Computational study of geometric effects of bottom wall microgrooves on cell docking inside microfluidic devices

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Abstract
Cells docking inside microfluidic devices is effective in studying cell biology, cell-based biosensing as well as drug screening. Moreover, single cell and regularly cells docking inside microstructure of the microfluidics system are advantageous in different analysis of single cell exposed to drugs or mechanical stimulus. In this study, we investigated the bottom wall microgrooves with semicircular and rectangular geometries with different sizes which are suitable for single cell docking in 2D microchannel and numerous cells docking regularly in a line in 3D microchannel. We used computational fluid dynamics to analyze the fluid recirculation area inside different microgrooves which can play important role in the cell attachment to the microgroove substrate. In addition, we analyzed the fluid drag force on the moving cell toward the microgroove. This parameter is proportional to the fluid velocities in x and y directions changing in different microgrooves geometries. Besides, this is important in the cell attachment to the microgroove substrate. The percentage of negative shear stress and average shear stress on the adhered cell surface which is important in the cell detachment, were also calculated. The results showed that in the constant fluid inlet velocity and microchannel height, microgroove geometry and ratio of cell size to the microgroove size play pivotal role in the cell initial adhesion to the substrate as well as the cell detachment.

Keywords: Microfluidic device, Simulation, Microgrooves, Cell docking

1 Introduction
Docking and capturing of cells are critical in various biomedical applications comprising diagnostics, cell biology and therapeutics(Gossett, Weaver et al. 2010). Nowadays, single cell analysis takes precedence over studying bulk population of cells due to discovering mechanisms related to genomics, proteomics and metabolomics. In other words, single cell analysis has advantages involving analyzing the cell antigen and chromosome, exposure to PHAs and analyzing RNA and DNA alteration, analyzing cytotoxicity of the cell and different cell fate(Di Carlo, Wu et al. 2006). In some biomedical applications, capturing irregular accumulation of cells cannot provide suitable results, so the experiment needs orderly and organized cell docking in order to achieving suitable results. Some of these biomedical applications are studying cell mechanotransduction including different cells sense and response to mechanical stimuli(Zhou and Niklason 2012, Polacheck, Li et al. 2013), embryonic stem cell undifferentiated growth and to form homogenous embryonic stem cell aggregates to enhance their differentiation for
therapeutic applications (Khademhosseini, Ferreira et al. 2006, Vining and Mooney 2017), studying cells interactions and adhesion (Sackmann, Fulton et al. 2014), and providing multiphenotype cell arrays for drug discovery experiments (Khademhosseini, Yeh et al. 2005, Di Carlo, Wu et al. 2006). Some of conventional techniques used for cell sorting and capturing are Fluorescence-activated cell sorting (FACS) and Magnetic-activated cell sorting (MACS) which utilize complementary fluorophore-conjugated antibodies to label cells of interests and magnetic beads to bind specific proteins on cells of interest, respectively. These two conventional techniques with labels may affect cell fate and function, and also they consume costly reagent (Kumar and Bhardwaj 2008).

Nowadays, microfluidic devices have different biological and biochemical applications. These microdevices can manipulate fluid flows, enable high-throughput experimentation while minimizing costly reagent consumption and reducing sample processing time (Weibel and Whitesides 2006, Tian and Finehout 2009). To capturing cells within microfluidic channels, a number of methods such as encapsulation within photocrosslinkable polymers and using magnetophoresis, dielectrophoresis and acoustophoresis have been widely used which may adversely affect cell biology studies (Koh, Revzin et al. 2002, Khademhosseini, Suh et al. 2004, Fidkowski, Kaazempur-Mofrad et al. 2005, Petersson, Åberg et al. 2007, Vahey and Voldman 2008). However, target cell sorting and docking due to fluid hydrodynamic force inside the microchannel do not affect cell biology studies and cell phenotype (Moehlenbrock, Price et al. 2006). Since hydrodynamic forces on the target cells, for example leukocytes, inside the microchannel are similar to the physiological condition inside the body (in-vivo), so the cells experience almost same condition in-vitro (Schaff, Xing et al. 2007).

