Manipulation of micro-objects using acoustically oscillating bubbles based on the gas permeability of PDMS

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This paper presents a novel manipulation method for micro-objects using acoustically oscillating bubbles with a controllable position based on the gas permeability of polydimethylsiloxane. The oscillating bubble trapped within the side channel attracts the neighboring micro-objects, and the position of the air-liquid interface is controlled by generating temporary pressure difference between the side channel and the air channel. To demonstrate the feasibility of the method in technological applications, polystyrene microparticles of 10 μm in diameter were successfully captured, transported, and released. The influence of pressure difference on the movement speed of the air-liquid interface was demonstrated in our experiments, and the manipulation performance was also characterized by varying the frequency of the acoustic excitation and the pressure difference. Since the bubble generation and the air-liquid interface movement in our manipulation method do not need any electrochemical reaction and any high temperature, this on-chip manipulation method provides a controllable, efficient, and noninvasive tool for handling micro-objects such as particles, cells, and other entities. The whole manipulation process, including capturing, transporting, and releasing of particles, spent less than 1 min. It can be used to select the cells and particles in the microfluidic device or change the cell culture medium. *Published by AIP Publishing.* <https://doi.org/10.1063/1.5028419>

I. INTRODUCTION

The manipulation of micro-objects such as cells, particles, and organisms in microfluidic environments is a significant issue in many biomedical applications.^{1–3} Diverse methods for capturing, transporting, rotating, and collecting of micro-objects have been developed based on magnetism,^{4–6} dielectrophoretic,^{7,8} optics,^{9–11} electrowetting,¹² acoustics,^{13–15} and hydrodynamics.¹⁶ Among the various available techniques, acoustically oscillating bubbles have been widely applied to microfluidic applications.^{17–21} When acoustic waves are applied to a bubble in a fluid, the neighbouring micro-objects around the bubble are captured by means of an acoustic radiation force generated by the oscillating bubble.²² It is found that this force is strong enough to hold the micro-objects when the bubble is being transported.²³ The technique based on oscillating bubbles can carry, transfer, and manipulate micro-objects without direct contact.

During the past few years, there have been many studies on the manipulation of micro-objects in microfluidic devices based on acoustically oscillating bubbles. Sang presented a manipulation method using oscillating bubbles sitting on a solid surface transported by

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electrowetting-on-dielectric actuation.²⁴ The directional movement of the bubbles can be achieved by sequential activation of the electrodes. Sang also designed 3-D manipulations of objects using an acoustically excited oscillating bubble deposited on a hydrophobic rod tip.²⁵ The captured objects can be manipulated in a 3-D space by controlling a traversing system connected to the rod. Kyung developed a micromanipulation system using acoustically oscillating identical twin bubbles,²⁶ in which an electrolytic method was used to generate bubbles by controlling the applied voltage and the time. Jae developed a hybrid technique for the manipulation of micro-objects by incorporating an optothermally and acoustically excited bubble.²⁷ The bubble is generated by a laser beam due to optothermal effects, and the particles can be delivered to a preferred position under both an optothermocapillary flow and acoustic radiation effects. However, the generation or the movement of the bubble is more complex in the above studies. A method for controlling bubbles' size by pressure difference through a polydimethylsiloxane (PDMS) membrane was presented by Geng *et al.*²⁸ The pressure difference was controlled by adjusting the liquid level in the out tube.

This paper presents an on-chip manipulation method for micro-objects using acoustically oscillating bubbles with a controllable position based on the gas permeability of polydimethylsiloxane (PDMS). When acoustic waves are applied to the bubbles trapped within the side channels, the oscillating air-liquid interfaces can capture particles due to the acoustic radiation force. The position of the meniscus and the volume of the bubble carrying the micro-objects are controlled by generating a temporary pressure difference between the side channel and the air channel based on the gas permeability of PDMS. The bubble generation and the meniscus movement in our manipulation method do not need electrochemical reaction and high temperature, which can avoid some potential damages to cells and solutions. The manipulation method can be applied to the cell culture device, which can transitorily store cells in the side channel to prevent them from being washed away in the replacement of cell culture medium. It can also be used to select cells and particles in microfluidic devices.

