Fig. 4(b8)) was observed 330 s later after the reaction with NaOH. The significantly delayed color change of the compound drop (Fig. 4(b5)–(b8)) due to the COHb-saturated RBCs<sup>19</sup> indicated that the reaction of the RBCs and the CO gas was effectively realized by EWOD of the compound drop.

# Conclusions

The oil, organic and gaseous chemicals in the aqueous shell can be manipulated by EWOD. This EWOD technology for aqueous-shell compound drops was demonstrated to realize the transportation of high viscous oil drops, the mixture of organic chemicals, and the reaction with bubbles, which are usually difficult to realize by the traditional EWOD. Thus, the EWOD can be used for more complex fluids besides water drops and it would be a helpful tool in droplet microfluidics. Additionally, the aqueous shell also provides protection preventing the reaction products that are confined in the drops from releasing into the surrounding environment.

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# Notes and references

- 1 J. Lee, H. Moon, J. Fowler, T. Schoellhammer and C. J. Kim, *Sens. Actuators, A*, 2002, **95**, 259.
- 2 S. K. Cho, H. Moon and C. J. Kim, *J. Microelectromech. Syst.*, 2003, 12, 70.
- 3 S. Y. Teh, R. Lin, L. H. Hung and A. P. Lee, *Lab Chip*, 2008, 8, 198.
- 4 B. Berge and J. Peseux, Eur. Phys. J. E, 2000, 3, 159.
- 5 R. Sista, Z. Hua, P. Thwar, A. Sudarsan, V. Srinivasan, A. Eckhardt, M. Pollack and V. Pamula, *Lab Chip*, 2008, 8, 2091.
- 6 A. H. C. Ng, K. Choi, R. P. Luoma, J. M. Robinson and A. R. Wheeler, *Anal. Chem.*, 2012, 84, 8805.
- 7 H. H. Shen, S. K. Fan, C. J. Kim and D. J. Yao, *Microfluid. Nanofluid.*, 2014, 16, 965.
- 8 M. Vallet, B. Berge and L. Vovelle, Polymer, 1996, 37, 2465.
- 9 M. Vallet, M. Vallade and B. Berge, *Eur. Phys. J. B*, 1999, 11, 583.
- 10 H. J. J. Verheijen and M. W. J. Prins, *Langmuir*, 1999, 15, 6616.
- 11 V. Peykov, A. Quinn and J. Ralston, *Colloid Polym. Sci.*, 2000, 278, 789.
- 12 D. Chatterjee, B. Hetayothin, A. R. Wheeler, D. J. King and R. L. Garrell, *Lab Chip*, 2006, 6, 199–206.
- 13 D. Brassard, L. Malic, F. Normandin, M. Tabrizian and T. Veres, *Lab Chip*, 2008, 8, 1342–1349.
- 14 H. Ren, R. B. Fair, M. G. Pollack and E. J. Shaughnessy, *Sens. Actuators, B*, 2002, **87**, 201–206.
- 15 J. Li, Y. Wang, E. Dong and H. Chen, *Lab Chip*, 2014, 14, 860–864.
- 16 Z. T. Gygan, J. T. Casbral, K. L. Beers and E. J. Amis, *Langmuir*, 2005, 21, 3629–3634.
- 17 N. Bremond, A. R. Thiam and J. Bibette, *Phys. Rev. Lett.*, 2008, **100**, 024501.
- 18 H. Chen, Y. Zhao, J. Li, M. Guo, J. Wan, D. A. Weitz and H. A. Stone, *Lab Chip*, 2011, 11, 2312.
- 19 S. Kaye, Handbook of Emergency Toxicology: A Guide for the Indentification, Diagnosis and Treatment of Poisoning, Charles C. Thomas, Springfield, IL, 5th edn, 1988, pp. 250–252.
- 20 J. Wan, W. D. Ristenpart and H. A. Stone, *Proc. Natl. Acad. Sci. U. S. A.*, 2008, 105, 16432.
- 21 M. L. Bishop, E. P. Fody and L. E. Schoeff, *Clinical Chemistry: Principles, Techniques, and Correlations*, Lippincott Williams & Wilkins, Philadelphia, 2013, pp. 651–652.