Microfluidic cell concentrator with a reduced-deviation-flow herringbone structure

Ji-chul Hyun,¹ Jongchan Choi,¹ Yu-gyung Jung,² and Sung Yang^{1,2,a)}

¹*School of Mechanical Engineering, Gwangju Institute of Science and Technology (GIST), Gwangju 500-712, South Korea*

²*Department of Biomedical Science and Engineering, Gwangju Institute of Science and Technology (GIST), Gwangju 500-712, South Korea*

(Received 2 May 2017; accepted 19 September 2017; published online 27 September 2017)

In this study, a microfluidic cell concentrator with a reduced-deviation-flow herringbone structure is proposed. The reduced-deviation-flow herringbone structure reduces the magnitude of deviation flow by a factor of 3.3 compared to the original herringbone structure. This structure shows higher recovery efficiency compared to the original herringbone structure for various particle sizes at high flow rate conditions. Using the reduced-deviation-flow herringbone structure, the experimental results show a recovery efficiency of 98.5% and a concentration factor of $3.4\times$ at a flow rate of 100 ml/h for all particle sizes. An iterative concentration process is performed to achieve a higher concentration factor for $10.2\text{-}\mu\text{m}$ particles and Jurkat cells. With two stages of the concentration process, we were able to achieve over 98% recovery efficiency and a concentration factor of $10\text{--}11\times$. Cell viability was found to be above 96% after iterative concentration. We believe that this device could be used to concentrate cells as a preparatory step for studying low-abundance cells. *Published by AIP Publishing.* <https://doi.org/10.1063/1.5005612>

I. INTRODUCTION

Low-abundance cells refer to cells that have cell concentrations less than 1000ml^{-1} .^{1,2} This cell type includes circulating tumor cells (CTCs) and human immunodeficiency virus (HIV)-infected T cells, which are related to cancer or HIV disease, respectively.^{3–5} To improve the diagnostic accuracy of these cells, the concentration process is an essential preparatory step in the diagnostic field.^{6,7} Currently, centrifugation is a widely used method for cell concentration, but it cannot be applied to small sample volume (few microliters) or low-concentration cell samples (e.g., low abundance cells) owing to target cell loss and sample contamination from the users. In this respect, microfluidic devices have been studied as an alternative to the centrifugation method. The advantage of microfluidic devices is that they are suitable for concentrating small amounts of samples without cell loss owing to the microscale, and they can also reduce sample contamination because of the ability to be integrated with other applications (e.g., cell cultures, drug screening chips).^{8–10}

Many researchers have developed various mechanisms for concentrating cells in microfluidic devices. These concentration methods can be divided into two categories: active concentration and passive concentration. Active concentration uses external equipment to generate a force to concentrate cells. Acoustic,^{11–13} magnetic,¹⁴ and electrical methods^{15–19} have been studied by many researchers. These methods have high concentration factors (above $10\times$) and recovery efficiencies (above 85%), but they require specific buffer or cell labeling processes to concentrate the cells. Furthermore, these devices are complicated and complex for fabrication. Passive concentration only relies on the hydrodynamic phenomena produced by the shape of the channel to concentrate the cells.^{20–23} These devices are easy to fabricate compared to active

^{a)}E-mail: syang@gist.ac.kr

concentration devices. Additionally, they do not require cell labeling processes or specific buffers, but they cannot easily handle large amounts of volume (high throughput).

The herringbone structure is a fishbone-shaped channel that is one of the features used in cell concentration methods. Mostly, it has been used for cell separation and focusing applications.^{24–28} Its mechanism of action relies on the lateral pressure gradient produced by the herringbone structure; the particles are then focused and separated by the size of the particles. Recently, parametric studies (cell diameter/channel height, angle of the herringbone structure, and channel width) on the herringbone structure have been reported for particle focusing and separation applications in high flow rate conditions.^{29,30} In high flow rate conditions (above $Re > 10$), particles were focused in the equilibrium position owing to the lateral pressure gradient produced by the herringbone structure (focusing flow and deviation flow) and the inertial force (wall-effect and shear-gradient lift force) produced by the fluid velocity. Although parametric studies have been performed with various structural parameters, there have been no reports of changing the shape of the herringbone structure.

