Collective behavior of living cells on patterned substrate

Haosheng Wen

Follow this and additional works at: https://digitalcommons.memphis.edu/etd

Recommended Citation
Wen, Haosheng, "Collective behavior of living cells on patterned substrate" (2021). Electronic Theses and Dissertations. 2196.
https://digitalcommons.memphis.edu/etd/2196

This Thesis is brought to you for free and open access by University of Memphis Digital Commons. It has been accepted for inclusion in Electronic Theses and Dissertations by an authorized administrator of University of Memphis Digital Commons. For more information, please contact khggerty@memphis.edu.
COLLECTIVE BEHAVIOR OF LIVING CELLS ON PATTERNED SUBSTRATES

by

Haosheng Wen

A Thesis
Submitted in Partial Fulfillment of the Requirements for the Degree of

Master of Science

Major: Physics

The University of Memphis

August 2021
I express my sincere appreciation to Professor Mohamed Laradji for being a mentor in my research and life. Without his wise guidance and continuous encouragement, I can't imagine how this thesis could be completed. I am also very grateful to him for helping me develop a rigorous attitude towards physics.

I would like to thank Professor Chenghui Pen and Professor Sunil Kumar for deep discussions. I thank all of the fellow students in our lab, especially Eric J Spangler for patient instructions on C++, and Kumar, Yu, and Abash for friendship. It was lucky for me to have all of you in the past two years!

Finally, I express my very deep gratitude to Hongchun Wang, my father and to Kefang Wu, my mother, who always encourage and love me, and to Shiran Yu, my girlfriend, who firmly stand by my side and support me, and to my three lovely little kittens and one cute puppy, Mimi, Taozi, Huahua, and Naonao.
ABSTRACT

Individual units of many biological systems can produce cooperative behaviors through direct interactions between individuals as well as interactions between the surrounding media and individuals. This cooperativity is characterized by amazing collective dynamical and structural behaviors at large scales. In this work, we use molecular dynamics simulations of a ring polymer model, to describe biological cells, to systematically investigate the collective behavior of disjoint semi-flexible cells and their dynamic assembly behavior. We particularly investigate the effect of cell motility, the areal density of the cells, and their interactions with the substrate on their collective behavior. Our results show that the collective behavior of cells strongly depends on an interplay among the driving force of cells, the strength of the circularly patterned substrate, and areal density. A rich phenomenology and dynamics of collective cells motion of cells is revealed from our simulations, including giant traveling bands, unidirectional vortices, and dynamic phase transitions from non-collective to collective behavior as a function of cells motility and areal density. Our study provides novel non-trivial insight into biological processes associated with cell motility, stiffness of the substrate, and cell areal density.
# Table of contents

LIST OF FIGURES...vii

1. INTRODUCTION.................................................................1

2. LITERATURE REVIEW..........................................................5
   2.1 Cellular Potts model .........................................................5
   2.2 Phase-field models..........................................................7
   2.3 Particle models .............................................................9
   2.4 Active network models: Vertex and Voronoi models .................10

3. RING-POLYMER MODEL AND QUANTIFICATIONS...........................13
   3.1 Theoretical model............................................................13
   3.2 Quantification methods....................................................17
   3.3 Schematics and table of parameters.....................................19

4. RESULTS ON UNIFORM SUBSTRATE ........................................24
   4.1 Simulations in the system with periodic boundary condition........24
      4.1.1 Effect of increasing driving force ....................................24
      4.1.2 Effect of increasing density ...........................................29
   4.2 Simulations in circular confinement.......................................33
      4.2.1 The effect of increasing driving force ................................33
      4.2.2 The effect of increasing density .......................................40
5. RESULTS IN CIRCULAR CONFINEMENT WITH PATTERNED SUBSTRATE .................................................46

5.1 The effect of driving force ..................................................................................................................46

5.2 The effect of stiffness of substrate ..................................................................................................55

5.3 The effect of density ........................................................................................................................62

6. RESULTS IN PERIODIC BOX WITH PATTERNED SUBSTRATE .........................................................69

6.1 The effect of driving force ..................................................................................................................69

6.2 The effect of stiffness of substrate ..................................................................................................78

6.3 The effect of density .........................................................................................................................85

7 CONCLUSIONS AND SUMMARY ....................................................................................................91

REFERENCE ..............................................................................................................................................Error! Bookmark not defined.
LIST OF FIGURES

Figure 1. Lattice Models: The Cellular Potts Model. (a), Lattice sites corresponding to two different cells are shown in different colors. Cell-cell and cell-medium interfaces have an interfacial energy $\alpha$ (black). Cell migration in the direction of the polarity is favored by a self-propulsion magnitude $P$. (b), Snapshots of the system and cell trajectories at the fluid ($\alpha = 1$) and solid ($\alpha = 4$) regimes. Neighboring cells are colored differently, with arbitrary colors. Cell boundaries are rougher and longer for smaller interfacial energy. The figure is adapted from ref. [32].................................................................6

Figure 2. Phase-field models a, Phase field of a cell. Adapted from [38]. b, The overlap between phase field, cell-cell interfaces are identified in white. The inset shows cell contours, $\phi_i = 0.5$. Adapted from [39]. Copyright (2019) by the American Physical Society. c-d, Collisions between deformable cells lead to velocity alignment(c) and collective motion(d). Adapted from [43]..................................................................................8

Figure 3. Particle models. a, Schematic representation of forces in particles models. Cells may experience various amounts of cell-cell repulsion(blue), adhesion(orange), cell-substrate active traction(red), or passive friction(green). Adapted from [46]. b, The CIL repolarization occurred in small cell cluster finally leads to the collective motion. The color code indicates the cell polarity angle $\theta_i$. Adapted from [47]......................................................................................................10

Figure 4. Active Network Models. a, Cell centers with positions $\{r_i\}$ are connected by the Delaunay triangulation(black). Voronoi tessellation(red) defines cell boundaries and vertices at position $\{\hat{h}\}$. Reprinted from [53]. b, Cells are confined by an area $A_i$ and a perimeter $P_i$. Net force of the cell motion can be calculated by the sum of a self-propulsion force $T_aP_i$ (orange) and an interaction force $F_i^{\text{int}} = -\nabla r_i F$ (dashed black). Adapted from [56]. c, Polarity-velocity alignment with a time scale $\tau$. Adapted from [56]. d, Phase diagram of the fluid-solid transition in the SPV model, in which shape index $p_0 = P_0/\sqrt{A_0}$ and self-propulsion speed $v_0 = T_a/\xi$ with persistence $D_\tau^{-1}$. Adapted from [53]. e, Schematic phase diagram of the SPV model with polarity-velocity alignment. Adapted from [58]..................................................................................................................12

Figure 5. Initial composition of single cell before polarization. Total number of monomers is 40. The length of spring that connects monomers $1r_b$. The initial radius $r \approx 6.37r_b$. Angle between two monomers $\theta = 9^\circ$. The bending angle of each monomer $\theta_b \approx 180^\circ$.............20

Figure 6. Initial configurations of cells without polarization moving in circular confinement (a) and periodic box (b), respectively. For circular confinement, the radius $r = 200r_b$. The length of periodic box $L = 400r_b$. ..................................................................................................................21

Figure 7. substrate force induced by circularly patterned substrate. The polarization of the cell is forced to rotate towards the tangent line of the curve. .................................................................21

Figure 8. Clustering of cells in circular confinement (a) and periodic box (b), respectively. If the distance of any monomers of two cells is less than $2r_b$, these two cells are considered in the same cluster. Different color represents different clusters. The circularly patterned substrate is located on the central region of systems. For circular confinement, the radius $r = 200r_b$. 
The length of periodic box $L = 400r_b$. In both two systems, the radius of patterned region $r_c = 100r_b$. Figure 9. Snapshots of central vortex induced by the substrate force in circular confinement (a) and periodic box (b). In the middle region, cells are self-organized into a rotating vortex at a moderate driving force of cells and strength of substrate. Figure 10. Configuration snapshots and velocity fields of the cells for $k_{mo} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1$, and $1.2k_B T/r_b$ at a given density $\rho = 0.003125 r_b^{-2}$. The length of the periodic box equals $400r_b$ as a measure of length. Figure 11. Quantifications of areal density distribution and average speed of the system. a, Areal density of the system as a function of radius for $k_{mo} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1$, and $1.2k_B T/r_b$ at a given density $\rho = 0.003125 r_b^{-2}$. b, Average speed of the system as a function of radius for $k_{mo} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1$, and $1.2k_B T/r_b$ at a given density $\rho = 0.003125 r_b^{-2}$. c, Average size of clusters as a function of driving force at a given motility force $k_{mo} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1$. The radius of the circular confinement $r = 200r_b$ as a measure of length. The radius of the periodic box $L = 400r_b$. Figure 12. Quantifications of clustering properties of the system. a, Average total number of clusters as a function of driving force coefficient at a given density $\rho = 0.003125 r_b^{-2}$. b, Average maximum size of clusters as a function of driving force coefficient at a given density $\rho = 0.003125 r_b^{-2}$. c, Average size of clusters as a function of driving force coefficient at a given density $\rho = 0.003125 r_b^{-2}$. Figure 13. Nematic order parameter of the systems of 250 cells and 500 cells. $\rho = 0.003125 r_b^{-2}$. Figure 14. Configuration snapshots and velocity fields of the cells for $\rho = 0.002188, 0.002500, 0.002813$, and $0.003125 r_b^{-2}$ at a given driving force $k_{mo} = 1k_B T/r_b$. The length of the periodic box equals $400r_b$ as a measure of length. Figure 15. Quantifications of areal density distribution and average speed of the system with increasing density. a, Areal density of the system as a function of radius for for $\rho = 0.001875, 0.002188, 0.0025, 0.002813, 0.003125$, and $0.003438 r_b^{-2}$ at a given driving force $k_{mo} = 1k_B T/r_b$. b, Average speed of the system as a function of radius for for $\rho = 0.001875, 0.002188, 0.0025, 0.002813 0.003125$, and $0.003438 r_b^{-2}$ at a given driving force $k_{mo} = 1k_B T/r_b$. Figure 16. Quantifications of clustering properties of the system. a, Average total number of clusters as a function of density at a given motility force $k_{mo} = 1k_B T/r_b$. b, Average maximum size of clusters as a function of driving force at a given motility force $k_{mo} = 1k_B T/r_b$. c, Average size of clusters as a function of driving force at a given motility force $k_{mo} = 1k_B T/r_b$. Figure 17. Nematic order parameter of the system at a given driving force coefficient $k_{mo} = 1k_B T/r_b$. Figure 18. Configuration snapshots and velocity fields of the cells for $k_{mo} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1$, and $1.2k_B T/r_b$ at a given density $\rho = 0.003981 r_b^{-2}$. The radius of the circular confinement $r = 200r_b$ as a measure of length. Figure 19. Quantifications of the effect of increasing driving force coefficient on areal density distribution and average speed of the system in circular confinement. a, Areal density of the
system as a function of radius for \( k_{mo} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1, \) and \( 1.2k_B T/r_b \) at a given density \( \rho = 0.003981r_b^{-2} \). b, Average speed of the system as a function of radius for \( k_{mo} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1, \) and \( 1.2k_B T/r_b \) at a given density \( \rho = 0.003981r_b^{-2} \). 37

Figure 20. Quantifications of clustering properties of the system. a, Average total number of clusters as a function of driving force at a given density \( \rho = 0.003981r_b^{-2} \). b, Average maximum size of clusters as a function of driving force at a given density \( \rho = 0.003981r_b^{-2} \). c. Average size of clusters as a function of driving force at a given density \( \rho = 0.003981r_b^{-2} \).………………………………………38

Figure 21. Tangential velocity of 38th layer of the system vs. driving force at \( k_{mo} \geq 0.6kBT/rb \). Density \( \rho = 0.003981r_b^{-2} \) and the radius of circular confinement is 200r.…..38

Figure 22. Vortical order parameter as a function of driving force coefficient of the system in circular confinement. The density \( \rho = 0.003981r_b^{-2} \)………………………………………40

Figure 23. Configuration snapshots and velocity fields of the cells for \( \rho = 0.002787, 0.003125, 0.003583, \) and \( 0.003981 \) at a given driving force \( k_{mo} = 1k_B T/r_b \). The radius of the circular confinement \( r = 100r_b \)……………………………………………………………..41

Figure 24. Quantifications of areal density distribution and average speed of the system with increasing density. a, Areal density of the system as a function of radius for for \( \rho = 0.001991, 0.002389, 0.002787, 0.003185, 0.003583, \) and \( 0.003981r_b^{-2} \) at a given driving force \( k_{mo} = 1k_B T/r_b \). b, Average speed of the system as as a function of density at a given driving force \( k_{mo} = 1k_B T/r_b \)…………………………………………………………………………..42

Figure 25. Quantifications of clustering properties of the system. a, Average total number of clusters as a function of density at a given motility force \( k_{mo} = 1k_B T/r_b \). b, Average maximum size of clusters as a function of driving force at a given motility force \( k_{mo} = 1k_B T/r_b \). c. Average size of clusters as a function of driving force at a given motility force \( k_{mo} = 1k_B T/r_b \)………………………………………………………………………….43

Figure 26. Tangential velocity of 38th layer of the system vs. density at \( k_{mo} = 1k_B T/r_b \). The radius of circular confinement is 200r.……………………………………………………………..44

Figure 27. Vortical order parameter as a function of density at a given driving force \( k_{mo} = 1k_B T/r_b \)………………………………………………………………………………………………..45

Figure 28. Configuration snapshots and corresponding velocity fields of the cells for \( k_{mo} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1, \) and \( 1.2k_B T/r_b \) at a given strength of substrate \( k_r = 100kBT \) and density \( \rho = 0.003979 \). The radius of circular confinement is 200r as a measure of length, while the radius of the patterned region is 100r. Both the system and the substrate are centered at the coordinates (200, 200)…………………………………………………………………………………………………..50

Figure 29. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at different values of driving force coefficient. The substrate force coefficient \( k_r = 100kBT \) and density \( \rho = 0.003979r_b^{-2} \). (b) Average speed of system as a function of driving force. The substrate force coefficient \( k_r = 100kBT \) and density \( \rho = 0.003979r_b^{-2} \)….51
Figure 30. Reversal rate (number of reversals per unit time), versus driving force coefficient, in layer 18 and layer 39. The substrate force coefficient \( k_r = 100kBT \) and density \( \rho = 0.003979r_b^{-2} \). 

Figure 31. (a) Average size of clusters (average number of cells per cluster) of the system as a function of driving force coefficient at a given substrate force coefficient \( k_r = 100kBT \) and density \( \rho = 0.003979r_b^{-2} \). (b) Maximum size of cluster (number of cells in biggest cluster) of the system as a function of driving force coefficient at a given substrate force coefficient \( k_r = 100kBT \) and density \( \rho = 0.003979r_b^{-2} \). (c) Number of clusters as a function of driving force coefficient at a given substrate force coefficient \( k_r = 100kBT \) and density \( \rho = 0.003979r_b^{-2} \). 

Figure 32. Vortical order parameter as a function of substrate force coefficient \( k_r = 100kBT \) and density \( \rho = 0.003979r_b^{-2} \). 

Figure 33. Configuration snapshots and velocity fields of the cells for \( k_r = 0, 40, 80, 100, 140, 180kBT \) at a given motility force \( k_m = 0.5k_BT/r_b \) and density \( \rho = 0.003979r_b^{-2} \). The radius of circular confinement is \( 200r_b \) as a measure of length, while the radius of the patterned region is \( 100r_b \). Both the system and the substrate are centered at the coordinates \((200, 200)\). 