II. MATERIALS AND METHODS

A. Microfluidic device design and fabrication

Figure 1 schematically shows the design of our microfluidic device. An inlet, a main channel, an outlet, an air port, and an array of side channels, semi-circular chambers, and air channels are designed in the PDMS chip. The depth of all the channels is 100 μm , the width of the side channels and the air channels is 100 μm , the length of the side channels is 2 mm, and the width of the main channel is 300 μm . Once the main channel is filled with solution, an air pocket remains trapped in one side channel forming an air-liquid interface. As shown in Fig. 1(b), the air port is connected to the atmosphere, a vacuum tank, and a compressed air through three valves by Teflon tubes, respectively. The juncture of the side channel and the air channel is shown in Fig. 1(c). When the air port is connected to different pressure sources, it can generate pressure difference between the side channel and the air channel, which makes the air diffuse from the side channel into the air channel or reverse through the PDMS wall based on the gas permeability of PDMS. Accordingly, the air volume in the bubble and the position of the air-liquid interface is then controlled by changing the pressure in the air channel.

The PDMS microfluidic device was fabricated using standard softlithography techniques.²⁹ Briefly, a negative master was fabricated on a silicon wafer with SU-8 photoresist (SU-8 2050; Micro-Chem Corp., Newton, MA, USA) using a transparency mask and a mask aligner (BGT-3B; Beijing Chuangweina Technology Co. Ltd., China). A PDMS mixture (Sylgard 184; Dow Corning) of 10:1 ratio of base to curing agent was thoroughly mixed and degassed in a vacuum chamber before being poured onto the SU-8 master. After baking at 65 °C for 2 h, the PDMS was peeled off from the mould and three holes with a diameter of 1.5 mm were punched for connection with tubes. The PDMS chip and the glass substrate were irreversibly bonded after being treated with oxygen plasma. The whole device was placed on a hot plate for 24 h at 70 °C to improve the bonding strength and stabilize the surface property of the device. Then, a piezoelectric transducer (YZ-20T-6.2A1; Yaoze Electronics) was attached to the glass substrate

FIG. 1. Schematic of the microfluidic system. (a) Schematic of the microfluidic device; (b) schematic of the connection of air port; and (c) the magnified view of an air diffusion region of the PDMS chip.

adjacent to the PDMS chip using a thin layer of epoxy (Gleihow 568; Gleihow New Materials Co. Ltd., China).

B. Principle of particle manipulation

When acoustic waves are applied to the bubble in the fluid, the oscillating bubble can radiate a secondary sound field and generate a streaming flow. The particles around the bubble are subjected to two forces: the acoustic radiation force from the sound field and a drag force by the streaming flow.

The acoustic radiation force F_r on a particle in the proximity of the oscillating bubble is expressed as below,³⁰

$$F_r = 4\pi u_o^2 \frac{R_b^4 R_p^3}{r^5} \frac{\rho_p - \rho_m}{2\rho_p + \rho_m} \rho_m, \quad (1)$$

where R_b is the radius of the bubble, R_p is the radius of the particle, ρ_p is the density of the particle, ρ_m is the density of the fluid, u_o is the surface velocity of the bubble, and r is the distance between the bubble and particle.

At a low Reynolds number, the drag force F_d originating from acoustic microstreaming can be estimated by Stokes' drag

$$F_d = 6\pi\eta R_p U_L, \quad (2)$$

where η is the viscosity of fluid, and U_L is the velocity of the particle relative to the ambient fluid.

The actual motion of particles depends on the relative magnitude of F_r and F_d . The ratio of F_r to F_d is expressed as below

$$\frac{F_r}{F_d} = \frac{4}{3} \pi \frac{f R_p^2}{\eta} \frac{\rho_p - \rho_m}{2\rho_p + \rho_m} \rho_m, \quad (3)$$

where f is the oscillating frequency of the bubble.