In this paper, we propose a new herringbone structure that enables the concentration of particles and cells at a high flow rate condition (100 ml/h) which is twenty times greater than the original herringbone structure.³⁰ This new herringbone structure produces a smaller magnitude of deviation flow compared to the original without sacrificing the focusing flow. This structure enables us to concentrate particles at 100 ml/h (Re : 230), because the magnitude of deviation flow is lower than that of the original structure. To compare the original herringbone structure and the new herringbone structure, the lateral pressure gradient is compared numerically. Additionally, the recovery efficiency is compared for various sizes of particles. Finally, an experiment is conducted to concentrate various sizes of particles and Jurkat cells under a high flow rate condition.

II. MATERIALS AND METHODS

A. Device design and fabrication

Figure 1(a) shows the configuration of the device that contains the reduced-deviation-flow herringbone structure. The device consists of one inlet and three outlets. Initially, the particles are randomly introduced into the inlet, and after passing through the reduced-deviation-flow herringbone structure, the particles are focused near the wall. This phenomenon occurs because of the force balance between the pressure gradient produced by the herringbone structure (focusing flow and deviation flow) and the inertial force produced by the fluid velocity (wall-effect and shear-gradient lift force). Outlets 1 and 3 are used to concentrate the particles, whereas outlet 2 is used to extract the diluted fluid. Figure 1(b) shows the geometric parameters of the original and reduced-deviation flow herringbone structures. The angle of the head herringbone structure (A_1) is 30° . The length of the herringbone (G_1) is $70\ \mu\text{m}$, and the gap between the herringbones (G_2) is $100\ \mu\text{m}$. The width of the device (W) is $200\ \mu\text{m}$. The height of the herringbone structure (h_{hr}) is $18\ \mu\text{m}$, and the channel height (h_{ch}) is $12.5\ \mu\text{m}$. The difference between the original and reduced-deviation-flow herringbone structures is the angle of the tail herringbone structure (A_2). The original herringbone structure has 30° , where the reduced-deviation flow herringbone structure has 0° .

The device was microfabricated by UV lithography and polydimethylsiloxane (PDMS) replica mold techniques. To fabricate the herringbone structure, SU-8 2025 (MicroChem, USA) was spun on a Si wafer (100) at 4000 rpm, and the wafer was then soft-baked at 95°C for 5 min. Subsequently, the wafer was exposed to UV light ($15\ \text{mJ cm}^{-2}\text{s}^{-1}$) for 10 s using a contact micro-aligner (MIDAS, Korea). After the exposing process, post-exposure baking (PEB) was performed at 95°C for 5 min. After the baking process, it was developed using an SU-8 developer (MicroChem, USA) for 2 min. After the fabrication process, the height of the herringbone structure (h_{hr}) was $18\ \mu\text{m}$. To fabricate the channel, SU-8 2010 (MicroChem, USA) was spun on a Si wafer (100) at 2000 rpm, and the wafer was then soft-baked at 95°C for 3 min. Subsequently, the wafer was exposed to UV light ($15\ \text{mJ cm}^{-2}\text{s}^{-1}$) for 9 s using a contact micro-aligner (MIDAS, Korea). After the exposing process, PEB was performed at 95°C for 4 min. After the baking process, it was developed using an SU-8 developer (MicroChem, USA) for 2 min. After the fabrication process, the height of the channel (h_{ch}) was $12.5\ \mu\text{m}$.

FIG. 1. Device configuration. (a) The particles are randomly introduced to the device from the inlet. The particles are focused on a nearby wall owing to the pressure gradient produced by the reduced-deviation-flow herringbone structure and the inertial force produced by the fluid velocity. (b) Structure parameters of the original and reduced-deviation-flow herringbone structures that have been used in the experiments.

Finally, a PDMS (Sylgard 184, Dow Corning, Korea) slab was fabricated using a conventional replica molding technique. Using the contact micro-aligner (MIDAS, Korea), the two PDMS blocks were aligned to form the device.

B. Experimental setup

A CCD camera (DP 73, Olympus, Japan) was mounted on an inverted microscope (IX72, Olympus, Japan) to capture the movement of the particles. A commercial syringe pump (neMESYS, Cetoni GmbH, Germany) was used to inject the fluid that contains the particles. The flow rate condition varied from 10 ml/h to 100 ml/h. A haemocytometer (Incyto, Korea) was used to calculate the recovery efficiency and concentration factor in later experiments.

C. Microparticles and cell culture

To evaluate the device, green fluorescent polystyrene particles (diameters: $4.8\ \mu\text{m}$, $7.3\ \mu\text{m}$, and $10.2\ \mu\text{m}$) (Thermofisher scientific, Spherotech Inc., USA) were suspended in a 0.5% Tween-20 solution. The particle concentrations were 5×10^4 to $1 \times 10^5\ \text{ml}^{-1}$.