Figure 34. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at different values of substrate force coefficient. The driving force coefficient \( k_m = 0.5k_BT/r_b \) and density \( \rho = 0.003979r_b^{-2} \). (b) Average speed of system versus radius (the distance between cell and origin point), at different values of substrate force coefficient. The driving force coefficient \( k_m = 0.5k_BT/r_b \) and density \( \rho = 0.003979r_b^{-2} \). 

Figure 35. Reversal rate (number of reversals per unit time), versus substrate force coefficient, in layer 18. The driving force coefficient \( k_m = 0.5k_BT/r_b \) and density \( \rho = 0.003979r_b^{-2} \). 

Figure 36. (a) Average size of clusters (average number of cells per cluster) of the system as a function of substrate force coefficient at a given driving force coefficient \( k_m = 0.5k_BT/r_b \) and density \( \rho = 0.003979r_b^{-2} \). (b) Maximum size of cluster (number of cells in biggest cluster) of the system as a function of substrate force coefficient at a given driving force coefficient \( k_m = 0.5k_BT/r_b \) and density \( \rho = 0.003979r_b^{-2} \). (c) Number of clusters as a function of substrate force coefficient at a given driving force coefficient \( k_m = 0.5k_BT/r_b \) and density \( \rho = 0.003979r_b^{-2} \). 

Figure 37. Vortical order parameter as a function of substrate force coefficient at a given driving force coefficient \( k_m = 0.5k_BT/r_b \) and density \( \rho = 0.003979r_b^{-2} \). 

Figure 38. Configuration snapshots and velocity fields of the cells for \( \rho = 0.002787, 0.003185, 0.003583, 0.003981 \) at a given driving force coefficient \( k_m = 0.5k_BT/r_b \) and substrate coefficient \( k_r = 100kBT \). The radius of circular confinement is \( 200r_b \) as a measure of length, while the radius of the patterned region is \( 100r_b \). Both the system and the substrate are centered at the coordinates \((200, 200)\). 

Figure 39. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at density. The driving force coefficient \( k_m = 0.5k_BT/r_b \) and substrate force coefficient \( k_r = 100kBT \) and density \( \rho = 0.003979r_b^{-2} \).
coefficient $k_r = 100k_BT$. (b) Average speed of system versus radius (the distance between cell and origin point), at different density. The driving force coefficient $k_{mo} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$ ..........................................................65

Figure 40. Reversal rate (number of reversals per unit time), versus density, in layer 18. The driving force coefficient $k_{mo} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$ ..................................................66

Figure 41. (a) Average size of clusters (average number of cells per cluster) of the system as a function of density at a given driving force coefficient $k_{mo} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. (b) Maximum size of cluster (number of cells in biggest cluster) of the system as a function of density at a given driving force coefficient $k_{mo} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. (c) Number of clusters as a function of density at a given driving force coefficient $k_{mo} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$ ..............................................................................................................67

Figure 42. Vortical order parameter as a function of density at a given driving force coefficient $k_{mo} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$ ..........................................................68

Figure 43. Configuration snapshots and corresponding velocity fields of the cells for $k_{mo} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1,$ and $1.2k_BT/r_r$ at a given substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003125r_b^{-2}$. The length of the periodic box equals $400r_b$ as a measure of length, while the radius of the patterned region is $100r_b$. Both the system and the patterned substrate are centered at the origin (200, 200) ........................................................................................................73

Figure 44. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at different values of driving force coefficient. The substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003125r_b^{-2}$. (b) Average layer speed (average speed per cell in the layer), versus radius at different values of driving force. The substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003125r_b^{-2}$. ........................................................................................................74

Figure 45. Reversal rate (number of reversals per unit time), versus driving force coefficient, in layer 18. The substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003125r_b^{-2}$. ........................................................................................................75

Figure 46. (a) Average size of clusters (average number of cells per cluster) of the system as a function of driving force coefficient at a given substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003125r_b^{-2}$. (b) Maximum size of cluster (number of cells in biggest cluster) of the system as a function of driving force coefficient at a given substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003125r_b^{-2}$. (c) Number of clusters as a function of driving force coefficient at a given substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003125r_b^{-2}$. ........................................................................................................77

Figure 47. Normalized tangential momentum (see method), versus radius for different values of driving force coefficient in the system with periodic boundary condition. The substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003125r_b^{-2}$ ........................................................................................................78

Figure 48. Configuration snapshots and velocity fields of the cells for $k_r = 0, 40, 80, 100, 140, 180k_BT$ at a given motility force $k_{mo} = 0.5k_BT/r_b$ and density $\rho = 0.003125r_b^{-2}$. The length of the periodic box is $400r_b$ as a measure of length, while the radius of the patterned region is $100r_b$. Both the system and the substrate are centered at the origin (200, 200) ........................................................................................................80
Figure 49. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at different values of substrate force coefficient. The driving force coefficient $k_m = 0.5k_BT/r_b$ and density $\rho = 0.003125r_b^{-2}$. (b) Average layer speed (average speed per cell in the layer), versus radius at different values of substrate force coefficient. The driving force coefficient $k_m = 0.5k_BT/r_b$ and density $\rho = 0.003125r_b^{-2}$ ..................................81

Figure 50. Reversal rate (number of reversals per unit time), versus substrate force coefficient, in layer 18. The driving force coefficient $k_m = 0.5k_BT/r_b$ and density $\rho = 0.003125r_b^{-2}$.82

Figure 51. (a) Average size of clusters (average number of cells per cluster) of the system as a function of substrate force coefficient at a given driving force coefficient $k_m = 0.5k_BT/r_b$ and density $\rho = 0.003125r_b^{-2}$. (b) Maximum size of cluster (number of cells in biggest cluster) of the system as a function of substrate force coefficient at a given driving force coefficient $k_m = 0.5k_BT/r_b$ and density $\rho = 0.003125r_b^{-2}$. (c) Number of clusters as a function of substrate force coefficient at a given driving force coefficient $k_m = 0.5k_BT/r_b$ and density $\rho = 0.003125r_b^{-2}$. .................................................................84

Figure 52. Normalized tangential momentum (see method), versus radius for different values of substrate force coefficient in the system with periodic boundary condition. The driving force coefficient $k_m = 0.5k_BT/r_b$ and density $\rho = 0.003125r_b^{-2}$ ........................................84

Figure 53. Configuration snapshots and velocity fields of the cells for $\rho = 0.002188, 0.0025, 0.002813, \text{and} 0.003125$ at a given driving force coefficient $k_m = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. The length of the periodic box is $400r_b$ as a measure of length, while the radius of the patterned region is $100r_b$. Both the system and the substrate are centered at the origin $(200,200)$. .................................86

Figure 54. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at different densities. The driving force coefficient $k_m = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. (b) Average layer speed (average speed per cell in the layer), versus radius at different densities. The driving force coefficient $k_m = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. .........................................................87

Figure 55. Reversal rate (number of reversals per unit time), versus radius (the distance between cell and origin point), at different densities. The driving force coefficient $k_m = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. .................................................................89

Figure 56. (a) Average size of clusters (average number of cells per cluster) of the system as a function of density at a given driving force coefficient $k_m = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. (b) Maximum size of cluster (number of cells in biggest cluster) of the system as a function of density at a given driving force coefficient $k_m = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. (c) Number of clusters as a function of density at a given driving force coefficient $k_m = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. .................................................................................................90

Figure 57. Normalized tangential momentum (see method), versus radius for different densities. The driving force coefficient $k_m = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. ..................................................90
1. INTRODUCTION

As an emerging scientific research hot point, collective motion is widely present in a plethora of life phenomena, such as the social collaboration of insects including ants and bees [1], the centripetal movement of fish schools and bird flocks [2], and swarms of locusts [3]. In addition to these living systems, scientists have also investigated the collective motion formed by artificially synthesized self-propelled units such as discs or bars [4] [5] [6]. Regardless of whether it is a living system or a man-made system, although the individual manifestations in these systems are not quite the same, they all have a common feature: Individual units can produce cooperative behaviors through direct interactions with each other and with medium, exhibiting interesting collective dynamic behaviors on a large scale. These interacting self-propelled particles are termed active matter.

In active matters systems, the spontaneous movement of active particles is mainly determined by their anisotropy. The orientation of active particles is not only determined by their movement but also by the interaction between active particles and the interaction between active particles and the surrounding medium [7]. Active particles are generally divided into self-propelled particles and self-rotating particles [8] [9]. In this work, we mainly focus on the collective motion of self-propelled active matter systems. While considerable investigations were launched into understanding the collective behavior of active matters, the mechanism behind the collective motion remains elusive to a large extent. There are various modeling approaches to the collective motion of living cells. These include lattice models [10] [11] [12], phase-field models [12] [13] [14], particle models [15] [16] [17], active network models [18] [19] [20], and continuum models [21] [22] [23].
Lattice models, a class of statistical-mechanical models, include the well-studied Ising model, Potts model, XY model, and Toda lattice. Within this approach, each cell is represented by an ensemble of contiguous lattice points [10] [11] [12] [17]. Lattice models have been widely used to investigate the mechanisms of cell rearrangements based on the detailed description of cell shape, cell-cell, and cell-substrate interactions. However, the difficulty of distinguishing cell-cell and cell-substrate interactions prevents the use of this approach to investigate the collective motion of cells as a result of strong dependence on neighboring sites to calculate the interfacial potential energy [18].

Phase-field model is another approach able to describe cell shape with sub-cellular details, in which each cell is represented by a phase field. Phase-field model will be reviewed detailly in Chapter 2, but what we need to address here is that the Phase-field model is more suitable to describe the cell-cell and cell-substrate adhesion of individual cells due to the force balancing providing physical dynamics [13] [14] [20]. Overmuch parameters of the phase-field approach which are difficult to relate to the system properties limit the use of this approach in the investigation of macroscopic collective migration of cells.

Particle models involve far fewer details, than either lattice or phase field models, of cell shape and its coupling to their polarity [15] [16] [17] [24]. Here, a cell is simply treated as one or two circular beads. The approach has been used to investigate the collective migration of epithelial and mesenchymal cells [17], but it ignores too many nontrivial cellular details such as bending flexibility of cell membranes, cell shape, and area constraint.

Active network models include two subtypes: Vertex and Voronoi models. For example, epithelial tissues, in this approach, are described as networks of polygonal cells [19]. Compared to previous models such as lattice models and phase-field models, the active network models
incorporate even fewer details, and are useful to the case where cells are packed densely, without free space between them, thus restricting these models to investigations of epithelial cell groups. Generally speaking, the active network models are suitable for the study of the role of cell geometry and topological rearrangements on cell motion [12].

Continuum models do not account explicitly for the microscopic details of cells [12] [21] [22] [23]. Instead, these models are based on kinetic Toner-Tu equations for flocking [26] [27] [28], which, on the one hand, cannot distinguish between the polarity and the velocity fields, and on the other hand, cannot incorporate effects such as hydrodynamic interactions. Thus, the flocking-type continuum model cannot be used to investigate collective cell migration.

In this thesis, we present a novel approach, namely a ring-polymer model, to investigate the emergence of collective motion (flocking) and non-equilibrium patterns. In this approach, each cell is treated as a semi-flexible two-dimensional ring and accounts for many single-cell properties, including deformability, bending rigidity, self-propulsion, short-range repulsion, polarity, and area constraint of cells to be altered. Extensive numerical investigations of this model through molecular dynamics (MD) simulations conveniently allowed us explore the positional and orientational correlations of the cells as a function of many important variables mentioned earlier, and the emergence of macroscopic flocking patterns induced by cell motility, anisotropic interaction with the substrate, and areal density. Our model can make a significant contribution to pursue the further understanding of the collective motion in systems of self-propelled active matter with short-range repulsion.

We considered two general boundary conditions, corresponding to periodic boundary conditions and circular confinement. We also investigated the effect of anisotropically circular patterns in the central region of the system on the collectivity of the cells. The monomers of the
cells boundary are moved using a molecular dynamics scheme with a Langevin thermostat, in which the equations of motion are derived from the Hamiltonian of the system and are integrated by using the Velocity-Verlet algorithm. Driving forces, regarded as a net force of cytoskeletal propulsion and the frictional force between the cells and the substrate, are applied to each cell to investigate the effect of cell motility on the physical properties of the system. The quantifications of the system include areal density distribution, the average speed of the system, tangential and radial momentum, clustering properties of the system, order parameter, and the frequency of reversals of the rotational band.

We emphasize that all the MD codes and quantification codes are developed and written by the candidate independently in C++. A detailed literature review of previous work done to investigate the collective motion of cells is found in Chapter 2. The explanations of our model and quantification methods are found in Chapter 3. In Chapters 4, 5, and 6, we present the results and analyse of our simulations. Finally, the conclusions are presented in Chapter 7. In this chapter, we also present a future outlook of this project.
2. LITERATURE REVIEW

What is the first step when a graduate student starts to do his research? The answer is absolutely to do some literature review. In this chapter, we will review some previous models used to investigate collective cell migration with their benefits and drawbacks including the cellular Potts model [10] [11] [12], Phase-Field Model [12] [14], Particle-Field Model [15] [16] [17], and Active network models [18] [19] [20].

2.1 Cellular Potts model

The cellular Potts model (CPM), initially proposed by Francois Graner and James Glazier to simulate cell sorting [24], is a computational approach to model cells and tissues. With similarity to other classical models of statistical mechanics such as paradigmatic Ising model[12], In the CPM, cells are generally described as deformable objects with a certain excluded volume, which can interact with other cells and the substrate. Each lattice site $i = 1, 2, ... , N$ is assigned a state variable $\sigma_i = 1, 2, ... , m$ corresponding to one of $m - 1$ cells, where $m < N$[24] [29]. The state of each lattice site is then updated through a state-exchange Monte Carlo scheme with Metropolis dynamics at a sufficiently low temperature and the Hamiltonian of the CPM is given by[24]

$$\mathcal{H} = \sum_{\langle i,j \rangle} J(\sigma_i, \sigma_j) + \lambda \sum_{\sigma=1}^{m-1} (A_{\sigma} - A_0)^2 - P \sum_{\sigma=1}^{m-1} R_{\sigma} \cdot \bar{p}_{\sigma}$$  \hspace{1cm} (2.1.1)

where the sum of the first term in Eq. (2.1.1) runs over neighboring sites $\langle i,j \rangle$, which accounts for the interfacial tension between neighboring cells as well as between cells and the medium (state $\sigma = m$), which are encoded in the interaction matrix $J$. The simple form of $J$ is given by
\[ J(\sigma_i, \sigma_j) = \alpha \left( 1 - \delta_{\sigma_i, \sigma_j} \right) \]  \hspace{1cm} (2.1.2)

where \( \alpha \) corresponds to the interfacial energy that controls the amplitude of the fluctuations of cell shape. The second term is the area constraints of the system, in which \( A_0 \) is the preferred area for each cell and \( \lambda \) is the controlling parameter to adjust the strength of the constraint force. The third term is devoted to assign the cell motility, in which \( \vec{R}_{\sigma} \) is center of mass, \( \vec{p}_{\sigma} \) corresponds to the cellular polarity [Fig. 1(a)] and \( P \) is the magnitude of the active polar force.