The working principle of particle manipulation using the oscillating bubbles with a controllable position is illustrated in Fig. 2. When the solution is introduced into the main channel, some air is trapped in the side channel to form an air bubble protruding into the main channel due to the hydrophobic–hydrophilic interaction between the PDMS and the fluid [Fig. 2(a)]. A piezoelectric transducer is used to acoustically oscillate the air-liquid interface to capture the neighbouring particles [Fig. 2(b)]. A negative pressure source is connected with the air port when one turns V3 on and turns V1 and V2 off, and the air trapped in the side channel will diffuse into the air channel through the thin PDMS wall. The volume of the bubble will decrease, and the air-liquid interface carrying particles will move toward the air channel along the side channel. When the air-liquid interface carrying the particles moves to a satisfied position, the air port is connected with the atmosphere by shifting the switch status of valves, that is, V1 is turned on, V2 and V3 are turned off. Due to the main channel and the air channel having the same pressure (atmosphere pressure), the air-liquid interface can stay at the specified position in the side channel [Fig. 2(c)]. Then, another solution is introduced into the main channel, which washes away the previous solution in the main channel. Meanwhile, the particles captured by the air-liquid interface can be stored in the side channel [Fig. 2(d)]. When a positive pressure source is connected with the air port as one turns V2 on and turns V1 and V3 off, the air inside the air channel will diffuse into the side channel through the thin PDMS wall. The volume of the bubble will increase, and the air-liquid interface will move toward the main channel. After the interface goes back to the main channel, the atmosphere is connected with the air port and the captured particles can be easily released from the interface by turning off the acoustic excitation [Fig. 2(e)]. A series of the operations (capturing, transporting, changing solutions, and releasing of particles) can be accomplished with the microfluidic device. The particle manipulation can also be used to select cells and particles in microfluidic devices.

III. RESULTS AND DISCUSSION

A. Experimental characterization of particle manipulation

A successful manipulation of particle is shown in Fig. 3. In the experiments, three solenoid valves (DELIXI 2V025–06) were adopted to control the pressure in the air port. The on/off algorithm of the valves was controlled by the programmable controller (MITSUBISHI,

FIG. 2. Schematic of the device's working process. (a) Particles flow in the main channel with its solution. (b) When the piezoelectric transducer is turned on, the oscillating air-liquid interface captures particles. Then, the air port is connected with a negative pressure source, and the air-liquid interface moves toward the air channel. (c) When the air port is connected with the atmosphere, the interface stops at the specified position. (d) The previous solution is washed away with another solution, and the captured particles are stored in the side channel. (e) When the air port is connected with a positive pressure source, the interface moves toward the main channel. The particles are released after the excitation is turned off.

MELSEC FX2N-48MT). Polystyrene microparticles (Dantec Dynamics A/S) with a diameter of $10\text{ }\mu\text{m}$ are chosen to suspend in the DI water. The surfactant Tween 20 (Qingdao Uself Co. Ltd., China) is added at 1% (w/w) to the solution to prevent the sticking of particles to the microchannel walls and avoid particles aggregation.³¹ After the solution was injected into the main channel with a syringe pump (SPLab01; Shenzhen, China) at a constant flow rate of $0.3\text{ }\mu\text{l/min}$, an air bubble was trapped in the side channel creating an air-liquid interface [Fig. 3(a) (multimedia view)]. The piezoelectric transducer, activated by a high frequency pulse generator (SP1631A), is used to acoustically oscillate the air-liquid interface. The acoustic wave was then transmitted to the air-liquid interface, and the neighbouring particle (circled in red) was captured around the oscillating interface [Fig. 3(b) (multimedia view)]. The resonant frequency of the oscillating air-liquid interface can be estimated using the Rayleigh–Plesset equation²⁶ as below,

$$f = \frac{1}{2\pi} \sqrt{\frac{1}{\rho_m R_b^2} \left[3\kappa \left(P_0 + \frac{2\sigma}{R_b} \right) - \frac{2\sigma}{R_b} \right]}, \quad (4)$$

where κ is the polytropic exponent (1.4), P_0 is the hydrostatic pressure ($101,325\text{ N/m}^2$), σ is the liquid's surface tension (0.0728 N/m), ρ_m is the density of solution (1001 kg/m^3), and R_b is the radius of air-liquid interface. From the image of the air bubble, we measured the radius of curvature of the air-liquid interface to be around $70\text{ }\mu\text{m}$. Experimentally, the optimized excitation frequency (42 kHz) to capture particle was determined by sweeping the excitation

FIG. 3. Sequential pictures showing that an oscillating air-liquid interface captures, transports, and releases the neighbouring particle. (a) Initial state; (b) the air-liquid interface captures a particle when the acoustic excitation (at 42 kHz , 8 V) turns on; (c) the oscillating interface carrying the particle stops at the specified position; and (d) the interface pushes the particle back to the main channel and releases the particle when the excitation is turned off. Multimedia view: <https://doi.org/10.1063/1.5028419.1>