Jurkat cells were cultured in RPMI-1640 (Sigma Aldrich, USA) contained with 10% fetal bovine serum (Gibco, USA). The initial cell concentrations were 1×10^4 to $5 \times 10^4 \text{ ml}^{-1}$. Cell staining (color: green) was performed with calcein (Thermofisher scientific, USA) to observe cell movement. After concentrating the cells, Trypan blue solution (Gibco, USA) was used to confirm the viability of the cells.

III. RESULTS AND DISCUSSION

A. Numerical simulations of the original herringbone structure and the reduced-deviation-flow herringbone structure

To compare the reduced-deviation-flow herringbone structure with the original herringbone structure, the commercial software COMSOL Multiphysics (Comsol, USA) was used for the numerical simulations. The simulated structure is shown in Fig. 2. Figure 2(a) shows the original herringbone structure, and Fig. 2(b) shows the reduced-deviation-flow herringbone structure. We have defined the geometry dimensions in the simulation to match those given in Sec. II A [Fig. 1(b)].

To compare these structures, the magnitudes of the velocity vector and pressure gradient have been compared. The boundary conditions were defined as a flow rate of 100 ml/h at the inlet and atmospheric pressure at the outlet. In addition, a no-slip condition was applied to the channel walls. Five regions were compared to observe the magnitude of the focusing flow and

FIG. 2. Simulation results of the magnitude of the velocity vector. (a) Original herringbone structure. (b) Reduced-deviation-flow herringbone structure. The design parameters used in the numerical simulations match those in Fig. 1(b). Focusing flow is defined as a flow that is produced to the central part of the microchannel (black arrow line). On the contrary, deviation flow is defined as a flow that is produced in the outer part (nearly the fluidic channel wall) of the microchannel (red arrow line).

deviation flow. In Fig. 2, regions 1 and 2 show the magnitude of the focusing flow, whereas regions 3, 4, and 5 show the magnitude of the deviation flow. There is no difference in the magnitude of the focusing flow between the original and reduced-deviation-flow herringbone structures, but the reduced-deviation-flow herringbone structure significantly reduced the magnitude of the deviation flow compared to the original herringbone structure. These results imply that the head herringbone structure's angle affects the focusing flow, while the tail herringbone structure's angle affects the deviation flow. We expect that the magnitude of deviation flow has been reduced due to the following hydrodynamic reason. When the fluid and particle escape from the original herringbone structure, there is gradual decrement of the height in the middle part depending on the angle of the tail herringbone structure (Fig. 2, original herringbone structure regions 3, 4, and 5). Due to the gradual height difference, there is a pressure drop in the x-axis direction (channel wall direction) which causes the deviation flow. For the reduced-deviation-flow herringbone structure, there is a sudden contraction of the height in the z-direction when the fluid or particle escapes from the herringbone structure (Fig. 2, reduced-deviation-flow herringbone structure regions 3, 4, and 5). So, there is a pressure drop to the z-axis (height direction) rather than to the x-axis (channel wall direction). Owing to this reason, the reduced-deviation-flow herringbone structure has a downward flow created instead of the deviation flow. With the change of the tail herringbone structure's angle, the magnitude of the deviation flow has been decreased compared to the original one.

For quantitative comparison, Figs. 3(a) and 3(b) show the pressure distributions of the original herringbone structure and the reduced-deviation-flow herringbone structure, respectively, in regions 1 and 3. The scale bar indicates the values for the pressure (Pa). The pressure gradients are shown in Fig. 3(c). The pressure gradients in region 1 are nearly identical, which means that the magnitude of focusing flow is similar for the two different structures. However, the pressure gradient for the reduced-deviation-flow herringbone structure in region 3 is 3.3 times smaller than that for the original. This demonstrates that the reduced-deviation-flow herringbone structure has a smaller magnitude of deviation flow without sacrificing the focusing flow. In order to validate this effect, experiments were conducted to compare the recovery efficiencies of the original herringbone structure and the reduced-deviation-flow herringbone structure with various sizes of particles.