Some previous studies on cell docking inside microfluidic channels have included substrate with microgrooves or microwells in order to improve cell docking (Khademhosseini, Yeh et al. 2005, Park, Berthiaume et al. 2005, Manbachi, Shrivastava et al. 2008, Khabiry, Chung et al. 2009, Cioffi, Moretti et al. 2010, Khabiry and Jalili 2015). These studies have been experimentally done by cell seeding inside microfluidic systems. Due to high cost of experiments, they have only studied rectangular microgrooves for cell positioning. Recent studies of microgrooves role in cell docking were accompanied by simulation of fluid shear stress and recirculation area at the bottom of microgrooves before cell seeding (Manbachi, Shrivastava et al. 2008, Khabiry, Chung et al. 2009, Cioffi, Moretti et al. 2010). In these studies the interaction between fluid and cells have not been simulated. In addition, they have studied rectangular microgrooves with large dimensions for capturing many cells inside microgrooves; therefore, irregular accumulation of cells inside a microgroove cannot provide the condition for aforementioned biomedical applications that need orderly cell docking.

In this work, we studied the cell docking in the flat microchannel and microchannels with semicircular and rectangular microgrooves with different dimensions. Assumed microgrooves dimensions were suitable for single cell docking in 2D microchannel, and cells positioning in one line in 3D microchannel. We used computational simulation to obtain the fluid velocity inside different microgrooves and shear stress on the attached cells inside the microgrooves. Computational fluid dynamics (CFD) revealed the effect of two kinds of microgrooves with different sizes on the fluid velocity and flow direction which are related to the drag force on the moving cell. In addition, fluid-structure interaction (FSI) modeling determined the fluid shear stress distribution on the adhered cell to predict the cell detachment. In this work, numerical simulation of microfluidic devices has the privilege of assessing the microdevice geometrical features and analyzing mechanical stress on the cells before the fabrication; therefore, it can
reduce the cost of different microdevices fabrication.

2 Materials and Methods

2.1 Microgrooves Geometries

In this work, microchannel with flat bottom wall and two kinds of grooved bottom wall have been studied. The assumed microgrooves sizes were appropriate for docking single cell in 2D or numerous cells regularly in a line in 3D microchannel. The target cells were spherical with radius of 10µm. In the microfluidic device, which its characteristic parabolic velocity, spherical cells such as leukocytes feel the greatest force are driven closest to the wall where flow velocity is the lowest (Yamada and Seki 2005). According to table 1, the radius of assumed semicircular microgrooves was between 12µm and 20µm. In microchannel with rectangular microgrooves, the depth of rectangles were between 12µm and 20µm, and for each depth we assumed two widths which one of them was equal to the width of corresponding semicircular microgroove, and another was representative of the same areas of corresponding semicircular and rectangular microgrooves. Fig. 1 shows two microchannel with two kinds of microgrooves (rectangular and semicircular) that length, height and width of the microchannel are in x, y and z directions, respectively.

The height and length of microchannel were assumed 50µm and 500µm, respectively, and the distance between two microgrooves were 40µm. As shown in Fig. 2, 2D computational domain was discretized into an unstructured mesh of triangular elements. The maximum mesh length was set to 2µm in the bulk and 0.8µm at all corners and microgroove edges and it was confirmed that the results of simulation were grid-independent.

2.2 Numerical simulation of fluid flow

2D computational fluid dynamics (CFD) was used to determine the fluid recirculation area and velocity direction inside the microgrooves. The laminar fluid flow was assumed Newtonian, incompressible and homogenous. The steady state Navier-Stokes equations for incompressible fluid were defined as:

\[ F + \nabla \cdot [ - PI + \mu(\nabla u + (\nabla u)^T) ] = \rho((u) \cdot \nabla)u \]  

(1)

\[-\nabla \cdot u = 0 \]  

(2)

where, \( I \) denotes the unit diagonal matrix, \( F \) is volume force affecting the fluid due to the gravity force, \( u \) is velocity vector, and \( P \) is fluid pressure. The fluid was assumed as water with viscosity of \( \mu=8.9 \times 10^{-4}\text{Pa.s} \) and density of \( \rho=1000\text{Kg/m}^3 \).

The boundary conditions at the walls and at the bottom of the microgrooves were set as no-slip boundary condition. The specified velocity condition equal to \( 2 \times 10^{-4}\text{m/s} \) was applied for the inflow boundary condition. Moreover, the specified pressure equal to 0Pa was used for the outflow boundary condition. Furthermore, the criteria for convergence (RMS residual) were
2.3 Analyzing fluid drag force on the moving cell toward the microgroove

The moving cells experience different forces from the fluid including drag force, buoyance force and weight (Duprat and Shore 2015). Weight and buoyancy force are independent of microgroove geometry and were assumed constant in different microchannels. Fig. 3 shows all the forces acting on the cell when the cell is about to enter the microgroove. In order to compare the moving cell toward different geometries of microgroove, the drag forces in x and y directions are important because other forces are constant in different geometries.