This model was used to study collective rotations [30], tissue spreading [31], and phase transitions from liquid to solid phase and glassy dynamics of cells[28] [Fig. 1(b)]. Fig. 1(a) shows two cells composed of many lattices, and Fig. 1(b) are snapshots and trajectories of the cells at the fluid and solid regimes, where different cells are in different colors with their unique state variables. Once again, CPM is useful to investigate the mechanism of cell rearrangement, however, it is not suitable for collective cell migration due to the artificial cell shape fluctuations and the difficulty of distinguishing cell-cell and cell-substrate interactions.

Figure 1. Lattice Models: The Cellular Potts Model. (a), Lattice sites corresponding to two different cells are shown in different colors. Cell-cell and cell-medium interfaces have an
interfacial energy $\alpha$ (black). Cell migration in the direction of the polarity is favored by a self-propulsion magnitude $P$. (b), Snapshots of the system and cell trajectories at the fluid ($\alpha = 1$) and solid ($\alpha = 4$) regimes. Neighboring cells are colored differently, with arbitrary colors. Cell boundaries are rougher and longer for smaller interfacial energy.

The figure is adapted from ref. [28].

2.2 Phase-field models

Phase field models were first introduced by Fix [32] and Langer [33] to study free boundary problems and interface dynamics. In the field of biophysics, phase-field models describe each cell $i = 1, 2, \ldots, N$ by a phase field $\phi_i(\vec{r}, t)$, which is 1 inside the cell and 0 outside [24] [Fig. 2(a)]. Regarding the cell-cell interactions, there are different ways to build the free energy functional of the phase field [34] [35] [36]. A possible form is given by [24]

$$F = F_{CH} + F_{area} + F_{cell-cell}, \tag{2.2.1}$$

where for the first term we have

$$F_{CH} = \sum_{i=1}^{N} \gamma \int [4\phi_i^2 (1 - \phi_i)^2 + \epsilon^2 |\nabla \phi_i|^2] d^2 \vec{r}, \tag{2.2.2}$$

Which corresponds to a double-well potential with minima at the cell interior ($\phi_i = 1$) and exterior ($\phi_i = 0$). The second term is the area constraint of the cells that penalizes departures of cell area from its preferred area $\pi R^2$ and given by

$$F_{area} = \sum_{i=1}^{N} \frac{u}{\pi R^2} \int [\phi_i^2 d^2 \vec{r}]^2, \tag{2.2.3}$$

The third term accounts for cell-cell interactions, which corresponds to a two-body repulsive force between cells and is given by
\[ F_{\text{cell-cell}} = \sum_{i=1}^{N} \sum_{j \neq i}^{K} \frac{1}{\varepsilon} \int \left[ \phi_i^2 \phi_j^2 - \tau \varepsilon^4 |\nabla \phi_i|^2 |\nabla \phi_j|^2 \right] d^2 \vec{r}, \]  

(2.2.4)

\( F_{\text{cell-cell}} \) is used to prevent cells from overlapping [Fig. 2(b)].

The phase field model can explicitly account for cell-cell and cell-substrate interactions as well as for active stresses [14] [34], which is better connected to tissue mechanics and kinetics of individual cells. We can also couple the cell polarity to cell shape asymmetry to study the cell-cell alignment and collective motion resulting from the collisions between deformable cells [37] [38][Fig. 2(c) and 2(d)]. However, we can see clearly that the phase-field model neglects the bending rigidity of the interface between cells.

Figure 2. Phase-field models a, Phase field of a cell. Adapted from [33]. b, The overlap between phase field, cell-cell interfaces are identified in white. The inset shows cell contours, \( \phi_i = 0.5 \).
Adapted from [39]. Copyright (2019) by the American Physical Society. \textbf{c-d}, Collisions between deformable cells lead to velocity alignment(c) and collective motion(d). Adapted from [37].

2.3 Particle models

All matter is made up of particles, which behave in various ways whether they are solid, liquid, or gas. In particle models, each cell is treated as one or two particles and the positional cell-cell interactions are usually implemented by a central interparticle potential. For example, a simple form of the potential energy can be given by

\[ V_{i,j} = \kappa (1 - \frac{r_{i,j}}{r_{eq}})^2, \]  

where \( r_{i,j} \) is the distance between particle \( i \) and \( j \), \( r_{eq} \) is the preferred distance between particles.

Cell motility accounts for an active polar force \( T_a \vec{p}_i \). Other forces such as cell-substrate viscous friction and \(-\xi \vec{v}_i\) and cell-cell friction with coefficient \( \xi_c \) can also be added to the force balance [Fig. 3(a)] in this model. Thus, the equation of motion of cell \( i = 1, 2, \ldots, N \) is given by[20]

\[ \xi \vec{v}_i = T_a \vec{p}_i + \sum_{(i,j)} \left[ -\vec{v}_i \left( |\vec{r}_i - \vec{r}_j| \right) + \xi_c |\vec{v}_i - \vec{v}_j| \right], \]  

Contact inhibition Locomotion (CIL) is widely used to model polarity interactions in the Particle model, which is the process whereby cell collide, cease migrating in the direction of the collision, and repolarized their migration machinery away from the collision [39] [40]. In cell clusters, CIL induces a coupling between the polarity and density fields, which gives rise to a spontaneous symmetry breaking towards collective motion [41] [Fig. 3(b)].

The particle models are able to incorporate the cell polarity, cell-cell and cell-substrate interactions, and active polar force, which is widely used to study the collective migration of epithelial and mesenchymal cells coordinated by weak and transient cell-cell contact[41].
However, too many details about cell shape and its coupling to polarity are almost entirely overlooked in these models.

Figure 3. Particle models. a, Schematic representation of forces in particles models. Cells may experience various amounts of cell-cell repulsion(blue), adhesion(orange), cell-substrate active traction(red), or passive friction(green). Adapted from [39]. b, The CIL repolarization occurred in small cell cluster finally leads to the collective motion. The color code indicates the cell polarity angle $\theta_i$. Adapted from [40].

### 2.4 Active network models: Vertex and Voronoi models

Vertex models treat epithelial tissues as networks of polygonal cells, thus incorporating more details about sub-cellular features of cell shape compared to Particle models. In Vertex models, the degrees of freedom are the vertices of the polygons. In Voronoi models, the network can be described by the cell centers, which reduces the number of degrees of freedom[24]. If we know the positions of the cell center, the cell-cell boundaries are delineated by the Voronoi tessellation [Fig. 4(a)]. The difference in the number of degrees of freedom can distinguish vertex and Voronoi models [20].

In both descriptions, an energy function for tissue is used to encode cellular properties and interactions in terms of the areas $A_i$ and perimeters $P_i$ of cells $i = 1, 2, \ldots, N$ and given by[24]
\[ F = \sum_{i=1}^{N} \left[ \frac{\kappa}{2} (A_i - A_0)^2 + \Lambda P_i + \frac{\Gamma}{2} P_i^2 \right], \]  

(2.4.1)

where \( \kappa \) is the modulus to penalize the departure of the area from preferred value \( A_0 \). \( \Lambda \) is the line tension of the cell-cell interfaces that connect the vertices. For foams, the energy function is given by[24]

\[ F = \sum_{i=1}^{N} \left[ \frac{\kappa}{2} (A_i - A_0)^2 + \frac{\Gamma}{2} (P_i - P_0)^2 \right]. \]  

(2.4.2)

Where preferred perimeter \( P_0 = -\Lambda/\Gamma \) and \( \Lambda \) is always a positive number in this case.

As to cell motility, we can apply active polar forces either on the vertices or on the cell centers, leading to Active Vertex Models (AVM) [42] [43] [44] and Self-Propelled Voronoi models (SPV) [45] [46], respectively. The form of this active polar force [Fig. 4(b)] is given by[20]

\[ \vec{F}_{i}^{\text{int}} = -\nabla r_{i} F, \]  

(2.4.3)

and in particular, polarity-velocity alignment is one of the most popular orientational interaction in SPV models [47] [48][Fig. 4(c)]. The SPV models can be applied to investigate many interesting phenomena. For example, Bi at al. studied the effect of cell motility on solid-fluid transition[45], showing that both self-propulsion speed and persistence favor the fluid phase [Fig. 4(d)]. What’s more, the onset of collective motion was also studied, showing that cell-autonomous polarity-velocity alignment [Fig. 4(c)] leads to the emergence of cell-cell alignment, which finally gives rise to coherent rotations[46] and flocking[47][48] [49]. All in all, SPV models make predictions in four phases: solid, liquid, solid flock, and liquid flock [24] [Fig. 4(e)].
Instead of typical collective cell migration, most active network models are used to
describe confluent tissues, where cells are closely packed together without free space between
them, thus restricting the models to collective migration of epithelial cell groups. Generally
speaking, network models are better connected to study the cell geometry and topological
rearrangements on cell motion.

Figure 4. Active Network Models. a, Cell centers with positions \{r_i\} are connected by the
Delaunay triangulation (black). Voronoi tessellation (red) defines cell boundaries and vertices at
position \{h_i\}. Reprinted from [45]. b, Cells are confined by an area \(A_i\) and a perimeter \(P_i\). Net
force of the cell motion can be calculated by the sum of a self-propulsion force \(T_a \vec{P}_i\) (orange) and
an interaction force \(\vec{F}^{\text{int}}_i = -\nabla_{\vec{r}_i} F\) (dashed black). Adapted from [48]. c, Polarity-velocity
alignment with a time scale \(\tau\). Adapted from [56]. d, Phase diagram of the fluid-solid transition
in the SPV model, in which shape index \(p_0 = P_0/\sqrt{A_0}\) and self-propulsion speed \(v_0 = T_a/\xi\)
with persistence \(D_f^{-1}\). Adapted from [45]. e, Schematic phase diagram of the SPV model with
polarity-velocity alignment. Adapted from [50].
3. RING-POLYMER MODEL AND QUANTIFICATIONS

In this chapter, we will first go through the details of the ring polymer model to see how we construct the cells step by step in section 3.1 including potential energies acting on cells, motility force, elongation of cells, circular pattern of substrate, and Molecular Dynamics method. Later in section 3.2, we proceed by presenting our quantifications of the properties of the system such as areal density distribution, average speed, normalized tangential and radial momentum, clustering, and frequency of reversals in flocking layers as a variety of constraints including cell motility, stiffness of substrate, density, and radius of the system.

3.1 Theoretical model

In this thesis, each cell is represented by its projected area on the substrate. To account for its flexibility, bending rigidity of its membrane, and self-avoidance between cells, we generalize a ring polymer model introduced earlier by Dr. Laradji’s group [51]. This model was used to investigate the conformational behavior of self-avoiding ring polymers and their self-assembly as a function of their density and bending rigidity. For the purpose of this thesis, each cell projected boundary in the xy-plane is described by a ring polymer consisting of N monomers in an implicit solvent. Due to their strong adsorption to the substrate, the cells are then modeled as two-dimensional ring polymers. The net potential energy acting on the system is given by

$$U_{\text{net}}(\{r_i\}) = \sum_i U_{\text{bond}}(r_{i,i+1}) + \sum_i U_{\text{bend}}(r_{i-1,i}, r_{i,i+1}) + \sum_{i,j} U_{\text{rep}}(r_{i,j})$$

$$+ \sum_{i,j} U_{\text{sub}}(r_{i,j}) + \sum_i U_{\text{area}}(i), \quad (1)$$

where $U_{\text{bond}}$ is a two-body bonding potential energy that maintains the connectivity between adjacent monomers and is given by
\[ U_{\text{bond}}(r_{i,i+1}) = \frac{k_{\text{bond}}}{2} (r_{i,i+1} - r_b)^2, \]  
(2)

where \( k_{\text{bond}} \) is the spring constant, \( r_{i,i+1} = |r_{i+1} - r_i| \) (\( r_i \) is the coordinates of bead \( i \)) and \( r_b \) is the preferred bond length. In our simulation, a single ring polymer is composed of \( N \) monomers, which means that for monomer \( i = N, i + 1 \equiv 1 \) in Eq. (2). \( U_{\text{bend}} \) is a three-body bending energy that maintains the semi-flexibility of ring polymers and given by

\[ U_{\text{bend}}(r_{i-1}, r_i, r_{i+1}) = -k_{\text{bend}}(\cos \theta_b + \cos \theta_i), \]
(3)

where \( k_{\text{bend}} \) is the bending stiffness of the polymers and \( \theta_b \) is the preferred bending angle of monomers that in most of cases equals roughly \( \pi \) except for two poles of each single ring polymer. \( \theta_i \) is the splay angle at bead \( i \) and \( \cos \theta_i \) is defined as

\[ \cos \theta_i = \frac{r_{i-1,i} \cdot r_{i+1,i}}{|r_{i-1,i}| |r_{i+1,i}|}, \]
(4)

In Eq. (1), \( U_{\text{rep}} \) is the two-body repulsive potential energy acting between any two non-connected monomers. This potential ensures that the cells remain disjoint. We use a simple quadratic repulsive potential energy given by

\[ U_{\text{rep}}(r_{i,j}) = \begin{cases} 
\frac{k_{\text{rep}}}{2} \left(1 - \frac{r_{i,j}}{r_{\text{eq}}} \right)^2, & \text{if } r_{i,j} \leq r_{\text{eq}}, \\
0, & \text{if } r_{i,j} \leq r_{\text{eq}}, 
\end{cases} \]
(5)

where \( k_{\text{rep}} \) is the strength of repulsive interaction and is positive. \( r_{\text{eq}} \) is the cutoff distance for beads \( i \) and \( j \). To account for the elongated geometry of cells, in general, we assign two poles to each ring polymer located at two indices 1 and \( N/2 + 1 \). The elongated shape of the cell is induced by assigning a preferred splay angle \( \theta_p \) at the poles that is smaller than \( \pi \). This is achieved by modifying the three-body interaction at 1 and \( N/2 + 1 \) to

\[ U_{\text{bend}}(r_{i-1}, r_i, r_{i+1}) = -\frac{k_{\text{bend}}}{2} (\cos \theta_i + \cos \theta_b)^2, \]
(6)
where \( \theta_b \) here is the preferred splay angle for poles 1 and \( N/2 + 1 \) and is chosen to \( \pi/2 \) in our simulation. \( U_{area} \) is the area constraint on each cell to maintain the cell shape, the potential energy is given by

\[
U_{area}^i = \frac{k_{area}}{2} (A_i - A_0)^2,
\]

(7)

where \( A_i \) is the area of cell \( i \) and \( A_0 \) is the preffered area for each cell. In this work, we focus on the case of a circularly patterned substrate with a tendency to align the cells along the pattern. This is achieved by the following additional interaction on the two poles \((i, j)\) of a cell

\[
U_{sub}(r_{i,j}) = \frac{k_r}{2} \sin^2 \phi_{i,j},
\]

(8)

where \( k_r \) is the stiffness of the substrate, \( i \) and \( j \) are the two poles of the ring polymer and \( \phi_{i,j} \) is the angle between the vector \( r_{i,j} \) and the tangent line to the circular pattern at the midpoint of the two poles.