B. Experiments with the original herringbone structure and the reduced-deviation-flow herringbone structure

To compare the original herringbone structure and reduced-deviation-flow herringbone structure experimentally, the recovery efficiencies were measured for three different sizes of particles (diameters: $4.8\ \mu\text{m}$, $7.3\ \mu\text{m}$, and $10.2\ \mu\text{m}$). Here, the recovery efficiency is defined as the number of particles obtained from outlets 1 and 3 divided by the number of particles acquired from all outlets. Figure 4(a) shows the movement of the $4.8\text{-}\mu\text{m}$ particles when the particles are injected at a flow rate of 100 ml/h. The microscopic images have been taken from the end of the herringbone structure and the outlet. In the case of the original herringbone structure, the defocusing phenomenon has been observed due to the higher deviation flow compared to the reduced-deviation-flow herringbone structure. (For a more detailed depiction, an animated avi file is available as supplementary file. The avi files have been taken in the end of the herringbone structure and the outlet with original and reduced-deviation-flow herringbone structures, Multimedia view.) For the original structure case, the particles travel through outlet 2 owing to the defocusing phenomenon of the particles. Figure 4(b) shows the comparison of the recovery efficiency with respect to the size of the particles with different herringbone structures. In the case of the original herringbone structure, the recovery efficiency increases as the particle size increases. As other researchers have already mentioned in their papers, the influence of the deviation flow decreases as the size of the particle increases ($D/h_{\text{ch}} > 0.5$; D : particle diameter and h_{ch} : channel height).^{29,30} Nevertheless, the recovery efficiencies for the original herringbone structure are lower (33%, 10%, and 5% for $4.8\ \mu\text{m}$, $7.3\ \mu\text{m}$, and $10.2\ \mu\text{m}$ particles, respectively) than those for the reduced-deviation-flow herringbone structure due to the higher magnitude of

FIG. 3. Simulation results for pressure value (Pa). (a) Original herringbone structure. (b) Reduced-deviation-flow herringbone structure. (c) Pressure gradients in regions 1 and 3, where focusing flow and deviation flow are dominant, respectively.

deviation flow. The recovery efficiencies of the three different sizes of particles were all above 98.5% for the reduced-deviation-flow herringbone structure.

C. Concentration of various sizes of particles

The concentration performance of the reduced-deviation-flow herringbone structure was studied with three different sizes of fluorescent particles. Three parts (herringbone channel inlet, end of herringbone structure, and outlet) were observed with fluorescent particles (Fig. S1, [supplementary material](#)). The flow rate condition was 100 ml/h. At the inlet, the particles were randomly distributed, and after flowing through the series of herringbone structures, the particles were focused into two streamlines owing to the pressure gradient created by the herringbone structure and the inertial force ($Re > 10$) caused by the velocity of the fluid. The particles were concentrated at outlets 1 and 3, and the diluted fluid traveled through outlet 2. In a previous paper, it was reported that the height of the channel should satisfy $D/h_{ch} > 0.5$ (D : particle diameter and h_{ch} : channel height) in order to focus the particles.^{29,30} However, in the case of the reduced-deviation-flow herringbone structure, even $4.8\text{-}\mu\text{m}$ particles ($D/h_{ch} > 0.38$) could be well-focused owing to the reduction of the deviation flow.

Figure S2 ([supplementary material](#)) shows a comparison of the focusing characteristics for three different particle sizes under various flow rate conditions (10–100 ml/h). To compare the focusing effect, two parameters have been defined. The focused x -position represents the x -coordinate with the highest intensity value among the intensity values in Fig. S2(b) ([supplementary material](#)). As the flow rate increases, the focused x -position moves closer to the wall.

FIG. 4. (a) Microscopic image of 4.8- μm particles in the herringbone channel inlet, end of the herringbone structure, and channel outlet. (b) Comparison of recovery efficiency using the original herringbone structure and reduced-deviation-flow herringbone structure with different sizes of particles. The design parameters used in the experiments match those in Fig. 1(b). All experiments were performed and observed three times. Multimedia views: <https://doi.org/10.1063/1.5005612.1>; <https://doi.org/10.1063/1.5005612.2>; <https://doi.org/10.1063/1.5005612.3>; <https://doi.org/10.1063/1.5005612.4>

This equilibrium position is determined by the force balance of the pressure gradient (focusing flow and deviation flow) and the inertial force (wall-effect and shear-gradient lift force). Another parameter is the full width at half maximum (FWHM), which is defined as the width of half the maximum intensity at the x-position [see the blue line in Fig. S2(b), [supplementary material](#)]. A smaller FWHM value indicates a better focusing effect. As the flow rate increases, the FWHM value decreases, which was confirmed in other articles as well.^{29,30}

