High-throughput sheathless and three-dimensional microparticle focusing using a microchannel with arc-shaped groove arrays

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Sheathless particle focusing which utilises the secondary flow with a high throughput has great potential for use in microfluidic applications. In this work, an innovative particle focusing method was proposed. This method makes use of a mechanism that takes advantage of secondary flow and inertial migration. The device was a straight channel with arrays of arc-shaped grooves on the top surface. First, the mechanism and expected focusing phenomenon are explained using numerical simulation of the flow field and force balance. A simulation of particle trajectories was conducted as a reference, and then a series of experiments was designed and the effects of changes in particle size, flow rate and quantity of the groove structure were discussed. The microscopic images show that this particle focusing method performed well for different size particles, and the results agreed well with the theory and simulated results. Finally, the channel successfully concentrated Jurkat cells, which showed a good compatibility in the biological assay field. In this work, the arc-shaped groove channel was demonstrated to have the ability to achieve high-throughput, sheathless and three-dimensional particle focusing with simple operations.

Over the past decades, microfluidic technology has attracted a great deal of attention due to its potential applications, especially in the field of biology and diagnostics1–3. Microfluidic technologies are suitable for a number of reasons. These include: (1) reduction in the number of samples and reagents necessary; (2) rapid analysis; (3) high sensitivity and resolution in detection; (4) low cost; (5) good portability due to its small size and (6) the potential to be a highly integrated and automated component to meet the experimental requirements4,5.

According to the focusing principle, the state-of-the-art microfluidic technologies can be categorized as either active or passive focusing methods. Active focusing technologies include acoustophoresis (AP)6, dielectrophoresis (DEP)7 and magnetophoresis (MP)8 which rely on external fields for their functionality. The active focusing technologies can offer precise particle manipulation and high focusing efficiency. A high-throughput acoustophoresis chip (HATC) has also been reported to separate blood cells from the whole blood using a temperature-stabilized standing ultrasonic wave at a flow rate up to 2 L h−19. Most of them, however, still have some shortcomings, such as the relatively complicated fabrication of introducing external fields. On the other hand, passive focusing technologies rely entirely on the channel geometry and intrinsic hydrodynamic forces for functionality. Unlike active approaches, passive approaches always offer a remarkably high-throughput with simple set-ups, in which the external fields are not required. These characteristics are extremely useful for some commercial particle and cell focusing applications.

Hydrodynamic focusing using sheath flow is one of the typical passive focusing methods which has been widely adopted in recent years.10,11. The focusing system of sheath flow typically introduces two neighbouring buffer streams with high flow rates to horizontally compress the middle stream containing particles to yield a two-dimensional focusing plane in a narrow width.12. The typical on-chip single-layer sheath flow focusing system can only concentrate particles in 2D format with a high throughput. Although this problem can be solved using the microfluidic shifting technique to achieve three-dimensional focusing, the logistically burdensome sheath flow still cannot be
In this paper, a sheathless, high-throughput and three dimensional particle focusing device consisting of a low aspect ratio straight channel ($AR = 0.2$) and an arc-shaped groove array pattern on the surface is presented. In this channel, the secondary drag force drives particles to a new equilibrium position with good focusing performance which is useful for many biological and diagnostic devices. This focusing approach has attracted a great deal of attention due to its excellent compatible characteristics in terms of both yielding and precision. Inertial microfluidics in a straight channel, however, has some limitations: (1) the cross-section of the channel (aspect of ratio) must be matched in size for effective focusing functionality; (2) particles must pass through a long enough distance in order to move laterally to the equilibrium positions; (3) there are always several equilibrium positions in one channel at the same time\(^{20-22}\). In order to solve these problems, secondary flow is introduced by transforming the geometry of the straight channels by adding a curved portion or disturbance obstacles\(^{23-25}\). The secondary flow induced by these structures exerts a secondary flow drag force on the particles, which: (1) eliminates the limitations of channel size, (2) enhances the efficiency of particle focusing and (3) reduces the requirement of long distance for sufficient displacement to equilibrium positions\(^{16}\). The flow rates of various particle focusing methods are listed in Table 1.