In Fig. 3, F1 and F2 are drag forces in x and y directions, respectively. The drag force on the moving sphere with radius a in the laminar flow can be defined as (Duprat and Shore 2015):

\[ F_{drag} = \int_S \mathbf{n} \cdot \mathbf{\sigma} \, dS = 6.\pi.\mu.a.V \]  

(3)

In equation (3), \( \mathbf{\sigma} \) is the fluid stress tensor which has contributions from both pressure and viscous stresses and \( \mathbf{n} \) is the unit normal direction from sphere surface into the fluid domain. The third side of equation (3) is known as the Stokes drag formula, and \( \mu, a \) and \( V \) denote fluid dynamic viscosity, radius of sphere and speed of the sphere related to the fluid, respectively. So, the drag force is proportional to \( V \). Therefore, we analyzed velocities in x and y directions and illustrated the different drag forces on the cell when the cell was entering different microgrooves.

2.4 Analyzing the cell rolling on the microchannel substrate

When the cell is initially adhering to the coated receptors of the surface, the adhesion force is important in stopping the cell rolling in contrast to fluid shear stress. Former experimental studies have discovered the adhesion force of different types of cells such as different leukocytes ligands and special proteins (receptors) in-vitro by aspiration force of micropipette (Ethier and Simmons 2007, Wang and Discher 2007). The normal stress distribution acting on the interface between cell ligands and surface receptors should be balanced by the fluid shear force resultant acting in the center of the cell in order to cell attachment. When resultant shear force due to fluid shear stress on the cell surface is higher than the micropipette aspiration force causing cell detachment, the cell will be detached from the surface (Ethier and Simmons 2007, Fung 2013). So, we analyzed the absolute cell detachment from surface by comparing experimental aspiration force with resultant shear force on the cell in different positions and conditions inside the microfluidic device.

2.5 Numerical simulation of fluid and the adhered cell interaction

2D finite element method was used to study the fluid-structure interaction (FSI) including fluid flow shear stress effect on adhered cells in microchannels with different microgrooves. The steady state Navier-Stokes equations for incompressible fluid were defined in equations (1) and
The cells were modeled elastic due to steady state flow, and the shear stress did not fluctuate by passing time. The cell attachment smoothing at the juncture with substrate was assumed by fillet of 0.2 cell radius (2µm)(Gaver and Kute 1998). The equation of viscous and pressure forces of fluid on the cell was defined as:

\[ \mathbf{F}_T = -\mathbf{n}. (P - \mu (\nabla \mathbf{u} + (\nabla \mathbf{u})^T)) \]  

where, \( \mathbf{n} \), \( \mathbf{F}_T \) and \( \mathbf{u} \) are the normal vector to the boundary, a sum of viscous and pressure forces and fluid velocity field, respectively(Païdoussis, Price et al. 2010).

In equation (5), \( \mathbf{F} \) illustrates the deformation gradient tensor which gives the relationship of a material line \( d\mathbf{X} \) before deformation to the line \( d\mathbf{x} \) after deformation, \( \nabla \) is the gradient operator with respect to , and \( \mathbf{u} \) is the displacement vector of the particle inside the body.

\[ F = \left( \frac{dx}{dx} \right)^T \equiv (\nabla x)^T = (\nabla u + I)^T \]  

Equations of stress on the cell can be defined as:

\[ d\mathbf{f} = \mathbf{S} \cdot d\mathbf{A} = F^{-1} \cdot d\mathbf{f} \]  

\[ J = \text{det}(F) \]  

\[ \mathbf{S} = J \cdot F^{-1} \cdot \sigma \cdot F^{-T} \]

where, the second Piola-Kirchhoff stress tensor (\( \mathbf{S} \)) is introduced as the stress tensor associated with the force \( d\mathbf{f} \) in the undeformed elemental area (\( d\mathbf{A} \)). In addition, the force \( d\mathbf{f} \) on the deformed elemental area (\( d\mathbf{a} \)) is related to the force \( d\mathbf{f} \) on the undeformed elemental area (\( d\mathbf{A} \)). In equation (8), the second Piola-Kirchhoff stress is related to Cauchy stress tensor \( \sigma \) (Reddy 2013).