In our model, we assume that a cell, \( k \), is acted upon by a non-conservative force, of magnitude \( \eta \), in the direction of its polarization and an orientation determined by the average velocity, i.e.,

\[
F_{k}^{mo}(t) = \eta \frac{P_k(t)}{P_k(t)} f[\mathbf{v}_k(t), \mathbf{P}_k(t)],
\]

(9)

Where \( f(A, B) = +1 \) or \(-1\) if \( A \cdot B > 0 \) or \( < 0 \), respectively. In the equation above \( \mathbf{P}_k(t) \) is the polarization of the cell \( k \) at time \( t \) and \( \mathbf{v}_k^{ave}(t) \) is the average velocity of the cell over the time interval \( [t - \tau, \tau] \), i.e.

\[
\mathbf{v}_k^{ave}(t) = \frac{1}{\tau_{mo}} \int_{t-\tau}^{t} \mathbf{v}_k(t')dt'.
\]

(10)
Therefore, while the direction of the motility force is along the cell's polarization, its orientation is determined by the average velocity of the cell during the time interval \( [t - \tau, \tau] \).

The beads of each cell move following a molecular dynamics scheme with a Langevin thermostat, i.e.,

\[
\dot{r}_i(t) = v_i(t), \quad \text{and}
\]

\[
m\dot{v}_i(t) = -\nabla_i U_{net} + F_i^{mo} - \Gamma v_i(t) + \sigma \xi_i(t),
\]

where \( m \) is the mass and \( v_i \) is the instantaneous velocity of bead \( i \). \( \Gamma \) is the friction coefficient and \( \sigma \xi_i(t) \) is a random force used to mediate the heat transfer from the thermal reservoir to the system. \( \sigma \xi_i(t) \) must obey

\[
\langle \xi_i(t) \rangle = 0,
\]

\[
\langle \xi_i^\mu(t) \xi_j^\nu(t') \rangle = \delta_{\mu\nu} \delta_{ij} \delta(t - t'),
\]

where \( \mu \) and \( \nu \) are equal to \( x \) or \( y \). Since the equipartition theorem must be satisfied, the dissipative and random force in Eq. (10) are inter-related, leading to the following relation between the coefficients \( \Gamma \) and \( \sigma \)

\[
\Gamma = \frac{\sigma^2}{2k_B T'},
\]

where \( k_B \) is Boltzmann’s constant and \( T \) is the temperature of the system.

Investigations on the collective motion of ring polymers are conducted in both periodic square box and circular confined domain, respectively. In both cases, there is circularly patterned substrate directing the motion of cells in the central region of the system. We must ensure that the parameters \( k_{bond} \) and \( k_{rep} \) in Eq. (2) and (5) are strong enough to preserve the topological disjointness condition of the cells. In order to confine the cells in a circular domain, we apply the
repulsive interaction between monomers and circular boundary, the harmonic potential energy of repulsive boundary condition is given by

\[
\mathcal{U}_{\text{wall}}(r_i) = \frac{k_{\text{wall}}}{4}(r_i - r_{eq})^4,
\]

where \( k_{\text{wall}} \) is the cell-wall interaction constant and \( r_i \) is the shortest distance between monomer \( i \) and the circular boundary. \( r_{eq} \) is the cutoff distance of the repulsive interaction.

For all simulations performed with periodic boundary condition and in circular confinement, the preferred bond length \( r_b \) sets the unit of length and the cutoff distance of repulsive force \( r_{eq} = r_b \). Total number of monomers making up one cell is \( N = 40 \). The length of periodic square box is \( 400r_b \) and the radius of circular confinement is \( 200r_b \). All energies are measured in units of \( k_B T \). The numerical values of the parameters used in our simulations are \( k_{\text{bond}} = 100k_B T/r_b^2 \), \( k_{\text{bend}} = 100k_B T \), \( k_{\text{rep}} = 50k_B T \), \( k_{\text{area}} = 100k_B T/r_b^4 \), and \( k_{\text{wall}} = 100k_B T/r_b^4 \). \( k_m \) is varied in the range between 0 and \( 1.2k_B T/r_b \) and \( k_{\text{sub}} \) is varied between 0 to \( 180k_B T \). \( \Gamma = 0.5m/\tau \) and the timescale \( \tau = \sqrt{m/k_B T} \). Single timestep of the integration of equations of motion \( \Delta t = 0.01\tau \). In particular, density is defined as \( \rho = N/A \), where \( N \) is total number of ring polymers and \( A \) is the area of the system. \( \rho \) is varied from 0.002185\( r_b^{-2} \) to 0.003125\( r_b^{-2} \) in the simulations with periodic boundary condition, and 0.002785\( r_b^{-2} \) to 0.003979\( r_b^{-2} \) in the simulations in the circular confinement. All simulations are run with \( 4 \times 10^6 \) steps, which is well beyond the timesteps for the system to reach the steady state.

### 3.2 Quantification methods

The system is divided into multiple layers as a function of distance \( d \) to the center point \( O(L/2, L/2) \), and the layer index of ring polymer \( i \), defined as \( \theta_i = r_i / d \), where \( r_i \) is the
distance between center of mass of the ring polymer and center point of the system. The physical properties of cells of different layers are quantified in terms of layer index, including their areal density distribution, average speed, and average tangential velocity. Furthermore, we quantify the number of reversals for the rotating layer in the patterned region as a function of different variables of the system, such as driving force, strength of substrate, and system density. As to the clusters of the system, if the distance between any monomers of two cells is less than $2r_b$, these two cells are considered in one cluster. The clustering properties of the system are also quantified, including its average size of clusters, maximum size of clusters, and average total number of clusters. For a specific layer $i$, the areal density distribution is defined as $\rho_i = N_i/A_i$, where $N_i$ is the total number of ring polymers and $A_i$ is area of the layer. Average speed of the layer is given by

$$\langle |V_i| \rangle = \frac{1}{N_i} \sum_{j=1}^{N_i} |v_j|,$$  \hspace{1cm} (16)

where $v_j$ is the average velocity for ring polymer $j$ in the layer. The average tangential velocity of the layer is given by

$$\langle V_{i}^{tan} \rangle = \frac{1}{N_i} \sum_{j=1}^{N_i} v_j^{tan},$$ \hspace{1cm} (17)

where $v_j^{tan}$ is the tangential velocity for cell $j$ in the layer. Similarly, the average radial velocity of the layer is given by

$$\langle V_{i}^{rad} \rangle = \frac{1}{N_i} \sum_{j=1}^{N_i} v_j^{rad},$$ \hspace{1cm} (18)

where $v_j^{rad}$ is the radial velocity for ring polymer $j$ in the layer. The number of reversals is recorded in terms of change of direction of average tangential velocity in the layer. Average size of clusters in a system can be calculated using total number of cells in clusters divided by the number of clusters. Maximum size of clusters in the system means the total number of ring
polymers in the cluster with biggest size. Number of clusters show how many clusters the system has. Vortical order parameters are calculated to directly quantify the collectivity of cells moving in a circular confinement and give by

\[ \alpha_c = \left| \frac{\sum_i v_i^{tan}}{N_c} \right|, \] (19)

where \( v_i^{tan} = +1 \) or -1 if the direction of tangential velocity of cell \( i \) is clockwise or counterclockwise, respectively. \( N_c \) is the total number of cells in circular confinement.

Alignment order parameter is used to quantify collectivity of the system with periodic boundary condition and given by

\[ \alpha_p = \left| \frac{\sum_i c_i \cos_i}{N_p} \right|, \] (20)

where \( \cos_i \) is the cosine between two vectors: the velocity of cell \( v_i \) and the average velocity of corresponding cluster \( v_j \). In particular, we use normalized tangential momentum to quantify the collectivity of the system with patterned substrate. The equation is given by

\[ a_i = \frac{1}{N_i} \left| \frac{\sum_i \rho_i v_i^{tan}}{v_i^{tan Base}} \right|, \] (21)

Where \( v_i^{tan} \) is the average tangential velocity of layer \( i \) and \( v_i^{tan Base} \) is the corresponding tangential velocity of the system without patterned region for layer \( i \). \( \rho_i \) is the areal density distribution of the same layer as tangential velocity. \( N_i \) is the total number of cells in the layer \( i \).

3.3 Schematics and table of parameters

In this section, we show some schematics of our model including the initial composition of single cell (Fig. 5), initial configurations of systems before polarization of cells (Fig. 6),
substrate force induced by circularly patterned substrate (Fig. 7), clustering of systems (Fig. 8), the position of patterned region (Fig. 8), and central vortex induced by substrate force (Fig. 9).

Figure 5. Initial composition of single cell before polarization. Total number of monomers is 40. The length of spring that connects monomers $1r_b$. The initial radius $r \approx 6.37r_b$. Angle between two monomers $\theta = 9^\circ$. The bending angle of each monomer $\theta_b \approx 180^\circ$. 
Figure 6. Initial configurations of cells without polarization moving in circular confinement (a) and periodic box (b), respectively. For circular confinement, the radius $r = 200r_b$. The length of periodic box $L = 400r_b$.

Figure 7. Substrate force induced by circularly patterned substrate. The polarization of the cell is forced to rotate towards the tangent line of the curve.
Figure 8. Clustering of cells in circular confinement (a) and periodic box (b), respectively. If the distance of any monomers of two cells is less than $2r_b$, these two cells are considered in the same cluster. Different color represents different clusters. The circularly patterned substrate is located on the central region of systems. For circular confinement, the radius $r = 200r_b$. The length of periodic box $L = 400r_b$. In both two systems, the radius of patterned region $r_c = 100r_b$.

Figure 9. Snapshots of central vortex induced by the substrate force in circular confinement (a) and periodic box (b). In the middle region, cells are self-organized into a rotating vortex at a moderate driving force of cells and strength of substrate.
At last, let us look at standard values of simulation parameters in our system. While varying the driving force coefficient, \( k_{mo} \), substrate force coefficient, \( k_r \), and system density, \( \rho \), we keep other simulation parameters constant.

Table of parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>( k_{rep} )</td>
<td>50</td>
<td>Coefficient of the repulsive force between monomers</td>
</tr>
<tr>
<td>( k_{bend} )</td>
<td>100</td>
<td>Coefficient of bending force between monomers</td>
</tr>
<tr>
<td>( k_{sp} )</td>
<td>100</td>
<td>Coefficient of elastic force between monomers</td>
</tr>
<tr>
<td>( \kappa_a )</td>
<td>1</td>
<td>Equi-preferred length of the spring connects monomers</td>
</tr>
<tr>
<td>( \kappa_c )</td>
<td>1</td>
<td>Coefficient of area constraint of cells</td>
</tr>
<tr>
<td>( m )</td>
<td>1</td>
<td>Mass of a single monomer</td>
</tr>
<tr>
<td>( \kappa_{cp} )</td>
<td>100</td>
<td>Equi-preferred area for cells</td>
</tr>
<tr>
<td>( N )</td>
<td>500</td>
<td>Total number of ring polymers in the standard system</td>
</tr>
<tr>
<td>( k_{area} )</td>
<td>100</td>
<td>Coefficient of area constraint on cells</td>
</tr>
<tr>
<td>( \kappa_{wall} )</td>
<td>100</td>
<td>Coefficient of cell-wall interaction</td>
</tr>
<tr>
<td>( \kappa_r )</td>
<td>100</td>
<td>Coefficient of the circular pattern of substrate (Varied from 0 to 180)</td>
</tr>
<tr>
<td>( \kappa_{mo} )</td>
<td>0.5</td>
<td>Coefficient of motility force of cells (Varied from 0 to 1.2)</td>
</tr>
<tr>
<td>( R )</td>
<td>200</td>
<td>Radius of circular confinement</td>
</tr>
<tr>
<td>( L )</td>
<td>400</td>
<td>Length of periodic box</td>
</tr>
<tr>
<td>( r_p )</td>
<td>100</td>
<td>Radius of patterned region</td>
</tr>
<tr>
<td>( \rho )</td>
<td>( N / \pi R^2 )</td>
<td>Density of the system</td>
</tr>
<tr>
<td>( \Delta t )</td>
<td>0.01</td>
<td>Each timestep</td>
</tr>
<tr>
<td>( T_{max} )</td>
<td>4000000</td>
<td>Total steps for each simulation</td>
</tr>
</tbody>
</table>
4. RESULTS ON UNIFORM SUBSTRATE

In this chapter, we investigate the spatiotemporal organization and physical properties of the cells when the system undergoes varied driving force \( (k_{mo}) \) and density \( (\rho) \) with periodic boundary condition (PBC) and in circular confinement, respectively. The substrate is isotropic with uniformity in all directions. The quantifications include average areal density distribution, average speed, alignment order parameter, vortical order parameter, and some clustering properties such as the total number of clusters, the maximum size of clusters, and the average size of clusters. All simulations are run up to 4 million steps and all results are averaged over 2 million steps.

4.1 Simulations in the system with periodic boundary condition

In this section, we show results of simulations on systems with periodic boundary condition at increasing driving force \( (k_{mo}) \) and density \( (\rho) \), where the length of the periodic box is \( 400r_b \). Initially, 500 cells are placed randomly without overlap.

4.1.1 Effect of increasing driving force

Configuration snapshots and corresponding velocity fields, at different driving force coefficients, are shown in Figure 10. The figure shows that increasing driving force induces a bigger size of traveling band with well-aligned velocities of the cells and the less total number of clusters. This is due to the fact that high values of \( k_{mo} \) lead to more collisions, which give rise to the velocity alignments of cells, thus increasing the size of clusters. \( 0.6k_BT/r_b \) seems to be a critical value to induce the emergence of the macroscopic collective dynamics of the system. The increasing average size of clusters also decreases the total number of clusters in the system since the number of cells is constant. In addition, we can see that cells in clusters are more aligned in high motility cases.
Figure 10. Configuration snapshots and velocity fields of the cells for $k_{m0} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1, \text{ and } 1.2k_B T/r_b$ at a given density $\rho = 0.003125r_b^{-2}$. The length of the periodic box equals $400r_b$ as a measure of length.
We now turn to quantify the spatial organization and physical properties of the cells in the system. Fig. 11(a) shows the average areal density distribution as a function of radius for different values of driving force coefficient. This figure shows that the average areal density does not change with the increasing driving force except for some oscillations caused by computational simulations. The reason is that cells are moving in a periodic box, where they can pass through one side and come back from another side of the box. Averagely speaking, the probability density for cells to be present should be uniform everywhere in the system. In addition, we expect smaller fluctuations if the simulation is run over longer time.

Fig. 11(b) depicts the relationship between the average speed of the system and the driving force of cells. The figure shows that the average speed of the system increases monotonically with the augment of the driving force. This is due to the fact that a greater driving force will induce bigger instantaneous acceleration and higher kinetic energy of the system. Again, as expected, the average speed of the system should also be uniform everywhere in the system as a result of periodic boundary condition. Also, the higher average speed of the system gives rise to substantial collisions among cells, thus increasing the collectivity of the system.

We also investigate the clustering properties of the system with periodic boundary condition. The average total number of clusters, depicted in Fig. 12(a), decreases with the augment of the cell motility force at \( k_{m_o} > 0.2k_B T/\tau_b \). This is in line with the observation in Fig. 1 that higher motility force will lead to giant clusters in the system as a result of substantial collisions. Furthermore, the system seems to reach a saturated state as we further increase the driving force over \( 0.8k_B T/\tau_b \). Fig. 12(b) shows the relationship between the average maximum size of the cluster and cell motility. There are more bigger size of clusters forming during the increase of the driving force, which agrees with our results from Fig. 12(a) because of more frequent local
collisions. The size of the biggest cluster can represent the collectivity of the system as well. As to average size of clusters of the system, shown in Fig. 12(b), it also can be used to quantify the collectivity of the system that increasing the average size of clusters means that more cells are moving together in the system.