Figure 5 details the recovery efficiency and concentration factor of the reduced-deviation-flow herringbone structure. Various particle sizes have been tested and it turns out that the recovery efficacy increases as the particle size increases. Nevertheless, the recovery efficiency exceeded 98.5% for all particle sizes. The concentration factor is defined as the average particle concentration obtained from outlets 1 and 3 divided by the particle concentration obtained from the inlet sample. For a recovery efficiency of 100%, the concentration factor can alternatively be determined as the amount of fluid obtained from the entire outlet divided by the amount of fluid obtained from outlets 1 and 3. Utilizing this method, the concentration factor value could be obtained from the numerical simulation and was found to be $3.26\times$ as shown in Fig. S3

FIG. 5. Performance evaluation of the reduced-deviation-flow herringbone structure. (a) Recovery efficiency and (b) concentration factor for various particle sizes at 100 ml/h. All experiments were performed and observed three times.

([supplementary material](#)) and the experimentally obtained concentration factor was about $3.4\times$ regardless of the particle size variation.

D. Iterative concentration of particles and cells

From Sec. III C, the experimental results show that the particles could be concentrated up to $3.4\times$ under high flow rate conditions (100 ml/h). In this section, the device has been used iteratively to further increase the concentration factor. The concentrated fluid in outlets 1 and 3 was again introduced into the inlet for iterative concentration. Particles with a diameter of $10.2\mu\text{m}$ and Jurkat cells were used for the experiment. The mean diameter of the Jurkat cells

FIG. 6. Iterative concentrations of 10.2- μm particles and Jurkat cells. (a) Recovery efficiency. (b) Concentration factor. All experiments were performed and observed three times.

was 10.64 μm [standard deviation: 1.114 μm ; Fig. S4 ([supplementary material](#))]. Figures 6(a) and 6(b) show the recovery efficiency and concentration factor, respectively, for multiple iterative concentration stages. After concentrating twice, the recovery efficiency for particles and cells was still above 98%. Additionally, it was confirmed that the concentration factor was 10–11 $\times$. Cell viability was studied using Trypan blue solution (dead cells) and calcein (live cells). It has been found to be above 96% after iterative concentration (Fig. S5, [supplementary material](#)).

The throughput is relatively high in the microfluidic environment, but it is relatively low compared to the centrifugation method. To overcome this fact, device parallelization could be the key factor to achieve sufficient throughput which has been already reported by other researchers.^{25,31}

IV. CONCLUSION

In this study, we have developed a new herringbone structure that reduces the magnitude of deviation flow by a factor of 3.3 compared to the original without sacrificing the focusing flow. This structure has shown the ability to concentrate various particle sizes (diameters:

4.8 μm , 7.3 μm , and 10.2 μm) under high flow rate conditions (100 ml/h; $\text{Re} = 230$) with the recovery efficiency above 98.5% and the concentrator factor around $3.4\times$ for all particle sizes. When the iterative concentration process was performed, a concentration factor of $10\text{--}11\times$ with 98% recovery efficiency was also achieved not only for 10.2- μm particles but also for 10.64- μm Jurkat cells. In addition, the viability of the Jurkat cells has been maintained up to 96%. We believe that this device could be used to concentrate cells as a preparatory step for studying low-abundance cells.

SUPPLEMENTARY MATERIAL

See [supplementary material](#) for microscopic images of three different particle sizes in three regions (Fig. S1), quantitative focusing characteristics for different particle sizes (Fig. S2) design of the outlet part (Fig. S3), Jurkat cell size distribution (Fig. S4), and Cell viability test after the iterative concentration process (Fig. S5).

ACKNOWLEDGMENTS

This material is based upon work supported by the National Research Foundation of Korea (NRF) grant funded by the Korea government (MSIP) (NRF-2016M3A7B4910556) and the Industrial Technology R&D program of MOTIE/KEIT [2016(No.10062533)].