The elasticity equations of the isotropic and symmetric cell can be defined as(Reddy 2013):

\[ \varepsilon_{ij} = \frac{1}{E} \left( (1 + \nu)\sigma_{ij} - \nu \delta_{ij}\sigma_{kk} \right) \]  

where \( \varepsilon_{ij} \), \( \sigma_{ij} \) and \( \sigma_{kk} \) are strain tensor, component of deviatoric stress tensor and hydrostatic stress tensor respectively(Reddy 2013). In this study, Poisson ratio and Young modulus of the cell were assumed as \( \nu = 0.3 \) and \( E = 300\text{Pa} \) (Fung 2013).

### 3 Results and Discussion

#### 3.1 Fluid flow recirculation area

Recirculation of fluid flow can have profound effect on the cells attachment to the coated proteins on the microchannel surface. This assumption was also proved experimentally in the former studies inside the microwells in a microfluidic channel(Khademhosseini, Ferreira et al.)
2006, Cioffi, Moretti et al. 2010). We assessed the recirculation area parameters in different dimensions of two kinds of microgrooves by numerical simulation.

Fig. 4 shows the fluid streamlines in different microgrooves. It is concluded that flow recirculation area inside the microgroove is related to the depth and width of microgroove. In the rectangular microgrooves, fluid recirculation can occur everywhere at the bottom of the microgrooves (Fig. 4A(i)) or only in the corners of the microgrooves (Fig. 4A(ii)). Recirculation at the all bottom of the microgroove can have a pivotal role in the cell attachment and the recirculation in the corner does not improve the cell attachment to the surface. In semicircular microgrooves, the recirculation area is at the bottom of microgroove but its height depends on the radius of the microgroove (Fig. 4B(i,ii)). If the recirculation area is deep and narrow inside the microgroove, the recirculation does not improve the cell attachment to the surface (Fig. 4B(ii)).

Fig. 5 shows our defined parameters of recirculation area in semicircular and rectangular microgrooves. Parameter (a) is the maximum height of recirculation area near the edge of the microgroove. Parameter (b) is the minimum height of the recirculation area in the center of the microgroove. Table 2 represents the recirculation parameters in the 6 microchannels with semicircular microgrooves which have different radiiuses. In the all assumed radiuses, we had recirculation area, and when the radius is lower than 12µm, the probability of cell docking inside the microgrooves is very low due to the assumed cell radius (10µm). The semicircular microgroove radius higher than 20µm not only have the possibility of two cells docking in x direction, but also have a narrow recirculation area (as shown in Fig. 4B(ii)). Table 2 also represents the recirculation parameters in the 10 microchannel with rectangular microgrooves which have different dimensions. The depth of the rectangles (from 12µm to 20µm) are equal to the radius of the semicircle microgrooves. It should be noted that for each depth of microgrooves, the width higher than maximum assumed widths does not have fluid flow recirculation at the all bottom surface of the microgrooves. We also assessed the deeper rectangular microgrooves, but they can whelm the cell because of high recirculation area.

Higher recirculation area can increase the possibility of cells separation from fluid flow and cells attachment to the surface receptors in microgrooves. In other words, higher recirculation area inside the microgroove can cause large separated region of fluid streamlines as well as cells separation from fluid flow. On the other hand, the height of recirculation area has an important role in delivering nutrients to the docked cells and cellular waste disposal which are important in cells lives. Higher recirculation area can whelm the larger volume of cells and reduce the delivering nutrients to the cells as well as cellular waste disposal. So, the recirculation areas’ heights should not be much high.

Maximum height of the recirculation area is close to edges of the microgroove. When the maximum height of recirculation area (a) is high, large portion of cell can experience the fluid flow recirculation which push the cell to the left side of microgroove. In other words, the negative velocity direction of the fluid flow in the recirculation area can push the cell to the left side of the microgroove. When the cell positions inside the microgroove, all surface of the cell should not be immersed in the recirculation flow due to receiving nutrients and materials, so the maximum height of recirculation area should be lower than cell diameter. According to the cell radius (10µm) and the attachment smoothing at the juncture with substrate by fillet of radius 2µm, the maximum recirculation height should be lower than 18µm. By comparing two assumed rectangular microgrooves with same depths and different widths, maximum and minimum heights of recirculation have a bit difference. In the following computations, in rectangular
microgrooves, we assume the microgroove with higher width between two different width with same depth mentioned in table 2 because the cell can experience longer time in a recirculation area which can lead to higher possibility of cell docking in a microgroove with large width. Therefore, due to lowering later computations we assume five rectangular dimensions from ten assumed dimensions in table 2.