Figure 11. Quantifications of areal density distribution and average speed of the system. a. Areal density of the system as a function of radius for \( \kappa_{m_0} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1, \) and \( 1.2k_BT/r_b \) at a given density \( \rho = 0.003125r_b^{-2} \). b. Average speed of the system as a function of radius for \( \kappa_{m_0} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1, \) and \( 1.2k_BT/r_b \) at a given density \( \rho = 0.003125r_b^{-2} \).
Figure 12. Quantifications of clustering properties of the system. a, Average total number of clusters as a function of driving force coefficient at a given density $\rho = 0.003125r_b^{-2}$. b, Average maximum size of clusters as a function of driving force coefficient at a given density $\rho = 0.003125r_b^{-2}$. c. Average size of clusters as a function of driving force coefficient at a given density $\rho = 0.003125r_b^{-2}$.

To confirm the previous results, we use the nematic order parameter to directly quantify the collective dynamics of the system as shown in Fig. 13. When the driving force of cells is under $0.4k_B T / r_b$, the order parameter keeps more or less constant, which means that the system is in a relatively disordered state. The sharpest slope is observed at $0.6k_B T / r_b$. This is expected since we see that macroscopic collective behavior of the system emerges at $k_{mo} = 0.6k_B T / r_b$ in Fig. 10. Furthermore, a motility-saturated state of the system is also observed here around $k_{mo} = 1k_B T / r_b$. We also compare the order parameter between systems of 250 cells and 500 cells. It is clear that if the system is at a disordered state, the addition of cell density just increases the chaos of the system while increasing density induces more collectivity of the system at high driving force coefficient.
In conclusion, increasing driving force leads to a higher average speed of cells. The cells moving with higher speed will collide with each other more frequently, which gives rise to more velocity alignments, finally inducing the giant clusters and coherent locomotion of the system.

Figure 13. Nematic order parameter of the systems of 250 cells and 500 cells. $\rho = 0.003125 r_b^{-2}$.

4.1.2 Effect of increasing density

This section is focused on the effect of density ($\rho$) on the collective motion of cells in a periodic box. Here, we change the density by manipulating the total number of cells at a given driving force coefficient of cells $k_{mo} = 1k_B T/r_b$. Spatial configuration snapshots and the corresponding velocity fields at different densities are shown in Figure 14. It is easy to see that on average, there are more cells moving in one cluster with well-aligned velocities as we increase the density of the system. The collisions between deformable cells and the resultant velocity alignments play the most important role in the collective dynamics of the system in this
case. Obviously, more cells will lead to more collisions, thereby inducing more collectivity of the system.

<table>
<thead>
<tr>
<th>$\rho = 0.002188$</th>
<th>0.002500</th>
<th>0.002813</th>
<th>0.003125</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="https://example.com/image1.png" alt="Image" /></td>
<td><img src="https://example.com/image2.png" alt="Image" /></td>
<td><img src="https://example.com/image3.png" alt="Image" /></td>
<td><img src="https://example.com/image4.png" alt="Image" /></td>
</tr>
<tr>
<td><img src="https://example.com/image5.png" alt="Image" /></td>
<td><img src="https://example.com/image6.png" alt="Image" /></td>
<td><img src="https://example.com/image7.png" alt="Image" /></td>
<td><img src="https://example.com/image8.png" alt="Image" /></td>
</tr>
</tbody>
</table>

Figure 14. Configuration snapshots and velocity fields of the cells for $\rho =$ 0.002188, 0.002500, 0.002813, and 0.003125$r_b^{-2}$ at a given driving force $\kappa_{\mu\nu} = 1k_BT/r_b$. The length of the periodic box equals $400r_b$ as a measure of length.

Fig. 15(a) shows the effect of density on the areal density distribution of the system. Surely as expected, the areal density of each layer just increases with the augment of the total number of cells, and the distribution over all the system is uniform due to the periodic boundary condition. Regarding the average speed of the system, depicted in Fig. 15(b), increasing density actually suppresses the motion of the cells since more cells lead to more local collisions, thereby causing more kinetic energy loss of the system. The more cells moving in the uniform system,
the lower the average speed the cells have. However, a lower average speed of the cells does not mean a decrease of the collectivity of the system.

\[ \rho_{c} = \frac{1}{v_{a}} \]

Figure 15. Quantifications of areal density distribution and average speed of the system with increasing density. a, Areal density of the system as a function of radius for \( \rho = 0.001875, 0.002188, 0.0025, 0.002813, 0.003125, \) and \( 0.003438 r_{b}^{-2} \) at a given driving force \( \kappa_{mo} = 1 k_{B}T/r_{b} \). b, Average speed of the system as a function of radius for \( \rho = 0.001875, 0.002188, 0.0025, 0.002813 0.003125, \) and \( 0.003438 r_{b}^{-2} \) at a given driving force \( \kappa_{mo} = 1 k_{B}T/r_{b} \).

We also quantify the clustering properties of the system here. In contrast to Fig. 12(a), the total number of clusters, shown in Fig. 16(a) increases with the increasing density up to \( \rho = 0.0025 r_{b}^{-2} \), then decreases as we further increase the density. To understand this, it is important to see that collectivity of the system has two contributing factors: the size of the cluster and velocity alignments of cells, respectively. Below the critical value \( \rho = 0.0025 r_{b}^{-2} \), the effect of cell collisions is not strong enough to induce the macroscopic collective motion of cells so if there are more cells in the system, there would be more small clusters just consisting of two or three cells. When \( \rho > 0.002813 r_{b}^{-2} \), increasing density induces more collectivity of the system forming giant clusters and that is why we find that even though there are more cells, the number of cells still
keeps going down. As to the maximum size of the cluster depicted in Fig. 16(b), increasing density reasonably raises the total number of cells in the biggest cluster. In addition, more cells and collectivity of the system surely increase the average size of the cluster as shown in Fig. 16(c).

Figure 16. Quantifications of clustering properties of the system. **a**, Average total number of clusters as a function of density at a given motility force $k_{mo} = 1k_B T/r_b$. **b**, Average maximum size of clusters as a function of driving force at a given motility force $k_{mo} = 1k_B T$. **c**, The average size of clusters as a function of driving force at a given motility force $k_{mo} = 1k_B T/r_b$.

We again calculate the nematic order parameter for the systems with increasing density here. Increasing order parameters, depicted in Fig. 17, confirm our previous results that increasing density surely induces more collectivity of the system.
In summary of this section, increasing density typically leads to more collisions and more corresponding velocity alignments as well, thereby promoting the collective motion of cells.

![Graph](image)

Figure 17. Nematic order parameter of the system at a given driving force coefficient $k_{mo} = 1k_{B}T/r_{b}$.

4.2 Simulations in circular confinement

In this section, we initially place 500 cells in a circularly confined domain with a strong repulsive force near the boundary, which prevents cells from passing through the area. Again, we investigate the effect of the driving force coefficient $k_{mo}$ and density $\rho$ on the collective motion of cells. In addition, the effect of confinement is also investigated here in contrast to the system with periodic boundary condition. The radius of the circular confinement is $200r_{b}$.

4.2.1 The effect of increasing driving force

In this subsection, we present the results of our simulations on a system composed of 500 cells confined in a circular domain. The configuration snapshots and corresponding velocity
fields, at different driving force coefficient, are shown in Fig. 18 at a given density $\rho = 0.003981r_b^{-2}$. At $k_{mo} = 0k_B T/r_b$, the system is in a completely disordered state where the only factor affecting the motion of cells is thermal fluctuations. As we further increase the driving force up to $0.5k_B T/r_b$, we see some small clusters forming with a few cells moving collectively in the system. $0.6k_B T/r_b$ is a critical value for the system to exhibit the macroscopic collectivity that there is a huge cluster forming at the edge of the circular boundary where a lot of cells rotates together in either clockwise or counterclockwise direction. At $k_{mo} > 0.6k_B T/r_b$, increasing cell motility typically increases the size of the rotating vortex and fewer moving cells are observed in the middle region of the circular confinement. From Fig. 18, we can absolutely get the conclusion that increasing the driving force of cells induces more collectivity of the system and there is a critical value for the system to exhibit global collective behavior. However, in contrast to the results in section 4.1.1, we observe a rotational vortex here instead of giant clusters with many cells moving with well-aligned velocity fields as a result of the repulsive circular boundary. The effect of system boundary also plays a crucial role in the collective dynamics of cells. At high motility force, increasing motility force typically increases the width of the rotational vortex, which means on average there are also fewer cells moving freely in the central region.
Figure 18. Configuration snapshots and velocity fields of the cells for $\kappa_{m_0} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1$, and $1.2k_B T/\tau_b$ at a given density $\rho = 0.003981r_b^{-2}$. The radius of the circular confinement $r = 200r_b$ as a measure of length.
Again, we quantify the areal density distribution [Fig. 19(a)] and average speed of cells [Fig. 19(b)] as a function of radius for different values of driving force coefficient in this case. At the low driving force of cells $k_{mo} \leq 0.5k_B T/r_b$, the density distribution seems to be uniform anywhere except for the edge of the boundary that has a slightly higher value since driving forces there are not strong enough to induce global collectivity. At $k_{mo} > 0.6k_B T/r_b$, the density distribution at the edge of boundary is getting higher and the total number of cells moving in the central region is decreasing with the augment of the driving force of cells, which is in line with our observations in Fig. 18. This is due to the combined effect of cell motility and repulsive boundary.

The average speed of cells, depicted in Fig. 19(b), increases monotonically with increasing driving force. What is interesting here is that there is a big difference between the system with periodic boundary condition and the system in circular confinement when the global collective behavior emerges. The average speed is uniform all over the system with periodic boundary condition [see Fig. 11(b)]. However, we see a distinct lower value of average speed for the layers near the rotational vortex at the edge of the circular boundary for higher values of driving force coefficient. The reason is that when cells moving from the middle region collide with the cells in the rotational vortex, there is kinetic energy loss for the cells during this process, thus decreasing the average speed of that layer.
Figure 19. Quantifications of the effect of increasing driving force coefficient on areal density distribution and average speed of the system in circular confinement. **a.** Areal density of the system as a function of radius for $\kappa_{m0} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1,$ and $1.2k_B T / r_b$ at a given density $\rho = 0.003981 r_b^{-2}$. **b.** Average speed of the system as a function of radius for $\kappa_{m0} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1,$ and $1.2k_B T / r_b$ at a given density $\rho = 0.003981 r_b^{-2}$.
c.

Figure 20. Quantifications of clustering properties of the system. a, Average total number of clusters as a function of driving force at a given density $\rho = 0.00398 1r_b^{-2}$. b, Average maximum size of clusters as a function of driving force at a given density $\rho = 0.00398 1r_b^{-2}$. c. Average size of clusters as a function of driving force at a given density $\rho = 0.00398 1r_b^{-2}$.

Figure 21. Tangential velocity of $36^{th}$ layer of the system vs. driving force at $k_{mo} \geq 0.6$. Density $\rho = 0.00398 1r_b^{-2}$ and the radius of circular confinement is $200r_b$. 
We now turn to the quantification of the clustering properties of the system. Similarly, as the previous section, increasing driving force on the one hand decreases the total number of clusters in the system [Fig. 20(a)], on the other hand, increases the size of the biggest cluster (rotational band) [Fig. 20(b)], thereby increasing the average size of clusters of the system [Fig. 20(c)].

Since we observe that most of the cells form a big rotating cluster at the edge of the circular boundary at $k_m \geq 0.6/r_b$, will this rotational behavior of the system change its direction? To answer this question, we quantify the tangential velocity of $38^{th}$ layer near the circular boundary as a function of time where the positive value of the tangential velocity represents the clockwise direction of the vortex and the negative value means that the direction of the rotating vortex is counter-clockwise. As shown in Fig. 21, there is no sign change of tangential velocity for every system at $k_m \geq 0.6/r_b$, which shows the unidirectional rotation behavior of the vortex. In addition, a higher driving force will definitely lead to a higher tangential velocity of the cells.

The order parameter of increasing densities, shown in Fig. 22, does not change too much at $k_m \leq 0.4k_B T/r_b$ where the motility force is not strong enough to induce the global collectivity of the system. At $k_m \geq 0.6k_B T/r_b$, increasing driving force actually increases the degree of the collectivity of the system, thus raising the order parameter. Also, we can see a clear phase transition around $k_m = 0.6k_B T$, which agrees with our observation in Fig. 18. We again compare the vortical order parameter between systems of 250 cells and 500 cells. It is pretty clear that increasing density slightly decreases the collectivity of the cells moving in circular confinement.
Figure 22. Vortical order parameter as a function of driving force coefficient of the system in circular confinement. The density $\rho = 0.003981 r_b^{-2}$.

4.2.2 The effect of increasing density

In order to confirm our observation in Fig. 22, we further investigate the effect of increasing density on the collective motion of cells on a system in circular confinement at a given driving force $k_{mo} = 1k_B T/r_b$. The configuration snapshots and corresponding velocity fields, at different densities, are shown in Fig. 23. The figure shows that as density is increased in circular confinement, there are more cells moving in the middle of the system as the boundaries become saturated. More monomeric cells and small clusters induce more chaos in the system.
Figure 23. Configuration snapshots and velocity fields of the cells for \( \rho = 0.002787, 0.003185, 0.003583, \) and 0.003981 at a given driving force \( k_{mo} = 1k_B{T}/r_b \). The radius of the circular confinement \( r = 100r_b \).

As to the areal density distribution [Fig. 24(a)], we can clearly see that increasing density increases the areal density in the middle region, which is in line with our observation in Fig. 22. In addition, the thickness of the rotational vortex is also expanding inwards due to the combined effect of circular boundary and increasing density. Fig. 24(b) shows the average speed of the system as a function of density. As expected, when there are more cells moving in the system, there is a less average speed of cells due to more frequently collisions existing among cells.
a. Quantifications of areal density distribution and average speed of the system with increasing density. a, Areal density of the system as a function of radius for for $\rho = 0.001991, 0.002389, 0.002787, 0.003185, 0.003583,$ and $0.003981r_b^{-2}$ at a given driving force $\kappa_{mo} = 1k_BT/r_b$. b, Average speed of the system as a function of density at a given driving force $\kappa_{mo} = 1k_BT/r_b$.

Figure 24. Quantifications of areal density distribution and average speed of the system with increasing density. a, Areal density of the system as a function of radius for for $\rho = 0.001991, 0.002389, 0.002787, 0.003185, 0.003583,$ and $0.003981r_b^{-2}$ at a given driving force $\kappa_{mo} = 1k_BT/r_b$. b, Average speed of the system as a function of density at a given driving force $\kappa_{mo} = 1k_BT/r_b$.

We now turn to quantify the clustering properties of the system with increasing density. Following the observations in Fig. 23, we know there are more monomeric cell and small clusters moving in the central region of the system with the augment of density, which is again confirmed in Fig. 25(a). The increasing density increases the total number of clusters in the system. The maximum size of the cluster, depicted in Fig. 25(b), or the biggest size of the cluster (rotational vortex) increases with more cells in the system. Thus as expected, we see a less value of the average size of the cluster with a higher density of the system [Fig. 25(c)]. Combined with these results, it is clear that increasing density actually suppresses the collectivity of the system with high motility force.
Figure 25. Quantifications of clustering properties of the system. a, Average total number of clusters as a function of density at a given motility force $k_{m0} = 1k_B T$. b, Average maximum size of clusters as a function of driving force at a given motility force $k_{m0} = 1k_B T$. c, Average size of clusters as a function of driving force at a given motility force $k_{m0} = 1k_B T$. 
We also want to see the effect of increasing density on the rotational direction of our system. The magnitude of tangential velocity of the rotating layer, depicted in Fig. 26, actually is independent of the density, which means that the degree of the collectivity is constant for a saturated layer near the circular boundary. Furthermore, all of the systems are also in unidirectional rotation states despite of the random choice of the orientation.