In this paper, a sheathless, high-throughput and three dimensional particle focusing device consisting of a low aspect ratio straight channel ($AR = 0.2$) and an arc-shaped groove array pattern on the surface is presented. In this channel, the secondary drag force drives particles to a new equilibrium position with good focusing performance at a moderate flow rate. Simulation by finite element software is carried out and calculated numerically to describe the focusing mechanism. This focusing approach has been demonstrated by a series of experiments on different size particles ($10\mu m$, $13\mu m$ and $24\mu m$) as well as Jurkat cells. All particles and cells of all sizes focus well at the moderated flow rates ($Re = 1$), and this agrees with the mechanism and simulations. The effect of groove structure quantity on particle focusing is also discussed and investigated by observing the performance of particle focusing after travelling through different numbers of groove structures. This innovative focusing approach shows good focusing performance which is useful for many biological and diagnostic devices.

### Theory

**Inertial migration.** When particles are dispersed randomly at the entrance of a straight channel, the inertia of the fluid and particles causes the particles to migrate laterally to several equilibrium positions within the cross-section of the channel after travelling a long enough distance\(^{26,27}\). This interesting phenomenon has been widely recognised by the counteraction of two dominated inertial effects: (1) the shear gradient lift force $F_{\text{Lsg}}$, originating from the curvature of the fluid velocity profile and its interaction with the particle, which directs particles away from the centre of the channel, and (2) the wall lift force $F_{\text{Lwa}}$ as a result of the flow field interaction between suspended particles and adjacent walls, which repels particles away from the wall\(^{28}\).

In a channel, the inertial lift force exerts onto flowing particles and forms some equilibrium positions, which is closely related to the geometry of the channel's cross-section. Due to limitations in microfabrication, the cross-sections of most channels utilised in microfluidic devices are rectangular. When particles travelling in a square straight channel ($AR = \text{height/width} = 1$), they form four equilibrium positions centred at the faces of the channel at a finite Reynolds number (Fig. 1a)\(^{29}\). As the aspect ratio (AR) decreases, the number of equilibrium positions reduces to two which are located along the longer faces as the velocity profile varies from a sharp parabolic curve to a blunted one\(^{15}\). As the AR decreases continuously to 0.2, however, the two equilibrium positions gradually spread out to two relatively wide bands parallel with longer faces of the channel (Fig. 1b). The particles migrate to the top and bottom of the channel along shorter faces because the parabolic velocity profile between two longer faces is sharp, while it is difficult to see the particles focusing along the longer face with an extremely weak shear gradient lift force due to the blunt velocity profile between two shorter faces\(^{30}\).

The net inertial lift force $F_L$ acting on a small rigid sphere is expressed by Asmolov as\(^{31}\):

$$F_L = \frac{2}{3} \pi R^2 \rho \mu \frac{d}{2}$$

### Table 1. List of the flow rates of different particle focusing methods.

| Classification | Method | Focusing mechanism | Flow rate |
|----------------|--------|---------------------|-----------|
| Active         | Acoustophoresis\(^{42}\) | Standing surface acoustic waves (SSAW) | 10μl/min   |
|                | Dielectrophoresis\(^{43}\) | Externally controlled positive Dielectrophoresis | 0.01μl/min |
|                | Hydrodynamics\(^{43}\) | Dean flow and shear flow | 330μl/min |
| Passive        | Inertial microfluidics in a straight channel\(^{42}\) | Inertial migration | 140μl/min |
|                | Inertial microfluidics in a curved channel\(^{17}\) | Inertial migration and Dean flow | ~20μl/min |
|                | Inertial microfluidics in a step channel\(^{17}\) | Secondary flow and Inertial migration | ~295μl/min |
|                | This | Secondary flow and Inertial migration | ~1100μl/min |

\(^{1}\) actuation methods are listed in Table 1.
This expression can be simplified as:

\[ F_L = f_L \rho_f \gamma \gamma a^4 \]  

(1)

This expression can be simplified as:

\[ F_L = f_L \rho_f U a^4 H^2 \]  

(2)

\[ \text{Re} = \rho_f U H / \mu \]  

(3)

\[ H = 2wh / (w + h) \]  

(4)

where \( \rho_f, U, \gamma \) and \( \mu \) are fluid density, maximum velocity, shear gradient and dynamic viscosity, respectively. \( a \) is the particle diameter, and \( H \) is the hydraulic diameter of the channel (\( w \) is the width and \( h \) is the height of the channel). The lift coefficient \( f_L \) is a function of the lateral position of particles \( x \) and the Reynolds number \( \text{Re} \). 