frequency with a 100 Hz increment near the theoretically calculated resonance frequency (45 kHz). The discrepancy between the experimental and the calculated resonance frequency can be attributed to the non-spherical shape of the bubble trapped within the side channel since a spherical shape was assumed for theoretical calculation.³² When the vacuum tank was connected to the air port, the oscillating air-liquid interface transported along the side channel because of the gas permeability of PDMS. It was characterized that the captured particle was retained around the interface in transportation. Once the atmosphere was connected with the air port, the pressure difference between the side channel and the air channel disappeared and the air-liquid interface stayed at the specified position in the side channel [Fig. 3(c) (multimedia view)]. When the compressed air was connected to the air port, the air-liquid interface in the side channel moved toward the main channel. Finally, after the interface reached the main channel, the atmosphere was connected to the air port again and the particle was released from the interface by removing the acoustical excitation [Fig. 3(d) (multimedia view)]. The whole manipulation process, including capturing, transporting, and releasing the particles, spent less than 1 min.

The flow rate of the fluid in the main channel was $0.3 \mu\text{l}/\text{min}$, and the speed of the particle was about $400 \mu\text{m}/\text{s}$. In our experiments, the air-liquid interface can capture particles with a distance of about $40 \mu\text{m}$. If the fluid in the main channel keep still and if there are many particles uniformly distributed in the fluid, the particles can be captured when the distance between the air-liquid interface and the particle is less than $40 \mu\text{m}$.

Supposing that the relative magnitude of F_r and F_d is equal, the critical excitation frequency can be calculated with Eq. (3). In our experiments, the density of polystyrene particles ρ_p is $1050 \text{ kg}/\text{m}^3$ and the viscosity of solution η is $0.723 \times 10^{-3} \text{ Pa}\cdot\text{s}$. The critical excitation frequency for the particle with a diameter of $10 \mu\text{m}$ is about 43.6 kHz. For an excitation frequency of greater than 43.6 kHz, the acoustic radiation force dominates over the drag force and the particles can be attracted around the air-liquid interface. On the contrary, for an excitation frequency of less than 43.6 kHz, the drag force dominates the system and the particles will follow the streaming pattern. The optimized excitation frequency in the experiments was 42 kHz, which is similar to the critical excitation frequency with theoretical calculation.

In our experiments, the manipulation of capturing, transporting, and releasing for particles with different size (with a diameter of $5 \mu\text{m}$, $10 \mu\text{m}$, and $20 \mu\text{m}$) were realized by adjusting the excitation frequency. As shown in Fig. 4, polystyrene microparticles with a diameter of $20 \mu\text{m}$ were captured and transported at the excitation frequency of 31 kHz. Multiple particles can also be handled at the same time. We have ever observed that more than 5 particles were captured and transported by the air-liquid interface at the same time in our experiments.

FIG. 4. Sequential pictures showing that an oscillating air-liquid interface captures and transports the neighbouring particles. (a) The air-liquid interface captures particles with a diameter of $20 \mu\text{m}$ when the acoustic excitation frequency is 31 kHz and (b) the oscillating interface carrying the particles stops inside the side channel.

FIG. 5. A comparison between the theoretical speed and experimental speed of the air–liquid interface. (a) The movement speed of interfaces with different thicknesses of the PDMS wall at various pressure differences. The diffusion area was 0.3 mm². (b) The movement speed of interfaces with different diffusion area at various pressure differences. The thickness of the PDMS wall was 100 μm.

B. Characterization of the movement speed of the interface

PDMS is one of the polymeric materials with certain gas permeation. For the steady-state gas flux through the PDMS wall, the flow rate is calculated by³³

$$\frac{dV}{dt} = \frac{PA\Delta p}{\delta} \frac{T}{273 p_{atm}}, \quad (5)$$

where $\frac{dV}{dt}$ is the volumetric flow rate, P is the permeability coefficient of PDMS wall for air ($480 \times 10^{-10} \text{ cm}^3(\text{STP}) \text{ cm/cm}^2 \cdot \text{s cmHg}$), A is the diffusion area, Δp is the pressure difference on both of the PDMS wall, δ is the thickness of the PDMS wall, T is the absolute temperature in kelvin, p_{atm} is the atmospheric pressure.

