RESEARCH ARTICLE

Printing channels with millimeter-scale curvature and deciphering their effect on the proliferation, morphology, orientation, and migration of M-22 cells

Supplementary File

Computational model description

In our study, the model for the cell migration simulation was modified based on the model described in reference [1], which was used to simulate the cell migration on confined 2D square region. The boundary and parameters were amended based on our experiment. The cells were described as a circle with a diameter of 20 μm corresponding to the width of M-22 cells.

The geometric shape of the boundary with radius R in the Cartesian coordinate system was described as Equation S1:

$$x^2 + y^2 = R^2 \ (x < 0, y > 0),$$
(S1)

R was taken as 1.5, 2, 2.5, and 3, respectively.

The calculation of cell migration was based on the force balance of the cell–cell, cell–substrate, and cell–boundary interactions on the node of the cells. The resultant force can be calculated as Equation S2:

$$\mathbf{F}_{\text{resultant}} = \boldsymbol{F}_{mem} + \boldsymbol{F}_{mod} + \boldsymbol{F}_{ext} + \boldsymbol{F}_{cell},$$
(S2)

where $\boldsymbol{F}_{mem}$ denotes the cell membrane tension; $\boldsymbol{F}_{mod}$ represents the substrate-derived forces acting on modules; $\boldsymbol{F}_{ext}$ is the external forces derived from the collision of the cell to cell and cell to boundary; $\boldsymbol{F}_{cell}$ is the substrate-derived forces acting on the central cell body. The quantities expressed in Equation S2 are all vectors.

$\boldsymbol{F}_{mem}$ was calculated as Equation S3:

$$\boldsymbol{F}_{mem} = \frac{\pi \beta L_{cell} \cos(\theta)}{2\gamma \sin(\alpha)}.$$
(S3)

$\boldsymbol{F}_{mod}$ was calculated as Equation S4:

$$\boldsymbol{F}_{mod} = \left\{ \frac{1}{\dfrac{1}{K_{cell}} + \dfrac{N_{on}}{K_{clu}} + \dfrac{1}{K_{sub}}} \right\} \bullet \sum_{j=1}^{N_{on}} x_{c,j}.$$
(S4)

$\boldsymbol{F}_{ext}$ was calculated as Equation S5:

$$\boldsymbol{F}_{ext} = \frac{K_{cell}}{2} \times S.$$
(S5)

$\boldsymbol{F}_{cell}$ was calculated as Equation S6:

$$\boldsymbol{F}_{cell} = \left\{ \frac{1}{\dfrac{N_{onc}}{K_{clu}} + \dfrac{1}{K_{sub}}} \right\} \bullet \sum_{i=1}^{N_{cc}} x_{c,i},$$
(S6)

The value and meaning of variables that appear in Eqs. S3–6 in simulation were illustrated in **Table S1**.

Table S1. Parameters setting in simulation

| Parameter	Description	Value
K_{sub}	Substrate spring stiffness	10^9 pN/nm
K_{cell}	Cell spring constant	30 pN/nm
K_{clu}	Module clutch spring constant	0.8 pN/nm
$K_{on,j}$	Module clutch on-rate constant	1/s
$K_{off,j}$	Module clutch off-rate constant	0.1/s
K_{mod}	Base module birth rate	0.5/s
K_{cap}	Module capping rate	0.001/s
Γ	Capping rate of membrane	1/s
B	Total number of nascent filaments	5/s
A	The range of angles affected by a single pseudopod	35°
V_m	Unloaded motor velocity	120 nm/s
V_{poly}	Maximum polymerization rate	125 nm/s
N_{motor}	Total number of motors shared by modules	1000
N_{clutch}	Total number of clutches shared by modules	750
N_{total}	Total available actin pool in polymerized length	10^5 nm
L_m	Minimum module length	100 nm
L_{cell}	Initial length of a module	10000 nm
T	Membrane force per unit length	1 pN/nm
F_b	Module clutch bond breaking force	2 pN
F_{stall}	Motor stall force	2 pN
θ	The angle between the node and the x-axis	Change with iteration
N_{on}	The number of engaged clutches of the ith modules	
$x_{c,j}$	The extension of the jth clutch of the ith module	
S	The sum of cell-to-cell and cell-to-boundary intersection area	
N_{cc}	The number of engaged clutches of cell	
$x_{c,i}$	The extension of the ith clutch of cell	
N_{onc}	The number of engaged clutches of cell	

Simulation program for simulating cell migration

The programs for cell migration simulation were packed in the folder "Simulation program." The description of the program was shown in **Table S2**.