**Secondary flow.** By adding arc-shaped groove arrays on top of the straight channel (AR = 0.2), a secondary flow appears when the fluid flow tends to be trapped in the groove structure due to a pressure gradient in the transverse direction. The secondary flow vorticity induced by the circulation of fluid can influence the particle distribution within cross-sections (Fig. 1c). It is expected that the flow will be required to turn to a larger extent, creating a larger transverse pressure gradient driving the secondary flow and generate a secondary flow drag force that is strong enough to manipulate the particles. To generate a strong enough secondary flow, the ratio of the height of the arc-shaped groove arrays and main channel is set as ~1.

Because the arc-shaped groove arrays are composed of the same arc-grooves, the computational fluid dynamics analysis can be simplified by simulating only one section with a single characteristic groove structure (Fig. 2). The vorticity in the flow is created within the cross-sections, as the flowing fluid is deflected by the arc-shaped groove (Fig. 2a–e). Thus, the secondary flow gradually transports the fluid to shift laterally along the vorticity.
rotational direction within the cross-sections. In this process, the direction of rotation and the magnitude of the secondary flow are the important factors which determine particle displacement. In these five cross-section plots, it can be seen that the direction of the vorticity in the main channel (except the groove part) is constantly directed in a (Y) direction, and the fluid in the arc-shaped groove part will simultaneously re-circulate in the opposite (−Y) direction due to the conservation of mass. The whole helical vorticity pattern is shown in Fig. 1c.

At a low Reynolds number (typically in microfluidic systems), the drag force originating from the transverse fluid flow which is exerted on the small rigid spherical particle can be expressed as follows:

\[ F_D = -6\pi a\eta (v_t - v_p) \]

where \( a \) is the radius of the particle, \( \eta \) is the viscosity of the fluid, \( v_t \) is the dynamics velocity of the fluid, and \( v_p \) is the velocity of the particle.33

Figure 2. Finite element analysis of flow field and characterization of rotational vorticity at modulated flow rate within cross-sections obtained by COMSOL Multi-physics 5.1. A 3D model of a straight channel (\( AR = 0.2 \)) with a single arc-shaped groove has been imported into COMSOL to investigate the alteration of flow field. The dotted line is used to distinguish the interface between the main channel and the arc-shaped groove. (a–e) These five schematic diagrams (a combination of the velocity arrow plot and the pressure contour plot) show the streamline displacement and pressure alternation. The vector arrows represent the flow velocity and the arrow size is proportional to the magnitude of the flow velocity. The expected process of particle focusing is depicted in these five images. (i–v) are the corresponding colour bars of (a–e) to show the colour scale. The unit on the colour bars is Pa.
The process of focusing is shown in Fig. 2a–e. After being distributed randomly at an entrance, the particles locate along the top and bottom surfaces of the channel and form two wide focusing bands due to the inertial migration. When particles are flowing through the parts with the groove structure, the transverse vorticity imposes vertical and lateral drag forces which cause a displacement (shown as grey arrows in Fig. 2) of the particles within the cross-sections. Because the vorticity in the main channel (apart from the groove part) is half of the vorticity circulation (another half occurs in the groove part), the corresponding drag force \( F_D \) on particles is constantly directed in the Y direction and the particles are gradually gathered to one side of the channel. Simultaneously, the shear gradient lift force \( F_{Ls} \) and the wall lift force \( F_{Lw} \) push particles away from the centre of the channel and repel the particles away from the wall of the channel, which will help them to achieve a balance and locate at an equilibrium position. It can be seen that the secondary flow vorticity is not symmetric and the region near the left wall (where particles gathered) is more stable than the other regions (as shown in the simulation results Fig. 2a–e), because it can be seen that the variation of the arrow size which depicts the magnitude of the flow field is smaller than other parts. This also means that particles tend to locate at the left region. This also shows that a trap tendency appears at the left upper corner where the inertial lift force (consisting of the shear gradient lift force \( F_{Ls} \) and the wall lift force \( F_{Lw} \)) counteracts the drag force \( F_D \) and achieves a balance. In addition, if the particle is large enough, particles will not be trapped in the groove part. As a result, this mechanism means that a three-dimensional, high-throughput particle focusing method can be realized using this arc-shaped groove channel at a moderated flow rate.