3.2 Fluid drag force on the moving cell toward the microgroove

In order to compare the moving cell toward different geometries of microgroove, the drag forces in x and y directions are important. According to equation (3), by changing \( V \) in different microgrooves, the drag force on the cell will change. When the cell is entering the microgroove, changing velocity and streamlines pattern can play important role in changing drag forces. Fluid velocities in x and y directions have effect on the cell moving to bottom of the microgroove and its attachment. We assessed the velocity change in the streamline near the wall going into the microgroove in order to compare drag forces in different geometries. The origin of assumed streamline is near the edge of the microgroove which the cell bottom is located in this streamline. Other parallel streamlines which cause the cell motion, have the similar change to the assumed streamline change. As shown in Fig. 6, we assumed 5 points in the streamline entering the microgroove to assess the velocity change of fluid entering the different microgroove geometries. In all geometries, the assumed streamline entering inside each microgroove has 0.2\( \mu \)m distance from the edge of microgroove, so all selected points have same conditions in all geometries. The assumed points have about 2\( \mu \)m difference in y direction. The little velocity difference in y direction in all geometries can be negligible. Fig. 7A represents the column chart of the assumed streamline velocities in x direction (\( V_x \)) at five points in semicircular microgrooves. Fig. 7B represents the length of last point in assumed streamline in x direction that represents the length of cell moving inside different microgrooves in the same height of assumed points. Fig. 8A represents the column chart of the assumed streamline velocities in x direction (\( V_x \)) at five points in rectangular microgrooves, but in two smaller microgrooves the four points were assumed due to the lower depth of the assumed streamline. Fig. 8B represents the length of last point in assumed streamline in x direction except two smaller rectangular microgrooves. This length shows the assumed streamline pattern, which is related to the cell movement inside the microgroove.

Lower velocity in x direction causes lower drag force on the cell in x direction. If the drag force in x direction decreases, the cell will move toward the bottom of the microgroove due to the drag force in y direction and cell weight. So, this can help the cell attachment to the substrate. In addition, the lower velocity in x direction provided that the cell moving time in x direction will decrease, so the cell can experience longer time inside the microgroove which increase the probability of cell attachment.

In addition, when the ratio of length of last assumed point in x direction to width of microgroove is low, so the cell can move longer distance in x direction with low velocity inside the microgroove, and the cell can have longer time to move toward the bottom of the microgroove. Therefore, the probability of cell attachment can increase. As it is shown in Fig. 7A and Fig. 8A, in the x direction, the velocities of assumed streamline inside the rectangular microgrooves are
considerable larger than corresponding semicircular microgrooves due to the sharp corners of the rectangle, but velocities in y direction do not have considerable differences. Besides, in the rectangular microgrooves the ratio of length of last point in x direction to the width of microgroove is bigger than this ratio in semicircular microgrooves. By increasing the radius of semicircular microgrooves, the velocity in x direction decreases, and also the ratio of length of last point in x direction to the diameter of microgroove decreases. Therefore, it is more possible that the cell enter the recirculation area due to the lower drag force in x direction of larger semicircular microgroove.

We also evaluated the rectangular microgrooves with recirculation area at the corners of the microgrooves. The results showed that the velocity in assumed streamline was larger than the velocity in assumed rectangular microgrooves in Fig. 8A. Due to the very low recirculation area in semicircular microgroove with radius larger than 20µm, the possibility of initial cell adhesion inside these microgrooves can be very low. In addition, the velocity in x direction inside these microgroove is positive, and the moving cell experiences drag force in x position that can worsen the cell attachment to the surface.