In order to confirm our results of clustering properties, we again calculate the order parameter for each system. Fig. 27 shows that increasing density actually decreases the collectivity of the system. As mentioned before, this is again expected since there are more cells moving in the middle of the system as the boundaries become saturated. More monomeric cell and small clusters cause more disorder, which decreases the collectivity of the system.
Figure 27. Vortical order parameter as a function of density at a given driving force $k_{mo} = 1k_B T$. 
5. RESULTS IN CIRCULAR CONFINEMENT WITH PATTERNED SUBSTRATE

In this chapter, we would like to in turn show the spatiotemporal properties of cells as functions of the driving force, the strength of the substrate, and density. Initially, cells are placed in random positions in circular confinement of radius $R = 200r_b$, and then we simulate the system up to 4 million steps to make sure that the system reaches its steady state. The quantifications include average areal density distribution, average speed, vortical order parameter, and some clustering properties such as the total number of clusters, maximum size of clusters, and average size of clusters. All results are averaged over 2 million steps. A rich phenomenology and dynamics of collective motion of cells is revealed during the process of our simulations.

5.1 The effect of driving force

In this section, we introduce the results of our simulations on systems with $N = 500$ cells at different values of the driving force coefficient $k_{mo}$ at a given strength of substrate coefficient $k_r = 100k_BT$ and density $\rho = 0.003979r_b^{-2}$ of the system. As defined in chapter 2, driving force of cells is proportional to the polarization of each ring polymer, which is controlled by the parameter $\kappa_{mo}$. Configuration snapshots and corresponding velocity fields, at different driving force coefficients of the cells, are shown in Figure 28. This figure shows that increasing motility force leads to a bigger size of depletion region and higher density distribution in the patterned region at low values of $\kappa_{mo}$, and as we further increase the motility force to 0.6, there is a phase transition happening that the vast majority of the collectivity of the system shift from the patterned region to the edge of the circular confinement where finally the cells are spatially organized into a giant vortex with strong vortical order. In addition, the higher the driving force coefficient we use, the fewer cells moving in the middle region we can observe at $k_{mo} > 0.6$. 
This is reasonably expected since at $k_{mo} = 0$, the thermal fluctuation is the only contributing factor to the motion of cells, which gives rise to the uniformly random distribution of the ring polymers all over the system. While the driving force coefficient is further increased up to 0.6, the collectivity in the patterned region is also increasing due to the dominance of the circularly patterned substrate, which is also the reason we see a bigger size and more complete rotating vortex in the middle region. 0.6 is a critical value for the system to undergo a phase transition. Below the value $k_{mo} = 0.6$, the ring polymers stuck in the patterned region can travel longer distances in the same amount of time $\Delta t$ with a higher motility force, at the same time, higher motility force exerted on the system induce more collectivity in the patterned region, therefore leading to a bigger size of the depletion region and higher density distribution within the patterned region. Story is different at $k_{mo} \geq 0.6$ that the substrate force can no longer dominate the motion of the system, in which case cells can move out of the patterned region easily and finally densely packed together at the edge of the circular boundary due to the combined effect of cell motility and circular confinement.
$$\kappa_{m o} = 0.2$$

$$\kappa_{m o} = 0.4$$

$$\kappa_{m o} = 0.5$$
<table>
<thead>
<tr>
<th>$\kappa_{mo}$</th>
<th>Image 1</th>
<th>Image 2</th>
<th>Image 3</th>
<th>Image 4</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.6</td>
<td><img src="image1.png" alt="Image" /></td>
<td><img src="image2.png" alt="Image" /></td>
<td><img src="image3.png" alt="Image" /></td>
<td><img src="image4.png" alt="Image" /></td>
</tr>
<tr>
<td>0.8</td>
<td><img src="image5.png" alt="Image" /></td>
<td><img src="image6.png" alt="Image" /></td>
<td><img src="image7.png" alt="Image" /></td>
<td><img src="image8.png" alt="Image" /></td>
</tr>
<tr>
<td>1</td>
<td><img src="image9.png" alt="Image" /></td>
<td><img src="image10.png" alt="Image" /></td>
<td><img src="image11.png" alt="Image" /></td>
<td><img src="image12.png" alt="Image" /></td>
</tr>
</tbody>
</table>
Figure 28. Configuration snapshots and corresponding velocity fields of the cells for \( \kappa_{mo} = 0, 
0.2, 0.4, 0.5, 0.6, 0.8, 1, \) and 1.2\( k_B T/r_b \) at a given strength of the substrate \( k_{sub} = 100 k_B T \) and density \( \rho = 0.003979 \). The radius of circular confinement is 200\( r_b \) as a measure of length, while the radius of the patterned region is 100\( r_b \). Both the system and the substrate are centered at the coordinates (200, 200).

Let us move on to quantify the spatial organization and mechanical properties of the cells in the system. Fig. 29(a) shows the area density distribution as a function of radius for different values of motility force coefficient. When \( \kappa_{mo} = 0 \), the area density of the system is considered homogeneous, which agrees with our results from the configuration snapshots that cells without motility force are distributed uniformly over all the circular confinement except for some impurities in certain zones of the box. At \( \kappa_{mo} > 0 \), the first observation is that the areal density in the central region of the substrate is decreasing and gradually reaching zero, which forms the depletion region of the system. The size of the depletion region and areal density distribution in the patterned region keeps going up as we further increase the motility force at \( \kappa_{mo} < 0.6 \). This is due to the combined effect of cell motility and substrate force that the larger driving force of the system leads to more cells moving from uniform region to patterned region. At \( \kappa_{mo} \geq 0.6 \), we see a clear phase transition as the peaks of the area density distribution of the system are decreasing. This simply follows the fact that cells travel in a straight line and when the driving force is strong enough, the dominance of the substrate force is no longer existing. With higher
kinetic energy, cells are able to move out of the patterned region easily and collide with the cells rotating near the boundary of circular confinement, which leads to the velocity alignment of the ring polymers there. Finally, most of the cells form a giant vortex at the edge of the circular confinement.

The average speed of the specific layer, defined by the summation of the speed of the cells divided by the number of the cells inside the layer, depicted in Fig. 29(b), increases monotonically with higher motility force. This is expected since the cells with a stronger driving force have larger kinetic energy.

![Figure 29.](image)

Figure 29. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at different values of driving force coefficient. The substrate force coefficient $k_{sub} = 100 \ k_B T$ and density $\rho = 0.003979 \gamma_b^{-2}$. (b) Average speed of system as a function of driving force. The substrate force coefficient $k_{sub} = 100 \ k_B T$ and density $\rho = 0.003979 \gamma_b^{-2}$.

Many cells with moderate motility forces rotate in the patterned region, and in contrast, most of cells undergoing a high motility force form a giant vortex at the edge of circular confinement. For some cases, we observe a plethora of reversals of the rotation, while there is barely reversal in the system with specific values of $k_{mo}$. At a given substrate force coefficient
and density, what is the effect of motility force on the reversal rate of the system in the region where the cells rotate collectively? Fig. 30 depicts the reversal rate as a function of motility force in both patterned region (18th layer) and the uniform region near the edge of circular confinement (39th layer). Regarding the reversal rate in the patterned region, we see a clear declination due to the fact that more frequently local interactions lead to more collectivity in the region. However, when the coefficient $k_{mo}$ is further increased over 0.5, more and more cells prefer to move at the edge of circular boundary with higher kinetic energy, thus increasing the collectivity there. This is also the reason for us to observe more numbers of reversals in the patterned region at high motility force. As to the uniform region near the edge of circular confinement, initially there are relatively fewer cells moving in the region at small value of $k_{mo}$, while the majority of cells are densely packed there at $k_{mo} \geq 0.6$. As expected, the number of reversals decreases all the way to zero with the augment of coefficient $k_{mo}$ at the edge of the system. Combined with our observations in Fig. 19 and Fig. 20, we conclude that where there is more collectivity, there are relatively fewer reversals.
Figure 30. Reversal rate (number of reversals per unit time), versus driving force coefficient, in layer 18 and layer 39. The substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003979r_b^{-2}$.

We also quantify the clustering properties of the system. The average size of clusters and maximum size of clusters, depicted in turn in Figure. 31(a) and Fig. 31(b), increase with a higher motility force of the system, while Fig. 31(c) shows that the total number of clusters of the system decreases with the augment of motility force of the ring polymers. All results are expected since there are more collectivities in the system undergoing a higher value of $k_{mo}$, where more and more cells travel together and form big clusters due to frequently local interactions, thereby decreasing the total number of clusters and in contrast increasing the average size and maximum size of clusters of the system when the density is constant.

Finally in this section, we use the vortical order parameter [Fig. 32] to directly quantify the degree of collectivity for systems with different driving forces. The figure shows that there is no much collectivity in the systems at $k_{mo} \leq 0.5k_BT/r_b$ and the order parameter increases monotonically with increasing driving force coefficient at $k_{mo} \geq 0.6k_BT/r_b$, which again comply with our observations in Fig. 19 and Fig. 20.
Figure 31. (a) Average size of clusters (average number of cells per cluster) of the system as a function of driving force coefficient at a given substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003979r_b^{-2}$. (b) Maximum size of cluster (number of cells in biggest cluster) of the system as a function of driving force coefficient at a given substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003979r_b^{-2}$. (c) Number of clusters as a function of driving force coefficient at a given substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003979r_b^{-2}$. 
Figure 32. Vortical order parameter as a function of driving force coefficient at a given substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003979r_b^{-2}$.

### 5.2 The effect of stiffness of substrate

In this section, we present results of our simulations on systems with $N = 500$ cells at different values of the substrate force coefficient $k_r$ at a given driving force coefficient $k_{mo} = 0.5k_BT/r_b$ and density $\rho = 0.003979r_b^{-2}$ of the system. As defined earlier, the substrate force of the system, controlled by the parameter $k_r$, leads to the tangential alignment of cells in the patterned region. Configuration snapshots and corresponding velocity fields, at different values of the substrate force coefficient, are shown in Figure 33. Depletion region is a salient feature of the system with circular pattern of substrate as a result of the complex interplay between motility force of the cells, strength of substrate, and density of the system. While keeping the $k_{mo}$ and $\rho$ constant, it is clear from the figure that the increasing strength of substrate leads to a smaller size of depletion region and a higher areal density distribution in the patterned region. The reason is that higher stiffness of the substrate on the one hand keeps more cells moving in the patterned
region, on the other hand, it leads to a stronger effect of tangential alignment. More collisions at the interface of the substrate force the rotating cells to move inwards, thereby decreasing the size of the depletion region.

<table>
<thead>
<tr>
<th>Strength of substrate</th>
<th>Snapshot</th>
<th>Velocity field</th>
</tr>
</thead>
<tbody>
<tr>
<td>( \kappa_r = 0 )</td>
<td><img src="image1.png" alt="Snapshot" /></td>
<td><img src="image2.png" alt="Velocity field" /></td>
</tr>
<tr>
<td>( \kappa_r = 40 )</td>
<td><img src="image3.png" alt="Snapshot" /></td>
<td><img src="image4.png" alt="Velocity field" /></td>
</tr>
</tbody>
</table>
\[ \kappa_r = 80 \]

\[ \kappa_r = 100 \]

\[ \kappa_r = 140 \]
Figure 33. Configuration snapshots and velocity fields of the cells for $k_{sub} = 0, 40, 80, 100, 140, 180k_B T$ at a given motility force $k_m = 0.5k_B T$ and density $\rho = 0.003979r_b^{-2}$. The radius of circular confinement is $200r_b$ as a measure of length, while the radius of the patterned region is $100r_b$. Both the system and the substrate are centered at the coordinates $(200, 200)$.

The area density distribution of the cells, depicted in Fig. 34(a), shows that area density distribution in flocking layers increases with the augment of strength of substrate. Moreover, the left shift of the graphs indicates that the thickness of depletion region of the system is keeping reduced with the increasing $\kappa_r$. This is expected since the cells are forced to move tangentially under the effect of the substrate. The stronger the substrate, the less able the cell is to escape from the patterned region. At the same time, more and more cells move from the uniform region to the patterned region and collide with the cells in flocking layers, which push the cells in the patterned region to move inwards, thereby reducing the size of depletion region and increasing the area density of the flocking layers.

The average speed of the patterned region (layer 15-20) and uniform region (layer 20-25), depicted in Figure. 34(b), does not change in the uniform region. However, we can observe that cells move faster in the patterned region in the form of rotating vortex. In addition, the average speed decreases as the radius of system expand outwards from patterned region to uniform
region. This is due to the fact that there are fewer collisions and more collectivity for the movement of the cells in the patterned region, which actually reduces the loss of the kinetic energy for the cells in rotation band compared to the ones in the uniform region. As to the decrease of the average speed near the edge of the patterned region, it is expected since there are many collisions at the interface of two regions as mentioned before, thereby decreasing the average kinetic energy of cells there.

![Spatial areal density distribution](image-a)

![Average speed](image-b)

Figure 34. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at different values of substrate force coefficient. The driving force coefficient $k_{mo} = 0.5k_B T/r_b$ and density $\rho = 0.003979r_b^{-2}$. (b) Average speed of system versus radius (the distance between cell and origin point), at different values of substrate force coefficient. The driving force coefficient $k_{mo} = 0.5k_B T/r_b$ and density $\rho = 0.003979r_b^{-2}$.

The reversal rate of rotating vortex in the patterned region (layer 18), depicted in Fig. 35, decreases all the way to zero as we increase the strength of tangential alignment. This is simply as expected since the increasing value of $\kappa_r$ keeps more cells rotating in the patterned region, which on the one hand rises the density distribution in the region, on the other hand, increases the collectivity there. As we mentioned before, where there is more collectivity, there are fewer reversals of the area.
Figure 35. Reversal rate (number of reversals per unit time), versus substrate force coefficient, in layer 18. The driving force coefficient $k_{m1} = 0.5k_BT/r_b$ and density $\rho = 0.003979r_b^{-2}$.

Fig. 36(a), 36(b), and 36(c) in turn show the average size of clusters, the maximum size of cluster, and number of clusters in the system as a function of substrate force coefficient $k_{r1}$. More collectivity induced by increasing strength of substrate leads to a higher areal density distribution in the patterned region of the system, which also means there are fewer cells in the uniform region of the system at a given density, thus, on the one hand, decreasing the total number of clusters in the system and on the other hand increasing the number of cells in the rotating vortex. However, we observe a small decrease of the average size of clusters and the increase of total number of clusters in the system at $k_{sub} \geq 0.6$. The reason is that many small clusters containing just two or three ring polymers are formed when the density of the uniform region is low enough, thereby increasing the total number of clusters slightly. At the same time, the total number of cells in clusters more or less remains constant or increases slowly in the system, which gives rise to a small decrease of average size of clusters of the system. Overall,
more collectivity of the system induced by increasing substrate force decreases the total number of clusters and inversely increases the average size of clusters. The maximum size of cluster (the number of cells in rotating vortex) increases monotonically with the augment of substrate force.

The vortical order parameter, depicted in Fig. 37, just increases monotonically with the increasing substrate force due to more collectivities are induced by the higher value of strength of substrate.