Results and Discussion
Simulation of particle trajectories. Figure 3a shows the simulation of particle trajectories in the arc-shaped groove channel, where 200 13μm particles were evenly distributed form the inlet at a flow velocity of 2.29 m/s (Re = 160.4). The performance of particle focusing was demonstrated by the contractive particle trajectories, where the colour of the trajectories denotes the particle velocity.

Figure 3b shows the Poincare maps of particle distributions in different cross-sections in the channel. The portion with the groove structure was divided into 10 sections with 11 cross-section plots. Within the first cross-section plot, particles were evenly distributed, but those particles within the following cross-section plots tended to concentrate step by step to one side. Finally, after travelling through the whole portion, all these particles gathered together at a small region within the last cross-sections.

Effects of the channel Reynolds number. To evaluate the effect of flow rate (channel Reynolds number) on the particle focusing performance, a series of validation experiments at various flow rates was conducted. Here, the 13μm particle suspensions were injected into the arc-shaped groove channel form the inlet. When the flow rate increased from 200μl/min to 1500μl/min (Reynolds number from 29.2 to 218.7), different particle focusing patterns and qualities occurred near the outlet of the arc-shaped groove channel shown by Fig. 4a–g. At relatively low flow rates (200μl/min and 500μl/min), the focusing patterns were still inconspicuous and shaped as wide bands (Fig. 4a,b) but the focusing performance was improved when the flow rate increased. The fluorescent images (Fig. 4c–e) show that the particle trajectories tend to concentrate at one side of the channel until the flow rate
increased to 1100 μl/min (Re = 160.4). The reason is that the magnitude of secondary flow drag force was the crucial factor to the particle focusing performance and a higher flow rate was able to induce a stronger secondary flow within the main channel. As a result, the induced stronger secondary flow exerted a stronger lateral drag force on the particles, which enabled them to assemble into a small region quickly and efficiently. When the flow rate exceeded 1200 μl/min (Re = 175.0), however, the stabilized concentrated particle focusing pattern was destroyed due to the violent interaction between particles in the narrow region (Fig. 4f,g). Since the particles in this series of experiments were 13 μm in diameter and the height of channel was 40 μm, they tended to interact with each other when gathering together. Figure 4i is a wall diagram including the particle focusing patterns at different flow rates (200 μl/min to 1500 μl/min) and was established by measuring the particle fluorescent intensity. This diagram shows that the 13 μm particles focusing efficiency was proportional to the flow rate and reached its optimal performance at 1100 μl/min (Re = 160.4). To assess the particle focusing performance numerically, the focusing band width was determined through measuring the length between two points where the normalized intensity profile crossed the 50% threshold. And focusing was attained when the band width was <2 times the diameter of the focused particles34. Figure 4j, the intensity profile at 1100 μl/min (Re = 160.4), shows that the length between two points was measured <25 μm, which means that particle focusing was achieved. On the other hand, to investigate the focusing type of this method, the PDMS channel could be located vertically onto the microscope stage in order to provide a clear enough side view of the arc-shaped groove channel (Fig. 4h). In the plot, particles were found to concentrate along the top of the main channel (the interface between the groove arrays and the main channel) and had a trend to be trapped into the groove structure. Because the size of particles (13 μm) was large enough compared with the gap of the groove (50 μm), particles could be retained in the fluid stream line and not be trapped by the groove structure. The side view of normalized intensity profile (Fig. 4k) showed that the particle distribution in vertical direction located in one streamline approximately as well. As a result, this particle focusing approach in the arc-shaped groove channel could be considered as a three-dimensional focusing.