Table S2. Description of the simulation program

Program name	Corresponding function
Main.m	The main program. All of the calculation started from this file. Initialize the position of cells.
CellMigrationSimulator	Initialize the parameters for clutch, modules, and cells.
RecordOV.m	Record parameters of the subject at iterations.
Time2Next.m	Calculate time step in each iteration.
NextEvent.m	Determines the next event to be occurred.
TranPoly.m	Calculate the degree of aggregation after aggregation and the updated position of pseudopodia after aggregation.
Execute.m	Execute the determined event.
ForceBalance.m	Calculate the cell positions after the force balance.
plotcell.m	Plot results.
auxpoint.m	Plot cell boundary.
collision.m	Calculate the external force from the overlap and add the force calculation for the next iteration.
getside.m	Set boundary conditions.
Plottrajectory.m	Output the migration speed, cell position, force, cell and boundary intersection area, and count of invalid modules at each step.

A)

B)
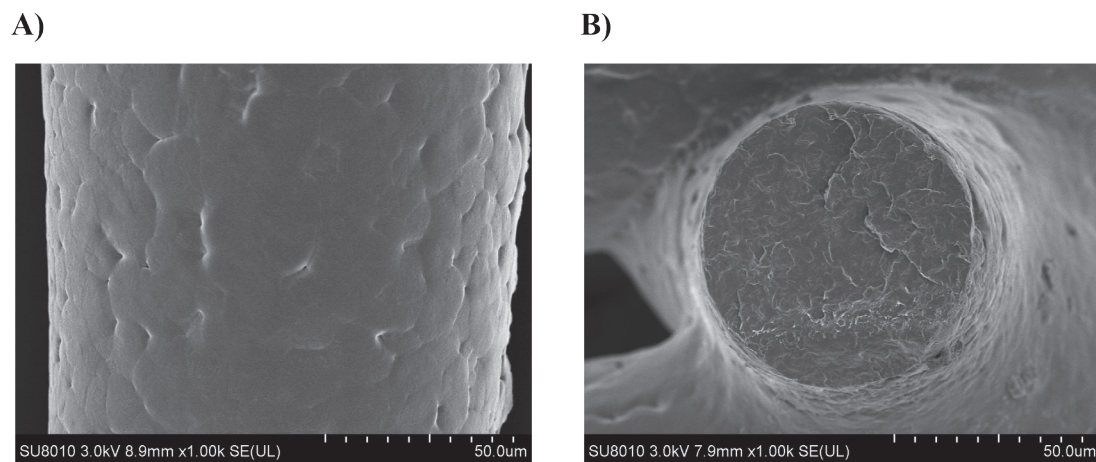
Figure S1. SEM images of printed fibers. (A) Surface image of the printed fiber. (B) Cross-section image of the printed fiber.
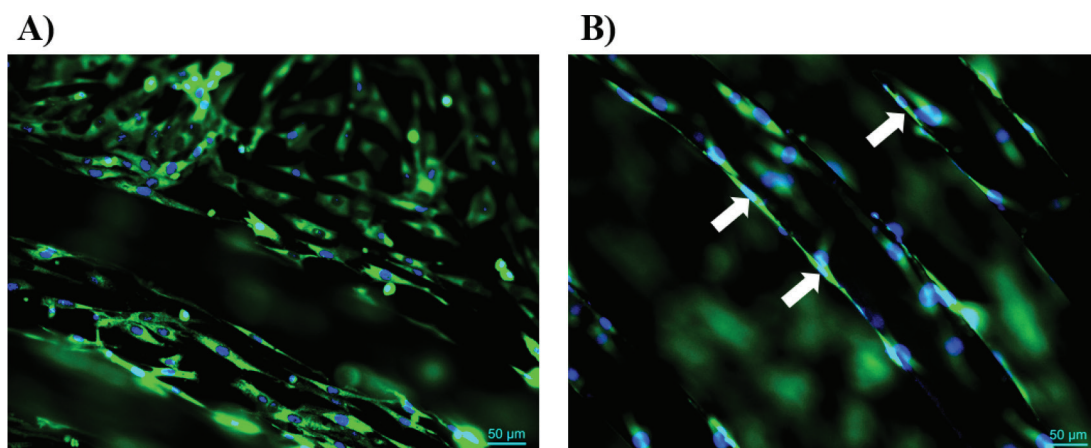
Figure S2. Fluorescence images of fibronectin (green). (A) Adhesion proteins on the bottom surface. (B) Adhesion proteins on the side wall.

Other files:

Video S1. Time-lapse video of cell migration in the curved channels with radius R = 1.5–2 mm.
Video S2. Time-lapse video of cell migration in the curved channels with radius R = 2–3 mm.
Video S3. Time-lapse video of cell migration in the straight channels.
Video S4. Simulation video of cell migration in the curved channel with radius R = 1.5 mm.
Video S5. Simulation video of cell migration in the curved channel with radius R = 2 mm.
Video S6. Simulation video of cell migration in the curved channel with radius R = 2.5 mm.
Video S7. Simulation video of cell migration in the curved channel with radius R = 3 mm.
Video S8. Simulation video of cell migration in the straight channel.

Reference

1. Chang C, Dai Z, Shih P, 2022. Modeling and simulation of cell migration on the basis of force equilibrium. Int J Numer Method Biomed Eng, 38:e3550.