In order to obtain the movement speed of the air–liquid interface in the side channel, we characterized the influence of various parameters of the PDMS wall. Multiple experiments were implemented for the different thicknesses and the various diffusion areas of PDMS wall with the pressure difference from 10 kPa to 80 kPa in 10 kPa increments. The movement speed of the interface in the side channel was calculated by measuring the displacement of interface over time by capturing images with a CCD camera (Panasonic WV-GP460) and analyzing the

distance using Image J visualization software. We measured the movement speed of different devices with PDMS wall thicknesses of $50\ \mu\text{m}$, $100\ \mu\text{m}$, and $200\ \mu\text{m}$ at different pressure difference, while the diffusion area was $0.3\ \text{mm}^2$. As shown in Fig. 5(a), it can be observed that the movement speed of interface is approximating proportional to the pressure difference. The movement speed decreases with the increase in the thickness of PDMS wall, which indicates that air volume that passed through the PDMS wall in a certain time decreases while the thickness of the PDMS wall increases. We also tested the movement speed of different devices with varying diffusion areas of $0.3\ \text{mm}^2$, $0.6\ \text{mm}^2$, and $0.9\ \text{mm}^2$, while the thickness of PDMS wall was $100\ \mu\text{m}$. As shown in Fig. 5(b), the movement speed of interface at different pressure difference is found to increase as the diffusion area increases.

C. Characterization of manipulation performance

After proving the proposed manipulation method and determining the relationship between the movement speed of the air-liquid interface and related parameters, we further investigated the influence of the pressure difference on the manipulation performance. Upon activating the transducer at the frequency of $42\ \text{kHz}$, while the thickness of PDMS wall was $100\ \mu\text{m}$ and the diffusion area was $0.6\ \text{mm}^2$, the experiments were repeated with various pressure differences and the number of polystyrene microparticles with a diameter of $10\ \mu\text{m}$ that were separated from the air-liquid interface was counted. Figure 6(a) shows the results of the particles lost at

FIG. 6. The manipulation performance with different pressure differences and different excitation frequencies. (a) Loss percentage of the particles versus pressure difference. The thickness of PDMS wall was $100\ \mu\text{m}$, the diffusion area was $0.6\ \text{mm}^2$, and the excitation frequency was $42\ \text{kHz}$. (b) Loss percentage of the particles versus excitation frequency. The thickness of PDMS wall was $100\ \mu\text{m}$, the diffusion area was $0.6\ \text{mm}^2$, and the pressure difference was $10\ \text{kPa}$.

various pressure differences. The captured particles can be held on the acoustically oscillating interface under low pressure difference, showing a better carrying performance at low movement speed of the interface. However, as the pressure difference and the movement speed increased, the number of particles lost from the oscillating interface in transportation increased. At the pressure difference of 70 kPa with the movement speed of 150 $\mu\text{m/s}$, almost all of the captured particles were lost from the oscillating interface in transportation.

Acoustic excitation frequency is also one of the important parameters with regard to the carrying performance in transportation. Thus, we investigated the influence of excitation frequency of the applied signal to the piezoelectric transducer on the carrying performance. The carrying performance was characterized by varying the excitation frequency with the pressure difference of 10 kPa, while the thickness of PDMS wall was 100 μm and the diffusion area was 0.6 mm^2 . Figure 6(b) shows the lost percentage of particles from the acoustically oscillating interface at different excitation frequency with the movement speed of 30 $\mu\text{m/s}$ and the transportation distance of 300 μm . When the excitation frequency was 42 kHz, more than 95% of captured particles were still held on the oscillating interface. As the excitation frequency was far from the resonant frequency, the captured particles easily departed from the oscillating interface in transportation, which indicates that the acoustic radiation force was stronger near the resonant frequency. According to the above results, it seems that the radiation force generated by the oscillating interface is responsible for the capturing and carrying of particles.

IV. CONCLUSIONS

We have demonstrated a novel manipulation method for micro-objects using acoustically oscillating bubbles with a controllable position based on the gas permeability of PDMS. The position of the air-liquid interface trapped within the side channel can be controlled by generating pressure difference between the side channel and the air channel. Successful manipulation of capturing, transporting, and releasing for particles is accomplished in the experiments. We also characterized the influence of various parameters on the gas permeability of PDMS wall, including the thickness and the diffusion area of the PDMS wall and the pressure difference. By controlling the pressure difference and excitation frequency, the carrying performance of particles in transportation can be optimized. Since the bubble generation and the air-liquid interface movement in our manipulation method do not need any electrochemical reaction and any high temperature, this on-chip manipulation method provides a controllable, efficient, and non-invasive tool for handling micro-objects such as particles, cells, and other entities. It can be applied to the cell culture device which can transitorily store cells in the side channels to achieve culture medium replenishment and waste removal in a short time. The particle manipulation can also be used to select a particular size of cells and particles in the microfluidic device.

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