Effects of the quantity of groove structure. For this particle focusing method, particles were concentrated by the secondary flow which was induced by the groove structure. So it was necessary for particles to pass through plenty of groove structures to achieve adequate lateral displacements for an effective particle focusing. In order to investigate the influence of quantity of the groove structure on particles focusing, 13 μm particle suspensions were injected from the inlet at a flow rate of 1100 μl/min (Re = 160.4) and fluorescent images of the particle flowing pattern after travelling
different quantity of the groove structure were captured (Fig. 5a–e). The microscopic images show that the focusing performance was improved significantly as particles passed more quantity of the groove structure. The particles were gradually pushed to one side and located in a line observing through the (0–5), (10–15), (20–30) and (35–40) segment of the channel. Finally, all the particles were able to be concentrated at the equilibrium position which agreed well with the simulation results achieved (Fig. 5e). The normalized intensity profile (Fig. 5f) showed a comparison of particle focusing patterns at different segments in the channel. According to the particle focusing assessment above, an effective particle focusing could be attained in the channel with more than approximate 40 groove structures when the focusing band width was <2 times the diameter of the focused particles (13 μm) (Fig. 5g,h).

Effects of the particle size. Figure 6a shows that 10 μm particles focused in the same way as 13 μm particles, and they were located precisely at the equilibrium position at the flow rate of 1100 μl/min (Re = 160.4). However, when the 24 μm particles travelled in the channel, the focusing pattern differed from the 10 μm and 13 μm particles (Fig. 6b). The 24 μm particles could concentrate at one side of the channel at only 500 μl/min (Re = 73.0) and 800 μl/min (Re = 116.4), while the focusing pattern was loose at 1000 μl/min (Re = 145.8). This phenomenon could be explained that the secondary flow drag force exerted on particles was proportional to particles size according to equation (5). Particles with diameter of 24 μm could receive a larger drag force than 13 μm particles, and tended to focus at lower flow rates. However, the 24 μm particles could not focus effectively at a flow rate of 1000 μl/min, because there was a greater possibility that the larger size could induce a stronger interaction.
The intensity profiles of fluorescent particles show the focusing performance of 10 μm and 13 μm respectively.

In conclusion, the critical Reynolds number (Re) of optimal focusing performance in the channel was predicted to be inversely proportional to the size of the particle (Fig. 6d). And there was also a critical particle size range which is appropriate for particle focusing in the selected channel. In the arc-shaped groove channel, the critical particle size range was predicted from 10 μm to 24 μm. It was not easy to identify the accuracy range, because the commercial microparticles had a relatively large coefficient of variation (CV), such as a CV 16% of 13 μm particles, and this error would influence the experiments.

To confirm the feasibility of this focusing method on cells, 14 μm Jurkat cell suspensions were introduced into the arc-shaped groove channel. According to the secondary flow drag force equation (5), the forces \( F_D \) and \( F_L \) exerted on the cells were expected to be similar to the forces on 13 μm particles. The microscopy image (Fig. 6c) shows that the arc-shaped groove channel performed well on focusing Jurkat cells at 1100 μl/min (Re = 160.4) and the normalized intensity profile also demonstrates that most Jurkat cells concentrated at one side of the channel. The experiment result agreed well with the focusing mechanism and the experimental results of rigid polystyrene microparticles.

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**Figure 6. Focusing patterns of different particles and Jurkat cells.**

(a) The optical microscopy images of 10 μm particles focusing at different flow rates respectively and the corresponding normalized intensity profile. (b) Images of 24 μm particles focusing at different flow rates respectively and the corresponding normalized intensity profile. (c) Focusing pattern of Jurkat cells at a flow rate of 1100 μl/min (Re = 160.4) and the corresponding normalized intensity profile. And the optical microscopy of Jurkat cells is shown at the corner. (d) The predicted relationship between particle size and corresponding optimal focusing Reynolds number.
Materials and Methods

**Design and fabrication of the microchannel.** Figure 7a is a schematic drawing of the microchannel with the arc-shaped grooves utilised in experiments. The microchannel is a stacked structure which consists of a main straight channel (AR = 0.2) and arc-shaped groove arrays. The main channel width (W) is 200 μm with a height (H) of 40 μm and a total length (L) of 7 mm. The groove arrays are located on the top of the main channel between 1 and 6 mm downstream from the inlet; they are composed of 50 arc-shaped grooves with 50 μm spacing (H1) between them. The groove is arc-shaped with a small curvature of 600 μm (R1) and a large curvature of 650 μm (R2). The height of the groove arrays (H2) is 38 μm (Fig. 7b).