3.3 Fluid and adhered cell interaction

After cell adhesion to the receptors coated on the surface, the fluid flow force can detach the cell from the surface, so the interaction of cell and flowing fluid is important. Therefore, fluid shear stress on the cell plays a pivotal role in cell detachment and direction of the flow affects the direction of the shear stress on the cell surface. Recirculating flow have negative shear stress on the cell, so the percentage of cell surface which experiences negative shear stress can be important in the cell deformation and detachment. In addition, average shear stress on the cell surface can be a representative for evaluating the cell detachment from surface. Fig. 9 represents the numerical simulation of adhered single cell and flowing fluid interaction. In Fig. 9, fluid velocity field, the recirculating fluid flow near the adhered cell and shear stress on the cell surface are shown. Fig. 9A and Fig. 9B illustrate the interaction of fluid flow and adhered cells in the rectangular and semicircular microgrooves which have the capacity of single cell docking in x direction. We also assessed the distances between the microgrooves, and when the gap between microgrooves is lower than 40µm, the fluid velocity fluctuates due to the interaction with the cell inside the microgroove, and it affects the shear stress on the cell inside the next microgroove. This also depends on the cell size, and if the cell size decreases, the distance between microgrooves can decrease.

We also studied the fluid interaction with attached cell in the flat microchannel. Former experimental studies have discovered the adhesion force of different types of cells such as different leukocytes ligands and special proteins (receptors) in-vitro. Different cells ligands and proteins bonds have broken when the micropipette aspiration force was between 45pN and 80pN (Ethier and Simmons 2007, Wang and Discher 2007). In the flat microchannel as well as the gap between microgrooves, cells surface experienced average shear stress about 96mPa. The resultant shear force in the center of the cell was 120.6pN which is larger than the detachment aspiration force. So, in this resultant shear force related to the inlet velocity, the cells cannot adhere to the flat surface, and the gaps between microgrooves can be empty of adhered cells. In fact, when the inlet velocity is between 10^{-4}m/s and 7\times10^{-4}m/s, the cells cannot attach to the gap between microgrooves, and also the resultant shear force on the cells inside microgrooves are lower than micropipette aspiration force causing detachment. For smaller inlet velocity, the cells
can attach to the gaps between microgrooves and the fluid velocity fluctuates due to the interaction with attached cells and effect the shear stress on the cell inside next microgroove. Besides, when the velocity decreases, the experiment time increases.

Fig. 10 and Fig. 11 represents the percentage of negative shear stress and average shear stress on the docked cell surface inside the rectangular and semicircular microgrooves, respectively. Due to the large differences of shear stress on the cell in different positions inside the semicircular microgroove, we assessed the cell and fluid interaction in 3 positions (left, right and middle) for the cell docking.

Lower average shear stress and negative shear stress percentage near 50% on the cell surface are in favor of the cell better attachment. According to the shear stress distribution on the cell surface, the negative and positive shear stresses are in the right and left sides of the docked cell. So, this stress distribution can cause low cell deformation in the direction of fluid flow. In semicircular microgrooves, the possibility of the cell attachment is high due to the cell docking in the left and right sides of the microgroove. In different rectangular microgrooves as shown in Fig. 10, the percentage of negative shear stress on the cell surface do not fluctuate. On the other hand, by increasing the width and depth of rectangular microgrooves, the average shear stress on the cell surface decrease. When the cell docks in the left side of the semicircular microgroove, the cell surface experiences lower average shear stress and higher percentage of negative shear stress. On the other hand, the right sided cell detachment is highly probable due to higher average shear stress (Fig. 11B).

4 Conclusion

The ratio of cell radius to the microgroove width and depth is important in possibility of cell docking. In this study, the microgroove sizes were assumed according to single cell docking with radius of 10µm. It is concluded that three parameters play pivotal role in the cell capturing by the coated receptors on the surface. First, maximum and minimum height of recirculation area should be high enough although the height of recirculation area should not be higher than adhered cell height (18µm). Second, the drag force in the flow direction exerted on the moving cell inside the microgroove should be low. Third, the cell should move slowly with long distance inside the microgroove. If the microgroove geometry has these three parameters, the cell attachment can be highly possible. Although in the large rectangular microgroove (s=20×35) the recirculation area is high enough, the drag force in x direction is high due to the large velocity in x direction. On the other hand, in the large semicircular microgroove (r=18 or 20 µm) in favor of cell docking, the recirculation area is large, and the drag force in x direction is low.

After initial adhesion of the cell, there can be two parameters against cell detachment. First, lower average shear stress on the cell surface is in favor of cell docking inside the microgroove. Second, the percentage of negative shear stress be near 50 percentages, and also negative and positive stresses distribution be on the left and right side of the cell surface are favorable for cell attachment. In semicircular microgrooves the negative stress distribution is on the right side of the cell due to the fluid recirculation area. On the contrary, in rectangular microgrooves the negative shear stress distribution is on the left and right sides at the bottom of the cell; therefore, the top of the cell experiences positive shear stress in flow direction leading to high probability
of cell deformation. When the cell is captured in the recirculation area, the flow direction changes; therefore, the possibility of cell docking in the left side of the microgrooves may be higher than other sides. In semicircular microgrooves, docked cells in the left side not only experience lower average shear stress, but also have large percentage of negative shear stress; therefore the probability of cell detachment can be low.