Figure 36. (a) Average size of clusters (average number of cells per cluster) of the system as a function of substrate force coefficient at a given driving force coefficient \( k_{mo} = 0.5k_B T/r_b \) and density \( \rho = 0.003979r_b^{-2} \). (b) Maximum size of cluster (number of cells in biggest cluster) of the system as a function of substrate force coefficient at a given driving force coefficient \( k_{mo} = \)
$0.5k_B T / r_b$ and density $\rho = 0.003979r_b^{-2}$. (c) Number of clusters as a function of substrate force coefficient at a given driving force coefficient $k_{mo} = 0.5k_B T / r_b$ and density $\rho = 0.003979r_b^{-2}$.

Figure 37. Vortical order parameter as a function of substrate force coefficient at a given driving force coefficient $k_{mo} = 0.5k_B T / r_b$ and density $\rho = 0.003979r_b^{-2}$.

5.3 The effect of density

In this section, we present the results of our simulations on systems at different densities $\rho$ at a given driving force coefficient $k_{mo} = 0.5k_B T / r_b$ and substrate force coefficient $k_r = 100k_B T$ of the system. Density is defined as total number of ring polymers divided by the area of the circular confinement. Spatial configuration snapshots and velocity fields of steady states of systems, at different densities, are shown in Fig. 38. The figure shows that as we increase the density of the system, the average size of the depletion region is getting smaller with a higher areal density distribution in the patterned region. This simply follows the fact that there are more local interactions with more cells in the system, directly leading to more collectivities in the patterned region.
<table>
<thead>
<tr>
<th>Density</th>
<th>Snapshot</th>
<th>Velocity field</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\rho = 0.002787$</td>
<td><img src="image1.png" alt="Snapshot" /></td>
<td><img src="image2.png" alt="Velocity field" /></td>
</tr>
<tr>
<td>$\rho = 0.003185$</td>
<td><img src="image3.png" alt="Snapshot" /></td>
<td><img src="image4.png" alt="Velocity field" /></td>
</tr>
<tr>
<td>$\rho = 0.003583$</td>
<td><img src="image5.png" alt="Snapshot" /></td>
<td><img src="image6.png" alt="Velocity field" /></td>
</tr>
</tbody>
</table>
\[ \rho = 0.003981 \]

Figure 38. Configuration snapshots and velocity fields of the cells for \( \rho = 0.00278, 0.003185, 0.00358, \) and \( 0.003981 \) at a given driving force coefficient \( \kappa_{mo} = 0.5k_B T/\tau_b \) and substrate coefficient \( \kappa_r = 100k_B T \). The radius of circular confinement is \( 200r_b \) as a measure of length, while the radius of the patterned region is \( 100r_b \). Both the system and the substrate are centered at the coordinates \( (200, 200) \).

The area density distribution, depicted in Fig. 39(a), increases with more cells in the system. In addition, the thickness of the depletion region also decreases with the augment of the density. This is due to the fact that the cells in uniform region tend to move everywhere, including the central patterned region. The higher is the density the more cells “invade” the patterned region, thereby reducing the size of depletion region. In the meantime, we also see the increase of the density in the patterned region, which is in line with our observations in Fig. 29.

The average speeds of the patterned region (layer 15-20) and uniform region (layer 20-25), depicted in Figure. 39(b) have inverse responses to the increasing density. From the former results in section 4.2, we know that more cells lead to more collisions of the uniform system, thereby decreasing the average speed, which is confirmed in Fig. 39(b) again. However in contrast, we do see an interesting increase of average speed of the patterned region as a result of more collectivity of the system induced by higher density. As expected, more collectivity in the
patterned region leads to a higher average speed. This is really a point showing the effect of patterned substrate on the physical properties of cells.

![Figure 39](image)

**Figure 39.** (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at density. The driving force coefficient $k_{m_0} = 0.5k_B T/r_b$ and substrate force coefficient $k_r = 100k_B T$. (b) Average speed of system versus radius (the distance between cell and origin point), at different density. The driving force coefficient $k_{m_0} = 0.5k_B T/r_b$ and substrate force coefficient $k_r = 100k_B T$. 

65
Figure 40. Reversal rate (number of reversals per unit time), versus density, in layer 18. The driving force coefficient $k_{mo} = 0.5k_B T / r_b$ and substrate force coefficient $k_r = 100k_B T$.

Once again, we quantify the reversal rate of rotating vortex as a function of density here. Obviously, larger density, as shown in Fig. 40, increases the collectivity in the patterned region, which leads to the decrease of number of reversals. This figure also confirms again that when there is enough collectivity of the patterned region, the rotating behavior of the vortex is unidirectional where the number of reversals vanishes to zero.

Fig. 41(a), 41(b), and 41(c) in turn show the average size of clusters, the maximum size of clusters, and the total number of clusters as a function of density in the system. With the increasing density of the system, the collectivity in the patterned region is strong enough to form a relatively more complete rotational band. While more and more cells from the uniform region join the rotating vortex in the patterned region, we observe a clear declination of the total number of clusters. Regarding the average size and maximum size of the clusters in the system, they just
keep increasing since there are more ring polymers in the system. In short, more collectivity of the system induced by increasing density decreases the total number of clusters and inversely increases the average size of clusters. The maximum size of the cluster (the number of cells in the rotating vortex) increases monotonically with the augment of substrate force.

To comply with our previous results in this section, we again quantify the vortical order parameter as a function of density [Fig. 42]. The figure perfectly tells us that increasing density does lead to more collectivity in the system.

![Graphs](image)

Figure 41. (a) Average size of clusters (average number of cells per cluster) of the system as a function of density at a given driving force coefficient $k_{mo} = 0.5k_B T/r_B$ and substrate force
coefficient $k_r = 100k_B T$. (b) Maximum size of cluster (number of cells in biggest cluster) of the system as a function of density at a given driving force coefficient $k_{m0} = 0.5k_B T/r_b$ and substrate force coefficient $k_r = 100k_B T$. (c) Number of clusters as a function of density at a given driving force coefficient $k_{m0} = 0.5k_B T/r_b$ and substrate force coefficient $k_r = 100k_B T$.

Figure 42. Vortical order parameter as a function of density at a given driving force coefficient $k_{m0} = 0.5k_B T/r_b$ and substrate force coefficient $k_r = 100k_B T$. 
6. RESULTS IN PERIODIC BOX WITH PATTERNED SUBSTRATE

In the preceding chapter, the collective behavior of ring polymers is investigated in the circular confinement. Here, we would like to in turn show the spatiotemporal properties of cells as functions of driving force, the strength of substrate, and density in a 400 × 400 periodic box. In the central region of the system, there is also a circularly patterned region with radius \( R = 100r_b \) centered at origin. The quantifications include average areal density distribution, average speed, normalized tangential momentum, and some clustering properties such as total number of clusters, maximum size of clusters, and average size of clusters. Again, we simulate systems up to 4 million steps to make sure that system reaches its steady state. All results are averaged over 2 million steps. Compared to the system in circular confinement, different phenomenology and dynamics of collective motion are revealed during the process of our simulations.

6.1 The effect of driving force

In this section, The effect of increasing value of driving force coefficient \( k_{mo} \) on the collective motion of cells in a periodic box with patterned substrate is our concern here at a given substrate force coefficient \( k_r = 100k_BT \) and density \( \rho = 0.003125r_b^{-2} \). Note that cells are more flexible to migrate in the system since they can pass through the boundary of periodic box and come back from another side, which gives rise to a different migratory pattern compared to the system in circular confinement. Configuration snapshots as well as corresponding velocity fields, at different driving forces of the cells, are shown in Fig. 43. The figure shows that at low motility force, increasing \( k_{mo} \) typically increases both the areal density distribution of cells and the size of the depletion region in the patterned region up to the value \( \kappa_{mo} = 0.6k_BT/r_b \). In addition, we see a maximum collectivity of the system at \( \kappa_{mo} = 0.6 \) However, we observe a reduction of both area density and the size of the depletion region at \( \kappa_{mo} > 0.6 \). All these observations follow the
fact that the collective motion of the cells emerges as a result of competition among driving force, strength of the substrate, and density. At $\kappa_{mo} = 0$, there are only thermal fluctuations affecting the movement of ring polymers that the areal density distribution is uniform in the system. As we keep increasing the motility force at $\kappa_{mo} \leq 0.6$, cells are able to travel longer distances in the same amount of time $\Delta t$. The velocity alignments resulted from more frequently local interactions finally lead to the aggregation of the cells into isotropic clusters forming rotating vortex in the middle region. In particular, substrate force is still strong enough to keep the cells staying longer in the patterned region when system undergoes relatively low motility force, thus increasing both the areal density distribution and the size of depletion in the patterned region. The story is different for the system at $\kappa_{mo} > 0.6$. Driving force is strong enough to dominate the motion of ring polymers that ring polymers are more able to either move closer to the origin of the system or move out of the patterned region based on their direction of movement, which leads to a smaller size of depletion region and lower areal density distribution in the patterned region.

<table>
<thead>
<tr>
<th>Motility force</th>
<th>Snapshots</th>
<th>Velocity fields</th>
</tr>
</thead>
<tbody>
<tr>
<td>$k_{mo} = 0$</td>
<td><img src="image.png" alt="Snapshots" /></td>
<td><img src="image.png" alt="Velocity fields" /></td>
</tr>
<tr>
<td>$k_{mo}$</td>
<td>Image 1</td>
<td>Image 2</td>
</tr>
<tr>
<td>---------</td>
<td>---------</td>
<td>---------</td>
</tr>
<tr>
<td>0.2</td>
<td><img src="image1.png" alt="Image" /></td>
<td><img src="image2.png" alt="Image" /></td>
</tr>
<tr>
<td>0.4</td>
<td><img src="image4.png" alt="Image" /></td>
<td><img src="image5.png" alt="Image" /></td>
</tr>
<tr>
<td>0.5</td>
<td><img src="image7.png" alt="Image" /></td>
<td><img src="image8.png" alt="Image" /></td>
</tr>
<tr>
<td>( k_{m_0} )</td>
<td>0.6</td>
<td>0.8</td>
</tr>
<tr>
<td>-----------</td>
<td>-----</td>
<td>-----</td>
</tr>
</tbody>
</table>

\[ k \_m \_o = 0.6 \]
\[ k \_m \_o = 0.8 \]
\[ k \_m \_o = 1 \]
$k_{mo} = 1.2$

Figure 43. Configuration snapshots and corresponding velocity fields of the cells for $k_{mo} = 0, 0.2, 0.4, 0.5, 0.6, 0.8, 1,$ and $1.2k_B T$ at a given substrate force coefficient $\kappa_{sub} = 100k_B T$ and density $\rho = 0.003125r_b^{-2}$. The length of the periodic box equals $400r_b$ as a measure of length, while the radius of the patterned region is $100r_b$. Both the system and the patterned substrate are centered at the origin $(200, 200)$.

Let's move on to quantify the spatial organization and mechanical properties of the cells in the system. Fig. 44(a) depicts the area density distribution as a function of radius in the system for different values of driving force coefficient. The figure shows both areal density distribution and size of depletion region increase with the augment of driving force below 0.6. At $\kappa_{mo} > 0.6$, increasing driving force conversely decreases the areal density and size of depletion in the patterned region, which agrees with the observations in Fig. 43. This again follows the fact that increasing motility force increases the kinetic energy of the cells leading to more collisions and velocity alignments near the boundary of patterned region. While cells have stronger ability to move inside the patterned region, more cells in the uniform region also are able to invade the patterned region at the same time, thereby increasing the size of depletion region and the area density of patterned region. At $\kappa_{mo} > 0.6$, high motility force dominates the cell motion since we know that the collective motion of the cells results from the interplay among the motility force, strength of the substrate, and density of the system. When the motility force of cells is strong enough, cells are more able to either move closer to the origin of the system or move out of the
patterned region based on their direction of movement, which leads to a smaller size of depletion region and lower areal density distribution in the patterned region.

Average speed of a specific layer, depicted in Fig. 44(b), increases monotonically as the augment of the motility force. This is expected since higher motility force leads to the faster movement of the cells. In addition, for the cases where there are lot of collectivity (rotating vortex in the middle region) happening, the average speed in the region is higher than the one in the uniform region. Once again, there are fewer collisions and more collectivity in the middle vortex leading to less energy loss, which causes a higher average speed of the region.

![Figure 44(a)](image1)

![Figure 44(b)](image2)

Figure 44. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at different values of driving force coefficient. The substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003125r_b^{-2}$. (b) Average layer speed (average speed per cell in the layer), versus radius at different values of driving force. The substrate force coefficient $k_r = 100k_BT$ and density $\rho = 0.003125r_b^{-2}$.

Fig. 45 depicts the reversal rate of the rotating vortex ($18^{th}$ layer) as a function of driving force coefficient. The figure shows that at the low value of $\kappa_{mo}$, there are a plethora of reversals. The direction of the cell’s movement in the patterned region remains more or less stable when the system undergoes a moderate motility force. Finally, more reversals come out at
high motility force. All these observations are expected since there is relatively less collectivity in the patterned region at small $\kappa mo$, while a full “donut” (vortex) is formed in the patterned region at moderate motility force. Finally, due to the higher kinetic energy resulted from higher motility force, the density of the patterned region decreases, which breaks the “donut” and leads to less collectivity of the motion causing a greater number of reversals in the patterned region.

The figure tells us that if there is a big enough collectivity in the middle region, the direction of the rotating vortex is unidirectional as always.

Figure 45. Reversal rate (number of reversals per unit time), versus driving force coefficient, in layer 18. The substrate force coefficient $k_r = 100 k_B T$ and density $\rho = 0.003125 r_b^{-2}$.
We again quantify the clustering properties of the system with the periodic boundary condition. Similarly, average size and maximum size of clusters, depicted in Fig. 46(a) and 46(b), initially increase with the augment of driving force at $k_{mo} \leq 1k_B T/r_b$ and then decrease a little bit, while Fig. 43(c) shows that the total number of clusters first increases slightly and then decreases monotonically with increasing driving force of the ring polymers. Combined the results with Fig. 24(a), these results are intuitively expected since the number of reversals is related to the collectivity in the region. At low value of driving force coefficient less than 0.5, we observe the formation of a greater size of giant vortex in the patterned region, which means fewer cells are moving in the uniform region and a smaller number of clusters in the system. Increasing motility typically increases the areal density distribution in the patterned region and collectivity in the whole system. Since average size of clusters is defined as the total number of cells in clusters divided by the total number of clusters in the system, it is not astonishing that average size of clusters keeps increasing in this case. Furthermore, the slopes of Fig. 46(a) and 46(b) become less and less shaper on account of less collectivity in the patterned region at $\kappa_{mo} > 0.6k_B T/r_b$. Finally, the average size and maximum size of clusters even tend to decrease at $\kappa_{mo} > 1k_B T/r_b$ due to more chaos in the middle region, which is also the difference between the system with periodic boundary condition and in circular confinement. Cells are freer to move in the periodic box.

Finally in this section, let us take a look at the change of collectivity of cells directly in the patterned region. Fig. 47 shows normalized tangential momentum of the middle region as a function of radius for different values of driving force coefficient. At $k_{mo} \leq 0.6k_B T/r_b$, the collectivity of the middle region just increase with the augment of driving force of cells. The collectivity of the patterned region is in its maximum at $k_{mo} = 0.6k_B T/r_b$, then begins to
decrease with higher values of driving force coefficient, which is exactly what we observe in Fig. 34. From this graph, it is also easy for us to see the different effects of confinements combined with Fig. 28 and Fig. 32.