- ¹U. Dharmasiri, M. A. Witek, A. A. Adams, and S. A. Soper, *Annu. Rev. Anal. Chem.* **3**, 409 (2010).
- ²Y. Chen, S. Li, Y. Gu, P. Li, X. Ding, L. Wang, J. P. McCoy, S. J. Levine, and T. J. Huang, *Lab Chip* **14**, 924 (2014).
- ³S. Nagrath, L. V. Sequist, S. Maheswaran, D. W. Bell, D. Irimia, L. Ulkus, M. R. Smith, E. L. Kwak, S. Digumarthy, A. Muzikansky, P. Ryan, U. J. Balis, R. G. Tompkins, D. A. Haber, and M. Toner, *Nature* **450**, 1235 (2007).
- ⁴C. A. Jansen, I. M. De Cuyper, B. Hooibrink, A. K. van der Bij, D. van Baarle, and F. Miedema, *Blood* **107**, 1427 (2005).
- ⁵E. Ramírez De Arellano, C. Martín, V. Soriano, J. Alcamí, and Á. Holguín, *Virus Genes* **34**, 111 (2007).
- ⁶A. J. Mach, O. B. Adeyiga, and D. Di Carlo, *Lab Chip* **13**, 1011 (2013).
- ⁷A. J. Mach, J. H. Kim, A. Arshi, S. C. Hur, and D. Di Carlo, *Lab Chip* **11**, 2827 (2011).
- ⁸J. Warrick, B. Casavant, M. Frisk, and D. Beebe, *Anal. Chem.* **82**, 8320 (2010).
- ⁹P. Sabounchi, A. M. Morales, P. Ponce, L. P. Lee, B. A. Simmons, and R. V. Davalos, *Biomed. Microdevices* **10**, 661 (2008).
- ¹⁰P. Gascoyne, C. Mahidol, M. Ruchirawat, J. Satayavivad, P. Watcharasil, and F. F. Becker, *Lab Chip* **2**, 70 (2002).
- ¹¹O. Jakobsson, S. S. Oh, M. Antfolk, M. Eisenstein, T. Laurell, and H. T. Soh, *Anal. Chem.* **87**, 8497 (2015).
- ¹²C. Mu, Z. Zhang, M. Lin, Z. Dai, and X. Cao, *Sens. Actuators, B* **215**, 77 (2015).
- ¹³M. Nordin and T. Laurell, *Lab Chip* **12**, 4610 (2012).
- ¹⁴P. Grodzinski, J. Yang, R. H. Liu, and M. D. Ward, *Biomed. Microdevices* **5**, 303 (2003).
- ¹⁵D. Chen and H. Du, *Microfluid. Nanofluid.* **9**, 281 (2010).
- ¹⁶R. Kwak, S. J. Kim, and J. Han, *Anal. Chem.* **83**, 7348 (2011).
- ¹⁷H. Moncada-Hernández and B. H. Lapizco-Encinas, *Anal. Bioanal. Chem.* **396**, 1805 (2010).
- ¹⁸M. Motosuke, K. Yamasaki, A. Ishida, H. Toki, and S. Honami, *Microfluid. Nanofluid.* **14**, 1021 (2013).
- ¹⁹D. Puchberger-Enengl, S. Podszun, H. Heinz, C. Hermann, P. Vulto, and G. A. Urban, *Biomicrofluidics* **5**, 044111 (2011).
- ²⁰A. E. Reece, K. Kaastrup, H. D. Sikes, and J. Oakey, *RSC Adv.* **5**, 53857 (2015).
- ²¹J. M. Martel, K. C. Smith, M. Dlamini, K. Pletcher, J. Yang, M. Karabacak, D. A. Haber, R. Kapur, and M. Toner, *Sci. Rep.* **5**, 11300 (2015).
- ²²J. M. Burke, R. E. Zubajlo, E. Smela, and I. M. White, *Biomicrofluidics* **8**, 024105 (2014).
- ²³Y.-Y. Chiu, C. K. Huang, and Y. W. Lu, *Biomicrofluidics* **10**, 011906 (2016).
- ²⁴K. Kim, H.-K. Seo, and Y.-J. Kim, *Microfluid. Nanofluid.* **16**, 159 (2014).
- ²⁵S. Choi, T. Ku, S. Song, C. Choi, and J.-K. Park, *Lab Chip* **11**, 413 (2011).
- ²⁶S. Choi, S. Song, C. Choi, and J.-K. Park, *Lab Chip* **7**, 1532 (2007).
- ²⁷S. Choi, S. Song, C. Choi, and J.-K. Park, *Anal. Chem.* **81**, 50 (2009).
- ²⁸S. Choi, S. Song, C. Choi, and J.-K. Park, *Small* **4**, 634 (2008).
- ²⁹S. Song and S. Choi, *Appl. Phys. Lett.* **104**, 074106 (2014).
- ³⁰S. Song and S. Choi, *J. Chromatogr. A* **1302**, 191 (2013).
- ³¹S. Choi, J. M. Karp, and R. Karnik, *Lab Chip* **12**, 1427 (2012).