A double-layer silicon master was fabricated by the two-step photolithography technique on a silicon wafer. The first layer was processed as the main channel and the second layer was processed as the arc-shaped groove arrays aligned to locate on top of the first layer (the main channel). The first layer photoresist (PR) (SU-8 2050, Microchem Corp., Newton, MA) was spun on a clean silicon wafer through a three-step coating cycle (500 rpm for 20 s, 2000 rpm for 20 s, and 4000 rpm for 40 s). Then, the silicon wafer with coated photoresist was heated at the temperature of 65 °C for 2 minutes and 95 °C for 7 minutes. After the first heating, the silicon wafer was exposed by UV light through the first photomask and subsequently heated by the second two-step hard baking (at 65 °C for 3 minutes and 95 °C for 7 minutes). Afterwards, the silicon wafer was developed in the SU-8 developer solution and rinsed with isopropyl alcohol (IPA). The remaining photoresist pattern attaching onto the wafer formed the first layer (the main channel). In the second-step photolithography, the second spin-coating photoresist on the previous mould was processed according to the first-layer process. Then, the second photomask was carefully aligned to the processed photoresist pattern (the first layer), and UV light was used to expose onto the second layer photoresist through the second photomask. After heating and developing the pattern, the double-layer mould was processed with trichlorosilane to deposit a monolayer onto the surface.

A mixture of poly(dimethylsiloxane) (PDMS) and curing agent (Dow Corning, Midland, MI) in a ratio of 10:1 was poured onto the double-layer mould, degassed and then cured in a convection oven at 65 °C for 2 hours. The cured PDMS was then peeled off from the mould and punched through the inlet and outlet holes with a custom needle tip. Finally, The PDMS replica was sealed with a glass slide after treatment in oxygen plasma (PDc-002, Harrick Plasma, Ossining, NY) for 3 minutes.

**Preparation of particles and cells.** Green fluorescent polystyrene particles with a diameter of 9.9 μm (CAT. NO. G1000, 5% CV), red fluorescent polystyrene particles with a diameter of 24 μm (CAT. NO. 36-5, 12% CV) and 13 μm (CAT. NO. 36-4, 16% CV) were purchased from Thermo Fisher Scientific, and then suspended in deionized (DI) water. The concentrations of the particle in these suspensions were ~5 × 10⁴ counts/ml, ~2 × 10⁵ counts/ml and ~5 × 10⁶ counts/ml respectively. To prevent the aggregation, sedimentation, and adhesion to the microchannel walls, 0.1% w/v Tween 20 (Sigma-Aldrich, Product NO. P9416) was added to the particle suspensions³⁴,³⁵,³⁶.

Jurkat cell lines were cultured in RPMI medium 1640 (Gibco) supplemented with 100 units/ml aqeous penicillin, 100 μg/ml streptomycin and 10% fetal bovine serum at 37 °C in an atmosphere of 100% humidity and 5% CO₂. The cell diameter ranges from 10 and 15 μm. For better observation and being captured using a CCD camera, the Jurkat cells were stained with Calcein AM (Thermo Fisher Scientific) for 30 minutes. The concentration of Jurkat cells used in the experiment was ~5 × 10⁵ counts/ml in the suspensions.