Figure 46. (a) Average size of clusters (average number of cells per cluster) of the system as a function of driving force coefficient at a given substrate force coefficient $k_r = 100k_B T$ and density $\rho = 0.003125r_b^{-2}$. (b) Maximum size of cluster (number of cells in the biggest cluster) of the system as a function of driving force coefficient at a given substrate force coefficient $k_r = 100k_B T$ and density $\rho = 0.003125r_b^{-2}$. (c) Number of clusters as a function of driving force coefficient at a given substrate force coefficient $k_r = 100k_B T$ and density $\rho = 0.003125r_b^{-2}$.
Figure 47. Normalized tangential momentum (see method), versus radius for different values of driving force coefficient in the system with periodic boundary condition. The substrate force coefficient $k_r = 100k_B T$ and density $\rho = 0.003125r_b^{-2}$.

6.2 The effect of stiffness of substrate

In this section, we focus on investigating the effect of circularly patterned substrate on the collective motion of cells in a periodic box at a given driving force coefficient $k_{mo} = 0.5k_B T/r_b$ and density $\rho = 0.003125r_b^{-2}$. Configuration snapshots and corresponding velocity fields, at different values of substrate force coefficient, are shown in Fig. 48. From our observation, it is pretty clear that increasing substrate force leads to a higher areal density distribution and a smaller size of depletion in the patterned region, which means that stronger substrate force induces more collectivity of the system. This simply follows the fact that more cells are moving collectively in the patterned region induced by higher values of substrate force coefficient. In addition, more collisions at the interface between the uniform substrate and
patterned region push the rotating cells to move inwards, thereby decreasing the size of the depletion region.

<table>
<thead>
<tr>
<th>Strength of substrate</th>
<th>Snapshots</th>
<th>Velocity field</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\kappa_r = 0$</td>
<td><img src="image" alt="Snapshot $\kappa_r = 0$" /></td>
<td><img src="image" alt="Velocity field $\kappa_r = 0$" /></td>
</tr>
<tr>
<td>$\kappa_r = 40$</td>
<td><img src="image" alt="Snapshot $\kappa_r = 40$" /></td>
<td><img src="image" alt="Velocity field $\kappa_r = 40$" /></td>
</tr>
<tr>
<td>$\kappa_r = 80$</td>
<td><img src="image" alt="Snapshot $\kappa_r = 80$" /></td>
<td><img src="image" alt="Velocity field $\kappa_r = 80$" /></td>
</tr>
</tbody>
</table>
Figure 48. Configuration snapshots and velocity fields of the cells for $\kappa_r = 0, 40, 80, 100, 140, 180k_BT$ at a given motility force $k_{mo} = 0.5k_BT/\tau_b$ and density $\rho = 0.003125r_b^{-2}$. The length of the periodic box is $400r_b$ as a measure of length, while the radius of the patterned region is $100r_b$. Both the system and the substrate are centered at the origin (200, 200).
The area density distribution of the cells, depicted in Fig. 49(a), increases monotonically with higher values of substrate force coefficient and in contrast, the thickness of depletion is decreasing, which is again expected since stronger substrate force tends to keep the ring polymers moving a relatively longer time in the patterned region. In the meantime, more cells from the uniform region collide with the cells rotating at the interface between the rotating vortex and uniform region, thus resulting in a smaller size of depletion.

The average speed of the patterned region (layer 15-20) and uniform region (layer 20-25), depicted in Fig. 49(b), directly show the effect of patterned substrate on the motion of cells. When \( k_r = 0 \), the average speed is uniform everywhere in the periodic box. At \( k_r > 0 \), the figure shows that stronger substrate force leads to a larger average speed in the patterned region, which again confirms our previous result that more collectivity will induce a higher average speed of the cells rotating in the patterned region. In addition, we do see a decrease of average speed around the interface of two regions (layer 20) since there are a lot of collisions of cells there.

![Figure 49](image)

Figure 49. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at different values of substrate force coefficient. The driving force coefficient \( k_{mo} = 0.5k_B T/r_b \) and density \( \rho = 0.003125r_b^{-2} \). (b) Average layer speed (average speed per cell in
the layer), versus radius at different values of substrate force coefficient. The driving force coefficient \( k_{m,o} = 0.5k_B T/r_b \) and density \( \rho = 0.003125r_b^{-2} \).

The reversal rate of rotating vortex in the patterned region (layer 18), depicted in Fig. 50, again decreases monotonically with the augment of substrate force. As mentioned before, more collectivity will induce a smaller number of reversals in the patterned region. The total number of reversals just decreases all the way to zero with the increasing collectivity of the patterned region.

![Figure 50](image-url)

**Figure 50.** Reversal rate (number of reversals per unit time), versus substrate force coefficient, in layer 18. The driving force coefficient \( k_{m,o} = 0.5k_B T/r_b \) and density \( \rho = 0.003125r_b^{-2} \).

Fig. 51(a), 51(b), and 51(c) in turn depict the average size, maximum size, and the total number of reversals in the system as a function of substrate force. The average size of clusters sharply increases to the maximum value then fluctuates in a small range there, while the total
number of cells inside the rotating vortex or maximum size of clusters keeps increasing with the augment of substrate force coefficient. As to the total number of clusters in the system, it also fluctuates in a small range as we further increase the value of $k_r$ at high substrate force. It seems that there is a threshold value above $100k_BT$ that the system reaches a substrate-saturated state.

In contrast to the order parameter of the system with increasing $k_r$ in circular confinement, normalized tangential momentum of the middle region, depicted in Fig. 52, initially increases monotonically with the increase of substrate force. However, we do see a saturated state of system here that the collectivity of the cells remains constant when the substrate force constant is strong enough. This is also a difference between the two boundary conditions.

\[a.\]

\[b.\]
Figure 51. (a) Average size of clusters (average number of cells per cluster) of the system as a function of substrate force coefficient at a given driving force coefficient $k_{mo} = 0.5k_B T/r_b$ and density $\rho = 0.003125 r_b^{-2}$. (b) Maximum size of the cluster (number of cells in biggest cluster) of the system as a function of substrate force coefficient at a given driving force coefficient $k_{mo} = 0.5k_B T/r_b$ and density $\rho = 0.003125 r_b^{-2}$. (c) Number of clusters as a function of substrate force coefficient at a given driving force coefficient $k_{mo} = 0.5k_B T/r_b$ and density $\rho = 0.003125 r_b^{-2}$.

Figure 52. Normalized tangential momentum (see method), versus radius for different values of substrate force coefficient in the system with periodic boundary condition. The driving force coefficient $k_{mo} = 0.5k_B T/r_b$ and density $\rho = 0.003125 r_b^{-2}$.
6.3 The effect of density

In this section, we present results of our simulations on systems at different densities $\rho$ at a given driving force coefficient $k_{\text{mo}} = 0.5k_B T/r_b$ and substrate force coefficient $k_r = 100k_B T$ of the system. Fig. 53 shows the spatial configuration snapshots and the corresponding velocity fields at different densities of the system. The figure demonstrates clearly that more cells increase the areal density distribution and conversely decrease the size of depletion in the patterned region since more local interactions will induce more collectivity. However, we do see in this case that the effect of density on the collective motion of cells is weaker than the case where cells move in a circular confinement. There is more freedom for cells to move in a periodic box compared to the circular confinement case.

<table>
<thead>
<tr>
<th>Density</th>
<th>Snapshots</th>
<th>Velocity field</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\rho = 0.002188$</td>
<td><img src="image.png" alt="Snapshots Image" /></td>
<td><img src="image.png" alt="Velocity field Image" /></td>
</tr>
</tbody>
</table>
\[ \rho = 0.0025 \]

\[ \rho = 0.002813 \]

\[ \rho = 0.003125 \]

Figure 53. Configuration snapshots and velocity fields of the cells for \( \rho = 0.002188, 0.0025, 0.002813, \) and 0.003125 at a given driving force coefficient \( k_{\text{mo}} = 0.5k_B T/r_b \) and substrate force coefficient \( k_r = 100k_B T \). The length of the periodic box is 400\( r_b \) as a measure of length, while the radius of the patterned region is 100\( r_b \). Both the system and the substrate are centered at the origin (200, 200).
Fig. 54(a) illustrates the areal density distribution as a function of radius for different densities in the system. It is clearly presented that the areal density increases monotonically with a higher density in the patterned region. The reason is again that the cells out of the patterned region or in the uniform region tend to move everywhere, including the central patterned region. The higher is the density the more cells “invade” the patterned region from the uniform region, thereby increasing the total number of cells and reducing the size of the depletion in the patterned region. Fig. 54(b) depicts the correlation between the density of the system and the average speed. The figure shows that in the patterned region, the average speed slightly increases with higher density, which also agrees with our previous results that more collectivity induces a higher average speed on account of less energy loss. Furthermore, we do see a decrease of the average speed of uniform region with more cells moving in the system. As mentioned in Chapter 4, more collisions are leading to more kinetic energy loss in this case at a constant driving force coefficient of the system.

Figure 54. (a) Spatial areal density distribution, versus radius (the distance between cell and origin point), at different densities. The driving force coefficient $k_{mo} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. (b) Average layer speed (average speed per cell in the layer),
versus radius at different densities. The driving force coefficient $k_{mo} = 0.5k_B T / r_b$ and substrate force coefficient $k_r = 100k_B T$.

As we increase the total number of cells in the system, the reversal rate of rotating vortex (layer 18), depicted in Fig. 55, decreases all the way to zero with more collectivity induced by frequently local interactions.

Fig. 56(a), 56(b), and 56(c) are in turn show the average size, maximum size, and total number of clusters as a function of density in the system. Interestingly, we observe an increase of number of clusters initially even with higher density. The reason is that the collectivity of the system is not strong enough for small densities where adding more cells just increases the chaos of the system. Cells are freer to move in the periodic box than a circular confinement that the rate of areal density change of uniform region is slightly higher than the one in the patterned region, which causes the aggregation of small clusters in the system although there are also more some cells moving inside the patterned region. When the density is large enough, increasing density absolutely increases average size and maximum size of clusters in the system simply because there are more cells joining the rotating vortex in the middle region. This figure also implies the different effects of confinement on the collective motion of cells.

Fig. 57 exactly confirms our results in this section that normalized tangential momentum doesn’t increase with the increasing density from $0.002188r_b^{-2}$ to $0.0025r_b^{-2}$ because these densities are not enough to induce big collectivity of the patterned region, while it just goes up with more cells moving in the system at $\rho > 0.0025r_b^{-2}$. 
Figure 55. Reversal rate (number of reversals per unit time), versus radius (the distance between cell and origin point), at different densities. The driving force coefficient $k_{mo} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$.

a. 

b. 

Average size of clusters vs. Density 

Maximum size of cluster vs. Density
Figure 56. (a) Average size of clusters (average number of cells per cluster) of the system as a function of density at a given driving force coefficient $k_{m_0} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. (b) Maximum size of cluster (number of cells in biggest cluster) of the system as a function of density at a given driving force coefficient $k_{m_0} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. (c) Number of clusters as a function of density at a given driving force coefficient $k_{m_0} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$.

Figure 57. Normalized tangential momentum (see method), versus radius for different densities. The driving force coefficient $k_{m_0} = 0.5k_BT/r_b$ and substrate force coefficient $k_r = 100k_BT$. 

90
7 CONCLUSIONS AND SUMMARY

In this work, we constructed a relatively simple ring-polymer model to investigate the collective motion of cells on uniform and patterned substrates. Molecular dynamics simulations of a system composed of many such cells are employed to investigate the spatiotemporal organization and collective dynamics of cells under many constraints, including the driving force of the cells, the strength of the interaction of the cells with the circularly patterned substrate, the areal density of the cells, and geometric confinement. The main conclusions of this study:

1. The effect of driving force

   • When cells move in a system with periodic boundary conditions along the x- and y- axes, which correspond to the case of a very large system, on a uniform substrate, increasing the driving force typically leads to a higher average speed of cells. The cells moving with higher speed will collide with each other more frequently, which gives rise to local alignments, inducing a giant traveling band and coherent locomotion of the cells.

   • When a circularly patterned region is introduced in the middle of the system with periodic boundary conditions, we observe a depletion in the very center of the patterned region. Moderate driving force induces a complete rotating vortex in the middle region of the system. High driving force helps reduce the dominance of patterned substrate on the collective motion of cells, thus transferring the majority of the collectivity from patterned region to uniform region.

   • When the cells are confined in a circular box with a uniform substrate, high driving force induces a giant unidirectional vortex at the edge of the circular boundary due to the combined effects of cell motility and repulsive circular wall.
• When the cells are confined in a circular box with a patterned substrate, substrate force dominates the collective motion of cells at an intermediate driving force where we observe a unidirectional vortex in the middle region. At high driving forces, the cells again form a giant rotating vortex at the edge of circular confinement.

• In summary, increasing driving force induces more collectivity of the system and there is a critical value leading to the global collective motion of cells. The presence of anisotropic substrate enhances the collectivity of the cells within the region of the patterned substrate, while leading to a depletion region in the very center of the pattern. However, at large driving forces, the increased collectivity of the cells outside the patterned region leads to a reduction of the degree of collectivity within the patterned region.

2. The effect of strength of substrate

• Depletion is a salient feature of the system with the circularly patterned substrate.

• At moderate driving force and high density of the system, with periodic boundary conditions, increasing the substrate's interaction with the cells leads to a higher areal density distribution in the patterned region and a depletion zone in the very center of the patterned region. The cells form a rotating vortex in the patterned region.

• At moderate driving force and high density in the case of circular confinement, increasing substrate force has a stronger effect on generating the rotating vortex in the middle region compared to the case of a system with periodic boundary conditions.
• In summary, increasing substrate force typically increases the collectivity of the cells in the patterned region.

3. The effect of density

• In the case of uniform substrate, increasing the density of the cells increases the total number of collisions of cells, thereby inducing more collectivity of the system where large coherent clusters are formed.

• In the case of a patterned substrate, increasing the density increases the size of the rotating vortex in the middle region, thus inducing more collectivity of the system.

• In summary, increasing density induces more collectivity of the system at a moderate driving force and substrate force.

This thesis represents a continuous exploration of the ring polymer model established by my PI's research group. In the previous work, Ph.D. student Yu Zhu, in our group, used molecular dynamics simulations to investigate the equilibrium spatial organization of the circular ring polymers, under various constraints including density and bending rigidity of polymers. In addition, the effect of the isotropic substrate along the y-direction on the spatial arrangements of circular ring polymers is also investigated. In this work, we apply the ring polymer model to investigate the collective motion of living cells on both uniform and patterned substrates, under three important constraints including the driving force of the cells, the strength of the substrate, and areal density. Here are some future outlooks on this project:

• We will add the frictional forces between cells based on two-body interactions among monomers.
• Different boundary conditions will be applied to our system such as square confinement and periodic boundary conditions with different length.

• Other substrate patterns will be embedded in the central region of the system.

• The effect of many other important constraints, including bending rigidity, cell shape, and area constraint, on the collective motion of the cells, will be investigated.
Reference


[33] B. Camley, Y. Zhang, Y. Zhao, B. Li, E. Ben-Jacob, H. Levine and W. J. Rappel, "Polarity mechanisms such as contact inhibition of locomotion regulate persistent rotational motion of mammalian cells on micropatterns", Proceedings of the National Academy of Sciences, vol. 111, 14770-14775, 2014.