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Table 1. Dimensions of assumed microgrooves at the bottom wall of microfluidic devices

| Semicircular microgroove | Rectangular microgroove dimensions |  |
|--------------------------|-----------------------------------|---|
| radius (µm)              | (Depth×Width (µm²))               |  |
| r=12                     | s=12×20                           |  |
|                          | s=12×24                           |  |
| r=14                     | s=14×28                           |  |
|                          | s=14×22                           |  |
| r=16                     | s=16×32                           |  |
|                          | s=16×26                           |  |
| r=18                     | s=18×35                           |  |
|                          | s=18×29                           |  |
| r=20                     | s=20×35                           |  |

Table 2. Maximum and minimum height of recirculation area inside semicircular microgrooves with different radiuses, and rectangular microgrooves with different dimensions

| Semicircular microgroove radius (µm) | a (µm) | b (µm) |
|--------------------------------------|--------|--------|
| 12                                   | 10.4   | 5.8    |
| 14                                   | 11.3   | 6.2    |
| 16                                   | 12.2   | 6.4    |
| 18                                   | 13.5   | 7.1    |
| 20                                   | 14.2   | 5.6    |

| Rectangular microgroove dimensions (depth×width µm²) | a (µm) | b (µm) |
|-----------------------------------------------------|--------|--------|
| 12×20                                                | 11     | 8.5    |
| 12×24                                                | 11     | 6.8    |
| 14×22                                                | 13.8   | 7.9    |
| 14×28                                                | 12.5   | 7.6    |
| 16×26                                                | 14.6   | 11.7   |
| 16×32                                                | 14.6   | 11.7   |
| 18×29                                                | 16.4   | 12.7   |
| 18×35                                                | 16.4   | 12.7   |
| 20×35                                                | 17.7   | 12.0   |
**Figure 1.** 3D view of two microchannels with different microgrooves. A) Rectangular microgrooves. B) Semicircular microgrooves.

**Figure 2.** Mesh structures with triangular elements for microchannel with rectangular microgrooves with depth of 20µm and width of 35µm.

**Fig. 3** The force diagram on the moving cell in the fluid (Duprat and Shore 2015)
Fig. 4 Fluid streamlines and recirculation area inside the semicircular and rectangular microgrooves. The flow direction and assumed coordinates x and y are shown. In each microgroove, the flow direction is shown by arrows. A) Rectangular microgroove dimensions (depth×width) i) 14×35 µm², ii) 16×26 µm². B) Semicircular microgroove radius i) 18µm, ii) 24µm.

Fig. 5 Velocity streamline showing the fluid flow recirculation area inside the semicircular and rectangular microgrooves. (a) and (b) are the recirculation area parameters.
Fig. 6 Five assumed points in the streamline entering the microgroove. A) The semicircular microgroove with radius 18µm. B) The rectangular microgroove with depth 18µm and width 35µm. Both sample microgrooves have same depth and area.
**Fig. 7** A) Velocity of fluid at five points in the assumed streamline in semicircular microgrooves in x direction. B) Length of last point in the assumed streamline in semicircular microgrooves in x direction.
Fig. 8  A) Velocity of fluid at the points in the assumed streamline in rectangular microgrooves in x direction. B) Length of last point in the assumed streamline in rectangular microgrooves in x direction.
Fig. 9 Simulation of fluid flow and the cell interaction including fluid velocity field, Shear stress on the cell surface and velocity streamlines in A) Rectangular microgrooves with depths of 24µm and width of 45µm. B) Semicircular microgrooves with radiuses of 18µm. The left color bar displays the shear stress between -0.2Pa and 0.2Pa, and the right color bar displays the velocity in x direction.

Fig. 10 In rectangular microgrooves: A) Percentage of negative shear stress on the cell surface. B) Average shear stress on the cell surface.
Fig. 11 In semicircular microgrooves: A) Percentage of negative shear stress on the cell surface in three possible position of the cell. B) Average shear stress on the cell surface in three possible position of the cell.