**Experimental setup.** To observe and record the particle focusing phenomenon, the microfluidic device was placed on an inverted microscope (CKX41, Olympus, Japan). A CCD camera (Optimos, Q-imaging, Australia) was used to observe and capture the fluorescent particle trajectory images, which were then post-processed and analysed using the image processing software, Q-Capture Pro 7 (Q-imaging, Australia). The exposure time was set as 10 ms. Particle fluorescent trajectories were obtained by stacking 50 consecutive frames in order to measure the focusing position of the fluorescent particles.
Numerical simulation. A numerical simulation was conducted to calculate the flow field characteristics and particle trajectories to investigate and predict the focusing phenomenon with finite element software (COMSOL Multi-physics 5.1, Burlington, MA). A 3D model of the microchannel with 50 arc-shaped grooves was built using the modelling function of COMSOL. A laminar flow model was then added to the component to calculate the flow field. In laminar flow physics, the boundary condition was set as no slip and the physical property of fluid was set as a steady incompressible flow. At the inlet, the normal inflow velocity was set as 2.29 m/s (the corresponding flow rate and Reynolds number were 1100 μl/min and Re = 160.4 respectively) and the pressure at the outlet was set as 0 Pa. Afterwards, a model of particle tracing for fluid flow was added to the same component and coupled with the previous physics to predict the particle trajectories in the microchannel. At the entrance, 200 μm particles with a density of 1050 kg/m³ were released randomly along the channel width. The time range was set from 0 to 0.019 s with an interval time of 10⁻⁵ s, in time settings. For the numerical solution, the order of finite element was set as P1 + P1, the default setting of COMSOL Multiphysics and the mesh was set as the free tetrahedral at finer level (1488007 elements). The equations to govern steady incompressible flow are as below:

\[ \rho \mathbf{v} \cdot \nabla \mathbf{v} = -\nabla P + \mu \nabla^2 \mathbf{v} \]  

Continuity equation:  

\[ \nabla \cdot \mathbf{v} = 0 \]  

Non-slip boundary condition:  

\[ \mathbf{v} = \mathbf{0} \]

where \( \mathbf{v} \) is the velocity vector and \( P \) is the pressure of flow field, respectively; \( \mathbf{v} \) is the fluid velocity vector at the channel walls; \( \nabla \) is the Nabla operator: \( \nabla = \frac{\partial}{\partial x} + \frac{\partial}{\partial y} + \frac{\partial}{\partial z} \), and \( \nabla^2 \) is the Laplace operator:  

\[ \nabla^2 = \nabla \cdot \nabla = \frac{\partial^2}{\partial x^2} + \frac{\partial^2}{\partial y^2} + \frac{\partial^2}{\partial z^2}. \]

For this particle focusing method, the secondary flow drag force works dominantly and guides the particle distribution within the channel, while the inertial affects only act as an assisting role to balance particles when they reach the equilibrium position. Meanwhile, the inertial lift force is much weaker compared with the secondary flow drag force, especially along longer faces of channel for the blunt velocity profile. As a result, we neglect the intricate inertial lift force and only consider the secondary flow drag force to reduce the computational time. On the other hand, to simplify the simulation, the fluid-structure interactions were neglected. In this work, the drag force dominates the external force exerted on particles at low Reynolds numbers, so the governing equations are according to equation (5).

Conclusions

A passive particle focusing approach has been developed in this paper by utilising an arc-shaped groove channel. The benefits of this approach are that different size particles can be focused in a three-dimensional type, all in a high-throughput manner, without the requirement of sheath flow. The focusing mechanism was explained and demonstrated by numerical simulation of the flow field. As the inertial focusing performance in a low aspect ratio channel (AR = 0.2) was not effective, the arc-shaped groove arrays were located on the top of the straight channel to induce a secondary flow rotating within the channel cross-sections and a new equilibrium position of particles was formed under the balance between the secondary flow drag force and inertial lift force. Meanwhile, through verification experiments, the focusing approach showed a good applicability for different size particles and cells (Jurkat cells). In conclusion, this research provides a sheathless and high-throughput particle focusing method in three-dimensions, which has the potential to be used for particle or cell concentration, filtration and flow cytometry.

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Author Contributions

Qianbin Zhao conducted the experiments, analyzed the data and wrote the manuscript with the assistance from Jun Zhang. Sheng Yan fabricated and design the microfluidic devices and Dan Yuan cultured the Jurkat cells. Weihua Li supervised the whole research work. All authors discussed the results and commented to the work.

Additional Information

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